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Hydrogen Cancer Control

Theory and Practice

Revolutionary Exploration of Tumor Rehabilitation

Natural Gas for Home Rehabilitation — Physiological Gas with Medicinal Effect

Editor: Xu Kecheng

Published by the International Society of Clinical Hydrogen Medicine

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About this edition

This document is a faithful, verbatim transcription of the published book *Hydrogen Cancer Control: Theory and Practice* (editor: Xu Kecheng), prepared as a searchable reference edition presented by Hydrogen Therapy H2E.

The text, figures described in captions, data tables, case reports, references, and all claims and opinions are those of the original authors and the original publisher. They are reproduced here for information and internal communication of health-care knowledge, and have not been altered, endorsed, or independently verified.

Not medical advice. Nothing in this book is a substitute for professional medical care. Within the book, hydrogen inhalation is presented as a complementary or adjunctive measure alongside — not in place of — conventional cancer treatment. Decisions about diagnosis and treatment should always be made together with a qualified oncologist or physician.

The original work carries its own publisher's notice, reproduced verbatim in the front matter that follows.

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Revolutionary Exploration of Tumor Rehabilitation

Natural Gas for Home Rehabilitation — Physiological Gas with Medicinal Effect

Editor: Xu Kecheng

Published by the International Society of Clinical Hydrogen Medicine

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Editorial Board

- **Editor:** Xu Kecheng
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- **Observed by:**
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  - Wang Zhenyi, academician of the Chinese Academy of Engineering
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About the Editors

Chief Editor — Xu Kecheng

Born in Nantong, Jiangsu, Xu Kecheng is a professor, a chief physician, and a doctoral supervisor. After graduating from the Department of Medicine of Nantong Medical College (now Medical College of Nantong University) in 1963, Xu Kecheng became a teacher in his Alma Mater, and moved up the career ladder from being a teaching assistant (resident physician), lecturer (attending physician), associate professor (vice chief physician) to professor (chief physician) successively. In the 1980s and 1990s, he went to several universities for advanced study and training in Japan and the U.S. successively. Later, Xu Kecheng founded Guangzhou Fuda Cancer Hospital, an affiliated hospital of Jinan University. Currently, he is the honorary chief president of Fuda Cancer Hospital and lifetime honorary president of Jinan University Institute of Biomedical Transformation. Xu Kecheng once edited and published *Modern Treatment of Digestive Diseases*, the earliest monograph on digestive diseases, and *Clinical Pancreatology*, the first book dedicated to pancreatic diseases in China. In the past few years, Xu Kecheng focused on researching cryoablation-led minimally invasive treatment of tumor, and tumor rehabilitation that centered on immunity and integrative Chinese and western medicine. So far, Xu Kecheng has authored nine monographs, of which *Modern Cryosurgery for Cancer* (Chinese and English versions) was the first of its kind in the world, while *New Technique of Tumor Ablation: Irreversible Electroporation* the first in China. In addition, he has published more than 500 papers, over 100 of which were science citation index (SCI) ones covering tumor ablation, immunity and rehabilitation. Xu Kecheng has also edited such popular science works as *Unusual Stories*, *Nothing But the Truth*, *Fight Cancer*

with Me, and *Practice China's Way of Cancer Control*. As the former president and current honorary president of International Society of Cryosurgery, Xu Kecheng is the founder, legal person and honorary president of Asian Society of Cryosurgery. Having won multiple major awards in international cryotherapy, Xu Kecheng was praised as an outstanding contributor to the world's cryo-technology by WHO Forum in Geneva. Moreover, he received multiple national-level honors, such as Top Ten People Inspiring Guangdong China, Bethune Medal winner, and Model of the Times.

Co-editor — Lu Youyong

Graduating from the Department of Cell Genetics at Beijing Medical University in 1982, Lu Youyong is a second-grade professor at Peking University Cancer Hospital, and a senior researcher and doctoral tutor of Beijing Research Institute for Cancer Prevention and Treatment. From 1986 to 1990, Lu Youyong was a postdoctoral student and associate researcher at National Cancer Institute (NCI) Laboratory of Molecular Oncology of U.S. National Institutes of Health (NIH). In 1991 to 1993, he became a visiting researcher of the Cell and Molecular Genetics Laboratory at Fox Chase Cancer Center. Lu Youyong has been awarded with multiple honors: The National Science Fund for Distinguished Young Scholars, Professor of the Outstanding Talent Award Program of Peking University (Medical Science), winner of the Outstanding Professor Award of USA Chinese Medical Board (1994), winner of the National Top 100 Young Medical Science and Technology Stars (1993), State Council Special Allowance Specialist (1992), to name but a few. He has successively served as the vice chairman and chairman of the Beijing Environmental Mutagen Society; a deputy editor of *World Journal of Gastroenterology* (WJG); the editorial boards of journals such as the *Journal of Pathology*; a visiting (honorary) professor at Chinese University of Hong Kong; an expert member of the National Key Basic Research Program of China (973 Program) Health Science Expert Advisory Group; an executive member of the International Cancer Genome Consortium (ICGC) Science and Technology Committee; and the Secretary General of Chinese Cancer Genome Consortium (CCGC). He has led major research projects under The National Science Fund for Distinguished Young Scholars as well as national 973 and 863 Programs. So far, Lu Youyong has published more than 200 papers and literature reviews in leading journals, such as *Cancer Research*, *Molecular and Cellular Biology*, *Nature*, *Gut*, and *Gastroenterology*. Moreover, he is the winner of more than 10 scientific and technological achievement awards at national, military and provincial levels.

Co-editor — Wu Peihong

Wu Peihong is famous both at home and abroad for his expertise in minimally invasive diagnosis & treatment and immuno-biological therapy of cancer. He was recognized as one of the Top Ten Persons of Year 2018 Healthy China. Apart from being the incumbent director of Medical Imaging and Minimally Invasive Intervention Center at Sun Yat-sen University Cancer Center, Wu Peihong is also a second-grade professor, first-grade chief physician, doctoral supervisor, and one of the first well-known doctors of Sun Yat-sen University. He was successively recognized as a Famous Doctor of Guangzhou and Famous Doctor in the South of the Five Ridges (south China's Guangdong and Guangxi provinces); and was ranked in Top 10 Distinguished Specialist Physicians in China. Moreover, Wu Peihong is a visiting professor of the Medical Schools at University of Rochester and University of Toronto. He is also the founder and incumbent chairperson of the specialized committee of minimally invasive intervention, World Association of Chinese Oncologist. His other titles include: chairman and executive director of Asia-Pacific Association of Image-Guided Therapy of Oncology

(APAITO); chairman and executive director of Asian Society of Cancer Ablation Therapy; executive director of China Anti-Cancer Association; founder, former chairperson and lifelong honorary chairperson of China Anti-Cancer Association Expert Committee for Cancer Minimally Invasive Treatment; chairperson of China Anti-Cancer Association Expert Committee for Liver Cancer Minimally Invasive and Comprehensive Treatment, and China Anti-Cancer Association Expert Committee for Cancer Minimally Invasive Treatment and Biotherapy; vice chairperson of Guangdong Medical Association Specialized Committee for Cell Therapy; deputy director-general of Guangdong Specialized Committee for Cell Therapy; and deputy director-general of Guangdong Elder Health Care Association; editorial board member and executive editors of several journals such as *Chinese Medical Journal* and *Chinese Journal of Oncology*. So far, Wu Peihong has published 200 academic papers in leading journals at home and abroad, including more than 60 SCI papers as the first author or correspondent author. He has been the editor-in-chief and chief reviewer of 11 monographs.

Co-editor — Shi Jiangguo

Shi Jiangguo is a professor, doctoral tutor and deputy director of the Institute for Cancer Research at Air Force Medical University (Fourth Military Medical University, Xi'an), as well as a chief physician of the Division of Oncology at Tangdu Hospital. He has long been engaged in the teaching, treatment and scientific research of cancer, cardiovascular diseases, mental health, and clinical pathology. He has published over 80 research papers; edited *Modern Rehabilitation of Tumor* and *Patients' Hundred Thousand Whys: Tumor Fascicule* and 53 episodes of *Professor Shi Talks About Health*, a health program on Xi'an TV. Moreover, Shi Jiangguo has received funding for tumor prevention projects from American Cancer Society and Susan G. Komen (originally known as The Susan G. Komen Breast Cancer Foundation). At present, he is an advisor to the Global Chinese Breast Cancer Organizations Alliance; vice chairman of Chinese Anti-Cancer Association Division of Cancer Rehabilitation; a member of Chinese Anti-Cancer Association Specialized Committee of Psychosocial Oncology; chairman of the Cancer Rehabilitation and Palliative Care Committee of the Shaanxi Anti-Cancer Association; chairman of Shaanxi Province Health Care Association Specialized Committee of Tumor Prevention; and president of Xi'an Cancer Rehabilitation Association. In addition to all the titles, Shi Jiangguo was recognized as a quality course teacher, and was given People's Liberation Army (PLA) General Logistics Department Award for Cultivating People of Ability.

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About the Book

This is a pathbreaking book with almost revolutionary exploration. Controlling cancer with hydrogen to prolong patient lifetime is a brand-new concept and technology. As the oldest and most abundant natural gas between heavenly bodies, hydrogen is applied to the benefit of human beings, reflecting correspondence between man and universe. As a physiological gas generated in the human body all the time, hydrogen offers a natural way to maintain health.

This is a perfect combination of theory and practice. From the creation of hydrogen molecular biology and hydrogen medicine, this book talk about how to maintain the health of hydrogen, and then introduce the scientific basis of hydrogen cancer control, and then lists experimental evidence and "real world" case survey. Also there are the objective observation and description of the reporter as the "third eye". All the descriptions are layered and ingenious, and in one go, reading the hand does not release the volume, for the disease and healthy the benefits are endless.

In particular, the book is full of real-world stories that offer close glimpse of real people with real feelings. The stories abound with scientific elements and represent witnesses to the benefits of superb technology and the power of life.

This book is highly praised by top masters in China's medical circle, such as academician Wu Mengchao, the father of hepatobiliary surgery; academician Wang Zhenyi, the pioneer of Tumor Therapy of Induced Differentiation; academician Tang Zhaoyou, the founder of small liver cancer research; and academician Zhong Nanshan, a famous respiratory disease specialist who is known to be "more like a soldier". All of them contributed their opinions from different perspectives regarding the theoretical basis and practicality and feasibility of hydrogen in cancer control. Their enthusiastic support to this emerging technology is deeply touching and indeed commendable.

This book is the result of the author's painstaking efforts. Chief editor Xu Kecheng is a distinguished oncologist in China. His deeds of saving patients' lives have spread far and wide at home and abroad, touching the heart of countless patients. This book is born thanks to his great dedication to the research and practice of hydrogen cancer control. He is a man who becomes more vigorous with age. Prof. Lu Youyong, Wu Peihong and Shi Jianguo co-edited the book; while Prof. Wang Xiaoning and Yin Zhinan contributed sincere and candid prefaces to the book. All of them are well-known scholars and clinical practice experts in the medical field in China. They have stepped out of tradition and embraced emerging disciplines with daring spirit, which reflects in them a bold vision to constantly develop broad thinking and keep up with the times. Prof. Sun Xuejun, who did main review of the book, is the earliest in China to study hydrogen molecular biology and hydrogen medicine. His full-hearted appreciation to and tremendous support for hydrogen cancer control later are highlights of the book.

Thanks to the author's trust. We are honored to have the privilege of publishing this book. We hereby highly recommend this book to readers.

by Yangcheng Evening News Publishing House, May 2019, in Guangzhou, China

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From the Editor

Why Did I Edit *Hydrogen Cancer Control: Theory and Practice*?

For what purpose are we living? Perhaps this question is of the greatest concern only to those who are facing death threats.

Exactly 13 years ago, one third of my liver was surgically removed because of cancer. After waking up from anesthesia, I insisted on having a look at the left lobe liver that was resected. I noticed a round yellow-white mass in the reddish-brown liver tissues. Judging based on decades of experience in liver research, I knew that it was a malignant cancerous tumor. Could I survive? According to literatures then, the probability of patients with such tumor surviving 5 years was lower than 5%.

At that time, I was over 60 years old when the happy life I wanted seemed to have just begun. Besides, I had not fulfilled my childhood commitment to my mother yet.

Nobel Prize winner Peter Kapitza once said: "The concept of life is established in kindergarten." My kindergarten happened to be on a battlefield filled with clouds of smoke in China in the 1940s. I will never forget that a good man from my hometown covered me from being shot by the enemy's bullets at that time. Since the time I could remember, my mother often told me: "It is the folks here who saved you and protected our home. After you grow up,

do good things for the people." Old enough to understand the words, I promised to my mother I would.

For decades, whenever I meet critically-ill patients, especially those in deep poverty, what my mother said echoes in my ears, and the good man who saved me comes to my mind.

After being diagnosed with cancer myself, I work up to the truth that doing good things as both a doctor and a patient means breathing the same air and sharing the same destiny with other fellow patients with cancer. I want to not only enjoy life myself, but also help them survive and live a colorful life.

Nobel laureate Bertrand Russell once said: "The trouble with the world is that the stupid are cocksure and the intelligent are full of doubt." WHO proposed to manage cancer as a controllable chronic disease, which requires efforts to question the current reality and explore the unknown. The treatment aimed at eradicating cancer is simply the first step to success. It is top priority to keep patients surviving for a long time. Post-treatment rehabilitation is a lifelong task for me and my cancer patients. In one word, cancer requires a lifetime of care.

I put forward the "ABC" (Applicable, Brief, Costless) principle of cancer rehabilitation.

Driven by the principle, I had been searching and exploring for years. Eventually, I found hydrogen medicine.

I was genuinely convinced by the potential effects of hydrogen medicine after reading a large number of basic and experimental research literatures. I was grateful for my personal experience of hydrogen inhalation. I was excited to visit each and every cancer patient who inhaled hydrogen at home. Therefore, I started working together with relevant experts and eventually completed this book.

As Nobel Prize winner Erwin Schrödinger once said, "The task is, not so much to see what no one has yet seen; but to think what nobody has yet thought, about that which everybody sees." The purposes of this book are to: introduce hydrogen, a natural gas, the Qi of the body and a common substance that everybody sees; present our observation, experience, investigation, description, thinking and research findings; and reveal how hydrogen can be used as a "non-medical medicine" to maintain health and control cancer.

For cancer patients, including me, hydrogen offers a way to enjoy health and happiness. As depicted by the famous poem, *Let life be beautiful like summer flowers and death like autumn leaves*.

In this book, hydrogen cancer control is positioned as a means of rehabilitation instead of treatment. Whether it can be a therapy requires specified tests and assessment according to regulated procedures. Currently, in the whole course of cancer treatment, hydrogen is not the main treatment but an assisting measure and plays a supplementary role. In the concept of general health, hydrogen is responsible for the overall care of the whole process of life.

This book will inevitably attract questioning. In fact, I have been questioning myself all the time. But I am aware that one of the basic attitudes of science is questioning, and its most fundamental spirit is criticism.

I am open to questioning and criticism.

Back to the original question: What is the purpose of living? I was inspired by a short poem on the Internet:

> Pick up the pen of hope
> Write eulogy for yesterday
> Draw an end for today
> Dye a color for tomorrow

That is what drove me to edit *Hydrogen Cancer Control: Theory and Practice* in the first place.

— Xu Kecheng, March 2019 in Guangzhou, China

Timeline of related works by the editor:

- *In 2010*, I proposed that doctors should tell the truth to cancer patients and do practical things for them during treatment. It is important that doctors and patients fully communicate with each other and understand each other's thoughts. (Xu Kecheng: *Truth to Cancer Patients*, Guangzhou Publishing House)
- *In 2013*, I proposed the treatment model "3C + P" (Cryoablation, Cancer Microvascular Intervention, Combined Immunotherapy for Cancer, Personalized Cancer Therapy) based on my own experience. The model is designed to integrate treatments and rehabilitation for patients to survive. (Xu Kecheng: *Fight Cancer with Me*, People's Medical Publishing House)
- *In 2016*, I proposed the concept "living with cancer", for conventional eradicative treatments never leads to the cure of cancer. Coexistence with cancer should be the main theme of cancer treatment. This is an ideal that can be achieved with hard work. (Xu Kecheng: *Living with Cancer*, Guangzhou Publishing House)
- *In 2018*, with the help of academician Tang Zhaoyou, I proposed to practice "Chinese Way of Cancer Control", which challenges the tradition and advocates an innovative path by integrating Chinese cultural elements with modern medicine. (Xu Kecheng: *Practice Chinese Way of Cancer Control*, Yangcheng Evening News Publishing House)

Words from the Main Observer

Hydrogen Cancer Control: I Am Fully Convinced

I am ninety and want nothing but to live a quiet life these days. Unexpectedly, Prof. Xu Kecheng asked me to write a preface for his forthcoming book *Hydrogen Cancer Control: Theory and Practice*. I gladly says yes.

I was very dubious about the use of hydrogen in cancer control when Prof. Xu showed me the first patient case. But when I was shown nearly 50 cases successively, I became convinced. In the cases, patients were diagnosed definitively with cancer. Eradicative means of treatment were exhausted but failed to effectively control their cancer. After several months' hydrogen inhalation, the patients still had tumors in their bodies, but resumed normal life like healthy people. Therefore, I believe that hydrogen has certain effect in cancer control.

At 2018 Chinese Conference on Oncology, I gave a speech about the "six integrations" in the fight against cancer. Now, it seems that the six integrations echo with the concept of hydrogen cancer control. The first is strategic integration of cancer elimination and transformation. Hydrogen inhalation does not directly kill cancer cells but transform them. The second is integration of individual operations and comprehensive treatments. Hydrogen inhalation alone hardly cures cancer but is helpful in the same way as exercise helps control cancer. However, once a person has cancer, exercise alone is never enough. Treatments to eliminate tumors

are required. The third integration is integration of fast operations and strategic long-term measures. Eradicative therapies (surgery, radiotherapy, chemotherapy and most molecular targeted therapies) deliver instant results. In contrast, it takes a few hours a day for a patient to inhale hydrogen, and several months to achieve the desired results. The fourth integration is integration of passive treatments and active participation. Cancer treatment is given to patients by doctors, while hydrogen inhalation is a process participated by patients. The latter requires confidence and perseverance. The fifth integration is integration of advanced and new therapies with fast and cost-saving measures. Some reports revealed that some molecular targeted therapies cost around 150,000 US dollars annually. Apparently, hydrogen inhalation is a fast and cost-saving method, which is absolutely necessary according to our current national conditions. The sixth integration is integration of western medicine and Chinese thinking. We need to learn from western approaches to cancer and use them in line with the national conditions in China. But to achieve transcendental results, it is necessary to apply Chinese thinking. According to Lao Zi, a great ancient Chinese thinker, the highest good is like that of water. Coincidentally, hydrogen comes from the electrolysis of water. Perhaps it has behind it the Chinese thinking "the soft overcoming the hard".

Indeed, any new things are more or less questioned. At the beginning, I questioned whether it was hydrogen or oxygen that achieved positive effects (two hydrogens and one oxygen are generated after water electrolysis). I questioned whether hydrogen inhalation was effective on all types of cancer. I wondered about the efficiency, mechanism, side effects, long-term effects and other aspects of hydrogen inhalation. After reading this book, you will find the answers to some questions. Yet, a lot is under research. And we will see how it turns out.

In fact, application of hydrogen to cancer treatment was published in the top journal *Science* in 1975. In 2007, *Nature Medicine* reported the antioxidant effects of hydrogen, but the study did not receive much attention. It is not uncommon that major discoveries pass us by as a result of our ignorance of accidental event. The reason might be related to the fact that people in general think that the right way to deal with cancer is to eliminate it. Strategically, fighting cancer requires renewed concepts. Cancer is an internal disorder of the body. Unlike infectious diseases that occur due to invasion of foreign enemies, cancer must be treated with not only eliminating measures but also transforming ones. This book came into being exactly as a result of the efforts to change cancer treatment approaches from cancer elimination to cancer transformation.

I strongly recommend this book to readers. I believe it will benefit cancer patients and inspire doctors.

— Tang Zhaoyou, March 2019 in Shanghai, China

> *Tang Zhaoyou: academician of the Chinese Academy of Engineering; professor; well-known oncologist; founder of small hepatocellular carcinoma research; winner of Ho Leung Ho Lee Foundation Prize for Scientific and Technological Progress in 1995; winner of Wu Jieping Medicine Prize in 2008; granted the title "Lifetime Honorary Professor" by Zhong Shan Hospital of Fudan University in 2018.*

Observations from Academician 1

Hydrogen Cancer Control: Revolutionary Exploration of Tumor Rehabilitation

On the eve of the Spring Festival in 2019, I received the first draft *Hydrogen Cancer Control: Theory and Practice* from Prof. Xu Kecheng. At first glance, I saw the title of the book: cancer control instead of cancer elimination. The difference lies in a single word. Control is a well-chosen word for the book. Having performed surgery in my whole career time, I've come to realize that tumors must not be only killed with scalpels but should be controlled to the level where cancer cells behave and change to good cells. As Prof. Xu wrote in his book *Living With Cancer*, it is of overriding importance to keep patients surviving and living happily. This might be achieved by the use of hydrogen.

As early as 1975, the American Journal *Science* published an article that revealed the function of high-pressure hydrogen in inhibiting skin squamous cell carcinoma. In 2007, *Nature Medicine* published a study, showing that inhaling low-concentration hydrogen improves cerebral ischemia-reperfusion injury and produces selective antioxidant effects. The Second Military Medical University where I work was the first institution in China to carry out the research in hydrogen molecular biology. Prof. Sun Xuejun, the University's researcher engaged in the study of diving diseases and hyperbaric medicine, first published an international paper about hydrogen medicine in 2008. In 2013, he edited and published the Chinese monograph *Hydrogen Molecular Biology*. The book was the first of its kind in China. In 2015, he published the English version of *Hydrogen Molecular Biomedicine*, which systematically introduced the main mechanism and research progress of hydrogen-based treatment of diseases. Many other scholars from our university have participated in hydrogen medicine research. Their efforts have contributed to promoting the establishment and development of hydrogen medicine in China.

Prof. Xu Kecheng was the first expert in China to study the application of hydrogen in cancer control from clinical perspectives. He and his team followed up on a large number of cancer patients in rehabilitation through hydrogen inhalation at home. These patients were mostly patients who had lost the chance of surgery, experienced relapse after surgery, or received chemotherapy and radiotherapy (including molecular targeted therapies) to no effect. However, after inhaling hydrogen (mainly hydrogen-oxygen mixture) for a long time, the patients demonstrated significant improvement (in terms of sleep, appetite and fatigue). In quite some cases, the patients' tumor markers dropped or became stable. In some other cases, the tumors shrank or stabilized. This is amazing and innovative exploration. It is gratifying to see the results of hydrogen inhalation in some patient cases.

Currently, the mechanism of action by which hydrogen inhibits cancer is not yet fully clear, except that hydrogen plays a role in modulating the microenvironment and inhibiting the growth, proliferation, movement and invasion of cancer cells. I am particularly interested in an article in this book. Titled *Modulating Cancer Cells: Intimate Contact between Hydrogen and Cancer Cells*, the article suggests that hydrogen may modify cancer cells in a way that makes cancer cells "break away from evil ways and return to the orthodox path". Another article by a Japanese scholar states that hydrogen restores cytotoxic T cells in burnout status by improving mitochondrial function. All these represent innovative discoveries worthy of further study.

Those who participated in the writing of this book are all experts and scholars from famous universities and hospitals. This shows that that the theme "hydrogen cancer control" has

attracted wide attention. China's research on hydrogen molecular biology and hydrogen medicine has made a good start and has been in the forefront of the world in many aspects.

It is important that we have firm confidence in our theories, the road ahead, system and culture; seize opportunities; and carry on earnest research and practice. In the context that hydrogen science, especially hydrogen energy, has become the topic of strategic projects for national development in China, I believe that hydrogen medicine is set to a good path of development.

I have been friends with Prof. Xu Kecheng for more than 40 years. I am now at the age of 90, while he is about to be 80. We share a common idea. That is, we do not seek to become superior to others but to be one of others. As doctors, we pursue no financial gains, but the joy and gratitude of patients. To explore the use of hydrogen in cancer control, Prof. Xu traveled thousands of miles and visited patients again and again to greet patients and collect real data. He did everything at his own expense. His spirit is truly commendable and worth learning by the younger generation. What he did touched me. I will definitely read this book to the end and learn from the real data in it.

Cancer is a systemic chronic disease that can be controlled. Surgery and radiotherapy are major means to locally eliminate tumors. As a systemic treatment, chemotherapy cannot be used for rehabilitation due to its toxicity and ultimate drug resistance. Considering its high safety, easy use, and roles in cancer cells and the body's systems, hydrogen can be useful for a long time or even lifetime. The discovery of hydrogen and its effects is a revolutionary exploration of tumor rehabilitation.

This book is the first one dedicated to hydrogen and cancer. Anything "first" is hardly perfect. I hope that the academic community will be supportive and tolerant; and help us correct defects and improve. This is also the wish from the chief editor of this book.

Congratulations on the publishing of this book. I hope to see progress on the research and development of hydrogen medicine driven by this book.

— Wu Mengchao, February 2019 in Shanghai, China

> *Wu Mengchao: academician of the Chinese Academy of Sciences; professor; distinguished expert in hepatic surgery; one of the founders of Chinese hepatobiliary surgery; winner of the National Science and Technology Progress Award in 2005; awarded with Asteroid Naming Certificate and the Asteroid Orbital Bronze Medal in 2011.*

Observations from Academician 2

Hydrogen Cancer Control: A Revolutionary Topic of Cancer Rehabilitation

I often say that cancer treatment should neither be complicated nor painful. I was very pleased to read the first draft of this book. Maybe my expectations will eventually become a reality, at least for some tumors or at certain stages of cancer treatment.

As early as more than 40 years ago, an American scholar reported that hydrogen, a natural gas, had effects on inhibiting cancer cells. Despite being published in the famous journal *Science*, the report did not attract much attention. This was probably because that hydrogen is too simple and too ordinary. It seems that ordinary things are not presentable enough. In the 1990s, a middle school teacher in Shandong Province of China suggested that hydrogen

helps control oxidation and inflammation; and predicted that it can be used to inhibit cancer. His views and research findings were published in a university journal in 1999. However, perhaps because the author was unknown to the public, what he published was forgotten soon and was not recognized by any important figure. This is truly a pity.

Today, we have Prof. Xu Kecheng's *Hydrogen Cancer Control: Theory and Practice*, in which he recorded cases about cancer patients with hydrogen inhalation from his and his team's follow-up visits. Most of the cancer patients had lost the chance of conventional treatment originally, but had improved their conditions and survived because of hydrogen inhalation, a seemingly simple treatment. Some may say that this is not evidence-based medical research. But patients have survived, and some even have survived and lived a very high quality of life in spite of cancer cells in their bodies. We cannot afford to turn a blind eye to the real-world cases in this book.

I once wrote a preface to Prof. Xu's book *Live with Cancer*. At that time, I said that I could and should write a preface, because Prof. Xu always tries to crack the most challenging cases from the perspectives of ethics and universe. I still remember that more than a decade ago, I met Prof. Xu after not seeing him for over 20 years. When I learned that he underwent surgery for liver cancer a few years before that, I said that cancer treatment must not be targeted merely at the tumor, but should be focused on systemic control with innovative approaches. Prof. Xu is very wise. He started studying the strategies and methods of cancer rehabilitation; and proposed that the top priority of cancer treatment is to keep patients surviving. According to Prof. Xu, what he did is not only for patients, but also for himself. So far, he has survived cancer, and has successfully treated many patients with most challenging tumors.

The greatest truths are the simplest. I appreciate the ABC (Applicable, Brief, Cost-saving) principle proposed by Prof. Xu. Hydrogen cancer control is in exact conformity with the principle.

The topic of this book is new, but is an innovative one worth exploring.

I am happy to recommend this book to readers. I also hope that medical professionals will be inspired by his book to further study hydrogen medicine.

— Wang Zhenyi, Febrary 2019 in Shanghai, China

> *Wang Zhenyi: academician of the Chinese Academy of Engineering; professor; academician of the French Academy of Sciences; expert in internal medicine hematology; one of the pioneers in establishing disciplines of thrombosis and hemostasis at university in China; known as the "father of induced differentiation of cancer"; winner of the National Science and Technology Progress Award in 2010. The Minor Planet Center named No. 432 59 asteroid "Wang Zhenyi Asteroid" permanently.*

Observations from Academician 3

Laudable Exploration of the Real World

Hydrogen molecular medicine (or hydrogen medicine) is an emerging discipline. Hydrogen molecules are widely recognized for their strong antioxidant activity, especially their high selectivity (only neutralizing toxic active oxygen species), high diffusion and great safety.

Antioxidation has been a hot topic for many years, resulting in the birth of many antioxidant drugs and health care products. But such drugs and products have failed to deliver desired effects in general. The reason is that there are two types of free radicals in the body: physiological and toxic ones. Physiological free radicals play a role in the body's metabolism; while toxic ones cause oxidative stress, tissue damage and eventually many diseases.

The occurence of common thorax, lung and heart diseases is often associated with oxidative stress. We did a hydrogen inhalation experiment using mouse models of smoking-induced COPD and found that hydrogen molecules relieved lung function deterioration, pulmonary emphysema, inflammatory cell infiltration, small airway reconstruction and tracheal epithelial goblet cell proliferation. They also down-regulated the activation of inflammation-related factor ERK1/2 and NK-κB. Based on the findings, we carried out clinical observations on chronic airway diseases and found that hydrogen inhalation had positive effects on the acute attack of chronic airway diseases, and alleviated dyspnea and shortness of breath in a short time. At present, there is an ongoing randomized, double-blind and multicenter study of hydrogen inhalation in chronic obstructive pulmonary diseases, including acute exacerbation, bronchial asthma and bronchiectasis.

In fact, it has been around decades since gas was applied to treating respiratory diseases. At the being, the gas used was a mixture of helium and oxygen. As an inert gas with relatively small molecular weight, helium can smoothly carry oxygen to the deep part of the airway. Hydrogen is a smaller molecule that eases airway resistance more effectively than helium. More importantly, the antioxidant effects of hydrogen make it a medicine for diseases.

The formation and development of tumors are associated with chronic oxidative stress and inflammation. Oxidative stress causes inflammation. And chronic inflammation promotes the production of reactive oxygen species and intensifies oxidative stress. Both oxidative stress and inflammation cause nuclear DNA damage and gene mutations, or damage mitochondria, resulting in abnormal energy metabolism. These abnormalities tend to accumulate, promoting the occurrence and progression of tumors. Therefore, the application of hydrogen to cancer control has its theoretical foundation. So far, quite some experiments have shown that hydrogen has inhibitory effects on the growth, movement and invasion of cancer cells.

This book presents the follow-up results of nearly 100 cases of cancer patients who inhaled hydrogen (hydrogen 66%, oxygen 34%) at home, and stories about many individuals in whom positive results were achieved from inhalation of hydrogen-oxygen mixture. Most of them were patients who lost the opportunities of conventional treatment. However, after a period of inhalation, some patients showed significant improvement, and some others had gradually stable conditions. This is the first exploration of the real world in this field and a laudable attempt.

Cancer treatment is a global challenge. Regarding hydrogen cancer control, I look forward to conducting more experimental research and translating it into clinical research as soon as possible. Hydrogen inhalation can be used for at least the following:

First, hydrogen inhalation may serve as an adjuvant treatment for cancer. As the book says, it is complementary and reinforcing to mainstream treatments. For example, in the case of lung cancer, can hydrogen inhalation slow down resistance to targeted drugs and reduce the adverse effects of chemotherapy? Studies indicate that hydrogen reduces the side effects of chemotherapy without affecting its therapeutic effect.

Second, as an alternative treatment for cancer, hydrogen inhalation is mainly applied to patients for whom treatment has failed or conventional treatment is no longer suitable. This is the case for some patients with successful treatment from hydrogen inhalation in this book. It is necessary to focus on several types of cancers and conduct real-world evidence (RWE) research or randomized controlled trials in terms of hydrogen dose, administration method, and optimal time or duration of inhalation.

Third, hydrogen inhalation can be used for symptomatic treatment. Sleep, appetite, fatigue and pain are not unique to cases of cancer, but represent the major problems that beset cancer patients. With positive effects on all systems of the body, hydrogen offers a good way to improve systemic conditions.

Fourth, hydrogen inhalation can be used to treat the "disease to come". Such cases include cancer prevention that starts treatment ahead of time, especially for high-risk groups such as those with family history of cancer, long-term smoking, long hours of work in polluted environment, or intentions to prevent cancer recurrence after effective treatment (level-3 prevention). The concept "home rehabilitation" proposed in this book accords with the connotation of general health.

With the above, I hope to initiate a discussion that leads to valuable contributions by my fellows in the same pursuit. Congratulations on the publishing of this book.

— Zhong Nanshan, February 2019 in Guangzhou, China

> *Zhong Nanshan: academician of the Chinese Academy of Engineering; professor; famous expert in respiratory diseases; honorary doctor of the University of Edinburgh, UK; director of the State Key Laboratory of Respiratory Diseases; winner of Ho Leung Ho Lee Foundation Prize for Scientific and Technological Progress in 2003; awarded as one of the "Top 100 Teaching China Since the Founding of New China" in 2009; awarded as "Pioneer of Reform" by the Party Central Committee and the State Council.*

Words from the Chief Reviewer

We Cannot Live Without Oxygen and Cannot Live Well without Hydrogen

I am very happy and excited that Prof. Xu Kecheng's *Hydrogen Cancer Control – Theory and Practice* is about to be published. This is a milestone in the field of hydrogen biomedicine.

In 2007, I started systematic research on hydrogen medicine at the Second Military Medical University, which was the first time in China. My research focused on cellular biological effects of hydrogen, studies of animal disease models, individual cases and human clinical trials, challenging the traditional concepts of medicine. I made continuous efforts to study and promote hydrogen medicine, but with little success. As a well-respected clinical medical expert, Prof. Xu, will sound the trumpets for the clinical application of hydrogen medicine and open a new landscape for clinical medicine by publishing this new book. This is indeed exciting. Prof. Xu asked me to be the chief reviewer of this book, for which I am very grateful and willing to contribute. This will be a new driving force for me to work harder on the research and popularization of hydrogen medicine.

There have long been extensive studies of diving medicine that confirmed the ultimate safety of hydrogen to human body. But the studies neglected the potential therapeutic effects of

hydrogen on diseases. As a safe, common and comprehensively-studied gas with possible curative effects, hydrogen has great application potential and research value. This soon became a consensus, which is reflected by the fact that hydrogen biomedicine quickly received widespread attention from international medical professionals. It has been discovered that hydrogen not only has selective antioxidant effects, but also relieves inflammation and affects apoptosis. Having made bold attempts and conducted studies of various diseases and conditions, many scholars found that hydrogen delivers unique effects on the prevention and control of metabolic chronic diseases such as arteriosclerosis and refractory skin diseases such as psoriasis-specific dermatitis. Compared with other areas, applying hydrogen to studies of cancer prevention and treatment is rare but very groundbreaking. Existing evidence suggests that hydrogen can prevent the toxic and side effects of radiotherapy and chemotherapy without affecting their therapeutic effects; and can significantly improve cancer patients' quality of life.

Cancer treatment is in a difficult predicament in the current medical world. In spite of typical therapies such as surgery, radiotherapy and chemotherapy, doctors find themselves quite helpless in front of most patients with advanced cancer. For years, Prof. Xu has been advocating and practicing the Chinese way of cancer control, which has offered Chinese wisdom to the international medical dilemma and witnessed fruitful results. Recently, Prof. Xu collected and sorted out the clinical data of many patients who tried to use hydrogen for cancer treatment and demonstrated positive outcomes. Better still, Prof. Xu visited the patients and verified their treatment results. Based on his years of clinical experience and objective analysis with evidence-based medicine logic, Prof. Xu pointed out that hydrogen Inhalation has positive effects on advanced cancer, features great safety; and brings certain value for improving patients' survival and quality of life. Above all, academician Tang Zhaoyou, a distinguished oncologist, commented on each and every patient case followed up on by Prof. Xu. Both academician Tang and Prof. Xu explicitly proposed hydrogen therapy as one of the feasible means for Chinese way of cancer control. The therapy promotes basic and clinical studies of hydrogen use in main and adjuvant treatments for cancer patients. This book systematically displays these research findings. Prof. Xu's pioneering work has not only brought new hope to many cancer patients, but also moved hydrogen medicine forward.

Of the more than 1,000 international academic papers on hydrogen medicine, only 50 are about the clinical studies of hydrogen medicine. This indicates that many bottlenecks and dilemmas remain in the clinical application of hydrogen medicine. Prof. Xu's new book adds more clinical academic evidence to hydrogen medicine and provides abundant clues for basic research in hydrogen medicine. This will definitely strengthen enthusiasm for hydrogen medicine research. Moreover, this academic and highly readable book provides first-hand objective data for accurate understanding of hydrogen's medical value. As an ordinary scholar in hydrogen medicine research, I want to thank Prof. Xu and admire him for his important academic contributions to hydrogen medicine.

"We cannot live without oxygen and cannot live well without hydrogen." This is what I said casually in an interview with CCTV in 2016. It represents a characteristic of hydrogen medicine. Hydrogen has biomedical effects but far lower status in the medical world. Once again, with the title for this section, I hope to send the following message to the hydrogen health industry and general public: Hydrogen medicine is miraculous but is not a panacea. To fully utilize the safe and effective value of hydrogen, we must always seek truth and refrain

from exaggerating or misleading others. Only the truth will eventually stand the test of time and keep its vitality.

— Sun Xuejun, March 1, 2019 in Shanghai, China

> *Sun Xuejun: professor and doctoral tutor of the Second Military Medical University; one of the pioneers of hydrogen medicine research in China; published over 260 academic papers (including 148 Science Citation Index papers); edited the Chinese and English versions of Hydrogen Molecular Biology; chairman of the Hydrogen Molecular Biomedicine Division of China Science and Technology Industry Association; part-time professor of Shanghai Jiaotong University and Xi'an Jiaotong University; and associate editor of Medical Gas Research; first researcher in the world to study the treatment of various diseases with hydrogen normal saline.*

Preface 1

Hydrogen Cancer Control: A New Health Civilization

In 2018, the Chinese Union of Life Science Societies appointed me as the executive secretary general and entrusted me with the tasks of coordinating the organization of the second World Life Science Conference and searching for collaborative projects for the Union's development. During this period, Prof. Lv Youyong from Peking University Cancer Research Institute told me that I could cooperate with a hydrogen medicine institution. Both Youyong and I are senior consultants in population health under the national 973 Program. Considering that Youyong is a rigorous and serious famous expert in the Chinese medical circle, the idea "Oh, health care product again" flashed in my mind the moment he started introducing hydrogen medicine to me. But then, I listened very carefully as he briefly told me about the progress of hydrogen medicine.

Ten days later, I met Lin Xinyong and Prof. Xu Kecheng, the inventors of a medical hydrogen machine, in Beijing. Prof. Xu looked very familiar to me. After short greetings, Prof. Xu took out his computer and gave us a clear introduction about the development of hydrogen medicine and its current mechanism, safety and application scope. I was deeply impressed by the typical cases of terminal cancer patients who inhaled hydrogen at home and in Xu Kecheng Care for Health Studio. The patients came from Taipei, Shanghai, Beijing and other parts of the country; and were visited by Prof. Xu and his team as a part of follow-up survey. I asked Mr. Lin about the research and development status of the medical hydrogen machine, only to realize that hydrogen has been used by human beings as a diving pressor for more than 100 years and is very safe to use. Scholars in the 1970s reported that hydrogen had inhibitory effects on cancer. In 2007, a Japanese scholar published a paper in *Nature Medicine*, expounding on the role of hydrogen intervention in cerebral apoplexy. Only since then, the inhibitory effects of hydrogen on cancer have been receiving extensive attention from academic and medical communities. Previously used hydrogen-rich water may have dosage ceiling due to limited solubility. However, the problem was resolved by Mr. Lin's Asclepius Meditec hydrogen-oxygen nebulizer. The machine has been initially validated in a clinical double-blind experiment on interventional chronic obstructive pulmonary disease (COPD), and has been qualified as a product of national innovation. Currently, the machine is undergoing pre-market approval by China Food and Drug Administration (CFDA) as a Class III medical device. Once approved, the machine may become the world's first medical hydrogen machine,

therefore providing a foundation for further exploration of hydrogen clinical application. So far, there have been over 1,000 published papers from at home and abroad, exploring the medical and health effects as well as potential mechanisms of hydrogen; and revealing multiple biological effects of hydrogen.

After returning to Guangzhou, Prof. Xu personally took me on a visit to his studio. On that day, the studio was full of cancer patients who came to inhale hydrogen for free. Prof. Xu, in high spirit, asked each patient present about their basic conditions. I stood in awe when learning that several of the patients were there for hydrogen inhalation simply because they believed Tang Zhaoyou and other senior experts' positive evaluation of hydrogen medicine. It seemed that they are determined to keep inhaling hydrogen until their tumors retreat. Prof. Xu told me that hydrogen inhalation is only an adjuvant treatment, and that his studio does not accept patients who give up standard medical treatment. The studio is not doing marketing promotion for anything.

Prof. Xu said that he and Prof. Lv Youyong, using Real World Evidence (RWE) design, are currently collecting extensive data about the effects of hydrogen inhalation on cancer patients, especially on patients with terminal cancer, in order to further explore the role of hydrogen in tumor adjuvant treatment.

In the flight back to Beijing, I finished reading Prof. Xu's book *Live with Cancer*, from which I came to know that Prof. Xu himself is a survivor of cholangiocarcinoma for 13 years and is nearly 80 years old. It does not take long for readers to see Prof. Xu's dedication to medicine, profound knowledge, and great care for patients. Moreover, the book includes abundant health and disease management concepts, which have been implemented long time ago but are promoted only recently. The concepts are highly forward-looking. It's no wonder that Prof. Xu still has the intellectual vigor in him that is not seen in many young people nowadays. Later, I recommended this book to the cancer patients I knew.

Prof. Xu sent me the manuscript of his new work *Hydrogen Cancer Control: Theory and Practice* and asked me to write a preface. I felt slight pressure seeing that observants of this book are all top medical experts in China. By this preface, I want to share my expectations for the future.

What exactly is the mechanism behind the health and medical effects of hydrogen? To answer the question, a fine device in vitro and in vivo is required to verify the biological effects of hydrogen. Also, there must be well-defined hydrogen markers that indicate where hydrogen is and what it has become. Fortunately, Prof. Wei Youzhen, a business partner of Shanghai Asclepius Meditec and member of Shanghai East Hospital Experiment Center, has done a lot of substantive work to find the answer. He developed cells and animal hydrogen chambers for experiments in vivo and vitro; and separated pure tritium for tracing. Prof. Gu Ning, a nano expert from Central South University, is willing to make hydrogen nanobubbles to explore the effects of directed release in the body. Academician Cheng Heping from Peking University is also willing to study the influence of hydrogen on mitochondrial flash by using more sophisticated devices. It is believed that more Chinese biologists will join the research on the physiological and medical effects of hydrogen. When the time comes, the mechanisms of action demonstrated in patient cases in Prof. Xu's book will be understood gradually. Perhaps in the near future, China will usher into an era of real hydrogen medicine, leading to the emergence of a tremendous health industry for the benefit of patients and mankind as a whole.

Civilizations are born along with water. Water is dissociated into oxygen and hydrogen; whereas hydrogen and oxygen are restored to water in the human body. Hydrogen health means the health of water. In this sense, hydrogen cancer control can be regarded as a new civilization of health.

— Wang Xiaoning, February 2019 in Beijing, China

Preface 2

Studying Hydrogen Cancer Control: A Noble Cause

Prof. Xu Kecheng is a teacher I highly respect. As early as I was still in the U.S., I heard about his deeds and academic achievements. I met him after returning to Guangzhou, and we felt like old friends. At that time, I was studying immunology and needed clinical support and cooperation. As a clinical doctoral tutor at Jinan University, Prof. Xu became the best partner for me. Some time ago, the paper that we co-authored about the treatment of liver cancer with gdT cells was published in an influential professional journal abroad. The paper was the result of a project that I desired when I was working at Yale University and have been working hard on for nearly 10 years. It is also the first of its kind in the world. The groundbreaking study cannot be completed without Prof. Xu's support and collaboration. Every time I presented the research findings at academic conferences at home and abroad, I felt grateful to Prof. Xu from the bottom of my heart.

In recent years, Prof. Xu has been studying tumor rehabilitation, with a focus on hydrogen use for cancer control. Since my work was not related to hydrogen molecules, and hydrogen was a topic rarely touched by others, I did not pay much attention to it. Yet, I trust Prof. Xu and therefore have firm belief in what he does. Once, I visited Prof. Xu's studio and happened to meet a few experiencers of hydrogen inhalation. Glowing with smile on their faces, the experiencers said that they felt completely refreshed and had better appetite, great energy and improved sleep after hydrogen inhalation. With further communication, I learned that most of them were cancer patients not eligible for other treatment methods. After a few weeks to months after hydrogen inhalation, some of the patients had tumor marker decrease and even tumor shrinking. The brain metastasis disappeared in one patient with breast cancer. With rigorous attitude towards his work, Prof. Xu kept records of all the experiencers of hydrogen inhalation. The records are open to visitors.

The results of hydrogen inhalation require further research and verification. But the real feelings of the patients I saw drove me to think about the deeper mechanism behind. I reckon that patients' improvement after hydrogen inhalation is related to energy, which involves mitochondria. Hydrogen, as a selective antioxidant, can improve mitochondrial function. Existing evidence suggests that mitochondrial function is associated with maintaining good immune status, while mitochondrial failure causes immune failure.

Recent research findings indicate that many cancers are closely related to immune function decline, whereas other cancers are more likely to be the results of immune system decline and multiple genetic mutations. This means that the key to preventing cancer may lie in the immune system instead of genetic mutation. Chen Lieping, a scientist of Chinese origin, pointed out that tumor immunotherapy has undergone a paradigm shift from immune

enhancement that systematically improves immune response to immune normalization that selectively corrects immunodeficiency in tumor microenvironment (immune normalization).

A Japanese scholar's research verified my speculation. The study disclosed that PD-1 positive anti-CD8 T cells in cancer patients were in a state of depletion. After 3 months of hydrogen inhalation, these T cells changed to PD-1 negative, that is, hydrogen saved the T cells. In other words, it was hydrogen that achieved immune normalization.

I was excited to know the research findings. Hydrogen seems to have opened a skylight for my lifelong research on immunology.

Science, especially natural science, serves to constantly seek and understand the objective world; explore new phenomena and new rules; and relentlessly pursue the truth. Hydrogen science is a very young discipline. In spite of his advanced age, Prof. Xu, started from scratch and delved into the real world of hydrogen cancer control. His spirit and lofty faith are indeed admirable and worth learning.

Prof. Lu Youyong and Wang Xiaoning, both of whom are famous experts in biological immunity, became fans of Prof. Xu long time ago. In the study of hydrogen cancer control, Prof. Xu has strong support from master medical professionals like academicians Wu Mengchao, Wang Zhenyi, Tang Zhaoyou and Zhong Nanshan. In contrast, I feel ashamed of my long-time hesitation to move forward. As an ancient Chinese poem said, "A time will come to ride the wind and cleave the waves, I'll set my cloudlike sail to cross the sea which raves." Things in the past are beyond redemption, but I can pursue what lies in the future. Not long ago, Prof. Xu was hired as the honorary dean of the Biomedical Translational Research Institute of Jinan University. I believe this will be the beginning of our new higher level of collaboration.

After going through the first draft of this book, I could not help but admire the contributions of Prof. Xu and other experts like him. I learned a lot from the reading. I want to congratulate Prof. Xu on the publishing of this book. I hope that readers will benefit from the book like I did.

— Yin Zhinan, March 2019 in Guangzhou, China

> *Yin Zhinan: graduated from Hebei Medical University in 1984; obtained a master's degree in immunology from Shanghai Second Medical University in 1988; went to the National Cancer Center of Italy for further study at the end of 1992; transferred to German Center for Rheumatology led by immunologist Av Mitchson in 1994 and received Ph.D. three years later; engaged in doctoral research at the Division of Rheumatology, Department of Internal Medicine, Yale University School of Medicine; promoted to be associate professor and won the Research Achievement Award of the Department of Internal Medicine of Yale University in 2006; hired as the Dean of Nankai University School of Life Sciences in March 2007; recognized as a distinguished professor under The Yangtze River Scholar program, national "High-level Talent Returning from Studying Overseas", and "Young and Middle-aged Expert with Outstanding Contributions" of the New Century Talents Project; invited to work at Jinan University to establish the Biomedical Translational Research Institute as the director of the Institute; published 118 papers in top internal journals such as Nature, Hepatology and JEM.*

Introduction

Soul of This Book: Truth

Hydrogen can be used to treat diseases? "That's right." said Zhong Nanshan, an academician of the Chinese Academy of Engineering.

It is said that there have been more than 1,000 literatures on hydrogen biology and hydrogen medicine. We have read hundreds of them. Some Japanese scholars stated that hydrogen is effective against 67 diseases or pathological conditions. One American scholar said that the number is 170. Another American expert believed that hydrogen plays certain role in the top ten causes of death, except suicide and accidents, in the United States. The findings are based primarily on animal experiments and a small number of clinical observations.

As to whether hydrogen controls cancer, only one paper published in the American Journal *Science* by Dole M. in 1975 clearly concluded that hydrogen helps treat cancer.

We were shocked. Is that true?

As a clinical doctor and cancer researcher, I think the most important thing is to see the patient in person; understand the patient's entire medical history and feelings; and obtain various clinical evidence. We established a survey team and started visiting patients with hydrogen inhalation. Eventually, we travelled to more than 20 cities at home and abroad on a follow-up visit to cancer patients who inhaled hydrogen at home in the past few years. In addition, we conducted prospective observations of hundreds of patients who had voluntary hydrogen inhalation in Xu Kecheng Care for Health Studio. It was particularly exciting to see the significant signs of rehabilitation and even miraculous results in some progressive cancer patients, for whom conventional treatment had failed or could hardly be effective, after hydrogen inhalation.

We discovered that hydrogen controls cancer. This is real.

We are touched by patients' joy from the effect of hydrogen inhalation; are grateful to patients for their courage to try hydrogen inhalation; and feel indebted to patients for their selfless sharing with us. With that in mind, we, together with media professionals as "third eyes", wrote one after another story from the real world. That has constituted the basic content of this book.

We are thankful to the patients who accepted our interviews. They are the best teachers. Full of gratitude, the patients told us about their treatment courses and hydrogen inhalation without reservation. They shared lots of details and data with us. Their stories were exciting to doctors and gave them more confidence.

Here, we would like to thank the masters for their guidance and encouragement.

Our thanks go to academician Tang Zhaoyou, who advocates the Chinese way of cancer control that values combined use of cancer elimination and transformation therapies. He regards cancer transformation as an important strategy for patients' long-term survival. He proposed that hydrogen can serve as a means of transforming cancer and stressed the importance of practice in testing truth. According to academician Tang, in today's world full of high-grade, refined and advanced technologies, we cannot let seemingly accidental events like hydrogen inhalation pass by. He once said with excitement: "I was dubious about the effect of hydrogen in cancer control when I saw the first case. But after seeing nearly 50 cases, I became fully convinced."

We want to also thank academician Wu Mengchao, a famous liver surgery expert and winner of the country's Highest Science and Technology Award. Academician Wu believes it is necessary to abandon the idea that surgery is omnipotent and to develop simple but effective rehabilitation means for patients' long-term survival. Hydrogen might be such an effective means. He considers hydrogen cancer control as a revolutionary exploration of tumor rehabilitation.

We are grateful to academician Wang Zhenyi, another winner of the national *Highest Science and Technology Award* and founder of tumor induced differentiation therapy. Academician Wang has long advocated simplified and painless cancer treatments. He said: "I was very pleased to read the first draft of this book. Maybe my expectations will eventually become a reality, at least for some tumors or at certain stages of cancer treatment." He refers to hydrogen cancer control as an innovative topic of tumor rehabilitation.

We would like to thank academician Zhong Nanshan, a distinguished expert in respiratory diseases. As the first expert to conduct clinical studies of hydrogen medicine in China, academician Zhong gave us enlightenment in the study of hydrogen cancer control. He believes that this field is the first real world exploration and is worthy of attempts.

We appreciate the efforts of experts who were the first to introduce hydrogen medicine to China and conducted basic research with outstanding achievements. Prof. Sun Xuejun is the chief reviewer of this book. Prof. Qin Shucun and Ma Xuemei contributed articles to this book. They regard the publishing of this book as a milestone event, which is overpraise yet enormous encouragement for us to continue clinical studies of hydrogen.

Last but not least, we want to thank the experts who are engaged in research on related disciplines. Prof. Wang Xiaoning, a well-known expert in geriatrics-rehabilitation and immunology; and Prof. Ying Zhinan, an immunologist, both contributed prefaces to this book and showed strong interest in further research on hydrogen biology and hydrogen medicine.

This book does not present hydrogen inhalation (or other hydrogen intake methods) as a therapy. We assume that if conventional surgery, chemotherapy (including molecular targeted therapy) and radiotherapy are considered as major cancer treatments, hydrogen inhalation is a means that reinforces the effects of major treatments. Under the current circumstances, hydrogen is not likely to be a dominant treatment, but can play a complementary role in treatment. In order for patients to survive, the first treatment is crucial, and proper rehabilitation is the key. We do not wish to see patients abandoning major treatments after reading this book. In view of its high safety and ease of use, hydrogen is listed as a means of post-treatment recovery, home rehabilitation, or health care. This seems to be more appropriate assessment of hydrogen, a natural and physiological gas, under the concept of general health.

Research on hydrogen cancer control has just started. There are too many uncertainties. Science itself is about constant questioning, denial, starting over, and then discovering true meaning. Your questioning is always welcome to me. Considering that medicine is a combination of science, technology and art, we prefer real-world research to obtain real data and collect real evidence.

In any case, hydrogen helps control cancer. This is a fact. The soul of this book lies in facts.

This book is simply an introduction intended to solicit valuable opinions. The content and viewpoints in the book are not perfect. With this book, we hope to offer a starting point and attract more researchers to the emerging field of tumor hydrogen medicine. We'd like to

receive candid feedback from readers, especially from researchers of hydrogen molecular biology and hydrogen medicine.

— Editor: Xu Kecheng; Co-editors: Lv Youyong, Wu Peihong, Shi Jianguo. May 2019

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PART 1 — History and Overview of Hydrogen Medicine

Research History of Hydrogen Biology and Its Medical Significance

Key points

- Hydrogen (or hydrogen molecule) is receiving increasing attention as a new medical gas, because hydrogen is a selective scavenger of toxic reactive oxygen species. Toxic reactive oxygen species are associated with the occurrence and progression of many pathological conditions, including cancer.
- In 1975, Dole M. and other scholars reported the role of hydrogen in cancer treatment in a paper published in the journal *Science*. In 2007, Shigeo Ohta from Japan published a paper in *Nature Medicine*, revealing that hydrogen molecules can improve cerebral infarction and have selective antioxidant effects.
- In recent years, there have been a large number of studies in the connections of hydrogen with human in-vivo environment, cytology, genetics, cell signaling, disease models, human diseases, treatment-related pathology, and pathophysiology. As a result, hydrogen molecular biology and hydrogen medicine have been established as disciplines.
- Du Yuanwei, a teacher from Shandong Province, China, presented in 1996 that hydrogen is a strong reducing agent, which can eliminate oxides naturally without side effects and achieve redox balance. His view happens to coincide with the current understanding of hydrogen molecular biology.
- Based on the observed and reported significance of hydrogen biology and hydrogen medicine, many medical institutions and technology companies in China have carried out research on the clinical value of hydrogen.

The Big Bang 4.6 billion years ago resulted in the formation of a vast and constantly moving galaxy, with diverse physical forms. In the entire universe, hydrogen is the most abundant element in terms of atomic percentage. The number of hydrogen atoms is around 100 times greater than the sum of all other element atoms.

Among all the elements that make up the human body, hydrogen accounts for only 9.5%, but provides vital energy to life. It is an indispensable substance for maintaining life and health.

Since the discovery of nitrogen, hydrogen, and oxygen as well as understanding of life and its activities from 1766 to 1800, human beings have created numerous miracles in many fields, including life science and medicine. However, human society is still in very young in the universe. And we are faced with too many unknown things, including hydrogen and its relationships with life and health.

Discovery and Existence of Hydrogen

Hydrogen was first discovered and described by Swiss chemist Philippus Aureolus Paracelsus as a flammable gas in 1520. In 1671, Robert Boyle, a famous Irish philosopher, chemist, physicist and inventor, described the nature of hydrogen. He was the first in the world to collect hydrogen using a mercury tank and conduct systematic experimental observation. In 1766, Boyle presented to the Royal Society a research report titled *On Artificial Air*, which focused on the characteristics of hydrogen. Referring to hydrogen as a flammable gas, Boyle found that the gas was 11 times lighter than normal air and insoluble in water or alkaline solutions. The more important finding was that hydrogen, once mixed with oxygen, generated water under the action of electric current.

In 1787, famous French chemist Lawrence Lavoisier verified that hydrogen is an element and named the gas "hydrogen", meaning "water element". Lavoisier pointed out that water is not an element, but a compound of hydrogen and oxygen. To this day, hydrogen is known as water element in Japanese.

On the earth, there is a very small amount of free-state hydrogen in the atmosphere. In the air on the surface of the earth, hydrogen accounts for around five ten-millionth (0.5 ppm) of the total volume. In the high-altitude atmosphere 20 to 25 kilometers from the ground, there are only helium and hydrogen. The soil contains around 1.5% hydrogen, which exists also in oil, natural gas, animals and plants. It can be seen that hydrogen plays a non-negligible role in life activities and health.

Proposal and Evaluation of Hydrogen Medicine

According to its characteristics, hydrogen was initially used as a carrier for airbags, and then was widely applied to spacecrafts. At present, hydrogen is being developed and utilized as a new energy with low pollution. Nowadays, in addition to its application value in the energy field, hydrogen is attracting more and more attention as a new medical-use gas. It is universally recognized as a selective scavenger of reactive oxygen species (ROS), especially hydroxyl (·OH). And hydroxyl is associated with the occurrence and progression of many pathological conditions, including cancer.

As early as 1937, hydrogen was first tested as a diving gas. The test results showed that hydrogen is the best breathing medium for moderate and deep diving; and is extremely safe for organisms without any side effects.

In 1969, Levit published a paper in *The New England Journal of Medicine*. The paper presented the results of hydrogen production by intestinal flora, showing that the colon produced 0.06 to 29 mL of hydrogen per minute in an average person in fasting state (0.24 mL on average). Based on the finding, it is estimated that the human body generates up to about 345 mL of hydrogen per day under fasting conditions, and hydrogen production increases by 7 to 30 times after eating.

In a 1975 paper in the journal *Science*, Dole M. et al. from Baylor College of Medicine and A&M reported the effects of hydrogen on cancer. In nude mouse models of UV-induced squamous cell carcinoma of the skin, the tumors shrank and retracted considerably in the chambers at eight atmospheres that contained 2.5% oxygen and 97.5% hydrogen.

In 1978, Roberts et al. reported that high-pressure hydrogen had inhibitory effects on leukemia cells. In 1988, Rostain and his fellow researchers in the U.S. discovered that Hydreliox, which contained 49% hydrogen, 50% helium and 1% oxygen, effectively protected divers from decompression sickness and nitrogen anesthesia when they were working 500 meters below sea level. In 2001, French researcher Gharib et al. reported that high-pressure hydrogen had anti-inflammatory effects on schistosomiasis-related chronic hepatitis in mouse models; and speculated that high-pressure hydrogen may have therapeutic effects on certain diseases.

Following a few studies of high-pressure hydrogen, researchers studied the functions of electrolytic alkaline water. American researcher Shirahata et al. reported that a hydrogen atom named active hydrogen can produce active hydrogen during electrolysis to scavenge ROS. Although hydrogen atoms do not exist in the human body for a long time, hydrogen does exist in electrolyzed water. According to a study by Li and his colleagues, electrolyzed water scavenges ROS and protects hamsters' pancreatic β cells from alloxan-induced cell damage.

A hemodialysis solution produced by the company Nihon Trim helped reduce oxidative stress in patients with hemodialysis. In 2008, researchers from Tohoku University School of Medicine and Nihon Trim started collaborative clinical research and established the Electrolyzed Water-Hemodialysis Research Group Association. They believed that the effect of electrolyzed water may come from the action of dissolved hydrogen molecules.

In 2005, Yanagihar et al. from Miz revealed that the hydrogen-rich neutral water produced by the company's unique electrolyzer reduced oxidative stress in mice. This was a groundbreaking achievement, because the researchers proved that it was the hydrogen in alkaline water rather than alkaline substances that exerted therapeutic effects.

In 2007, Shigeo Ohta and his colleague from Nippon Medical School published a paper in *Nature Medicine*, reporting that hydrogen had obvious therapeutic effects on rat models of cerebral infarction. Experimental data showed that inhalation of 1% to 4% hydrogen narrowed the areas of cerebral infarction in rats. The researchers further pointed out that hydrogen had specific functions in eliminating ·OH and peroxynitrite without affecting hydrogen peroxides or superoxides. The discovery sparked a wave of interest in the biological role of hydrogen and its clinical significance in various human diseases. As a result, the value of hydrogen for life and health attracted increasing attention from the fields of medicine, science, industry and capital.

In recent years, scholars mainly from Japan, China, and the United States have conducted extensive research on hydrogen and its connections with human in-vivo environment, cytology, genetics, cell signaling, disease models, human diseases, treatment-related pathology, and pathophysiology. Hydrogen molecules were found to have strong antioxidant and anti-inflammatory effects. Meanwhile, their effects on cell signaling, gene expression regulation, and cell protection were analyzed. Research findings further confirmed that hydrogen is useful for intervening in and improving many diseases related to peroxidation and inflammation.

Throughout the history of medical development, any new scientific discovery and technological invention must be supported with solid scientific research. In particular, when it comes to methods or products with medical effects, it is necessary to identify their impact on normal physiological functions. This fundamental scientific matter requires a long process of accumulating knowledge and experience. The concept of hydrogen medicine has been through this process before being gradually established.

Based on the aforementioned research progress, the U.S. Office of Food Additive Safety issued a statement in 2014, stating that it is safe to use hydrogen dissolved in water. In 2015, China promulgated national standards, specifying hydrogen as a food additive.

Relation between Hydrogen and Traditional Medicine

Zhang Xichun (1860–1933), a famous doctor in the Qing Dynasty in China, wrote in his book *Records of Tradition Chinese and Western Medicine in Combination*: "One can find hydrogen in the center of a furnace. There is also hydrogen in the human abdomen. Dried rehamnnia root or astragalus membranaceus leads hydrogen to the lungs, where hydrogen combines with inhaled oxygen and energizer (the portion located below the stomach) without causing frequent urination." Therefore, which coincides with the current understanding that there is hydrogen in the abdomen, functions of dried rehamnnia root or astragalus membranaceus

described in the book seem to be consistent with the current evidence that hydrogen can improve diabetes symptoms.

In 1996, Du Yuanwei, a teacher from Shandong, China, first put forward the significance of exploring hydrogen to life. In the abstract of a paper used for an international conference, he presented the important role of hydrogen in living organisms. He believed that the hydrogen generated after water electrolysis boosts vital movement of plants (lilac branches), animals (white mice), and the human body. Moreover, Du Yuanwei pointed out that the nature of carcinogenesis, no matter based on which theory, lies in the oxidation and peroxidation of the substrate in the process of biological metabolism; and that hydrogen may play the role as a scavenger in organisms. From 2002 to 2013, he published several papers elaborating the idea that "a proper amount of hydrogen in the human body may have inestimable effects on various diseases and the anti-aging process", including on the germination and growth of wheat seeds. In an experiment, Du Yuanwei placed wheat seeds at the anode (producing oxygen) and cathode (producing hydrogen) respectively after water electrolysis. Results showed that the germination rate of wheat seeds at the cathode in Day 8 and Day 15 was higher than those at the anode. In addition, Du Yuanwei did an experiment in which hydrogen was injected into the central medullae of clove and peach tree branches. Results showed that the flower buds opened up earlier and withered later than those of the control group. In another experiment, Du Yuanwei injected hydrogen subcutaneously into mice. Compared with the control group, the mice recovered from anesthesia in shorter time, became more resistant to hunger, and grew faster in the later stage. Afterwards, Du Yuanwei gave himself subcutaneous and intramuscular injections of hydrogen. He felt that he had stronger vitality without adverse reactions.

In 1999, Du Yuanwei et al. published a paper in the Journal of Shandong Normal University, presenting that hydrogen exerts reductive and antioxidative effects in the body. According to Du Yuanwei, excessive peroxides are accumulated in the human body during the metabolic process, causing aging and many diseases. The human body must have a certain mechanism that controls peroxides. As a strong reducing agent, hydrogen eliminates peroxides without side effects and therefore achieves redox balance. The aforementioned viewpoints of Du Yuanwei happen to be consistent with our current understanding of the biology of hydrogen molecules. If the paper is regarded as the beginning of his research on the role of hydrogen biology, Du Yuanwei's discovery came 3 years earlier than that of the French diving medical scientist, who proved in 2001 that breathing eight atmospheres of high-pressure hydrogen gas created anti-inflammatory effects. Du Yuanwei was also 8 years ahead of Shigeo Ohta in discovering the specific functions of hydrogen molecules in scavenging hydroxyl radicals in 2007. It is a pity that the journals where his studies were published had very limited influence. If the studies had been published in domestic English

journals such as Scientific Reports and Chinmed, the situation would have been different today; and the journals would probably have gained the first right to publish about hydrogen biomedicine. It is noteworthy that Du Yuanwei pointed out the relationships between traditional Chinese medicine (TCM) Qi and hydrogen. According to *Inner Canon of the Yellow Emperor*, a medical classic of Chinese medicine, nutrient Qi is a cereal essence. And those with nutrient Qi secrete body fluids, which nourish the pulse and eventually turn into blood. TCM holds that "Man comes to life through the Qi of heaven and earth." and "Qi is the origin of the human body." Believing that Qi is hydrogen, Du Yuanwei proposed the concept "biological hydrogen energy". Hydrogen, like ideal clean energy, has high power in a living body and can be

complementary to organic sources of energy in normal circumstances. Du Yuanwei boldly speculated that the energy that sustains life activities for people who fast and only drink a small amount of water comes from biological hydrogen energy.

To further verify whether hydrogen can spread rapidly to the whole body in 2013, Du Yuanwei and his follow researchers produced tritium gas by electrolyzing tritiated water (a hydrogen isotope that can be specifically labeled and distinguished from other hydrogen elements), and blew tritium gas instead of hydrogen gas into the environment where mice lived. In the end, tritium was detected in various tissues and organs of the mice, which proved that tritium participates in the life activities of organisms, is transformed into tritium ions, and exist generally in organisms. This also indirectly proved that the hydrogen generated in the air and intestinal tracts is both a component and a substance of energy source for life.

In 2017, in response to previous research findings, Du Yuanwei and his fellow researchers put forward the hypothesis that tissue fluid circulation exists in the human body. Considering that senses of air inflation and airflow are felt during internal martial art practice, Du Yuanwei believed that the "genuine Qi and primordial Qi" as well as "healthy Qi and middle Qi" in TCM are neither physical nor from pipelines; but merely represent certain passageways, medullary channels or intervals for Qi. It is hydrogen that runs in meridians. According to Du Yuanwei, tissue fluid is probably the Qi referred to in TCM; and the interval channels for circulating tissue fluid are meridians. Tendons, ligaments, and fascia function as parts of a transport network. Du Yuanwei's hypothesis that hydrogen is mainly transmitted with tissue fluid is the first of its kind in human history.

According to a paper in the journal Science Reports on March 27, 2018, two doctors at the Yobe Israel Medical Center in the United States discovered a type of reticular tissues beneath the skin, around blood vessels, and in the underlining of lungs, digestive organs and urinary system. There was fluid passing through the tissues. The doctors called the tissues new organs. Does this coincide with Du Yuanwei's hypothesis? Are the tissues equivalents to the meridians in TCM? Is it possible that the fluid contains hydrogen? All of this is worthy of study.

Du Yuanwei's ideas have theoretical value in terms of academic thought. However, it requires systematic observation and analysis to verify the ideas via scientific experiments. It can be expected that scientific confirmation of his ideas will open up a new path for the study of the biological effects of hydrogen, and inject modern scientific meaning to the TCM concept "meridian" that has been explored for years without real answers.

Clinical Observation and Evaluation of Hydrogen Efficacy

Based on the research papers on hydrogen biology and medicine, many medical institutions and technology companies in China have carried out research on the clinical value of hydrogen. A total of six multicenter randomized, double-blind and controlled studies of hydrogen therapy are being and will be conducted, covering: a) hydrogen-assisted treatment for allergic rhinitis; b) hydrogen-assisted treatment for acute chronic obstructive pulmonary disease (COPD); c) hydrogen-assisted treatment for severe COPD in stable stage; d) hydrogen-assisted treatment for severe asthma; e) hydrogen-assisted treatment for bronchiectasis; f) adjuvant therapy for non-small-cell lung cancer. The research work is carried out by 16 hospitals from Shanghai, Tianjin, Guangzhou, Hebei and other places; and involves hundreds of cases.

There are many single-center studies of hydrogen therapy across China, involving the therapeutic effects of hydrogen on airway stenosis, Alzheimer's disease, Parkinson's disease, spinal cord injury rehabilitation, PM2.5 haze-caused harm, breathing-related sleep disorder & metabolism disorder, respiratory rhythm regulation, postoperative cognitive impairment in elderly patients, postherpetic neuralgia, non-alcoholic fatty liver disease, diabetes, hyperlipidemia, pulmonary fibrosis, and depression.

Topics and Outlook on Hydrogen Medicine Research

Hydrogen medicine research needs to be approached from the following three aspects: clinical issues and scientific hypotheses; research methods and tools; and market potential and capital. Looking at the development history of scientific research, it is extremely challenging to raise creative scientific issues and identify important scientific topics. In particular, appropriate research methods are the key to scientific discovery. In today's society, the support of funding and capital also plays a vital role.

1. Considerations of clinical challenges and scientific hypotheses

In the past decade, China's academic and industrial circles have started paying close attention to the scientific significance and application value of hydrogen. In particular, the biological importance, physiological significance and clinical application of hydrogen molecules aroused wide concern from the medical industry. Experimental observations have preliminarily confirmed that hydrogen is potentially effective in regulating physiological functions; maintaining health; and treating radiation damage, brain trauma, burn shock, sepsis, pancreatitis, decompression sickness, oxygen poisoning, liver cirrhosis, Alzheimer's disease and other diseases.

The Second Military Medical University was the earliest academic institution in China to conduct hydrogen medicine research. In 2011, academician Wu Mengchao and his team discovered that hydrogen can protect against liver damage and inhibit the process leading to cirrhosis and hepatic cell compensatory hyperplasia. Academician Xia Zhaofan was the first in the world to put forward the concept of hydrogen recovery. Prof. Sun Xuejun edited *Hydrogen Molecular Biology* and co-authored *Hydrogen Molecular Biology and Medicine* with Japanese professor Shigeo Ohta. Both the book presented the mechanism of hydrogen in treating diseases, therefore providing important references for the study of hydrogen biology and its applications.

In 2014, a research team of Guangzhou Medical University led by academician Zhong Nanshan started research on hydrogen medicine, and conducted a number of clinical multi-center trials related to COPD. The team proposed ideas such as "Hydrogen is means of etiological treatment and helps prevent diseases." and "Hydrogen treatment mainly targets at chronic diseases." Their research points the way for the healthy development of hydrogen biomedicine.

It was Xu Kecheng and his team that first linked hydrogen to cancer. Their retrospective and prospective investigation of hydrogen control of progressive cancer is pioneering research that has received extensive attention.

2. Considerations of research methods and tools

Technical innovation is an important driving force for scientific discovery. Using new water electrolysis technology, Shanghai Asclepius Meditec Co., Ltd. produced the world's first

medical-use hydrogen-oxygen nebulizer. The machine was defined by China Food and Drug Administration as a Class III medical instrument (category ID: 6826) in 2018; and has obtained 118 patents for invention and utility models from at home and abroad. Designed with the features "fresh made, instant use, zero pneumatization" to avoid accumulation of hydrogen and oxygen gases, the machine is equipped with atomized water facilities to avoid dry static electricity and spontaneous combustion, therefore ensuring safe use.

3. Considerations of market potential and capital

In consideration of its biological significance and potential clinical value, hydrogen has drawn extensive attention from the fields of fund, industry and investment and financing. From 2008 to 2017, the Chinese National Natural Science Foundation increased hydrogen biomedical research projects and funding year by year. By 2018, it approved a total of 69 projects covering the following research topics: the mechanism of action of hydrogen molecules; protective effects of hydrogen inhalation on various diseases (especially nervous, respiratory, cardiovascular and metabolic diseases) and pathological conditions (radiation damage, wounds, inflammation); and other topics. In addition, hydrogen medicine was studied on more than 100 projects under the support of provincial and municipal governments as well as universities, hospitals and research institutions. In early 2019, Shanghai Jiao Tong University established Hydrogen Scientific Research Center for the studies of hydrogen energy, hydrogen agriculture and hydrogen medicine. The Center attracted the attention from relevant state departments and the society.

The good news is that hydrogen research started from companies and has been driven forward by resources, technology and talents from universities and research institutes. The aforementioned hydrogen-oxygen nebulizer manufactured in Shanghai has unique advantages and offers a reliable means for randomized controlled multi-center studies of the natural population and diseased groups. It has attracted attention at home and abroad.

Summary

Hydrogen is the earliest, oldest, and most basic element in the universe. The original universe consisted almost exclusively of hydrogen. Moreover, the bacteria in human intestines produce hydrogen all the time. In spite of these facts, human understanding of hydrogen in life activities and health maintenance remains preliminary.

In the early stage of any science, different hypotheses and theories are often mutually challenging and even exclusive. This makes it difficult for different schools of thought to communicate with and understand each other. Historical experiences are reminders that good things are generally objected at the beginning. We believe that as time goes by, people will eventually acquire true knowledge through observation and experiments. It is reassuring that scientific workers in different areas have joined the undertaking of hydrogen medicine and gradually established strong research groups in the course of exploration. They are not only engaged in basic scientific experiments, but also have transformed their studies into clinical research. The real world evidence-based reseach, which has received wide attention, is expected to play a role in clinical transformation.

We believe that as long as we follow appropriate academic views; adopt correct research thinking, methods and techniques; carry out serious exploration; and incorporate traditional Chinese culture elements in our exploration and research, China's hydrogen medical research will make considerable headway.

— Lv Youyong, Xu Kecheng, Wu Peihong, Li Jiaping

References

1. 2013. Sun Xuejun. *Hydrogen Molecular Medicine*. Shanghai Second Military Medical University Publishing House, 2013.
2. Ohta S. Molecular hydrogen as a novel antioxidant: overview of the advantages of hydrogen for medical applications. Elsevier Inc, 2015.
3. Ohta S. Recent progress toward hydrogen medicine: potential of molecular hydrogen for preventive and therapeutic applications. Curr Pharm Des, 2011, 17(22): 2241-2252.
4. Levitt MD. Production and excretion of hydrogen gas in man. N Engl J Med, 1969, 281(3): 122-127.
5. Dole M, Wilson FR, Fife WP. Hyperbaric hydrogen therapy: a possible treatment for cancer. Science, 1975, 190(4210): 152-154.
6. Roberts BJ, Fife WP, Corbett TH, Schabel FM, Jr. Response of five established solid transplantable mouse tumors and one mouse leukemia to hyperbaric hydrogen. Cancer Treat Rep, 1978, 62(7): 1077-1079.
7. Rostain JC, Gardette-Chauffour MC, Lemaire C, Naquet R. Effects of a H2-He-O2 mixture on the HPNS upto 450 msw. Undersea Biomed Res, 1988, 15(4): 257-270.
8. Gharib B, Hanna S, Abdallahi OM, et al. Anti-inflammatory properties of molecular hydrogen: investigation on parasite-induced liver inflammation. C R Acad Sci III, 2001, 324(8): 719-724.
9. Shirahata S, Hamasaki T, Teruya K. Advanced research on the health benefit of reduced water. Trends Food Sci Tech, 2012, 23: 124-131.
10. Yanagihara T, Arai K, Miyamae K, et al. Electrolyzed hydrogen-saturated water for drinking use elicits an

antioxidative effect: a feeding test with rats. Biosci Biotechnol Biochem, 2005, 69(10): 1985-1987.

11. Ohsawa I, Ishikawa M, Takahashi K, et al. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med, 2007, 13(6): 688-694.
12. Ichihara M, Sobue S, Ito M, et al. Beneficial biological effects and the underlying mechanisms of molecular hydrogen — comprehensive review of 321 original articles. Med Gas Res, 2015, 5: 12.
13. Ostojic SM. Targeting molecular hydrogen to mitochondria: barriers and gateways. Pharmacol Res, 2015, 94: 51-53.
14. Dixon BJ, Tang J, Zhang JH. The evolution of molecular hydrogen: a noteworthy potential therapy with clinical significance. Med Gas Res, 2013, 3(1): 10.
15. Du Yuanwei. Thinking on Qi from a Material Perspective. Qigong and Science, 1996, 11: 34.
16. Du Yuanwei. Hydrogen Energy and Life Hydrogen. Solarmedia, 2002, 5: 15.
17. Du Yuanwei. Hydrogen Energy and Life Hydrogen. Teaching of Middle School Biology, 2002, 1-2: 42.
18. Du Yuanwei, Li Peng, Zhao Xianfeng. Experimental Confirmation of the Role of Hydrogen in Life. Journal of Zibo Normal College, 2013, 1: 81-82.
19. Du Yuanwei, Wei Cuigui. A New Subject of Science—First Exploration of the Significance of Hydrogen in Life Activities. Journal of Shandong Normal University, 1999, 2: 196-197.
20. Wang Yusheng, Jiang Jinxu, Zhang Guangfu, Du Yuanwei. Tissue Fluid Circulation: A Human Body System More Accurate Than Blood Circulation. Chinese Journal of Health Care Nutrition, 2017, 27(10): 97.
21. Benias PC, Wells RG, Sackey-Aboagye B, et al. Structure and Distribution of an Unrecognized Interstitium in Human Tissues. Sci Rep, 2018, 8(1): 4947.
22. Sun H, Chen L, Zhou W, et al. The protective role of hydrogen-rich saline in experimental liver injury in mice. J Hepatol, 2011, 54(3): 471-480.
23. Zheng XF, Sun XJ, Xia ZF. Hydrogen resuscitation, a new cytoprotective approach. Clin Exp Pharmacol Physiol, 2011, 38(3): 155-163.

Hydrogen: A New Option to Prevent and Treat Different Diseases

Key points:

- Hydrogen can easily spread in organisms and enter the cytoplasm, nuclei and mitochondria. This is the foundation for hydrogen to maintain health and treat various diseases of the body.
- Molecular hydrogen is usually administered through hydrogen inhalation, oral intake, or injection of hydrogen-rich (saline) water. It controls oxidation (by eliminating toxic reactive oxide species), controls inflammation, maintains metabolic balance, and participates in gene expression regulation. This represents the main principle that explains hydrogen's effects on diseases related to peroxidation and uncontrollable chronic inflammation.
- Molecular hydrogen has certain adjunctive therapeutic effects on most diseases of human systems and organs; and helps treat different diseases.
- In terms of health management, molecular hydrogen can remove discomforts of unknown origin in the body, improve sleep, relieve fatigue, enhance beauty, and slow down the aging process. It might be a new means to prevent diseases.
- The human colon produces up to 12 liters of hydrogen per day, indicating that hydrogen is necessary to maintain normal physiological functions. Hydrogen inhalation serves only as a supplement to insufficient hydrogen generated in the body.

Usually, human cells stay in oxidative stress due to the abnormal oxidation potential of excess reactive oxygen species (ROS). Most of the oxygen inhaled into the body turns into non-toxic ROS with physiological functions; and a small amount becomes toxic ROS. If not cleared in time, the toxic ROS exerts destructive effects in cells (especially in mitochondria). Toxic ROS may originate from a variety of conditions, such as intense exercise, inflammation, ischemia reperfusion (I/R) injury, bleeding, tissue and organ transplantation. Persisting chronic oxidative stress is closely associated with many lifestyle-related diseases, aging and cancer. In spite of their various clinical types, anti-oxidant supplements are ineffective if used in small doses, but generate highly toxic side effects if used in large doses. As a result, it is difficult to apply the supplements based on individual differences. Therefore, finding non-toxic or less toxic antioxidants for maintaining health or treating certain diseases has always been the goal of medical professionals' exploration and pursuit.

Unremitting efforts in recent decades led to the discovery that hydrogen (H_2) is a non-toxic, natural, simple, convenient and effective selective antioxidant molecule. As early as 1975, Dole M. et al. described the inhibitory effects of H_2 on squamous cell carcinoma of the skin in mice. In 2001, French researcher Gharib B et al. reported that hydrogen inhalation helps treat hepatitis caused by parasitic infection of human liver. In 2007, Japanese scholar Shigeo Ohta et al. found that hydrogen inhalation selectively neutralizes hydroxyl (·OH) and peroxynitrite ($ONOO^-$) and protects mouse

models of radiation and stroke from brain damage.

It is not a new concept that gases deliver therapeutic effects on diseases. Oxygen is an indispensable substance for human survival; and its application to medicine is universally known. In the 1980s, nitric oxide (NO) was discovered to be the molecular mechanism behind nitroglycerin-based treatment for angina. NO originates from coronary vascular endothelial cells and induces hemangiectasis. Its researchers were awarded the Nobel Prize in Physiology or Medicine in 1998. Since then, research on medical-use gases has made rapid progress. Both carbon monoxide (CO) and hydrogen sulfide (H_2S) have strong antioxidant effects and are considered to be very important neurotransmitters. These aforementioned

three types of gas molecules are referred to as gas signal molecules in the body (see the table below).

Table — Features of medical-use gases

Gas	NO	CO	H_2S	H_2
Color & odor	None & none	None & none	None & stimulating odor	None & none
Flammability	–	–	+	++
Toxicity	+	++	++	–
Generated in the body	By NOS and XO	By HO	By CBS and CSE	Intestinal flora
Hemangiectasis	++	+	+	–
Anti-inflammatory effect	+	+	+	+
Anti-apoptosis effect	+	+	+	+

(NOS: nitric oxide synthase; XO: xanthine oxidase; HO: heme oxygenase; CBS: cystathionine-β-synthase; CSE: cystathionine-γ-lyase)

NO has been used clinically to treat heart and lung diseases. In clinical studies, CO has been applied to treating patients with kidney transplantation and severe respiratory failure. And H_2S has been used to treat patients with renal insufficiency and history of coronary artery bypass surgery. However, for a long time, people find it hard to believe that H_2 has any biological effect. That is because H_2, compared with other gases, is a non-free radical, non-reactive, non-polar, and highly diffusible neutral gas. It is unlikely that H_2 has specific binding site, or specific interaction with specific receptors.

Characteristics of Molecular Hydrogen

Molecular hydrogen has multiple biological functions, which lie in its characteristics that are not found in general drugs and biomolecules:

1. High penetrability and fast diffusion

With high biofilm penetrability and intracellular dispersion, hydrogen molecules can enter the organelle structures in the nuclei and cytoplasm, including mitochondria. They can pass into both normal tissues and ischemic areas; and penetrate the blood brain barrier, which is not possible for common antioxidants and chemical drugs. Detection results of hydrogen content in expiration showed that inhaled hydrogen was distributed throughout the body within 10 minutes.

In 2019, Japanese researcher Yamamoto et al. studied the distribution of hydrogen in mice after the mice was given consecutive inhalation of 3% hydrogen (see Figure 1 below). The length of time for hydrogen in muscles to reach a saturation level was 20.2 minutes, which was significantly longer than 6.3 to 9.4 minutes for other organs. Moreover, hydrogen concentration gradually increased with time. By organ, the liver had the highest concentration of hydrogen (29.0 ± 2.6 μmol/L), and kidney the lowest (18.0 ± 2.2 μmol/L).

2. High selectivity and protective effect on functional ROS

With weak reducibility, hydrogen only selectively neutralizes active and highly toxic reactive oxygen (such as ·OH). It has no adverse effects on other free radicals involved in normal signal regulation, such as O_2^- and H_2O_2. Low concentrations of O_2^- and H_2O_2 may act as signaling molecules that regulate cell proliferation, differentiation, and apoptosis. In spite of

their strong reducibility, antioxidants such as vitamin C or E may destroy normal ROS and intervene in normal cell signaling. Some studies have shown that H_2O_2 at a high concentration is converted to hypochlorous acid under the action of myeloperoxidase, which prevents bacterial invasion. NO acts as a neurotransmitter and is essential for hemangiectasis. Excess antioxidants destroy the important defense mechanisms, thereby increasing the incidence and mortality of diseases including cancer. However, molecular hydrogen does not cause these side effects.

3. Great safety in spite of high concentration

In the past few decades, some gas molecules, including NO, CO and H_2S, have been recognized for their biological functions. Inhalation of exogenous CO or H_2S may benefit most vital organs. However, these gases can be highly toxic at high concentration levels. NO reacts with O_2^- and generates highly oxidized peroxynitrite ($NO + O_2^- \rightarrow ONOO^-$), which strengthens oxidative stress. Even O_2, the most widely used gas in clinical practice, causes oxygen poisoning if used at high concentrations.

H_2 is different. It has no even cytotoxicity even at a high concentration. After reacting with free radicals, H_2 generates water, and all the redundant H_2 is excreted out of the body through breathing. High-pressure H_2 is used in gas mixtures for deep diving and helps prevent decompression sickness and arterial gas thrombosis. Hydreliox, which is applied to deep diving, is a mixture of exogenous breathing gases containing 49% H_2, 50% He and 1% O_2.

With very low activity, H_2 manifests as an inert gas in the absence of catalysts or body temperature. It does not react with most chemical compounds, including oxygen at room temperature. H_2 is flammable only at a temperature of above 527℃. In addition, it explodes through a rapid chain reaction with O_2 only within the explosion range of H_2 concentration (4% to 75%).

Biological Effects of Molecular Hydrogen

1. Antioxidant effects

In the Chinese medicine classic *Inner Canon of the Yellow Emperor*, cancer is described as a large gathering of toxins, including heat and damp toxins. The most common therapy in TCM is detoxification. What are exactly toxins? According to modern biomedicine, the most powerful toxin in the body is toxic ROS, which can be generated by a many pathogenic factors ranging from

physical factors (such as ionizing radiation and ultraviolet light) and chemical factors (such as chemical toxins) to biological factors (such as bacteria, virus infections and their released biotoxins) and abnormal immunity (such as rheumatoid arthritis). Hydrogen molecules eliminate toxic ROS by the following means:

1) Hydrogen molecules activate the Nrf2-Keap1 system; induce the activation of antioxidant response elements (ARE); and secrete various cytoprotective proteins such as glutathione, catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase and heme-1 oxygenase.

2) Hydrogen molecules activate the transcription factor FoxO1; up-regulate the expression of manganese superoxide dismutase (MnSOD) and CAT; and reduce damage to mitochondria and massive generation of ROS.

3) Hydrogen molecules activate the pathway of AMP-activated protein kinase (AMPK); regulate cell growth; and reshape energy metabolism, thereby achieving anti-apoptotic effects.

2. Anti-inflammatory effects

Inflammation is one of the most important causes of many diseases and sub-health status. Cancer is also considered to uncontrollable chronic inflammation. Some believe that cancer is actually an inflammatory wound that does not heal for a long time.

In 2001, French researcher Gharib et al. reported that inhalation of high-concentration H_2 cured liver inflammation caused by parasites, suggesting that H_2 had anti-inflammatory properties. In 2013, Japanese scholar Ishibashi et al. presented that hydrogen inhalation had a residual effect in the treatment of rheumatic arthritis, which continued to recede 4 weeks after hydrogen inhalation was stopped. Many cytological tests have shown that cells are still resistant to attacks (such as attacks from toxins, radiation, or damage) long after H_2 has disappeared from the culture system, indicating that H_2 has lingering anti-inflammatory effects. The anti-inflammatory effects of H_2 are achieved mainly by down-regulating various pro-inflammatory and inflammatory cytokines including interleukin (IL)-1β, IL-6, tumor necrosis factor (TNF)-α, intercellular adhesion molecule (ICAM)-1, high mobility group box (HMGB)-1, nuclear factor (NF)-κB, and prostaglandin (PG) E2.

> *Picture 1. Time needed for organs to reach hydrogen saturation levels in mice after hydrogen inhalation (hydrogen concentration stops increasing after reaching the saturation level). [Source: Yamamoto R, et al. Sci Rep, 2019, 9(1): 1255.]*

3. Anti-apoptotic effects and cell protection

Apoptosis is programmed cell death, representing a slow and natural way of dying for cells. Cell apoptosis and cell regeneration balance each other. Too little apoptosis and constant cell regeneration result in the formation of tumors. However, excessive apoptosis impairs organ structure and functions, causing diseases and failure to maintain vital functions. In 2017, Chinese researcher Ge et al. reviewed the basic and clinical data about the roles of H_2 in significantly reducing apoptosis induced by ischemia, infarction, poisoning and inflammation; and in protecting cells.

The mechanism by which H_2 inhibits cell apoptosis is primarily its inhibition on the activity of Caspase-3 and -12. In addition, H_2 suppresses the expression of pro-apoptotic factors and Bax; and up-regulates the expression of anti-apoptotic factors and Bcl-2. According to a study by Chinese scholar Hou et al. in 2012, H_2 activates the anti-apoptotic protein kinase B in neurons and has neuroprotective effects. In 2011, Japanese scholar Nakai et al. summarized that oral hydrogen-rich water changes the functions of more than 200 biomolecules, which in turn affects the expression of more than 1,000 genes.

4. Signal pathway regulation

Life consists of matters (flesh), energy, and signals. Without the transmission, connection and communication of information, the life process cannot proceed. H_2 affects multiple signal pathways. As a gas signal regulator, the action of H_2 involves a network of signal molecules. For example:

1. H_2 regulates NF-κB. NF-κB controls hundreds of genes involved in cell growth, differentiation, development, inflammation, and apoptosis. H_2 activates NF-κB, thereby activating a range of cell behavior.

2. H_2 promotes the expression of Nrf2 in the body, especially in organs such as the lung, liver and kidney. Nrf2 is a new regulator of innate immune response. H_2 up-regulates Nrf2 and maintains the body's immune function.
3. H_2 induces heme oxygenase-1 (HO-1) and its enzymatic reaction. In addition to their significant anti-inflammatory effects, HO-1 and its product CO eliminate infections by increasing phagocytosis and endogenous antibacterial reactions; and alleviate inflammation caused by hyperoxia and improve blood oxygenation.

Hydrogen: Treatment of Different Diseases and Preventive Treatment

TCM holds that human health is maintained by "keeping the head cool, feet warm and bowel movement smooth". Stagnation of Qi and blood stasis lead to obstruction and cause pain. The current human health problems are basically closely related to the fact that "Once Qi stagnates,

blood flow is restrained." Researcher Ge et al. once summarized literatures officially published as of March 2017. According to the summary, H_2 is potentially effective in terms of biology and medicine in treating more than 100 types of diseases. Some believe that H_2 has therapeutic effects (see Picture 2 below) on the conditions that are top 10 causes of death in the United States, except for suicide and accidents. The following is a detailed list by disease type.

1. Central nervous system diseases

Oxidative stress and nervous system inflammation cause neurological diseases, such as brain damage, neurodegenerative diseases, and other neurological diseases secondary to diseases. In 2009, Japanese researcher Nagata et al. experimented on mice and found that H_2 penetrated the blood brain barrier through diffusion and exerted therapeutic effects on some central nervous diseases.

(1) Ischemia reperfusion (I/R) injury: also known as vascular occlusion (stroke, cerebral thrombosis or cerebral embolism) – recanalization (thrombolysis) – tissue damage caused by blood reperfusion. This condition is usually more harmful than stroke itself. Application of H_2 to I/R injury treatment is the most reported of all research on H_2. In 2007, Japanese scholar Shigeo Ohta et al. first reported that hydrogen inhalation significantly inhibited focal I/R brain damage in rats, proving that the therapeutic effect was achieved because H_2 eliminated excessive toxic ROS caused by I/R injury.

(2) Cerebral hemorrhage: an acute condition with extremely high mortality and often a complication of high blood pressure. In 2013, American researcher Manaenko et al. induced intracerebral hemorrhage by intracerebral injection of collagenase. The animals showed evidently better brain barriers and neurological functions after hydrogen inhalation than the group without hydrogen inhalation. Meanwhile, researchers discovered that mast cell activation is associated with cerebral hemorrhage and can be blocked by H_2. In 2013, Chinese researcher Shao et al. verified with rabbit experiments that injection of hydrogen-rich water can lessen brain edema and blood brain barrier destruction; reduce neuronal apoptosis; and improve neurological functions. Relief of oxidative damage is related to activation of the NF-κB pathway. In 2016, Shao et al. repeated the experiments in rat models and came to similar findings. The results indicated that in addition to NF-κB pathway activation, the formation of NLRP3 inflammasome was also involved.

(3) Parkinson's disease: The main pathological change is abnormality in dopaminergic neurons in substantia nigra and nigrostriatal pathways. Such abnormality is connected with

excessive oxidation of the substantia nigra and deterioration of endogenous antioxidant function. The incidence of Parkinson's disease increases sharply with age. Around 1.5% of all human beings are exposed to the risk of developing the disease. Currently, the antioxidants used clinically have not shown significant therapeutic effects. In 2009, Japanese researchers Fu et al. and Fujita et al. experimented on rats and mice respectively; and verified that hydrogen-rich water can reduce nigrostriatal degeneration and inhibit the loss of dopaminergic neurons. In a randomized, double-blind and placebo-controlled trial in 2013, Japanese scholar Yoritaka et al. gave participants one liter of hydrogen-rich or normal water every day for 48 weeks consecutively. The results showed that Parkinson's disease rating scale for the hydrogen-rich water group improved, whereas that for the placebo group worsened.

(4) Alzheimer's disease: So far, there is no effective treatment for Alzheimer's disease. In 2010, Chinese researcher Li et al. injected hydrogen-rich saline into rat models of Alzheimer's disease; and found that the rats' cognitive and memory functions improved afterwards. According to a study by Wang et al. from the same research group in 2011, the mechanism of action behind the improvement might be that hydrogen-rich saline improved the abnormal signaling related to Alzheimer's disease.

(5) In 2009, Japanese scholar Nagata et al. experimented on mice and discovered that injection of hydrogen-rich water lessened neuronal damage caused by uterine I/R in the hippocampus of fetal mice and blocked endotoxin-induced activation of microglial cells in fetal mice's brains. As a result, neuronal damage and brain damage caused by such cell activation were reduced.

2. Cardiovascular diseases

(1) Myocardial infarction: is an acute condition caused by persisting coronary ischemia and has a high mortality rate. In 2008, Hayashida et al. prepared rat models of myocardial infarction through coronary artery clipping, and found that myocardial infarction was alleviated after hydrogen inhalation. In 2015, Jing et al. induced myocardial infarction in rats with isoproterenol. In 2016, Gao et al. induced myocardial damage in rats with doxorubicin, and obtained similar experimental results.

(2) Cardiac arrest/resuscitation: has extremely high motality rate. In 2014, Huo et al. verified with rat models that hydrogen-rich saline can significantly improve the survival rate of cardiac arrest and reduce the sequelae of nervous system injury. Currently, Japan has listed cardiac arrest/resuscitation as a statutory indication for hydrogen inhalation therapy.

(3) Heart transplantation: has been increasingly applied to treating heart failure and heart diseases. In 1997, Knight et al. conducted a heart transplantation experiment on rats. The results verified that the preservation solution containing hydrogen-rich saline was better than ordinary preservation solutions, and led to longer survival time after heart transplantation. In 2009, Sun et al. experimented on rat models and proved that adding hydrogen-rich saline during extracorporeal circulation in extracardiac surgery helped reduce myocardial damage caused by extracorporeal circulation and blocked myocardial apoptosis. In 2011, the same research team found that this effect was achieved by activating the JAK2/STAT3 signaling pathway.

(4) Cardiac hypertrophy: may result in primary or secondary cardiomyopathy. In 2018, Fan et al. verified from rat models that hydrogen-rich saline protected the myocardium and reduced hypertrophy and damage.

(5) Atherosclerosis: is a common chronic disease that often causes high blood pressure. Apolipoprotein E plays an important role in preventing atherosclerosis. In 2008, Ohsawa et al. prepared mice whose apolipoprotein E genes were knocked out, and created models of atherosclerosis. The researchers observed that after multiple injections of hydrogen-rich water, endarteritis was eased; atherosclerotic plaques became more stable; and less macrophage-derived foam cells were generated. As a result, various complications of atherosclerosis were prevented. In 2018, Matsuoka et al. prepared rat models of atherosclerosis by using high-salt diet, and obtained similar results after hydrogen inhalation.

(6) Hypertension: is the most common disease in modern society and has an incidence rate

higher than 40%. In 2019, Guan et al. from experiments on rat models of spontaneous hypertension, verified that hydrogen inhalation prevented hypertension attacks to rat models.

> **Picture 2. Therapeutic effects of hydrogen on diseases in animal models and human studies** (organized by body system; central figure labeled H_2)
> *Cardiovascular system:* Acute myocardial infarction; Heart transplantation, Myocardiopathy; Atherosclerosis; Hypertension; awaking from sudden cardiac arrest.
> *Central nervous system:* Cerebral infarction; Parkinson's disease; Alzheimer's disease; Carbon monoxide poisoning of the brain; Transient cerebral ischemia; Cerebral blood supply insufficiency, subarachnoid hemorrhage; Cerebral injury after sudden cardiac arrest.
> *Respiratory system:* Acute lung injury; Bronchial asthma; Chronic obstructive pulmonary disease; Bronchiectasis, Radiation pneumonia; Intestinal ischemia-reperfusion lung injury; Lung transplantation.
> *Gastric, intestinal, hepatic, gallbladder and pancreatic systems:* Multiple liver injuries; Hepatic fibrosis; Hepatic ischemia-reperfusion; Hepatic-radiation-induced liver injury; Liver transplantation; acute gastric mucosal injury; Acute pancreatitis, Fatty liver.
> *Renal, urinary and reproductive systems:* Chemotherapy-induced renal injury; Kidney transplantation; Chronic kidney disease; Renal ischemia-reperfusion; Radiation-induced testicular injury; Radiation-induced ovarian injury; Uveositis.
> *Metabolic, skeletal, joint and muscular systems:* Diabetes; Hyperlipidemia; Obesity; Myopathy; Joint disorder; Osteoporosis; Osteonecrosis.
> *Inflammation/infection system:* Allergic reaction; Septicemia; Acute and chronic graft-versus-host disease; Aplastic anemia.
> *Cancer:* Parenchyma tumor; Side effects of radiotherapy/chemotherapy; Precancer prevention.
> *Fetal and neonatal diseases:* Cerebral anoxia, cerebropathy; Threatened abortion; Fetal inflammatory response syndrome.
> *Skin & sensory systems:* Physicochemical damage of the skin; Retinal ischemia, Retinal degeneration; Hearing disorder.

3. Hepatobiliary and pancreatic diseases

(1) Viral hepatitis: In 2001, French researcher Gharib et al. reported a study of professional deep-sea divers. According to the findings, inhaling high-pressure oxygen-hydrogen gas (oxygen cylinder) had therapeutic effects on hepatitis and various types of liver damage, leading to antioxidase increase in the blood, decrease in lipid peroxides, and less liver fibrosis. In 2009, American researcher Kajiya et al. confirmed that hydrogen inhalation improved liver functions in mice with hepatitis induced by concanavalin A. In 2013, Chinese researcher Xia et al. reported that drinking hydrogen-rich water helped reduce oxidative stress, improve liver functions and reduce viral load in patients with chronic hepatitis B.

(2) Liver damage: can be caused by many factors. In 2007, Japanese researcher Fukuda et al. reported that hydrogen inhalation treated I/R-induced liver injury in mice by reducing

oxidative stress in hepatic cells. In 2011, American researcher Kang et al. revealed that patients with liver tumor radiotherapy had significantly higher quality of life after drinking hydrogen-rich water. In 2012, Japanese researcher Kawai et al. presented that injection of hydrogen-rich water slowed down the progression of non-alcoholic fatty liver and prevented liver cancer. In 2014, Chinese scholar Tan et al. stated that liver failure was prevented after subtotal hepatectomy by injecting hydrogen-rich saline into mouse models. In 2014, Chinese researcher Liu et al. reported that hydrogen-rich saline helped treat I/R-induced liver injury in rat models by reducing oxidative stress in hepatic cells and HMGB1 release. In 2015, Chinese researcher Lee et al. reported that the injection of hydrogen-rich saline into rats with acetaminophen-induced cirrhosis improved the blood supply status of the whole body, spleen and liver. In 2016, another Chinese researcher Liu et al. reported that injection of hydrogen-rich saline helped improve hepatocellular mitochondria functions and reduce hepatic cell apoptosis in mice with obstructive jaundice. In 2016, Chinese researcher Shi et al. reported that injection of hydrogen-rich saline improved acute liver injury caused by acute necrotizing pancreatitis by inhibiting hepatic cell inflammation and apoptosis. As a result, JNK and p38-MAPK signaling pathways were reduced, resulting in less ROS being generated. In 2017, China's Zhai et al. reported that injection of hydrogen-rich saline treated non-alcoholic fatty liver in rats, which was attributed to the mechanism that oxidative stress was relieved; and liver PPAR α and γ were activated.

(3) Liver fibrosis: is an important pathological process of cirrhosis. In 2014, Koyama et al. verified with mouse experiments that oral intake of hydrogen-rich water alleviated liver fibrosis caused by chronic hepatitis B.

(4) Pancreatitis: According to a study by Zhang et al. in 2013, acute pancreatitis induced by taurocholate in rats can be alleviated by injection of hydrogen-rich saline. In 2015, Luo et al. reported that injection of hydrogen-rich saline reduced the severity of acute pancreatitis after pancreas transplantation and relieved pancreatic I/R injury.

4. Gastrointestinal diseases

As the home to complex and diverse microflora, the gastrointestinal tract has a symbiosis with the

human host and maintains human health. Most gastrointestinal microorganisms have the genetic ability to encode metabolic H_2, which means that H_2 may affect the composition of enteric microorganisms. In 1980, American researcher Levitt reported that 3000 mL H_2 was generated daily by intestinal bacteria in a normal adult. It can be seen that H_2 helps relieves intestinal diseases.

(1) Gastric ulcer: In 2012, Chinese researcher Liu et al. reported that injection of hydrogen-rich saline can treat pressure-related gastric ulcer in mice. In 2014, another Chinese researcher Zhang et al. presented that injection of hydrogen-rich water treated aspirin-induced gastric damage. In 2015, McCarty et al. observed that drinking hydrogen-rich water helped increase the expression and secretion of growth hormone releasing peptide (GHRP) and significantly boost gastric digestion functions in mice. In the same year, Japanese researcher Ichihara et al. made a summary of 321 literatures, confirming the conclusion that hydrogen mitigates gastric damage.

(2) Intestinal injury: In 2008, American researcher Buchholz et al. reported that hydrogen inhalation can alleviate oxidative stress; ease mucosal erosion and mucosal barrier

destruction; and reduce the possibility of postoperative intestinal obstruction in rat models of intestine transplantation. In 2009, Chinese researcher Zheng et al. reported that injection of hydrogen-rich saline mitigated intestinal I/R injury in rats. In 2011, Chen et al. from the same researcher team revealed that the injection boosted intestinal contractility and prevented intestinal structural changes after I/R injury. Similar findings were reported by Japanese researcher Shigeta et al. in 2015.

5. Respiratory diseases

(1) Acute lung injury: is a very common disease. In 2013, Chinese researcher Zhou et al. reported that hydrogen inhalation mitigated I/R-induced damage in rat models of lung transplantation. In 2015, Chinese researcher Zhai et al. reported that injection of hydrogen-rich saline can inhibit acute lung injury induced by sepsis, eliminate extravascular pulmonary edema, and reduce sepsis-related lung failure in rats. As indicated by animal experiments on many other factors, such as hyperoxia, lipopolysaccharide (LPS), smoking, paraquat, monocrotaline, and lung damage caused by extensive burns, injection of hydrogen-rich water or saline protects the airway mucosa and facilitates mucus production.

(2) Asthma: is a common chronic airway disease worldwide. Oxidative stress is considered to be important pathogenesis of asthma. In 2013, Chinese researcher Xiao et al. reported that injection of hydrogen-rich saline in mice with allergic asthma reversed severe inflammatory infiltration and goblet cell proliferation; significantly reduced the numbers of eosinophilic granulocytes and lymphocytes as well as the levels of such inflammatory factors as IL-4, IL-13 and TNF-α; and meanwhile, up-regulated the activity of SOD.

(3) Chronic obstructive pulmonary disease (COPD): is an inflammatory lung disease caused by harmful gases or particles. It features progressive airway remodeling and airflow obstruction; high incidence; and difficulty in treatment. In 2017, Chinese researcher Liu et al. prepared mouse models of COPD using smoke from cigarettes, and then injected hydrogen-rich saline into the mice. Results showed that airway inflammation and excessive secretion of bronchial mucosa were inhibited under the anti-apoptotic action. In the same year, China researcher Liu et al. prepared the same mouse models and treated them with hydrogen inhalation. It was found that the method improved symptoms of emphysema, chronic bronchitis, small airway remodeling and lung function decline. Also in the same year, Japan's Suzuki et al. reported that this protective effect was the result of hydrogen's anti-inflammatory and anti-oxidative effects.

6. Urinary system diseases

It is often difficult to avoid I/R injury during kidney transplantation. In particular, the kidney that comes from a donor with cardiac death usually has excessively long cold ischemia, leading to potential risk of lower kidney survival after transplantation. The main cause of kidney damage caused by I/R is ROS production. According to Chinese researcher Liu et al. in 2015, preserving rat kidneys with hydrogen-rich saline helped reduce renal cell death during storage and transportation; and mitigate I/R damage after transplantation. This is attributable to the mechanism by which DNA oxidative stress and overexpression of PPM-1 are inhibited. In 2015, Chinese researcher Wang et al. used the same preprocessing method and found that the ganglion cell activity of the kidney improved significantly, which directly led to higher efficiency of transplantation.

(1) Kidney damage: In 2009, Japanese researcher Nakashima-Kamimura et al. reported that both hydrogen inhalation and oral hydrogen water helped weaken cisplatin-induced renal toxicity without affecting the antitumor activity of tumor-bearing mice. A number of recent studies reveal that hydrogen inhalation and oral hydrogen relieve renal damage induced by iron (III) nitroacetate, dicarbonyl compounds, unilateral ureteral obstruction, spontaneous hypertension, glycerinum, infectious shock, acute pancreatitis and burns.

(2) Bladder pain: In 2013, Japanese researcher Matsumoto et al. reported that drinking hydrogen water alleviated the symptoms of interstitial cystitis and treated bladder pain syndrome.

7. Metabolic syndrome

(1) Obesity: Obesity has become the most common modern lifestyle-related disease. In 2011, Japanese researcher Kamimura et al. reported that for mice with spontaneous obesity db/db, drinking hydrogen-rich water helped reduce liver oxidative stress, significantly mitigate fatty liver induced by high-fat diet, and lower the levels of plasma glucose, insulin and triglyceride. The mechanism of action is to improve parameters of obesity and metabolism at the molecular level. This includes enhancing the expression of fibroblast growth factor (FGF21), which functions to strengthen fatty acid and glucose consumption.

(2) Hyperlipemia: It is one of the most common metabolic syndromes and is usually followed by atherosclerosis. In 2008, Japanese researcher Kajiyama et al. reported that patients with Type 2 diabetes, after taking oral hydrogen water for 8 weeks, had significantly lower LDL, urinary 8-isoprostanes, and insulin dosage. In 2012, Chinese scholar Zong et al. induced hyperlipemia in hamsters with high-fat diet, and gave the hamsters injections of hydrogen-rich saline. As a result, the hamsters had lower plasma LDL and better HDL functions. In 2013, Japanese researcher Nishimura et al. discovered that oral intake of fructosan facilitated the production of more hydrogen in the colon and reduced inflammation-induced oxidative stress in rats. In 2013, Chinese researcher Song et al. reported that patients with metabolic syndrome, after taking oral hydrogen for 8 weeks, had 39% more superoxide dismutase (SOD) in the urine, 43% less thiobarbituric acid

reactive substances (TBARS) in the urine, 8% more high density lipoprotein (HDL), and 13% lower ratio of total cholesterol/HDL.

(3) Diabetes: In 2009, Japanese researcher Suzuki et al. observed that oral acarbose induced the production of endogenous hydrogen, improved glucose tolerance, and reduced the risks of cardiovascular disease in patients with Type 2 diabetes. In 2013, Japanese researcher Amitani et al. reported the effects of hydrogen on two tumor cell lines (hydrogen involved in cell culture) and three mouse models of diabetes (oral or injection of hydrogen-rich water), suggesting that hydrogen delivered metabolic effects like insulin and can even be promising as a new alternative to the treatment of Type 1 diabetes.

8. Reproductive system diseases

(1) Testicular injury: It might be caused by radiotherapy and induce azoospermatism or infertility. For γ-ray induced rat models of testicular injury, hydrogen-rich saline has protective effects, leading to lower loss of germ cells in male rats and protection of sperm production. Studies of rat models have shown that hydrogen-rich saline prevents testicular damage caused by I/R and spinal cord hemisection. In 2014, Chinese researcher Li et al. reported that

long-term injection of hydrogen-rich saline reduced nicotine-induced oxidative stress damage in mouse testis.

(2) Erectile dysfunction: In 2013, Chinese researcher Fan et al. found that rat models of streptozotocin (STZ)-induced diabetes often showed erectile dysfunction, which were prevented through injection of hydrogen-rich saline.

(3) Ovarian injury: According to Chinese researcher Meng et al. in 2015, injection of hydrogen-rich saline helps reduce chemotherapy-induced ovarian injury in female rats.

(4) Eclampsia: It is the most serious stage of pregnancy-induced hypertension. Eclampsia refers to convulsions that cannot be explained by other reasons on the basis of pregnancy-induced hypertension. It may occur at different time points before, during, and after childbirth. In 2011, Chinese researcher Yang et al. reported that injection of hydrogen-rich saline prevented eclampsia in pregnant rats with hypertension, and had no impact on pregnant rats with normal blood pressure.

9. Sensory system

(1) Vision: Retinal ischemia is a common cause of visual impairment and blindness. At the cellular level, ischemic retinal damage is composed of a self-reinforcing destructive cascade, including neuronal depolarization, calcium influx, oxidative stress caused by energy failure, and glutamatergic stimulation. In 2010, Japanese researcher Oharazawa et al. reported that eye drops containing hydrogen mitigated retinal I/R injury caused by elevated intraocular pressure in rats, inhibited the increase of ·OH, reduced the number of retinal apoptotic cells, and prevented gliocyte activation accompanied by retina thinning. In 2012, Chinese researcher Wei et al. reported that the glutamate-induced excitotoxic retinal damage prepared in guinea pigs was relieved by eye drops containing hydrogen-rich saline. In 2014, Chinese researcher Sun et al. observed that in rat models of optic nerve injury, eye drops with hydrogen-rich saline increased the survival of retinal ganglion cells. In 2016, Chinese researcher Chen et al. discovered that eye drops containing hydrogen-rich saline had good therapeutic effects on retinal photoreceptor degeneration induced by N-methyl sub-N-nitrourea in rats. In 2011, Japanese researcher Kubota et al. reported that in mouse models of corneal alkali burn, hydrogen-rich saline eye drops with dissolved N-acetyl-L-cysteine alleviated oxidative stress on the surface of cornea and inhibited the formation of blood vessel clusters.

(2) Hearing: In 2014, Japanese researcher Kikkawa et al. reported that when the ear was exposed to a high-volume sound for a long time, hair cells suffered from over-stimulation. Consequently, ROS were generated, which in turn led to cell death. Hydrogen had the effects of protecting hair cells. In explant cultures of auditory tissues, H_2 attenuated oxidative stress induced by epirubicin and cisplatin, thereby protecting hair cells and preventing inner ear damage. A series of studies indicate that hydrogen inhalation, oral intake of hydrogen water, and injection of hydrogen-rich saline help protect inner ear hair cells and prevent hearing loss caused by noise in guinea pigs.

(3) Smell: In 2017, Chinese researcher Yu et al. reported that for guinea pig models of allergic rhinitis, injection of hydrogen-rich saline reduced eosinophilic granulocyte activation and oxidative stress on the surface of nasal mucosa; and relieved allergic rhinitis.

(4) Touch: In 2012, Chinese researcher Guo et al. reported that intraperitoneal injection of hydrogen-rich saline alleviated symptoms in rat models of UV-induced dermatitis and skin

damage. In 2014, Japanese researcher Watanabe et al. presented that hydrogen inhalation mitigated symptoms in γ-ray-induced models. In 2014, Chinese researcher Mei et al. obtained similar findings after the models were induced by X-rays and given subcutaneous injections of hydrogen-rich water.

In 2012, American researcher Ono et al. reported about four patients with acute erythematous skin disease accompanied by fever and/or pain. After given intravenous infusion of hydrogen-rich water, the patients' erythema and related symptoms subsided notably without recurrence. In 2013, Chinese researcher Zhao et al. reported that if intraperitoneal injection of hydrogen-rich saline was given to rats 10 minutes before skin flap transplantation, the flap survival rate and flap blood flow perfusion were significantly higher than the group with injections of normal saline. In 2015, Chinese researcher Liu et al. observed that the rats for whom the skin flap was washed with hydrogen-rich saline before skin flap transplantation had considerably higher flap survival rate and flap blood flow perfusion than those of the normal saline group. In 2015, Japanese researcher Ishibashi et al. reported about an experiment in which patients with psoriasis were treated with instillation of hydrogen-rich saline, hydrogen inhalation, and oral hydrogen water respectively. After treatment, all the three groups' psoriasis disappeared, indicating that the therapeutic effect was no different.

(5) Taste: The development of oral plaque biofilm is the main cause of oral diseases. According to a study by Japanese researcher Kasuyama et al. in 2011, flushing the oral cavity with hydrogen-rich water for rat models of periodontitis helped inhibit dental plaques formed by streptococcus and alleviate periodontitis symptoms. In 2017, another Japanese researcher Yoneda et al. reported that for rats on high-fat diet, long-term intake of hydrogen-rich water relieved oxidative stress on the gums and inhibited alveolar bone dissolution.

10. Musculoskeletal system diseases

(1) Joint diseases: In 2011, Japanese researcher Hanaoka et al. reported that hydrogen prevented

NO from being converted to $ONOO^-$; protected cartilage cells from oxidative stress, indirectly altered gene expression, and prevented or treated joint diseases. In 2014, Japanese researcher Ishibashi et al. revealed that in patients with rheumatoid arthritis, instillation of hydrogen-rich saline for 4 weeks helped significantly reduce the level of rheumatoid factor in the blood, while the saline control group showed increase in the rheumatoid factor. Osteoporosis of the jaw is a serious complication after radiotherapy for head and neck cancer. Most radiation-induced symptoms occur due to ROS produced from water radiation decomposition. In 2013, Chinese researcher Sun et al. reported that osteoporosis occurred to rats under simulated microgravity conditions. Injection of hydrogen-rich water reduced bone oxidative stress and bone loss. In 2016, Chinese researcher Huang et al. reported that intraperitoneal injection of hydrogen-rich saline had preventive effects on rabbit models of steroid-induced osteonecrosis. In 2017, Chinese researcher Li et al. reported similar findings in their experiments, and discovered that hydrogen prevented hormone-induced osteonecrosis in rabbits by suppressing oxidative damage, vascular injury and apoptosis.

(2) Muscular diseases: In 2011, Japanese researcher Ito et al. reported about the treatment for 5 cases of progressive muscular dystrophy (PMD), 4 cases of polymyositis/dermatomyositis (PM/DM), and 5 cases of mitochondrial myopathy (MM). The patients drank 1 liter of hydrogen-rich water per day for 12 weeks. It was found that hydrogen-rich water mitigated mitochondrial dysfunction and relieved PM/DM inflammation. In 2012,

Japanese researcher Aoki et al. reported that oral hydrogen-rich water for athletes with acute sport injuries helped accelerate the relief of muscle fatigue. In 2014, Chinese researcher Gu et al. reported that glycerinum induced rhabdomyolysis and acute kidney injury models in rats. However, if the models were injected intraperitoneally with hydrogen-rich saline for 7 consecutive days, remodeling would not be successful. In 2017, Japanese researcher Hasegawa et al. reported that for mdx mice with congenital myodystrophy, long-term consumption of hydrogen-rich water inhibited the attack of the disease.

11. Blood system diseases

(1) Acute graft-versus-host disease (GVHD): This is a fatal complication of hematopoietic stem cell transplantation, and usually occurs within a few months after transplantation. According to the findings by Chinese researcher Qian et al. in 2013, for mouse models of bone marrow transplantation that was incompatible with major histocompatibility complex (MHC), injection of hydrogen-rich saline before or after transplantation prevented the occurrence of GVHD and improved the mice's survival and clinical scores. In 2015, Chinese researcher Yuan et al. reported the same results, which verified that hydrogen-rich saline improves the survival rate of mice after bone marrow transplantation, facilitates the recovery of white blood cells, reduces serum cell levels, and reverses tissue damage.

(2) Chronic graft-versus-host disease (GVHD): It usually occurs months or years after transplantation, and is the most challenging complication of bone marrow transplantation. Inflammation and fibrosis play dominant roles in the pathogenesis of chronic GVHD. In 2017, Chinese researcher et al. prepared mouse models of chronic bone marrow transplantation, and gave long-term intraperitoneal injection of hydrogen-rich saline to the recipient mice. As a result, the symptoms of scleroderma caused by rejection were significantly alleviated, and graft rejection was slowed down.

(3) Aplastic anemia (AA): In 2013, Chinese researcher Zhao et al. prepared mouse models of AA and gave the mice intraperitoneal injections of hydrogen-rich saline. Afterwards, it was found that the mice's body weight and peripheral blood cell levels improved considerably; bone marrow microenvironment became markedly better; and tissue repair accelerated. Gradually, the treatment group had normal numbers of peripheral blood CD4+ and CD8+ cells as well as CD4/CD8 ratio; and lower levels of TNF, IFN and IL-6 in the serum.

In addition, oral hydrogen water has been found to enhance the blood alkalinity of healthy men; inhibit collagen-induced platelet aggregation in healthy human beings and rats; and improve serum antioxidant levels in purebred horses.

12. Infection

(1) Sepsis: It features severe inflammatory response to infection and is the leading cause of death for criticallyed patients. In 2014, Chinese researcher Xie et al. reported that zymosan was a substance derived from the walls of saccharomyces cerevisiae cells, and may induce a variety of inflammatory mediators, resulting in systemic inflammation. Inhalation of H_2 in 1 and 6 hours respectively after injection of zymosan significantly improved the 14-day survival rate in mice from 10% to 70%, and reduced liver and kidney function impairment. In 2016, Chinese researcher Zheng et al. made a summary of literatures on the protective effects of molecular hydrogen therapy on sepsis patients, and believed that the protective effects were manifested in basic vital signs, functions of organs (the brain, lung, liver, kidney, small intestine, etc.) and survival rate. Molecular hydrogen therapy significantly decreases the release of inflammatory

factors and oxidative stress damage, thereby reducing damage of sepsis to the functions of organs and improving survival rate.

(2) Sepsis complications: In 2018, Chinese researcher Dong et al. prepared mouse models of sepsis by means of cecal ligation. It was found that hydrogen inhalation improved the chance of 14-day survival to 100% for mice with moderate sepsis, and increased 7-day survival rate from 0% to 70% for mice with severe sepsis. Meanwhile, hydrogen inhalation resulted in less damage to the lung, liver and kidney in the mice with moderate and severe sepsis. The authors believed that the therapeutic mechanism was related to the regulation of mitochondrial function.

13. Neonatal diseases

(1) As an important cause of death in newborns and infants, neonatal encephalopathy often leads to autism, cerebral palsy, mental retardation, and other neurological and cognitive disorders. Perinatal asphyxia is an important cause of brain damage. Inflammation and oxidative stress are the main mechanisms resulting in hypoxia-ischemia and nerve cell apoptosis. In 2009, Chinese researcher Cai et al. prepared neonatal rat models of hypoxia-ischemia by ligation of the left common carotid artery. It was discovered that intraperitoneal injection of hydrogen-rich saline inhibited nerve cell apoptosis and improved neurological and neurobehavioral functions in the long term. In 2014, American researcher Dohi et al. presented that oral hydrogen-rich water for pregnant women mitigated brain damage caused by neonatal intrauterine ischemia.

(2) Anesthesia injury: Administration of anesthetics to immature brains may lead to pathological changes and long-term behavioral abnormalities. A basic study has indicated that exposure to

sevoflurane (SEV) induces abnormal behavior similar to autism. In 2017, Chinese researcher Shu et al. reported that combined use of hydrogen inhalation inhibited oxidative stress, nerve cell apoptosis and subsequent neurobehavioral defects caused by exposure to SEV; and reduced untoward effects from exposure to anesthetics in neonatal mice.

14. Cancer rehabilitation

An growing number of studies have indicated that ROS are involved in the formation, proliferation, angiogenesis, invasion, and distant metastasis of human tumor cells. Molecular hydrogen is being increasingly studied for its application to cancer treatment, prevention and rehabilitation. In 2009, Japanese researcher Nakashima-Kamimura et al. reported that cisplatin had great renal toxicity as an anticancer drug widely used in cancer treatment. After being given cisplatin and then hydrogen inhalation, experimental mice showed markedly less complications from cisplatin. Giving the mice oral hydrogen water after use of cisplatin created similar effects to hydrogen inhalation. According to the findings by Japan's Kitamura et al. in 2010, the group that consumed hydrogen water 7 days before injection of cisplatin showed significant improvement in both CT imaging results and serum creatinine clearance rate compared to the group that consumed normal drinking water. In 2011, Chinese researcher Kang et al. stated that cancer patients receiving radiotherapy often had fatigue and lower quality of life. However, the researchers discovered that drinking hydrogen-rich water exerted antioxidant effects, which led to considerably higher quality of life score during radiotherapy without affecting tumor treatment.

Maintaining Health and Treating "Disease to Come"

Modern lifestyle has greatly increased our body's oxidative stress, leaving the body in imbalance and with growing sub-health conditions. With safe and effective therapeutic effects on diseases, molecular hydrogen also helps cells and organs restore homeostasis, suppresses serious interference from adverse environment, and plays a unique role in maintaining the overall health of the human body.

Molecular hydrogen overcomes sub-health status and improves the quality of life from the following aspects:

1. Allaying fatigue

Chronic fatigue syndrome is a major recurring ailment that plagues modern city dwellers. It is often accompanied by increase in inflammatory cytokines (including TNF-α, IL-1β and IL-6) and Th-17 cell-mediated proinflammatory cytokines (such as IL-23 and IL-17); and by interruption of energy homeostasis. Continuous or repetitive exercise may over-consume glycogen and ATP. In 2018, South Korean researcher Ara et al. reported that feeding mice with hydrogen-rich water helped block the production of blood lactic acid; reduce the levels of urea nitrogen, ROS and inflammatory factors; boost the activity of lactate-degrading enzyme and glutathione peroxidase; strengthen the endurance capacity in tested animals; and promote hepatic glycogen synthesis, thereby exerting anti-fatigue effects.

2. Improving the quality of life

Everyone pursues a healthy and energetic life, that is, high quality of life. In healthy individuals, aging, poverty, and work stress all lead to more oxidative stress. Molecular hydrogen improves the quality of life by controlling antioxidant activity, preventing inflammation and eliminating discomforts. In 2017, Japanese researcher Mizuno et al. reported that drinking hydrogen-rich water regularly helped alleviate mood, anxiety and autonomic nerve dysfunction in adults; and improve their quality of life.

3. Cosmetic effect

In 2018, Chinese researcher Zhou et al. prepared skin wound models by applying electron beam irradiation to rats (simulating injury from ultraviolet rays such as sunlight); and gave the models ordinary water spray, hydrogen-rich water spray and oral hydrogen-rich water. After monitoring the time and speed of wound healing, the researchers found that hydrogen-rich water spray created similar results to oral hydrogen-rich water. The two groups of animals showed markedly lower levels of inflammatory factors secreted, and notably better wound healing than that of the ordinary water group. Moreover, the therapeutic effects were positively correlated with hydrogen concentration.

4. Anti-aging effect

Although aging is an unavoidable process, many wish to slow down aging as much as possible. Cell cycle arrest is a prerequisite for cell aging. Studies suggest that the genes p53 and p16 play important roles in the aging process. P53 is a stress protein that rapidly upregulates when cells are stimulated, prevents cell cycle progression, and accelerates cell aging. The protein p16 competitively binds with cell cycle-dependent kinases, preventing cell cycle progression and triggering aging. In 2016, Chinese researcher Zhu et al. reported that with age, p53 and p16 tend to show up-regulation. And H_2 can inhibit its expression and promote cell cycle progression.

In 2017, Indian researcher Ghoshal et al. presented a meaningful survey that measured H_2 concentration in the exhaled breaths of centenarians in order to reflect the H_2 generated by intestines. Results showed that H_2 concentration of the centenarians was significantly higher than that of the healthy control group and elderly patients with diabetes. The survey also suggested that higher levels of H_2 generated in the intestinal tract may prolong the lifetime of the elderly, and might be associated with diet and intestinal flora.

Safety of Hydrogen

According to a report by Guardian, the human body itself, from DNA to atoms, is a marvel of science. Strangely-behaving quantum particles, such as atoms and their constituents, make up a solid object like the human body. Every atom in the human body has a history of billions of years. Hydrogen, the most common element in the universe, is the main component of the human body and was generated during the Big Bang 13.7 billion years ago. Atoms heavier than hydrogen, such as carbon and oxygen, were formed in fixed stars between 7 billion and 12 billion years ago. They filled the whole universe

during explosion of stars. This means that the composition of the human body is very old. In fact, human beings are just stardust.

According to Taoism, heaven is nature; and man is a part of nature. Zhuangzi, an ancient Chinese philosopher, said: "Human beings appear because of nature. Nature appears also because of nature. There is a unity between man and nature." Laozi, another ancient Chinese philosopher, remarked: "Man follows the earth. The earth follows heaven. Heaven follows the Tao. Tao follows what is natural."

As a part of our body, hydrogen is probably a very important element. As early as 1969, *Production and Excretion of Human Hydrogen*, a paper by Levitt published in the New England Journal of Medicine, reported that H_2 was detected in the intestines of ten healthy subjects. Results showed that 0.06-29 ml H_2 was generated per minute in one test subject on empty stomach, with an average of 0.24 ml. After infusion of lactulose into the intestine, H_2 production increased rapidly, and reached 1.6 ml/minute on average at the highest level. After eating, H_2 production increased by 7 to 30 times. Under normal conditions, 99% H_2 generated in the colon. In a patient with excessive small intestinal flora, the small intestine produced notably more H_2. Around 14% H_2 produced in the body was excreted by the lungs; and the amount of excretion was positively correlated with the amount produced.

Therefore, hydrogen inhalation or other methods (oral hydrogen water, hydrogen bubble bath, and hydrogen saline infusion) offer a way to take molecular hydrogen into the body. But simply, it is a biological process that adapts to the living body and is a type of compensation for the body.

In 1980, Levitt published a paper in the New England Journal of Medicine (right side of Picture 3), criticizing the reports that deliberately introduced H_2 research into the hot air field. Levitt pointed out that H_2 production in the intestine is an integral part of human physiology. According to Levitt's calculation of the rate of H_2 production, the average daily H_2 production is 345.6 ml on average and 41,740 ml at the highest level. After eating, H_2 production is increased by 7 times to 292.18 liters at the highest level; and may rise by 30 times to 1252.8 liters. Currently, the hydrogen-oxygen nebulizer used for hydrogen inhalation has a gas flow rate of up to 3 liters per minute. And the gas produced contains 66% hydrogen and 34% oxygen. The actual inhalation in the lung is estimated to be 20%. The daily intake of hydrogen

is 47.52 liters if hydrogen inhalation continues for 2 hours, and reaches 95.4 liters if hydrogen inhalation continues for 4 hours. Both the volumes are much lower than the amount of hydrogen generated in the body after eating.

From an evolutionary perspective, it is not surprising that molecular hydrogen produces biological effects. In addition to its role in the origin of the universe, hydrogen is also involved in the origin of life and plays a positive role in the evolution of eukaryotes. In the evolution of millions of years, plants and animals develop reciprocal relationships with hydrogen-producing bacteria, which keeps molecular hydrogen in the eukaryotic system at a certain basic level. It is reasonable to assume that eukaryotes may have preserved the sensitivity to and dependence on molecular hydrogen during millions of years of evolution.

> *Picture 3. Levitt's papers published in the New England Journal of Medicine in 1969 & 1980 ("Production and Excretion of Hydrogen Gas in Man", 1969; and "Intestinal Gas Production—Recent Advances in Flatology", 1980).*

Summary

Is there a panacea for treating all diseases? In recent years, people favor two drugs: melbine and aspirin. The former is considered to be effective for both diabetes and cancer. The latter is considered to be an "evergreen tree" that has survived 120 years of wind and frost. It has preventive effects on cardiovascular disease and cancer.

At the European Society of Cardiology (ESC) annual meeting in 2018, Harvard University and Oxford University research teams presented the findings of two large-scale clinical trials: For middle-aged and elderly people with moderate cardiovascular risks and diabetes, aspirin brought basically no benefit to the prevention of cardiovascular disease and stroke. The papers were published in Lancet and New England Journal of Medicine.

Although melbine has not been confronted with such bad luck, the previous glory of aspirin seems to have been blown away by the wind. In the end, history is a long river that runs endlessly. Time has washed off the frivolous dregs and kept the shining essence. Perhaps, chemical drugs, which are never parts of the human body, will always be only "companions".

Molecular hydrogen is different. It is not a medicine, but represents natural and physiological existence that benefits people. It seems that hydrogen offers a preventive and therapeutic option for diseases.

— Xu Kecheng, Chen Jibing, Piao Xianghao, Shi Jianguo, Chen Xianming

References

1. Dole M, Wilson FR, Fife WP. Hyperbaric hydrogen therapy: a possible treatment for cancer. Science, 1975, 190(4210): 152-154.
2. Gharib B, Hanna S, Abdallahi OM, et al. Anti-inflammatory properties of molecular hydrogen: investigation on parasite-induced liver inflammation. C R Acad Sci III, 2001, 324(8): 719-724.
3. Ohsawa I, Ishikawa M, Takahashi K, et al. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med, 2007, 13(6): 688-694.
4. Ohta S. Recent progress toward hydrogen medicine: potential of molecular hydrogen for preventive and therapeutic applications. Curr Pharm Des, 2011, 17(22): 2241-2252.
5. Hayashida K, Sano M, Ohsawa I, et al. Inhalation of hydrogen gas reduces infarct size in the rat model of myocardial ischemia-reperfusion injury. Biochem Biophys Res Commun, 2008, 373(1): 30-35.

6. Yamamoto R, Homma K, Suzuki K. Hydrogen gas distribution in organs after inhalation: real-time monitoring of tissue hydrogen concentration in rat. Sci Rep, 2019, 9(1): 1255.
7. Liu H, Colavitti R, Rovira II, et al. Redox-dependent transcriptional regulation. Circ Res, 2005, 97(10): 967-974.
8. Salganik RI. The benefits and hazards of antioxidants: controlling apoptosis and other protective mechanisms in cancer patients and the human population. J Am Coll Nutr, 2001, 20(5 Suppl): 464S-475S; discussion 473S-475S.
9. Winterbourn CC. Biological reactivity and biomarkers of the neutrophil oxidant, hypochlorous acid. Toxicology, 2002, 181-182: 223-227.
10. Murad F. Discovery of some of the biological effects of nitric oxide and its role in cell signaling. Biosci Rep, 2004, 24(4-5): 452-474.
11. Bjelakovic G and Gluud C. Surviving antioxidant supplements. Journal of the National Cancer Institute, 2007, 99(10): 742-743.
12. Ge L, Yang M, Yang NN, et al. Molecular hydrogen: a preventive and therapeutic medical gas for various diseases. Oncotarget, 2017, 8(60): 102653-102673.
13. Kajimura M, Fukuda R, Bateman RM, et al. Interactions of multiple gas-transducing systems: hallmarks and uncertainties of CO, NO, and H_2S gas biology. Antioxid Redox Signal, 2010, 13(2): 157-192.
14. Szabo C. Hydrogen sulphide and its therapeutic potential. Nat Rev Drug Discov, 2007, 6(11): 917-935.
15. Abraini JH, Gardette-Chauffour MC, Martinez E, et al. Psychophysiological reactions in humans during an open sea dive to 500 m with a hydrogen-helium-oxygen mixture. J Appl Physiol (1985), 1994, 76(3): 1113-1118.
16. Fontanari P, Badier M, Guillot C, et al. Changes in maximal performance of inspiratory and skeletal muscles during and after the 7.1-MPa Hydra 10 record human dive. Eur J Appl Physiol, 2000, 81(4): 325-328.
17. Sano M, Suzuki H, Homma K, et al. Promising novel therapy with hydrogen gas for emergency and critical care medicine. Acute Med Surg, 2018, 5(2): 113-118.
18. Diakos CI, Charles KA, McMillan DC, et al. Cancer-related inflammation and treatment effectiveness. Lancet Oncol, 2014, 15(11): e493-503.
19. Ishibashi T. Molecular hydrogen: new antioxidant and anti-inflammatory therapy for rheumatoid arthritis and related diseases. Curr Pharm Des, 2013, 19(35): 6375-6381.
20. Zhang JY, Liu C, Zhou L, et al. A review of hydrogen as a new medical therapy. Hepatogastroenterology, 2012, 59(116): 1026-1032.
21. Elmore S. Apoptosis: a review of programmed cell death. Toxicol Pathol, 2007, 35(4): 495-516.
22. Du H, Sheng M, Wu L, et al. Hydrogen-Rich Saline Attenuates Acute Kidney Injury After Liver Transplantation via Activating p53-Mediated Autophagy. Transplantation, 2016, 100(3): 563-570.
23. Cai J, Kang Z, Liu WW, et al. Hydrogen therapy reduces apoptosis in neonatal hypoxia-ischemia rat model. Neurosci Lett, 2008, 441(2): 167-172.
24. Kawamura T, Huang CS, Tochigi N, et al. Inhaled hydrogen gas therapy for prevention of lung transplant-induced ischemia/reperfusion injury in rats. Transplantation, 2010, 90(12): 1344-1351.
25. Hong Y, Shao A, Wang J, et al. Neuroprotective effect of hydrogen-rich saline against neurologic damage and apoptosis in early brain injury following subarachnoid hemorrhage: possible role of the Akt/GSK3beta signaling pathway. PLoS One, 2014, 9(4): e96212.
26. Hou Z, Luo W, Sun X, et al. Hydrogen-rich saline protects against oxidative damage and cognitive deficits after mild traumatic brain injury. Brain Res Bull, 2012, 88(6): 560-565.
27. Nakai Y, Sato B, Ushiama S, et al. Hepatic oxidoreduction-related genes are upregulated by administration of hydrogen-saturated drinking water. Biosci Biotechnol Biochem, 2011, 75(4): 774-776.
28. Liu Q, Shen WF, Sun HY, et al. Hydrogen-rich saline protects against liver injury in rats with obstructive jaundice. Liver Int, 2010, 30(7): 958-968.

29. Xiao M, Zhu T, Wang T, et al. Hydrogen-rich saline reduces airway remodeling via inactivation of NF-kappaB in a murine model of asthma. Eur Rev Med Pharmacol Sci, 2013, 17(8): 1033-1043.
30. Nagata K, Nakashima-Kamimura N, Mikami T, et al. Consumption of molecular hydrogen prevents the stress-induced impairments in hippocampus-dependent learning tasks during chronic physical restraint in mice. Neuropsychopharmacology, 2009, 34(2): 501-508.
31. Manaenko A, Lekic T, Ma Q, et al. Hydrogen inhalation ameliorated mast cell-mediated brain injury after intracerebral hemorrhage in mice. Crit Care Med, 2013, 41(5): 1266-1275.
32. Zhuang Z, Sun XJ, Zhang X, et al. Nuclear factor-kappaB/Bcl-XL pathway is involved in the protective effect of hydrogen-rich saline on the brain following experimental subarachnoid hemorrhage in rabbits. J Neurosci Res, 2013, 91(12): 1599-1608.
33. Shao A, Wu H, Hong Y, et al. Hydrogen-Rich Saline Attenuated Subarachnoid Hemorrhage-Induced Early Brain Injury in Rats by Suppressing Inflammatory Response: Possible Involvement of NF-kappaB Pathway and NLRP3 Inflammasome. Mol Neurobiol, 2016, 53(5): 3462-3476.
34. Fu Y, Ito M, Fujita Y, et al. Molecular hydrogen is protective against 6-hydroxydopamine-induced nigrostriatal degeneration in a rat model of Parkinson's disease. Neurosci Lett, 2009, 453(2): 81-85.
35. Fujita K, Seike T, Yutsudo N, et al. Hydrogen in drinking water reduces dopaminergic neuronal loss in the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine mouse model of Parkinson's disease. PLoS One, 2009, 4(9): e7247.
36. Yoritaka A, Takanashi M, Hirayama M, et al. Pilot study of H(2) therapy in Parkinson's disease: a randomized double-blind placebo-controlled trial. Mov Disord, 2013, 28(6): 836-839.
37. Li J, Wang C, Zhang JH, et al. Hydrogen-rich saline improves memory function in a rat model of amyloid-beta-induced Alzheimer's disease by reduction of oxidative stress. Brain Res, 2010, 1328: 152-161.
38. Wang C, Li J, Liu Q, et al. Hydrogen-rich saline reduces oxidative stress and inflammation by inhibit of JNK and NF-kappaB activation in a rat model of amyloid-beta-induced Alzheimer's disease. Neurosci Lett, 2011, 491(2): 127-132.
39. Jing L, Wang Y, Zhao XM, et al. Cardioprotective Effect of Hydrogen-rich Saline on Isoproterenol-induced Myocardial Infarction in Rats. Heart Lung Circ, 2015, 24(6): 602-610.
40. Gao Y, Yang H, Fan Y, et al. Hydrogen-Rich Saline Attenuates Cardiac and Hepatic Injury in Doxorubicin Rat Model by Inhibiting Inflammation and Apoptosis. Mediators Inflamm, 2016, 2016: 1320365.
41. Huo TT, Zeng Y, Liu XN, et al. Hydrogen-rich saline improves survival and neurological outcome after cardiac arrest and cardiopulmonary resuscitation in rats. Anesth Analg, 2014, 119(2): 368-380.
42. Knight RJ, Dikman S, Liu H, et al. Cold ischemic injury accelerates the progression to chronic rejection in a rat cardiac allograft model. Transplantation, 1997, 64(8): 1102-1107.
43. Sun Q, Kang Z, Cai J, et al. Hydrogen-rich saline protects myocardium against ischemia/reperfusion injury in rats. Exp Biol Med (Maywood), 2009, 234(10): 1212-1219.
44. Zhang Y, Sun Q, He B, et al. Anti-inflammatory effect of hydrogen-rich saline in a rat model of regional myocardial ischemia and reperfusion. Int J Cardiol, 2011, 148(1): 91-95.
45. Fan Z, Gao Y, Huang Z, et al. Protective effect of hydrogen-rich saline on pressure overload-induced cardiac hypertrophy in rats: possible role of JAK-STAT signaling. BMC Cardiovasc Disord, 2018, 18(1): 32.
46. Ohsawa I, Nishimaki K, Yamagata K, et al. Consumption of hydrogen water prevents atherosclerosis in apolipoprotein E knockout mice. Biochem Biophys Res Commun, 2008, 377(4): 1195-1198.
47. Matsuoka H, Miyata S, Okumura N, et al. Hydrogen gas improves left ventricular hypertrophy in Dahl rat of salt-sensitive hypertension. Clin Exp Hypertens, 2018, 1-5.
48. Guan P, Lin XM, Yang SC, et al. Hydrogen gas reduces chronic intermittent hypoxia-induced hypertension by inhibiting sympathetic nerve activity and increasing vasodilator responses via the antioxidation. J Cell Biochem, 2019, 120(3): 3998-4008.

49. Kajiya M, Sato K, Silva MJ, et al. Hydrogen from intestinal bacteria is protective for Concanavalin A-induced hepatitis. Biochem Biophys Res Commun, 2009, 386(2): 316-321.
50. Xia C, Liu W, Zeng D, et al. Effect of hydrogen-rich water on oxidative stress, liver function, and viral load in patients with chronic hepatitis B. Clin Transl Sci, 2013, 6(5): 372-375.
51. Fukuda K, Asoh S, Ishikawa M, et al. Inhalation of hydrogen gas suppresses hepatic injury caused by ischemia/reperfusion through reducing oxidative stress. Biochem Biophys Res Commun, 2007, 361(3): 670-674.
52. Kang KM, Kang YN, Choi IB, et al. Effects of drinking hydrogen-rich water on the quality of life of patients treated with radiotherapy for liver tumors. Med Gas Res, 2011, 1(1): 11.
53. Kawai D, Takaki A, Nakatsuka A, et al. Hydrogen-rich water prevents progression of nonalcoholic steatohepatitis and accompanying hepatocarcinogenesis in mice. Hepatology, 2012, 56(3): 912-921.
54. Tan YC, Xie F, Zhang HL, et al. Hydrogen-rich saline attenuates postoperative liver failure after major hepatectomy in rats. Clin Res Hepatol Gastroenterol, 2014, 38(3): 337-345.
55. Liu Y, Yang L, Tao K, et al. Protective effects of hydrogen enriched saline on liver ischemia reperfusion injury by reducing oxidative stress and HMGB1 release. BMC Gastroenterol, 2014, 14: 12.
56. Lee PC, Yang YY, Huang CS, et al. Concomitant inhibition of oxidative stress and angiogenesis by chronic hydrogen-rich saline and N-acetylcysteine treatments improves systemic, splanchnic and hepatic hemodynamics of cirrhotic rats. Hepatol Res, 2015, 45(5): 578-588.
57. Liu Q, Li BS, Song YJ, et al. Hydrogen-rich saline protects against mitochondrial dysfunction and apoptosis in mice with obstructive jaundice. Mol Med Rep, 2016, 13(4): 3588-3596.
58. Shi Q, Chen C, Deng WH, et al. Hydrogen-Rich Saline Attenuates Acute Hepatic Injury in Acute Necrotizing Pancreatitis by Inhibiting Inflammation and Apoptosis, Involving JNK and p38 Mitogen-Activated Protein Kinase-dependent Reactive Oxygen Species. Pancreas, 2016, 45(10): 1424-1431.
59. Zhai X, Chen X, Lu J, et al. Hydrogen-rich saline improves nonalcoholic fatty liver disease by alleviating oxidative stress and activating hepatic PPARalpha and PPARgamma. Mol Med Rep, 2017, 15(3): 1305-1312.
60. Koyama Y, Taura K, Hatano E, et al. Effects of oral intake of hydrogen water on liver fibrogenesis in mice. Hepatol Res, 2014, 44(6): 663-677.
61. Zhang DQ, Feng H, Chen WC. Effects of hydrogen-rich saline on taurocholate-induced acute pancreatitis in rat. Evid Based Complement Alternat Med, 2013, 2013: 731932.
62. Luo ZL, Cheng L, Ren JD, et al. Hydrogen-rich saline protects against ischemia/reperfusion injury in grafts after pancreas transplantations by reducing oxidative stress in rats. Mediators Inflamm, 2015, 2015: 281985.
63. Levitt MD. Intestinal gas production—recent advances in flatology. N Engl J Med, 1980, 302(26): 1474-1475.
64. Liu X, Chen Z, Mao N, et al. The protective of hydrogen on stress-induced gastric ulceration. Int Immunopharmacol, 2012, 13(2): 197-203.
65. Zhang JY, Wu QF, Wan Y, et al. Protective role of hydrogen-rich water on aspirin-induced gastric mucosal damage in rats. World journal of gastroenterology: WJG, 2014, 20(6): 1614-1622.
66. McCarty MF. Potential ghrelin-mediated benefits and risks of hydrogen water. Med Hypotheses, 2015, 84(4): 350-355.
67. Ichihara M, Sobue S, Ito M, et al. Beneficial biological effects and the underlying mechanisms of molecular hydrogen — comprehensive review of 321 original articles. Med Gas Res, 2015, 5: 12.
68. Buchholz BM, Kaczorowski DJ, Sugimoto R, et al. Hydrogen inhalation ameliorates oxidative stress in transplantation induced intestinal graft injury. Am J Transplant, 2008, 8(10): 2015-2024.
69. Zheng X, Mao Y, Cai J, et al. Hydrogen-rich saline protects against intestinal ischemia/reperfusion injury in rats. Free Radic Res, 2009, 43(5): 478-484.

70. Chen H, Sun YP, Hu PF, et al. The effects of hydrogen-rich saline on the contractile and structural changes of intestine induced by ischemia-reperfusion in rats. J Surg Res, 2011, 167(2): 316-322.
71. Shigeta T, Sakamoto S, Li XK, et al. Luminal injection of hydrogen-rich solution attenuates intestinal ischemia-reperfusion injury in rats. Transplantation, 2015, 99(3): 500-507.
72. Zhou H, Fu Z, Wei Y, et al. Hydrogen inhalation decreases lung graft injury in brain-dead donor rats. J Heart Lung Transplant, 2013, 32(2): 251-258.
73. Zhai Y, Zhou X, Dai Q, et al. Hydrogen-rich saline ameliorates lung injury associated with cecal ligation and puncture-induced sepsis in rats. Exp Mol Pathol, 2015, 98(2): 268-276.
74. Sun Q, Cai J, Liu S, et al. Hydrogen-rich saline provides protection against hyperoxic lung injury. J Surg Res, 2011, 165(1): e43-49.
75. Muramatsu Y, Ito M, Oshima T, et al. Hydrogen-rich water ameliorates bronchopulmonary dysplasia (BPD) in newborn rats. Pediatr Pulmonol, 2016, 51(9): 928-935.
76. Liu S, Liu K, Sun Q, et al. Consumption of hydrogen water reduces paraquat-induced acute lung injury in rats. J Biomed Biotechnol, 2011, 2011: 305086.
77. Wang Y, Jing L, Zhao XM, et al. Protective effects of hydrogen-rich saline on monocrotaline-induced pulmonary hypertension in a rat model. Respir Res, 2011, 12: 26.
78. Fang Y, Fu XJ, Gu C, et al. Hydrogen-rich saline protects against acute lung injury induced by extensive burn in rat model. J Burn Care Res, 2011, 32(3): e82-91.
79. Liu Z, Geng W, Jiang C, et al. Hydrogen-rich saline inhibits tobacco smoke-induced chronic obstructive pulmonary disease by alleviating airway inflammation and mucus hypersecretion in rats. Exp Biol Med (Maywood), 2017, 242(15): 1534-1541.
80. Liu X, Ma C, Wang X, et al. Hydrogen coadministration slows the development of COPD-like lung disease in a cigarette smoke-induced rat model. Int J Chron Obstruct Pulmon Dis, 2017, 12: 1309-1324.
81. Suzuki Y, Sato T, Sugimoto M, et al. Hydrogen-rich pure water prevents cigarette smoke-induced pulmonary emphysema in SMP30 knockout mice. Biochem Biophys Res Commun, 2017, 492(1): 74-81.
82. Sun L, Dutta RK, Xie P, et al. Myo-Inositol oxygenase overexpression accentuates generation of reactive oxygen species and exacerbates cellular injury following high glucose ambience: a new mechanism relevant to the pathogenesis of diabetic nephropathy. J Biol Chem, 2016, 291(11): 5688-5707.
83. Liu H, Hua N, Xie K, et al. Hydrogen-rich saline reduces cell death through inhibition of DNA oxidative stress and overactivation of poly (ADP-ribose) polymerase-1 in retinal ischemia-reperfusion injury. Mol Med Rep, 2015, 12(2): 2495-2502.
84. Wang R, Wu J, Chen Z, et al. Postconditioning with inhaled hydrogen promotes survival of retinal ganglion cells in a rat model of retinal ischemia/reperfusion injury. Brain Res, 2016, 1632: 82-90.
85. Nakashima-Kamimura N, Mori T, Ohsawa I, et al. Molecular hydrogen alleviates nephrotoxicity induced by an anti-cancer drug cisplatin without compromising anti-tumor activity in mice. Cancer Chemother Pharmacol, 2009, 64(4): 753-761.
86. Li FY, Zhu SX, Wang ZP, et al. Consumption of hydrogen-rich water protects against ferric nitrilotriacetate-induced nephrotoxicity and early tumor promotional events in rats. Food Chem Toxicol, 2013, 61: 248-254.
87. Katakura M, Hashimoto M, Tanabe Y, et al. Hydrogen-rich water inhibits glucose and alpha,beta-dicarbonyl compound-induced reactive oxygen species production in the SHR.Cg-Leprcp/NDmcr rat kidney. Med Gas Res, 2012, 2(1): 18.
88. Xu B, Zhang YB, Li ZZ, et al. Hydrogen-rich saline ameliorates renal injury induced by unilateral ureteral obstruction in rats. Int Immunopharmacol, 2013, 17(2): 447-452.
89. Xin HG, Zhang BB, Wu ZQ, et al. Consumption of hydrogen-rich water alleviates renal injury in spontaneous hypertensive rats. Mol Cell Biochem, 2014, 392(1-2): 117-124.
90. Gu H, Yang M, Zhao A, et al. Pretreatment with hydrogen-rich saline reduces the damage caused by glycerol-induced rhabdomyolysis and acute kidney injury in rats. J Surg Res, 2014, 188(1): 243-249.
91. Liu W, Dong XS, Sun YQ, et al. A novel fluid resuscitation protocol: provide more protection on acute kidney injury during septic shock in rats. Int J Clin Exp Med, 2017, 7(4): 919-926.

92. Shi Q, Liao KS, Zhao KL, et al. Hydrogen-rich saline attenuates acute renal injury in sodium taurocholate-induced severe acute pancreatitis by inhibiting ROS and NF-kappaB pathway. Mediators Inflamm, 2015, 2015: 685043.
93. Guo SX, Fang Z, You CG, et al. Effects of hydrogen-rich saline on early acute kidney injury in severely burned rats by suppressing oxidative stress induced apoptosis and inflammation. J Transl Med, 2015, 13: 183.
94. Matsumoto S, Ueda T, Kakizaki H. Effect of supplementation with hydrogen-rich water in patients with interstitial cystitis/painful bladder syndrome. Urology, 2013, 81(2): 226-230.
95. Kamimura N, Nishimaki K, Ohsawa I, et al. Molecular hydrogen improves obesity and diabetes by inducing hepatic FGF21 and stimulating energy metabolism in db/db mice. Obesity (Silver Spring), 2011, 19(7): 1396-1403.
96. Kajiyama S, Hasegawa G, Asano M, et al. Supplementation of hydrogen-rich water improves lipid and glucose metabolism in patients with type 2 diabetes or impaired glucose tolerance. Nutr Res, 2008, 28(3): 137-143.
97. Zong C, Song G, Yao S, et al. Administration of hydrogen-saturated saline decreases plasma low-density lipoprotein cholesterol levels and improves high-density lipoprotein function in high-fat diet-fed hamsters. Metabolism, 2012, 61(6): 794-800.
98. Nishimura N, Tanabe H, Adachi M, et al. Colonic hydrogen generated from fructan diffuses into the abdominal cavity and reduces adipose mRNA abundance of cytokines in rats. J Nutr, 2013, 143(12): 1943-1949.
99. Song G, Li M, Sang H, et al. Hydrogen-rich water decreases serum LDL-cholesterol levels and improves HDL function in patients with potential metabolic syndrome. J Lipid Res, 2013, 54(7): 1884-1893.
100. Suzuki Y, Sano M, Hayashida K, et al. Are the effects of alpha-glucosidase inhibitors on cardiovascular events related to elevated levels of hydrogen gas in the gastrointestinal tract? FEBS Lett, 2009, 583(13): 2157-2159.
101. Amitani H, Asakawa A, Cheng K, et al. Hydrogen improves glycemic control in type1 diabetic animal model by promoting glucose uptake into skeletal muscle. PLoS One, 2013, 8(1): e53913.
102. Colpi GM, Contalbi GF, Nerva F, et al. Testicular function following chemo-radiotherapy. Eur J Obstet Gynecol Reprod Biol, 2004, 113 Suppl 1: S2-6.
103. Chuai Y, Shen J, Qian L, et al. Hydrogen-rich saline protects spermatogenesis and hematopoiesis in irradiated BALB/c mice. Med Sci Monit, 2012, 18(3): BR89-94.
104. Chuai Y, Gao F, Li B, et al. Hydrogen-rich saline attenuates radiation-induced male germ cell loss in mice through reducing hydroxyl radicals. Biochem J, 2012, 442(1): 49-56.
105. Jiang Z, Xu B, Yang M, et al. Protection by hydrogen against gamma ray-induced testicular damage in rats. Basic Clin Pharmacol Toxicol, 2013, 112(3): 186-191.
106. Jiang D, Wu D, Zhang Y, et al. Protective effects of hydrogen rich saline solution on experimental testicular ischemia-reperfusion injury in rats. J Urol, 2012, 187(6): 2249-2253.
107. Ge L, Wei LH, Du CQ, et al. Hydrogen-rich saline attenuates spinal cord hemisection-induced testicular injury in rats. Oncotarget, 2017, 8(26): 42314-42331.
108. Li S, Lu D, Zhang Y. Long-term treatment of hydrogen-rich saline abates testicular oxidative stress induced by nicotine in mice. J Assist Reprod Genet, 2014, 31(1): 109-114.
109. Fan M, Xu X, He X, et al. Protective effects of hydrogen-rich saline against erectile dysfunction in a streptozotocin induced diabetic rat model. J Urol, 2013, 190(1): 350-356.
110. Meng X, Chen H, Wang G, et al. Hydrogen-rich saline attenuates chemotherapy-induced ovarian injury via regulation of oxidative stress. Exp Ther Med, 2015, 10(6): 2277-2282.
111. Yang X, Guo L, Sun X, et al. Protective effects of hydrogen-rich saline in preeclampsia rat model. Placenta, 2011, 32(9): 681-686.
112. Oharazawa H, Igarashi T, Yokota T, et al. Protection of the retina by rapid diffusion of hydrogen: administration of hydrogen-loaded eye drops in retinal ischemia-reperfusion injury. Invest Ophthalmol Vis Sci, 2010, 51(1): 487-492.
113. Wei L, Ge L, Qin S, et al. Hydrogen-rich saline protects retina against glutamate-induced excitotoxic injury in guinea pig. Exp Eye Res, 2012, 94(1): 117-127.
114. Sun JC, Xu T, Zuo Q, et al. Hydrogen-rich saline promotes survival of retinal ganglion cells in a rat model of optic nerve crush. PLoS One, 2014, 9(6): e99299.

115. Chen T, Tao Y, Yan W, et al. Protective effects of hydrogen-rich saline against N-methyl-N-nitrosourea-induced photoreceptor degeneration. Exp Eye Res, 2016, 148: 65-73.
116. Kubota M, Shimmura S, Kubota S, et al. Hydrogen and N-acetyl-L-cysteine rescue oxidative stress-induced angiogenesis in a mouse corneal alkali-burn model. Invest Ophthalmol Vis Sci, 2011, 52(1): 427-433.
117. Kikkawa YS, Nakagawa T, Taniguchi M, et al. Hydrogen protects auditory hair cells from cisplatin-induced free radicals. Neurosci Lett, 2014, 579: 125-129.
118. Taura A, Kikkawa YS, Nakagawa T, et al. Hydrogen protects vestibular hair cells from free radicals. Acta Otolaryngol Suppl, 2010, 563: 95-100.
119. Lin Y, Kashio A, Sakamoto T, et al. Hydrogen in drinking water attenuates noise-induced hearing loss in guinea pigs. Neurosci Lett, 2011, 487(1): 12-16.
120. Chen L, Yu N, Lu Y, et al. Hydrogen-saturated saline protects intensive narrow band noise-induced hearing loss in guinea pigs through an antioxidant effect. PLoS One, 2014, 9(6): e100774.
121. Kurioka T, Matsunobu T, Satoh Y, et al. Inhaled hydrogen gas therapy for prevention of noise-induced hearing loss through reducing reactive oxygen species. Neurosci Lett, 2014, 89: 69-74.
122. Yu S, Zhao C, Che N, et al. Hydrogen-rich saline attenuates eosinophil activation in a guinea pig model of allergic rhinitis via reducing oxidative stress. J Inflamm (Lond), 2017, 14: 1.
123. Guo Z, Zhou B, Li W, et al. Hydrogen-rich saline protects against ultraviolet B radiation injury in rats. J Biomed Res, 2012, 26(5): 365-371.
124. Watanabe S, Fujita M, Ishihara M, et al. Protective effect of inhalation of hydrogen gas on radiation-induced dermatitis and skin injury in rats. J Radiat Res, 2014, 55(6): 1107-1113.
125. Mei K, Zhao S, Qian L, et al. Hydrogen protects rats from dermatitis caused by local radiation. J Dermatolog Treat, 2014, 25(2): 182-188.
126. Ono H, Nishijima Y, Adachi N, et al. Hydrogen(H_2) treatment for acute erythematous skin diseases. A report of 4 patients with safety data and a non-controlled feasibility study with H_2 concentration measurement on two volunteers. Med Gas Res, 2012, 2(1): 14.
127. Zhao L, Wang YB, Qin SR, et al. Protective effect of hydrogen-rich saline on ischemia/reperfusion injury in rat skin flap. J Zhejiang Univ Sci B, 2013, 14(5): 382-391.
128. Liu YQ, Liu YF, Ma XM, et al. Hydrogen-rich saline attenuates skin ischemia/reperfusion induced apoptosis via regulating Bax/Bcl-2 ratio and ASK-1/JNK pathway. J Plast Reconstr Aesthet Surg, 2015, 68(7): e147-156.
129. Ishibashi T, Ichikawa M, Sato B, et al. Improvement of psoriasis-associated arthritis and skin lesions by treatment with molecular hydrogen: A report of three cases. Mol Med Rep, 2015, 12(2): 2757-2764.
130. Kasuyama K, Tomofuji T, Ekuni D, et al. Hydrogen-rich water attenuates experimental periodontitis in a rat model. J Clin Periodontol, 2011, 38(12): 1085-1090.
131. Yoneda T, Tomofuji T, Kunitomo M, et al. Preventive Effects of Drinking Hydrogen-Rich Water on Gingival Oxidative Stress and Alveolar Bone Resorption in Rats Fed a High-Fat Diet. Nutrients, 2017, 9(1).
132. Hanaoka T, Kamimura N, Yokota T, et al. Molecular hydrogen protects chondrocytes from oxidative stress and indirectly alters gene expressions through reducing peroxynitrite derived from nitric oxide. Med Gas Res, 2011, 1(1): 18.
133. Ishibashi T, Sato B, Shibata S, et al. Therapeutic efficacy of infused molecular hydrogen in saline on rheumatoid arthritis: a randomized, double-blind, placebo-controlled pilot study. Int Immunopharmacol, 2014, 21(2): 468-473.
134. Sun Y, Shuang F, Chen DM, et al. Treatment of hydrogen molecule abates oxidative stress and alleviates bone loss induced by modeled microgravity in rats. Osteoporos Int, 2013, 24(3): 969-978.
135. Huang SL, Jiao J, Yan HW. Hydrogen-rich saline attenuates steroid-associated femoral head necrosis through inhibition of oxidative stress in a rabbit model. Exp Ther Med, 2016, 11(1): 177-182.

136. Li J, Ge Z, Fan L, et al. Protective effects of molecular hydrogen on steroid-induced osteonecrosis in rabbits via reducing oxidative stress and apoptosis. BMC Musculoskelet Disord, 2017, 18(1): 58.
137. Ito M, Ibi T, Sahashi K, et al. Open-label trial and randomized, double-blind, placebo-controlled, crossover trial of hydrogen-enriched water for mitochondrial and inflammatory myopathies. Med Gas Res, 2011, 1(1): 24.
138. Aoki K, Nakao A, Adachi T, et al. Pilot study: Effects of drinking hydrogen-rich water on muscle fatigue caused by acute exercise in elite athletes. Med Gas Res, 2012, 2: 12.
139. Hasegawa S, Ito M, Fukami M, et al. Molecular hydrogen alleviates motor deficits and muscle degeneration in mdx mice. Redox Rep, 2017, 22(1): 26-34.
140. Qian L, Mei K, Shen J, et al. Administration of hydrogen-rich saline protects mice from lethal acute graft-versus-host disease (aGVHD). Transplantation, 2013, 95(5): 658-662.
141. Yuan L, Chen X, Qian L, et al. Administration of hydrogen-rich saline in mice with allogenic hematopoietic stem-cell transplantation. Med Sci Monit, 2015, 21: 749-754.
142. Qian L, Liu X, Shen J, et al. Therapeutic effects of hydrogen on chronic graft-versus-host disease. J Cell Mol Med, 2017, 21(10): 2627-2630.
143. Zhao S, Mei K, Qian L, et al. Therapeutic effects of hydrogen-rich solution on aplastic anemia in vivo. Cell Physiol Biochem, 2013, 32(3): 549-560.
144. Ostojic SM. Serum alkalinization and hydrogen-rich water in healthy men. Mayo Clin Proc, 2012, 87(5): 501-502.
145. Ostojic SM and Stojanovic MD. Hydrogen-rich water affected blood alkalinity in physically active men. Res Sports Med, 2014, 22(1): 49-60.
146. Takeuchi S, Wada K, Nagatani K, et al. Hydrogen may inhibit collagen-induced platelet aggregation: an ex vivo and in vivo study. Intern Med, 2012, 51(11): 1309-1313.
147. Tsubone H, Hanafusa M, Endo M, et al. Effect of Treadmill Exercise and Hydrogen-rich Water Intake on Serum Oxidative and Anti-oxidative Metabolites in Serum of Thoroughbred Horses. J Equine Sci, 2013, 24(1): 1-8.
148. Xie K, Liu L, Yu Y, et al. Hydrogen gas presents a promising therapeutic strategy for sepsis. Biomed Res Int, 2014, 2014: 807635.
149. Zheng Y and Zhu D. Molecular Hydrogen Therapy Ameliorates Organ Damage Induced by Sepsis. Oxid Med Cell Longev, 2016, 2016: 5806057.
150. Dong A, Yu Y, Wang Y, et al. Protective effects of hydrogen gas against sepsis-induced acute lung injury via regulation of mitochondrial function and dynamics. Int Immunopharmacol, 2018, 65: 366-372.
151. Cai J, Kang Z, Liu K, et al. Neuroprotective effects of hydrogen saline in neonatal hypoxia-ischemia rat model. Brain Res, 2009, 1256: 129-137.
152. Dohi K, Kraemer BC, Erickson MA, et al. Molecular hydrogen in drinking water protects against neurodegenerative changes induced by traumatic brain injury. PLoS One, 2014, 9(9): e108034.
153. Houck CS and Vinson AE. Anaesthetic considerations for surgery in newborns. Arch Dis Child Fetal Neonatal Ed, 2017, 102(4): F359-F363.
154. Chung W, Park S, Hong J, et al. Sevoflurane exposure during the neonatal period induces long-term memory impairment but not autism-like behaviors. Paediatr Anaesth, 2015, 25(10): 1033-1045.
155. Shi Y, Wang G, Li J, et al. Hydrogen gas attenuates sevoflurane neurotoxicity through inhibiting nuclear factor kappa-light-chain-enhancer of activated B cells signaling and proinflammatory cytokine release in neonatal rats. Neuroreport, 2017, 28(17): 1170-1175.
156. Gunther S, Ruhe C, Derikito MG, et al. Polyphenols prevent cell shedding from mouse mammary cancer spheroids and inhibit cancer cell invasion in confrontation cultures derived from embryonic stem cells. Cancer Lett, 2007, 250(1): 25-35.
157. Kitamura A, Kobayashi S, Matsushita T, et al. Experimental verification of protective effect of hydrogen-rich water against cisplatin-induced nephrotoxicity in rats using dynamic contrast-enhanced CT. Br J Radiol, 2010, 83(990): 509-514.
158. Roerink ME, Buckland M, Lloyd AR, et al. Cytokine signature in chronic fatigue syndrome. Proceedings of the National Academy of Sciences of the United States of America, 2017, 114(45): E9435.

159. Ara J, Fadriquela A, Ahmed MF, et al. Hydrogen Water Drinking Exerts Antifatigue Effects in Chronic Forced Swimming Mice via Antioxidative and Anti-Inflammatory Activities. Biomed Res Int, 2018, 2018: 2571269.
160. Mizuno K, Sasaki AT, Ebisu K, et al. Hydrogen-rich water for improvements of mood, anxiety, and autonomic nerve function in daily life. Med Gas Res, 2017, 7(4): 247-255.
161. Zhou P, Lin B, Wang P, et al. The healing effect of hydrogen-rich water on acute radiation-induced skin injury in rats. J Radiat Res, 2019, 60(1): 17-22.
162. Al-Khalaf HH, Nallar SC, Kalvakolanu DV, et al. p16(INK4A) enhances the transcriptional and the apoptotic functions of p53 through DNA-dependent interaction. Mol Carcinog, 2017, 56(7): 1687-1702.
163. Carrasco-Garcia E, Moreno M, Moreno-Cugnon L, et al. Increased Arf/p53 activity in stem cells, aging and cancer. Aging Cell, 2017, 16(2): 219-225.
164. LaPak KM and Burd CE. The molecular balancing act of p16(INK4a) in cancer and aging. Molecular cancer research: MCR, 2014, 12(2): 167-183.
165. Zhu Y, Liao W, Niu L, et al. Nitric oxide is involved in hydrogen gas-induced cell cycle activation during adventitious root formation in cucumber. BMC Plant Biol, 2016, 16(1): 146.
166. Ghoshal UC, Shukla R, Ghoshal U. Small Intestinal Bacterial Overgrowth and Irritable Bowel Syndrome: A Bridge between Functional Organic Dichotomy. Gut Liver, 2017, 11(2): 196-208.
167. Levitt MD. Production and excretion of hydrogen gas in man. N Engl J Med, 1969, 281(3): 122-127.
168. Gaziano JM, Brotons C, Coppolecchia R, et al. Use of aspirin to reduce risk of initial vascular events in patients at moderate risk of cardiovascular disease (ARRIVE): a randomised, double-blind, placebo-controlled trial. Lancet, 2018, 392(10152): 1036-1046.
169. Bowman L, Mafham M, Wallendszus K, et al. Effects of Aspirin for Primary Prevention in Persons with Diabetes Mellitus. N Engl J Med, 2018, 379(16): 1529-1539.

China's Way of Cancer Control and Hydrogen Cancer Control

Key points:

- Cancer has become a global challenge. New treatments have limited effects on inhibiting disseminated or metastatic cancer and prolonging lifetime. Breakthrough in concepts is required to improve the serious landscape of cancer treatment.
- Based on the characteristics of eastern and western thinking, Academician Tang Zhaoyou proposed to combine cancer elimination and transformation methods during cancer treatment. The strategy is of Chinese characteristics and integrates both Chinese and western medicine. It advocates cancer control rather than cancer elimination, for cancer control is about both eliminating and transforming cancer.
- Like a double-edged sword, the elimination approach may cause accelerated progression of residual cancer, cancer stem cell resistance suppression, immunosuppression and other issues. The transformation approach involves transforming cancer cells themselves and their surrounding microenvironment as well as improving the anti-cancer capacity of the entire body.
- Molecular hydrogen inhibits the formation, proliferation, invasion and metastasis of cancer cells. In the practice of Chinese-style cancer control, molecular hydrogen truly plays the role of transforming cancer cells, microenvironment and the entire body.

Serious Challenge

Cancer has become the most primary disease threatening human beings. Dealing with cancer has become a global challenge. The current status of cancer treatment is in a mixed contradiction, just as British writer Dickens said, "It was the best of times, it was the worst of times."

In a 2015 article titled *Timeline: Chemotherapy and the War on Cancer*, American researchers Chabner and Roberts talked about cancer treatment: "60% of lung cancer patients die within a year. Only 15% can survive for 5 years." According to the article, after 1975, breast cancer mortality has decreased; but if metastasis occurs, only 27% of patients can survive for 5 years. The article also stated that pancreatic cancer is a catastrophic cancer, with no change in mortality since 1975. And its 5-year survival rate is lower than 5%.

Although the concept of precision medicine is appealing to patients and institutions that support cancer research, "Precision treatments have not brought benefits to a majority of patients.", as stated in a paper by American researcher Prasad in Nature in 2016. Another paper by Canadian researchers Tannock and Hickman was published in New England Journal of Medicine. The paper pointed out that precision drugs are eventually found for only 3% to 13% of patients; and that only a small percentage (30%) of the patients who use matched drugs benefit from the drugs. According to

the paper, patients should be given a clear message: individualized treatment of tumor has not been proven with substantial effects.

In 2017, Annals of Oncology published a latest study from the UK Cancer Drugs Fund. It stated, "We conclude the Cancer Drugs Fund has not delivered meaningful value to patients or society. From 2010 when it started, to 2016 when it closed, the Cancer Drugs Fund cost the UK taxpayer a total of £1.27 billion, the equivalent of one year's total spend on all cancer drugs in the NHS." In the study, researchers examined 29 drugs available through the fund for 47 different cancer indications. They concluded that the drugs showed no definitive therapeutic effect on a majority of (62%) cancer patients. Of the 47 CDF approved indications, only 18 (38%) reported a statistically significant overall survival benefit, with an overall median survival of 3.1 months. However, this extended 3.1 months of life were full of toxic side effects of drugs. In the face of the challenges of cancer treatment, it is necessary to make breakthroughs in concepts and develop new thinking and strategies in order to improve the serious situation of cancer treatment.

China's Way of Cancer Control

The goal of cancer treatment is patients' survival, that is, enable patients to survive, not to live for a few more months but for a few or many years with high quality and happiness. That is the top priority of cancer treatment.

Richard Eugene Nisbet, a famous American cultural psychologist, believes that the development of eastern and western cultures has their own trajectories. Western civilization is based on ancient Greek traditions, and features Aristotle's logic and analytical thinking. Eastern culture, represented by China, is based on oriental cultural traditions deeply influenced by Confucianism and Taoism. Regarding the way of thinking, eastern culture is characterized by bialectical and holistic thinking. Approaching from the characteristics of eastern and western thinking, academician Tang Zhaoyou, a distinguished Chinese oncologist, proposed Chinese way of cancer control that integrates Chinese and western medicine in his book *Combined Use of Elimination and Reforming Methods — Academician's New Perspectives on Cancer Treatment* (Shanghai Scientific & Technical Publishers, 2011).

According to Tang Zhaoyou, the key is not cancer elimination but cancer control, which is aimed not only at eliminating but also transforming cancer. Only by maximally eliminating the tumor can the body be effectively preserved. Surgery, ablation (cryo, ablation radiofrequency,

irreversible electroporation), radiotherapy, and chemotherapy are all methods of elimination. For early-stage non-metastatic tumors, surgery is the most important means among elimination methods to achieve radical cure. However, for tumors in progressive and especially metastatic stages, elimination methods are double-edged swords and may lead to the following issues:

(1) Cancer metastasis may occur in the early stage. Almost all elimination methods accelerate the metastasis and development of cancer.

(2) Cancer cells originate from the division of cancer stem cells, which are not sensitive to standard treatments. Even if chemotherapy achieves clinical remission, latent cancer stem cells remain, making recurrence inevitable and leading to drug resistance in many cases.

(3) Cancer is constantly evolving. After chemotherapy or radiotherapy eliminates sensitive cancer cells, the original insensitive cancer cells gain room for development and reversible growth, accelerating cancer progression.

(4) Chemotherapy or radiotherapy may cause cancer cells to mutate again and form "outliers", which are not only resistant but also more malignant.

(5) Chemotherapy and radiotherapy inhibit the body's immune function and weaken immune monitoring.

In the past more than 100 years, cancer treatment has been based on pathology; focused on eliminating tumors while ignoring patients' actual conditions; and neglecting the human body's own capacity to fight cancer. As a result, there has been limited progress in the extension of lifetime and improvement of survival rate.

China's way of cancer control is a concept that advocates combined use of cancer elimination and transformation methods. Results of cancer elimination are shown in the number of visible tumors in CT, ultrasound and MRI images; and indicate local removal of cancer. However, cancer transformation measures applied after cancer elimination determine the foundation of cancer presence and root of recurrence. Cancer transformation strategies involve transforming cancer cells themselves and their microenvironment as well as improving the body's capacity to fight cancer.

1. Transforming cancer cells

(1) The so called induced differentiation is a therapy that helps transform evil cancer cells and bring them back to the path of differentiation.

(2) Cancer cells can be induced to restrict and fight against each other. They compete with each other. The harsh treatment with high-dose chemotherapeutic drugs eliminates both sensitive cells and cancer cell "competitors". Short-course adaptation therapy is designed to counteract this.

(3) It is necessary to weaken the malignancy of cancer cells. Usually, cancer cells are less malignant in the early stages, and become increasingly malignant in later stages. This is particularly true in the case of cancer cells that have been through chemotherapy and radiotherapy.

2. Transforming the microenvironment

Inflammatory cells and toxic ROS in the microenvironment are important factors that cause oncogene instability as follows:

(1) Tumor-inhibiting T lymphocytes in tumor tissues are inhibited by regulatory T cells and lose their functions.

(2) Tumor vascular endothelial cells are incomplete or absent, and extracellular interstitial fluid increases, causing tumor interstitial hypertension.

(3) Tumor cells have increased demand for substances such as oxygen and glucose, causing tumor tissue hypoxia and anaerobic glycolysis that leads to release of lactic acid in large volumes. As a result, tumor tissues become acidic inside.

These factors promote the growth of cancer cells. The tumor microenvironment can be improved by eliminating inflammation and toxic ROS, and relieving hypoxia.

3. Transforming the whole body

Cancer is a systemic disease. Transforming the body involves maintaining and improving functions

of the nervous, immune, endocrine and metabolic systems. The key to transform the body's cancer-fighting capability is to strengthen its immune function.

Hydrogen Cancer Control Follows China's Way of Cancer Control

In his new book, *Cancer Control Rather Than Anti-Cancer Warfare — 'On Protracted War' and Cancer Prevention and Control Strategy* (Shanghai Scientific and Technical Publishers, 2018), Tang Zhaoyou proposed cancer control as a systematic project with the following main content:

(1) Internal environment stability: requires multi-system interventions.

(2) External environment imbalance: requires efforts to remove environmental factors that cause cancer.

(3) In-vivo disorder: requires measures to not only eliminate cancer cell but also transform cancer cells, the microenvironment and body.

(4) Cancer caused by multiple factors: requires measures to eliminate carcinogenic factors in many ways.

(5) Cancer that involves multiple genes: requires treatments that target at multiple genes, and comprehensive treatments.

(6) Cancer formed in multiple stages: requires appropriate measures from the early stage to advanced stage of cancer.

(7) Systemic disease: requires treatments that are focused on the whole body and maintaining the function of each organ system.

(8) Chronic disease: requires treatments for the whole lifetime and great attention to lifestyle.

(9) Dynamic changes of cancer: requires dynamic intervention according to the development of the disease.

For the above-mentioned conditions, molecular hydrogen may play a very important role in controlling cancer, which is manifested in:

(1) Tumor cells themselves produce a large number of ROS. By removing ROS, molecular hydrogen transforms cancer cells; suppresses cancer cell proliferation, invasion and movement; and weakens cancer.

(2) Molecular hydrogen selectively eliminates toxic ROS; down-regulates pro-inflammatory and inflammatory factors; removes chronic inflammation; and transforms the microenvironment.

(3) Molecular hydrogen protects mitochondria, maintains energy production, helps correct hypoxia, and protects cardiovascular, endocrine, and nervous systems, thereby transforming the entire body.

(4) Molecular hydrogen regulates immune function, selectively down-regulates inflammatory cytokine gene expression, up-regulates anti-inflammatory cytokine gene expression, restores depleted CD8+ T cell function, and strengthens the body's own anti-cancer effect.

Molecular hydrogen features strong antioxidation, high dispersion and great safety, all of which ensure that its translation into clinical use is feasible and effective. According to existing studies, molecular hydrogen serves as an important means to practice Chinese way of cancer control. In the strategy that requires combined use of cancer elimination and transformation therapies, molecular hydrogen may play a role in many aspects, mainly in terms of cancer transformation as summarized in the table below.

Table — China's way of cancer control and role of molecular hydrogen

Strategy	Method	Role of Molecular Hydrogen
Transforming residual cancer	Differentiation induction	To reduce ROS levels in tumor cells and weakens the cancerous effect
Transforming the microenvironment	Peroxidation control	To eliminate toxic ROS; down-regulate cell division and vascular endothelial growth-related gene expression; and inhibit tumor progression.
Transforming the microenvironment	Inflammation control	To down-regulate pro-inflammatory and inflammatory factor gene expression through gene regulation.
Transforming the body	Nerval intervention	To protect nerves, down-regulate the activity of apoptotic enzymes, and mitigate hypoxic-ischemic brain damage.
Transforming the body	Immune intervention	To selectively down-regulate pro-inflammatory cytokine gene expression, up-regulate anti-inflammatory cytokine gene expression, and restore functions of depleted CD8+ T cells.
Transforming the body	Metabolic intervention	To protect mitochondria, improve aerobic respiration, and regulate bioenergy.
Transforming the body	Nutritional intervention	To reduce the side effects of radiotherapy and chemotherapy; boost appetite; prevent decline of white blood cells in the blood, and improve quality of life.
Comprehensive transformation	Systemic nursing	To strengthen vitality, eliminate cancerous fatigue, boost appetite, improve sleep, smoothes breathing, maintain heart & kidney functions, and alleviate liver damage.

Tang Zhaoyou's Comments on Hydrogen Cancer Control

Academician Tang Zhaoyou has given brief comments on each story about hydrogen cancer control in this book.

In the fight against cancer, the western approach advocates "confronting the tough with toughness", while the eastern one often "conquers the tough with softness". The western approach aims to "kill all", while the eastern approach often "leaves a way out for the hard-pressed enemy" (excerpted from *Chinese Way of Fighting Cancer—Wisdom in The Art of War*, Shanghai Scientific and Technical Publishers, 2014). Chemotherapy and hydrogen inhalation are clearly two different treatments. The former can be compared to elimination therapy, and the latter to transformation therapy. The former can be understood as "positional warfare" and the latter "guerrilla warfare". How to properly apply the two different treatments varies from person to person and from case to case. Both methods (hard and soft ways) complement each other and are indispensable. Nowadays, tactics are neglected. This is a defect in cancer control and is worth pondering.

Hydrogen inhalation is obviously not a treatment for mass killing of tumors, but builds on cancer elimination treatments and achieves "small victories with accumulative effect" as in guerrilla warfare. This may once again indicate the importance of integrating cancer elimination and transformation therapies.

Nowadays, people give more attention to the so-called expensive "specific medicine". Simple and easy methods such as hydrogen inhalation are rarely taken seriously. This deserves reflection. Practice is the only criterion for testing truth. In today's medical world that blows strong winds of high, sophisticated and cutting-edge technologies, we cannot afford to let such accidental events like hydrogen inhalation pass by. A cancer patient can be compared to a drowning person desperate to survive. The moment when the patient is only

one foot off the shore and about to be drowned, he or she can be saved if someone offers a hand with gentle pull. Such a pulling hand must not be underestimated. Hydrogen inhalation is exactly a "gentle pull."

Cancer is a chronic disease, making cancer control a long-lasting battle about eliminating and transforming cancer cells. Hydrogen inhalation can certainly play a vital role in it.

Summary

The research on hydrogen molecular biology is still in its infancy; and that on its value for preventing and treating cancer has just started. More basic and preclinical studies are needed to understand the biological effects that drive molecular hydrogen's induction, and its upstream and downstream regulatory mechanisms. Well-designed multi-center clinical trials should be conducted gradually to identify the optimal mode of administration and optimal hydrogen dosage regimen.

In fact, it is undoubtedly reasonable and practical to give patients hydrogen inhalation leisurely so that they enjoy physical and mental satisfaction from hydrogen, the simplest, easiest, most natural and safest small molecule. It does not matter whether hydrogen inhalation happens in a hospital, in a health center or at home. Nor does it matter whether hydrogen inhalation serves as a neoadjuvant therapy before cancer-eliminating treatment; as a supplementary treatment; or as a means of rehabilitation. Hydrogen inhalation is worth trying even for those who are in advanced stages of cancer and have lost the chance of conventional treatment or refuse to receive conventional treatment.

According to an ancient Chinese poem, "When reaching the great peak (of Mount Tai), we hold all mountains in a single glance." We believe that with the deepening and further practice of hydrogen molecular medicine, hydrogen cancer control will definitely prove its unique charm as both a concept and a strategy in Tang Zhaoyou's China's way of cancer control.

— Xu Kecheng, Lu Tianyu, Chen Jinfei

References

1. Chabner BA and Roberts TG, Jr. Timeline: Chemotherapy and the war on cancer. Nature reviews. Cancer, 2005, 5(1): 65-72.
2. Prasad V. Perspective: The precision-oncology illusion. Nature, 2016, 537(7619): S63.
3. Tannock IF and Hickman JA. Limits to Personalized Cancer Medicine. N Engl J Med, 2016, 375(13): 1289-1294.
4. Cutbacks at England's Cancer Drugs Fund. Cancer Discov, 2015, 5(4): 338-339.
5. Haines I. The war on cancer: time for a new terminology. Lancet, 2014, 383(9932): 1883.
6. Jiang Y, Liu G, Zhang L, et al. Therapeutic efficacy of hydrogenrich saline alone and in combination with PI3K inhibitor in nonsmall cell lung cancer. Mol Med Rep, 2018, 18(2): 2182-2190.
7. Yang Y, Zhu Y, Xi X. Anti-inflammatory and antitumor action of hydrogen via reactive oxygen species. Oncol Lett, 2018, 16(3): 2771-2776.
8. Wang D, Wang L, Zhang Y, et al. Hydrogen gas inhibits lung cancer progression through targeting SMC3. Biomed Pharmacother, 2018, 104: 788-797.
9. Huang L. Molecular hydrogen: a therapeutic antioxidant and beyond. Med Gas Res, 2016, 6(4): 219-222.
10. Hu Z, Wu B, Meng F, et al. Impact of molecular hydrogen treatments on the innate immune activity and survival of zebrafish (Danio rerio) challenged with Aeromonas hydrophila. Fish Shellfish Immunol, 2017, 67: 554-560.
11. Grazioli S and Pugin J. Mitochondrial Damage-Associated Molecular Patterns: From Inflammatory Signaling to Human Diseases. Front Immunol, 2018, 9: 832.
12. Li HM, Shen L, Ge JW, et al. The transfer of hydrogen from inert gas to therapeutic gas. Med Gas Res, 2017, 7(4): 265-272.

Extra: "Holy Water"

> *Banas Holy Water (picture by Chen Xianming)*

Nordenau in Germany

Nordenau is a small village on the east side of Dusseldorf in northwestern Germany. The village had a cave where a stream of sweet spring water gushed from the wall. After drinking the cave spring water, the locals were full of vitality. And their various sub-health symptoms, including insomnia, anxiety, waist & leg pain and gastrointestinal discomforts, disappeared. Their skin became smooth and delicate. Symptoms were alleviated in many patients with chronic diseases. Some patients even recovered miraculously.

On June 13, 1998, the Asahi TV program *Exploring the Truth* played an episode of *Treating a Hundred Diseases – The Truth of Magical Water*, reporting the magical effect of the water from the cave in Nordenau, Germany. According to the program, the magical water contains abundant hydrogen, which means that hydrogen is the main mechanism behind the healing water. At the fifteenth Meeting of Japanese Association for Animal Cell Technology on September 22, 2006, a Japanese scholar presented the therapeutic effects of the magical water from Nordenau cave on patients with Type 2 diabetes.

Lourdes in Southern France

This is a water spring that has never dried up. More than 5 million people from more than 150 countries have come here to seek holy water. According to legend, on February 11, 1858, Bernadette, a 14-year-old shepherd, came to collect firewood near a cave on the bank of The Po river. Virgin Mary suddenly appeared in front of her, calling the little girl to the river to drink some water and wash her face. As the girl dug the ground near the cave, spring water gushed out. Since then, many ill people had their illness cured after drinking the spring water.

In the past more than 100 years, at least 67 people have been reported to have fully recovered from years of pain or disease after drinking the holy water. Ms. Anna Santaniro waAt the age of 40 in 1952, Anna suffered from a serious heart attack. Her brother and sister suffered from the same illness and had died successively. Anna came to Lourdes with a glimmer of hope and washed herself with the holy water. Amazingly, shortly after soaking herself in the spring water, Anna felt a warm current in her chest. Then, her breathing began to flow smoothly and legs gained consciousness.

What has long been puzzling is that the holy water has therapeutic effects only in the local area. Once being taken out of the locality, the water loses its effect. It is now understood that

> **To Holy Water**
> *— Zhong Zimei*
> From France, Germany or Nordenuguan in China;
> Comes the gospel;
> An invisible hand reaches out from the clear spring water, bringing patients back to life;
> Smoothing the pain and bringing longevity and health;
> Misty fairy air;
> The secret of this remains unknown for a long time;
> But now, medical pioneers identify the effect of the spring water; inventors have developed a hydrogen mask device that allows expiration and inspiration of the holy spring water;
> For thousands of households,
> People becoming desperate because of cancer;
> Are able to conquer death and feel the miracle of being reborn.
> A new era has arrived;
> Nature's most common ingredients gently lift the heavy cover of human seipathia;
> Holy spring water swirls, rejoices and bounces in the garden of good health…

the effect of the holy water comes from hydrogen molecules.

Bama in China's Guangxi Province

The Bama Yao autonomous county in the northwestern mountainous region of Guangxi, China, is a magical and beautiful place. Since ancient times, there have been elderly people over 100 years old. The county is known as a place of longevity.

It is spring water that explains why people in Bama live long lives. The spring water, which is generated from hundreds of millions of years of karst stratum, runs four times underground and flows out of the earth's surface four times, making up natural wonders. Through seven years of research, the International Society for Naturopathic Medicine has proved that Bama spring water can enter cell nuclei and DNA; activate cellular enzyme tissues; and stimulate vitality.

Studies suggest that Bama water disperses red blood cells that conglomerate into groups; reduces blood viscosity; promotes microcirculation; alleviates or eliminates nausea,

heartache, limb atony, shortness of breath, headache, head swelling, dizziness, insomnia, cold limbs, limb numbness, tinnitus, blurred vision and other symptoms. It also helps lower blood fat, reduce blood pressure, and mitigate or eliminate cardiac and cerebral atherosclerosis. Young girls who use Bama water as a toner have more radiant skin. In the case of diabetes, patients have normal blood sugar levels after drinking 4 liters of Bama water every day for 3 months consecutively. Long-term consumption of Bama Shouzhen spring water contributes significantly to enhancing physical fitness and slowing down aging.

Bama Water delivers similar magical effects to the above-mentioned holy water from Germany and France. It is no exaggeration to call it Bama holy water. Opinions vary as to the nature of Bama water. Does hydrogen also play a role in the effect of Bama water? This is worthy of study.

PART 2 — Scientific Foundation of Hydrogen Cancer Control

Antioxidant and Anti-inflammatory Effects of Molecules Hydrogen

Key points

- In 1975, American researcher Dole M used high-pressure hydrogen-oxygen mixture for the first time to treat squamous cell carcinoma of the skin in mice and achieved significant results, creating a new era of hydrogen cancer control.
- In 2007, Japanese researcher Shigeo Ohta first discovered that a low concentration (2%) of positive-pressure hydrogen scavenged reactive oxygen species and exerted antioxidant and anti-inflammatory effects. This represents considerable progress in the research of molecular hydrogen biology and hydrogen medicine, blazing a way into studying the mechanism of hydrogen cancer control.
- The mechanism of hydrogen cancer control is yet to be clarified. The probable mechanism is mainly to remove toxic reactive oxygen species; control oxidation and inflammation; maintain mitochondrial function and stability; and regulate immunity especially by rescuing depleted T cells.
- With antioxidant and anti-inflammatory effects, hydrogen reduces the adverse reaction of chemotherapy and radiotherapy without compromising the anti-tumor effect of these treatments. This provides a basis for applying combined anti-tumor therapies, including hydrogen therapy.

Discovery of the Role of Hydrogen Cancer Control

In recent years, the biological effects of molecular hydrogen has attracted attention. Its clinical availability started with cancer treatment. In 1975, American researcher Dole M et al. first proposed that hydrogen (H_2) had direct inhibitory effects on cancer. In their experiment, hairless mice with squamous cell carcinoma of the skin inhaled eight atmospheres of a gas mixed with 97.5% hydrogen and 2.5% oxygen in 2 weeks. As a result, the cancer in the mice shrank significantly, and manifested as tumor blackening and fall-off. Biopsy of the rat models receiving hyperbaric oxygen-helium demonstrated no such change (see Picture 1). The subcutaneous tumors showed no sign of squamous cell carcinoma.

> *Picture 1. "Hyperbaric Hydrogen Therapy: A Possible Treatment for Cancer" by Dole et al., published in the 1975 issue of Science. The picture showed three groups of animals: (i) control group; (ii) group with inhalation of high-pressure helium-oxygen; (iii) group with inhalation of high-pressure hydrogen-oxygen, and respective tumor sizes (cm) at the beginning and after 10 days. Numbers in the brackets refer to tumor sizes at the end of Day 10 (at the end of Day 9 for the helium-oxygen group).*

In 1978, American researcher Roberts et al. studied the effects of high-pressure H_2 on five mice with transplanted tumors and one mouse with leukemia. It was found that hydrogen inhalation inhibited the growth of tumor cells and leukemia cells.

In 2008, Japanese researcher Saitoh et al. studied the effects of hydrogen-rich electrolyzed water with neutral pH value on tumor cells. It was found that the water inhibited the colony formation efficiency or colony size of human tongue cancer cell line HSC-4, while normal human tongue epithelioid cells were not much inhibited. For human fibrosarcoma cell line HT-1080, hydrogen-rich water also causes growth inhibition and cell degeneration; and weaken

its invasiveness. In 2009, Saitoh et al. reported that similar inhibitory effects were achieved on the colony formation and size of human HSC-4 tongue cancer cells by enhancing hydrogen water in colloidal platinum.

In 2010, Chinese scholars published three reports about applying molecular hydrogen to radiation injury. According to a review by Liu C et al., hydrogen inhalation had antioxidant and anti-apoptotic activity; and protected the brain from ischemia-reperfusion injury and cerebral stroke by selectively reducing hydroxyl (·OH) and peroxynitrite ($ONOO^-$). Reducing hydroxyl radicals helped significantly strengthen the protective effects of cells on radiation damage. The authors believed that hydrogen therapy might be an effective, specific, and unique treatment for acute radiation syndrome. Qian et al. reported that processing cells with H_2 before irradiation significantly inhibited irradiation-induced apoptosis of human lymphocyte ah-1 cells and enhanced cell viability in vitro. Hydrogen inhalation protected the gastrointestinal endoderm from radiation damage in mice; reduced the levels of plasma malondialdehyde (MDA) and intestinal 8-hydroxydeoxyguanosine (8-OHDG); and increased plasma endogenous antioxidants. Qian et al. also studied the protective effects of hydrogen-rich water on mice with damage from ionizing radiation. The researchers proved that hydrogen-rich water protected against myocardial degeneration; reduced myocardial MDA and 8-OHDG levels; and increased the content of myocardial endogenous antioxidants in the body.

In a study by Chinese researcher Zhao et al. in 2011, lymphoma was induced in mice after multiple exposures to radiation. The mice group with intraperitoneal injections of hydrogen-rich saline had significantly lower incidence of lymphoma and markedly longer survival. Such preventive effects were not found in the mice group with injections of normal saline.

In 2015, Japanese researcher Runtuwene et al. reported that Colon-26 cell line induced intestinal cancer in mice; and that intraperitoneal injection of hydrogen water increased the survival rate of tumor-bearing mice and induced cancer cell apoptosis. Hydrogen water injection, when combined with the application of chemo drug 5-fluorouracil (5-FU), delivered better anti-tumor effects. This indicated that the two methods had synergistic effects on the treatment of colon cancer.

Basic Principles of Hydrogen Cancer Control

In 2007, Japanese researcher Shigeo Ohta et al. reported that in rat models of ischemia-reperfusion brain injury induced by surgical clamping of the carotid artery, inhaled hydrogen rapidly spread to cell membranes and buffered the action of oxidative stress, which significantly inhibited brain injury. This pioneering study provided a theoretical basis for the application of molecular hydrogen to the treatment of clinical diseases.

Molecular hydrogen has therapeutic effects on many disease models related to ROS and human diseases. The effects are mainly attributed to hydrogen's selective antioxidation and anti-inflammation, as reported by many articles and papers such as Chinese researcher Huang et al. in 2010 and Japanese researcher Shigeo Ohta et al. in 2011 and 2014. Cancer is a multi-stage process defined by cancer initiation, promotion, and progression. And oxidative stress plays a role in all the three phases of the process. Peroxidation and inflammation mediated by ROS and their mutual promotion are key factors in the development and progression of cancer. It is generally believed that H_2 may control the occurrence and development of cancer mainly by containing oxidation and inflammation. However, recent studies suggest that hydrogen molecules act directly on the mitochondria of tumor cells and immune cells; and play a role in killing tumors and reviving immune cells.

Picture 2. Relationships between inflammation and tumor [Excerpt from: Kim J and Bae JS. Exp Mol Med, 2016, 48(11): e269.]
The balance of ROS production versus the body's antioxidant system: *Few ROS* — change the normal functions of lipids, proteins and DNAs (cell signal, cell proliferation, angiogenesis); *Many ROS* — DNA damage causes normal cell death or apoptosis; *A great many ROS* — DNA damage causes normal cell death or apoptosis, ROS adapts to new environment conditions, tumor cells survive and become resistant to drugs (occurrence of tumor). ROS can be systematically cleared by the body's antioxidant system.

1. Molecular hydrogen controls cancer by removing ROS

(1) ROS causes cancer

As the products of imbalance between free radicals and active metabolites, ROS include oxygen radicals [such as superoxide anion (O^{-}_{2}), hydroxyl (·OH), peroxy (RO·) and alkoxy (RO−)] and non-oxygen free radicals. The molecules that can be converted into ROS and act as oxidants include hydrogen peroxide (H_2O_2), hypochlorous acid (HOCl), ozone (O_3) and singlet oxygen (1O2). ROS promote DNA synthesis, cell proliferation, cell survival, cell migration and cell invasion as well as tumor metastasis and angiogenesis. In endogenous metabolic reaction, aerobic cells may produce multiple types of ROS. Continuous exposure of mitochondria to high levels of ROS results in mitochondrial DNA damage and apoptosis. If the mitochondria of normal stem cells in tissues are damaged, production of cancer stem cells might be induced, leading to the formation of tumors. If the risk factors remain, the damage is gradually aggravated. And the tumors grow bigger and become resistant to drugs (see Picture 2).

Reactive nitrogen radicals (RNS) are generated by nitrogen-containing oxidants such as nitric oxide (NO) and peroxynitrite ($ONOO^{-}$). The mitochondrial respiratory chain produces RNS under hypoxic conditions; and RNS further generate other active substances. It is now known that cells produce ROS and RNS consecutively, which is the result of exposure to carcinogens, inflammation and mitochondrial respiration.

Radiation, smoking, air pollution, chemicals, mental stress, inflammation, strenuous exercise, myocardial infarction, blood flow stop and organ transplantation induce the body to produce ROS and RNS, of which ·OH and $ONOO^{-}$ are the most toxic. Free radicals act on DNA, lipids and proteins by inducing cellular signal transduction pathways. As a result, DNA damage, lipid peroxidation and albuminous degeneration occur, triggering cell mutations and cancer cell production, proliferation, survival and migration.

(2) ROS promote tumor progression

Tumor cells produce more abundant ROS than normal cells and cause elevated oxidative stress. ROS damage to DNA involves a variety of chronic inflammatory diseases and many types of cancer, including bladder cancer, brain tumor, breast cancer, cervical cancer, stomach cancer, liver cancer, lung cancer, melanoma, multiple myeloma, leukemia, lymphoma, oral cancer, ovarian cancer, pancreatic cancer, prostate cancer, and sarcoma.

ROS initiate oncogenicity and subsequent tumor progression by inducing DNA damage. Oxidative stress interacts with the initiation, promotion and progression of cancer. During the initiation phase, ROS introduce gene mutations and structural changes into DNA and cause DNA damage. During the promotion phase, ROS facilitate cancer cell proliferation or reduce cancer stem cell apoptosis by causing abnormal gene expression, blocking cell

communication and modifying the second messenger system. In the end, oxidative stress may increase DNA changes in the initiating cell population and accelerate cancer progression.

(3) ROS exert influence on cancer by regulating gene expression

Under intracellular and extracellular environment conditions, ROS regulate gene mutations and balance cell proliferation and apoptosis by stimulating cellular signaling pathways. Cancer signals originate from various autocrine and paracrine elements in the hypoxic microenvironment, including vascular endothelial growth factor (VEGF), hepatocyte growth factor (HGF), hypoxia-inducible factor 1 alpha (HIF-1 alpha), NO and H_2O_2. Cancer signals generate positive feedback loops that activate protein kinase B (Akt) gene expression. Oxidative stress activates a range of transcription factors including NF-κB, P53, HIF-1α, matrix metalloproteinase (MMP), peroxisome proliferator-activated receptor γ (PGC-1α), β-catenin/Wnt signal and erythrocyte nucleus-related factor 2 (Nrf2). These effector molecules are activated by persistent ROS-related chronic inflammation and are involved in the malignant transformation, immunity, inflammatory response, and gene expression of cancer occurrence and metastasis.

(4) Molecular hydrogen removes ROS

H_2, dissolved in the culture medium selectively reduces the strongest oxidants (such as ·OH and $ONOO^-$) in cell signaling transduction. But it does not interfere with the levels of O_2, NO or H_2O_2 near where it affect oxidation in cell-free systems (see Picture 3). Studies show that H_2 neutralizes ·OH produced by radiation decomposition or photolysis of water, and significantly reduces ·OH levels in cultured cells, thereby protecting mitochondria. With the capacities to penetrate biofilms and diffuse into organelles, H_2 reduces the level of synthetic adenosine triphosphate (ATP) in mitochondria and cell nuclei. In addition, H_2 can boost the activity of superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPx); decrease ROS; and promote the expression of phosphoinositide 3-kinase (pi-3k), Akt and Srf2 in human immortalized epidermal cells (HaCaT) induced by chronic ultraviolet radiation. H_2 exerts neuroprotective effects by weakening cyclooxygenase-2 (Cox-2) activity in immunoreactive neurons.

> *Picture 3. H_2 enhances the activity of SOD, CAT and GPx.* (O_2 → [superoxide dismutase] → H_2O_2 → [peroxidase] → ·OH → H_2O)

2. Molecular hydrogen controls cancer by containing inflammation

Metastasis is a characteristic of cancer cells. As early as more than 100 years ago, British pathologist Paget suggested that cancer cells only metastasize in a suitable environment. This is the "Seed and Soil Theory." Further studies revealed that cancer cell metastasis depends on the microenvironment (soil), which is a hotbed for the formation, development and metastasis of cancer cells. In the cancer microenvironment, there are at least four factors that facilitate cancer cell growth, namely, inflammation, hypoxia, macrophages, and immune cells. Inflammation is the most important cancer-promoting factor in the microenvironment (see Picture 4).

ROS may also cause chronic inflammation, resulting in increased organ cell damage, increased cell division, increased cell mutations, and increased cancer cell formation. Molecular hydrogen exerts anti-inflammatory and anti-allergic effects by inhibiting

inflammatory cytokines and cell signaling factors, thereby suppressing tumor growth and progression.

It is known that H_2 reduces the expression of many pro-inflammatory factors, including tumor necrosis factor alpha (TNF-α), interleukin (IL)-6, IL-1β, IL-10, IL-12, chemotactic factor ligand 2 (CCL-2), intercellular adhesion molecule 1 (ICAM-1), NF-κB, high-mobility group protein 1 (HMG-1) and prostaglandin E 2 (PGE 2). In addition, H_2 has functions to reduce the activity of serum diamine oxidase (DAO), TNF-α, IL-1β, IL-6, tissue malondialdehyde (MDA), protein carbonyl (PCO) and myeloperoxidase (MPO); and inhibit apoptosis-promoting factors, including amino-terminal kinase (JNK) and caspase-3. In 2018, Chinese researcher Zhang et al. reported that in mouse models of allergic asthma, hydrogen inhalation significantly reduced the numbers of eosinophilic granulocytes and lymphocytes in bronchoalveolar lavage fluid; increased the levels of IL-4, IL-13, TNF-α and CCL-15; and mitigated airway inflammation.

> *Picture 4. Tumor microenvironment. The green elements represent tumor-killing cells, and the red ones indicate tumor-protecting cells.*

3. Molecular hydrogen maintains the normal function of mitochondria

Mitochondria are the centers of oxidative metabolism and the main sites where ROS are generated. Mitochondrial genome DNA (mtDNA) or mitochondrial life function disorders (such as the production of ATP and ROS) have been proven to be involved in the occurrence of many diseases. Maintaining the mitochondrial functions of normal cells helps lower the possibility of mutation into tumor cells. Maintaining the mitochondrial functions of tumor cells leads to weakened tumor invasiveness and drug resistance. Maintaining the mitochondrial functions of immune cells helps keep immune cells at normal function levels. In 2016, German researcher Otto et al. observed cancer fermented glucose in the presence of O_2, suggesting that mitochondrial respiratory defects might be the root cause of cancer. Radiation, carcinogenic chemicals, viruses, and chronic inflammation induce ROS, which damage mitochondria and aerobic respiration. As a result, fermentation genes are activated, forming a fermentation-based energy-generating process and further causing cancer cells. Some people think that cancer is actually a metabolic disease. Some refer to cancer as mitochondrial disease. This will be discussed in detail in a separate chapter below.

Driven by concentration gradients, H_2 rapidly penetrates cell membranes into mitochondria. In 2011, Japanese researcher Ohta reported that in addition to protecting mitochondria from ROS such as ·OH, H_2 protects mitochondrial functions, including by maintaining mitochondrial membrane potential, increasing ATP production, reducing organelle swelling, and maintaining sound production of mitochondria bioenergy (see Picture 5).

> *Picture 5. H_2 passes through the cell membrane, and enters the nucleus, endoplasmic reticulum and mitochondrion, and neutralizes ·OH. [Quoted from: Ohta S. Curr Pharm Des, 2011, 17(22): 2241-2252.]*

4. Molecular hydrogen directly regulates immune function

CD8+ T cell (cytotoxic T lymphocyte) is an important immune killer cell and plays an important role in tumor immune control. However, its immune function is closely related to cell surface molecules. As a marker of depleted T cells, programmed death receptor 1 (PD-1) is abundantly expressed in cancer patients' CD8+ T cell circulation and tumor-infiltrated lymphocytes. It is associated with the poor prognosis of various cancers (including breast,

pancreatic and gastric cancers). The mechanism by which H_2 restores depleted T cell function remains unclear. But this phenomenon was reported by Akagi and Baba in Japan in early 2019. This part will be elaborated in a separate

chapter below.

Molecular hydrogen reduces the side effects of radiotherapy and chemotherapy

The three major treatments for cancer are surgery, radiotherapy, and chemotherapy. Patients receiving radiotherapy and chemotherapy often have to discontinue treatments for a short or long period of time due to vomiting, diarrhea, liver & kidney toxicity, and low levels of white blood cells or platelets to avoid fatal side effects. In recent years, there have been more and more reports about the effects of hydrogen on alleviating the side effects of tumor radiotherapy or anti-tumor drugs. And the conclusions are increasingly clear.

1. Molecular hydrogen reduces radiotherapy side effects

In 2011, Chinese researcher Kang et al. conducted a study of 49 patients with liver radiotherapy. It was found that drinking hydrogen-rich water daily helped ease oxidative stress caused by radiotherapy and improve quality of life (using QLQ-C30 quality of life measurement scale questionnaire). Moreover, the therapeutic result for the radiotherapy site was no different from that of the control group, indicating that hydrogen-rich water had no impact on anti-tumor effects. The authors also detected liver function changes in patients whose livers were irradiated, and immune cell changes in patients whose spleens were irradiated. The findings showed that indicators of the hydrogen water group were not significantly different from those before irradiation, while the control group had significant damage.

In 2014, Chinese researcher Zhao et al. reported that irradiation caused immune damage to mice; and intraperitoneal injection of hydrogen-rich saline exerted the following effects: a) Increasing spleen index and reducing radiation damage to the spleen; b) Significantly lowering spleen ROS expression levels, decreasing apoptotic cells, and reducing the expression of pro-apoptotic proteins; c) Increasing the numbers of CD4+ and CD8+ T cell subsets in the spleen, so that the ratio of the two is close to normal; d) Restoring the polarization pattern and cytokine secretion level of spleen CD4+ T cells.

In 2016, Chinese researcher Guo et al. reported that low-dose long-term radioactive irradiation caused cumulative damage to mice, which was reflected in body mass index, forced swimming test, open box experiment, chromosome aberration assay, peripheral blood cell parameter analysis, sperm abnormality analysis, and lymphocyte deformation experiments. Long-term consumption of hydrogen-rich water by irradiated mice gradually restored almost all indicators to normal levels, indicating that hydrogen can reduce damage from low-dose long-term irradiation and protect multiple systems throughout the body.

2. Molecular hydrogen reduces chemotherapy side effects

As a cell cycle non-specific drug with cytotoxicity, cisplatin may inhibit the DNA replication process of cancer cells and damage their structures on cell membranes, exerting strong broad-spectrum anti-cancer effects. Cisplatin is clinically effective in treating multiple solid tumors, such as ovarian tumor, prostate tumor, testicular tumor, lung tumor, nasopharyngeal tumor, esophageal tumor, malignant lymphoma, head & neck squamous cell carcinoma, thyroid tumor and osteosarcoma. In 2009, Japanese researcher Kamimura-Kamimura et al.

reported that severe complications occurred after injection of chemo drug cisplatin in mice. On the second day, some of the mice died, and only 60% mice survived to Day 6. However, the hydrogen inhalation group had markedly lower mortality. All the mice survived to Day 5, and 80% of them survived to Day 9. Their body weight changed a little, and nephrotoxicity was small (serum creatinine and urea nitrogen levels were basically normal). The sensitivity of the two groups of tumor-bearing mice to cisplatin was not affected by hydrogen. In 2010, the research team again reported the effects of intragastric administration of hydrogen-rich water on tumor-bearing rats' recovery from chemotherapy. And similar results were observed.

As one of the most effective anti-tumor drugs, doxorubicin is widely applied to adults and children with tumors. But its clinical application is significantly limited due to strong cardiac and renal toxicity. In 2016, Chinese researcher Gao et al. reported that intraperitoneal injection of hydrogen-rich saline mitigated rats' complications induced by doxorubicin: a) After 30 days of treatment, 30 rats in the hydrogen-rich saline group survived, whereas only 18 in the normal saline group survived. b) Cardiotoxicity, histopathological changes, liver function indicators, and ROS decreased. c) Heart and liver inflammation was inhibited; and the expression of apoptosis-related proteins in organs was reduced.

From the results above, it can be concluded that hydrogen has the potential to be an auxiliary means for reducing the side effects of radiotherapy and chemotherapy in cancer patients.

Summary

Hydrogen medicine is a young yet fast-developing division of science. The discovery of molecular hydrogen's effects on cancer control is earlier than the generally conceived hydrogen medicine. More than 40 years ago, the famous American journal *Science* reported that the application of high-pressure hydrogen and oxygen mixture effectively controlled cancer.

For technical reasons, high-pressure hydrogen is difficult to apply. Or because of misconception, hydrogen-based treatment is considered to be too simple. Until recent years after the antioxidant and anti-inflammatory effects of molecular hydrogen were discovered, people remembered and started to examine the paper published in Science, only to find that hydrogen, a simplest inexhaustible natural substance, does inhibit cancer. Then, they came to believe that controlling cancer with hydrogen is not nonsense but has profound scientific foundation. Encouragingly, molecular hydrogen is involved in the body's immune function. It maintains mitochondrial functions, saves T cells in depletion, and restores the anti-tumor efficacy of T cells, thereby improving the prognosis of patients with advanced cancer.

Medical science is not simply a natural science, because it implies people's awe of human life. Science-based medical technology is inevitably the best for serving life and caring for life. Hydrogen cancer control is such a technology.

— *Xu Kecheng, Lu Youyong, Chen Jibing, Du Duanming*

References

1. Dole M, Wilson FR, Fife WP. Hyperbaric hydrogen therapy: a possible treatment for cancer. Science, 1975, 190(4210): 152-154.
2. Roberts BJ, Fife WP, Corbett TH, et al. Response of five established solid transplantable mouse tumors and one mouse leukemia to hyperbaric hydrogen. Cancer Treat Rep, 1978, 62(7): 1077-1079.

3. Saitoh Y, Okayasu H, Xiao L, et al. Neutral pH hydrogen-enriched electrolyzed water achieves tumor-preferential clonal growth inhibition over normal cells and tumor invasion inhibition concurrently with intracellular oxidant exposure. Oncol Res, 2008, 17(6): 247-255.
4. Saitoh Y, Yoshimura Y, Nakano K, et al. Platinum nanocolloid-supplemented hydrogendissolved water inhibits growth of human tongue carcinoma cells preferentially over normal cells. Exp Oncol, 2009, 31(3): 156-162.
5. Liu C, Cui J, Sun Q, et al. Hydrogen therapy may be an effective and specific novel treatment for acute radiation syndrome. Med Hypotheses, 2010, 74(1): 145-146.
6. Qian L, Cao F, Cui J, et al. Radioprotective effect of hydrogen in cultured cells and mice. Free Radic Res, 2010, 44(3): 275-282.
7. Qian L, Cao F, Cui J, et al. The potential cardioprotective effects of hydrogen in irradiated mice. J Radiat Res, 2010, 51(6): 741-747.
8. Zhao L, Zhou C, Zhang J, et al. Hydrogen protects mice from radiation induced thymic lymphoma in BALB/c mice. Int J Biol Sci, 2011, 7(3): 297-300.
9. Runtuwene J, Amitani H, Amitani M, et al. Hydrogen-water enhances 5-fluorouracil-induced inhibition of colon cancer. PeerJ, 2015, 3: e859.
10. Ohsawa I, Ishikawa M, Takahashi K, et al. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med, 2007, 13(6): 688-694.
11. Huang CS, Kawamura T, Toyoda Y, et al. Recent advances in hydrogen research as a therapeutic medical gas. Free Radic Res, 2010, 44(9): 971-982.
12. Ohta S. Recent progress toward hydrogen medicine: potential of molecular hydrogen for preventive and therapeutic applications. Curr Pharm Des, 2011, 17(22): 2241-2252.
13. Ohta S. Molecular hydrogen as a preventive and therapeutic medical gas: initiation, development and potential of hydrogen medicine. Pharmacol Ther, 2014, 144(1): 1-11.
14. Kim J and Bae JS. ROS homeostasis and metabolism: a critical liaison for cancer therapy. Exp Mol Med, 2016, 48(11): e269.
15. Kumari S, Badana AK, G MM, et al. Reactive Oxygen Species: A Key Constituent in Cancer Survival. Biomark Insights, 2018, 13(1177271918755391).
16. Yang Y, Zhu Y, Xi X. Anti-inflammatory and antitumor action of hydrogen via reactive oxygen species. Oncol Lett, 2018, 16(3): 2771-2776.
17. Liou GY and Storz P. Reactive oxygen species in cancer. Free Radic Res, 2010, 44(5): 479-496.
18. Zhang G, Miura Y, Yagasaki K. Suppression of adhesion and invasion of hepatoma cells in culture by tea compounds through antioxidative activity. Cancer Lett, 2000, 159(2): 169-173.
19. Lei XG, Zhu JH, Cheng WH, et al. Paradoxical Roles of Antioxidant Enzymes: Basic Mechanisms and Health Implications. Physiol Rev, 2016, 96(1): 307-364.
20. Trush MA and Kensler TW. An overview of the relationship between oxidative stress and chemical carcinogenesis. Free Radic Biol Med, 1991, 10(3-4): 201-209.
21. Wang J and Yi J. Cancer cell killing via ROS: to increase or decrease, that is the question. Cancer Biol Ther, 2008, 7(12): 1875-1884.
22. Klaunig JE, Xu Y, Isenberg JS, et al. The role of oxidative stress in chemical carcinogenesis. Environ Health Perspect, 1998, 106 Suppl 1(289-295).
23. Sun H, Chen L, Zhou W, et al. The protective role of hydrogen-rich saline in experimental liver injury in mice. J Hepatol, 2011, 54(3): 471-480.
24. Zhang N, Deng C, Zhang X, et al. Inhalation of hydrogen gas attenuates airway inflammation and oxidative stress in allergic asthmatic mice. Asthma Res Pract, 2018, 4(3).
25. Giorgi C, Marchi S, Simoes ICM, et al. Mitochondria and Reactive Oxygen Species in Aging and Age-Related Diseases. Int Rev Cell Mol Biol, 2018, 340(209-344).
26. Otto AM. Warburg effect(s)-a biographical sketch of Otto Warburg and his impacts on tumor metabolism. Cancer Metab, 2016, 4(5).
27. Sabharwal SS and Schumacker PT. Mitochondrial ROS in cancer: initiators, amplifiers or an Achilles' heel? Nature reviews. Cancer, 2014, 14(11): 709-721.
28. Dermani FK, Samadi P, Rahmani G, et al. PD-1/PD-L1 immune checkpoint: Potential target for cancer therapy. J Cell Physiol, 2019, 234(2): 1313-1325.
29. Akagi J and Baba H. Hydrogen gas restores exhausted CD8+T cells in patients with advanced colorectal cancer to improve prognosis. Oncol Report, 2018.

30. Kang KM, Kang YN, Choi IB, et al. Effects of drinking hydrogen-rich water on the quality of life of patients treated with radiotherapy for liver tumors. Med Gas Res, 2011, 1(1): 11.
31. Zhao S, Yang Y, Liu W, et al. Protective effect of hydrogen-rich saline against radiation-induced immune dysfunction. J Cell Mol Med, 2014, 18(5): 938-946.
32. Guo J, Zhao D, Lei X, et al. Protective Effects of Hydrogen against Low-Dose Long-Term Radiation-Induced Damage to the Behavioral Performances, Hematopoietic System, Genital System, and Splenic Lymphocytes in Mice. Oxid Med Cell Longev, 2016, 2016(1947819).
33. Nakashima-Kamimura N, Mori T, Ohsawa I, et al. Molecular hydrogen alleviates nephrotoxicity induced by an anti-cancer drug cisplatin without compromising anti-tumor activity in mice. Cancer Chemother Pharmacol, 2009, 64(4): 753-761.
34. Kitamura A, Kobayashi S, Matsushita T, et al. Experimental verification of protective effect of hydrogen-rich water against cisplatin-induced nephrotoxicity in rats using dynamic contrast-enhanced CT. Br J Radiol, 2010, 83(990): 509-514.
35. Gao Y, Yang H, Fan Y, et al. Hydrogen-Rich Saline Attenuates Cardiac and Hepatic Injury in Doxorubicin Rat Model by Inhibiting Inflammation and Apoptosis. Mediators Inflamm, 2016, 2016(1320365).

Hydrogen Molecules Maintain Mitochondrial Functio and Stability

Key points

- According to the Warburg effect, cancer occurs due to strengthened anaerobic glycolysis of cellular sugar and decrease in oxygen consumption. For this reason, some believe that cancer is a mitochondrial disease.
- Mitochondria are the generators of the body's energy. The mitochondria of cancer cells are different from those of normal cells in terms of microstructure and metabolic function.
- Reactive oxygen species cause oxidative damage to mitochondrial proteins, membranes and DNA; impair mitochondrial capability to synthesize ATP; and adversely affect ATP metabolic functions, including tricarboxylic acid cycle, fatty acid oxidation, urea cycle, amino acid metabolism, and heme synthesis.
- Hydrogen molecules, due to their small sizes and high dispersion, can quickly enter the mitochondria and protect them (and cell nuclei) from acute oxidative stress damage.

With regard to hydrogen cancer control, the academic community generally focuses on hydrogen's antioxidant and anti-inflammatory effects. Life is not only about existence of the body, but also depends on energy and information. The production of biological energy in the body relies on mitochondria. What role do mitochondria play in cancer? What are the impact and role of hydrogen molecules on mitochondria?

Warburg Effect

In 1924, German physiologist Otto Warburg proposed that when mitochondrial function is impaired, cells provide energy by enhancing anaerobic glycolysis. According to Warburg, after being metabolized to pyruvic acid, glucose no longer performs aerobic oxidation through the tricarboxylic acid cycles of mitochondria; but converts into lactic acid to drain cells through lactic dehydrogenase (see Picture 1). Warburg believed that the occurrence of cancer is caused by increase in cellular sugar anaerobic glycolysis and decrease in oxygen consumption. This view is known as the Warburg Effect. In spite of many controversies on the Warburg Effect, the relationship between cancer and mitochondria has been verified by many facts later. Knowing this is of great significance to understanding the effects of hydrogen molecules on cancer and health.

> *Picture 1. Cancer cell self-sufficiency — a schematic of altered cancer metabolism: protein is hydrolyzed to gluconeogenesis; oxygen and cancer-suppressor-gene mutation drive*

> *chronic glycolysis; glutamine/glutamic-acid decomposition feeds the intermembrane gradient; high glycolysis consumes NAD+, which needs regeneration of lactic acid to maintain glycolysis.*

What Is Mitochondrion?

As cell generators in the cytoplasm, mitochondria are in large quantities and open the chemical bonds inside food molecules to obtain energy. The structure of a mitochondrion consists of an outer membrane, an inner membrane and intramembranous space (see Picture 2 on the left). A chemical bond consists of a positive charge called proton and a negative charge called electron. Protons and electrons are closely interdependent. Mitochondria pull electrons away from protons, and then leak electrons through the electron transport chain to generate current. This electrical energy is used to produce adenosine triphosphate (ATP) molecules, each of which contains a high-energy phosphate bond. ATP can be compared to a tiny chemical battery. Cells separate ATP phosphate bonds when energy is needed. Oxygen is waiting to capture cascaded electrons at the end of the ATP assembly line, and then binds with them, generating a harmless by-product: water. Since this process requires oxygen and produces high-energy phosphate bonds, it is called "oxidative phosphorylation", also known as "breathing" (see Picture 2 on the right).

> *Picture 2. Structure of a mitochondrion (left) — outer membrane, inner membrane, intermembrane space, ground substance, ridge (cristae) — and its metabolic function (right): glycolysis → pyruvate → acetyl coenzyme A → electron transport chain.*

Mitochondria of Cancer Cells

The mitochondria of cancer cells have the following characteristics compared with normal cells:

- a) Less mitochondria per cell;
- b) Often deformed; unsmooth inner surface;
- c) Low activity of key respiratory enzyme;
- d) Less and deformed cardiolipin, a key mitochondrial fat;
- e) Less DNA inside;
- f) Leakage-like shape and non-coordinated electron transport chain: Energy is consumed as heat instead of ATP.
- g) Significantly lower respiratory rate.

> *Picture 3. Metabolic difference between normal cells and proliferative or cancer cells. The left picture illustrates two physiological processes in normal tissues. The pyruvate generated by glucose under aerobic conditions mainly enters mitochondria and reacts with O_2, producing CO_2. The picture on the right shows hyperplasia or cancerous tissues. Regardless of aerobic or anaerobic environment, the pyruvate generated by glucose rarely enters mitochondria, but mainly produces lactic acid, an acidic metabolite. (Oxidative phosphorylation vs. anaerobic glycolysis vs. aerobic glycolysis / Warburg Effect.)*

What Is the Difference Between Normal and Cancer Cells in Energy Production?

The most important fundamental difference between normal cells and cancer cells is how they produce energy. Normal cells use complex breathing processes to effectively convert any nutrient (fat, carbohydrate or protein) into high energy. These processes require oxygen to completely break down food into harmless carbon dioxide and water. Cancer cells use a

primitive process called fermentation, which ineffectively converts glucose (mainly from carbohydrates) or amino acids glutamine (from proteins) into small amounts of energy. This process does not require oxygen and can only partially decompose food molecules into toxic wastes such as lactic acid and ammonia. If there is a temporary shortage of oxygen, normal cells occasionally have to resort to fermentation. However, when there is enough oxygen around, normal cell adopts fermentation. Cancer cells are different. Even if there is abundant oxygen around, cancer cells mainly use fermentation. This is the Warburg Effect described above, and is considered to be the metabolic feature of cancer cells (see Picture 3). The reason for this difference is that the mitochondria of cancer cells are damaged.

> *Picture 4. Mitochondria-derived ROS cause oxidative damage to mitochondrial proteins, membranes and DNA; impair mitochondrial capacity to synthesize ATP; and adversely affect mitochondrial metabolic functions, including tricarboxylic acid cycle, fatty acid oxidation, urea cycle, amino acid metabolism, and heme synthesis. If mitochondria have oxidative damage, mitochondrial outer membrane permeabilization (MOMP) occurs and results in the release of intermembrane space proteins such as cytochrome c (cyt c) to the cytoplasm, thereby activating apoptosis mechanisms.*

Mitochondrial Abnormalities Caused By ROS

Radiation, carcinogenic chemicals, viruses and chronic inflammation enable mitochondria to produce ROS, which impair breathing and cause a process called reverse reaction in mitochondria. Under normal conditions, DNA within the cell nucleus issues commands and sends commands to the mitochondrion in the cytoplasm. However, if the mitochondrion is damaged and breathing is threatened, the mitochondrion will apply information to the nucleus to activate the fermenting gene. As a result, a fermentation-based energy generation process is generated (see Picture 4). ROS cause severe damage to surrounding molecules. Some genes most closely related to cancer

> *Picture 5. The dual functions of extension (SNPH) in cancer. In a good microenvironment (left) and when nutrients and oxygen are present, high levels of SNPH 1 in mitochondrial inner and outer membranes support tumor cell proliferation through oxidative phosphorylation; and ROS decrease. At the same time, they prevent mitochondrial transport to the cortical cytoskeleton and tumor cell movement. Conversely, in an unfavorable microenvironment, low nutrition and hypoxia (right) reduce SNPH levels, resulting in less production of ATP, more production of ROS, and relief of inhibition on mitochondrial transport. As a result, tumor cell proliferation is reduced; but tumor cell movement and invasiveness are strengthened. [Quoted from: Altieri DC. Cell Mol Life Sci, 2018.]*

(oncogenes) are activated; and tumor suppressor genes are inhibited or mutate.

Cancer Is "Mitochondrial Disease"

As a result of mitochondrial damage, DNA becomes unstable in the nucleus; cells lose their unique shape; and cytoskeleton and extension (SNPH) fall into disorder and begin to replicate in an uncontrollable manner. Cancer is thus formed. Therefore, some people refer to cancer as a mitochondrial disease (see Picture 5). Reducing mitochondrial damage and protecting mitochondrial function help inhibit cancer cells, and also ensure that the body has sufficient energy supply to improve the patient's symptoms and quality of life.

How Does Molecular Hydrogen Enter Cells and Mitochondria?

Molecular hydrogen is the simplest, lightest and most abundant molecule in the universe, with a length of only 0.74 angstrom. The small size allows H_2 to rapidly penetrate cell membranes,

transfer itself to cytoplasm, and then enter mitochondria. This is a non-selective fast transfer process. Ilat experiments indicate that the hydrogen content in the myocardium increases immediately after a few minutes of hydrogen inhalation. In addition to molecular size, another factor affecting the entry of molecular hydrogen into mitochondria is its concentration gradient between the cytoplasm and mitochondrial matrix. Inhaled molecular hydrogen first enters blood circulation. Therefore, the greater the concentration gradient between the circulation and cells as well as mitochondria, the faster the transfer of hydrogen to mitochondria. This explains why in the treatment of diseases, in order to deliver fast biological effects, the concentration and amount of hydrogen supplied must not be too small.

> *Picture 6. Factors affecting the entry of molecular hydrogen into the mitochondria (molecule size, concentration gradient, electrochemical gradient, hepatic glycogen attraction). Green and red arrows indicate the facilitation and obstruction of entry respectively.*

Another factor that may affect mitochondria absorption of molecular hydrogen is its reactivity, which has the following characteristics:

1) In theory, molecular hydrogen can react with many biologically related elements and compounds (such as active and transition metals, non-metals, and organic compounds). But at room temperature or in the absence of a catalyst, the reaction rate is usually very low and can even be ignored.

2) Hydrogen molecules do not react with oxygen at room temperature, and do not interfere with redox reaction. In the presence of hydroxyl radicals (·OH), the biological reactivity of molecular hydrogen increases, neutralizing ·OH effectively.

3) Excessive elimination of ·OH in the cytoplasm increases the consumption of hydrogen molecules and reduces the speed of hydrogen delivery to mitochondria.

4) Molecular hydrogen imported in vitro accumulates with glycogen in the liver. Glycogen has a unique affinity for the accumulation and gradual release of molecular hydrogen. Since glycogen is mainly stored in the cytoplasm (many types of cells), the ability of carbohydrates (including glycogen) to polymerize hydrogen may have a negative effect on the transfer of hydrogen to mitochondria (see Figure 6).

Molecular Hydrogen Protects Mitochondria

Hydrogen protects mitochondria and nuclei from acute oxidative stress damage. In 2017, Serbian researcher Ostojic et al. reported that molecular hydrogen boosts mitochondrial function by maintaining mitochondrial membrane potential, increasing ATP production, and reducing organelle swelling. Hydrogen molecules maintain robust mitochondrial bioenergy through a range of possible mechanisms (see Picture 7).

> *Picture 7. Hydrogen regulates mitochondrial bioenergy in the following way: (1) Promote the release of growth hormone releasing peptide. On the one hand, hydrogen promotes phosphorylation under a series of mechanisms. On the other hand, hydrogen upregulates the activation of glucose transporter 1 and facilitates glucose uptake. (2) Up-regulate the expression of fibroblast growth factor 21, and promote oxidative phosphorylation. (3) Promote the activation of glucose transporter 4 by AMP activating enzyme; (4) Boost the activity of phosphatidylinositol 3-hydroxykinase; (5) Facilitate glucose transporter 4 by activating botulinum toxin substrate 1 or directly promoting glycogen synthase kinase gene, thereby promoting glucose metabolism. [Quoted from: Ostojic SM. Theranostics, 2017, 7(5): 1330-1332.]*

Mitochondria Regulate Immune Function

American researcher Buck et al. (2016) and Angajala et al. (2018) had detailed and precise exposition of the relationship between T cells and mitochondrial function. In anti-tumor immunity, T cell-mediated protective immunity plays a very important role and has a unique ability to proliferate at an unparalleled rate in adult organisms. A young T cell can clone millions of "armed" effector T cells in merely a few days. Mitochondria are important hubs for metabolic activity, anti-tumor immune response, and cell death. They reshape their structures to meet different needs. Effector T cells are associated with punctiform separate mitochondria, and memory T cells with confluent mitochondria. The former is the same as tumor cells. The metabolic characteristics of the Warburg Effect rely on aerobic glycolysis to provide energy. The latter is dependent on the process of oxidative phosphorylation. Once antigens are cleared, most effector T cells die. But long-lived memory T cells sustain and respond quickly when cancer recurs.

In 2014, Mexican researcher Bonifaz et al. expounded on the relationship between antigen-presenting cells (dendritic cells and macrophages) and mitochondrial function, believing that mitochondria control the functions of these two cells. Dendritic cells are professional antigen-presenting cells that help and enhance T cell anti-tumor immunity. Macrophages are classified into M1 and M2 types. M1 macrophages play an important role in anti-tumor immunity and inhibit tumor growth. M2 macrophages are the opposite. However, from the point of view that healthy mitochondria are essential for controlling cancer cell growth, mitochondria are of great significance to keeping normal immune balance in the body. It is also based on this view that hydrogen molecules are considered to play an evidently regulatory role in anti-tumor immunity by maintaining mitochondrial function.

Summary

From the perspective of treatment, exogenous hydrogen molecules should be perceived as valuable biomedical preparations. In certain sense, cancer is a metabolic disease rather than a genetic disease. Further research on the causal relationship between hydrogen molecules and metabolic abnormalities regulated by specific genes will undoubtedly contribute to the creation of innovative treatments for cancer.

— Xu Kecheng, Chen Jibing, Lv Youyong, Fei Sujuan

References

1. San-Millan I and Brooks GA. Reexamining cancer metabolism: lactate production for carcinogenesis could be the purpose and explanation of the Warburg Effect. Carcinogenesis, 2017, 38(2): 119-133.
2. Grazioli S and Pugin J. Mitochondrial Damage-Associated Molecular Patterns: From Inflammatory Signaling to Human Diseases. Front Immunol, 2018, 9: 832.
3. Zong WX, Rabinowitz JD, White E. Mitochondria and Cancer. Mol Cell, 2016, 61(5): 667-676.
4. Sabharwal SS and Schumacker PT. Mitochondrial ROS in cancer: initiators, amplifiers or an Achilles' heel? Nature reviews. Cancer, 2014, 14(11): 709-721.
5. Murphy MP. How mitochondria produce reactive oxygen species. Biochem J, 2009, 417(1): 1-13.
6. Jia D, Park JH, Jung KH, et al. Elucidating the Metabolic Plasticity of Cancer: Mitochondrial Reprogramming and Hybrid Metabolic States. Cells, 2018, 7(3).
7. Signorile A, Sgaramella G, Bellomo F, et al. Prohibitins: A Critical Role in Mitochondrial Functions and Implication in Diseases. Cells, 2019, 8(1).
8. Altieri DC. Mitochondrial dynamics and metastasis. Cell Mol Life Sci, 2018.

9. Ohsawa I, Ishikawa M, Takahashi K, et al. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med, 2007, 13(6): 688-694.
10. Ostojic SM. Targeting molecular hydrogen to mitochondria: barriers and gateways. Pharmacol Res, 2015, 94: 51-53.
11. Ostojic SM. Does H_2 Alter Mitochondrial Bioenergetics via GHS-R1alpha Activation? Theranostics, 2017, 7(5): 1330-1332.
12. Angajala A, Lim S, Phillips JB, et al. Diverse Roles of Mitochondria in Immune Responses: Novel Insights Into Immuno-Metabolism. Front Immunol, 2018, 9: 1605.
13. Buck MD, O'Sullivan D, Klein Geltink RI, et al. Mitochondrial Dynamics Controls T Cell Fate through Metabolic Programming. Cell, 2016, 166(1): 63-76.
14. Bonifaz L, Cervantes-Silva M, Ontiveros-Dotor E, et al. A Role For Mitochondria In Antigen Processing And Presentation. Immunology, 2014.

Hydrogen Molecules Save T cells and Regulate Immunity

Key points

- Hydrogen controls cancer by directly inhibiting tumor growth and probably also by relieving the body's immunosuppressive status. For the first time in 2019, we provided detailed clinical evidence that old T lymphocyte (to be apoptotic; with high expression of PD-1 on the surface) can be revived (PD-1 down-regulated), thereby prolonging progression-free survival (PFS) and overall survival (OS) for patients with advanced colon cancer.
- After hydrogen inhalation, the patients with elevated coenzyme (Co) Q10 had significantly higher OS. This indicates that hydrogen may improve T cell mitochondrial function and T cell survival rate by increasing CoQ10 content.

In recent years, the controlling effect of hydrogen on cancer has received great attention. It is generally believed that the antioxidant and anti-inflammatory effects of molecular hydrogen are the main mechanisms for inhibiting cancer cells and their microenvironment. However, studies lack as to whether molecular hydrogen has immunomodulatory effect. Our research team published a paper in 2019, presenting that hydrogen can restore cytotoxic T cells in depletion by improving mitochondrial function, and relieve the body's immunosuppressive status, thereby exerting a controlling effect on cancer progression.

What Is an Immune Cell?

The therapeutic effects and prognosis of cancer patients are strongly dependent on the patients' own immune function status. For immune function monitoring, an ideal and simple method is to detect the immune cell function by blood sampling (see Picture 1).

Cells in the peripheral blood of humans can be divided into three categories: red blood cells, white blood cells and platelets. The cells that exert immune function exist in white blood cells. Leukocytes can be divided into granulocytes, monocytes and lymphocytes according to volume and weight. Granulocytes are mainly responsible for controlling infection and removing foreign pathogenic microorganisms. Like scouts on constant patrol throughout the body, monocytes are mainly responsible for detecting tumors and reporting to lymphocytes. Lymphocytes locate tumors according to directions provided by monocytes and kill tumor cells in the body. By function, lymphocytes are further divided into three types, which are T cells from the thymus, B cells from the bone marrow, and NK cells (natural killer cells).

The formation of a tumor in the human body takes 10 to 20 years, during which time a single stem cell turns into a cancer stem cell and breaks up into billions of clusters that eventually

form tumors recognizable by the naked eye. During this period, there is a constant struggle between the killer T cells and tumor cells. If cells are depleted first, the patient is cured. If killer T cells are depleted first, the tumor grows rapidly and metastasizes.

> *Picture 1. Classification, biological markers and functions of human white blood cells (from the bone marrow hematopoietic stem cell): natural killer cell (anti-tumor); B lymphocyte (anti-infection); dendritic cell (anti-infection and anti-tumor); neutrophil (anti-infection); T lymphocyte (anti-tumor) → helper T cell, killer T cell, immature T cell. Lymphocytes identify cells mainly based on the difference of glycoprotein molecules (CD) expressed on the surfaces of cells. So far, nearly 300 CD molecules have been discovered. If labeled with one CD molecular marker, some cells can be distinguished from other cells (for example, CD19+ represents B cells). Some other cells require two or more molecular markers (for example, CD3+ and CD8+ represent killer T cells) to be distinguished.*

What Is an Immune Checkpoint and Its Ligand (PD-1/PD-L1)?

During evolution, human lymphocytes form immune checkpoints to detect whether the visitor is a friend or enemy. Lymphocytes let friends go and destroy enemies. This is a stabilizing mechanism in the body's immune system. The mechanism prevents activation of excessive T cells, which cause strong inflammation and injury to the immune system. Recognizing the characteristics of the human immune system, tumor cells disguise themselves fully when passing through checkpoints. They can manage to bypass and escape from human immune surveillance and killing. In 1992, Tasuku Honjo, a professor at Kyoto University in Japan, discov-

> *Picture 2. T cell expresses PD-1 molecules during activation; and there exist corresponding ligand PD-L1 on the surfaces of tumor cells. When PD-1 and PD-L1 bind, T cells stop attacking cancer cells and lose the ability to produce cytokines and activate immunity. (T cell receptor / tumor antigen; CD8+ T cell / cancer cell; PD-1 / PD-L1.)*

ered programmed death protein 1 (PD-1) on the surfaces of T cells. For this discovery, the professor won the Nobel Prize in Medicine and Physiology in 2018. In 1999, Chinese scientist Chen Lieping discovered the B7-H1 (also known as PD-1 ligand or PD-L1) protein on the surfaces of tumor cells. This key finding laid a foundation for applying PD-1 inhibitors to tumor immunotherapy. After being formed, tumors cells use this inhibitory pathway to suppress PD-1 T cell activation, thereby circumventing the immune system. Once the activity of immune checkpoints is inhibited, the immune brakes in the tumor microenvironment are released and re-activate the immune response of T cells to tumors; therefore achieving anti-tumor effects. The premise of using this category of drugs is to identify whether the enemy has been camouflaged (that is, immunohistochemical detection of tumor tissues), in order to ensure strong anti-tumor effects.

Lymphocytes are usually stored in the spleen and lymph nodes of the body, just as the army stays in a military camp in peaceful time. When monocytes detect tumors and report them to T lymphocytes, lymphocytes are dispatched from the military camp to destroy the tumors. After contact with the cells expressing the task, PD-1 is generated on the surface of the T cell. After contact with the cells expressing PD-L1, PD-1 is destroyed in their original places. That is, PD-1 enters apoptosis (exhaustion) state. For this reason, PD-1 is also regarded as a marker of T cells in depletion. Like a vulnerable point of T cells, PD-1 is easily caught and struck hard by tumor cells (see Picture 2).

Effect of Hydrogen on the Immune Cells of Cancer Patients

If PD-1 is widely expressed on the surfaces of killer T cells, it usually indicates that the patient's immune system has collapsed and survival is short. This conclusion has been verified true in patients with breast cancer, pancreatic cancer and gastric cancer. In 2016, American researcher Scharping et al. reported that there were metabolic defects in depleted T cells in both the tumors and blood. Such defects were characterized by easy aggregation of T cells, continuous deterioration of mitochondrial function, and continuously lower expression of PGC-1α. In the same year, Japanese researcher Kamimura et al. reported that hydrogen strongly activated signal molecule PGC-1α, which in turn up-regulated the respiration of mitochondria in T lymphocytes. In 2006, Handschin and Spiegelman reported that PGC-1α signaling enhanced adaptive thermogenesis action and glyconeogenesis; and reversed the dysfunction of killer T cells. The next question is: If the functions of killer T cells are restored, can the cells prolong progression-free survival (PFS) from tumors and overall survival (OS) of patients?

Clinical Outcome of Hydrogen Therapy for Stage IV Colon Cancer

In 2018, our research team reported the results of hydrogen inhalation by patients with stage IV colon cancer. A total of 55 patients participated in the study. Each patient inhaled hydrogen for 3 hours every day. The patients were followed up on for 3 consecutive years and were given regular blood tests to examine the expression of PD-1 on the surfaces of killer T cells. After continuous hydrogen inhalation, the proportions of PD-1+ T cell subtypes in the patients' blood dropped continuously by around 60% in a few months. The reason was that cells of PD-1+ subtype were continuously induced by hydrogen to be apoptotic; and PD-1 cells continued to be replenished. The patients with high proportions of exhaustive (PD-1+) CD8+ T cells in their peripheral blood had short PFS and OS (both 18 months). But the patients with high proportions of non-exhaustive (PD-1-) CD8+ T cells showed significantly prolonged PFS (> 40 months) and OS (46 months) (see Picture 3).

> *Picture 3. Patients with high levels of PD-1+ (depleted) CD8+ T cells in the peripheral blood have markedly shorter PFS and OS than those with low expression of PD-1+ CD8+ T cells. (Kaplan-Meier curves for tumor progression-free survival and overall survival; log rank p < 0.0001 for PFS, p = 0.004 for OS.)*

Mechanism of Hydrogen on Immune Function

From blood tests, we found that hydrogen treatment resulted in a decrease in the proportions of PD-1+ (exhaustive) CD8+ T cells and an increase in the proportions of PD-1- (non-exhaustive) CD8+ T cells in the peripheral blood of cancer patients (Picture 4). Hydrogen has two characteristics. First, it can only remove ·OH from four types of ROS. Known as a "villain" type of ROS, ·OH has the strongest oxidative capacity and is associated with cancer, diabetes and hypertension. Secondly, hydrogen boosts the function of lymphocyte mitochondria by activating PGC-1α. ·OH destroys mitochondrial DNA, causing mitochondrial dysfunction. Therefore, hydrogen protects CD8+ T cell mitochondria from DNA damage by clearing ·OH, and maintains the state of PD-1- for a long time. As the proportion of PD-1-

> *Picture 4. Change in the proportions of PD-1+ and PD-1- CD8+ T cells before and after treatment for 55 patients enrolled in the test (expression of PD-1+ CD8+ decreased in 35 patients; expression changes in 39 patients).*

cells increases, the proportion of terminal PD-1+ cells gradually decreases, therefore improving prognosis. This is attributed to hydrogen-induced activation of mitochondria.

> *Picture 5. CoQ10 is a transporter on the mitochondrial inner membrane, carrying electrons released by NADH2 (complex I) and FADH2 (complex II); and activates cytochrome C on compound III. After being activated, cytochrome C catalyzes the formation of water molecules by hydroxyl. Coenzyme Q10 can also activate hydrogen molecules on compound IV and enable hydrogen molecules to enter mitochondria on compound IV. (Electron transport chain: Complex I–V, CoQ, cytochrome C, ATP synthesis.)*

Coenzyme (Co) Q10 is an indispensable electron carrier in the mitochondrial respiratory chain (see Picture 5). It transfers the electrons in compounds I and II to compound III, and catalyzes the binding of ·OH with molecular hydrogen to form water molecules, thereby eliminating ·OH. Therefore, we believe that CoQ10 concentration in the peripheral blood can be regarded as a useful marker of mitochondrial function and as positively correlated with mitochondrial function.

Afterwards, we selected 25 patients and tested the expression of CoQ10 in their blood. Results showed that 18 cases (72%) of the patients had higher CoQ10 concentrations after hydrogen treatment. Compared with the patients (median OS = 8 months) with decreased CoQ10, the patients with elevated CoQ10 had markedly longer OS (>50 months). The test suggested that hydrogen might improve the mitochondrial function of T lymphocytes by increasing CoQ10 content, thereby improving patient survival (see Figure 6).

> *Picture 6. Changes of CoQ10 content in 25 patients before and after hydrogen treatment (left, 18 of 25 patients = 72% showed an increase); and correlations of the changes with survival period (right): high-CoQ10 group (n=18) vs. low-CoQ10 group (n=7), log rank p < 0.0001, median survival time 8 months.*

Summary

We first discovered immunological evidence that hydrogen inhalation prolongs the survival of tumor patients by increasing the number of highly active (PD-1-) T cells in the blood, reducing the proportion of exhaustive (PD-1+) T cells, and prolonging the survival and action of killing of killer T cells. As a result, patients' tumor progression-free survival and overall survival are significantly extended. The mechanism for achieving this effect is probably to increase the level of CoQ10 expression in the inner membranes of T lymphocyte mitochondria so that more molecular hydrogen enters mitochondria and neutralizes ·OH to protect the normal functions of mitochondria.

— (Japan, Akagi Junji). (Remarks: The article above is translated and sorted by Chen Jibing.)

References

1. Akagi J and Baba H. Hydrogen gas restores exhausted CD8+ T cells in patients with advanced colorectal cancer to improve prognosis. Oncol Rep, 2019, 41(1): 301-311.
2. Barber DL, Wherry EJ, Masopust D, et al. Restoring function in exhausted CD8 T cells during chronic viral infection. Nature, 2006, 439(7077): 682-687.
3. Ahmadzadeh M, Johnson LA, Heemskerk B, et al. Tumor antigen-specific CD8 T cells infiltrating the tumor express high levels of PD-1 and are functionally impaired. Blood, 2009, 114(8): 1537-1544.
4. Sun S, Fei X, Mao Y, et al. PD-1+ immune cell infiltration inversely correlates with survival of operable breast cancer patients. Cancer Immunol Immunother, 2014, 63(4): 395-406.
5. Zarour HM. Reversing T-cell Dysfunction and Exhaustion in Cancer. Clin Cancer Res, 2016, 22(8): 1856-1864.
6. Lu X, Yang L, Yao D, et al. Tumor antigen-specific CD8(+) T cells are negatively regulated by PD-1 and Tim-3 in human gastric cancer. Cell Immunol, 2017, 313: 43-51.

7. Scharping NE, Menk AV, Moreci RS, et al. The Tumor Microenvironment Represses T Cell Mitochondrial Biogenesis to Drive Intratumoral T Cell Metabolic Insufficiency and Dysfunction. Immunity, 2016, 45(2): 374-388.
8. Kamimura N, Ichimiya H, Iuchi K, et al. Molecular hydrogen stimulates the gene expression of transcriptional coactivator PGC-1alpha to enhance fatty acid metabolism. NPJ Aging Mech Dis, 2016, 2: 16008.
9. Handschin C and Spiegelman BM. Peroxisome proliferator-activated receptor gamma coactivator 1 coactivators, energy homeostasis, and metabolism. Endocr Rev, 2006, 27(7): 728-735.

Transforming Cancer Cells: Intimate Contact of Hydrogen with Cancer Cells

Key points:

- Cancer stem cells account for only a small part of the whole tumor, but play an important role in tumor survival, proliferation, metastasis and recurrence.
- When cultured in a culture container, ordinary cancer cells die due to lack of nutrition, but tumor stem cells form tumor spheres.
- The tumor spheres of ovarian cancer stem cells are reduced significantly in number and diameter after hydrogen is applied, indicating that hydrogen has certain inhibitory effect on tumor growth.

Many people turn pale at the mentioning of cancer. Although there have been constant efforts in the field of modern medicine, many lives are lost to cancer daily under the attack of cancer cells. Most cancer treatments focus on how to kill cancer cells. However, common treatments such as radiotherapy and chemotherapy after surgical resection kill cancer cells and meanwhile, cause great damage to normal cells and trigger serious side effects. A number of animal studies and preliminary studies of cancer patients suggest that hydrogen has potentially important application value in supplementing cancer treatment and mitigating the side effects of radiotherapy and chemotherapy. What is the interaction between hydrogen and tumor cells?

The seemingly identical tumors are actually composed of different types of cancer cells. There is a special type of cancer cells known as cancer stem cell, which accounts for only a small part of the whole tumor but plays an important role in tumor survival, proliferation and metastasis. Cancer stem cells have strong capacity to proliferate. On the one hand, they produce new cancer stem cells through cell division. On the other hand, they produce other types of tumor cells, that is, differentiated cells or cells in differentiation. As a result, tumor cell clusters are formed. Cancer stem cells maintain the vitality of tumor cell clusters through self-renewal and immortaliza-

Picture 1. Cancer Stem cell and Evolution (tumor stem cell → tumor cells → tumor).

tion. They represent the root causes of tumor recurrence and metastasis (see Picture 1).

In consideration of each tumor cell's capability of infinite multiplication, tumors treatments are generally aimed at killing as many tumor cells as possible. If the gross tumor volume shrinks, the corresponding treatment regimen is considered to be effective. However, most tumors recur after a period of time after treatment. Conventional treatments fail to completely kill tumor stem cells. Despite taking up only a small part of the tumor population, tumor stem cells survive because of their strong resistance to drugs. In addition, they are capable of unlimited

proliferation, sowing the seeds for subsequent recurrence. Therefore, it is necessary to find a way to treat cancer stem cells in the course of tumor treatment. If subdued, cancer stem cells have only limited ability to proliferate other cancer cells. In this case, even if the tumor size remains the same, the tumor will gradually degenerate and shrink, making it possible to eventually cure the tumor (see Figure 2).

> *Picture 2. Treatment targeted at tumor stem cells: precision treatment of tumor stem cells → tumor stem cell clearance → tumor suppression; versus traditional tumor treatment → tumor stem cell survives → tumor recurrence.*

We conducted certain studies of cancer stem cells. Cancer stem cells share one characteristic: When cultured in a culture container and if such nutrients as serum are not added to the container, ordinary cancer cells die due to insufficient nutrition, but cancer cells with stem cell characteristics form cell spheres known as tumor spheres. The effects of hydrogen on cancer stem cells can be observed by studying the capacity of tumor sphere formation. Our study of ovarian cancer, published in Translational Cancer Research (2018), indicated that hydrogen treatment significantly reduced the numbers and sizes of tumor spheres formed (see Figure 3). At the same time, we found that although the growth of cells did not change significantly in the presence of serum, some cells were not in the same state as the original cells. Similar phenomena have been observed in the studies of various tumor cells. Existing available findings suggest that putting hydrogen in "intimate" contact with cancer cells may help transform cancer cells in a way that the cancer cells lose certain properties possessed by cancer cells. Our deepened research is still ongoing regarding the long-term effect of this change, and the potential and practical applicability of hydrogen in combination with other methods to treat cancer stem cells.

> *Picture 3. Hydrogen inhibits tumor sphere formation (cancer cell PA-1 and cancer cell Hs38.T; non-hydrogen group vs. hydrogen group). Charts show the number of tumor spheres and the tumor average diameter (μ) are reduced in the hydrogen group. (Note: The pictures in the articles above are provided by Yves Mawulikplimi Adzavon and Zhao Pengxiang.)*

— Ma Xuemei

Hydrogen Molecules Scavenge Reactive Oxygen and Prevent Tumor

> **Key points**
> - Compared with general antioxidants, hydrogen is selective, diffusible and harmless. It might play a unique role in the prevention and treatment of tumors, such as avoiding the so-called oxidation-promoting effects shown by some antioxidants.
> - Due to the heterogeneity of tumors, anticancer drugs have their own scope of application. Hydrogen has demonstrated its effective, ineffective, and potentially harmful effects on tumor control.

In 1975, Dole et al. from Baylor University in the United States published an astonishing paper in the internationally known academic journal Science. The authors discovered that hydrogen at a high concentration of 97.5%, inhibited squamous cell carcinoma in mice. The discovery overturned the traditional perception that hydrogen was a physiologically inert gas. However, there had been no progress in the molecular mechanism by which hydrogen inhibited cancer.

Until 2007, Ota Satoshi et al., researchers from the Institute of Geriatrics of Nippon Medical School, found that a safe dose of hydrogen as low as 2% exerted biological effects; and proved that hydrogen had selective antioxidation. This opened a floodgate to research on the biological functions of hydrogen molecules.

It is well known that reactive oxygen species (ROS) maintain a dynamic balance between production and clearance; and keep at a relatively low level in normal cells. However, ROS metabolism in tumor cells is abnormal due to genetic changes. A high level of generated ROS facilitates tumor occurrence and development. This provides a possibility for hydrogen to prevent and treat tumors by eliminating ROS mechanism. In 2009, Japanese researcher Itoh et al. proposed the hypothesis that hydrogen can be used as a second messenger. The hypothesis was not recognized due to insufficient evidence. In fact, some ROS themselves can act as second messengers that activate intracellular proliferation and antioxidation information channels. The treatment targeted at tumor cell ROS is called oxidation-reduction therapy, which involves further increasing ROS (oxidation therapy) and decreasing ROS (reduction therapy). In a broad sense, the action of hydrogen on tumors is regarded as reduction therapy, because hydrogen is harmless to the human body and easily penetrates various biofilms. There are reasons to speculate that hydrogen, compared with other reducing agents, has promising prospects in tumor prevention and treatment.

Hydrogen and ROS

Hydrogen is considered to be an inert gas for a long time. For this reason, hydrogen is mixed with oxygen under pressure and used as a gas for divers to breathe underwater. Shigeo Ohta et al. used cell-containing and cell-free pure solutions; combined fluorescent probes, electron spin resonance and nitrite anions ($ONOO^-$). Studies of $ONOO^-$ are limited only to pure solutions without cells; and its effect is weak. Therefore, the primary target molecule of hydrogen is ·OH. This result confirms the long-standing speculation that hydrogen has reducing effects on tissue cells. More importantly, the antioxidant effect of hydrogen is limited only to direct clearance of ·OH and $ONOO^-$. Hydrogen cannot clear other reactive oxygen molecules (such as O_2 and H_2O_2) or reduce oxidation products generated from the combination of OH^- and other molecules. Also, hydrogen has no reducing effect on molecules in oxidative state in the oxidation respiratory chains of mitochondria (i.e. no interference with mitochondrial function). It is speculated that hydrogen may play a certain role in the free radical metabolism of cancer cells. Studies using intestinal adenocarcinoma Caco-2 cells indicate that hydrogen not only significantly reduces antimycin A-induced elevation of intracellular OH^- but also O_2 and H_2O_2 levels. At least in terms of malignant cells, this finding raises questions about the high selectivity of hydrogen in scavenging free radicals. From a study using inflammatory cells, Japanese researcher Itoh et al. found that hydrogen inhibited the activation of mast cells and it can be seen that hydrogen either had no effect on ROS elevation (macrophages), or inhibited NADPH oxidase activation (mast cells) while mainly reducing O_2 and H_2O_2 levels. According to Shigeo Ohta's research findings, hydrogen does not directly clear O_2 and H_2O_2. Therefore, the decrease in O2 and H_2O_2 levels is the result of hydrogen's inhibition of NADPH oxidase. Hydrogen has inhibitory effects on the information channels for activation of both inflammatory cells. Therefore, after ruling out the possibility that hydrogen directly removes ROS, researcher Itoh et al. proposed the hypothesis that hydrogen is a second messenger like CO, H_2S and NO in cells. Unfortunately, this hypothesis remains to be verified, because no specific target molecules have been identified. Considering the complexity of intracellular information transduction pathways, it can be expected that this will

be a difficult task and explains why there have been few studies and satisfactory results in this regard so far.

In addition, according to principles of biological pathology, the idea that hydrogen increases antioxidant molecules such as Nfr2 and SOD in cells seems to contradict the role of hydrogen in scavenging free radicals. Strengthened expression of aforementioned proteins is the result of cell reaction to ROS. If ROS are cleared by hydrogen in time, Nfr2 and SOD levels are expected to drop correspondingly. It seems unconvincing to explain this experimental phenomenon by stating that hydrogen maintains the ability of cells to respond to ROS by alleviating oxidative damage. Therefore, hydrogen may have other mechanisms of action in addition to directly scavenging free radicals. Inspired by the fact that metals bind with hydrogen molecules directly to produce stable compounds in organometallic chemistry, Chinese researcher Shi et al. put forward that the target molecules of hydrogen might be metal ions in the body's metalloprotein.

Observing hydrogen from the perspective of ROS may help understand the experimental phenomena of hydrogen. For example, ROS have a wide range of effects on cells, including the effects of causing direct oxidative damage and indirectly regulating signaling pathways. The influence of hydrogen on the pathophysiology of various tissue cells may ultimately be explained based on the biological effects of ROS. It can be expected that the studies of hydrogen and ROS complement and facilitate each other.

ROS and Tumor

ROS are mainly products of partial reduction of O_2 and include O_2, H_2O_2 and OH. Mitochondria are the major sites where ROS are produced. Certain biological oxidation reaction, such as β-oxidation of peroxisomes, also generates ROS. In addition, cells have a system for scavenging ROS, including enzyme antioxidants such as superoxide dismutase (SOD). Non-enzymatic antioxidants, such as NADPH, mainly provide reducing equivalents for the reduction reaction. ROS influence oxidation-reduction potential in cells and vice versa. The ratio of GSH to its oxide GSSG (GSH / GSSG) is the primary buffer pair for maintaining redox potential. NADPH is mainly produced by the phosphopentose pathway and represents an important factor in maintaining the stability of GSH / GSSG. The biochemical processes of glucose phosphopentose pathway (producing NADPH), oxidative phosphorylation (generating ROS), and protein anabolism (consuming NADPH) are coordinated with each other to maintain a balance of redox equivalents in cells. It can be seen that the regulation of intracellular ROS is a complex system.

(1) ROS abnormalities in some tumor cells: In the cell cycle, intracellular ROS levels increase steadily; and antioxidants can block cells in the late G1 stage. Early studies revealed that H_2O_2 in tumor cells increases steadily. Compared with progeny tumor cells, cancer stem cells have less ROS because of their strong antioxidant capacity. Gene mutation is the most important cause of ROS abnormalities (ROS increase) in tumor cells. Oncoproteins, such as RAS and MYC, also increase intracellular ROS. For example, RAS increases O_2 by strengthening NADPH oxidase (NOX) and reducing antioxidant molecule sestrin1. Excessive ROS increase cyclin D1 through ERK1/2, which in turn promotes RB phosphorylation. As a result, E2F1 is released, facilitating cell transformation from G1 to S phase. Tumor suppressor proteins such as RB and BRCA1 inhibit the production of ROS and promote their metabolism. For example, BRCA1 lowers intracellular ROS levels by up-regulating antioxidant gene glutathione S-transferase. Some oncoproteins and tumor suppressor proteins exert two-way

regulation on ROS and help stabilize ROS levels. In non-stress state, oncoprotein BCL-2 increases O_2 by strengthening oxidative phosphorylation of mitochondria. In stress state, BCL-2 suppresses excessive increase in O_2. Low and medium degrees of ROS elevation boost P53 expression and activate ROS clearance. If ROS levels are too high, P53 activates the pathway for ROS production and induces apoptosis by further increasing ROS. Moreover, tumor cell infiltration and metastasis (EMT) raise ROS levels due to changes in gene expression. Infiltration of tumor microenvironment cells, hypoxia and nutritional imbalance all contribute to an increase in ROS in tumor cells.

(2) Influence of ROS on cells (oxidation response of cells): The information channels activated by ROS not only regulate redox metabolism, but also exert effects on cell proliferation, apoptosis and aging. Therefore, ROS serve as second messengers. The important mechanism is that the cysteine sulfhydryl groups of the key structural domains of some proteins are easily oxidized reversibly by ROS, generating disulfide bonds (-SH+-SH→-S-S-). These proteins include transcription factors such as p53, information transduction proteins such as RAS, and proteasomes of inactivated proteins. The involved information molecules, such as HIF1, TGF-β/SMADs and KEAP1 / NRF2 compounds, activate cell stress channels, protect cells, inhibit apoptosis and promote angiogenesis. Activation of NF-κB is associated with synthesis and release of inflammatory factors. PKC-MAPK channels are involved in cancer cell proliferation. ROS synergistically activate proliferation channels and antioxidant/stress channels, allowing tumor cells to proliferate while avoiding apoptosis caused by excessive ROS. Therefore, some scholars regard the anti-oxidation system such as NRF2 in tumor cells as an oncoprotein component. Acute high levels of ROS directly disrupt macromolecular structures of cells and promote cell death, in addition to activating apoptotic channels. Tumor suppressor genes such as P53, some chemo drugs, and radiotherapy induce apoptosis by increasing ROS. ROS have different effects on tumor cells of different types and in different stages. For example, the decrease of O_2 suppresses melanoma but strengthens the invasion of fibrosarcoma cells. In early-stage tumors, ROS levels rise and induce apoptosis by activating P53 or other means. In progressive and advanced tumors, inactivation of the aforementioned pathways strengthens tumor cell tolerance to ROS. Moreover, refractory tumors or cancer stem cells are more resistant to ROS because of their strong antioxidant capacity.

(3) Prevention and treatment of tumors with ROS: ROS promote the occurrence and development of tumors, which provides a basis for reduction therapy. Compared with normal cells, tumor cells have high ROS levels and are more sensitive to further elevation of ROS. And oxidation therapy becomes an applicable option. Reduction therapy is designed to prevent the development of tumors by reducing ROS and inhibiting the activation of proliferation channels and DNA mutations. ROS scavengers, pro-ROS enzyme inhibitors, and anti-ROS enzyme activators can be used in reduction therapy. Significant effects have been observed in cell and animal experiments on reduction therapy; but the results from clinical studies (intake of vitamins A & E, selenium, and β-carotene) vary widely. Some studies even indicate that these antioxidants increase the incidence of cancer and weaken the effects of other anti-cancer treatments. This might be attributable to the diversity of tumor cell oxidation-reduction disorders. Oxidation therapy uses cytotoxicity of ROS to directly kill tumor cells, including by facilitating production of ROS and/or inhibiting clearance of ROS. With low ROS levels, cancer stem cells might be insensitive to treatments and survive eventually. Higher ROS levels might also induce more mutations in tumor cells and even normal cells, resulting in drug resistance. All the aforementioned scenarios put limits on the application of oxidation

therapy. Theoretically, it is speculated that if the tumor cells with ROS disorder are in early stages without metastasis, and DNA changes are small, anti-oxidation treatment might be helpful to prevent the occurrence and development of tumors. If DNA changes are big, and invasive metastasis has already occurred, a potentially more effective option is to increase ROS to promote apoptosis. At present, some indicators for predicting therapeutic effects have been selected. For example, monitoring the plasma thioredoxin and VEGF levels in patients with pancreatic and hepatic cancers helps evaluate the efficacy of thioredoxin inhibitors. PET scans monitor the glucose uptake rate and degree of hypoxia of tumor cells, therefore helping make ROS-promoting drugs related to metabolism and hy-

poxia more targeted. The screening and application of such indicators contribute to personalizing oxidation-reduction anti-tumor treatments.

Hydrogen and Tumor Prevention & Treatment

Some existing research findings have proven the role of hydrogen in cancer prevention and treatment.

In terms of prevention, the application of hydrogen water to mouse models of thymoma, hepatocellular carcinoma, and renal cell carcinoma induced by 60Co, streptozotocin and nitrogen triacetate lowers the incidence of tumors, reduces the volumes of tumors generated, and extends tumor latency. In the study where intraperitoneal injections of nitrogen triacetate were used to induce renal cell carcinomas in Wistar rats, short-term (1 week), medium-term (12 weeks) and long-term (6 weeks) observations were made respectively. Short-term intervention by hydrogen reduced the DNA uptake of 3-thymidine and activity of ornithine decarboxylase in renal homogenate. In addition, less $OONO^-$ and MDA were generated; and NADPH oxidase, xanthine oxidase, HO-1 and other enzymes showed weaker activity. Medium-term intervention of hydrogen reduced the expression of VEGF and PCNA in renal homogenates, as well as phosphorylation of STAT3. Long-term intervention of hydrogen reduced the incidence of renal cell carcinoma and average size of tumors. The above studies suggest that hydrogen lowers the incidence and severity of tumors in animal models through an antioxidation mechanism.

In terms of treatment, Dole et al. experimented on hairless mice with albino in 1975; induced squamous cell carcinoma of the skin in the mice by ultraviolet radiation; and observed the effects of high-pressure hydrogen (97.5% hydrogen, 2.5% oxygen, 8.28 barometric pressures) on the mice. The findings were surprising: Tumors shrank and some of them fell off and disappeared. Healing effect was achieved. But equally surprising is that there have been no similar reports afterwards. Studies in recent years used tumor cells mostly, and a few of them used mice as experimental subjects. The tumor cell strains used included colon adenocarcinoma, human tongue cancer, human lung adenocarcinoma, human fibrosarcoma, Ehrlich ascites tumor and the like. After hydrogen was added to the culture solution, the ability of tumor cells to metastasize (reflected in the number of colonies and number of cells per colony) and infiltrate (reflected in the number of cells passing through the reconstituted basement membrane) weakened; and the rates of cell degradation and apoptosis improved. Importantly, hydrogen reduced the number of colonies of human tongue cancer cell strain HSC-4 and the number of cells per colony, but did not impact colony formation from normal human tongue epithelial-like cell DOK. This indicated that hydrogen has selective anti-tumor effects. The anti-cancer effects of hydrogen involve decrease in intracellular ROS levels, weakened activity of MAPK, and strengthened activity of apoptotic enzymes such as Caspase

3. Hydrogen also reduces angiogenesis induced by A549 culture solution by inhibiting the transcription and secretion of VEGF in human lung adenocarcinoma cell strain A549.

It is worth noting that in spite of its function to promote apoptosis in cell strain colon 26, hydrogen does not increase the numbers and volumes of tumors induced by colon 26 injection; and does not improve the survival of tumor-bearing BALB/c mice. Hydrogen has no inhibitory effect on hepatoma cell strain HepG2. In oxidative stress induced by antimycin A, hydrogen protects intestinal adenocarcinoma Caco-2 cells instead. Moreover, as a means of auxiliary intervention, hydrogen helps reduce the side effects of other anti-tumor treatments. For example, hydrogen weakens renal toxicity of cisplatin in the treatment of transplanted tumors in C57BL/6 mice, and improves physical conditions and quality of life for patients with liver tumor radiotherapy. As shown by these studies, hydrogen does not exert any effect on tumors themselves.

The aforementioned studies reveal that hydrogen has preventive and therapeutic effects on some tumors; and exhibits selectivity for tumor cells. It has no therapeutic effect on some other tumors. However, under acute oxidative stress, hydrogen may protect tumor cells instead.

Summary

There are only a small number of studies on the prevention and treatment of tumors by hydrogen. And not many of the studies involved tumor cells and animals. Therefore, there are no clinical trial reports of randomized control. Future efforts can be made as follows: 1) Strengthen research at the cellular level; conduct studies on more and various types of tumor cells under conditions of oxidative stress and anti-oxidative stress; focus on therapeutic effects; and classify therapeutic effects into effective, ineffective and harmful types. 2) Strengthen the overall study on animals, and explore the biomedical effects of hydrogen in different stages of tumor development in terms of genes and phenotypes. 3) Strengthen clinical research, and carry out research on real data about the reality of tumor populations, especially the group with malignant tumors. In other words, it is important to keep complete records of each individual's treatment course, collect a certain number of medical records, and truly identify the therapeutic effects on certain types of tumor. 4) Explore the combined application of hydrogen and other anti-tumor treatments, such as hydrogen therapy combined with chemotherapy, radiotherapy or hyperthermia. Some animal and human experiments involve a wide range of topics and require multidisciplinary researchers to work together.

— *Qin Shucun, Zhou Xiao*

References

1. Dole M, Wilson FR, Fife WP. Hyperbaric hydrogen therapy: a possible treatment for cancer. Science, 1975, 190(4210): 152-154.
2. Ohsawa I, Ishikawa M, Takahashi K, et al. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med, 2007, 13(6): 688-694.
3. Itoh T, Fujita Y, Ito M, et al. Molecular hydrogen suppresses FcepsilonRI mediated signal transduction
and prevents degradation of mast cells. Biochem Biophys Res Commun, 2009, 389(4): 651-656.
4. Itoh T, Hamada N, Terazawa R, et al. Molecular hydrogen inhibits lipopolysaccharide/interferon gamma-induced nitric oxide production through modulation of signal transduction in macrophages. Biochem Biophys Res Commun, 2011, 411(1): 143-149.

5. Shi P, Sun W, Shi P. A hypothesis on chemical mechanism of the effect of hydrogen. Med Gas Res, 2012, 2(1): 17.
6. Diehn M, Cho RW, Lobo NA, et al. Association of reactive oxygen species levels and radioresistance in cancer stem cells. Nature, 2009, 458(7239): 780-783.
7. Kopnin PB, Agapova LS, Kopnin BP, et al. Repression of sestrin family genes contributes to oncogenic Ras-induced reactive oxygen species up-regulation and genetic instability. Cancer Res, 2007, 67(10): 4671-4678.
8. Chen ZX and Pervaiz S. Bcl-2 induces pro-oxidant state by engaging mitochondrial respiration in tumor cells. Cell Death Differ, 2007, 14(9): 1617-1627.
9. Vousden KH and Ryan KM. p53 and metabolism. Nature reviews. Cancer, 2009, 9(10): 691-700.
10. Watson J. Oxidants, antioxidants and the current incurability of metastatic cancers. Open Biol, 2013, 3(1): 120-144.
11. Kurz EU, Douglas P, Lees-Miller SP. Doxorubicin activates ATM-dependent phosphorylation of multiple downstream targets in part through the generation of reactive oxygen species. J Biol Chem, 2004, 279(51): 53272-53281.
12. Dempsey EC, Newton AC, Mochly-Rosen D, et al. Protein kinase C isozymes and the regulation of diverse cell responses. Am J Physiol Lung Cell Mol Physiol, 2000, 279(3): L429-L438.
13. Gorrini C, Harris IS, Mak TW. Modulation of oxidative stress as an anticancer strategy. Nat Rev Drug Discov, 2013, 12(12): 931-947.
14. Hurd SL, Grant JW, Ridnour LA, et al. Increased manganese superoxide dismutase expression suppresses the malignant phenotype of human melanoma cells. Proc Natl Acad Sci USA, 1993, 90(7): 3113-3117.
15. The alpha-tocopherol, beta-carotene lung cancer prevention study: design, methods, participant characteristics, and compliance. The ATBC Cancer Prevention Study Group. N Engl J Med, 1994, 4(1): 1-10.
16. Marinho HS, Real C, Cyrne L, et al. Hydrogen peroxide sensing, signaling and regulation of transcription factors. Redox Biol, 2014, 2(23): 535-562.
17. Kawai D, Takaki A, Nakatsuka A, et al. Hydrogen-rich water prevents progression of nonalcoholic steatohepatitis and accompanying hepatocarcinogenesis in mice. Hepatology, 2012, 56(3): 912-921.
18. Saitoh Y, Yoshimura Y, Nakano K, et al. Platinum nanocolloid-supplemented hydrogendissolved water inhibits growth of human tongue carcinoma cells preferentially over normal cells. Exp Oncol, 2009, 31(3): 156-162.
19. Runtuwene J, Amitani H, Amitani M, et al. Hydrogen-water enhances 5-fluorouracil-induced inhibition of colon cancer. PeerJ, 2015, 3: e859.
20. Zhao P, Jin Z, Chen Q, et al. Local generation of hydrogen for enhanced photothermal therapy. Nat Commun, 2018, 9(1): 4241.

Extra: Jiuhua Mountain

Jiuhua Mountain is one of the four Buddhist holy mountains in China. Li Bai, a well-known Chinese poet from Tang Dynasty, praised Jiuhua Mountain with a poem: "Sailing down the Jiujiang River the other day, I saw the Jiuhua Peaks in the distance. Looking like a heavenly river hanging in heaven, its green water embroidering cottonrose hibiscuses." From the foothills to the Tiantai Peak, Jiuhua Mountain is home to many ancient temples and rare historical relics. It is particularly world-famous for the whole-body Buddhist relics from Buddhist spiritual masters. After passing away, Eminent monks remain in the same original physical shape and as natural as though they were living. Such bodies are whole-body Buddhist relics. For example, Master Haiyu lived in a cave in Jiuhua Mountain for 100 years. He never went downhill, receive visitors, or trained any apprentices. He lived until 126 years old. Master Haiyu had strict self-discipline and found companionship in mist and clouds in the twilight throughout

the year. Living a life of transcendency and purity, he ate sealwort and roots of kudzu vines when hungry; and drank

> *(Picture by Ni Shengpei)*

mountain spring water when thirsty. According to legend, one-time consumption of raw sealwort was sufficient to keep Master Haiyu from feeling hungry for 7 days.

Zhang Xichun, a famous doctor from the Qing Dynasty in China, wrote in his book *Records of Tradition Chinese and Western Medicine in Combination*: "One can find hydrogen in the center of a furnace. There is also hydrogen in the human abdomen. Dried rehmannia root or astragalus membranaceus leads hydrogen to the lung, where hydrogen binds with inhaled oxygen and transforms into water. Moreover, hydrogen drives liquid in the stomach upwards and gasifies the lower energizer (the portion located below the stomach) without causing frequent urination. Therefore, it can be used to treat diabetes." Master Haiyu did not feel hungry for 7 days after eating sealwort. Is sealwort possibly astragalus membranaceus? Astragalus membranaceus leads hydrogen to the lung. Is that why Master Haiyun had no hunger after eating it?

> **Tribute to Jiuhua Mountain**
> — *Zhong Zimei*
> The legend of immortal body
> Is a metaphor for Buddhist relics from the human body?
> Or is an astonishing representation of Zen in reality?
> Refreshing mountain and gentle breeze
> A journey without end for millennial years
> The clarity and wonder of the soul
> Not subject to stain, dirt or damage
> Permanent or not
> As condescendingly as Jiuhua Mountain
> Seen through the mortal world

PART 3 — Experimental Study

Experimental Study of Hydrogen Cancer Control

> **Key points**
> A large number of experiments have been conducted on the effects of hydrogen molecules, including the effects of hydrogen gas and hydrogen-rich water on cancer cells and their microenvironment. The experiments involved tumors of the liver, kidney, tongue, thymus, lung and colon. According to experimental findings, the effects can be summarized as follows: a) Facilitate the clearance of reactive oxygen species (ROS); inhibit the formation, division, migration and infiltration of tumor cells; and promote cancer cell apoptosis; b) Protect normal cells and exert antagonistic effects on oxidative stress, apoptosis, inflammation and tumor occurrence; c) Regulate the expression and signaling pathways of related molecules; and protect mitochondria; d) Down-regulate inflammation-related factors and inhibit inflammation; e) Hydrogen combined with chemo drugs boost the tumor suppressing effect of the latter; f) Relieve the adverse effects of radiotherapy and chemotherapy.

Tumor cells generate a large quantity of ROS, which far exceed the number of ROS in normal cells. These ROS promote tumor cell proliferation, DNA synthesis and angiogenesis, leading to invasive growth and distant metastasis of cancer cells. The first report about hydrogen's role in directly inhibiting tumor cell growth was published in 1975. American researcher Dole et al. discovered that the squamous cell carcinoma inoculated on mice's skin shrank under the action of high-pressure hydrogen. In recent years, literature reviews on hydrogen's direct inhibitory effects on tumor formation or progression have been published continuously in professional medical journals at home and abroad.

In 2009, Japanese researcher Saitoh et al. used hydrogen-rich water or cisplatin separately or in combination on the test group to intervene in human tongue cancer cell (HSC-4) growth in vitro. Normal tongue epithelioid cells (DOK) were applied to the control group. The experiment led to the following findings: 1) Hydrogen-rich water used alone or in combination with cisplatin (combination group) accelerated the clearance of ROS in HSC-4. The combination group showed faster ROS clearance. 2) Combined use of hydrogen-rich water and cisplatin suppressed the growth rate and narrowed the growth area of HSC-4 cells. It had no inhibitory effect on DOK cell growth. It can be seen that with the help of hydrogen-rich water, cisplatin delivers stronger killing effects on tumors, indicating that cisplatin and hydrogen-rich water have great synergistic effects on tumor suppression. Cisplatin, if used together with hydrogen-rich water, only kills tumors instead of normal cells, indicating that hydrogen-rich water can offset the side effects of chemo drugs on normal cells. In terms of mechanism: 1) Since ROS is involved in the occurrence of almost all tumors, using hydrogen-rich water alone or together with cisplatin accelerates the clearance of ROS in HSC-4. This is probably because hydrogen-rich water inhibits tumor mutation and metastasis and strengthens anti-tumor effects. 2) Using hydrogen-rich water only is not enough to achieve good therapeutic effects on tumors. Combined treatments deliver significantly better anti-tumor effects than hydrogen alone. 3) Cisplatin and other chemo drugs cause side effects mainly because they induce normal cells to produce a large number of ROS. This in turn causes cytotoxicity, which justifies the necessity for patients with chemotherapy to take antioxidant therapy for rehabilitation.

In 2011, Chinese researcher Zhao et al. induced tumors in the thymus of mice through multiple times of irradiation with radioactive particles and meanwhile, gave the mouse models intraperitoneal injections of hydrogen-rich saline. The condition of thymic lymphoma was monitored. After 30 weeks, the following discovery was made: 1) The mice with hydrogen-rich saline injection had significantly lower rate of tumorigenesis (38%) than the group with mere irradiation (60%), and most of their tumors did not recur. 2) Different methods were used to detect the levels of intracellular and extracellular ROS for the hydrogen-rich saline group and control group respectively 4 hours after irradiation. Blood was drawn from the mice; and peripheral blood mononuclear cells were separated. Results showed that the mice treated with hydrogen-rich saline had notably less ROS in cells than the control group. Moreover, the former group showed significantly higher serum SOD and total GSH concentrations as well as markedly lower MDA concentration than the latter group. The results above indicated that the antioxidant status of the hydrogen-rich saline group improved significantly, leading to lower degree of DNA damage. In terms of the mechanism of action: 1) Tumors were induced by radiotherapy for the reason that a large number of toxic ROS were continuously generated in normal cells. Hydrogen-rich saline inhibited tumor mutation and metastasis by controlling ROS, which probably represented an important reason for the decline in radiation-caused tumorigenesis. 2) The use of mere hydrogen-rich water significantly reduced the level of ROS generated by peripheral blood leukocytes and plasma, thereby slowing down and even stopping tumor formation. This indicated that molecular hydrogen was resistant to radiation damage and protective of normal tissues.

In 2012, Japanese researcher Kawai et al. prepared STAM mouse models of fatty liver and liver cancer, which were induced by single or combined use of hydrogen-rich water and pioglitazone (PGZ, an anti-diabetic and anti-inflammatory drug to prevent fatty liver) as well as by MCD diet. The blank control group was given injections of drinking water. After 8 weeks, it was found that: 1) Liver function damage of the hydrogen-rich water group and hydrogen-rich water + PGZ group was significantly less than that of the control group. The combination group exhibited the best therapeutic effects and least increase in transaminase. 2) Liver tumors in the hydrogen-rich water group and hydrogen-rich water + PGZ group were markedly smaller than those of the control group. The hydrogen-rich water group showed the best effect, with its biggest tumor being smaller than those of the other two groups. This indicated that hydrogen-rich water exerted antagonistic action on the oxidative stress, apoptosis, inflammation and tumor occurrence of liver cells. In terms of the mechanism of action: 1) Hydrogen water was not as effective as PGZ in decreasing cholesterol but was better in antioxidation and prevention of liver cancer, indicating that hydrogen water can be a new means to prevent fatty liver. There have been reports about similar effects on models of acute liver injury induced by diabetes and carbon tetrachloride. 2) The hydrogen water group showed significant down-regulation of lipid metabolism-related genes AOX and FAT in modelled animals.

This indicated that hydrogen water may play a role in inhibiting PPAR signaling pathway, thereby suppressing the enhancement of mitochondrial oxidase and fatty acid oxidation. 3) In the experiment, the hydrogen water + PGZ group showed stronger tumor-suppressing capacity than the PGZ group, suggesting that there existed synergy between the two methods. The hydrogen water group had better anti-tumor results than the combination group, possibly because hydrogen had many effect targets and sometimes interplayed with PGZ. For example, single use of hydrogen water delivered the best effects in reducing liver 8-OhdG and peripheral blood ROM, indicating that hydrogen water used alone was stronger in protecting

DNA from oxidative damage than hydrogen water used in combination with PGZ. 4) TNF-α and IL-6 played a central role in liver cell apoptosis and fatty liver occurrence. This revealed that the mechanism of action of hydrogen water involved suppressing gene expression of the two cytokines. 5) PCNA expression reflected the proliferative activity of liver cells. Of all the groups, only the hydrogen water group showed significantly lower PCNA expression in liver cells, indicating that hydrogen water exerted strong effects on controlling cell proliferation.

In 2013, Chinese researcher Li et al. reported that drinking hydrogen-rich water helped prevent renal damage and renal cell carcinoma induced by ferric nitrilotriacetate (Fe-NTA) in mouse models. Findings of the experiment were as follows: 1) The hydrogen-rich water group had noticeably lower degrees of renal damage and normal levels of serum creatinine and urea nitrogen. 2) Hydrogen-rich water protected renal function by: a) reducing the formation of lipid peroxidase and peroxynitrite, and inhibiting oxidative stress; b) activating NAPDH oxidase and xanthine oxidase to boost the activity of catalase and restore the functions of mitochondria in kidney cells; c) reducing the expression of NF-kB, IL-6 and MCP-1 in renal cells as well as the accumulation of macrophages in the kidney, thereby suppressing inflammation induced by Fe-NTA. 3) The expression of VEGF and PCNA in kidney cells as well as STAT3 phosphorylation were inhibited. 4) The incidence of renal cancer (22%) in the hydrogen-rich water group was lower than that (54.8%) in the control group. The former group had lower onset rate than the latter. Regarding the mechanism of renal cancer pathogenesis: 1) The mechanism of Fe-NTA induction model is continuous iron deposition in renal tubular cells. In this study, hydrogen water reduced iron content in the kidneys of model animals and lowered the levels of $OONO^-$ and MDA in the kidney, demonstrating strong ability to control oxidative stress. 2) HO-1 gene is highly expressed in various tumor models, and promotes angiogenesis and tumor proliferation and metastasis. This was greatly illustrated by the observation in the experiment that HO-1 expression in the kidneys of hydrogen water-injected animals dropped significantly. 3) Inflammation plays a central role in the pathogenesis of renal cancer. In the experiment, the expression of IL-6 and MCP-1 in the kidneys of model animals decreased significantly after hydrogen-rich water intake. This reflects the anti-cancer ability of hydrogen-rich water. 4) Abnormal elevation of VEGF in tissues is associated with accelerated growth and metastasis of renal carcinoma. Another mechanism of action might be responsible for VEGF expression decrease in the kidneys of model animals after consumption of hydrogen-rich water in the experiment. 5) Activation of the 5STAT signal is widely present in a variety of tumor cell lines and primary tumors. After treatment with hydrogen water, model animals showed less STAT expression, which resulted in a decline in IL-6 level. This is considered to be one of the mechanisms of action of hydrogen.

In 2015, Japanese researcher Runtuwene et al. applied high-pressure hydrogen-rich water separately or in combination with 5-fluorouracil (5-FU) to model animals with tumors from colon cancer cell line (Colon 26) that was induced to grow in vitro and induced in vivo. The control group consisted of model animals without such intervention. The results showed: 1) Oral intake of high-pressure hydrogen water alone facilitated the apoptosis of Colon 26 tumor cells, because the expression of p-AMPK, AIF and Caspase 3 that induced apoptosis in cells increased significantly. When used together with 5-FU, high-pressure hydrogen water exerted stronger killing effects on cancer cells in vitro. 2) The animals that took ordinary drinking water, ordinary hydrogen water, and high-pressure hydrogen water survived for 9, 11 and 13 days respectively on average. Survival time was prolonged by 18 to 20 days for the animals to which high-pressure hydrogen water and 5-FU were applied in combination. From the aforementioned results, it can be concluded as follows: 1) Since the tumor of this model

progresses rapidly, hydrogen water alone is not enough to achieve good therapeutic effects. It must be used together with 5-FU to considerably prolong survival of test animals. 2) When hydrogen is applied in low doses, its free radical-scavenging effect helps protect various cells, including tumor cells. At high concentrations of hydrogen, there are significantly more free radicals in tumors than normal cells. This induces tumor cell apoptosis. 3) In experiments in vitro, both types of hydrogen water increased the expression of p-AMPK, AIF and caspase 3 in Colon 26 cells, which might be attributed to the mechanism that hydrogen water induces apoptosis of Colon 26 cells. It is highly likely that the tumor generates a large number of ROS, which are cleared by H_2. Then, caspase 3 pathway and lipid oxidative stress are initiated to induce tumor cell apoptosis.

In 2018, Chinese researcher Wang et al. reported that: 1) After a hydrogen-oxygen mixture at different concentrations was introduced into two lung cancer cell culture systems (A549 and H1975) through hydrogen-oxygen nebulizers, cell division, migration and infiltration were greatly suppressed; and cell apoptosis accelerated. The reason was that the expression of NIBPL, SMC3/5/6, Cyclin D1, and CDK4/6 genes in the cells decreased. 2) Mice with lung cancer were treated with hydrogen nebulizers (2 hours a day for 4 weeks consecutively) and intraperitoneal injections of chemo drug cisplatin respectively. The control group was given injections of saline. Compared with the control group, the cisplatin group had tumors reduced by 75% and the H_2 group by 25%. Both groups showed lower expression of proteins Ki-67, VEGF and SMC3, which were positively correlated with tumor growth. The cisplatin group had bigger decrease in expression of the proteins.

— *Chen Jibing, Mu Feng*

References

1. Gunther S, Ruhe C, Derikito MG, et al. Polyphenols prevent cell shedding from mouse mammary cancer spheroids and inhibit cancer cell invasion in confrontation cultures derived from embryonic stem
cells. Cancer Lett, 2007, 250(1): 25-35.
2. Maulik N and Das DK. Redox signaling in vascular angiogenesis. Free Radic Biol Med, 2002, 33(8): 1047-1060.
3. Nonaka Y, Iwagaki H, Kimura T, et al. Effect of reactive oxygen intermediates on the in vitro invasive capacity of tumor cells and liver metastasis in mice. Int J Cancer, 1993, 54(6): 983-986.
4. Dole M, Wilson FR, Fife WP. Hyperbaric hydrogen therapy: a possible treatment for cancer. Science, 1975, 190(4210): 152-154.
5. Saitoh Y, Yoshimura Y, Nakano K, et al. Platinum nanocolloid-supplemented hydrogendissolved water inhibits growth of human tongue carcinoma cells preferentially over normal cells. Exp Oncol, 2009, 31(3): 156-162.
6. Szatrowski TP and Nathan CF. Production of large amounts of hydrogen peroxide by human tumor cells. Cancer Res, 1991, 51(3): 794-798.
7. Dhar A, Young MR, Colburn NH. The role of AP-1, NF-kappaB and ROS/NOS in skin carcinogenesis: the JB6 model is predictive. Mol Cell Biochem, 2002, 234-235(1-2): 185-193.
8. Yao X, Panichpisal K, Kurtzman N, et al. Cisplatin nephrotoxicity: a review. Am J Med Sci, 2007, 334(2): 115-124.
9. Zhao L, Zhou C, Zhang J, et al. Hydrogen protects mice from radiation induced thymic lymphoma in BALB/c mice. Int J Biol Sci, 2011, 7(3): 297-300.
10. Little JB. Radiation carcinogenesis. Carcinogenesis, 2000, 21(3): 397-404.
11. Siegler R, Harrell W, Rich MA. Pathogenesis of radiation-induced thymic lymphoma in mice. Journal of the National Cancer Institute, 1966, 37(2): 105-121.

12. Barcellos-Hoff MH, Park C, Wright EG. Radiation and the microenvironment - tumorigenesis and therapy. Nature reviews. Cancer, 2005, 5(11): 867-875.
13. Liu C, Cui J, Sun Q, et al. Hydrogen therapy may be an effective and specific novel treatment for acute radiation syndrome. Med Hypotheses, 2010, 74(1): 145-146.
14. Qian L, Cao F, Cui J, et al. Radioprotective effect of hydrogen in cultured cells and mice. Free Radic Res, 2010, 44(3): 275-282.
15. Qian L, Cao F, Cui J, et al. The potential cardioprotective effects of hydrogen in irradiated mice. J Radiat Res, 2010, 51(6): 741-747.
16. Kawai D, Takaki A, Nakatsuka A, et al. Hydrogen-rich water prevents progression of nonalcoholic steatohepatitis and accompanying hepatocarcinogenesis in mice. Hepatology, 2012, 56(3): 912-921.
17. Kamimura N, Nishimaki K, Ohsawa I, et al. Molecular hydrogen improves obesity and diabetes by inducing hepatic FGF21 and stimulating energy metabolism in db/db mice. Obesity (Silver Spring), 2011, 19(7): 1396-1403.
18. Sun H, Chen L, Zhou W, et al. The protective role of hydrogen-rich saline in experimental liver injury in mice. J Hepatol, 2011, 54(3): 471-480.
19. Yakaryilmaz F, Guliter S, Savas B, et al. Effects of vitamin E treatment on peroxisome proliferator-activated receptor-alpha expression and insulin resistance in patients with non-alcoholic steatohepatitis: results of a pilot study. Intern Med J, 2007, 37(4): 229-235.
20. Wigg AJ, Roberts-Thomson IC, Dymock RB, et al. The role of small intestinal bacterial overgrowth, intestinal permeability, endotoxaemia, and tumour necrosis factor alpha in the pathogenesis of non-alcoholic steatohepatitis. Gut, 2001, 48(2): 206-211.
21. Anty R, Bekri S, Luciani N, et al. The inflammatory C-reactive protein is increased in both liver and adipose tissue in severely obese patients independently from metabolic syndrome, Type 2 diabetes, and NASH. The American journal of gastroenterology, 2006, 101(8): 1824-1833.
22. Li FY, Zhu SX, Wang ZP, et al. Consumption of hydrogen-rich water protects against ferric nitrilotriacetate-induced nephrotoxicity and early tumor promotional events in rats. Food Chem Toxicol, 2013, 61(248-254).
23. Toyokuni S. Iron-induced carcinogenesis: the role of redox regulation. Free Radic Biol Med, 1996, 20(4): 553-566.
24. Nuhn P, Kunzli BM, Hennig R, et al. Heme oxygenase-1 and its metabolites affect pancreatic tumor growth in vivo. Mol Cancer, 2009, 8(37).
25. Mantovani A. Molecular pathways linking inflammation and cancer. Current molecular medicine, 2010, 10(4): 369-373.
26. Li C, Liu B, Dai Z, et al. Knockdown of VEGF receptor-1 (VEGFR-1) impairs macrophage infiltration, angiogenesis and growth of clear cell renal cell carcinoma (CRCC). Cancer Biol Ther, 2011, 12(10): 872-880.
27. Huang M, Page C, Reynold RK. Constitutive activation of STAT3 oncogene product in human ovarian carcinoma cells. Gynec Oncol, 2000, 79(67-73).
28. Lin TS, Mahajan S, Frank DA. Stat signaling in the pathogenesis and treatment of leukemias. Oncogene, 2000, 19(2496-2504).
29. Runtuwene J, Amitani H, Amitani M, et al. Hydrogen-water enhances 5-fluorouracil-induced inhibition of colon cancer. PeerJ, 2015, 3(e859).
30. Ishikawa T, Tanaka Y, Ishitsuka H, et al. Comparative antitumor activity of 5-fluorouracil and 5'-deoxy-5-fluorouridine in combination with radiation therapy in mice bearing colon 26 adenocarcinoma. Japanese journal of cancer research: Gann, 1989, 80(6): 583-591.
31. Tominaga T, Yoshida Y, Kosaki G, et al. Effect of 5-fluorouracil and UFT on experimental liver metastasis model of colorectal cancer using mouse colon 26 cells. Japanese journal of cancer research: Gann, 1993, 84(7): 783-786.
32. Nakashima-Kamimura N, Mori T, Ohsawa I, et al. Molecular hydrogen alleviates nephrotoxicity induced by an anti-cancer drug cisplatin without compromising anti-tumor activity in mice. Cancer Chemother Pharmacol, 2009, 64(4): 753-761.
33. Simone CB, 2nd, Simone NL, Simone V, et al. Antioxidants and other nutrients do not interfere with chemotherapy or radiation therapy and can increase kill and increase survival, Part 2. Altern Ther Health Med, 2007, 13(2): 40-47.

34. Jin C and Reed JC. Yeast and apoptosis. Nat Rev Mol Cell Biol, 2002, 3(6): 453-459.
35. Farmer EE and Mueller MJ. ROS-mediated lipid peroxidation and RES-activated signaling. Annual review of plant biology, 2013, 64(429-450).
36. Wang D, Wang L, Zhang Y, et al. Hydrogen gas inhibits lung cancer progression through targeting SMC3. Biomed Pharmacother, 2018, 104(788-797).

Cytological and Animal Experiments of Hydrogen in Inhibiting Lung Cancer

Key points

- The effect of hydrogen molecules on human lung adenocarcinoma cell A549 was studied in terms of cell proliferation, cell tumor formation, cell viability, cell cycle & apoptosis, and cell metastasis.
- In an experiment on nude mice with femted subgingival tumors, it was found that the nude mice that inhaled hydrogen-oxygen mixture had notably smaller tumor masses than the normal group. From the perspective of cell morphology, cancer cells decreased in number and exhibited vacuolization after hydrogen was applied.
- Cytological research findings indicate that hydrogen molecules reduce the proliferation rate of A549 cells and inhibit the capacity of cell migration. The experiments below suggest that hydrogen inhalation has adjuvant therapeutic effects on patients with non-small cell lung cancer. This has provided certain experimental basis for applying hydrogen molecules to preventing and treating lung cancer.

Effects of Hydrogen on Experimental Tumor Formation

Method: Six BALB/c nude mice were divided into Group A and Group B, which represented normal group and hydrogen inhalation group (inhalation of hydrogen-oxygen mixture) respectively.

Tumors formed from injection of tumor cells: After proliferating to a certain number, a549 cells were dissociated and collected by trypsin. After being observed in SPF environment for 3 days, the test nude mice were in good conditions and ready to be inoculated. In a super clean bench, 75% alcohol was used to disinfect the skin on the injection sites of the nude mice. Then, each of the mice received injection into the hind limb with a cell suspension containing 5×10^6 cells. With the piercing point around 1cm from the injection point, skin bumps appeared on the mice. After measures were taken to prevent the liquid from leaking out, the mice were given skin disinfection and were monitored for tumor formation status.

Intervention method: Group A nude mice were not given any treatment after tumor formation, and were kept under normal environment. Group B nude mice were given inhalation of hydrogen-oxygen mixture (66.7% H_2 + 33.3% O_2) every day after tumor formation for 6 hours per day at the flow rate 3 liter/min for a total of 14 days.

Experimental results: Tumors of the normal group grew slowly and gradually 3 weeks ago, but exhibited fast growth after 3 weeks. Compared with the normal group, the hydrogen inhalation group showed continuously slow growth of tumors (see Figure 1). The tumors eventually formed in hydrogen inhalation group mice were significantly smaller than those of the normal group (see Picture 2).

> *Picture 1. Growth curve of tumors in nude mice of the normal group and hydrogen inhalation group. (Accompanied by a data table of weekly tumor length (mm), width (mm), weight (g) and volume (mm^3) over weeks 1–5 for the normal and 66.7%-hydrogen groups; the per-cell*

> *values are too small to transcribe reliably from the photograph.) With the passage of time after tumor formation, the difference in tumor growth rate between the two groups became more and more significant. The hydrogen-inhalation group showed much slower formation of tumor than the normal group.*

> *Picture 2. The left picture shows the tumor masses of the normal group (A) and the hydrogen inhalation group (B). Group A has markedly bigger tumors than Group B. The right picture shows a nude mouse in the hydrogen inhalation group; and no clear sign of tumor is observed.*

> *Picture 3. Significant differences in the appearance of organs (such as the lung, liver and intestine) between the normal group (A) and the hydrogen inhalation group (B) nude mice.*

> *Picture 4. Significant differences in vascular proliferation of tumors between the normal group (A) and the hydrogen inhalation group (B) nude mice.*

Moreover, during the experiment, it was found that the normal group ate much less than the hydrogen inhalation group. In terms of anatomy generally, the lungs, livers and intestines of the normal group were significantly enlarged, whereas the organs of the hydrogen inhalation group appeared relatively normal (see Picture 3).

In addition, it was discovered that there was more microvascular proliferation in the tumor growth areas of the normal group, but the hydrogen inhalation group had fewer microvessels (see Picture 4).

Cell Morphological Observation

Method: After proliferating to a certain number, A549 cells were dissociated and collected by trypsin; and then put in two 35mm dishes. The cells were cultured in two groups: normal cell incubator group and hydrogen-oxygen mixture incubator group (66.7% H_2 + 33.3% O_2). The culture solution was changed every two days. Upon cell overgrowth, cell passage cultivation followed. Passage A549 cells were cultured in the same environment as their parent cells. The two groups of cells were observed continuously until the 10th generation.

Results: The two groups showed no significant difference in first-generation cultured cells, with similar proliferation and good cell status (see Figure 5a). In the 5th generation, the A549 cells in the hydrogen-oxygen mixture group displayed inhibited proliferation, larger cell volumes, and much cell death (see Picture 5b). In the 10th generation, more vacuolization occurred to the cells of the hydrogen-oxygen mixture group (see Picture 5c).

> *Picture 5. Growth of A549 cells in normal environment and environment with hydrogen-oxygen mixture (first-, fifth- and tenth-generation cultured cells). It was found that after the 5th generation, A549 cell growth was inhibited and cell vacuolization increased significantly.*

Cell Immunofluorescence

Method: A549 cells in logarithmic growth phase were collected and counted. After being diluted to 5 × 10^4 /mL with whole serum culture medium, the cell suspension was inoculated on the cell slides of six-pore plates. The cells were then placed in CO_2 cell incubators for culture for 24 hours. According to experimental grouping, the culture medium in each pore was replaced with normal culture medium or hydrogen-containing medium. The cultivation continued for 24 hours. Then, the cells were added with anti-Vimentin antibody (PBS and

antibody were added for the blank control group and experiment group respectively) and diluted fluorescent secondary antibody successively. After that, Hoechst was added for nuclear staining. The cell slides were taken out, observed under a fluorescence microscope, and photographed.

Results: This experiment showed the effects of hydrogen molecules on the growth of A549 cells. Compared with the control group cultured in ordinary medium, the hydrogen group (treated with hydrogen molecules) demonstrated slower growth of cancer cell strains (treated with staining) from the first to tenth generation of A549 cells. Red fluorescence (BrdU) of A549 cells decreased significantly, indicating that hydrogen inhibited cell growth (see Picture 6).

> *Picture 6. Immunofluorescence of A549 cells that grew in two different conditions. It was found that cell growth slowed down and was inhibited after the fifth generation (P5). Control: Control group, normal medium culture; H2: cultured in environment with hydrogen-oxygen mixture. BrdU: marks S-phase cells; Hoechst: mainly stains cell nuclei and functions to locate; Merge: overlap of two types of staining.*

A549 Cell Cycle Detection

Method: A549 cells in logarithmic growth phase were collected and counted. The cell suspension was diluted to 2 × 10^5 /mL with whole serum culture medium, and then was inoculated on six-pore plates. The cells were then placed in CO_2 cell incubators for culture for 24 hours. According to experimental grouping, the culture medium in each pore was replaced with normal culture medium or hydrogen-containing medium. The cultivation continued for 24 hours. After that, the fixed cells were taken out and centrifuged, with stationary liquid removed. The cells were then added with 200 μl of cell cycle detection reagent for flow cytometry.

Results: As shown in Picture 7, compared with normal medium culture, hydrogen-treated A549

> *Picture 7. Differences in the growth of A549 cells in two different gas environments (normal culture conditions vs. hydrogen environment; flow cytometry of DNA content). The cell cycle is blocked at G2/M phase: the hydrogen treatment group had considerably less S-phase cells and more G2/M-phase cells.*

cells decreased significantly in Phase S and increased notably in Phase G2/M. This suggests that hydrogen may have affected cell cycle, inhibited cancer cell proliferation and promoted cell apoptosis by reducing cell division in Phase S and cell block in Phase G2/M.

Observation of Cell Migration

Method: Cell scratch test was adopted. Cells in logarithmic growth were sampled and counted after dissociation. Then, the cells were cultured in an incubator containing 5% CO_2 saturated humidity. After the cells grew to a level that fully covered the bottom of the plate, 200 μl yellow gun head was put perpendicularly to the pore plate to create cell scratch wounds along the straight line. Each scratch wound was made as equally wide as possible. After that, normal culture medium with of 1% FBS and hydrogen-containing culture medium were added. The culture plate was placed in the incubator to continue culture. Photos of the cells were taken 0 and 24 hours respectively after the scratch. The effects of hydrogen on the migration capacity of tumor cells were observed through time-lapse recording of cell migration in the first 24 hours.

Results: After 24 hours of culture, the cells began to move toward the middle. After hydrogen-containing culture medium was added, the migration capacity of A549 cancer cells weakened significantly.

Conclusion

This experiment tested the effects of hydrogen-oxygen mixture prepared by the hydrogen-oxygen nebulizer manufactured by Shanghai Asclepius Meditec on human lung adenocarcinoma cell A549 from the aspects of cell proliferation, tumor formation, activity, cycle & apoptosis and metastasis & invasion. The findings indicated that hydrogen helps significantly change the morphology of tumor cells; change cell cycles; facilitate apoptosis; and inhibit tumor cell growth, movement, invasion and migration.

Appendix

Wang Dongchang and other researchers from China conducted a similar experiment using a hydrogen-oxygen nebulizer manufactured by Shanghai Asclepius Meditec, and explored the inhibitory effects of hydrogen on the growth and invasiveness of non-small cell lung cancer. Since the researchers used the same hydrogen-oxygen nebulizer as in the experiment above, the abstract of their paper is excerpted below (the original paper was published in International Journal of Respiration, issue 38: 561-565, 2018) with the authors' consent:

> *Picture 8. Tumors formed after inoculation of tumor cells in two groups of nude mice (hydrogen group vs. control group). Compared with the control group, the hydrogen group had obviously smaller tumors.*

Method:

Tumor-bearing nude mouse models were established. A549 cell suspension from cultured human lung cancer cell strain was inoculated subcutaneously into the back of the forelimb of BALB/c male nude mice. When the transplanted tumor reached an average diameter of around 4mm, the nude mice were divided into control group and hydrogen group. The control group was given inhalation of air, and hydrogen group 66% hydrogen plus 34% oxygen (using Asclepius Meditec nebulizer model: AMS-H-01) for 2 hours daily on average. After 4 weeks, the nude mice were sacrificed. Tumors in their bodies were taken out and measured in size and weight. Tissues of the mice were sampled for histological, immunohistochemical and molecular tests.

Results:

(1) Growth and weight of transplanted tumors: In the first week after tumor cell inoculation, there was no statistically significant difference in the sizes of tumors between the control group and hydrogen group. As hydrogen intervention was applied for a longer time, the sizes of tumors gradually displayed difference, which became of statistical significance up to 2 weeks. Four weeks after intervention, the tumors in the hydrogen group were considerably smaller than those in the control group (see Picture 8). And the difference between the two groups in tumor growth became bigger with time (see Picture 9).

(2) Histological and immunohistochemical changes of the tumors: Hematoxylin and eosin (H&E)-stained

> *Picture 9. Upper: Tumor size (cm^2) changes in two groups of nude mice — over time, the difference between the hydrogen group and control group became more significant, with the hydrogen group changing slower than the control group (a: $p < 0.05$). Lower: Tumor body*

> *weight (g) — tumors in the hydrogen group weighed clearly less than those in the control group (a: $p < 0.05$).*

> *Picture 10. Histology of tumors in two groups of nude mice: A and B, control group; C and D, hydrogen group (H&E staining; A and C × 100, B and D × 400). For the control group, histology showed typical images of cancer. Cancer cells in the hydrogen group were arranged neatly and were less carcinogenic.*

tumor tissues in the control group showed cells closely arranged at relatively large density and with significant heterogeneity in morphology, which were typical manifestations of cancer. The hydrogen group had cells with relevantly small density and less heterogeneity, indicating that the tumors were less carcinogenic than those in the control group (see Picture 10).

(3) The content of Ki-67, COX-2 and VEGF protein in tumor tissues was detected. The control group showed many deeply stained nuclei; while less deep staining of the three proteins was found in the hydrogen group (see Picture 11).

> *Picture 11. Immunohistochemical staining of tumors in two groups of nude mice (control group vs. hydrogen group; rows: Ki-67, COX-2, VEGF). The hydrogen group had lower content of Ki-67, COX-2 and VEGF protein (degree of staining), which reflect the invasiveness or malignancy of cancer, than the control group. COX-2: Cyclooxygenase-2; VEGF: Vascular endothelial growth factor.*

Conclusion:

Hydrogen inhibits the growth of non-small cell lung cancer and weakens the invasiveness of cancer. The inhibitory effects are associated with the down-regulation of Ki-67, COX-2 and VEGF expression.

— *Wang Ying, Lai Peizhen*

Experimental Study of Hydrogen in Controlling Ovarian Cancer

> **Key points:**
> - Ovarian cancer, breast cancer, and cervical cancer are the top three gynecological cancers, of which breast cancer has the highest incidence and ovarian cancer highest mortality.
> - Our study suggests that hydrogen inhibits tumor cell invasion and migration.

Ovarian cancer is often regarded as "an invisible killer" among female tumors. Since the ovaries are located deep in the pelvis, and initial symptoms of ovarian cancer are mild, only ovarian cysts can be detected generally in routine gynecological examinations. Ovarian cysts are caused by many factors. For example, irregular menstrual cycles may cause ovarian cysts, which disappear after the menstrual period. However, cysts with pathological changes are different and do not disappear after menstruation. In this case, further examination is required to determine whether the cysts are merely normal cysts or representations of ovarian cancer. In many cases, even if ovarian cancer progresses to an advanced stage, the ovary itself does not grow bigger. As a result, ovarian cancer is easily overlooked. Ovarian cancer is usually definitively diagnosed at its terminal stage when the optimal opportunity of treatment has been lost. According to statistics, more than 220,000 women worldwide suffer from ovarian cancer each year; and around 60% of them die of ovarian cancer. In the West, there is a saying

among gynecologists: If "cancer" is one of the most terrible words in the English language, then "ovary" is the worst attribute before it for most women. In China, the treatment of ovarian cancer is also not optimistic, with 5-year survival rate lower than 40%.

Despite the rapid development of new treatments and emergence of one after another new drug for most types of cancer, there have not been new first-line drugs for ovarian cancer in the past nearly 30 years. It can be said that ovarian cancer is difficult to detect, difficult to diagnose, and difficult to treat, making it a cancer with low survival rate.

The potential therapeutic effects of hydrogen on cancer has long been studied. As early as 1975, a study from the United States showed that high-pressure hydrogen exerted therapeutic effects on skin squamous cells. In the study, researchers exposed tumor-bearing mice to eight atmospheres of 97.5% hydrogen for 16 consecutive days. The results showed that hydrogen had good anti-tumor effects. Within 10 days of hydrogen treatment, tumor tissues turned black, and tumors shrank or even fell off respectively in some mice. In addition, no mice displayed adverse effects. The aforementioned study was published in the famous journal Science. Nevertheless, since high-pressure hydro-

gen must be stored using special equipment and poses potential safety hazards, there have been few subsequent studies on high-pressure hydrogen being used for cancer treatment. In 2007, Japanese scholar Shigeo Ohta discovered that even a small amount of hydrogen exerted biological effects. Since then, the application of hydrogen to cancer-related areas has gradually become a topic of cancer research. Our team conducted a study with animal and cell experiments. The results indicated that hydrogen has potential value for treating ovarian cancer. Our research findings have been published in the journal Translational Cancer Research. The following is a brief introduction to the study.

To study the effects of hydrogen on ovarian cancer, we first established mouse models of ovarian cancer. Two types of ovarian cancer cells Hs38.T and PA-1 were inoculated subcutaneously into experimental mice. Tumor growth was detected two weeks later. Then, these tumor-bearing mice were randomly divided into two groups. One group (hydrogen group) was given hydrogen inhalation 3 times a day (30 minutes each time) for 6 weeks consecutively (66% hydrogen + 34% oxygen mixture supplied with Shanghai Asclepius Meditec nebulizer). The other group (control group) breathed normal air. The two groups of mice were kept under the same conditions except for the difference of air inhaled. As shown as in Picture 1, the results indicated that the 6-week-long hydrogen inhalation significantly inhibited tumor growth and reduced the tumor volume by 32.3% on average. The expression of Ki67, one of the indicators that characterizes the degree of malignant tumor proliferation, decreased by 30%. This indicated that tumor growth was significantly suppressed. Moreover, it was found that CD34, a marker for tumor angiogenesis, dropped by 74%, indicating that hydrogen reduced tumor angiogenesis. Blood vessels are responsible for transporting oxygen and nutrients. The organs and tissues of the human body are entangled with dense blood vessels. Tumor tissues are no exception. Since cancer cells grow and thrive, their demand for oxygen and nutrients is particularly high. Therefore, during growth, tumors release some signals that promote blood vessel growth, allowing more blood vessels to extend into the tumors and transport more oxygen and nutrients to the tumors. We speculate that after hydrogen inhalation is applied, tumor blood vessels decrease significantly, leading to less oxygen and nutrients supplied to the tumors. Ultimately, tumor growth is slowed down. That might be one of the reasons that hydrogen inhalation inhibits ovarian cancer growth.

> *Picture 1. Effects of hydrogen inhalation on tumor growth and tumor-related indexes in mice with ovarian cancer. (A) Tumor volume (cm^3) over time, control vs. hydrogen inhalation group; (B) Ki67 immunostaining, control vs. hydrogen group; (C) proportion of Ki67- and CD34-positive cells, with CD34 staining shown.*

In addition to superior reproductive capacity, tumor cells have many "skills" that normal cells do not possess. For example, tumor cells invade adjacent tissues and metastasize to distant places, which is the main cause of tumor recurrence and metastasis as well as ultimate death of patients. In addition to its function of inhibiting tumor growth, can hydrogen be used in other ways to treat tumor cells? Our team carried out a cell experiment to study the effects of hydrogen on the biological functions of tumor cells. We prepared a hydrogen-rich cell culture medium. Then, we cultured ovarian cancer cells with a normal cell culture solution and a hydrogen-rich culture solution respectively. As shown in Picture 2, the experimental results suggested that hydrogen significantly inhibited the growth of tumor cells in vitro (see Figure 2, C, D) and migration (see Figure 2, E, F) showed that ovarian cancer cells, after being cultured in hydrogen-rich culture medium, had considerably weakened abilities to invade and migrate. This suggests that hydrogen inhibits the metastasis of ovarian cancer cells. It is important to know that tumor recurrence and metastasis are the main causes of

> *Picture 2. Effects of hydrogen on the biological functions of ovarian cancer cells (cancer cell PA-1 and cancer cell Hs38.T): (A, B) cancer cell growth rate over time, non-hydrogen vs. hydrogen group; (C, D) invasion assay and number of invading tumor cells; (E, F) tumor cell migration rate and migration photos at 0 hours and 24 hours, non-hydrogen vs. hydrogen group.*

> *Picture 3. Effects of hydrogen on ovarian cancer stem cells (cancer cell PA-1 and cancer cell Hs38.T): (A) tumor sphere photos, non-hydrogen vs. hydrogen group; (B) number of tumor spheres and average diameter of tumor spheres (μ).*

> *Picture 4. Effects of hydrogen on cervical cancer stem cells (non-hydrogen group vs. hydrogen group): (A) tumor sphere photos; (B) number of tumor spheres; (C) average diameter of tumor spheres.*

patients' death. How to prevent tumor recurrence and metastasis has always been the biggest challenge in cancer treatment. Although our study revealed the effects of hydrogen on inhibiting tumor cell invasion and migration, more experimental evidence is needed, especially evidence from animal and clinical trials. If it can be verified in the future that hydrogen inhibits the recurrence and metastasis of tumors, cancer patients will undoubtedly benefit tremendously.

In cell experiments, we also made an important discovery. That is, hydrogen has significant inhibitory effects on cancer stem cells. As shown in Picture 3, the experiment on tumor sphere formation in vitro indicated that hydrogen therapy significantly reduced the number and size of tumor spheres, suggesting that hydrogen potentially suppressed cancer stem cells. Tumor stem cells are the cells with stem cell characteristics in tumors. They feature strong proliferation and self-renewal as well as the potential of multi-directional differentiation. In other words, tumor stem cells not only self-replicate but also transform into other types under external stimulation. Current theories about cancer stem cells maintain that tumors are actually composed of many different types of cells, of which only cancer stem cells are highly carcinogenic and highly invasive. Despite their small proportions in tumor cells, cancer stem

cells play an important role in tumor recurrence and metastasis. The formation of new tumors after tumor recurrence and metastasis requires the participation of cancer stem cells. We discovered inhibitory effects of hydrogen on cancer stem cells in cervical cancer cells, in addition to those in ovarian cancer cells. As shown in Picture 4, the experiment on tumor sphere formation in vitro demonstrated that the number and size of tumor spheres decreased significantly in cervical cancer-derived HeLa cells cultured in a hydrogen-rich medium than in normal cultured cells.

Considering the findings of current research on hydrogen in other tumors, we believe that hydrogen may play an important role as an adjuvant treatment for existing cancer treatments.

First, hydrogen has direct effects on tumor cells themselves. Based on the above experimental results, inhaling hydrogen has considerably inhibit tumor growth in mice with ovarian cancer. The anti-cancer effects of hydrogen might be closely related to hydrogen's functions in controlling tumor angiogenesis; weakening tumor cell invasion and migration; and inhibiting tumor stem cell growth. It should be noted that our research findings are merely the results of animal and cell experiments. There are many types of ovarian cancer; and the pathological process of each patient is not necessarily the same. Because of such diversity and complexity, more experimental evidence, especially clinical research evidence, is needed for applying hydrogen to ovarian cancer treatment.

Second, hydrogen might help patients strengthen their own immunity and overcome cancer by themselves. Clinical studies from Japanese scholars revealed that T lymphocytes responsible for killing cancer in cancer patients are prone to aging and depletion. After patients inhale hydrogen, T lymphocytes are reactivated and increased in number. This change highly concerns patients' prognosis. In other words, the anti-cancer effects of hydrogen might be achieved by improving patients' immunity.

Third, hydrogen mitigates the toxic side effects of tumor radiotherapy and chemotherapy. There have been many research reports in this regard. For example, clinical studies long time ago suggested that hydrogen eases the side effects of radiotherapy and chemotherapy. There is more evidence in animal experiments, for example, evidence showing the capacity of hydrogen to control renal toxic injury caused by chemo drug cisplatin.

In short, the effects of hydrogen on cancer prevention and treatment are worth looking forward to, but must not be overstated. In the future, it is necessary to carry out basic and clinical research on hydrogen-based cancer prevention and treatment in a more in-depth and systematic way. This is of great significance and value for both hydrogen medical research and cancer patients.

— Xie Fei, Zhao Pengxiang, Ma Xuemei

References

1. Dole M, Wilson FR, Fife WP. Hyperbaric hydrogen therapy: a possible treatment for cancer. Science, 1975, 190(4210): 152-154.
2. Ohsawa I, Ishikawa M, Takahashi K, et al. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med, 2007, 13(6): 688-694.
3. Shang L, Xie F, Li JL, et al. Therapeutic potential of molecular hydrogen in ovarian cancer. Transl Cancer Res, 2018, 8(1).
4. Shang Lei, Li Jiala, Su Zehua, et al. Effects of Molecular Hydrogen on Cervical Cancer Cells. Biotechnology Advances, 2018, 8: 1-9.
5. Akagi J and Baba H. Hydrogen gas restores exhausted CD8+ T cells in patients with advanced colorectal cancer to improve prognosis. Oncol Rep, 2019, 41(1): 301-311.

6. Nakashima-Kamimura N, Mori T, Ohsawa I, et al. Molecular hydrogen alleviates nephrotoxicity induced by an anti-cancer drug cisplatin without compromising anti-tumor activity in mice. Cancer Chemother Pharmacol, 2009, 64(4): 753-761.

PART 4 — Case Studies

"Real World" Survey of Hydrogen Cancer Control: Follow-up Report on 82 Cases of Sdvanced Cancer Patients with Hydrogen Inhalation

Summary

Background and purpose: The treatment of progressive cancer is a huge challenge and requires new ideas and strategies. Treatment means beyond drugs have become the targets of people's exploration. Hydrogen has been proven with antioxidant and anti-inflammatory effects. And the occurrence and progression of cancer are closely associated with peroxidation and inflammation. Therefore, hydrogen may play a role in controlling cancer.

Cases and methods: A retrospective and prospective follow-up study of cancer patients with self-hydrogenation recovery, mainly from progressive cancer, was performed with reference to the Real World Evidence (RWE) approach. A total of 82 patients who inhaled hydrogen by themselves were observed. The patients had stage III and IV cancers, mainly including lung cancer, liver cancer, gynecological malignant tumor, pancreatic cancer, gastrointestinal malignant tumor and urinary malignant tumor. All the patients met the following conditions: 1) The patients had been definitively diagnosed with and pathologically confirmed to have stage III and IV cancers. 2) The patients had received conventional treatments, including surgery, radiotherapy and/or chemotherapy; but did not respond well to the treatments or experienced recurrence. Or conventional treatments were not suitable to the patients due to general physical conditions or complicating disease. Or the patients rejected conventional treatments. 3) The patients had different symptoms, such as poor appetite, fatigue, insomnia or pain. 4) The patients had existing tumors or/and abnormal tumor markers. 5) The patients, ranging from 20 to 80 years old, showed no obvious signs of heart, brain, lung or kidney failure, or mental disorder; and were capable of expressing themselves. 6) The patients inhaled hydrogen-oxygen mixture (66.7% hydrogen + 33.3% oxygen) from hydrogen-oxygen nebulizers, with a gas flow rate at 3000 mL/min. They inhaled hydrogen for at least 1.5 hour a day for more than 3 months consecutively.

Results: 1) The follow-up continued for 3 to 46 months, with a median time of 6 months. During the follow-up period, 12 patients died in stage IV, including 4 patients with pancreatic cancer. The death was caused by tumor progression in 5 cases; infection in 2 cases; and liver failure, intestinal obstruction, upper gastrointestinal bleeding, PD-1 antibody-related pneumonia and euthanasia in one case, respectively. 2) The life quality of 30 patients was prospectively evaluated according to EORTC QLQ-C30 assay. As hydrogen inhalation continued, the patients exhibited more significant improvements in their functional areas and symptoms. All aspects improved after 4 weeks of hydrogen inhalation, with the most notable improvement in fatigue, insomnia, loss of appetite and pain. 3) Physical fitness assessment was performed on all patients according to Zubrod-ECOG-WHO (ZPS, 5-point method). The results showed that 41.5%, 34.1% and 24.4% of the patients had improved, stable and worse physical conditions, respectively. There was no significant difference in the improvement rate between patients with merely hydrogen inhalation and patients with hydrogen inhalation combined with other therapies; and between patients with stage III and IV cancers. However, significant difference was observed between groups of different tumors. Patients with lung cancer showed the highest improvement rate, whereas those with pancreatic cancer and gynecologic cancer had the lowest. 4) The follow-up covered changes in tumor markers, including alpha-fetoprotein (AFP), carcinoembryonic antigen (CEA), CA19-9, CA125, CA153, and CA724. Among the 58 cases in which one or more tumor markers rose, 36.2%, 15.5% and 48.3% had tumor marker decline after hydrogen inhalation, stable tumor markers, and worse tumor markers, respectively. Tumor markers began to fall in Day 13 to 45, with a median time of 23 days. There was no significant difference in the degree of tumor marker decline between patients with hydrogen inhalation

and those treated with hydrogen combined with therapies. However, patients with different types of tumors demonstrated significant differences. The highest decline of tumor markers was found in patients with lung cancer; and the lowest in patients with pancreatic and hepatic malignant tumors. 5) Among the 80 cases with visible tumors on medical images, one case (1.3%) showed complete remission (CR); 15 cases (18.8%) partial remission (PR); 30 cases (37.5%) stable disease (SD); and 34 cases (42.5%) progressive disease (PD) after hydrogen inhalation. The overall control rate was 57.5%. Patients exhibited CR and PR in Day 21 to 80, with a median time of 55 days after hydrogen inhalation. The disease control rate displayed no statistically significant difference between patients who had merely hydrogen inhalation and those receiving hydrogen inhalation in combination with other treatments. Disease control rates were 83.0% and 47.7% in stage III and IV patients, respectively. And the difference was significant. Except for pancreatic cancer, other types of tumors showed no significant difference in disease control rate. The control rate was the lowest in patients with pancreatic cancer. 6) Adverse effects: No hematological toxicity was found. Stomach upset, dizziness, headache, and epistaxis were seen in individual cases at the beginning of hydrogen inhalation and disappeared spontaneously soon.
Conclusion: According to the findings of the follow-up above, inhalation of hydrogen (hydrogen and oxygen mixture) improved the quality of life and physical conditions as well as controlled disease progression for patients with progressive cancer. To ensure good therapeutic effects, the inhaled hydrogen must be supplied at a sufficient concentration so that it enters tissues as soon as possible. Moreover, hydrogen inhalation must be continued for sufficient time so that the doses accumulate to deliver certain effects. At the same time, inhaled oxygen might exert synergistic effects. In view of the great difficulty in treating progressive cancer and high safety of hydrogen, the simple method of inhaling hydrogen is worthy of further study and should be applied to clinical rehabilitation and treatment as soon as possible.

Introduction

Cancer has become the leading cause of human death. Its incidence is still on the rise. In the past half century, significant progress has been achieved in basic and clinical cancer research. The incidence and mortality of certain cancers have declined, mainly because of primary prevention (such as smoking cessation) and early screening of cancer. The decline is rarely attributed to a particular drug.

Treatment is like a double-edged sword. All common treatments, including radiotherapy, chemotherapy, fine needle aspiration and surgery, lead to increase in circulating tumor cells and accelerated progression and distant metastasis of cancer. Precision treatments based on molecular detection results have not benefited a majority of cancer patients. Precision drugs can be found for only 3% to 13% patients. Some studies suggest that targeted therapy results in cancer metastasis by inducing a large secretion of proteomes.

To overturn the harsh reality of current cancer treatment, there must be new ideas and strategies for cancer treatment. Treatment means beyond drugs have become the targets of people's exploration.

Hydrogen molecular biology and hydrogen medicine are young yet fast-developing disciplines. The occurrence and progression of cancer are closely associated with peroxidation and inflammation. Therefore, hydrogen is potentially effective in controlling cancer. Hydrogen has been proven with antioxidant and anti-inflammatory effects.

In recent years, many cancer patients inhale hydrogen by themselves. Referring to the Real World Evidence (RWE) research method, we conducted a retrospective and prospective

follow-up study of cancer patients who rehabilitated from hydrogen inhalation. The patients were mainly those with progressive cancer. Our research report is as follows.

Cases and Methods

Follow-up subjects: The follow-up included two parts. (1) A retrospective survey was conducted on the patients who inhaled hydrogen at home from April 2014 to April 2018. Mainly, we made house visits to patients; found and read relevant medical records in hospitals; and carried out retrospective studies. (2) After May 2018, patients inhaled hydrogen mainly in our health studio and office of the rehabilitation association. Some of them inhaled hydrogen by themselves at home. Specialized doctors were assigned as volunteers to take care of and guide patients; review relevant information as needed; and conduct prospective observational study. A total of 68 patients were involved in the prospective study.

All the patients who agreed to participate in the follow-up study and were included in this statistical analysis met the following conditions: 1) The patients had been definitively diagnosed with and pathologically confirmed to have stage III and IV cancers. 2) The patients had received conventional treatments, including surgery, radiotherapy and/or chemotherapy; but did not respond well to the treatments or experienced recurrence. Or conventional treatments were not suitable to the patients due to general physical conditions or complicating disease. Or the patients rejected conventional treatments. 3) The patients had different symptoms, such as poor appetite, fatigue, insomnia or pain. 4) The patients had existing tumors or/and abnormal tumor markers. 5) The patients, ranging from 20 to 80 years old, showed no obvious signs of heart, brain, lung or kidney failure, or mental disorder; and were capable of expressing themselves.

Hydrogen inhalation method: A hydrogen-oxygen nebulizer [AMS-H-01 hydrogen-oxygen nebulizer (Shanghai Asclepius Meditec Co., Ltd., China)] was used. The patient took a sitting or lying position; put on a nasal tube or a face mask; and inhaled hydrogen-oxygen mixture through breathing. The mixture contained 66.7% hydrogen and 33.3% oxygen. Gas flow rate during inhalation was 3000 mL/min. Each patient spent minimum 1.5 hours on hydrogen inhalation daily for 3 or more months consecutively.

Tumor staging: as per the TNM Staging System.

Tumor size changes: evaluated according to imaging results and the Response Evaluation Criteria in Solid Tumors (RECIST) criteria. Complete remission (CR): All target lesions disappear; no new lesion appears; and tumor markers keep normal for at least 4 weeks. Partial remission (PR): The sum of the maximum diameters of target lesions decreases by 30% or more, and the decrease is maintained for at least 4 weeks. Stable disease (SD): The sum of the maximum diameters of target lesions decreases by a level lower than that in PR, or increases by a level lower than PD. Progressive disease (PD): The sum of the maximum diameters of target lesions increases by at least 20%, or a new lesion appears. Disease control rate (DCR): CR + PR + SD.

Quality of life scoring: The European Cancer Research and Treatment Organization (EORTC) QLQ-C30 scale is applied. The QLQ-C30 scale consists of 30 items, including 1) five functional subscales: physical function, role function, cognitive function, emotional function and social function; 2) three symptom scale: fatigue, pain, nausea/vomiting; 3) six single measurement items and one overall quality of life scale. Evaluation was performed every other week before

and after hydrogen inhalation respectively. The questioner was a third party who was not aware of patients' conditions.

According to the U.S. Eastern Cooperative Oncology Group Zubrod-ECOG-WHO (ZPS, 5-point method), physical conditions were scored (1 to 5 points). Decrease in the preset score by 1 or more points indicates improvement. Increase in the preset score by 1 or more points indicates deterioration. A score the same as the preset score indicates no change or stable disease.

Statistical analysis: SPSS data statistical analysis tools were used.

Patients who participated in the prospective follow-up survey all signed an informed consent, stating that they rehabilitated from self-initiated hydrogen inhalation rather than clinical treatment and that they voluntarily partook in the follow-up study.

Results

1. Overview

(1) A total of 82 patients were followed up, including 29 males and 53 females with an average age of 56 years. There were 8, 48 and 26 patients from age groups 20-40, 41-60, and 61-85 respectively.

(2) Types of tumors: The patient cases included 19 cases of lung cancer (23.2%); 11 cases of liver cancer (13.4%); 16 cases of gynecological malignant tumors (19.5%); 10 cases of pancreatic cancer (12.2%); 6 cases of breast cancer (7.3%); 6 cases of gastrointestinal tumors (7.3%); 6 cases of urinary tumors (7.3%), and 8 other cases (2 cases of oral malignant tumors, one case of lymphoma, one case of basal cell carcinoma, one case of gallbladder carcinoma, one case of thymoma, one case of leiomyosarcoma and one case of mediastinal inflammatory myofibroblast).

(3) Tumor staging: All the patients had stage III and IV cancers, including 21 in stage III (25.6%) and 61 in stage IV (74.4%).

(4) Combined treatment: Of the patients, 28 had only hydrogen inhalation and did not receive any

anti-cancer treatment during the period of hydrogen inhalation. The other 54 patients inhaled hydrogen and meanwhile, received chemotherapy, targeted therapy, endocrine therapy and other anti-tumor treatments.

Table 1 — Overview of the 82 patients in the follow-up study

Type of Tumor	No.	Family Name	Gender	Age	Home City/Province/Country	Diagnosis	Stage	Follow-up Time
Lung cancer (19 cases)	1	Huang	Male	63	Taiwan	Squamous carcinoma	III	12 months
	2	Zhong	Female	54	Taiwan	Adenocarcinoma	III	8 months
	3	Zhou	Female	58	Shanghai	Small cell carcinoma	IV	9 months
	4	Chen	Female	43	Urumchi	Adenocarcinoma	IV	9 months
	5	Wang	Female	46	Dongying	Adenocarcinoma	IV	7 months
	6	Chang	Female	71	Dongying	Adenocarcinoma	IV	7 months

Type of Tumor	No.	Family Name	Gender	Age	Home City/Province/Country	Diagnosis	Stage	Follow-up Time
	7	Li	Female	57	Dongying	Adenocarcinoma	IV	10 months
	8	Zhang	Female	54	Dongying	Adenocarcinoma	IV	9 months
	9	Yue	Male	65	Chongqing	Adenocarcinoma	IV	4 months
	10	Zhu	Male	65	Guangzhou	Small cell carcinoma	IV	3 months
	11	Zhao	Male	78	Hebei	Adenocarcinoma	IV	6 months
	12	Feng	Female	65	Guangzhou	Adenocarcinoma	III	3 months
	13	Hua	Male	71	Guangzhou	Adenocarcinoma	IV	7 months
	14	Wang	Male	68	Nantong	Adenocarcinoma	III	8 months
	15	Li	Female	50	Guangzhou	Adenocarcinoma	IV	8 months
	16	Yue	Male	61	Harbin	Adenocarcinoma	IV	4 months
	17	Wu	Female	78	Beijing	Adenocarcinoma	IV	5 months
	18	Wu	Female	59	Guangzhou	Squamous carcinoma	IV	5 months
	19	Wu	Male	35	Zhongshan	Adenocarcinoma	III	26 months
Liver cancer (11 cases)	20	Zeng	Male	56	Taiwan	Hepatocellular carcinoma	IV	6 months
	21	Tian	Male	48	Beijing	Hepatocellular carcinoma	IV	3 months
	22	Wang	Female	71	Yantai	Hepatocellular carcinoma	III	7 months
	23	Huang	Female	48	Guangzhou	Hepatocellular carcinoma	IV	6 months
	24	Hou	Female	58	Yuxi	Hepatocellular carcinoma	IV	6 months
	25	Liao	Male	76	Taiwan	Cholangiocellular carcinoma	IV	3 months
	26	Yang	Female	49	South Korea	Cholangiocellular carcinoma	III	3 months
	27	Chen	Female	55	Zhongshan	Cholangiocellular carcinoma	IV	7 months
	28	Liang	Female	50	Beijing	Cholangiocellular carcinoma	IV	7 months
	29	Zhang	Male	77	Guangzhou	Hepatocellular carcinoma	IV	6 months
	30	Hu	Female	40	Hengshui	Hepatocellular carcinoma	III	8 months
Gynecological tumor (16 cases)	31	Feng	Female	75	Shanghai	Endometrial cancer	IV	5 months
	32	Wang	Female	46	Guangzhou	Endometrial cancer	IV	5 months
	33	Liu	Female	60	Guiyang	Cervical cancer	IV	3 months
	34	Zhang	Female	62	Weinan	Cervical cancer	IV	4 months
	35	Quan	Female	60	Guangzhou	Ovarian cancer	III	5 months

Type of Tumor	No.	Family Name	Gender	Age	Home City/Province/Country	Diagnosis	Stage	Follow-up Time
	36	Pan	Female	54	Guangzhou	Ovarian cancer	III	5 months
	37	Pan	Female	52	Guangzhou	Ovarian cancer	III	4 months
	38	Li	Female	59	Guangzhou	Ovarian cancer	III	3 months
	39	Zhang	Female	60	Shanghai	Ovarian cancer	IV	3 months
	40	Huang	Female	44	Zhanjiang	Ovarian cancer	IV	6 months
	41	Wang	Female	47	Foshan	Ovarian cancer	III	4 months
	42	Lei	Female	50	Zhengzhou	Ovarian cancer	IV	3 months
	43	Qiu	Female	51	Zhuhai	Ovarian cancer	IV	4 months
	44	Yu	Female	51	Urumqi	Ovarian cancer	III	4 months
	45	Chen	Female	48	Shanghai	Ovarian cancer	III	3 months
	46	Du	Female	79	Nanjing	Ovarian cancer	III	3 months
Pancreatic cancer (10 cases)	47	Zheng	Female	51	Shanghai	Adenocarcinoma	IV	7 months
	48	Peng	Female	50	Qingdao	Adenocarcinoma	IV	7 months
	49	Bai	Male	53	Xinyang	Adenocarcinoma	IV	3 months
	50	Sun	Male	47	Xinxiang	Adenocarcinoma	III	6 months
	51	Liang	Female	53	Taiwan	Adenocarcinoma	IV	14 months
	52	Fu	Male	85	Taiwan	Adenocarcinoma	IV	7 months
	53	Chen	Female	49	Dongguan	Neuroendocrine tumor	IV	7 months
	54	Li	Male	60	Tianjin	Adenocarcinoma	IV	5 months
	55	Tang	Female	39	Dongguan	Adenocarcinoma	IV	4 months
	56	Wang	Female	66	Shanghai	Adenocarcinoma	IV	7 months
Breast cancer (6 cases)	57	Sun	Female	39	Shanghai	Breast cancer	IV	7 months
	58	Yu	Female	58	Shanghai	Breast cancer	IV	7 months
	59	Zhao	Female	42	Shenzhen	Breast cancer	IV	5 months
	60	Liang	Female	36	Meizhou	Breast cancer	IV	4 months
	61	He	Female	41	Guangzhou	Breast cancer	IV	5 months
	62	Su	Female	43	Guangzhou	Breast cancer	IV	3 months
Gastrointestinal tumor (6 cases)	63	Lin	Female	84	Taiwan	Gastric cancer	III	46 months
	64	Zheng	Male	60	Guangzhou	Gastric cancer	IV	7 months

Type of Tumor	No.	Family Name	Gender	Age	Home City/Province/Country	Diagnosis	Stage	Follow-up Time
	65	Yang	Male	64	Guangzhou	Rectal cancer	IV	4 months
	66	Liu	Female	58	Suzhou	Rectal cancer	III	3 months
	67	Liang	Female	48	Zhaoqing	Rectal cancer	IV	7 months
	68	Song	Male	60	Chengdu	Duodenal adenocarcinoma	IV	8 months
Urinary tumor (6 cases)	69	Chu	Male	53	Nanning	Kidney cancer	IV	6 months
	70	Chen	Male	69	Guangzhou	Prostate cancer	IV	4 months
	71	Tan	Male	64	Guangzhou	Kidney cancer	IV	7 months
	72	Liu	Male	66	Beijing	Bladder cancer	III	3 months
	73	Wang	Female	82	Beijing	Pelvic ureteral urothelial carcinoma	IV	7 months
	74	Shi	Male	56	Nantong	Kidney cancer	IV	9 months
Other tumors (8 cases)	75	Ling	Male	43	Zhanjiang	Posterior mediastinal inflammatory myofibroblastic tumor	IV	3 months
	76	Zhi	Female	50	Shanghai	Oral cancer	IV	7 months
	77	Zhao	Male	37	Shandong	Thymoma	IV	13 months
	78	Li	Female	54	Dongying	Leiomyosarcoma	IV	7 months
	79	Wang	Female	22	Dongying	Lymphoma	IV	11 months
	80	Pan	Female	72	Quanzhou	Gallbladder adenocarcinoma	IV	3 months
	81	Liu	Male	75	Beijing	Oral floor carcinoma	IV	8 months
	82	Zeng	Male	47	Guangzhou	Perianal basal cell carcinoma	III	6 months

Table 2 — Causes of death in 12 patients during the follow-up period

Type of Cancer	Cause of Death
Pancreatic cancer	Tumor progression in 3 cases; and euthanasia in one case
Liver cancer	Liver failure and concurrent infection in one case each
Lung cancer	PD-1 antibody-associated interstitial pneumonia in one case
Gynecological tumor	Intestinal obstruction, abdominal infection, and tumor progression in one case each
Gastrointestinal tumor	Upper gastrointestinal bleeding in one case
Other tumors	Infection in one case

The information of the follow-up patients was summarized as follows (see Table 1).

2. Follow-up duration and patients' survival

All the patients were followed up for 3 to 46 months in total, with a median follow-up time of 6 months. Among the patients, 45 cases (54.9%) were followed up for 3-6 months; 33 cases (40.2%) for 7-12 months; one case for 13, 14, 26 and 46 months each respectively.

During the follow-up period, 12 patients, all with Stage IV cancer, died. The patients included 4 with pancreatic cancer, 2 with liver cancer, one with lung cancer, 3 with gynecological tumor, one with gastrointestinal tumor, and one with other tumor. The causes of death are shown in Table 2.

3. Changes in the quality of life and physical fitness

Thirty patients were prospectively evaluated according to the EORTC QLQ-C30 assay. The results showed that after 2 weeks of hydrogen inhalation, the patients showed certain improvement in shortness of breath and appetite; and significant improvement in physical function, role function, emotional function, fatigue, nausea, vomiting and insomnia. After 4 weeks of hydrogen inhalation, the patients had better cognitive functions and markedly relieved pain, poor appetite, constipation and diarrhea (see Table 3).

Table 3 — Quality of life QLQ-C30 symptom scale score results

Indicator	Score Before Hydrogen Inhalation	After 2 Weeks (P value)	After 4 Weeks (P value)
Physical function	55.06 ± 23.64	63.98 ± 20.3 (0.0017**)	68.06 ± 22.93 (0.001***)
Role function	59.14 ± 33.57	69.89 ± 28.68 (0.0045***)	76.88 ± 30.63 (0.0004***)
Cognitive function	52.15 ± 26.78	62.22 ± 29.4 (0.285)	65.05 ± 29.61 (0.0035***)
Emotional function	51.08 ± 33.32	62.65 ± 30.42 (0.0004***)	65.59 ± 29.56 (0.0024**)
Fatigue	61.65 ± 30.2	43.37 ± 27.42 (0.0002***)	37.99 ± 29.65 (0.0001***)
Pain	35.48 ± 34.89	32.80 ± 31.47 (0.3617)	21.50 ± 26.59 (0.0009***)
Nausea & vomiting	23.12 ± 32.68	11.29 ± 23.33 (0.0019**)	10.75 ± 22.99 (0.0374*)
Shortness of breath	30.11 ± 29	19.35 ± 24 (0.039*)	18.28 ± 24.1 (0.0463*)
Insomnia	65.59 ± 29.17	51.61 ± 28.34 (0.0003***)	41.93 ± 28.5 (0.0012**)
Loss of appetite	48.39 ± 38.34	37.63 ± 36.25 (0.0305*)	26.88 ± 33.8 (0.0024**)
Constipation	36.56 ± 34.81	32.26 ± 31.6 (0.3542)	23.65 ± 28.79 (0.0435*)
Diarrhea	19.35 ± 28.25	15.05 ± 22.51 (0.2547)	9.68 ± 17.62 (0.0367*)

(Note: P < 0.05 indicates statistical difference; **P < 0.01 and** *P < 0.001 indicate statistically significant difference.)*

Physical performance was assessed for all the patients, with patients on retrospective survey assessed based on their medical history. After 3 months of hydrogen inhalation, 41.5%, 34.1% and 24.4% of the patients had improved, stable, and worse physical conditions respectively. The improvement rates of physical performance scores for patients who inhaled hydrogen and patients who received hydrogen inhalation combined with other therapies were 32.1% and 44.4% respectively (see Table 4). The combination therapy group scored higher in improvement rate than the hydrogen inhalation group. However, there was no statistical difference between the groups.

After 3 months of hydrogen inhalation, the patients' physical performance was scored. Compared with before treatment, the improvement rate of Stage III patients was 57.1%, and that of Stage IV patients was 36.1%. The improvement rate of physical performance in Stage III patients was higher than that in Stage IV patients. However, there was no statistical difference between the groups (see Table 5). After 3 months of hydrogen inhalation, patients

with different tumors showed different levels of physical performance improvement. Among them, lung cancer patients had the highest improvement rate (68.4%). And the lowest improvement rate was seen in patients with pancreatic cancer (0%) and gynecological tumors (12.5%) (see Table 6). Compared with lung cancer patients, gynecological and

pancreatic cancer patients exhibited significant differences in physical performance improvement. In terms of physical improvement, hydrogen treatment was the most effective for lung cancer patients, and least effective for gynecological and pancreatic cancer patients.

Table 4 — Physical performance scores for patients with mere hydrogen inhalation and patients with hydrogen inhalation combined with other treatment

Treatment	Total Number of Cases	Improvement	Stable Disease Rate	Deterioration	P Value
Mere hydrogen inhalation	28	32.1%	32.1%	35.8%	0.295
Hydrogen inhalation + other treatment	54	44.4%	35.2%	20.4%	

Table 5 — Physical performance scores for patients with Stage III and Stage IV cancers

Stage	Total Number of Cases	Improvement	Stable Disease Rate	Deterioration	P Value
Stage III	21	57.1%	38.1%	4.8%	0.572
Stage IV	61	36.1%	32.8%	31.1%	

Table 6 — Physical performance scores for patients with different types of tumors

Tumor Type	Total Number of Cases	Improvement	Stable Disease Rate	Deterioration	P Value
Lung cancer	19	68.4%	21.1%	10.5%	
Liver cancer	11	36.4%	36.3%	27.3%	0.2178
Gynecological tumor	16	12.5%	56.3%	31.3%	0.0039**
Breast cancer	6	25.0%	75.0%	0.0%	0.2159
Pancreatic cancer	10	0.0%	60.0%	40.0%	0.0020**
Gastrointestinal tumor	6	33.3%	16.7%	50.0%	0.1037
Urinary tumor	6	33.3%	66.7%	0.0%	0.1037
Other tumors	8	25.0%	50.0%	25.0%	0.1166

4. Tumor marker changes

In the follow-up study, tumor markers, including alpha-fetoprotein (AFP), carcinoembryonic antigen (CEA), CA19-9, CA125, CA153, and CA724, were monitored. Before hydrogen therapy was applied, 58 patients had elevated (one or more) tumor markers; and 24 had normal tumor markers. For patients with elevated tumor markers, the rate of tumor marker decline was 36.2% after 3 months of hydrogen inhalation. Those without tumor marker changes and with worse tumor markers accounted for 15.5% and 48.3% respectively. Tumor markers started to decline in Day 13 to 45, with a median time of 23 days. Compared with before treatment, the rates of tumor marker decline 3 months after hydrogen inhalation were 22.2% and 42.5% for patients who took only hydrogen inhalation and patients who received

hydrogen inhalation combined with other treatments, respectively. The difference between the two groups was not significant (see Table 7). Those with normal tumor markers before treatment showed no rise in tumor markers after hydrogen inhalation.

From the perspective of tumor type, 58 patients with elevated tumor markers before hydrogen inhalation were analyzed. Three months after hydrogen inhalation, 75% of the patients with lung cancer exhibited tumor marker decline, whereas patients with pancreatic cancer and liver cancer showed no decline (see Table 8). There was significant difference in tumor marker decrease between patients with lung cancer and those with gynecological tumors, liver cancer and pancreatic cancer. Regarding serum marker decline, the best therapeutic effect was found in patients with lung cancer; and worst in patients with gynecological tumors, liver cancer and pancreatic cancer.

Table 7 — Tumor marker changes during the follow-up period

Treatment	Total Number of Cases	Decrease	No Change	Increase	P Value
All the patients in the follow-up study	58	36.2%	15.5%	48.3%	
Mere hydrogen inhalation	18	22.2%	16.7%	61.1%	0.3131
Hydrogen inhalation + other treatment	40	42.5%	15.0%	42.5%	

Table 8 — Tumor marker changes in patients with different types of tumors

Tumor Type	Total Number of Cases	Decrease	No Change	Increase	P Value
Lung cancer	16	75.0%	6.3%	18.7%	
Gynecological malignant tumor	16	15.4%	15.4%	69.2%	0.0059**
Hepatic malignant tumor	7	0	28.6%	71.4%	0.0041**
Pancreatic malignant tumor	8	0	25.0%	75.0%	0.0025**
Gastrointestinal malignant tumor	5	40.0%	0	60.0%	0.1939
Breast cancer	4	25.0%	50.0%	25.0%	0.0680
Other malignant tumors	2	50.0%	0	50.0%	0.5886

5. Tumor response

Of the 82 patients who participated in the study, 80 had existing tumors before enrollment; and the other 2 had only elevated tumor markers. Of the 80 patients with tumors, there were one case (1.3%) of complete remission (CR); 15 cases (18.8%) of partial remission (PR); 30 cases (37.5%) of stable disease (SD); and 34 cases (42.5%) of progressive disease (PD) after 3 months of hydrogen inhalation. The disease control rate was 57.5%. Patients showed CR and PR in Day 21 to 80, with a median time of 55 days. Three months after hydrogen inhalation, the disease control rates of patients with mere hydrogen inhalation and patients with hydrogen inhalation combined with other treatments were 53.8% and 59.3% respectively. There was no significant difference between the two groups (see Table 9).

From the perspective of tumor staging, the disease control rates were 83% in Stage III and 47.7% in Stage IV. The difference was significant (see Table 10).

In terms of tumor pathology, lung cancer showed the highest disease control rate (78.9%), while pancreatic cancer the lowest (20%). Compared with lung cancer, only pancreatic cancer was statistically different (see Table 11).

6. Adverse effects

No hematological toxicity was detected in the 29 patients who only inhaled hydrogen as treatment.

One of the patients had stomach discomfort and chills. Another one experienced dizziness (disappeared after a few days). No other discomfort was observed. Of the 53 patients who received hydrogen inhalation combined with other treatment, one patient had headache, which disappeared within 3 to 5 days; one patient had occasional nasal discharge with small amounts of blood, which disappeared on its own; one patient had dry nasal cavity, which appeared during hydrogen inhalation and vanished a few hours after hydrogen inhalation was completed; and one patient complained of nasal sinusitis without obvious symptoms, which was indicated to be nasal sinusitis by a CT scan.

Discussion

1. Controlling effects of hydrogen on cancer

As early as 1975, Dole et al. discovered that hydrogen inhibited cancer. Nude mice with squamous cell carcinoma of the skin were given inhalation of a mixture (97.5% hydrogen and 2.5% oxygen) at 8 atmospheres. After 2 weeks, tumors in the mice shrank significantly. But such change was not found in the control group and mouse models that were given high-pressure oxygen-helium. In 2008, Japanese researcher Saitoh et al. reported that neutral-pH hydrogen-rich electrolyzed water inhibited the colony formation efficiency or colony size of human tongue cancer cell line HSC-4, but had no significant inhibitory effect on normal human tongue epithelioid cells. According to the researchers, hydrogen exerted the same effects on the growth and migration of human fibrosarcoma HT-1080. In 2009, Saitoh et al. reported that enhanced platinum colloidal hydrogen-containing water suppressed the colony formation and size of human tongue cancer cell HSC-4. In 2011, Chinese scholar Zhao et al. found that hydrogen prevented the occurrence of radiation-induced lymphoma. In 2015, Runtuwene et al. revealed that hydrogen increased the survival rate of mice with tumors induced by colon cancer, induced cancer cell apoptosis, and strengthened the sensitivity of colon cancer cells to 5-fluorouracil.

Chinese researcher Wang Dongchang et al. studied the effects of mixed hydrogen inhaled (66% hydrogen + 34% oxygen) on transplanted tumors from human lung cancer cell strain. According to the findings, the volume and weight of tumors in the hydrogen group dropped significantly; and mRNA and protein expressions of Ki-67, COX-2, and VEGF decreased considerably. In terms of pathological morphology and cell heterogeneity, the hydrogen group exhibited weaker tumor invasiveness relative to the control group. Similarly, Shang Lei et al. studied the effects of hydrogen-oxygen inhalation on mouse models of ovarian cancer. It was found that 6-week-long hydrogen inhalation inhibited tumor growth significantly. Moreover, both Ki67, which indicates the degree of tumor malignant proliferation, and CD34, which is indicative of tumor angiogenesis, decreased considerably. This suggested that hydrogen

weakened the invasion and migration of tumor cells; and might have controlled anti-tumor angiogenesis.

From our observations of 82 patients with progressive cancer in Stage III & IV and hydrogen inhalation, the functions of hydrogen mainly manifest in the following four aspects:

(1) Hydrogen improves the quality of life. After 2 weeks of hydrogen inhalation, shortness of breath and poor appetite are eased to a certain degree; and such symptoms as fatigue and insomnia are relieved significantly. After 4 weeks of hydrogen inhalation, pain, constipation, and diarrhea are mitigated greatly (see Table 3).

(2) Hydrogen enhances physical performance. After 3 months of hydrogen inhalation, patients with lung cancer show the most significant improvement in physical fitness, whereas those with gynecological and pancreatic cancer benefit the least (see Table 6).

(3) Hydrogen reduces tumor markers. After 3 months of hydrogen inhalation, patients with lung cancer show the most significant decline in tumor markers; while patients with gynecological, hepatic and pancreatic cancer exhibit the least (see Table 8).

(4) Hydrogen controls cancer progression. After 3 months of hydrogen inhalation, the tumor control rate of Stage III patients was much higher than that of Stage IV patients (see Table 10). The best controlling effect was achieved on lung cancer, whereas the worst on pancreatic cancer (see Table 11).

The treatment of progressive cancer is a great challenge. Of the anti-cancer drugs approved by the U.S. Food and Drug Administration (FDA) and European Medicines Agency (EMA), 67% and 57% have not shown evidence of long-term survival and improved quality of life respectively. According to existing studies, neoadjuvant chemotherapy induces breast cancer metastasis through TMEM-mediated mechanisms; chemotherapy increases metastatic niche formation and promotes liver metastasis; and radiotherapy facilitates the metastasis of residual liver cancer by inducing epithelial-mesenchymal transition. Liang et al. discovered that cancer cells became more malignant and turned into stem cell-like cancer cells in the environment with chemical drugs and radiation. Amelot et al. analyzed 413 patients with brain metastasis caused by different factors, and found that anti-cancer drugs, especially paclitaxel and its derivatives, promoted brain metastasis and reduced disease-free survival. A recent review suggests that all commonly-used treatments, including radiotherapy, chemotherapy, fine needle aspiration and surgery, might cause increase in circulating tumor cells and facilitate cancer progression and distant metastasis.

In consideration of the above facts, it is encouraging to see the good effects of hydrogen on cancer patients.

2. Factors related to cancer control

According to our follow-up observation, the controlling effects of hydrogen inhalation on cancer concern not only the type of tumor, but also factors and administration of hydrogen itself.

(1) Hydrogen concentration: The group in this study inhaled hydrogen at the concentration of 66% and flow rate of 3000 mL/min. In 2007, Shigeo Ohta et al. reported that inhalation of hydrogen at the low concentration of 2% is sufficient to prevent brain ischemia/reperfusion injury. Since then, most researchers have studied the biological effects of hydrogen molecules using positive-pressure hydrogen at the concentration of 1% to 4%. There were limited reports about the role of high-pressure hydrogen or high-concentration hydrogen. Only Dole et al.

reported that skin squamous cell carcinoma was successfully inhibited by using 8 atmospheres of 97.5% hydrogen. In the two experimental studies by Wang Dongchang and Shang Lei, 60% and 66.7% hydrogen were used respectively. It was found that regardless of the inhibition of cancer cells in vitro or inhibition of tumor growth in vivo, the effect of hydrogen was dose-dependent. That is, the higher the concentration of hydrogen, the greater the effect.

(2) Duration of hydrogen inhalation: In this study, most patients inhaled hydrogen daily for over

Table 9 — Tumor response in patients with hydrogen inhalation alone and patients with hydrogen inhalation combined with other treatment

Treatment	Total Number of Cases	CR	PR	SD	PD	Disease Control Rate	P Value
All the patients in the follow-up study	80	1.3%	18.8%	37.5%	42.5%	57.5%	
Mere hydrogen inhalation	26	0	11.5%	42.3%	46.2%	53.8%	0.5917
Hydrogen inhalation + other treatment	54	1.9%	22.2%	35.2%	40.7%	59.3%	

Table 10 — Tumor response in patients with Stage III and Stage IV cancers

Stage	Total Number of Cases	CR	PR	SD	PD	Disease Control Rate	P Value
Stage III	19	5.3%	21.1%	57.9%	15.8%	84.2%	0.0167*
Stage IV	61	0	18%	31.1%	50.8%	49.2%	

Table 11 — Post-treatment response to tumors in patients with different types of tumors

Tumor Type	Total Number of Cases	CR	PR	SD	PD	Disease Control Rate	P Value
Lung Cancer	19	5.3%	36.8%	36.8%	21.1%	78.9%	
Gynecological tumor	14	0.0%	14.3%	57.1%	28.6%	71.4%	0.3677
Liver cancer	11	0.0%	9.1%	36.4%	54.5%	45.5%	0.1766
Pancreatic Cancer	10	0.0%	0	20.0%	80.0%	20.0%	0.0161*
Breast cancer	6	0.0%	16.7%	50.0%	33.3%	33.3%	0.1382
Gastrointestinal tumor	6	0.0%	33.3%	0	66.7%	33.3%	0.7103
Urinary tumor	6	0.0%	16.7%	50.0%	33.3%	66.7%	0.7105
Other tumors	8	0.0%	25.0%	25.0%	50.0%	50.0%	0.4777

3 hours, and some for as long as 8 hours. Tumor markers began to decrease in Day 13 to 45, with a median time of 23 days. CR and PR appeared in Day 21 to 80, with a median time of 55 days. In the aforementioned study, the tested mice were given hydrogen inhalation for 4 and 6 consecutive weeks respectively. Yamamoto et al. studied the distribution of hydrogen in mice after the mice had continuous inhalation of 3% hydrogen. It took notably longer time

for hydrogen to reach the saturation point in the muscle (after 20 minutes) than in other organs. With time, the concentration of hydrogen in the muscle gradually increased. From the perspective of organ, the concentrations of hydrogen were the highest in the liver and lowest in the kidney.

(3) Inhalation of hydrogen-oxygen mixture instead of pure hydrogen: A lack of oxygen causes metabolic modification, which in turn supports the progression of malignant tumors. Improving oxygen supply and relieving hypoxia help inhibit cancer progression. Some studies suggest that hydrogen can "load" oxygen. Because of their extremely small sizes, hydrogen molecules can carry oxygen into the deepest part of the tumor. Inhaling oxygen while absorbing hydrogen contribute to the control of cancer progression.

(4) Pathway for introducing hydrogen into the body: In current experiments and clinical practice, hydrogen is administered by means of hydrogen inhalation, oral hydrogen-rich water, and injection of hydrogen-rich normal saline. Drinking hydrogen water does not necessarily lead to a sufficiently high concentration of hydrogen in tissues of the body. It has been reported that hydrogen is not detected in the brain after hydrogen-rich water is taken orally. Although the injection of hydrogen saline results in a quick increase in hydrogen concentration in the blood, the method is minimally invasive. Continuous infusion of a large amount of saline causes water retention. Inhalation is the easiest way to raise hydrogen concentration in tissues in a short time. In general, hydrogen concentration reaches a peak after 30 minutes, and maintains a high level of equilibrium during continuous inhalation. In theory, when 55.7% hydrogen is inhaled, the hydrogen concentration for equilibrium is around 520 μM, which is equivalent to hydrogen concentration at 600 μM that is required to inhibit cancer cells in vitro. The disadvantage of hydrogen inhalation is that it is subject to execution operations. For example, hydrogen inhalation by a nasal tube does not ensure sufficient hydrogen inhalation like that through a mask. The amount of hydrogen inhaled depends on whether the patient's breathing is even and stable; and on the status of basic lung functions.

Although the amount of hydrogen inhaled by patients cannot be accurately measured, patients' experience suggests there must be sufficient hydrogen concentration to ensure good therapeutic effects. Sufficient concentration allows hydrogen to enter tissues quickly and stay for a sufficient period of time to generate sufficient cumulative dose. The hydrogen-oxygen nebulizer used by patients in the follow-up study basically meets the above requirements. The effects of other hydrogen generators remain to be observed.

3. Evaluation of hydrogen cancer control

In 2014, Haines criticized the journal Lancet for using the term "war on cancer", arguing that comparing cancer treatment to war was the reason that many incurable solid cancers were treated with highly toxic and excessively aggressive chemotherapy. As a result, many patients with terminal cancer were denied access to early palliative treatments, including the treatments that were proven beneficial by strong evidence. Haines believed that it is time to stop using the term "war on cancer". Tang Zhaoyou advocated that the term "cancer elimination" should be abandoned and replaced with "cancer control". He proposed to treat cancer with cancer-killing therapies combined with cancer transformation measures. The former includes surgery, radiotherapy and chemotherapy. And the latter involves transforming cancer cells themselves, tumor micro-environment and the entire body.

Hydrogen exerts selective antioxidant, anti-inflammatory and signal-regulating effects. Although it is not ruled out that hydrogen has direct effects on cancer cells, hydrogen might

mainly improve tumor microenvironment and play a transforming role in cancer treatment. In addition, it has recently been discovered that hydrogen molecules are involved in the body's immune function; and save T cells in depletion and restore their anti-tumor efficacy by maintaining mitochondrial function. As a result, hydrogen molecules improve the prognosis of patients with advanced cancer. Therefore, hydrogen helps transform the body.

Hydrogen is easy to use and affordable without causing adverse effects. Hydrogen inhalation represents an important step towards achieving a truly patient-centered and family-based home re-

habilitation.

4. Existing challenges

This article shows only observation and description; but is not evidence-based research in general sense. In addition, this article focuses on rehabilitation and supplementary means rather than treatment in general sense. There are still more topics to be studied in the future. For example, is the true mechanism of hydrogen cancer control realized only by removing reactive oxide species and controlling inflammation? Considering that most studies so far center on hydrogen molecules, do hydrogen ions exert any effect? What is the optimal dose of hydrogen that should be applied to the body? How long is the course of hydrogen treatment to achieve the best effect? How to apply hydrogen-based rehabilitation in combination with mainstream treatments? What is the long-term effect of hydrogen? The list goes on. This requires researchers to broaden thinking and strengthen experimental and clinical research.

Conclusion

According to observations of the follow-up study, hydrogen inhalation helps improve the quality of life and physical performance as well as control cancer progression for patients with progressive cancer. To ensure good effect of hydrogen inhalation, the concentration of hydrogen inhaled must be kept at a sufficient level so that hydrogen enters tissues as soon as possible. It is also necessary to keep inhaling hydrogen for a time long enough to generate accumulated dose effect. Moreover, oxygen inhalation may exert a synergistic effect. In view of the great difficulty in progressive cancer treatment and high safety of hydrogen, hydrogen inhalation is worthy of further study as a simple treatment. It can be extended to clinical rehabilitation and even treatment as soon as possible. Given the large number of studies indicating that hydrogen delivers good effects on all systems of the body, the prospects of hydrogen research are promising.

— Xu Kecheng, Kong Xiaofeng, Lu Tianyu, Chen Jibing

References

1. Brawley OW. Avoidable cancer deaths globally. CA Cancer J Clin, 2011, 61(2): 67-68.
2. Siegel RL, Jemal A, Wender RC, et al. An assessment of progress in cancer control. CA Cancer J Clin, 2018, 68(5): 329-339.
3. Neeman E and Ben-Eliyahu S. Surgery and stress promote cancer metastasis: new outlooks on perioperative mediating mechanisms and immune involvement. Brain Behav Immun, 2013, 30 Suppl: S32-40.
4. Karagiannis GS, Pastoriza JM, Wang Y, et al. Neoadjuvant chemotherapy induces breast cancer metastasis through a TMEM-mediated mechanism. Science translational medicine, 2017, 9(397).
5. Zenitani M, Nojiri T, Hosoda H, et al. Chemotherapy can promote liver metastasis by enhancing metastatic niche formation in mice. J Surg Res, 2018, 224: 50-57.

6. Li T, Zeng ZC, Wang L, et al. Radiation enhances long-term metastasis potential of residual hepatocellular carcinoma in nude mice through TMPRSS4-induced epithelial-mesenchymal transition. Cancer gene therapy, 2011, 18(9): 617-626.
7. Hodson R. Precision medicine. Nature, 2016, 537(7619): S49.
8. Tannock IF and Hickman JA. Limits to Personalized Cancer Medicine. N Engl J Med, 2016, 375(13): 1289-1294.
9. Obenauf AC, Zou Y, Ji AL, et al. Therapy-induced tumour secretomes promote resistance and tumour progression. Nature, 2015, 520(7547): 368-372.
10. Sano M, Suzuki M, Homma K, et al. Promising novel therapy with hydrogen gas for emergency and critical care medicine. Acute Med Surg, 2018, 5(2): 113-118.
11. Ge L, Yang M, Yang NN, et al. Molecular hydrogen: a preventive and therapeutic medical gas for various diseases. Oncotarget, 2017, 8(60): 102653-102673.
12. Ohta S. Molecular hydrogen as a preventive and therapeutic medical gas: initiation, development and potential of hydrogen medicine. Pharmacol Ther, 2014, 144(1): 1-11.
13. Booth CM, Karim S, Mackillop WJ. Real-world data: towards achieving the achievable in cancer care. Nat Rev Clin Oncol, 2019.
14. Krause JH and Saver RS. Real-World Evidence in the Real World: Beyond the FDA. American journal of law & medicine, 2018, 44(2-3): 161-179.
15. Clarke LP, Sriram RD, Schilling LB. Imaging as a Biomarker: Standards for Change Measurements in Therapy workshop summary. Academic radiology, 2008, 15(4): 501-530.
16. Bjordal K, de Graeff A, Fayers PM, et al. A 12 country field study of the EORTC QLQ-C30 (version 3.0) and the head and neck cancer specific module (EORTC QLQ-H&N35) in head and neck patients. EORTC Quality of Life Group. Eur J Cancer, 2000, 36(14): 1796-1807.
17. Dole M, Wilson FR, Fife WP. Hyperbaric hydrogen therapy: a possible treatment for cancer. Science, 1975, 190(4210): 152-154.
18. Saitoh Y, Okayasu H, Xiao L, et al. Neutral pH hydrogen-enriched electrolyzed water achieves tumor-preferential clonal growth inhibition over normal cells and tumor invasion inhibition concurrently with intracellular oxidant repression. Oncol Res, 2008, 17(6): 247-255.
19. Saitoh Y, Yoshimura Y, Nakano K, et al. Platinum nanocolloid-supplemented hydrogendissolved water inhibits growth of human tongue carcinoma cells preferentially over normal cells. Exp Oncol, 2009, 31(3): 156-162.
20. Zhao L, Zhou C, Zhang J, et al. Hydrogen protects mice from radiation induced thymic lymphoma in BALB/c mice. Int J Biol Sci, 2011, 7(3): 297-300.
21. Runtuwene J, Amitani H, Amitani M, et al. Hydrogen-water enhances 5-fluorouracil-induced inhibition of colon cancer. PeerJ, 2015, 3: e859.
22. Wang Dongchang, Zhao Yunxia, Zhao Zhifang, et al. Action of Hydrogen in Intervening in the Suppression of Non-small Cell Lung Cancer and Its Mechanism. International Journal of Respiration, 2018, 38(8): 561-565.
23. Shang L, Xie F, Li J, et al. Therapeutic potential of molecular hydrogen in ovarian cancer. Transl Cancer Res, 2017, 7: 988-995.
24. Prasad V. Do cancer drugs improve survival or quality of life? BMJ, 2017, 359: j4528.
25. Prasad V, De Jesus K, Mailankody S. A further strategy to combat the high price of anticancer drugs. Nat Rev Clin Oncol, 2017, 14(10): 629.
26. Liang Y, Zhang Z, Huang Y, et al. Stem-like cancer cells are inducible by increasing genomic instability in cancer cells. J Biol Chem, 2010, 285(7): 4931-4940.
27. Amelot A, Terrier LM, Mathon B, et al. Can anticancer chemotherapy promote the progression of brain metastases? Med Oncol, 2018, 35(3): 35.
28. Martin OA and Anderson RL. Editorial: Therapy-induced metastasis. Clin Exp Metastasis, 2018, 35(4): 219-221.
29. Ohsawa I, Ishikawa M, Takahashi K, et al. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med, 2007, 13(6): 688-694.
30. Yamamoto R, Homma K, Suzuki K. Hydrogen gas distribution in organs after inhalation: real-time monitoring of tissue hydrogen concentration in rat. Sci Rep, 2019, 9(1): 1255.
31. Kakkal S, Krishnamachary B, Jacob D, et al. Molecular and functional imaging insights into the role of hypoxia in cancer aggression. Cancer Metastasis Rev, 2019.

32. Al Tameemi W, Dale TP, Al-Jumaily RMK, et al. Hypoxia-Modified Cancer Cell Metabolism. Front Cell Dev Biol, 2019, 7: 4.
33. Fujita K, Seike T, Yutsudo N, et al. Hydrogen in drinking water reduces dopaminergic neuronal loss in the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine mouse model of Parkinson's disease. PLoS One, 2009, 4(9): e7247.
34. Ono H, Nishijima Y, Adachi N, et al. A basic study on molecular hydrogen (H_2) inhalation in acute cerebral ischemia patients for safety check with physiological parameters and measurement of blood H_2 level. Med Gas Res, 2012, 2(1): 21.
35. Hanahan D. Rethinking the war on cancer. Lancet, 2014, 383(9916): 558-563.
36. Haines I. The war on cancer: time for a new terminology. Lancet, 2014, 383(9932): 1883.
37. Tang Zhaoyou. Chinese Way of Cancer Control. Shanghai Scientific & Technical Publishers. 2015, Shanghai.
38. Xu Kecheng. Practice Chinese Way of Cancer Control. Yangcheng Evening News Publishing House. 2018, Guangzhou.
39. Iida A, Nosaka N, Yumoto T, et al. The Clinical Application of Hydrogen as a Medical Treatment. Acta medica Okayama, 2016, 70(5): 331-337.
40. Akagi J and Baba H. Hydrogen gas restores exhausted CD8+ T cells in patients with advanced colorectal cancer to improve prognosis. Oncol Rep, 2019, 41(1): 301-311.

"Real World" of Hydrogen Cancer Control: Case Reports and Comments on the Cases

In the cases of some patients included in this section, the lesions were inoperable or recurred after surgery, or radiotherapy/chemotherapy failed. After single hydrogen inhalation (hydrogen-oxygen mixture) or hydrogen inhalation combined with "mild" radiotherapy/chemotherapy, the patients' tumors became surgically resectable, shrank, disappeared (completely remission), or became stable. Eventually, the patients survived in spite of cancer in their bodies. The cases are listed below by hydrogen use timing and purpose.

Neoadjuvant Therapy Before Surgery

Case 1: Squamous cell lung carcinoma

The patient is female, 54 years old. In October 2016, the patient had chest discomfort, dry cough, and a sense of suffocation during breathing. On October 27, 2016, she took a PET-CT scan in Taiwan. CT images displayed a 4cm space-occupying lesion in the right middle lung; and the standard uptake value (SUV) was as high as 18.57. The tumor was adhered to the pericardium, with metastasis in mediastinal lymph nodes. Bronchoscopy revealed that the bronchus of the right middle pulmonary lobe was obstructed by the tumor (see Picture 1). Local tissues were sampled for biopsy. Pathological and immunohistochemical staining results indicated squamous cell carcinoma. Genetic test indicated no mutation.

The patient received radiotherapy, during which the tumor was irradiated 30 times. In the meanwhile, she underwent chemotherapy for 3 cycles, and inhaled hydrogen by herself for at

> *Picture 1. PET-CT scan and endoscopy for Case 1. A, PET-CT scan on October 27, 2016 showed a tumor in the right middle lung and tumor invasion in the hilus; the right hilar lymph node N1 was affected (black arrows). B, Bronchoscopy showed that edema was present in bronchial mucosa, and the bronchus of the right middle pulmonary lobe was blocked by a mass. C, Biopsy indicated squamous cell carcinoma. D & E, PET-CT scan on January 13, 2017 showed no mass (black arrows) in lymph nodes N1, 2, and 3 in the right middle lung area. Compared with the PET-CT findings on October 27, 2016, the original lesion of the right middle lung was not obvious.*

least 4 hours per day. After a week, the patient had less cough and smoother breathing. Two and a half months later, the patient received a PET-CT scan on January 13, 2017. The original lesion in the lung was not found in CT images. On February 23, 2017, the patient underwent thoracoscopic resection of the upper middle lobe of the right lung. During the surgery, the pericardium and mediastinum were examined; and lymph nodes (LN2, 4, 7, and 10) were removed. No sign of cancer cell was noted in a postoperative pathological examination. So far, the patient has lived free of disease for nearly two and a half years.

Comments: The patient was definitively diagnosed with squamous cell lung carcinoma that affected the pericardium and mediastinum. As a result, the tumor could not be surgically resected. After radiotherapy and chemotherapy, the tumor was completely removed (CR). Eventually, patient received surgery, but no sign of tumor was detected. In general cases, Stage IIb squamous cell lung carcinoma is mainly treated with chemotherapy using primarily platinum or platinum-based combination chemotherapy. However, regardless of chemotherapy regimen, the benefits of survival are limited. In a meta-analysis of six RCT reports, 847 patients with non-small cell lung cancer (including squamous cell carcinoma) underwent chemotherapy. There was no difference in the efficacy of mono-chemotherapy and combination chemotherapy. The median overall survival periods for the two groups of patients were only 37.3 weeks and 34.7 weeks respectively. And their median progression-free survival periods were 14 weeks and 11.7 weeks respectively. In spite of its good effect on early-stage isolated carcinoma of the lung, radiotherapy has very limited effect on non-small cell lung cancer with carcinomas bigger than 5cm in diameter. Although the patient in Case 1 received chemotherapy and radiotherapy, these treatments alone could barely lead to disappearance of the lesion (CR). Hydrogen inhalation definitely played a role.

Case 2: Esophagus cancer

The patient is male, 62 year old. In February 2017, the patient felt unwell during eating. He took gastroscopy at a hospital in Beijing. The findings showed a mass on the posterior wall of the esophagus 31 to 35cm from the incisors. The mass was oozing blood, had brittle texture, and was prone to bleeding. Ultrasonic endoscopy showed an ulcerative mass in the 31-35 cm section of the esophagus. The lesion was mainly located in the muscularis propria of the esophageal wall and had fully invaded the outer membrane of the esophageal wall. CT scan displayed a mass in the lower esophagus, with potential lymph node metastasis in the left area of the stomach. Biopsy was performed on the esophageal mass. Pathological examination of the mass indicated squamous cell carcinoma. On March 12, the patient underwent neoadjuvant chemotherapy using paclitaxel liposome and cisplatin for a total of 2 cycles (21 days in one cycle). In the meanwhile, the patient inhaled hydrogen by himself for 3 hours daily. The inhaled gas included 66% hydrogen and 34% oxygen. The CT scan on May 11 showed no sign of apparent space-occupying lesion and stenosis in the esophagus, and no markedly enlarged lymph node in the fixed gaps of the mediastinum. On May 15, the patient underwent radical surgery for esophageal cancer. Postoperative pathology showed a hard section that measured 2.5 × 1.0 × 0.8cm in the resected esophagus. Histological examination results were as follows: squamous cell carcinoma Grade II; cancer cells shrank and invaded the fibrous membrane; lymphocyte response.

Comments: It has been reported that for resectable progressive esophageal cancer, the clinical response rate of neoadjuvant chemotherapy is 76%, and the pathological response rate is 33%. The patient in Case 2 had esophageal cancer. Preoperative ultrasonic endoscopy

and CT scan showed that the ulcerous carcinoma in the lower middle part of esophagus was 5cm long and involved the esophageal wall membrane. The patient received chemotherapy combined with hydrogen inhalation. Two months later, the tumor shrank to 2.5cm. Pathology showed that the lesion invaded only the fibrous membrane. This effect is difficult to achieve as a result of chemotherapy alone. And hydrogen molecules may have played a role in the effect. The patient said that he did not experience any adverse effect during chemotherapy. Studies have revealed that hydrogen inhalation reduces the side effects of chemotherapy.

Applied Together with Radiotherapy & Chemotherapy

Case 3: Gastric cancer

The patient is female, 84 years old. In February 2014, the patient choked during eating and vomited. On May 21, she took gastroscopy. A bulging mass was detected under the cardia of stomach and measured around 3cm to 4cm. Medical images displayed erosive bleeding. Biopsy indicated poorly differentiated adenocarcinoma. The patient also received an abdominal CT scan, which showed a bulge under the cardia. The bulge was suspected to be a stomach neoplasm. Blood tumor marker CA19-9 was at a level of 1,450 units. The patient rejected surgery and radiotherapy. Instead, she inhaled hydrogen for 3 to 4 hours daily.

One month later, the patient's dysphagia was mitigated; and CA19-9 level decreased to 307 units in August. The patient was recommended to take oral xeloda. She had gastroscopy on October 26; and results showed a white scar at the site of the original tumor. The patient used xeloda for 9 months in total until May 2015 when she discontinued due to side effects. However, she continued with hydrogen inhalation, and took a CT scan once every 6 months. According to the findings of recent CT scans, the cardia was unobstructed; and original tumor signs had disappeared. CT scan and other tests indicated no sign of cancer metastasis (see Picture 2).

In particular, the patient's blood CA19-9 level gradually decreased. Her CA19-9 level fell to 307 U/L in August 2014, and gradually decreased afterwards. By May 2017, 3 years after onset, the patient's CA19-9 level was 17.68 U/L, which was a normal level (see Picture 3). As of February 2019, the patient considered herself to be completely healthy.

> *Picture 2 (in the thick circle). D, CT scan on March 22, 2016 showed that the stomach wall was thinner in the upper part of the stomach body than before; and the original thickened area became smaller (in the thin circle).*

Comments: A study in South Korea compared the therapeutic effect of single xeloda with

that of xeloda combined with oxaliplatin in the treatment of progressive stomach cancer. For the patients who used xeloda alone, the median progression-free survival and median overall survival were 2.6 months and 6.3 months respectively. For the patients who used xeloda combined with oxaliplatin, the median progression-free survival and median overall survival were 7.1 months and 11.1 months respectively. The patient in Case 3 did not receive surgery and took only oral xeloda. Her symptoms improved in a short time, and lesions disappeared according to endoscopic findings. The patient's tumor marker CA19-9 gradually decreased to a normal level. So far, she has lived free of disease for 4 years. Such therapeutic effect is clearly associated with hydrogen.

> *Picture 3. Blood CA19-9 changes for Case 3, showing a year-on-year decline and eventually a normal level.*

Case 4: Hepatocellular carcinoma

The patient is male, 54 years old. In October 2016, the patient felt pain in the upper-right abdomen. He took a CT scan and was then diagnosed with liver cancer. Biopsy indicated hepatocellular carcinoma. At that time, the intrahepatic tumor measured 14cm, accompanied by portal vein tumor thrombus. The patient received intravenous chemotherapy once a week for 6 times in total, and underwent hepatic arterial chemoembolization 3 times successively. At the same time, the patient inhaled hydrogen by himself for 2 to 3 hours a day. After 5 months, the patient took a CT scan on March 23, 2018. The findings showed a quasi-circular low-density focus, which measured 7.8 × 7.5cm and exhibited clear margins and small deposition of iodized oil. Without significant enhancement, the focus exhibited liquefactive necrosis (see Picture 4). Portal vein tumor thrombus vanished. The patient had normal general conditions and liver functions; and resumed normal work. As of February 2019, the patient said that he was living healthily.

> *Picture 4. CT scan images for Case 4. A & B: Before treatment, there were space-occupying lesions in the right posterior hepatic lobe, with the maximum one measuring 14cm in diameter. The lesions displayed irregularities on the margins and were surrounded by sub-foci. Portal vein tumor thrombus was noted. C (plain scan) and D (lag phase): The CT scan on March 23, 2018 showed a quasi-circular focus at low density and with clear margins and pseudo-capsules in S7 and S8 of the right hepatic area. The lesion exhibited no enhancement inside; had a CT value of 27HU; and displayed low-density areas of necrosis inside (indicated by the arrow). The findings suggested loss of tumor activity and tumor liquefactive necrosis.*

Comments: For terminal liver cancer with a huge carcinoma, transhepatic arterial chemotherapy and embolization (TACE) and targeted therapy are the main treatments. Systemic chemotherapy has not been proven to prolong survival. TACE helps reduce tumors. Its main role is to serve as a bridge to create conditions for transition to radical treatment (surgical resection or ablation). If applied alone, TACE does not bring benefits of survival. Based on nine literature reviews, the median progression-free survival after TACE was 2.4 to 6.7 months. TACE is not effective for portal vein tumor thrombus. So far, the aforementioned patient has lived free of progression for 2.5 years. His liver cancer tissues have basically been necrotic and liquefied; and portal vein tumor thrombus has disappeared. The role of hydrogen cannot be ignored in his case.

Adjuvant Therapy After Surgery

Case 5: Non-small cell lung cancer

The patient is male, 64 years old. On November 6, 2017, the patient underwent an operation that completely removed the right middle pulmonary lobe due to a nodule (around 2.5cm in diameter; Stage Ic) in the right middle lobe. Pathology indicated lymphoepithelioma-like carcinoma (large cell lung cancer subtype) of poor differentiation. Genetic test results suggested mutation of gene TP53, and showed that no targeted drug can be applied. Five single cells were detected during the moni-

> *Picture 5. Oligometastasis in the 12th thoracic vertebral body after surgery in Case 5. A & B: Active focus (yellow bright area) on PET-CT scan images before hydrogen inhalation. C: Calcification of the metastatic focus 2 months after hydrogen inhalation.*

> *Picture 6. Blood CA125 level in Case 5: CA125 was maintained at the previous level within 50 days after hydrogen inhalation; and began to gradually drop to a normal level starting*

> *from Day 50 after hydrogen inhalation. (Timeline annotated with surgery (手术), chemotherapy (化疗), radiotherapy (放疗) and hydrogen (H_2); CA125 values fall from 200 to 18 U/L.)*

toring of circulating tumor cells; and a 10 × 6 mm nodule was found on the left anterior border of the 12th thoracic vertebral body. Three weeks after the operation, the patient received two courses of chemotherapy using the TP regimen (taxol + nedaplatin). Medical reexamination showed that the focus in the 12th thoracic vertebral body grew bigger to 17 × 9 mm and exhibited significant enhancement. The focus was considered to be oligometastasis. After that, the patient was given two courses of chemotherapy, using pemetrexed plus nedaplatin, and TOMO radiotherapy (18 times; around 45Gy). Consequently, the patient had severe decrease in white blood cells and blood platelets. He stopped receiving chemotherapy. Starting from April 30, the patient inhaled hydrogen for 6 hours daily. After hydrogen inhalation, his appetite became stronger. One month after hydrogen inhalation, the patient took PET-CT scan, which showed calcification of the original lesion in the 12th thoracic vertebral body and no sign of F-18 deposition (see Picture 5).

The patient's blood CA125 level was 200UL after surgery; dropped to 45U/L after chemotherapy/radiotherapy; remained at 45U/L one month after hydrogen inhalation; fell to 24U/L 50 days after hydrogen inhalation; and then further decreased to 18U/L 60 days after hydrogen inhalation (see Picture 6). As of the beginning of 2019, the patient stated that he had stable conditions and no recurrence.

Comments: Generally, platinum-based combination chemotherapy is applied to non-small cell lung cancer patients with negative ALK and no EGFR mutation. Researcher Gridelli et al. reported that patients on combination chemotherapy with cis-platinum and without cis-platinum had median overall survival of 9.6 months and 7.5 months respectively; and progression-free survival of 4.6 months and 3 months respectively. The patients given combination chemotherapy with cis-platinum experienced more severe hematological toxicity and fatigue. Socinski et al. came to similar findings, and reported that patients' median progression-free survival and overall survival were 6.3 months and 12.1 months respectively. During the period of chemotherapy for 5 months, the patient in Case 5 above had continuously elevated CA125 level and thoracic metastasis. Two months after hydrogen inhalation alone, his metastatic focus was calcified and CA125 gradually dropped to a normal level. It is worth noting that the tumor marker did not change within one month after hydrogen inhalation, but dropped to a normal level after 50 days, suggesting that the effect of hydrogen may have a dose cumulative effect.

Case 6: Ovarian cancer after surgery

The patient is female, 28 years old. In December 2017, the patient underwent surgery that removed the uterus and adnexa uteri due to cancer in the left ovary. Her postoperative diagnosis was Stage IA ovarian adenocarcinoma. Two consecutive tests in the last 3 months indicated that the tumor marker was on the rise. On May 9, 2018, the patient started inhaling hydrogen for 3 to 4 hours per day. Two weeks later, the same tumor marker mentioned above was examined, and was found to be at a normal level (see Picture 7). After one month, another examination showed CEA level at 1.9 μg/L and CA19-9 level at 10.3 U/L; and no other symptoms. The patient's CT scan, ultrasonic examination and endoscopy showed no evidence of cancer. As of the beginning of 2019, the patient did not experience cancer recurrence.

> *Picture 7. Tumor marker changes before and after hydrogen inhalation in Case 6 (CEA in μg/L and CA19-9 in U/L, measured across 2018.1.3–2018.6.25; both markers fall to normal levels).*

Comments: CA19-9 and CEA rise after ovarian cancer surgery should be first attributed to postoperative residual ovarian cancer or occult metastasis, which is generally treated with chemotherapy. However, patients are reluctant to accept chemotherapy due to its adverse effects. Hydrogen inhalation, a very simple and safe treatment, exerted magical effects on the aforementioned patient in Case 6. This is very comforting for both the patient and doctor.

Case 7: Thymic carcinoma

The patient is male, 51 years old. On June 29, 2017, the patient took a CT scan at a cancer hospital in Shandong due to unexplained chest distress. The scan showed: irregular nodules and masses (the biggest cross section measured 101×38mm) in the anterior mediastinum; multiple nodular masses in the left pleura; and shadows in strip shape in the lower lobe of the left lung. On July 5, 2017, the patient was put on general anesthesia and underwent thoracoscopic biopsy of the mediastinal mass at a hospital in Beijing. During the operation, multiple hard white nodules were found on the surface of the splanchnic layer pleura. In addition, a 6 × 5 × 5cm mass was detected in the anterior mediastinum and was closely adhered to tissues of the left lung. The nodules on the surface of the parietal pleura were partially removed and sent for frozen biopsy. Results suggested thymoma (mainly Type B 2). Then, the nodules on the splanchnic layer pleura in the lower-left pulmonary lobe were subjected to

> *Picture 8. Chest and abdomen CT scan for Case 7 showed: the thymic tumor is stable and shrinks by 2cm compared with that in CT scan images 4 months ago. The abdomen, including liver, exhibits no sign of metastasis.*

G-shaped resection with a linear cutting stapler. There were widely-spread implantation metastases in the pleural cavity of the patient's thymoma, making it impossible to conduct radical resection. As a result, the operation was terminated. After the operation, the patient received chemotherapy. After four cycles of chemotherapy, he discontinued chemo drugs due to serious adverse effects, including insomnia, loss of appetite, severe fatigue and rapid weight loss.

In early October 2017, the patient started inhaling hydrogen for 8 to 10 hours a day. After 4 months, CT scan results showed that the tumor shrank by 0.1 × 0.3 cm, and pleural effusion decreased. After 10 months, the patient received a CT scan again. And results indicated that the tumor shrank further, and pleural effusion disappeared. The patient resumed normal work. Another CT scan in July 2018 showed that the mediastinal lesion was smaller than before (at least stabilized) and exhibited no obvious sign of metastasis (see Figure 8). As of the follow-up time in early 2019, the patient had been in stable conditions.

Comments: According to WHO classification, thymoma is divided into subtypes A, AB, B1, B2 and B3 based on the form of thymic epithelium. Thymic carcinoma mainly manifests as squamous cell carcinoma, for which surgery is the only means of cure. Around 50% to 60% Type B2 thymoma, 60% to 80% Type B3 thymoma, and thymic carcinoma are in stage III to IV. Thymic tumors that cannot be surgically removed are usually progressive and difficult to treat. Radiotherapy and chemotherapy are the primary palliative therapies. After chemotherapy, the objective response rate is 47.5%, and median progression-free survival is

6.5 months. The difference between different chemotherapy regimens is small. There were two phase II trials, in which tyrosine kinase inhibitor imatinib that targeted at KIT was used to treat thymic carcinoma. But no beneficial effect was achieved. In another Phase II trial, bevacizumab was applied in combination with erlotinib to 11 cases of thymoma and 7 cases of thymic carcinoma. Eventually, no tumor response was observed. The patient in Case 7 had thymic tumor that belonged to subtype B2 pathologically. From the perspective of biological behavior, the thymic tumor was affirmatively malignant. The patient's mediastinal tumor stabilized after hydrogen inhalation. His overall survival since disease onset has been 23 months; and survival starting from hydrogen inhalation has been 19 months. His case indicates that hydrogen exerts inhibitory effects on thymic tumor.

Alternative Treatment (Palliative Treatment) for Patients not Fit for Surgery, Chemotherapy or Radiotherapy, or with Failed Chemotherapy or Radiotherapy

Case 8: Pancreatic cancer

The patient is male, 84 years old. In May 2017, the patient had poor appetite, abdominal pain, and yellow staining of the skin and sclera. CT scan showed a 7-8cm space-occupying lesion in the pancreatic head, and pancreatic duct dilation in the pancreatic tail. Multiple small lymph nodes were found around the pancreas and surrounded the blood vessels. The common bile duct and intrahepatic bile duct were dilated. The patient was diagnosed with pancreatic cancer. Later, he received endoscope-guided choledochal catheterization. After the operation, the patient's jaundice vanished gradually. The patient inhaled hydrogen by himself for minimum 5 hours per day. During the period, his appetite increased.

The patient took a PET-CT scan in January 2018. Results showed a 6-7cm space-occupying lesion in the pancreatic head, and a stent in the common bile duct. Moreover, a 2cm quasi-circular focus of high activity was detected in the left hepatic lobe, suggesting liver metastasis. The patient continued with daily hydrogen inhalation and did not undergo any other anti-cancer treatment.

The patient did not receive any anti-tumor treatment. One year later, his tumor remained stable; liver metastasis even disappeared; and general conditions including appetite and weight improved. Such effects were achieved as a result of only hydrogen inhalation. It can be speculated that hydrogen played a key role in this patient case.

On March 23, 2018, a follow-up CT scan showed a 5.9 × 3.2cm space-occupying lesion in the pancreatic head, and a stent in the common bile duct. Compared with the findings from the PET-CT scan 2 months earlier, the tumor was stable and seemed to be smaller. The metastasis in the left hepatic lobe was not seen (see Picture 9). The patient had very good appetite and mental state; and gained 2kg in the past 2 months. His circulating tumor cells were at the level of only 1/mL, and various lymphocytes and cytokines in the blood (reflecting immune function) were completely normal.

> *Picture 9. Medical image changes for Case 8 (the open circle in each picture indicates biliary stent). A: PET-CT scan in January 2018 showed a tumor in the pancreatic head (yellow highlighted area). B: PET-CT scan in January 2018 showed metastasis in the liver (yellow highlighted area). C: CT scan on March 25, 2018 showed that the tumor in the pancreatic head was similar (yellow circle) in size to the one detected in the above PET-CT scan. D: CT scan showed no sign of intrahepatic metastasis.*

Comments: Inoperable pancreatic cancer is treated primarily with chemotherapy. Gemcitabine combined with albumin-bound paclitaxel or FOLFIRINOX (leucovorin, fluorouracil, irinotecan, and oxaliplatin) is the most widely used regimen of chemotherapy, leading to a median overall survival of around 11 months. Once liver metastasis occurs, the median overall survival decreases to 7.4 months. Moreover, a retrospective study of 224 patients indicated that less than half of the patients were eligible for full-dose chemotherapy. As an American doctor once remarked about the treatment of metastatic pancreatic cancer, one solution might be better than another one; but in the end, no solution is the winner.

Case 9: Hepatocellular carcinoma recurrence after liver transplantation

The patient is male, 54 years old. In July 2017, the patient suffered pain in the liver area. CT scan showed a huge mass in the liver. Then, the patient received liver transplantation. Three months after the operation, a "new liver" appeared, with multiple metastases in the lungs, spine, bones, brain and subcutaneous tissues. One of the metastatic tumors grew out of the left nostril, with ulceration and bleeding on the surface. The tumor blocked the nasal cavity, affecting breathing. With pain all over the body, the patient could not walk and ate a little, resulting in sharp weight loss. He did not receive

any special treatment. Since March 2018, the patient has been inhaling hydrogen for 3 to 4 hours a day.

In May 2018, the patient felt that he had less pain and better appetite; and was able to walk. The metastatic tumor in the left nostril gradually shrank and disappeared eventually (see Figure 10).

> *Picture 10. A: Metastatic tumor grew out of the patient's left nostril. B: The metastatic tumor in the nostril disappeared 2 months later.*

Comments: It is extremely difficult to treat recurrent liver cancer after liver transplantation. Originally, the patient had systemic metastasis of hepatocellular carcinoma after liver transplantation, which made surgery and chemotherapy impossible. In order to prevent rejection of the transplanted liver, immunotherapy cannot be performed on the patient. After hydrogen inhalation, the patient's general conditions improved; and cancer progression stabilized. In particular, his nasal metastasis disappeared. This case showed evident effects of hydrogen.

Case 10: Rectal cancer

The patient is female, 84 years old. The patient complained of blood in the stool and incomplete defecation for 2 years, accompanied by pain in the sacral tail for 1 year. At the beginning of 2016, the patient had mucous and bloody stool with no apparent cause. She visited a hospital in Jinhua, south China's Zhejiang province. Anal finger examination findings indicated a cauliflower-like mass at 3 to 6 o'clock of the left lateral position and 2 to 6cm from the anal edge. The mass accounted for 1/4 of the intestine. The patient refused to take surgery. In May 2016, she had a large amount of blood in the stool, which consisted of a mass of blood clots around 500ml in volume. Afterwards, the patient discharged similar bloody stool 3 times successively, for which no special treatment was performed. In September 2016, the patient was found to have a mass at the entry of the anus, accompanied by more serious pain in the sacral tail. CT scan revealed: space-occupying lesions in the upper-right mediastinum; infectious lesions in the middle upper lobe of the right lung; rectal cancer; and small lymph nodes next to the abdominal aorta. The patient was considered to have rectal cancer with

mediastinal metastasis and pulmonary infection. Anorectal examination in a knee-chest position showed a cauliflower-like mass protruding from the inside of the anus. The mass measured around 3 × 2cm and exhibited unsmooth surface stained with a small amount of blood. In February 2018, the patient took a CT plain scan and contrast-enhanced scan of the abdomen. The report presented the following: The intestinal wall in the distal rectum and anal area is thickened and shows formation of soft tissue masses. Bilateral musculus levator ani are adhered to the posterior wall of the vagina, without clear dividing lines. Multiple and slightly large lymph nodes are detected next to bilateral iliac vessels and in groin areas (see Picture 11).

Findings from colonoscopy: A 10cm wide-pedicle polyp is detected in the anus. The polyp measured around 1 × 0.6cm; and exhibited congestion, edema and erosion on the surface. The mucous membrane within 4cm from the rectum to anus showed congestion, edema and erosion with cauliflower-like uplift, involving 3/5 of the perianal area (see Figure 12). Pathological biopsy indicated moderately differentiated adenocarcinoma.

> *Picture 11. CT scan showed a space-occupying lesion in the distal rectum (yellow circle), and the lesion blocked the anus (yellow arrow).*

> *Picture 12. Colonoscopy showed tumors embedded in the anus.*

Since May 2018, the patient has been inhaling hydrogen for at least 4 hours per day. Before that, she had intolerable pain and great difficulty in defecation; and could only stay in bed. More than one month after hydrogen inhalation, the pain was significantly eased. Patient was able to defecate and walk outside. The anal mass disappeared. As of the beginning of 2019, the patient was in stable conditions.

Comments: The patient had low rectal cancer, with metastases in the mediastinum and lymph nodes around the rectum. Her anus was incarcerated by tumors, causing severe pain. For the patient, surgery was impossible. The only conventional treatments were radiotherapy and chemotherapy, which were difficult to perform on the patient due to her advanced age and poor general physical conditions. The patient also refused to take chemotherapy and radiotherapy. One month after hydrogen inhalation, her pain was relieved and local tumors retracted. Undoubtedly, the improvement of the patient's symptoms is attributed to the effects of hydrogen.

Case 11: Non-small cell lung cancer with brain metastasis

The patient is female, 47 years old. In September 2015, the patient had sudden dry cough, which became increasingly serious later. CT scan showed right lung cancer with metastases in the hilar, mediastinal and left supraclavicular lymph nodes; cancerous lymphangitis in the lower-right lung; and abnormally-enhanced nodules in the right occipital lobe. Bronchoscope-guided biopsy indicated: adenocarcinoma; no EGFR mutation; and fluorescent chromosome hybridization in situ ALK (2p23) chromosomal translocation (ALK positive). On September 29, the patient started chemotherapy. From October 21, 2015 to February 8, 2017, the patient was given

> *Picture 13. Treatment course of the patient in Case 11. (Timeline: Diagnosis 2015.9 → Chemotherapy 2016.1–2016.6, with stabilization then significant progression → Radiotherapy 2017.1, progression → Hydrogen inhalation + "Crizotinib" from 2017.6, rescue from progression and improvement through 2018.6.)*

two drugs in combination and then a single drug for maintenance, followed by other different two drugs in combination and then a single drug for maintenance. The whole course lasted 15 months. After that, the patient received radiotherapy 32 times with a needle within 48 days. During the period, the CT scan 6 months after chemotherapy indicated stable disease. Two months after that, CT scan showed significant progression. Five months later, the patient took another CT scan and MR, both of which suggested disease progression with brain metastasis and cerebral hemorrhage (see Figure 13).

From May 2017, the patient started oral administration of the crude drug crizotinib, and oxygen inhalation for 8 to 10 hours per day. After 3 months, CT scan showed that the lung tumor and lymph node metastases shrank; and pleural and pericardial effusion disappeared; and a new metastatic tumor appeared in the brain. Six months after that, CT scan indicated that the lung tumor and brain metastasis were smaller than before. One year later, CT scan indicated that the lung carcinoma and brain metastasis further shrank (see Picture 14). The patient had gradually improved conditions and eventually resumed normal work. By the beginning of 2019, the patient was in stable conditions.

> *Picture 14. Brain MR and chest CT for Case 11 showed that the brain metastasis (yellow arrow) and lung tumors (yellow circle) were significantly relieved respectively.*

Comments: The patient received chemotherapy for as long as 15 months. Despite being stable for a while, the patient's disease progressed very quickly. That is because the patient used the unconventional crizotinib (an unconventional crude drug) in spite of positive ALK. Shaw et al. reported a median survival of 7.7 months in the group treated with crizotinib and 3 months in the chemotherapy group. However, there was no significant difference in overall survival between the two groups. The adverse effects of crizotinib must not be ignored, and mainly include visual impairment, liver damage and gastrointestinal reaction. There have been reports about fatal interstitial pneumonia caused by crizotinib.

Lung cancer with positive ALK fusion-mutation progresses faster. For patients with ALK-positive non-small cell lung cancer, the incidence of brain metastasis is 20% to 30%; and that during the course of treatment is 60% to 90%. Due to the low possibility of crizotinib passing through the blood brain barrier, the central nervous system is still the main site of acquired resistance to crizotinib in patients with or without brain metastasis. Most patients still inevitably acquire resistance within 1 year. And intracranial progression is the first manifestation in 40% to 50% cases. The aforementioned patient had brain metastasis at the time of onset. Crizotinib hardly penetrates the blood brain barrier, and therefore has little effect on brain metastasis. During the course of treatment, the patient had cerebral hemorrhage once and was given emergency treatment. After hydrogen inhalation, the patient's conditions gradually improved. Since the time of hydrogen inhalation, the patient has survived for more than 20 months without any progression. It is self-evident that hydrogen delivered good effects.

Case 12: Stage IV adenocarcinoma cT2aN2M1b in the superior lobe of the right lung with brain metastasis

The patient is female, 43 years old. In the second half of November 2015, the patient was admitted to a hospital in Xinjiang province for dizziness with unstable standing for 2 weeks. On November 27, 2015, she underwent surgery in which the metastatic tumor in the left cerebellum was removed. Postoperative pathology indicated metastatic lung adenocarcinoma. Genetic test results indicated that EGFR fragment 19 was positive for mutation. From December 2015 to January 2016, the patient received radiotherapy on the

whole brain. The lung CT scan on April 20, 2016 showed a space-occupying lesion in the upper lobe of the right lung. The patient took targeted therapy with iressa (gefitinib tablets). She went to a local hospital for re-examination on September 11, 2017. Results suggested disease progression. The patient was given combined chemotherapy with pemetrexed and erlotinib. During the period, the patient experienced Grade III degree bone marrow suppression. She stopped receiving chemotherapy on October 13, 2017, and continued to take erlotinib orally as targeted treatment. Her MRI report on March 6, 2018 revealed the following: There are changes after metastasis resection in the left cerebellar hemisphere and after chemotherapy. Compared to previous findings on September 18, 2017, the residual cavity of the surgical area exhibits no significant change of scope. The nodule with abnormal enhancement on the residual cavity edge is bigger than before. The left frontal and parietal lobes display smaller scope of lesions with more significant nodular enhancement on the edge, indicating disease progression. Multiple abnormal signals are

> *Picture 15. Brain MR (red arrow indicates frontal lobe tumor; green arrow indicates cerebellar tumor) and lung CT scan (tumor within yellow circle) for the patient in Case 12.*

noted in the remaining part of the brain, suggesting the presence of new metastasis. The patient's chest CT scan showed: The lesion in the upper lobe of the right lung is bigger than before. The nodule in the basal segment of the lower-right pulmonary lobe is larger and considered to be metastasis. Multiple lymph nodes are noted in the mediastinum and hilus pulmonis; and some of them are enlarged compared to before. The left adrenal gland exhibits a metastatic nodule that is bigger than before. Then, the patient was given oral icotinib hydrochloride. She discontinued it by herself due to side effects after half a month. At that time, the patient had shortness of breath, expiratory dyspnea, chest tightness, inability to articulate, and difficulty in walking. She started hydrogen inhalation for 3 to 4 hours a day. Afterwards, her symptoms were alleviated. Five months later on August 30, 2018, the patient took a follow-up examination. Results showed that the brain metastasis and right lung lesion shrank considerably (see Picture 2). Gradually, patient was able to walk by herself, breathe normally, think clearly and answer questions to the point. As of February 2019 during a follow-up phone call, the patient had been in stable conditions.

Comments: With nervous system abnormality as the initial symptom, the patient was first found with brain metastasis. Six months later, a lesion was detected in her lung. The patient received brain surgery, radiotherapy, chemotherapy and targeted therapy successively. Over 2 years later, the lesions in her lung and brain were aggravated. The symptoms gradually retracted after hydrogen inhalation. A brain MRI after half a year of hydrogen inhalation showed notably less metastases, smaller lesion in the lung, and good physical conditions. Judging by the treatment result of this case, hydrogen inhalation has significant effects on intracerebral metastasis.

Hydrogen molecules are extremely small with a high degree of diffusion, and can easily penetrate the blood brain barrier. Brain tissues and cell damage are more susceptible to oxidative stress and inflammation. Therefore, hydrogen may exert special therapeutic effects on intracranial lesions, including brain metastases.

Case 13: Gallbladder cancer

The patient is female, 73 years old. In November 2017, the patient went to a hospital for examination due to swelling pain in the upper-right abdomen. Gallbladder cancer was detected, with metastases in the liver, hepatic hilum and pancreatic head. Biopsy indicated

moderately differentiated adenocarcinoma of the gallbladder (see Picture 16) in Stage IIIa. The patient received ablation and took tegafur, gimeracil and oteracil potassium orally, but to no good effect. On September 21, 2018, the patient was admitted to a hospital because of asthma, shortness of breath, and vomiting. At that time, she had severe anemia. Her levels of hemoglobin, serum albumin, red blood cells, and hematokrit were 37g/L, 25.6 g/L, 1.49 × 10^{12}/L, and 0.14% respectively. Tumor markers CA19-9, AFP, and CEA were at the levels of 88.18 U/mL, 14.11 IU/mL, and 39.68 ng/mL respectively, all of which were higher than before. With cardiac insufficiency, the patient had tumors pressing against the duodenum, causing tumor-intestinal fistula and intestinal obstruction. Her liver was half covered by metastatic tumors. The patient was given emergency symptomatic treatments, including oxygen inhalation; infusion of red blood cells and plasma; fasting; stomach tube insertion for the purpose of gastrointestinal decompression; and parenteral nutrition. Considering the patient's extremely serious conditions, doctors in the ward repeatedly explained the situation to the patient's family. However, the patient's family insisted on receiving treatment in the hospital.

At the beginning of October, the patient was given one injection of anti-PD-1, after which she had severe fatigue, diarrhea, itchy skin, and hypokalemia. As a result, she stopped taking the injec-

> *Picture 16. Gallbladder tumor biopsy for the patient in Case 13: moderately differentiated adenocarcinoma of the gallbladder.*

> *Picture 17. The patient in Case 13 had gradually higher levels of red blood cells, hematokrit, hemoglobin and albumin after hydrogen inhalation.*

> *Picture 18. In Case 13, the patient's tumor markers (CA19-9 in U/mL, CEA in ng/mL, AFP in IU/mL) gradually dropped to normal levels after hydrogen inhalation.*

> *Picture 19. Changes of the primary tumor and metastases under CT imaging before and after hydrogen inhalation by the patient in Case 13 (primary tumor of gallbladder cancer; metastatic tumors in the liver).*

tion.

Knowing that conventional treatments would fail to treat the patient, the patient's family asked to try hydrogen inhalation and brought a hydrogen-oxygen nebulizer (66% hydrogen + 34% oxygen; manufactured by Shanghai Asclepius Meditec). The patient inhaled hydrogen via a nasal tube at the rate of 3L/minute. She had hydrogen inhalation for 2 hours a day at the beginning; and for 4 and 8 hours gradually in a few days. Except symptomatic treatment for maintaining life, the patient did not receive any other anti-tumor treatment.

After half a month of hydrogen inhalation, the patient's general conditions improved. Her stomach tube was removed 61 days after insertion. Gradually, the patient was able to eat by herself, sit up and walk. Her anemia was relieved gradually; blood red blood cells and hemoglobin increased gradually; and serum albumin rose to a normal level (see Picture 17). At the same time, the patient's three tumor markers decreased gradually to normal levels (see Picture 18).

Follow-up CT scan showed that both the gallbladder tumor and intrahepatic metastases shrank (see Picture 19). Among them, the primary tumor of the gallbladder shrank by 2/3, and

liver metastases were reduced by more than 40%. On January 18, 2019, the patient was discharged from the hospital after nearly 4 months of hospitalization.

Comments: Gallbladder cancer has poor prognosis and can be treated only with surgical resection. Due to a lack of special symptoms in the early stage, gallbladder cancer is often accompanied by benign lesions (such as gallstones) and is usually misdiagnosed as chronic gallbladder disease. As a result, many patients have lost the chance of radical surgery at the time of diagnosis. There is no effective adjuvant therapy for gallbladder cancer, whose prognosis is closely related to cancer staging at the time of diagnosis. According to statistics from the U.S., the 5-year survival rates of gallbladder cancer are 50% in Stage I, 28% in Stage II, 8% in Stage IIIa, 7% in Stage IIIb, 4% in Stage IVa, and 2% in Stage IVb respectively. A British report revealed that the higher the staging, the shorter the survival period. Patients with Stage IIIa and IIIb gallbladder cancer can survive for only 5 and 4 months on average respectively. As shown by a Canadian review in 2014, the overall median survival of patients with gallbladder cancer is only 6 months, and their 5-year survival rate is only 5%. Since there is no serosa between the gallbladder and liver, gallbladder cancer easily invades or metastasizes to the liver in its early stage. That is why gallbladder cancer has poor prognosis.

In this case, the patient had metastases around the liver, porta hepatis and pancreatic head at the onset of the gallbladder cancer. Her cancer was in Stage IVa, for which surgery was impossible. The patient once received anti-PD-1 treatment but discontinued it due to serious side effects. Without any other anti-tumor treatment and taking only hydrogen-oxygen inhalation, the patient found herself with significantly improved conditions. Her red blood cells and albumin increased; tumor markers dropped to normal levels; and primary tumor and metastases shrank evidently, all of which indicated that hydrogen exerted controlling effects on gallbladder cancer.

With good systemic conditions and able to walk freely, the patient returned to the hospital for medical examination on April 13, 2019. Results showed normal levels of tumor markers. CT scan indicated further shrinking of tumors in the liver, gallbladder and adjacent lymph nodes. Immunohistochemical staining of tumor tissues indicated that microsatellite instability (MSI+IHC) was low, suggesting that the tumor was insensitive to PD-1 antibody drugs. This further indicated that the improvement in the patient was achieved mainly by hydrogen inhalation.

Case 14: Ureteral adenocarcinoma

The patient is female, 84 years old. With hematuria for 3 months, the patient took a CT scan in December 2016. CT images showed a 3 × 4cm space-occupying lesion at the junction of the right ureter's upper part and pelvis. Biopsy indicated ureteral epithelial cancer. Twelve years ago, the patient suffered from damage and atrophy of the left kidney due to drug allergy, leaving her with only 25% of kidney function. Her creatinine level stood at 170 μmol/L. The patient refused to take nephrectomy. In March 2017, she underwent catheterization in the right ureter and then CT-guided irreversible electroporation ablation of the tumor. Her creatinine level dropped to 110 μmol/L. CT scan showed shrinking of the ureteral tumor. Six months later, the tumor recurred in the upper part of the right ureter and invaded the area toward the pelvis. The patient received CT-guided irreversible electroporation ablation again. After that, she inhaled a mixture of hydrogen (66%) and oxygen (34%) for 3 to 4 hours per day at home. Her MR report in January 2019 showed that the tumor in the right kidney and adjacent metastatic lymph nodes were smaller than before (see Picture 20). The patient had good general conditions, with creatinine level stabilizing at below 150 μmol/L.

> *Picture 20. The renal tumors and metastatic lymph nodes (indicated by the red or green arrow) in Case 14 gradually became smaller as indicated by MR imaging before and after hydrogen inhalation (DWI and T2 fat suppression sequence; April 8, 2017; January 16, 2018; January 4, 2019).*

Comments: The standard treatment for progressive or metastatic ureteral malignancies that cannot be surgically removed is systemic chemotherapy. And platinum-based combination chemotherapy is the "gold standard" means. In a Phase III clinical trial, 248 patients with nephro-ureterectomy were randomly divided into two groups and given gemcitabine-cisplatin combination treatment and intensive care respectively. The 2-year disease-free survival of the groups were 70% and 51% respectively. However, there is limited information about upper ureteral cancer, especially about chemotherapy for such patients in progressive stage.

Bevacizumab, an anti-vascular endothelial growth factor (VEGF) monoclonal antibody, was reported in a Phase II trial. In the trial, bevacizumab was used in combination with gemcitabine-carboplatin; and led to a total survival of 13.9 months. But the total survival for patients in the single chemotherapy group was 9 months.

Because of her advanced age and impaired kidney function, the patient was hardly eligible for chemotherapy. She received only local ablation of the tumor. She had severe fatigue for a while. Six

months ago, she developed dermatomyositis and was treated with corticosteroids (prednisolone). However, her primary tumor did not progress significantly; lymph nodes with metastases did not increase further; and original lymph nodes with metastases shrank. Currently, the patient has good general conditions. She has been inhaling hydrogen for more than one year. In her case, the role of hydrogen cannot be ignored.

Summary

From the aforementioned "real world" typical cases, it can be seen that the effects of hydrogen inhalation (mixture of 66% hydrogen and 34% oxygen) are promising. Hydrogen can be used for cancer control in at least the following five aspects, including:

(1) Before surgery, hydrogen can be used as a supplement to neoadjuvant therapy. It can be applied alone or in combination with chemotherapy to reduce tumors so that surgery is easier to perform afterwards.

(2) After surgery, hydrogen can be used as a supplement to adjuvant therapy. It can be applied alone or in combination with chemotherapy to eliminate residual tumors and reduce recurrence. Experimental hydrogen inhalation can be an option of rehabilitation for patients with symptom-free elevation of tumor markers (such as AFP, CEA, CA19-9, CA125, CA153, and PSA) and with an increasing number of circulating tumor cells (CTCs) but without indications for chemotherapy.

(3) Hydrogen can be used as a supplement to and applied in the same period as chemotherapy or radiotherapy in order to reduce adverse effects from chemotherapy and radiotherapy.

(4) Hydrogen can be a means of palliative care for terminal cancer patients who are not eligible for surgery, have unresectable tumors, or cannot tolerate radiotherapy and chemotherapy. In such cases, hydrogen helps alleviate symptoms, improve the quality of life, and prolong survival.

(5) Hydrogen can be used preventively (Level 3 prevention) to reduce long-term recurrence for cancer patients who have been effectively treated.

Surely, all the cases are individual cases. To obtain high-level evidence, it is necessary to conduct long-term massive clinical observation and studies, especially multi-center study. In the final version of the 21st Century Cures Act adopted by the U.S. congress in 2016, Real-World Evidence (RWE) was adopted to replace traditional clinical trials and expand indications. RWE is an approach for patient-centered research. It imposes no special restrictions on testing subjects and values the voice from patients and patient-doctor communication. It is designed to be eventually applied to clinical practice.

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References

1. Di Maio M, Chiodini P, Georgoulias V, et al. Meta-analysis of single-agent chemotherapy compared with combination chemotherapy as second-line treatment of advanced non-small-cell lung cancer. J Clin Oncol, 2009, 27(11): 1836-1843.
2. Tekatli H, van 't Hof S, Nossent EJ, et al. Use of Stereotactic Ablative Radiotherapy (SABR) in Non-Small Cell Lung Cancer Measuring More Than 5 cm. J Thorac Oncol, 2017, 12(6): 974-982.
3. Hayata K, Ojima T, Nakamori M, et al. Neoadjuvant Chemotherapy with Docetaxel, Cisplatin and S-1 for Resectable Advanced Esophageal Cancer. Anticancer Res, 2018, 38(9): 5267-5273.
4. Peng J, Tan C, Zeng X, et al. Cost-effectiveness analysis of capecitabine monotherapy versus capecitabine plus oxaliplatin in elderly patients with advanced gastric cancer. PLoS One, 2018, 13(6): e0199553.
5. Lee EW and Khan S. Recent advances in transarterial embolotherapies in the treatment of hepatocellular carcinoma. Clinical and molecular hepatology, 2017, 23(4): 265-272.
6. Li L, Zhao W, Wang M, et al. Transarterial chemoembolization plus sorafenib for the management of unresectable hepatocellular carcinoma: a systematic review and meta-analysis. BMC Gastroenterol, 2018, 18(1): 138.
7. Liu X, Wang Z, Chen Z, et al. Efficacy and Safety of Transcatheter Arterial Chemoembolization and Transcatheter Arterial Chemotherapy Infusion in Hepatocellular Carcinoma: A Systematic Review and Meta-Analysis. Oncol Res, 2018, 26(2): 231-239.
8. Carmichael JA, Wing-San Mak D, O'Brien M. A Review of Recent Advances in the Treatment of Elderly and Poor Performance NSCLC. Cancers, 2018, 10(7).
9. Gridelli C, Morabito A, Cavanna L, et al. Cisplatin-Based First-Line Treatment of Elderly Patients With Advanced Non-Small-Cell Lung Cancer: Joint Analysis of MILES-3 and MILES-4 Phase III Trials. J Clin Oncol, 2018, 36(25): 2585-2592.
10. Socinski MA, Bondarenko I, Karaseva NA, et al. Weekly nab-paclitaxel in combination with carboplatin versus solvent-based paclitaxel plus carboplatin as first-line therapy in patients with advanced non-small-cell lung cancer: final results of a phase III trial. J Clin Oncol, 2012, 30(17): 2055-2062.
11. Zhang W, Wang L, Xin Z. Combination of serum CA19-9 and CA125 levels and contrast-enhanced ultrasound parametric data facilitates to differentiate ovarian serous carcinoma from ovarian malignant epithelial cancer. Medicine (Baltimore), 2018, 97(16): e0358.
12. Moro F, Pozzati F, Mascilini F, et al. Ultrasound characteristics of ovarian metastases from low-grade appendiceal mucinous neoplasms. Ultrasound Obstet Gynecol, 2018, 51(5): 699-700.
13. Xu XL and Mao WM. Current Drug Targets for Thymic Neoplasms. Curr Cancer Drug Targets, 2015, 15(7): 593-603.
14. Girard N. Chemotherapy and targeted agents for thymic malignancies. Expert Rev Anticancer Ther, 2012, 12(5): 685-695.
15. Merveilleux du Vignaux C, Maury JM, Girard N. Novel Agents in the Treatment of Thymic Malignancies. Curr Treat Options Oncol, 2017, 18(9): 52.

16. Vivaldi C, Caparello C, Masettini G, et al. First-line treatment with FOLFOXIRI for advanced pancreatic cancer in clinical practice: Patients' outcome and analysis of prognostic factors. Int J Cancer, 2016, 139(4): 938-945.
17. Jean M, Yoo-Joung K, Akmal G, et al. Standard clinical practice of FOLFIRINOX in advanced metastatic pancreatic cancer (PC) patients: a Canadian registry. Annals of Oncology, 2015.
18. Zimmerman MA, Ghobrial RM, Tong MJ, et al. Recurrence of hepatocellular carcinoma following liver transplantation: a review of preoperative and postoperative prognostic indicators. Arch Surg, 2008, 143(2): 182-188; discussion 188.
19. Holliday EB, Allen PK, Elhalawani H, et al. Outcomes of patients in the national cancer database treated non-surgically for localized rectal cancer. J Gastrointest Oncol, 2018, 9(4): 589-600.
20. Shaw AT, Kim TM, Crino L, et al. Ceritinib versus chemotherapy in patients with ALK-rearranged non-small-cell lung cancer previously given chemotherapy and crizotinib (ASCEND-5): a randomised, controlled, open-label, phase 3 trial. Lancet Oncol, 2017, 18(7): 874-886.
21. Loganadane G, Dhermain F, Louvel G, et al. Brain Radiation Necrosis: Current Management With a Focus on Non-small Cell Lung Cancer Patients. Front Oncol, 2018, 8(336).
22. Solomon BJ, Mok T, Kim DW, et al. First-line crizotinib versus chemotherapy in ALK-positive lung cancer. N Engl J Med, 2014, 371(23): 2167-2177.
23. Yang P, Kulig K, Boland JM, et al. Worse disease-free survival in never-smokers with ALK+ lung adenocarcinoma. J Thorac Oncol, 2012, 7(1): 90-97.
24. Liu CL, Zhang K, Chen G. Hydrogen therapy: from mechanism to cerebral diseases. Med Gas Res, 2016, 6(1): 48-54.
25. Smith GC, Parks RW, Madhavan KK, et al. A 10-year experience in the management of gallbladder cancer. HPB (Oxford), 2003, 5(3): 159-166.
26. Hundal R and Shaffer EA. Gallbladder cancer: epidemiology and outcome. Clinical epidemiology, 2014, 6(99-109).
27. Bianconi M, Cimadamore A, Faloppi L, et al. Contemporary best practice in the management of urothelial carcinomas of the renal pelvis and ureter. Ther Adv Urol, 2019, 11(1756287218815372).
28. Birtle AJ, Chester JD, Jones RJ. Results of POUT: a phase III randomised trial of perioperative chemotherapy versus surveillance in upper tract urothelial cancer (UTUC). J Clin Oncol, 2018, 36(Suppl 6): 407.
29. Bislar AV, Aoplo AB, Ostrovnaya I. Phase II study of gemcitabine, carboplatin, and bevacizumab in patients with advanced unresectable or metastatic urothelial cancer. J Clin Oncol, 2013, 31(724).

Extra: Rudong Coast

Rudong coast is located in a beautiful town where the Yangtze River and Yellow Sea meet. Coastal wetlands unfold in endless vista, revealing surreal mystery. Fishermen embrace the rising sun and collect clams in each other's company. A view of them gives a taste of the infinite wonder of original ecology and great romance. The countless wind power vehicles scattered along the sea not only provide the power that never runs dry, but also present a modern and beautiful picture of Rudong life.

> *(Picture by Wu Jianhuang, Xu Lili)*

> **Tribute to Rudong Coast**
> — *Zhong Zimei*
> An elegant coastline
> Divides sky blue and green.

> The all-encompassing original ecology comes into view.
> Unconstrained clouds in the distance;
> Reeds attract the wind.
> Meeting with the egrets.
> In the heart of everyone rings an eternal song.
> Against such background,
> Striding forward for a hundred years;
> Such a state of mind,
> Straight and strong for one hundred years.

PART 5 — "Real World" Stories About Hydrogen Cancer Control

Hydrogen and Cancerous Suffering: Casting off Suffering and Drawing Happiness

Suffering from Cancer

On the afternoon of November 13, 2018, I accompanied Prof. Wu, a famous oncologist from a domestic cancer hospital, on a visit to my health studio (Guangdong Xu Kecheng Care for Health Studio). As soon as we stepped in, the volunteer Yingzi came up and led me to Mr. Liu, a Hong Kong-born businessman doing business in Changsha, Hunan province. Yingzi told me that Mr. Liu is a friend of hers for 20 years.

Next to a hydrogen-oxygen nebulizer sat a thin man, with a hat on his head and a nasal tube in his nose. He was inhaling hydrogen. Seeing me and Prof. Wu, Mr. Liu immediately stood up and shook hands with us while greeting us.

His wife came over and said: "Thank you." She seemed to be a native of Hunan province. She said with tears in her eyes: "Now, my husband has less pain. He can eat and sleep well. He has very sound sleep."

In April 2018, Mr. Liu was found to have tongue cancer. Unwilling to receive surgery, he took Chinese medicine to nurse his health. In October 2018, his conditions started to deteriorate, with symptoms like tongue pain, blistering, swelling and ulceration. Unable to open the mouth and move the tongue, Mr. Liu could take only liquid food. In addition, he had insomnia, irritability, and weight loss. Just to try his luck, Mr. Liu came to Guangzhou with his wife and started hydrogen inhalation in my studio. A week later, his sleep, appetite and mental state improved. He found himself breathing freely and walking briskly.

Prof. Wu and I examined Mr. Liu's tongue and recommended him to visit a hospital after improvement of general conditions and especially nutrition status. We told him to receive conventional treatments including surgery or minimally invasive surgery. Mr. Liu nodded with agreement.

According to Mrs. Liu, they were desperate originally and did not want to receive any treatment. Mrs. Liu had even written a will, chosen a place in the cemetery, and made final arrangements. Mrs. Liu said: "We have been through the despair and helplessness in the face of death."

As a cancer survivor, I am deeply aware of cancer patients' suffering, that is, "cancerous suffering" from: pain, anorexia, fatigue, insomnia, constant treatment, fear, anxiety, helplessness, no dignity, loneliness, being deceived in the process of seeking treatment, and financial strain. The suffering also includes the helplessness as mentioned by Mrs. Liu.

Mrs. Liu said: "Now that my husband can eat, sleep, and walk. He has the hope to live. Hydrogen inhalation is an amazing method. At least, it has relieved our suffering for now. We have the confidence to seek treatment from the hospital."

Original Purpose of Hydrogen Inhalation

Hearing what Mrs. Liu said, I was very pleased and even felt a little sense of achievement. Guangdong Xu Kecheng Care for Health Studio is a non-profit organization approved by the provincial Bureau of Civil Affairs and is established with support from leaders of Guangdong province. My goal is to use the organization as a platform to do more for the society. As both a doctor and a cancer patient, I want to do things beneficial to the rehabilitation of cancer patients.

The studio is dedicated to health care, with primary missions to practice Chinese way of cancer control and find and study effective means of cancer rehabilitation.

I have found hydrogen, not only for my studio but also for myself.

Having had cancer for 13 years, I am always worried that cancer will come back to me. Regarding cancer, doctors often speak of 5-year survival rate or 10-year survival rate rather than cure rate. That is because cancer is a chronic disease that recurs at any time. Even breast cancer, which has a 5-year survival rate as high as 70%, may still recur after 20 years. My cancer is much more malignant than breast cancer. Therefore, I have to take particular care to prevent recurrence. I have read lots of literatures, and in particular, studied Japanese research achievements about hydrogen and health. I came to realize that hydrogen molecules are good for health; help prevent and treat many diseases and pathological conditions; and prevent cancer recurrence.

Hydrogen is the most abundant gas in nature. It cannot be felt, because it is excessively light,

> *Picture 1. The author is running on a treadmill and watching TV while inhaling hydrogen at home.*

> *Picture 2. As to who has more delicate skin, I am comparing myself with a lady from the royal family of a European country.*

> *Picture 3. The royal lady is happily trying hydrogen inhalation.*

distributed in the upper air, colorless, and tasteless. Some people say that the greatest contribution of Natural Medicine in the past 20 years might be the discovery that hydrogen molecules are the secrets to stronger health.

The test started with me myself first. I had hydrogen inhalation for at least an hour a day. Usually, I inhaled hydrogen while running on a treadmill and watching TV or listening to music. I call this practice "home rehabilitation three in one". After a while, I felt that I had better sleep, more appetite and less fatigue. Colleagues and friends commented that my skin became more delicate. I believed that, because I knew the Japanese use hydrogen water for cosmetic purposes.

I went to Taipei, Shanghai, Beijing, Shandong, Xinjiang, Tokyo and other places successively on trips to visit more than 100 patients in total. The patients have been inhaling hydrogen for months and even years. I witnessed miraculous recovery in some patients and listened to their exciting stories. I was very happy and deeply touched by them.

That is why I wanted to set a hydrogen inhalation experience room in my studio in the first place.

Hydrogen has attracted attention from the public, including cancer patients, patients with common chronic diseases, healthy people, and people in sub-health conditions. The hydrogen-oxygen nebulizers in the hydrogen inhalation room of our studio keep operating from morning till night. I invited Peng Ximei, one of my former patients, to be a volunteer to help those who come for hydrogen inhalation. Ximei was a patient with terminal ovarian cancer. We rescued her from Zhanjiang city, Guangdong, in December 2009, and removed 55 kilograms of tumors and cystic fluid from her body through surgery. Ximei has fulfilled her promise to be a lifetime volunteer as long as she is alive. Ximei and other volunteers have specially set a WeChat group under the name Sunshine Hydrogen for hydrogen inhalers. Almost every day, members of the WeChat group share exciting news as follows:

> *Picture 4. Members of Light of Life Cancer Rehabilitation Association, the first group of people benefiting from hydrogen medicine, are listening to the author's speech.*

> *Picture 5. Volunteer Peng Ximei (middle) is providing service to hydrogen inhalers.*

"It was raining today, but the rain did not stop us three to come for hydrogen inhalation. One of us first came to the studio yesterday and took hydrogen inhalation for two hours. Her cough was eased and throat felt better."

"Hydrogen has improved my sleep. I am grateful."

"My allergic rhinitis is gone and has not occurred in a week."

"Thanks to hydrogen inhalation. I had suffered from joint pain for four or five years. It stopped causing pain after I inhaled hydrogen only four times. This is unbelievable. I am so grateful."

.....

One afternoon while in the outpatient clinic of the hospital, I heard a sudden bustle and hustle outside. As soon as the nurse told me that a woman insisted on seeing me without an appointment, the woman invited herself in and took me by the hand, saying: "Professor, what should I do? I just felt comfortable. I cannot stop inhaling hydrogen."

I recalled that she was an outpatient of mine and had recurrent uterine cancer after surgery. At that time, she had many symptoms. I advised her to be hospitalized for cryotherapy and vascular interventional chemotherapy. She said that she had received dozens of chemotherapy sessions, only to find herself without any appetite; mentally and physically exhausted; and with constant pain in the abdomen. She did not want to be hospitalized again.

Then, she complained that it was hard to get in the queue for hydrogen inhalation in my studio due to a large number of people. Trying to comfort her, I said: "Hydrogen inhalation is only a complementary means. For your cancer, you need conventional treatment."

"I know. I will listen to you in the future. You know, I suffered so much from the pain and problems with defecation and urination. I inhaled hydrogen and finally got out of this suffering. Professor, you are a good man. Help me again." She cried.

On second thought, the life of a cancer patient is often filled with bitterness, distress and helplessness.

Cancer is an internal disorder of the body. It is impossible to completely wipe out cancer cells. "Living with cancer" is the purpose of cancer treatment and a strategy to control cancer. We remove tumors in order to survive. However, to live a life means more than just survival.

In a certain sense, the measures that effectively relieve patients' symptoms can be considered as parts of treatment and create conditions for cancer treatment.

As we often say, we need to live, not to merely survive.

Distinguished Journalist Is Satisfied

Mr. T is a veteran journalist holding a high official post (division commander) and is only 50 years old. A year ago, Mr. T had sudden pain in the upper abdomen, and went for a medical examination. He was found to have huge liver cancer. Soon, he received liver transplantation at a famous transplantation center in Beijing. Unfortunately, the liver cancer recurred after 3 months, with tumors metastasizing rapidly to the new liver, lung, bones, subcutaneous areas and brain. Perhaps because of pressure from modern treatments, cancer

> *Picture 6. The first time I saw Mr. T, he had a metastatic tumor growing out of the left nostril.*

metastasis shows no regular pattern anymore. Take liver cancer for example. According to the regular pattern of development, extrahepatic metastasis mainly occurs to the nearest areas: lungs and bones. But in Mr. T's case, the tumor not only metastasized to multiple organs, but also moved to a remote place: the left nostril. The tumor grew from the deep part of the nose and emerged from the nostril like a snake. It makes one heartbroken and scared to even look at it.

Mr. T found himself in overwhelming sorrow with suffering from: obstruction of the nasal cavity by a metastatic tumor; periosteal pain caused by bone metastases; headache and dizziness induced by brain metastasis; metastasis and ulceration in subcutaneous areas of the abdomen; and local infection and fever.

What can the science of medicine do for Mr. T? Surgery was impossible. Chemotherapy was not effective for hepatocellular carcinoma. Mr. T received only radiotherapy, which targeted at the brain metastasis. Since he already had liver transplantation, anti-immunization was needed to prevent transplant rejection reaction. As a result, the popular immunotherapy was not suitable for Mr. T.

At the beginning of 2018 when I was in Beijing, Mr. T visited me together with his wife. I read his medical records and found that almost all of the prescriptions were about painkillers. As a doctor with a dozen years of clinical work experience in digestive and hepatic diseases, I could not think of an alternative medicine for Mr. T. With no other solution in mind, I suggested to Mr. T that he inhale hydrogen. To be exact, the gas inhaled is a mixture of hydrogen and oxygen.

A month later, Mr. T, in the company of his wife, came to Guangzhou by high-speed train. The next morning after his arrival, I went to his ward room, feeling uneasy. At a glance at his nostril, I saw that the metastasis had disappeared; and the nasal cavity was basically unobstructed. I checked the CT scan and MR examination data he brought with him. The data indicated that there still existed tumors in Mr. T's body. Nevertheless, Mr. T told me: "I have less pain, better mental state, stronger physical strength and good appetite. Now I can even walk 100 meters by myself."

"Hydrogen inhalation eased much of my pain. I am satisfied." said Mr. T slowly, full of emotion.

Mr. T said: "Prof. Xu, I have been fighting between life and death in recent years. My biggest hope is to live with cancer." His wife took out the book *Live with Cancer*, one of my books, and told me: "I read the book to him. Mr. T has finished the book by listening to me reading several times."

"Living with cancer" is a normal state of the life process. The concept entails living with high quality.

I took Mr. T by the hand and said: "The time before us is invaluable. Regardless of one month, one year or one decade, time is worth cherishing. I will help you to confront the disease. We can do it together."

Due to brain metastasis, Mr. T's eye movements were limited. Hearing what I said, he stood up, slightly raised his head, and looked at me, saying, "Yes, we can do it together. I believe that as long as we keep cheerful in the fight against death, life continues."

Two weeks later, before being discharged from the hospital, Mr. T came to my office with his wife and had a small talk with me. He said that he did not wish to live long, but only to live without much pain. He believed that hydrogen inhalation was a good method and would continue to take it, hoping to get suggestions from me about how to proceed with hydrogen inhalation.

I told him that there is currently no consensus in this regard in the industry. According to my research, I would give the following suggestions:

(1) Hydrogen inhalation is about inhaling hydrogen-oxygen mixture, not pure hydrogen. This is because hydrogen and oxygen go hand in hand. Hydrogen molecules are small and can easily enter cells, possibly bringing oxygen in together. Hypoxia is a promoter of cancer cell proliferation and progression. And oxygen inhibits cancer cells.

(2) It is important to inhale a sufficient amount of the mixture so that it goes deep into tissues. The gas flow rate for inhalation must not be low.

(3) An atomizing device is attached to the hydrogen-oxygen nebulizer. It can be used to take drugs to be inhaled through the respiratory tract.

(4) It is necessary to choose a nebulizer with stable performance. The nebulizer has to be in operation for 24 hours continuously, with a gas output fluctuation rate lower than 1%.

With a smile on his face, Mr. T said: "Only those who understand the true meaning of life constantly gain wisdom and strength. Can we take a picture together? I will always remember what you said." As he finished in a low voice, I noticed tears in his eyes.

At the door as he was about to leave, Mr. T turned back and held me by the hand, saying: "Prof. Xu, pls continue with your research. From my personal experience, hydrogen may have more in-depth significance."

Mr. T is more than just a journalist. He is also a philosopher. Life is not only about survival. The source and power of thoughts will make our lives infinite.

> *Picture 7. (UP) One month after hydrogen inhalation, Mr. T's metastatic tumor in the left nostril basically disappeared. (DOWN) Mr. and Mrs. T with the author.*

Afterword

Upon finishing writing the above, I did not feel relaxed, because I am always worried about patients. Their cancer cannot be cured and are even difficult to relieve in the long run. I only hope that they will suffer less from cancer.

What is happiness? It is a feeling. To enjoy a happy time is a kind of happiness. Facing pain and confronting challenges represent another kind of happiness. The nature of medical treatment is to take care. With his tomb on the bank of Saranac Lake in northeast New York, Dr. Edward Trudeau

had the following in his epitaph: "To cure sometimes, to relieve often, to comfort always."

It reminded me of Mr. Le Junren, the president of Shanghai Putuo District Cancer Rehabilitation Association. Once, I received a letter from him. In the letter, he said that he was deeply touched by my speech *Practice Chinese Way of Cancer Control and Research of Hydrogen Medicine* at Yangtze River Delta Forum on Cancer Rehabilitation in Wuxi, south China's Jiangsu province —

> .....
> A date with life;
> A stick of incense from the heart;
> Rids of suffering and draws happiness;
> Being towards death;
> .....

Rid of suffering and draw happiness. That shows a tolerant attitude towards doctors and gives them motivation and encouragement.

When I was about to finish this article, I saw children swinging in the courtyard from out of the window. I found myself remembering the poem *Swinging* by Han Wo:

> The pond is resting amidst clear Ching Ming rain,
> Next to it lies a dust-free courtyard,
> Swinging with a five-wire rope to beyond the wall,
> Gaining a peek of the neighbor's garden, with all strength exhausted,
> Getting down from the swing, out of breath,
> Leaning against the vermilion fence, speechless at the view up on the swing,
> Lost for words, wrapped in thoughts,
> Looking up to the sky, with a long sigh.

The moment when one swings high in the sky, a broad horizon comes into view. Perhaps the horizon is bigger by only a little than before. And perhaps the distance we see is where the intractable disease lies. Nevertheless, it is the distance that keeps us moving forward always further.

Just like we swing ourselves to increasingly higher heights gradually, our vision is widened little by little. Hydrogen inhalation (or hydrogen-oxygen inhalation) is the not-so-far distance that we have reached in our journey of exploring cancer rehabilitation. But since we are already here, why not look up to the sky and into the future?

— *Xu Kecheng*

> **Comments from Tang Zhaoyou**

> I was filled with emotions from reading the above article by Prof. Xu, especially from reading about Mr. T's case. I have been engaged in liver cancer research for half a century. The last time I saw patients with such extensive metastases of liver cancer like Mr. T was as early as in the 1960s and 1970s. At that time, such a tumor was treated with high-dose chemotherapy (such as methotrexate; MTX) and shrank in a very short time afterwards. But it grew bigger at a faster speed a few weeks later. Soon, extensive metastases grew at an unprecedented level. Other doctors and I were helpless in the face of such cases. However, in Mr. T's case, the metastatic tumors decreased significantly after hydrogen inhalation, which is truly unbelievable. Nowadays, people pay more attention to the so-called valuable specific drugs, and care nothing for simple and handy methods like hydrogen inhalation. This deserves introspection. Prof. Xu struck me as a person who always finishes what he starts. In the past, the new thing for him was cryotherapy, and he was steadfast to the end. Currently, he has consistent follow-through in the research and practice of hydrogen inhalation therapy. This article has given me a lot of basic knowledge about hydrogen inhalation.

Hydrogen and Lung Cancer: Surgery for Nothing

Taipei, So Close

Guangzhou is so close to Taipei. On the morning of March 5, 2018, my secretary Rita and I took China Southern Airlines plane and landed at Taoyuan Airport of Taiwan in less than two hours. Not far from the cabin door, we saw Ms. Zhang, the director of Shang Asclepius Meditec Co., Ltd., greeting us. He took China Eastern Airlines from Shanghai and arrived earlier than us. Instead of going through customs first, he rushed from another terminal building to pick us up. With long queues in the customs, I was given fast track out of the airport because of my advanced age. Mr. Lin, the general manager of Shang Asclepius Meditec, and Dr. Wang from Shanghai Tongji University, were waiting for us. Born in Taiwan, Mr. Lin has been doing business for 26 years in Shanghai. In the early years, he was involved in the manufacturing of automobile parts and was a well-known expert in the world. In the past few years, he entered the health industry, with a keen interest in hydrogen molecules-based health care and rehabilitation. His efforts have attracted a group of experts to participate in research. Dr. Wang is one of them. Young and beautiful, she was excited and curious on her first trip to Taiwan.

After getting into the car, Mr. Lin, looking excited, shared that he went to a crystal clean resort and climbed a mountain with several classmates yesterday. Among them was a female classmate, who once suffered from lung cancer without any hope of survival. But now, she is healthy full of energy. Looking at the expression of Mr. Lin, I guessed that the recovery of this classmate must be related to hydrogen molecules that he was studying.

Meeting with Taiwan Experts

In a private room decorated in red in Taipei's The Grand Hotel that night, I had dinner with Mr. Lin and several well-known medical experts that he invited. Among the experts, Prof. Liu was the vice president of a local hospital and also a distinguished thoracic surgeon. He showed me the WeChat account of Dr. Niu Lizhi from our hospital. On the spot, I initiated a video chat with Dr. Niu, who greeted Prof. Liu. Dr. Niu praised Prof. Liu as the founder of thoracic surgery on both sides of the Taiwan Straits. Mr. Lin said that Prof. Liu is good at innovation and has an "obsession"-like enthusiasm for hydrogen molecules.

The introduction immediately brought us closer at the dinner table and gave us common topics. Naturally, we shared with each other stories about hydrogen cancer control. The most amazing case was a patient with lung cancer. The patient inhaled hydrogen during the periods of radiotherapy and chemotherapy, followed by "an operation in vain". According to Prof. Liu, what a surgeon is most afraid in surgery is "opening the door to nothing". Fortunately, the patient did not complain but was grateful to the doctor.

Mr. Lin said that the aforementioned patient was among the classmates with whom he went to climb the mountain yesterday. She is currently very energetic and healthy.

It was an unbelievable story. The next morning, we met the patient in a hydrogen inhalation experience area near the National Taiwan University Hospital.

She was Found to have Lung Cancer

The patient was ZYJ, 54 years old. In October 2016, the patient had chest discomfort, dry cough, and suffocation during breathing. On October 27, 2016, she took a PET-CT scan in a hospital of Taipei Linkou District. The results showed a 4cm space-occupying lesion in the right middle lung. With a standard uptake value (SUV) as high as 18.57, the tumor was adhered to the pericardium and had metastasized to mediastinal lymph nodes. Then, ZYJ underwent bronchoscopy, which revealed that the bronchus in the middle right pulmonary lobe was blocked by the tumor. The doctor took local tissues for biopsy. Pathology indicated non-small cell lung cancer. Afterwards, ZYJ was given immunohistochemical staining, whose results showed squamous cell carcinoma. Genetic test findings indicated no mutation.

Ms. Z first went to see a respiratory doctor and was told that standard intake value (SUV) higher than 2.5 indicated the presence of a malignant tumor. SUV is associated with the degree of tumor malignancy. SUV greater than 10 shows that the degree of tumor malignancy is high. Ms. Z's SUV was close to 20, indicating that her tumor was very malignant. In addition, the tumor had invaded the pericardium and mediastinal lymph nodes, making surgical resection difficult. Originally, molecular targeted therapy could be an option for Ms. Z. But her tumor type was squamous cell carcinoma; and no target spot was found during genetic test. Chemotherapy could be performed on Ms. Z, but the effect was not expected to be good. Afterwards, Ms. Z made an appointment with a senior respiratory expert. After carefully reading her medical records, the doctor said: "Take radiotherapy and chemotherapy to control the tumor." Looking helpless, the doctor then said: "Inhale hydrogen. It

> *Picture 1. PET-CT scan (imaging No.: 16AT33F000×××) on October 27, 2016 in a Linkou hospital: A mass is noted in the right middle lung and has invaded the hilar area. The regional right hilar lymph node (tumor node N1) is involved (SUV 18.57, score 4). [SUV (standard uptake value) greater than 2.5 indicates malignant tumor; SUV 2–2.5 indicates critical lesion; and SUV less than 2 suggests potential inflammation or benign tumor.]*

might be helpful."

Linkou × × Hospital that Ms. Z visited is a famous hospital in Taiwan. If the expert there could not think of a good solution for her case, who else can help her? After going home from the hospital, Ms. Z was full of worries, frustration and disappointment, as if there was a piece of iron pressing against her heart. She did not tell her family the truth. She was still young and had to take care of the elderly and young in her family. She did not want to leave the world like this.

A Classmate Friendship

As the saying goes, the friendship between those who study together in the same school for a few years is always there. Learning that Ms. Z had tumor, Mr. Lin, who was then in Shanghai, flew back to Taiwan to visit Ms. Z and brought a hydrogen-oxygen nebulizer he was developing. He told Ms. Z: "No matter what treatment you will have, follow the expert's advice. Start with hydrogen inhalation. This costs nothings and causes no harm."

Mr. Lin gave Ms. Z some literatures on the inhibition of cancer by hydrogen molecules. He also showed her three video pictures. One of the pictures showed normal cells with regular arrangements and orderly arrangement. The second picture demonstrated cancer cells of different sizes, chaotic arrangement and rapid reproduction. The third picture showed also cancer cells. The difference was that the cancer cells seemed to be behaving and arranged in several rows, like bad guys had been brought back to the right path.

After a devastating earthquake hit Japan on March 11, 2011, Mr. Lin went to Japan with members of the Compassion Relief Tzu Chi Foundation to help with disaster relief. In Japan, he saw that people who participated in rescue work for the Fukuoka nuclear power leak accident bathed in hydrogen-containing water. They believed that the water helped remove radiation and prevent cancer. Afterwards, Lin paid a visit to Prof. Shigeo Ohta from Nippon Medical School. In 2007, Ohta published a paper in the well-known journal Nature Medicine, stating that hydrogen molecules are selective antioxidants that exert therapeutic effects on many diseases related to reactive oxygen species. The paper attracted the world's biomedical community to the study of this small gas molecule. Because of the paper, Ohta was recognized as the founder of hydrogen molecular medicine.

> *Picture 2. Bronchoscopy (16AK313000×××) on October 19, 2016 in a Linkou District hospital: Edema is noted in the bronchial mucosa. The bronchus of the right middle pulmonary lobe is blocked by the mass. Middle right lobe transbronchial biopsy (pathological examination number 28000-4-M80×××): indicates non-small cell lung cancer. Immunohistochemical staining results: p40 (+); TTF-1 (8G7 G3/1) (-): squamous cell carcinoma.*

> *Picture 3. PET-CT scan in Linkou × × Hospital (imaging number 17103F000×××) on January 13, 2017. The SUV of the right middle lung is 1.9 (score 2). Regional lymph nodes N1, 2, and 3 are not displayed (score 0). There is a small nodule in the upper-right lung (SUV 2.6, score 2). Compared with findings on October 27, 2016, the original space-occupying damage in the right middle lung appears insignificant.*

Perhaps because of fate, Ohta was very happy to see Mr. Lin. Sighing with emotion, Ohta said that he had been trying to create a hydrogen generator for a few years but was far from being successful. As a specialist in mechanical engineering himself, Mr. Lin listened with close attention and understood tacitly. In 3 months after returning to Taiwan, Mr. Lin successfully developed a hydrogen-oxygen nebulizer that used water to generate hydrogen. Lin sent a prototype of the nebulizer to Ohta. Looking at the prototype, Ohta, the initiator of hydrogen research, stroked the machine and appreciated that it was the best such machine in the world. Meanwhile, he was so touched by the machine that he had to constantly wipe his wet eyes with a paper towel.

Ms. Z inhaled hydrogen for at least 4 hours a day. A week later, she had less cough, smoother breathing, and better mental state. One month later, she received radiotherapy for a total of 30 times and meanwhile, underwent chemotherapy. With her home close to the hospital, Ms.

Z rushed home for hydrogen inhalation every time the treatment in the hospital was completed. Since hydrogen inhalation was limited to civilian use, Ms. Z did not tell her doctor about it. According to the doctor, Ms. Z had good physical constitution and therefore did not experience vomiting or white blood cell decline in spite of much exposure to radiation during radiotherapy. Ms. Z noticed that the other patients with whom she shared one room in the hospital did not have any appetite; and some of them vomited constantly and were frequently given injections (to increase white blood cells). She thought that her case was different surely because of hydrogen inhalation. She was grateful to the expert who recommended to her hydrogen inhalation. She also appreciated the help of Mr. Lin, the classmate who gave her a hydrogen-oxygen nebulizer.

The Tumor has Gone

Two and a half months later on January 13, 2017, Ms. Z had a follow-up examination and received a PET-CT scan in Linkou × × Hospital. A few days later, she went to the hospital for results in great fear. The doctor checked the results repeatedly and looked at Ms. Z from time to time. After a few minutes, the doctor asked Ms. Z slowly and seriously: "You had only chemotherapy and radiotherapy?"

"Yes." Ms. Z panicked for a moment. She was so nervous that she felt her heart was about to pop out. Could it be that my conditions are more serious and I will not survive?

"Your lung is free of any tumor. This is almost impossible." The doctor did not seem to believe his judgment and told Ms. Z to go to the clinical department for another check.

Ms. Z went to the respiratory and surgical departments, where doctors discussed her case together and told her: prepare for surgery. Ms. Z asked why surgery was necessary if there was no tumor anymore. The doctors said that they did not believe it and needed surgery to cut the part off, because there had been almost no such precedent so far. A seemingly senior doctor patted Ms. Z on the shoulder and said: "Your disease is very malignant lung cancer. Without surgery, we can never rest assured."

Surgery "in Vain"

On February 23, 2017, Ms. Z underwent thoracoscope-guided pulmonary lobectomy on the right side. The operation was smooth. President Liu, who was mentioned in the text above, was a famous expert in thoracoscopy at Linkou × × Hospital. After the operation, Ms. Lin was told that the upper and middle lobes of the right lung as well as affected lymph nodes were removed. No tumor was detected. In the end, it all depended on pathological section results to know whether carcinoma was present in the resected tissues. After all, pathology is the "golden standard."

Perhaps it was a difficult case for the pathologist. Ms. Lin waited 12 days until March 7, 2017 when she received her pathological examination results, which suggested no sign of tumor.

The pathological report for Ms. Lin on March 7, 2017 (S2017-012×××) stated: The following tissues were received: upper-right pulmonary lobe, two pieces of lung tissues, and tissue slices. Foreign body giant cells and reactive organizing pneumonia were found under the microscope. No tumor was present.

> *Picture 4. On February 23, 2017, Ms. Z received surgery on the uncertain malignant neoplasm in the bronchus or lung. Surgical records (No.: 21952×××) were as follows: The right middle pulmonary lobe presented consolidation. Multiple nodules were seen in the upper-right pulmonary lobe and were then given wedge-shaped excision. The right middle*

> *pulmonary lobe was resected. The mass was closely adhered to the pericardium, which was later opened by the surgeon. The pulmonary vein, pulmonary artery and branches were explored. The mediastinum was opened, and lymph nodes (LN2, 4, 7 and 10) were removed. A F24 thoracic duct was placed for drainage purpose. (The records above are taken from Ms. Z's surgical records in Linkou × × Hospital. The picture, also from the surgical records, shows a section of upper lungs.)*

"Miraculous Effects of Hydrogen Verified in My Case"

When meeting us on the morning of March 6, 2018, Ms. Z was particularly happy. Her look and gestures were like those in their forties. She shared with us the details before and after disease. I asked her: "You underwent an operation for nothing. Do you regret it?"

"Slightly, but I'm still happy, because I testified to the magical effects of hydrogen." She took my hand and continued: "My experience also verified your idea of combining cancer killing and transformation measures as described in your book *Practice Chinese Way of Cancer Control*. In my treatment, cancer transformation measures were taken before cancer killing treatment. Hydrogen inhalation is one means of transformation." said Ms. Z full of wisdom.

I signed my name in a copy of *Live with Cancer*, one of my books published 3 years ago, and gave the book to Ms. Z. She took a pen and wrote on the title page: "I am a living example of Prof. Xu's idea 'living with cancer'." For doctors, patients are the best teachers. Ms. Z's life and death experience shows that hydrogen has at least two effects on cancer. First, hydrogen transforms cancer cells, making cancer cells more compliant and therefore enhancing the effects of radiotherapy and chemotherapy. This has been proven in experiments. Second, hydrogen reduces the side effects of radiotherapy and chemotherapy. Ms. Z said that she had no problem going hiking during the periods of radiotherapy and chemotherapy. Hydrogen might have helped her.

> *Picture 5. Ms. Z (left) excitedly shared her miraculous experience with us (Dr. Wang on the right).*

— Xu Kecheng

> **Comments from Tang Zhaoyou**
> The story in this article struck a chord in me. Three years ago, one of my relatives had non-small cell lung cancer with metastasis to mediastinal lymph nodes; and passed away 2 weeks after being hospitalized. I could do nothing to help. However, in the case of Ms. Z, surgery confirmed that the tumor had completely disappeared. It is incredible. The history of modern cancer treatment is a history of eliminating tumors. Although much has been achieved, we have not won a complete victory yet. The reason is that cancer is regarded as an invading enemy. And consequently, treatment is focused on eliminating cancer instead of transforming cancer. Ms. Z's case adds to the evidence that justifies combined use of cancer elimination and transforming measures.

Hydrogen and Lung Cancer: Live On

Worried about Wang

During May Day holiday 2018, I stayed in my home in Shanghai. Soon after waking up in the morning, I thought of Wang, who said that he would return to the hospital on May 1 for a follow-

up examination. I picked up the phone and wanted to send him a message. Strangely, I was afraid to continue after typing half of the message. What if his conditions already deteriorated?

Wang was once a government official from my hometown city. In recent years, I have been highly concerned with his lung cancer. On the other hand, I enjoyed the joy from successfully treating him. It was the morning after the New Year's Day in 2017. Wang laughed and walked into my office, handing me a test report and a pile of CT films. He asked again and again: "Is this true? Is it?"

From the test report, I saw that tumor marker CA153 was in a normal range. According to CT scan images, the original tumor in the left lung had disappeared. The report stated: No obvious sign of tumor.

Wang had been suffering from non-small cell lung cancer for 8 years. When cancer was first detected in him, bone metastasis already occurred; and EGFR mutation was positive. Wang became better after using TKI inhibitor tarceva. But he developed resistance to the drug in less than one year. He switched to another inhibitor and became resistant to it after a short time. In August 2016, Wang came to our hospital. At that time, the tumor in his upper-left lung was as large as 8cm in diameter. In our hospital, Wang received cryoablation and thoracoscopic surgery, during which 80% of the tumor was removed. The remaining tumor was treated with non-specific immunotherapy.

Wang returned to his home to recover, without expecting that he could survive. But to our surprise, his tumor disappeared. Was it real? I could not believe my eyes for a while.

The test report and CT films that he showed me came from Shanghai Chest Hospital. The results were 100% real. I remember that we were all very excited at that time. I even asked our photographer to take quite some pictures to remember the happy moments.

> *Picture. On January 2, 2017, Wang (right) excitedly shared the joy of his successful treatment in the author's office.*

Another Type of Cancer Grew

In 2018, Wang's lung cancer recurred. It was strange that the recurring tumor was found not in the lung, but in the cervical lymph nodes. It was lymph node metastasis. Lung cancer, especially lung adenocarcinoma, is characterized by heterogeneity. That is, there are many different genotypes in cancer cell populations. Wang's lymph node metastasis actually indicated anther type of cancer. The metastatic tumor grew very fast. At the beginning, it was the size of a soybean. In less than a month, the tumor grew bigger to the size of a walnut. As a result, Wang's left neck was swollen, shiny red and hot. In the end, the neck seemed to be "frozen" by the tumor, making it difficult for Wang to turn and causing serious pain. Bacterial culture was performed, and the result indicated negative. Antibiotics were applied experimentally, but no response occurred.

This is typical inflammatory cancer or cancerous inflammation. In fact, one theory states that cancer is an unhealed inflammatory wound.

Cancer cells are "traitors" formed due to gene mutation during replication of normal cells under the influence of their own genetic defects or microenvironment. The number of cancer cells generated equals the number of cell division times multiplied by the number of mutant genes per division. Because of inflammation, organ cell damage increases; cell division increases,

and more mutations occur, leading to the formation of more cancer cells. Therefore, in theory, controlling inflammation helps control cancer.

Another Heart-to-Heart Talk

Laden with anxiety, Wang wanted to talk with me again. Two years ago, we had a heart-to-heart talk; and I made a decision to give him non-specific immunotherapy. The decision led to successful treatment, which we celebrated on January 2, 2017 as mentioned in the text above (see the author's *Practice Chinese Way of Cancer Control*, Page 125–132).

I went to Wang's ward room and sat on the bedside. With wet eyes, Wang held me by the hand tightly and did not let go: "Help me, please. You have helped me survive more than two 8 months." Before Wang visited our hospital in 2016, he was expected to live for only 8 months. So far, he had lived for 18 months. We had the following conversation:

Wang: "Professor, I still want to live for at least another 8 months. Two years ago, you saved me. What better way do you have? I don't want to have chemotherapy anymore."

Xu: "I suggest that you take a gene test to check if there is any new mutation. If necessary, we'll use targeted drugs."

Wang: "I already did one such test. No new target has been found. I have used several generations of targeted drugs, but the effect is not that good. I heard that hydrogen helps control cancer. What is the principle behind?"

Xu: "The world is exploring and studying hydrogen nowadays. Currently, it is a means of natural rehabilitation and control."

I introduced to Wang the connections between inflammation and cancer and between inflammation and free radicals. According to the theory of free radicals, many carcinogenic factors, including smoking, environmental pollution, chemical drugs, radiation, chronic infections and obesity, cause the body to produce toxic reactive oxygen free radicals, especially hydroxy. Hydroxyl as a free radical is very unstable and can react with most biomolecules indiscriminately and rapidly, causing DNA mutations in cell nuclei or abnormal mitochondrial energy metabolism and mitochondrial DNA mutations. This leads to cancer cell growth and progression, and especially facilitates cancer cell metastasis.

Wang: "Does it mean that if free radicals are removed, cancer can be controlled? I have taken a lot of health supplements, including large doses of vitamins C and E, as well as antioxidants that I bought from abroad."

Xu: "General antioxidants are not necessarily effective. A few years ago, The New England Journal of Medicine published a short paper by two doctors. According to the paper, some antioxidants are not only ineffective against cancer, but may promote tumor growth."

Wang: "Yeah? But the instructions for these health supplements clearly state that they control active free radicals."

Xu: "Not all active free radicals are bad. Normal cells produce such oxides as hydrogen peroxide, which are essential for normal human metabolism. General antioxidants eliminate normal oxidative free radicals and interfere with human energy metabolism, especially mitochondrial function. This is apparently harmful."

Wang: "What about hydrogen? I read many articles saying that hydrogen is an antioxidant."

Xu: "Yes. Hydrogen molecules have characteristics that other antioxidants do not possess. First, hydrogen molecules feature high specificity. They exert controlling effects only on toxic active oxygen such as hydroxy and have no effect on normal free radicals. Second, hydrogen molecules feature high permeability. Once entering the body, hydrogen goes everywhere and can freely enter the mitochondria, cell nuclei and blood–brain barrier. In particular, hydrogen is effective in treating brain lesions. Hydrogen molecular medicine was founded as a discipline starting from the time when Japanese Prof. Shigeo Ohta published a paper about hydrogen molecules in reducing cerebral infarction."

Wang: "Is it toxic? I have been sick for many years. And I am very scared of the side effects from drugs."

Xu: "The third characteristic of hydrogen is great safety. The bacteria in our large intestines produce hydrogen all the time. One expert in the United States found that an average person produces up to 1,200 liters of hydrogen in one day after meals."

Wang: "Oh, I see. When was the earliest published paper about hydrogen cancer control?"

Xu: "In 1975, the first paper on hydrogen cancer control was published in the journal Science. The paper reported the successful treatment of squamous cell carcinoma of the skin with high-pressure hydrogen."

Start of Hydrogen Inhalation

Perhaps out of professional habits over the years as a government official, Wang was cautious about taking any type of treatment. For example, he purchased a PD-1 inhibitor from abroad, but never used it after reading an article warning patients with positive EGFR mutation to be discreet in using the drug. Despite knowing that hydrogen was not a drug but a natural substance, Wang personally visited several cancer patients who inhaled hydrogen at home to understand the real effect. Eventually, he borrowed a hydrogen–oxygen nebulizer and inhaled hydrogen–oxygen mixture (66.6% hydrogen + 33.4% oxygen) for at least 4 hours per day.

"I Might be Saved Again."

Finally, I encouraged myself to send a message to Wang, asking what happened to his neck tumor. Two hours later, Wang called back and said: "Thank you." several times. He told me about his conditions: there were three metastatic lymph nodes in the neck. One of them had subsided. The other two oozed blood originally, but gradually disappeared one by one in the past week. I asked him to continue. He said outright: "Of course, this is the most important thing." He added: "Professor, thank you very much, maybe I can be saved again." His voice was a little hoarse, possibly because the metastatic lymph nodes in the neck pressed against neck nerves. But judging by the emotions revealed from his laughter and speech, I thought that he would surely survive and live for not only another 8 months he expected.

Afterword

Over five months after I finished the article above, I received news from Wang's family member: "Wang's conditions are severely aggravated; and the odds of survival will be very small." I asked why, and his family member replied that Wang used PD-1 inhibitors and the side effects were so big that Wang's body was crushed overnight.

Not long ago, The Lancet reported complications of anti-PD-1 therapy, especially the complications of immune inflammation in most patients. According to the report, 101 patients were complicated with severe myocarditis and were subject to a mortality rate as high as 46%.

Among those who used PD-1 inhibitors alone, the mortality rate was 36%. For the patients using PD-1 inhibitors combined with ipilimumab, the mortality rate reached as high as 67%.

How did this complication occur to Wang?

I looked at the photos taken with him in my office. I remembered saying to Wang repeatedly in the past few years: "Do not hope to eradicate cancer. Just live with cancer. Regardless of the size of the tumor, survival is the top priority."

In spite of his concerns about using PD-1 inhibitors once, Wang eventually used the "magical drug", probably because he was convinced by the marketing of the drug...

— *Xu Kecheng*

> **Comments from Tang Zhaoyou**
> Prof. Xu's article above reminded me of another difference between eastern and western thinking. Western thinking flows from mechanism to practice. In terms of molecular targeted therapy, the first thing is to understand relevant molecules. Then, drugs are designed for specific molecules. Finally, drugs are put into use and tested in practice. On the contrary, eastern thinking begins with practice. If a drug is proven effective, efforts will be made to understand the mechanism of action if conditions allow. For example, arsenic trioxide was first applied to treating a type of leukemia and achieved good effects. After that, its action of mechanism was found to be differentiation induction that brought cancer cells back to the right path. If only western thinking is adopted, a great deal of actual results from practice might be completely ignored. Although the principle behind the concept "hydrogen is effective in cancer treatment" comes from abroad, the concept is well-practiced in China. What we need is to integrate western and eastern thinking.

Hydrogen and Lung Cancer with Bone Metastasis: Surviving Bitter Cold

Seeking Help from a Peer

Y is a radiologist and the director of the radiology department of a famous medical university-affiliated hospital in China. On May 27, 2018, he called me, telling me that he was diagnosed with lung cancer on November 6, 2017, and had surgery, chemotherapy and radiotherapy successively. During the period, oligometastasis (single metastatic tumor) was found in the 12th thoracic vertebra. On April 30, 2018, Y started hydrogen inhalation for 6 hours a day. He asked me for some advice.

Once suffering from cancer themselves, doctors have more considerations. In 1986, McGill Cancer Center in Canada conducted a questionnaire survey among doctors. One of the questions was as follows: If you are a 60-year-old oncologist diagnosed with non–small cell lung cancer with metastases in the liver and bones, would you take chemotherapy? It is said that 81% of the doctors surveyed chose not to use cisplatin-based chemotherapy. Eleven years later in 1997, the National Comprehensive Cancer Network (NCCN) conducted an identical survey. The proportion of doctors willing to take cisplatin-based chemotherapy increased from 19% to 64.5%; and that of nurses was 67%. Apparently, Director Y is a very "traditional" doctor. He received all the treatments that should and could be done, including cisplatin-based chemotherapy. However, the troubled part of Director Y's case was the occurrence of bone metastasis during chemotherapy. It was likely that the harsh treatment of

chemotherapy induced genomic disorder of cancer cells. As a result, new genomes resistant to chemotherapy were formed and eventually became "outliers" that resisted drugs or made the originally slowly-progressing cancer more aggressive. In this case, application of chemotherapy again was generally ineffective.

From the phone call, I knew that Director Y was very anxious. I encourage him: "Don't be disappointed. If the standard treatment does not work, why not switch the path?"

Metastasis and Progression

Director Y sent me the following WeChat message that described his course of treatment. (see picture 1)—

> *During a physical examination at the beginning of November 2017, I was found to have a 2.5cm nodule in the middle lobe of the right lung. Then, I received an operation in which the right middle pulmonary lobe was completely removed. Postoperative pathology indicated poorly differentiated lymphoid epithelial carcinoma (large cell lung cancer subtype). Genetic test results showed positive mutation of TP53 and no other mutation. No targeted drug could be applied to my case. My disease was in Stage Ic. Circulating tumor cells (CTC) monitoring revealed five single cells and no invasive tumor cell mass or mesenchymal cell. CT scan showed a 10 × 6mm nodule on the left anterior edge of the 12th thoracic vertebral body. The nodule was not defined before surgery. In the third week after surgery, I started chemotherapy with the TP regimen (taxol + nedaplatin). The chemotherapy lasted for two courses.*
> *A follow-up CT scan showed that the lesion in the 12th thoracic vertebral body grew to 17 × 9mm and displayed enhancement. It was considered as oligometastasis. My chemotherapy regimen was then changed to pemetrexed combined with nedaplatin for another two courses. I also received TOMO radiotherapy (18 times; around 45Gy). Afterwards, my white blood cell count decreased to 1800mm3, and platelet level dropped to 49,000/μL at the lowest level, for which I had leukocyte-boosting injections for more than a month. However, my white blood cell count was still 2400mm3. Therefore, I stopped chemotherapy.*
> *In middle March 2018, one month after the end of radiotherapy, Director Y took a MRI contrast-enhanced scan. The findings showed no change in the lesion of the 12th thoracic vertebra. During a follow-up examination in middle April, the lesion was found to be slightly smaller than previous month and with moderate enhancement, suggesting that the metastasis was not controlled.*

Director Y said that after reading the article I wrote about the follow-up on hydrogen treatment in Taiwan, he managed to buy a hydrogen nebulizer. Starting from April 30, he had hydrogen inhalation for 6 hours a day. He said: "The good news is that I have better appetite after hydrogen inhalation."

I suggested that Director Y continue with hydrogen inhalation and follow correct inhalation methods to ensure that sufficient hydrogen enters the body. I recommended him to read an experimental study on the successful treatment of skin cancer with hydrogen in the 1975 issue of Science. I told him about our findings from animal model experiments and cytological experiments to be published. All the aforementioned experiments indicated that high-concentration hydrogen has stronger inhibitory effects on cancer than low-concentration hydrogen; and that the inhibitory effects are positively correlated with accumulated hydrogen doses within certain limits.

Before Hydrogen Inhalation

Via WeChat, Director Y sent me pictures of the 12th thoracic vertebra metastasis before hydrogen inhalation.

> *Picture 1. May 27, 2018 WeChat message from Director Y describing his course of treatment.*

> *Picture 2. Metastasis in the 12th thoracic vertebra as shown in PET-CT scan and MRI.*

> *Picture 3. June 3, 2018 WeChat message from Director Y describing his conditions in Day 23 after hydrogen inhalation.*

> *Picture 4. Single metastatic tumor in the 12th thoracic vertebra:*
> *A&B: The tumor was displayed as an active focus (yellow bright area) in a PET-CT scan before hydrogen inhalation.*
> *C&D: The metastatic tumor showed calcification 2 months after hydrogen inhalation.*

The pictures were from the PET–CT scan (before surgery, chemotherapy and radiotherapy) in early November 2017 and MRI (plain scan + contrast–enhanced scan) in the end of December 2017. The results showed significant enhancement of the left anterior lesion in the 12th thoracic vertebra. Bone scan findings indicated notable isotope deposition (after two sessions of chemotherapy with the TP regimen). Tumor marker CA125 was not high before surgery but went up one month after surgery to 200 units at the highest level. During chemotherapy, CA125 was maintained at a level of 45 units.

23 Days after Hydrogen Inhalation

Director Y informed me via WeChat: PET–CT scan still showed presence of bone metastasis; CA125 level did not decrease and kept at a level of 45 units.

50 Days after Hydrogen Inhalation

On July 2, 2018, Director Y sent me another WeChat message, happily telling that CA125 level dropped to 24 units (normal value: less than 35 units); MRI contrast–enhanced scan showed slight decrease in the size of the lesion in the 12th thoracic vertebra and slightly weaker enhancement of the lesion.

65 Days after Hydrogen Inhalation

After another two weeks, Director Y sent me PET–CT photos, which showed calcification in the lesion area. His CA125 level further fell to 18 units, which was completely normal.

> *I started inhaling hydrogen on April 30 when my CA125 level was 45u. Mr. Lin encouraged me to keep hydrogen inhalation for 300 hours cumulatively. In Day 50 when I reached around 300 hours of hydrogen inhalation, my CA125 decreased to 24u. Last week, the level was 18u. I want to thank you and Mr. Lin for your care.*

> *Picture 5. Director Y sent me a WeChat message (A) and a picture about his CA125 dynamic changes on the afternoon of July 14, 2018.*

Good Luck and Sting in the Heart

Director Y suffered from poorly differentiated lymphoid epithelioid carcinoma, which was very malignant. Without target spots, there were limited drugs for him. The oligometastasis in the 12th thoracic vertebra may have existed before surgery and was apparently insensitive to chemotherapy. As a result, the metastatic tumor grew bigger during the period of chemotherapy. It was still not well-controlled by radiotherapy.

In Day 50 after hydrogen inhalation, Director Y's tumor marker CA125 decreased to a normal level and metastatic tumor showed calcification. The results are amazing; and Director Y is very lucky.

According to the timeline of hydrogen inhalation, metastasis changes and CA125 decrease, it can be said that hydrogen definitely played a positive role in the good therapeutic effects for Director Y.

As if one mishap is followed by another, Director Y found himself unhappy. He had been bothered by fatigue. In his WeChat message, Director Y said that he was extremely tired, and that the unbearable fatigue made it difficult for him to eat, sleep and walk well. He eagerly hoped me to offer certain treatment.

I fell into anxiety. That day, I sat in the office, thinking. The image of Director Y with great fatigue flashed in my mind.

What can I do for him?

I caught a glimpse of *Words at Hand*, a book given to me by a Beijing-born writer with pancreatic cancer. I thumbed through the book and stopped at a short essay titled *Trapped*, one paragraph of which reads like this:

> *..... Sudden events come into life one after another. The sun shines again after rain, but the traces remain. After all, there are always something broken under howling winds and heavy showers. With despotic power, a violent rain always leaves certain scars behind. Hence, my heart is doomed to sorrow. That is a thorn that cannot be pulled out, but has been deeply pricked in the heart .....*

Cancer metastasis is a "violent storm" for anybody, not to mention for a senior medical expert with decades of experience in treating others. Director Y has been through a "violent storm". Surely, there is an intractable thorn marked in his heart like an indelible "sting".

This is indeed the case. Director Y told me that he was bearing very heavy psychological burden and might have depression.

Fatigue, especially cancerous fatigue, is mainly associated with increased toxic free radicals and inflammation in the body. Studies in recent years suggest that depression is not only a psychological disorder, but also intensifies with inflammatory factors and inflammatory response. Hydrogen helps eliminate toxic free radicals and control oxidation and inflammation. Theoretically, hydrogen is effective in alleviating fatigue and depression. Animal experiments indicate that hydrogen inhalation significantly relieves stress–induced depression and anxiety–like behavior in mice.

It is very likely that the "sting in the heart" aggravates fatigue and fatigue intensifies "sting in the heart".

Hydrogen inhalation seems to lead to double gain. I encouraged Director Y to continue with hy-

drogen inhalation, because hydrogen has dose–accumulation effects.

7 Months after Hydrogen Inhalation

On November 20, 2018, Director Y sent me a WeChat message, telling me that he practiced Qigong (Guo Lin Qigong) in the morning, swam (1000 meters per day) in the afternoon, and

inhaled hydrogen at night. His conditions were much better than before, and appetite returned to the same level as before disease. The metastasis of in the 12th thoracic vertebra was stable. The tumor marker was in a normal range.

Meng Jiao, a Chinese poet during the Tang Dynasty, once wrote in the poem *Bitter Cold*: "*Cold sky a deep and distant grey. This north wind withers the mulberry trees.*" I hope that Director Y has already survived the bitter cold. "*Good rain knows its right time; It will fall when comes spring.*" (*Happy Rain on a Spring Night.* by Du Fu). I wish to see Director Y enjoying the spring of life.

— *Xu Kecheng*

> **Comments from Tang Zhaoyou**
> The radiologist in the case above had metastasis after lung cancer surgery. Chemotherapy was not effective for him. After 50 days of hydrogen inhalation, his tumor marker fell to a normal range; and bone metastasis showed calcification. From the perspective of time, hydrogen definitely played a role, indicating that hydrogen can be used for adjuvant therapy. Since 1960 when Rudolf Virchow introduced the origin of cancer cells, all efforts have been directed at eliminating cancer cells. However, cancer is different from infectious diseases, which are the results of invasion by "foreign enemies". Cancer is an internal disturbance of the body caused by imbalance between the internal and external environment. For this reason, both cancer elimination and transformation treatments (cancer control instead of cancer elimination) are needed and complement each other.
> Cancer is a chronic disease. And cancer control is a long-lasting battle to both eliminate and transform cancer. Hydrogen therapy is not used in traditional methods for killing tumors but represents a means of transforming the nature of cancer. It is worthy of long-term attention.

Hydrogen and Lung Cancer with Brain Metastasis: "Ten Times Better"

Flying to Snow-coated Xinjiang

On the morning of September 5, 2018, my secretary Rita and I boarded a flight to Urumqi. When the plane entered the high altitude of Xinjiang, pure blue and spotless sky came into view. White clouds outside the cabin flowed in waves, rushing and chasing each other. In the distance was a mountain covered with snow. The uncanny workmanship of the nature is so fascinating and intoxicating, making one lost in wild thoughts. Xinjiang, a vast and mysterious piece of land, is home to profound Kunlun loess civilization.

> *Picture 1. Plane up in the sky of Xinjiang.*

I had long been looking forward to this trip to Xinjiang. One day half a year ago, I received a call from a friend in Shanghai. She hoped me to give more attention to Ms. Chen, an inpatient hospitalized in our hospital. I went to a patient room in the third ward and saw Ms. Chen snuggling up in her bed, panting for breath. She was accompanied by her mother. Seeing me, the mother said with tears in her eyes: "Here is our last hope."

Ms. Chen was inarticulate and could not see clearly. Sometimes, she saw shadows from one object, which was typical double vision. I helped her stand up on the ground. It was difficult for her to stand up by herself. Her mother said: "The brain metastatic tumor is too big. From the airport to hospital, it was your hospital service staff who took her here with a wheelchair."

Ms. Chen's Lung Cancer

I checked Ms. Chen's medical history. In late November 2015, she visited a hospital in Xinjiang due to dizziness and inability to stand steadily. CT scan and MRI showed a space–occupying lesion in the left cerebellum. On November 27, 2015, Ms. Chen underwent brain tumor excision. Postoperative pathology indicated metastatic lung adenocarcinoma. Genetic test results revealed that the EGFR Fragment 19 was positive for mutation.

From December 2015 to January 2016, Ms. Chen received whole brain radiotherapy 18 times in total. Her lung CT scan on April 20, 2016 showed a lesion in the upper–right pulmonary lobe. She took iressa (gefitinib tablets) until September 2017 when the disease progressed. Then, she received chemotherapy using pemetrexed combined with erlotinib. She stopped chemotherapy on October 13, 2017 due to Grade III bone marrow suppression, and continued to take erlotinib for targeted treatment.

On March 6, 2018, Ms. Chen's MRI scan of the head exhibited nodules of abnormal enhancement on the residual cavity margin in the surgical area of the left cerebellar hemisphere; and significant nodular enhancement on the edge of the original lesion in the left frontoparietal lobe, suggesting disease progression and occurrence of new metastasis. Her CT scan showed the following: the lesion in the upper lobe of the right lung was bigger than before; the nodule in the basal segment of the lower–right pulmonary lobe grew bigger and was suspected to be metastasis; there were multiple lymph nodes in the mediastinum and hilus pulmonis, and some of them were enlarged; the metastatic nodule in the left adrenal gland was bigger. Later, Ms. Chen was given oral icotinib hydrochloride instead, but stopped taking it 20 days later due to side effects.

> *Picture 2. As soon as Rita stepped in the living room, Ms. Chen hugged her, beaming with a smile like a brilliant flower.*

Ms. Chen came to our hospital in April 2018. At the time of admission, she had dyspnea, chest tightness, palpitation, incoherent speech, and difficulty in walking. Her admission diagnosis was Stage IV adenocarcinoma (cT2aN2M1b) in the superior lobe of the right lung with brain metastasis. She had cerebral edema. In the following week, she was able to articulate after dehydration treatment.

Ms. Chen was not eligible for special treatment. At the time of discharge from the hospital, I suggested that she continue to take AZD9291 orally and inhale hydrogen for at least 6 hours a day. I asked her to keep me updated of her conditions and promised to visit her in Xinjiang a few months later.

In the past two months, Ms. Chen sent messages to my secretary Rita every few days, saying that she was much better and hoped us to visit Xinjiang. "The grapes are ripe. So sweet and good. Come and eat grapes."

> *Picture 3. Ms. Chen's brain metastasis shrank and improved constantly. (Imaging series labelled "Frontal lobe" and "Cerebellar tumor", with rows dated November 20, 2015 and June 28, 2017.)*

"Am I getting better?"

As the plane approached Urumqi, I felt that I smelled the fragrance of sweet and juicy grapes. At the same time, I had an uneasy mind: If Ms. Chen's conditions were not as good as she told us but progressed, how should we deal with it?

After getting off the plane, a friend picked us up, and we went directly to Ms. Chen's home. On the way, I saw cars of all varieties coming and going on a wide and flat street, like a colorful river running nonstop. Golden leaves lied on both sides of the street and decorated the street with their short-lived years. An hour later, the car arrived at the neighborhood where Ms. Chen lived. Ms. Chen's mother was waiting for us in front of the building. Following her, we went quickly to the third floor. The door was open. Standing at the door, Ms. Chen saw Rita and gave her a hug instantly.

It was an apartment with two bedrooms and two living rooms. On the tea table was a string of crystal-clear grapes, like pieces of agate. Ms. Chen and her mother took us to the couch and sat us down, handing us grapes. I could not help but put one grape in my mouth. I was impressed by the delicious juice. The grape was sweet and sour, with pleasant and refreshing fragrance.

I looked at Ms. Chen. She looked at me and smiled: "What do you think? Dr. Xu, am I getting better?" She started pacing in the room. I took her by the hand and tried to test her muscular strength. It was very strong. Then, I gave her a finger matching test and found that her coordination ability was basically normal. I also examined her eye movements and engaged her in a cheek-bulging test. I did not notice any abnormality in the corresponding cranial nerve function.

I checked Ms. Chen's films of CT scan and MRI performed from October 2015 to August 2018. The MRI on August 27, 2018 showed that the brain metastases were significantly smaller. And CT scan indicated that the space–occupying lesion in the right lung was basically stable.

"The 9291 I took was a crude drug I bought online. I don't know whether it works." Ms. Chen said, "The authentic drug is too expensive for me."

Ms. Chen said she started hydrogen inhalation from October 2017 using a hydrogen–oxygen nebulizer. Initially, she did not inhale hydrogen every day. After being discharged from our hospital, she had hydrogen inhalation for 6 to 8 hours on a daily basis. She also inhaled hydrogen before sleep. "I often fall asleep after a few minutes of inhalation." said Ms. Chen.

> *Picture 4. Blood brain barrier: The endothelial cells in capillaries separate blood and brain tissues, forming a barrier that prevents chemo drugs (orally and intravenously) from entering the brain. (Diagram labels: Brain, Smooth muscle cell, Ink cell, Enveloping cell, Endothelium, Blood, Input artery, Arteriole, Blood capillary.)*

Brain: "Harbor" for Lung Cancer

Around half of the patients with non–small cell lung cancer experience brain metastasis. Once the tumor metastasizes to the brain, patients have significantly lower quality of life, neurological dysfunction, and cognitive impairment, with poor prognosis and a median survival of 3 to 6 months. Traditional treatments include surgery and radiotherapy. Since there is a layer of vascular endothelial cells between the blood and brain tissues, cells have certain closely–connected structure in between, forming the so-called blood brain barrier that prevents large molecules from passing freely. Therefore, chemo drugs in the blood, be them

orally applied or intravenously infused, are difficult to penetrate through this barrier. That is, the drugs cannot enter the brain parenchyma and cannot efficaciously exert effects on metastatic cancer cells in the brain. For this reason, the brain is known as a "safe haven" for cancer cells to evade chemotherapy.

In spite of its function to prolong the survival of patients with non–small cell lung cancer, the molecularly targeted drug EGFR–TK inhibitor (TKI) has limited effects on patients with brain metastases. A researcher once found that the concentrations of erlotinib and its metabolites in the cerebrospinal fluid of patients with non–small cell lung cancer accompanied by brain metastases were only 5.1% and 5.8% of plasma concentration respectively. Gefitinib has a lower penetration rate in cerebrospinal fluid, and its concentration in cerebrospinal fluid is only 1.1% to 1.3% that of plasma (J ThoracOncol, 2015; Mol Cancer. 2018; 17: 38).

As a third-generation TK inhibitor, AZD9291 (osimertinib and tagrisso) can pass the blood brain barrier according to a literature report (Medicine [Baltimore], 2017). However, its actual effects are limited.

Ms. Chen Full of Confidence

Ms. Chen's treatment was very standard and complete. She received brain metastasis surgery and radiotherapy, followed by the use of targeted drug erlotinib for 17 months. After developing resistance to erlotinib, she used icotinib instead. Due to intolerance of side effects, she stopped icotinib after only 20 days of use. Her disease progressed. Five months ago, she started taking "9291".

Ms. Chen has been suffering from lung cancer for three years. She is very contented and told us: "Thank you for coming to visit me from afar. I am getting better and better. I am very happy. And I am confident to continue with treatment." (See Picture 5)

The median remission period for treatment with EGFR–TKI is 10 to 14 months. After the use of erlotinib and gefitinib, Ms. Chen's response duration was 23 months, which is in line with the general pattern. In addition, these drugs are difficult to pass through the blood brain barrier. As a result, Ms. Chen's brain metastasis progressed, which was no surprise. Later, she accepted "9291". Although "9291" is not a standard drug, one cannot rule out its potential therapeutic effects on lung tumors, especially on brain metastasis.

Was Ms. Chen's brain metastasis relieved because of AZD9291 or hydrogen inhalation?

As the saying goes, prolonged illness makes a doctor of a patient. I asked Ms. Chen to evaluate her treatment. She said: "Hydrogen is effective. After hydrogen inhalation, at least I can eat well, sleep well, and have certain physical strength." She held out her hand, wanting to compare arm strength with me.

> *Picture 5. Ms. Chen told the author about her daily hydrogen inhalation.*

> *[WeChat message, translated:] Mr. Lin, how have you been? Take good care of yourself. Recently, I found myself with increasingly better complexion. Seeing my ruddy complexion, my colleagues think that I'm no different from a healthy person. Wang Yun said that I have a complexion ten times better than the first time during our video talk.*

> *Picture 6. WeChat message from Ms. Chen to Mr. Lin.*

Since the start of the Hydrogen Tour in 2017, I sincerely hope to see positive effects, and I am always happy for the patients who showed positive results from hydrogen inhalation. Nevertheless, the foundation of medicine is science. As an honest doctor, I want to also see negative effects, that is, cases that indicate hydrogen is ineffective.

Whether hydrogen has brought beneficial effects to Ms. Chen is difficult to conclude based on existing available data. But because of its high safety, hydrogen can be of good use as an auxiliary measure.

One of the biggest features of hydrogen is its selective neutralization of toxic reactive oxygen species, which promote the formation and progression of cancer. In addition, hydrogen is highly dispersive and can pass through the blood brain barrier. Therefore, for patients like Ms. Chen, hydrogen has at least the following functions:

1. Hydrogen alleviates brain damage. As early as 2007, Japanese scholar Shigeo Ohta discovered that hydrogen can ease cerebral ischemic–reperfusion injury and reduce cerebral infarction. In 2017, Ohta's team published a study proving that hydrogen has protective effects against trauma, hemorrhage, inflammation, radiation and brain damage caused by tumors.
2. Hydrogen has inhibitory effects on metastatic cancer cells. Researchers at Chongqing Medical University found that hydrogen can down-regulate Akt phosphorylation, inhibit the PI3K pathway, strengthen the effects of PI3K–specific inhibitors on non–small cell lung cancer cells, and promote cancer cell apoptosis (Mol Med Rep, 2018). Using animal models and cell culture, scholars at Hebei Medical University also discovered that hydrogen can inhibit the growth, proliferation, invasiveness and migration of non–small cell lung cancer cells (Biomed Pharmather, 2018).
3. Hydrogen boosts immune function. CD8+ cytotoxic T lymphocytes have the capacity to kill tumors and play an important role in anti-tumor immunity. However, if the expression of inhibitory receptor PD–1 increases on the surfaces of T cells, the antitumor activity of T cells will be inhibited, putting T cells in depletion. In cancer patients, the T cells that express this PD–1 increase. Japanese researcher Akagi Junji recently tested peripheral blood CD8+ T cells/PD–1 positive cells in 55 patients with Stage IV colorectal cancer. It was found that 39 of the patients had an increase in the number of such cells. After treatment with hydrogen, such cells decreased in 35 cases, suggesting that hydrogen saved depleted T cells and improved the immune functions of T lymphocytes (Oncology Report, 2018).

The research findings are very gratifying. We have discovered the inhibitory effects of hydrogen on PD–1 in experimental studies (based on unpublished data).

4. Hydrogen mitigates systemic symptoms, including by boosting appetite and eliminating insomnia and fatigue. Korean scholars have found in animal experiments that the animals with hydrogen inhalation had more endurance and were less likely to get tired in swimming (Biomed Res, 2018). According to American scholars, hydrogen can effectively relieve various adverse effects caused by radiotherapy and chemotherapy (International Journal of Clinical Medicine, 2016).

Afterword

Two months after the article above was finished, I received a message forwarded by Mr. Lin from Ms. Chen: "Recently, I found myself with increasingly better complexion. Seeing my ruddy complexion, my colleagues think that I'm no different from a healthy person. Wang Yun said that I have a complexion ten times better than the first time during our video talk."

— Xu Kecheng, Lu Tianyu

> **Comments from Tang Zhaoyou**
> Thoughts thronged my mind after reading the article above. My wife had breast cancer with positive Her–2 and metastasis to underarm lymph nodes. She did not receive radiotherapy

> and chemotherapy after surgery. She discontinued herceptin half way due to worries about cardiotoxicity (causes cerebral infarction later). After surgery, she persisted in swimming and oxygen inhalation. After 11 years of breast cancer, she passed away without cancer recurrence. In her case, I do not deny the role of herceptin (using half the dose leads to no effect according to literatures). And it is also difficult to rule out the effects of swimming and oxygen inhalation. In Ms. Chen's case, her conditions improved probably because of AZD9291. However, such significant improvement must be somehow attributable to hydrogen inhalation. Cancer is a chronic disease that involves multiple genes. For this reason, long-lasting holistic treatment is the only way out. Cancer transformation methods (such as hydrogen inhalation) can be applied after cancer elimination treatments. The two might be mutually complementary. As Mao Zedong said, "In long-lasting and cruel wars, guerrilla warfare will show its great power."

Hydrogen and Lung Cancer with Brain Metastasis: Farewell to Suffering

Flying to Dongying

On July 20, 2018, my secretary Rita and I flew from Guangzhou to Jinan, from where we would transfer to Dongying, a city on the northern coast of Shandong province.

There are probably not many people who know the city Dongying. But everyone knows about Shengli Oil Field. Dongying is a new city that emerged with the flowing of oil. In recent years, there has been an advertisement played on CCTV in the prime time: "Yellow River into the sea, Damei Wetland". The advertisement is about Dongying.

The climate on the earth has indeed changed. Nowadays, the farther north you go, the hotter it gets in summer in China. As soon as we got out of Jinan Airport, a heat wave in the air sprayed into our faces. Mr. Shao, who had made a special trip from Dongying, greeted us loudly, came up to help us with luggage and took us to the parking lot. The moment we were seated in the car, Mr. Shao could not wait to tell me: "Ms. Wang's cotton cloth shop opened again."

I came to Dongying at the invitation of the Dongying Medical Association and Dongying Cancer Hospital to give a speech about practicing Chinese way of cancer control. But I would be happy to visit Ms. Wang. I had been worried about her conditions: Is her brain metastasis controlled? Did the enlarged lymph nodes on the left clavicle disappear?

I clearly remember the first time I met Ms. Wang at the end of March 2018. It was in a medical center near Zhongguancun in Beijing. Accompanied by Mr. Shao, Ms. Wang came specially to consult me about her case. The first thing she said to me was: "For how many months can I still live?"

Miraculous Survival

> *Picture 1. Images before and after brain metastasis in Ms. Wang's case. The MRI in 2018 showed notable improvement. (Imaging dated September 2017, March 2018, July 2018.)*

Shortly after breakfast the next day, Mr. Shao and his wife drove the hotel and took us to the eastern part of the city.

The morning in Dongying was quiet. The straight and wide avenue as well as unique ginkgo trees and green shade give first–time visitors unlimited imagination. Ginkgo trees along the road stretched out with great grace, swaying. I saw people walking or jogging. With typhoon about to come according to weather forecast, the air was extraordinarily fresh and gave off a pleasant coolness. The gentle mist was slowly dissipating.

After over 20 minutes' drive, the car stopped at a shop facing the street. Ms. Wang was waiting at the door. Seeing us, she greeted us, helped open the door, took me by the hand, and walked me into the shop. The tables in the shop were full of various cotton cloth. The walls were covered with curtain fabrics of different specifications. Ms. Wang told me that she opened the shop again after her conditions got better in the past few months. I asked her whether the business was good. She smiled and said: "I don't know why. But business is much better than before."

> *Picture 2. Mr. Wang's CT scan showed that the lesion stabilized and shrank slightly. (Imaging dated September 2017, March 2018, July 2018.)*

Mr. Shao smiled, saying: "Dongying is a small place. People bump into each other easily. Now that Ms. Wang's cotton shop has opened again, everyone comes to buy things. On the one hand, they want to help her. On the other hand, they want to see Ms. Wang who has survived cancer miraculously."

I looked at Ms. Wang. She had rosy cheeks and had gained more weight than last time when I saw her in Beijing a few months ago. I asked her if she had cough and headache. Then, I examined her left supraclavicular lymph nodes. She said, "The symptoms you said are gone." She smiled and pointed to a sewing machine inside the house, saying that she got up early every day. She was so busy from day to night that she had no time to think about her disease. It was only because I came to visit that she went to the hospital for a follow-up examination.

> *Picture 3. Ms. Wang's original enlarged lymph nodes on the left clavicle disappeared. Dr. Wang, the president of Dongying Cancer Hospital, examined her (author: first from the left).*

I checked Ms. Wang's CT scan and MRI reports, which indicated significant improvement compared to the medical reports in March. According to the imaging scans, there were still tumors in the lung and metastases in hilar lymph nodes and the brain. But all the lesions became smaller than before.

I held the hand of Ms. Wang and said, "Congratulations. Your experience and treatment results have given me much support." With tears in her eyes, Ms. Wang said: "When I was rescued in the ICU a year ago, the doctor said that I had only dozens of days to live. My husband even had graveclothes prepared for me." She pointed at the Mr. and Ms. Shao, saying: "Without them, I am already long gone. It is because of them that I started inhaling hydrogen on June 6, 2017."

The Shao couple were classmates of Ms. Wang in middle school. Mrs. Shao said: "At that time, it was heart-wrenching to see that Wang was not able to eat and walk and had serious headache." Knowing that there was no other better option, the Shao couple borrowed a hydrogen–oxygen nebulizer from a friend in Shanghai and sent it to Ms. Wang. After that, Ms. Wang inhaled hydrogen for at least 6 hours a day.

Mr. Shao is a fan of hydrogen. He used to study chemical engineering at school. In recent years, he has been obsessed with hydrogen and has applied for five inventive patents on

hydrogen applications. He showed me a photo, in which there were two daffodil plants. The plant on the left was short without any daffodil bud, but the one on the right was twice taller with bright daffodil buds. I was told that the plant on the left was watered with tap water, while the one on the right was watered with hydrogen–rich water.

I asked Ms. Wang: "Aren't you taking crizotinib?" Crizotinib should be a specific drug for Ms. Wang whose ALK was positive. She answered: "I started taking crizotinib in May 2017. A month later, my cough was eased but headache became worse. I was found to have bleeding from the brain tumor. That was the most dangerous time since I was sick. Later, my doctor told me that crizotinib had no effect on brain metastasis." Ms. Wang took out a medicine bottle labeled "crizotinib" with handwritten "270 mg × 60". There was no batch number, place of origin or date of manufacture on the bottle. Ms. Wang said that it was a crude drug from the black market. She said embarrassingly: "Without money, I can only afford such a drug. I don't know whether it's authentic or not."

According to Mr. Shao, on September 6, 2017, three months after hydrogen inhalation, Ms. Wang took a CT scan. Results showed that the lung carcinoma and metastatic lymph nodes were smaller than before; and that the original plural and pericardial effusion on the right side had disappeared. The only disturbing thing was the appearance of a new metastatic tumor in the brain. Mr. Shao thought the only option at that time was to extend hydrogen inhalation time. "She inhaled hydrogen for more than 10 hours a day. Maybe there is a membrane between the brain and outside world. So, it might take longer time for hydrogen to enter the brain." said Mr. Shao with a smile. "I am not a doctor. If I am wrong, don't laugh at me."

"You are right. There is a natural barrier between the brain and blood. It is known as blood brain barrier. Why is crizotinib not effective in brain metastasis? Because it cannot pass through the blood brain barrier. Hydrogen molecules are small and may pass through this barrier." I explained.

"What do you think turned the situation of your disease around?" I asked Ms. Wang, trying to understand the key of her complicated treatment.

"It was hydrogen inhalation. It was definitely hydrogen, without any doubt." Ms. Wang sounded firm and grateful. "A week after hydrogen inhalation, I was able to eat normally. Two weeks later, I could stand up by myself and go shopping outside. My headache was eased too."

Misery from Treatment

The 47–year–old Ms. Wang had been running a cotton fabric shop for a few years and was responsible for both selling and processing curtains. She was both the boss and employee, earning only a few thousand RMB yuan a month. Her husband worked in the logistics industry in another city and did not earn much money. They had two children at school age. In spite of poverty, Ms. Wang and her family lived happily together.

In September 2015, Ms. Wang had unexplained cough, which became increasingly serious. On September 21, she went to Dongying Hospital for examination. She received chest X-ray and then a CT scan. After reading Ms. Wang's imaging reports and CT films, the radiologist asked her to visit an oncologist.

Oncologist? Ms. Wang panicked slightly and started reading the medical reports carefully. The reports stated: right lung cancer with metastasis in the hilus pulmonis, mediastinum and left supraclavicular lymph nodes; cancerous lymphangitis in the lower–right lung; and abnormally

enhanced nodule in the right occipital lobe. After reading, Ms. Wang had turbulence in the heart and felt that a disaster just befell her. Then, tears started trickling down her face.

Ms. Wang was hospitalized and then took bronchoscopy, during which sample tissues were taken under the microscope for biopsy. Biopsy results indicated adenocarcinoma. On September 29, Ms. Wang received chemotherapy. Three weeks later, her husband came back from another city and took her to Beijing by train the next day. After waiting for a few days, Ms. Wang was finally admitted to the Cancer Hospital of Chinese Academy of Medical Sciences.

Several days later, Ms. Wang was given chemotherapy. From October 21, 2015 to February 8, 2018, she had chemotherapy using two drugs, followed by administration of a single drug for maintenance. Then, she had chemotherapy with another two drugs, followed by use of a single drug for maintenance. The process lasted for 17 months. After that, Ms. Wang received radiotherapy 32 times in 48 days. Eight months after chemotherapy, she had a CT scan, which showed stable disease. However, another CT scan two months later showed significant progression of disease. Five months after that, Ms. Wang had a follow-up CT scan and MRI. The results indicated disease progression combined with cerebral hemorrhage…

I read the summary of Ms. Wang's medical records provided by Dongying People's Hospital as follows.

Ms. Wang remembered being so happy when the doctor told her that her conditions were stable as indicated by the CT scan on July 7, 2016. All her hair was gone; and the vomiting after chemotherapy once made her wish to die. Nevertheless, she seemed to see hope after the doctor told her that stable disease meant successfully treatment.

"Why did it progress in such a short time?" Ms. Wang was referring to the findings of a CT scan 2 months later on September 7, 2016. The scan showed significant progression of disease. "I have suffered such much from chemotherapy. Eventually, the disease still progressed. It's so sad." said Ms. Wang.

Table: Ms. Wang's medical record abstracts

Date	Conditions and Examinations	Treatment
2015/09/21	The patient came for examination due to cough and headache. CT scan findings showed right lung cancer with metastases in the hilus pulmonis, mediastinum, and left supraclavicular lymph nodes; cancerous lymphangitis in the lower–right lung and abnormally enhanced nodule in the right occipital lobe.	
2015/09/24	Bronchoscopy revealed new organisms in the right bronchus. Biopsy: adenocarcinoma.	Chemotherapy: pemetrexed 0.8g (d1) + cisplatin 40mg (d1–3).
2015/09/29	Genetic test: no EGFR mutation.	Chemotherapy: same regimen as above; one cycle every 21 day; 4 cycles in total.
2015/10/20–2015/12/01	Vomiting, stomach pain, and pain in the limbs.	
2015/12/22–2016/08/19	On April 15, 2016, the fluorescence in situ hybridization of ALK (2p23) showed chromosomal translocation (ALK positive). The CT results of EGFR re–examination indicated no mutation. The CT scan on July 7, 2016 led to the	Chemotherapy: pemetrexed 0.8g; once every 3 weeks; maintenance with a single drug.

Date	Conditions and Examinations	Treatment
	following comprehensive evaluation: stable disease.	
2016/09/07	CT scan showed right lung cancer with metastases in the brain and mediastinal lymph nodes, and lymphangitis in the lower–right lung. Compared with the CT scan findings two months earlier, lesions in the lung and brain exhibited significant progression.	Crizotinib was recommended but was rejected by the patient due to financial problems.
2016/09–10–2016/11/17	Vomiting, severe headache.	Chemotherapy with docetaxel 100mg (d1) + cisplatin 40mg (d1–3) for 3 cycles (every 21 days for a cycle); followed by administration of docetaxel 100mg (d1) once every 3 weeks for maintenance purpose.
2017/02/08	CT scan showed effusion in the right thoracic cavity and pericardium in addition to the aforementioned lesions. MRI showed multiple metastases in the occipital and parietal lobes with hemorrhage.	Radiotherapy (on the right occipital lobe metastasis): total dose 95% PTV 60 Gy/30F. Dehydration reduced intracranial pressure.
2017/02/14–2017/03/28	Headache, vomiting, unable to walk.	The patient was given emergency treatment in the ICU.
2017/04/12	Sudden headache, aggravated vomiting, lethargy.	The patient purchased crizotinib without batch number by herself and took 2 pills per day continuously.
2017/05/07	Reduced cough, headache, dizziness, aggravated vomiting.	(continued)
2017/06/06	After a week, the headache was relieved, and the patient could walk by herself gradually.	Hydrogen inhalation (hydrogen and oxygen mixture); flow rate at 3 liters / minute; 8 hours a day; continuous inhalation.
2017/09/07	Follow–up CT scan showed that the lung cancer and lymph node metastases shrunk; thoracic and pericardial effusion disappeared; but a new metastatic tumor appeared in the brain.	Hydrogen inhalation time was extended to 10 hours a day.
2018/03/07	CT scan showed that the lung cancer and brain metastases were smaller than before.	Continued hydrogen inhalation.
2018/07/21	CT scan showed that the lung cancer and brain metastasis had shrunk further.	Continued hydrogen inhalation.

Changing the Way

The question asked by Ms. Wang is about a commonly seen issue. According to literature reports, terminal lung cancer patients like Ms. Wang have a median survival time less than one year. The only treatments for them are chemotherapy and radiotherapy. The treatments Ms. Wang received are very standard and in line with all the available official guidelines. These treatments extend the median survival time, but the final outcome is still disease progression. Therefore, median survival time is also known as "survival time to disease progression". Ms. Wang was actually in "survival time to progression" after her conditions became stable after chemotherapy. From the start of chemotherapy in September 2015 to disease progression in May 2017, Ms. Wang survived for 18 months, which is already a very good treatment result.

Fortunately, Ms. Wang's anaplastic lymphoma kinase (ALP) is positive. Due to chromosome inversion, a gene called echinoderm microtubule–related protein 4 (EML4) is rearranged with ALK genes (EML4–ALK). The rearrangement promotes lung cancer development and

progression. This mainly occurs in cases of non–small cell lung cancer, with an incidence rate of 3% to 5%. Patients with such gene rearrangement are prone to brain metastasis. At the onset of her cancer, Ms. Wang had brain metastasis. As a targeted drug for ALP–positive lung cancer, crizotinib is effective in 40% to 50% cases and helps prolong the median survival of patients to 12 months. However, crizotinib cannot easily pass through the blood brain barrier and therefore has very limited effects on brain metastasis. At the American Society of Clinical Oncology (ASCO) annual conference in June 2018, there were reports stating that alectinib, the third–generation ALP–targeted drug, could pass through the blood brain barrier and was undergoing a global multi–center phase–3 clinical trial. But for Ms. Wang, the drug is far from being accessible.

In order for patients like Ms. Wang to survive for a long time and live with high quality, treatment must be switched according to different thinking. That is, the key is not whether the tumor disappears, but lies in the overall health of the patient (coexistence with cancer).

Academician Tang Zhaoyou proposed Chinese way of cancer control, which advocates the use of cancer killing techniques in combination with cancer transformation measures. The treatments that Ms. Wang received are all targeted at killing cancer. They are effective but only in a limited way. Ms. Wang has survived and is now living like a healthy person mainly because of the cancer transformation measure she has taken. Hydrogen inhalation is a cancer transformation measure. Hydrogen molecules can selectively fight against and eliminate toxic free radicals (ROS); and transform cancer cells, the microenvironment and the entire body. It is hydrogen that made it possible for Ms. Wang to live with cancer.

Afterword

Ms. Wang reopened her shop and bid farewell to suffering. On the evening when I met Ms. Wang, I stood on the corridor on the highest floor of the hotel I stayed in, watching the blue sky decorated by floating clouds. I looked in the direction where the Yellow River flowed into the sea, with the round moon rising above me. It seemed that I saw the Yellow River, the mother river of the Chinese nation, emitting a glow of light from thousands of years of Chinese traditional culture.

> *Picture 4. Significant improvement after hydrogen inhalation. (Timeline: Diagnosis 2015.9 → Chemotherapy → 2016.6 Stabilization → 2017.1 Progression [Significant progression] → Radiotherapy → 2017.6 Rescue from progression → Hydrogen inhalation / "Crizotinib" → 2018.1 Improvement → 2018.6 Improvement.)*

"Chinese way of cancer control" is a concept rich in Chinese elements and Chinese philosophy. Hydrogen molecules serve to transform cancer cells and represent an important strategy and means to practice "Chinese way of cancer control". Using hydrogen molecules symbolizes a changed way for treating the most challenging and tricky cases.

As a famous Chinese poem says, "If you desire for a grander view, ascend to a higher floor." Success belongs to the wise and brave who dare to innovate and ascend.

— Xu Kecheng

> **Comments from Tang Zhaoyou**
> No one would object to the saying: "Practice is the sole criterion for testing truth." And most people would agree that "Out of accidental events come the inevitable." Nevertheless, when faced with specific issues in real life, doctors (including me) who have received Western medicine training will ask if there is support by evidence–based medicine proof. As a result,

> they miss out on the potential points for innovation. Scientific progress is usually the result of continuous development based on the hard work of our predecessors. For over a century, cancer has been treated following the killing strategy. Much has been achieved in cancer treatment, but a full victory is not won yet. Nowadays, we see patient cases in which unexpected results were attained as a result of cancer transformation measures. Such cases deserve great attention and are worth studying.

Hydrogen and Liver Cancer: A Surprise

Mr. Zeng Visits Guangzhou

Mr. Zeng Congxian is a resident of Taiwan. I invited him to Guangzhou for examination, because I believed that if his liver cancer had been really cured as he stated, his case would be an example that verifies the scientific nature of the concept "China's way of cancer control".

> *Picture 1. Academician Tang Zhaoyou said: "Cancer is a dynamic disease. Cancer treatment varies with timing and individual patient; and should be focused on transforming cancer cells and the entire body."*

Mr. Zeng flew from Taipei to Guangzhou on March 22, 2018. That afternoon, Yangcheng Evening News Publishing House held a press conference for the launch of my new book *Practice Chinese Way of Cancer Control*. The press conference was attended by many people, including Academician Tang Zhaoyou who specifically came from Shanghai and made a speech about Chinese way of cancer control. Originally, I expected Mr. Zeng to come and listen to academician Tang's speech, because Mr. Zeng had inhaled hydrogen for a few months in Taipei. The main strategy of Chinese way of cancer control proposed by Academician Tang is to combine cancer elimination and transformation treatments. And hydrogen inhalation might be a means of cancer transformation. Unfortunately, due to flight delay, Mr. Zeng did not make it to the press conference. He expressed that it was a pity. He came to Guangzhou together with Mr. Lin, an entrepreneur from Taiwan, and Mr. Fu Daren and his wife. As a well-known TV host in Taiwan, Fu Daren was suffering from pancreatic cancer. He was ready to go to Switzerland for euthanasia. Mr. Lin and I urged him to abandon euthanasia and eventually convinced him to visit our hospital for treatment.

The Guangzhou trip was not easy for Mr. Zeng. He said that it was his first time to go out of Taiwan island. Seeing the big and nice Baiyun Airport and many beautiful high-rise buildings in Guangzhou, Mr. Zeng joked: "If it were not for your invitation, how could a country folk like me have the opportunity for this absolutely eye-opening experience?"

Visit to Taoyuan, Taiwan

March 7th, 2018 was the last day of our Hydrogen Tour in Taipei. We planned to go shopping in the afternoon, because some of us came to Taiwan for the first time. But after hearing about a liver cancer patient whose originally large tumor had almost disappeared, all team members became interested and wanted to visit this patient. Our host Mr. Lin told us that the patient lived in a small place far away and reminded that the trip might be tiring. Hoping to see the daily life of ordinary residents in Taiwan, we asked the driver to take us there. Then, we went straight to Zhongli in Taoyuan County, which was 50 kilometers away from Taipei and a typical settlement for multi-ethnic groups in Taiwan.

Maybe the weather was about to change. It was very hot that afternoon, and there were drizzles from time to time. The car drove for more than an hour and stopped in front of a shop next to a street. Mr. Zeng came up, greeting and shaking hands with us. Upon entering the shop, we were asked to sit down at a simple square table. Mr. Zeng had set up teacups, and the water in the teapot on the electric stove was boiling. The tea used was the famous Taiwanese high mountain tea. Mr. Zeng poured tea for each of us, in the same way as the Cantonese procedure of tea drinking. I said: "It seems that we are back in Guangdong." Mr. Lin smiled: "We are all descendants of the dragon and one family on both sides of the strait."

Measuring around 50 square meters, this house was one part of a row of houses facing the street and had two floors. In the house were piles of planks and several tool stands with various woodworking ware on them. Mr. Zeng told us that he originally opened a couch shop. In recent years, he switched to advertising and decoration boards business. The business was not good. However, even during his hospitalization, Mr. Zeng still came back here to run the shop.

Discovery of Huge Liver Carcinoma

Mr. Zeng's medical history was not complicated. Due to pain in the upper-right abdomen, he went to the Linkou ×× Hospital for examination in October 2016 and was diagnosed with liver cancer. At that time, the intrahepatic tumor measured 14cm and was accompanied by portal vein tumor thrombus. Mr. Zeng was given chemotherapy with intravenous injection of chemo drugs once a week. Afterwards, he received hepatic artery interventional embolization 3 times. A few months ago, the doctor told Mr. Zeng that the tumor was very small and needed no further treatment.

> *Picture 2. CT scan (plain scan) (No18131) in Taiwan Linkou XX Hospital on November 16th, 2016: A large space–occupying lesion is noted in the right lobe of the liver and shows irregularities on the edge. The lesion is surrounded by focal lesions. The portal vein shows tumor thrombus.*

"Did you receive any other treatment?" we asked at the same time. Dr. Wang from Tongji University opened his eyes in surprise and smiled at me. He added: "Is it so miraculous?" Mr. Zeng took out his medical records provided by Linkou xx Hospital. In the records, liver function test results indicated slightly high levels of transaminase and alkaline phosphatase; normal levels of albumin and bilirubin; and positive marker of hepatitis B virus infection. According to the records, Mr. Zeng's alpha–fetoprotein (AFP), the main marker of hepatocellular carcinoma, showed no increase. This was not surprising, because 30% of the patients were false negative in AFP. The most important record was liver biopsy report, which indicated hepatocellular carcinoma. The diagnosis was unquestionable. "Oh, I inhale hydrogen every day." Mr. Zeng pointed to Mr. Lin and said, "Thanks to the help of Mr. Lin. Thank you!!"

Mr. Zeng's brother worked in a Taiwan–funded enterprise in Shanghai and knew Mr. Lin personally. He also knew that Mr. Lin was studying hydrogen medicine. He heard about a case in which the 80–year–old mother of another Taiwanese businessman suffered from gastric cancer; insisted on not taking surgery but only hydrogen inhalation; and had managed to survive 4 years and live well. He hoped to get help for his brother from Mr. Lin. Mr. Lin introduced Mr. Zeng to the hydrogen inhalation experience center next to Linkou ×× Hospital. Mr. Zeng took it seriously and had hydrogen inhalation in the center for 2 to 3 hours a day, even during the period of chemotherapy. Basically, once his treatment was completed, Mr. Zeng went to the center for hydrogen inhalation. He said: "The service staff there is very nice. They let him inhale hydrogen for free and sometimes offer me food."

"You secretly came back to run the shop during the period of chemotherapy. How did you make it?" I asked.

"I didn't have any discomfort. I ate and slept like a healthy person. And I had the physical strength to work. Other people always say that chemotherapy is hard. But it was smooth for me." Mr. Zeng pointed to the half–made plates in the house. "Nowadays, life is not easy for Taiwanese people. How can I afford to live a life without working? I have to support my family."

In the interviews over the past days, almost all of the respondents reported that they could eat and sleep well and gained physical strength after hydrogen inhalation. Moreover, the respondents did not complain of hair loss or vomiting during chemotherapy. Mr. Zeng looked very healthy, with a strong body and rosy cheeks. I felt his strength when shaking hands with him. It seems that all of this was the result of hydrogen inhalation. But what I cared most about was Mr. Zeng's exact status of tumor.

I asked him to go to the hospital for a checkup or at least a CT scan. He looked reluctant and said nothing. Was it because of money? I heard that everyone in Taiwan is covered by national health insurance. Mr. Lin whispered to me: "Taiwan's health insurance does not cover everything."

I took Mr. Zeng by the hand and said, "Have you been to the mainland?"

He shook his head: "There is never a chance."

I said to Mr. Zeng: "Mr. Lin and I want to invite you to Guangzhou and come to our hospital for a CT scan, ultrasound and blood test." Seeing that he was hesitant, I decided on behalf of Mr. Lin, "Mr. Lin will cover your flight ticket, and I will pay for all the checkups in our hospital. Okay?" I held Mr. Zeng's hand tightly and said: "Guangzhou is quite beautiful now. Promise to come. I'll see you in Guangzhou."

As mentioned in the text above, when Mr. Zeng, Mr. Lin and Mr. Fu Daren and his wife arrived in Guangzhou, the press conference for my new book had been over. I invited them to dinner in Soluxe Hotel Guangzhou and then sent Mr. Zeng to our hospital for hospitalization. Since I had to go to Beijing for a meeting the next day, I worked with the ward doctor that night to make an examination plan for Mr. Zeng.

Surprising Examination Results

The next night when I was having dinner with my friends after the meeting in Beijing concluded, I received a WeChat message from Mr. Zeng's doctor: "Mr. Zeng's CT scan was done and showed that the lesions are significantly smaller with significantly lower activity. It seems that the treatment is effective." Everyone at the dinner table could not help but stood up and cheered at the good news with me.

> *Picture 3. At my invitation to Guangzhou for examination, Mr. Zeng held my hand tightly and said, "Professor, you are a good person."*

The following day, I flew back to Guangzhou without any delay and went directly to the hospital where I checked Mr. Zeng's imaging films together with our radiologists. The CT scan revealed the following: The S7 and S8 of the liver displays a quasi–circular low–density focus, which measures 7.8 × 7.5cm. The focus shows clear edges, deposition of very little iodized oil, and insignificant enhancement. Liquefaction and necrosis are noted in the lesion. Compared with

the CT scan findings in November 2016 in Taiwan, the findings this time indicated that the tumor shrank considerably and its activity decreased significantly.

> *Picture 4. CT scan in Guangzhou Fuda Cancer Hospital on March 23, 2018: The right S7 and S8 of the liver displays a quasi–circular low–density focus with clear edges and pseudo envelopes. The lesion exhibits no enhancement inside, and its CT value is 27HU. Inside the lesion is a low–density necrosis area (in the circle indicated by the arrow), indicating that the tumor is inactive with liquefaction and necrosis. (Images: Plain scan, Arterial phase, Venous phase, Lag phase.)*

Seeming "Disease–free Survival"

Hepatocellular carcinoma is one of the most common malignant tumors in China. And the preferred treatment is surgical resection. However, due to absence of symptoms in the early stage, patients often have no chance of surgery at the time of diagnosis. Minimally invasive ablation (such as cryosurgery and irreversible electroporation or nanoknife) effect a radical cure for some patients with inoperable liver cancer. Nevertheless, more than 60% patients are not eligible for surgery and ablation. For such patients, chemotherapy does not prolong lifetime. As the main means of treatment, percutaneous transhepatic arterial chemoembolization and targeted drugs (such as sorafenib) are generally believed to be effective in only prolonging progression–free survival rather than overall survival. Studies show that these treatments promote tumor growth after producing positive effects for a period of time.

Mr. Zeng's liver cancer was pathologically confirmed to be hepatocellular carcinoma. According to the findings of his first CT scan, the carcinoma was giant liver cancer combined with cirrhosis and portal vein tumor thrombus, which could not be treated surgically. In Taiwan, Mr. Zeng received only chemotherapy and hepatic arterial chemoembolization. In general cases, patients like Mr. Zeng have a survival period of around 6 months. However, surprisingly, Mr. Zeng has not only survived, but is also in good overall conditions. He seems to be free of disease, with almost complete liquefaction and necrosis in the original space–occupying lesion of liver cancer. Though it is not clear whether Mr. Zeng is cured, his conditions have undoubtedly improved significantly. Judging by experience, I do not think this result can be achieved simply by several sessions of chemoembolization and systemic chemotherapy, which Mr. Zeng received. During the period of treatment, Mr. Zeng had hydrogen inhalation. Did hydrogen molecules play a role in his case?

> *Picture 5. The author went directly to the hospital after landing in Guangzhou and checked Mr. Zeng's imaging films with a radiologist.*

As early as 1975, Dole et al. published a study in the journal Science. In the study, animals were given continuous inhalation of 97.5% hydrogen at eight atmospheres, which effectively inhibited squamous cell carcinoma of the skin. The authors of the study were the first to propose that hydrogen inhibited tumor growth through antioxidation. Japanese scholars found that feeding hydrogen-containing water to animals with cirrhosis helped prevent liver cancer. Some researchers once processed tongue cancer cells with hydrogen water and discovered that cancer cell growth was suppressed. According to the research findings of scholars from Shanghai Jiaotong University, hydrogen inhibits animal models of intestinal cancer, and promotes cancer cell apoptosis and suppresses cancer cell proliferation by regulating redox microenvironment and interfering with gene expression related to cancer cell proliferation.

Hydrogen has proven to be a selective, non–toxic, residue–free, and highly affordable antioxidant. In the "elimination + transformation" strategy for Chinese way of cancer control, hydrogen molecules seem to function to transform cancer cells and the microenvironment. Although we cannot say that hydrogen plays a leading role in the treatment of cancer, including liver cancer, it can be used as a supporting and complementary means in comprehensive treatment.

With liver cancer that cannot be surgically resected, Mr. Zeng received conventional treatments. Eventually, the carcinoma was not only unexpectedly controlled, but the whole treatment for Mr. Zeng was smooth. In his case, hydrogen made undeniable contributions to the therapeutic effect.

(On March 5, 2019, which was exactly one year after our visit to Mr. Zeng in Taiwan, we called him. Mr. Zeng was very happy to hear from us and said that he goes to the hospital for ultrasound every three months. The result is "Still the same as before. It's good.")

— Xu Kecheng

> **Comments from Tang Zhaoyou**
> When it comes to cancer treatment nowadays, people hope to see the problem solved with a little "hammering here and battering there". They hardly realize that cancer is a chronic, systemic, and dynamic disease. It takes a few days for a cold to develop and also several days to fully recover from it. However, the formation of cancer often takes ten or even decades. And recovery from cancer will definitely require a period of time by year. To this end, cancer transformation treatments are indispensable at the same time as and after tumor killing methods are applied. This is why I proposed to supplement "quick actions to eliminate the tumor" with "protracted measures of cancer elimination combined with cancer transformation". Hydrogen exerts inhibitory effects on cancer cells through antioxidation and anti–inflammation mechanisms. I believe that hydrogen inhalation will have a place in "protracted treatment with cancer elimination combined with cancer transformation methods".

Hydrogen and Cholangiocarcinoma: "Birth of a Pearl from a Sick Clam"

"Beckoning" of Wuyi Mountain

I have long wanted to write a story about hydrogen and cholangiocarcinoma. But it was postponed because I wanted to go to southeast China's Yunnan to visit Mr. Chen. A while ago, I went to Wuyi mountain in Fujian to attend the Cancer Rehabilitation Conference. Driven by two things in mind, I immediately started writing.

> *Picture 1. Writer Xu Niang, the author, and famous journalist Ms. Li (left to right).*

One thing is my meeting with Miaoxuan, a bright, spiritual, lively and open woman. Seeing me, she gave me a hug right way, without thinking about the Chinese social code that renders body contact between man and woman in public inappropriate. As the saying goes, there is no unrivalled beauty in a thousand years; and a beautiful woman is always pleasing to the eye. Miaoxuan was my patient. But today, she looked beautiful enough to feast the eyes. She took my hand and jumped like a girl, saying affectionately: "Thank you. Thank you very much, my savior."

The other thing that inspired me to write this story is my meeting with a beautiful female writer. She is tall and a little chubby in a way without losing the slightest artistic temperament. With thick black hair and wearing a long coat, she looked as if one of the ladies depicted in a famous poem: "The weather's fine in the third moon on the third day. By riverside so many beauties in array. Each of the ladies has a fascinating face. Their skin is delicate, their manner full of grace." She gave me a copy of *Birth of a Pearl from a Sick Clam*, a book in which she recorded her experience, thoughts and feelings in her encounter with "Mr. Tumor". Having the same family name as me, she used the pen name Xu Niang. She said: "We were one family five hundred years ago." All of sudden, the distance between us narrowed.

Birth of a Pearl from a Sick Clam

I had a quick reading of the preface for *Birth of a Pearl from a Sick Clam* and thumbed through the whole book. Like the author's nice figure and temperament, the book makes a great reading written in freely flowing style and full of inspirations, vivid expressions and sparkling ideas.

Xu Niang was found to have breast cancer in 2013. The book mentioned above is about her over 4 years of battle with tumors. She received surgery only without chemotherapy. Till today, she keeps her elegance as before. She presented many of her sentiments and insights in the book, which "gives readers a glimpse of the sunshine from the haze of the canopy."

Ms. Xu loves pearls, because she believes that "Only the story of the pearl and clam can make the closest interpretation and most accurate analogy for those who have battled with tumors."

A clam is swimming freely in the water and opening its big shell from time to time. Suddenly, a grain of sand enters its body with rushing waves. Unable to send out the invading sand, the poor clam mobilizes all the force in the body and wraps the sand with blood and tears. The two start growing together. Eventually, the grain of sand turns into a beautiful pearl. "Rising and gathering together, rosy clouds enter the mortal world along with the breeze, giving off a dazzling brightness."

With her wisdom, Xu Niang, like the clam, has mobilized all the power in her. Eventually, she tamed the cancer cells that invaded her body and found back her territory of health, sunshine, elegance and comfort. She herself has become a "precious pearl of achievement".

Miaoxuan's Fight

On the morning of December 2, 2018, Miaoxuan and I, holding the banner "Light of Life", walked shoulder by shoulder on the lush highway beside Wuyi Mountain together with hundreds of people. We walked fast. It was an event called Ten Thousand Meter Walk organized by the conference sponsor. While walking, we breathed the fresh air of the mountain and enjoyed the natural beauty of green hills and clear water, experiencing a sense of unity with the nature.

> *Picture 2. Walking on a road next to Wuyi Mountain together with Miaoxuan (right) and Ms. Quan (left).*

Walking next to me, Miaoxuan did not seem to have any sign of cancer. She is a perfect example of the shiny pearl depicted in Xu Niang's book.

At the Cancer Rehabilitation Forum held in Honglin Hotel in Wuyi Mountain the next morning, Miaoxuan went to the stage with medical experts. She spoke on the stage: "In 2010, I received

an operation on cholangiocarcinoma. My cancer recurred once every year after the operation. And I had to take an operation again and again. In total, I have had three operations and three sessions of chemotherapy, which almost crashed me. However, in the past 4 years, my CT scan, MRI, ultrasound and blood tumor marker results have been normal. I am happy and energetic every day." Looking at me in the audience, Miaoxuan continued: "Thanks to the correct rehabilitation methods that Prof. Xu Kecheng made for me."

Compared with the breast cancer that Xu Niang had, Miaoxuan's tumor was ten times more malignant. I remember one day in 2014, I was at the specialist clinic. A tall lady came and handed her medical records to me. She was Miaoxuan from Shenzhen. Without telling me her medical history yet, Miaoxuan had tears rolling down her cheeks, gently asking: "Do you think there is still hope to save me?"

Like me, Miaoxuan was suffering from cholangiocellular liver cancer. Every time I saw a patient with such cancer in the past few years, I was struck by pain that grabbed the heart. After all, I suffered from the disease myself and have special sympathy for such patients. I remember that on a previous ward round, I saw a patient with cholangiocellular liver cancer, which recurred after 12 years. I was worried about the patient for a few days afterwards. Almost every time during my consultation at the specialist clinic, I met several patients with cholangiocellular liver cancer. All of them had relapse shortly after surgery or were found to have extensive metastases at the time when their cancer was detected. Pancreatic cancer is known as the "king of cancer". In fact, cholangiocellular liver cancer is similarly malignant as pancreatic cancer, with less than 10% of the patients surviving for 5 years.

Cancer recurs due to many reasons, which are extremely difficult to identify for individual patients. Apart from potential residual cancer cells, I speculated that Miaoxuan's relapse was associated with chemotherapy. Miaoxuan received more than 20 sessions of chemotherapy involving more than 10 drugs. On the one hand, cholangiocarcinoma is naturally insensitive to chemotherapy. On the other hand, the possibility that cancer cells make desperate moves after being cornered by chemotherapy cannot be ruled out. Cancer cells come from the mutation of normal cells. Chemotherapy drugs induce cancer cells to mutate again, forming "outliers" and making cancer cells more malignant.

Miaoxuan asked: "Prof. Xu, is there any treatment for me?"

I answered: "No chemotherapy."

"Without using any drug?" Miaoxuan looked at me with her eyes wide open. I could not tell whether she was desperate or shocked.

"Yes. No chemotherapy is treatment itself." For a doctor, it takes both wisdom and courage to make treatment decisions. But the decision for Miaoxuan seemed a little unconventional. After finishing, I regretted what I said for a short moment. But then, my rational mind came back to me – I was telling the truth.

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One day a year ago, the image of Miaoxuan flashed in my memory. Then, I sent her a message, asking about her health status instead of her disease.

I received a reply from her very soon: "I'm very well, without disease or pain. Thank you."

"Did you receive any treatment?"

"Didn't you tell me not to take chemotherapy some time ago? I listened. Afterwards, the tumor has not recurred." She was very happy.

Half a year ago, I went to Shenzhen for a lecture and gave a speech titled China's Way of Cancer Control and Hydrogen Molecular Medicine. As the lecture just came to an end, a lady came to the stage and took me by the hand. Wasn't she Miaoxuan? Standing in front of me, Miaoxuan was wearing a red top. I saw her full of joy and with ruddy complexion. She said: "Prof. Xu, you have sent us a gift. I definitely will try the hydrogen that you just spoke about."

This time, I met Miaoxuan in Wuyi Mountain. Looking excited, she told me that her sleep, appetite and physical strength became good after hydrogen inhalation. But she was worried as well. After all, her disease had not recurred for only 4 years. Cholangiocarcinoma is dreadful. Miaoxuan said: "What a struggle for me! In Shenzhen, I knew several patients with the same liver cancer as me. They passed away in less than 2 years." Then, she asked me whether I knew about any patient whose cholangiocellular liver cancer was successfully controlled through hydrogen inhalation.

I told her that I was going to visit Mr. Chen, who lived in Yunnan. He suffered from mixed cholangiocellular and hepatocellular carcinoma. He had signs of recurrence after surgery and had been inhaling hydrogen for 4 months. He called me and said that his conditions improved.

Mr. Chen's Misfortune and Good Fortune

Both Mr. Chen and his wife are teachers and have been working in a middle school in a city in Yunnan for many years. In September 2017, Mrs. Chen was diagnosed with breast cancer and received surgical resection. In November the same year, Mr. Chen was found to have a 5cm carcinoma in the liver. A month later, he received hepatic lobectomy. Postoperative pathological examination of the surgically removed specimens indicated mixed liver cancer of poor differentiation. He received six sessions of chemotherapy. Mr. Chen taught Chinese and had strong understanding of things. He knew from medical books that his liver cancer was not sensitive to chemotherapy and had a high incidence of recurrence.

Tumor markers are early indicators of recurrence. In May 2018, several tumor markers in Mr. Chen's blood began to rise. And CA19–9 level rose to 99 units, which was nearly three times higher than the upper limit of the normal range and exceeded the pre-operation level at 57 units. His alpha–fetoprotein doubled compared to the normal level. Mr. Chen started having tension in the mind and came to my outpatient clinic in Guangzhou. I asked him what his doctor in Yunnan said. He told me that the local doctor suggested continuous chemotherapy, which he refused. Mr. Chen said: "I have read relevant literatures. The conclusion is always that chemotherapy achieves tumor response but does not prolong lifetime."

I gave Mr. Chen an examination and did not find any space–occupying lesion in the liver or any indication for ablation. He wanted me to set a treatment plan for him no matter what. Mr. Chen had two sons. The older one was studying in the United States and was about to graduate with a post-doctoral degree. The younger son was studying at school and had not married yet. In the past few years, both Mr. and Mrs. Chen were sick with cancer and had spent all their savings on treatment. Even so, they were in debt with hundreds of thousands of RMB yuan.

While telling me his story, Mr. Chen almost cried. His wife was standing next to him, crying. What could I do? Mr. Chen had already received all the conventional treatments for liver cancer.

I suggested that he have injections of interferon. Then I added: "One more thing, there is a rehabilitative treatment. I cannot say for sure that it will be effective. But you can try it." I asked him to try hydrogen inhalation in my health studio.

After that, Mr. and Mrs. Chen rented an apartment in the neighborhood nearby and came to the Xu Kecheng Care for Health Studio every day for hydrogen inhalation. A month later, they went back home in Yunnan. Mr. Chen rented a nebulizer so that he could inhale hydrogen at home. Two months later, he sent a WeChat message to me, saying that his CA19–9 level dropped to 38 units, and that there was no recurrence in the liver according to latest CT scan.

Perhaps out of sympathy, I, once as a patient with similar disease, was excited at Mr. Chen's successful treatment. Moreover, I wanted to learn from Mr. Chen's experience. I knew that Mr. Chen's therapeutic outcome was hard to achieve in general cases. I knew too much about the nature of cholangiocellular liver cancer. I immediately sent Mr. Chen a WeChat message and asked him what treatments he had received. He said that he followed my advice and took interferon injection twice a week. But he discontinued the injection after a few weeks due to serious side effects. In his view, the main treatment he had was hydrogen inhalation for at least 6 hours every day. Moreover, he was careful about his dietary structure and adopted a primarily vegetarian diet, avoiding red meat, fried food and dietary bias to ensure energy balance in the body.

I have heard a speech by an expert of Chinese traditional and western medicine. The speech was about Qi promoting blood circulation. According to the expert, Qi is vitally important. Smooth Qi brings about unobstructed meridians and blood circulation. Once the three are smooth, the body is dominated by healthy vital energy, and its pathogenic factors subside. In traditional Chinese, the character 氣 (Qi) comprises 气 (gas) and 米 (rice). With "gas" and "rice" (nutrition), the body is well-grounded and blessed with good order of the five internal organs (the heart, liver, spleen, lung and kidney) as well as unhindered operation of the six hollow organs (the gallbladder, stomach, large intestine, small intestine, bladder and Sanjiao).

In the sense above, Mr. Chen's intake of hydrogen ("gas") and nutrition ("rice") put together represent exactly the Qi (氣) much needed.

This is only my imagination from philosophical and psychological aspects. Whether the idea is scientific remains to be studied. Practice is the sole criterion for testing truth. I decided to visit Mr. Chen in his home in Yunnan and see for myself how he actually lived a life. It is not the hospital but the patient's home where doctors should evaluate whether the patient is really cured. I thought that if I visited Mr. Chen, I could at least give him a few words of comfort. I already bought plane tickets to Yunnan, but my trip was postponed due to my schedules of meetings in a row in Guangzhou. Recently, an editor from the publishing house called me and asked for the final version of this article, which was the only missing part of my to-be-launched new book Hydrogen Cancer Control—Theory and Practice.

Coincidence can be remarkable sometimes. That day while on a business trip in Xi'an, I planned to fly to Yunnan after the trip and finish my patient visit before the New Year's Day. The next day, my secretary Rita told me: "There is something I need to tell you. It's not good news."

I was a little startled. Rita said in a low voice: "Maybe it's not serious. Please don't worry too much." Having been my secretary for 10 years old, Rita often knows what I think and always

shares my cares and burdens. Then, she said: "Mr. Chen from Yunnan will fly to Guangzhou to meet you tomorrow. He had blood from the mouth."

"Is the blood bright red or black? Did he vomit or cough out the blood? How much blood?" Feeling really worried, I asked several questions in a row. Mr. Chen had an "old liver". Considering that he had blood coming out of the mouth and was eager to come a long distance to Guangzhou to see me, I was sure that something bad happened to him. Perhaps his liver cancer recurred. The next day after my return to Guangzhou from the business trip, I rushed to the ward. Seeing me, Mr. Chen cracked a big smile and explained that it was a false alarm: gum bleeding. He found himself with a lot of bright red blood in the mouth one morning after getting up. In fact, he had gum bleeding for many years, but never took it seriously. Because of liver cancer, the blood from gum bleeding became a big alarm for him.

Mr. Chen received complete blood biochemical tests, tumor marker detection, CT scan and MRI, none of which showed any evidence of cancer recurrence.

I told Mr. Chen to relax, let go, live in the present and live in the present, and embrace mortality. There is a couplet in Huangzhou Lingyin Temple (a Buddhist temple) that says: "Half of our life is in our own hands, while the other half is up to nature." I asked Mr. Chen to learn from Miaoxuan, who inhaled hydrogen at home every night, and danced and participated in the daytime voluntary activities in a rehabilitation association in the daytime. We are all fellow travelers in life. Our life varies because of different mentality. Happiness befalls those who catch glimpses of the magic in the mundane everyday life and remain true to themselves. Everyone experiences ups and downs. When we fall, we get up, dust off and continue our journey.

> *Picture 3. On a special visit to Mr. Chen in his home in Yuxi, Yunnan, the author is reliving the past moments with Mr. Chen in great warmth and pleasure.*

Afterwards, Mr. Chen seemed like a different person. He and his wife visited relatives and friends, and shared memories happily with those who had similar experience.

On the early morning of 2019 New Year's Day, I turned on my mobile phone, and the first WeChat message popped out. It was from Mr. Chen. He quoted from an ancient Chinese poem: "Where the hills and streams end and there seems no way out, amidst shading willows and blooming flowers another village appears." He said that he looked forward to meeting me in Yunnan and that I definitely should visit the beautiful city of Yuxi there.

The city where Mr. Chen lives is a scenic spot with four seasons like spring and with clear periods of dry and wet days. It is the hometown of great musician Nie Er. I feel as if Nie Er's composition trailbreaker is lingering in my ears: "I am not afraid of layers after layers of heavy mountains. Fossils of thousands of years form a mountain peak." In spite of being a tremendous danger, cholangiocarcinoma can be overcome by combined use of conventional treatments and supplementary measures, as illustrated by the following in Sun Tzu's Art of War: "In all fighting, direct methods may be used for engaging in battles, but indirect methods will be needed in order to secure victory." Then, instead of spending the rest of life mourning in pain and sadness, we will eventually emerge as "shiny pearls born out of suffering clams", as depicted by the beautiful writer Xu Niang at the beginning of this article.

(In March 2019, Mr. Chen came to Guangzhou again for examination due to liver pain. The results indicated no problem with the liver and further decline of tumor markers. After learning

about the results, Mr. Chen laughed and said that he was too sensitive to any slightest sign and tended to take it seriously.)

— Xu Kecheng

> **Comments from Tang Zhaoyou**
> I enjoy reading Prof. Xu's articles for their fluent style, well-chosen words, and easy-to-understand in-depth ideas of philosophy. Some people might say that the cases in the article are accidental events without evidence-based medicine proof. However, there is often the inevitable in coincidences. Major discoveries pass us by usually as a result of our ignorance of coincidences. "Birth of a Pearl from a Sick Clam" is such a pertinent metaphor. In the past 200 years, the anti-cancer war has been aimed at eliminating tumors and preferably at annihilating each and every tumor. Few people think about the possibility to live peacefully with cancer. It is worth noting that the article implied methods for achieving "coexistence with tumor": keeping an open mind, enjoying life, "no use of chemotherapy anymore", maintaining a balanced diet, and inhaling hydrogen. These seemingly small measures are easily neglected because of cultural influence. I recently published the book The West Learning from Chinese, Creating New Chinese Medicine. "East" in the title of the book refers to not only traditional Chinese concepts but more to the essence of Chinese civilization. Laozi, an ancient Chinese thinker, believed that the soft overcomes the hard. If small measures eventually tame the tumor, isn't it a good example of Laozi's idea?

Hydrogen and Colorectal Cancer: Enormous Delight

In the past few months, I have been looking forward to visiting Jinhua, Zhejiang. It is not that I have never been to the city that is famous for Jinhua ham. But I feel sorry for Ms. Yang and her son Mr. Shen, both of whom live in Jinhua.

Ms. Yang is 84 years old and has rectal cancer. I feel apologetic towards her out of an introspection myself, not because of any medical negligence or complaint from her or her son.

Ms. Yang Being Hospitalized

On February 10, 2018, the director of the Ward 5 came to me, saying that a patient came from Zhejiang out of admiration for me and was admitted to our hospital. The patient insisted on seeing me and hoped me to set a treatment plan for her.

I went through the patient's medical records as follows:

> *The patient had bloody mucous stool with no obvious predisposing cause two years ago. She had poor stool output, and the stool was thinned than before. The symptoms were not accompanied by pain or abdominal distension. After being admitted to a central hospital in Jinhua, Zhejiang, the patient was given finger examination of the anus. A cauliflower-like mass of 2cm to 6cm was detected from the anal verge. The patient refused surgery and took oral Chinese medicine. Her hemafecia was not alleviated. One year ago, the patient had paroxysmal dull pain in the appendix, and took herbal medicine by herself. No positive effect was achieved. In May 2016, the patient passed a large amount of blood (around 500mL) in the stool. After that, she had similar bloody stool three times successively, but did not receive any special treatment. In September 2016, a mass appeared in the perianal area, and the pain in the sacral tail was aggravated. The patient was admitted to a hospital in Hangzhou and took a CT scan. The findings showed a space-occupying lesion in the upper-right mediastinum; an infectious lesion in the middle upper lobe of the right lung; rectal carcinoma; and small lymph nodes next to the aorta abdominalis, indicating rectal*

> *cancer with mediastinal metastasis and lung infection. The patient continued to take Chinese medicine for more than 5 months, but her conditions did not improve considerably. On the second day after being admitted our hospital, Ms. Yang had a full-abdomen CT plain scan + contrast-enhanced scan. The results were as follows: (1) The distal rectum and anal area exhibit thickened walls and formation of a soft tissue mass. (2) Bilateral musculus levator ani and the posterior wall of the vagina are adhered without clear borders. (3) Multiple small and slightly enlarged lymph nodes are detected beside bilateral iliac vessels and in inguinal regions; (4) The lower-right lung shows a small shadow, which is considered to be an inflammatory lesion. (5) Signs of multiple cysts are noted in the liver.*
> *Colonoscopy findings: A 1.0 × 0.6cm wide-stem polyp is found 10cm from the anus and displays congestion and edema on the surface. The mucosa of the rectal section within 4cm from the anus shows congestion, edema, erosion, and cauliflower-like bulge, involving 3/5 of the perianal area. Biopsy indicated moderately differentiated adenocarcinoma.*

> *Picture 1. CT scan showed a space-occupying lesion in the distal rectum and blocking the anus.*

> *Picture 2. Enteroscopy showed the tumor incarcerated at the entry of the anus.*

Dilemma

Dr. Shi, Ms. Yang's attending doctor, has more than 10 years of clinical experience in cancer treatment and has always been able to overcome challenges. However, in the case of Ms. Yang, Dr. Shi found herself in a dilemma: Ms. Yang had low rectal cancer, for which surgery was challenging due to her advanced age and poor overall conditions. In addition, the tumor had invaded the structure around the rectum and pelvic cavity. Chemotherapy was not possible, because Ms. Yang's family rejected it. The greatest trouble was that the tumor grew outwards, fell out of the anus, and was incarcerated at the entry of the anus, causing severe pain, especially during defecation.

I went to Room 524, Ms. Yang's ward room. After entering the room, I saw a gray-haired old lady lying on the bed near the window and constant groaning. Standing next to the bed were the old lady's son and daughter. Seeing me, the son took me by the hand and said, "Finally, I meet you in person. Please save my mother. She is in great pain." He stopped, with tears in the eyes, and then said: "As long as she does not feel pain, we will be happy."

What special treatments could I offer her?

To reduce tumor load and remove parts of the tumor, doctors put Ms. Yang on intravenous anesthesia and performed cryoablation on the rectal & anal canal tumor for Ms. Yang on February 14, 2017.

The next day, I went to visit Ms. Yang, and found that her pain was not eased. Cryosurgery helped ablate only a part of the tumor exposed at the entry of the anus. I said nothing but felt sorry for her.

On February 16, I went to visit Ms. Yang again in the ward. Mr. Shen, Ms. Yang's son, showed grave expressions and handed me a discharge bill, saying that they planned to leave the hospital and return to their home in Zhejiang.

Three Months of Telephone Communication

In the following three months, Mr. Shen gave me a call every few days, but I did not have much to say. I was not able to respond to his requests out of professional considerations as a doctor. Our typical conversations were as follows:

Mr. Shen: "Prof. Xu, the tumor in my mother's anus is growing again. What should I do?"

Xu: "Put a stent in the anus. She can receive surgery in a hospital in Shanghai or Hangzhou."

Mr. Shen: "Prof. Xu, my mother is in such big pain that she groans every day and cannot fall asleep, what should I do?"

Xu: "She can use morphine."

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By later May, Mr. Shen sounded more and more anxious on the phone: "Prof. Xu, my mother's pain cannot be stopped with morphine anymore. You must help me find a way..."

In fact, it was heartbreaking for me to hear that. On the phone with Mr. Shen, I kept silent for a few minutes and then said, "Ask your mother to inhale hydrogen for at least 4 hours every day. Keep doing it. Maybe..."

"Gem" from the Sky

On June 30, 2018, I received the following WeChat message from Mr. Qin:

> *The 84-year-old mother (patient with intestinal cancer) of Mr. Shen from Jinhua, Zhejiang inhaled hydrogen for no less than 4 hours a day and continued it for over a month. Before the treatment, the patient was in so much pain that she could not defecate and could only lie in bed. Recently, her pain has been significantly eased and defecation is back to normal. Yesterday, she went outside to buy food on the street…*

Later, Mr. Qin sent me two short videos, showing Ms. Yang inhaling hydrogen and playing Tai Chi outdoors.

As a friend of Mr. Shen, Mr. Qin rented a hydrogen-oxygen nebulizer for Ms. Yang to inhale hydrogen. For me, the news from Mr. Qin was like a gem from the sky. I sent Mr. Shen a WeChat message, telling him that I was happy to hear the good news and hoped his mother to continue with hydrogen inhalation. I also said that I would take time to visit his mother.

I got reply from Mr. Shen immediately. He said that he felt lucky and wished me to visit and encourage his mother.

Mr. Shen had very different mood in WeChat communication with me this time compared with how he sounded in our previous phone calls. As a doctor, I have always regarded the recovery of patients as the biggest reward for myself. The medical value of hydrogen needs further research. Maybe hydrogen will offer us a new scientific way of treatment. I sincerely thank Mr. Shen for his courage to follow my advice. I am also grateful to Mr. Shen's mother, for her "good luck" has given me the confidence to study hydrogen cancer control.

I would definitely visit Ms. Yang to thank her in person.

Exclusive Interview in Jinhua

On August 11, 2018, my secretary Rita and I boarded a high-speed train heading to the north. Soft music was played in the train carriage. The music, together with the regular sounds of

train wheels pouncing on the rails outside the carriage, gave passengers unlimited pleasure and imagination. Life is like a train that moves continuously forward. Every stage of life is like a train station platform. As a doctor, I know it's important to keep fighting disease together with patients in every stage of the treatment journey.

Six hours later as twilight scattered quietly like a gray big net, the train stopped at Jinhua Railway Station. At that time, it was 8 o'clock in the evening. Lights were shining on the street like colorful fireworks splashing into the world. A string of bright automotive lights was glittering on the street like a flashy long river running nonstop.

Mr. Shen was waiting for us at the exit. When I got in the car, I could not wait to ask about his mother. Mr. Shen said: "The old lady has been looking forward to seeing you for a long time. Knowing that you are coming today, she is so happy that she got better by one third."

Mr. Qin, who arrived from Shanghai one hour earlier, greeted us at the hotel. Eventually, the patient's family, Mr. Qin (helper of the patient's family), and us doctors met each other and were very happy. We congratulated the patient on her therapeutic results. The scene felt like a storm troop had just won a war.

> *Picture 3. Ms. Yang is inhaling hydrogen (picture taken at the beginning of her hydrogen inhalation).*

> *Picture 4. Ms. Yang was walking and Playing Tai Chi*

Shortly after breakfast the next morning, Mr. Shen came to the hotel to pick us up. Jinhua, situated in the central part of Zhejiang, has a history of 2,200 years. It was given the name Jinhua because of its location in the place where the Jinxing (Venus) and the Wunu (four stars in the constellation Aquarius) competed for splendor. Ancient and modern buildings stood alternately on both sides of the street and reinforced the charm of each other. The view reminded me of a poem by the famous poet Chen Fengchun describing Jinhua: "Pavilions lie alongside the river. To the north, one sees coralline flowers and jasper plants of the fairyland".

After walking more than 200 meters, we arrived at Mr. Shen's house, an old building with several floors. According to Mr. Qin, Mr. Shen is a very dutiful son. He lived with his own family in a luxury residential area of the city. But he moved here to take care of his elderly mother 2 years ago.

We went to the third floor. The door was already open. We went straight into the room. Ms. Yang was lying on the couch and inhaling hydrogen. Seeing us, she sat up and greeted us warmly. Mr. Shen's wife was a native of Jiangsu, the same province as Rita and me. Naturally, we felt close to each other. Then, Mrs. Shen went to make tea for us.

> *Picture 5. The author's WeChat message to Mr. Shen*

> *Picture 6. Mr. Shen's reply to the author*

I looked at Ms. Yang carefully. Her complexion was not good but was still much better than the time when she stayed in our hospital. I gave her a simple physical examination, without finding any superficial lymph nodes and swelling or tenderness in the abdomen. Rita examined Ms. Yang's anus and found that the tumor protruding from the anus was gone. When I was about to suggest that Ms. Yang receive a CT scan and colonoscopy in a hospital, Mr. Shen

probably read my thoughts and said: "The old lady can eat and sleep well now. So far, she has gained 2.5kg. We don't want her to go through the trouble of another medical examination."

Mr. Shen made a point. The fundamental purposes of cancer treatment are to extend patients' lifetime and help patients live with good quality. An 84-year-old patient like Ms. Yang is living with cancer. Why do we still care too much about the size of the tumor?

> *Picture 7. Ms. Yang and the author (first from the right) were talking, with Mr. Shen in the middle back and Mr. Qin on the left.*

Ms. Yang stood in the living room and chatted with us. She spoke highly of her son and daughter-in-law for taking good care of her. She also told us the food she ate every day. She thanked us and wanted to invite us to lunch. I said: "No need to say thanks. You are now recovering, and that is the best thanks to us."

Half a year ago, Ms. Yang discharged herself from our hospital, about which I felt bad. In the following 3 months, I kept communicating with her son Mr. Shen over the phone. I was helpless. One and a half month ago, I received videos of Ms. Yang walking in the garden by herself. Eventually, I visited Ms. Yang at her home, and she wanted to invite us to lunch. The whole story felt like what is depicted in the poem: "By no means can the hazy moon recognize the changes in things and people. I see people leaving as if in a dream." Life is too short to waste. Thinking about the experience with Ms. Yang and her family, a sense of happiness welled up in my heart.

People regard the profession of doctor as a lofty one. At that moment at Ms. Yang's home, I started to appreciate my own profession as a doctor. It has allowed me to enjoy impressive joy from the meaning of life.

Mr. Shen insisted that we stay for lunch, which we politely declined. We planned to go to Lanxi, a town more than 40 kilometers away from Jinhua. There was an old lady expecting us. She had a huge tumor in the mediastinum and also inhaled hydrogen. Her daughter called me in the morning excitedly and said that her mother had been inhaling hydrogen for two weeks and her conditions showed improvement…

Final Thoughts

Ms. Yang still has rectal cancer. At the beginning, she had a tumor incarcerated at the entry of the anus. The tumor caused severe pain that could not be eased even by morphine. But now, Ms. Yang can eat, sleep and walk well. The change in her conditions is almost a miracle. Is this a result of spontaneous relief of the tumor, or of hydrogen inhalation?

There are very few cases in which cancer is cured by itself. According to estimates, self-cure of cancer happens to around one patient out of 100,000 and primarily to children with tumors. Ms. Yang's rectal cancer had invaded the rectum and pelvic cavity. It was not in early stage. Moreover, Ms. Yang had not received chemotherapy or radiotherapy. Her disease was alleviated in one month after hydrogen inhalation. It is almost certain that hydrogen inhalation played an effective role in her case.

Currently, our follow-up observation of cancer patients with hydrogen inhalation shows that the effects of hydrogen are brought out one month after hydrogen inhalation. Like anti-cancer drugs, hydrogen appears to be dose-dependent to deliver effects. In particular, for tumors at the distal ends of tissues, it takes longer time for hydrogen to exert positive effects. That is

because hydrogen is inhaled from the nose and travels to the lung before entering blood circulation and tissues, which is a long journey. In other cases, we have discovered that when the hydrogen flow outlet is directed at the tumor wound, the tumor and its wound shrink after a few days of hydrogen inhalation.

How to administer hydrogen? What is the effective dose of hydrogen? What is the principle of hydrogen cancer control? All these questions require further research. Sometimes, dreams are seen by others as fanciful illusions. However, dreams are like bamboo roots that are buried underground and invisible to people. Nevertheless, bamboo roots never stop exploring, but keep growing and popping out new shoots.

— Xu Kecheng , Lu Tianyu

> **Comments from Tang Zhaoyou**
> Prof. Xu is nearly 80 years old and suffered from cancer himself. His care and enthusiasm for patients are rare and commendable. A few days ago, I read in Reference News a report titled Highly Costly Life-saving Drugs Become the Unspeakable Pain of American society. According to the report, a genetic drug costs 470,000USD per administration, and a targeted drug costs 150,000USD (about 1 million yuan) per year. In light of the national conditions, I think it is necessary to promote the use of high-tech, sophisticated and new therapies together with fast, good and affordable ones. Currently, patients tend to look down upon cheap drugs and treatments. Furthermore, practice is the only criterion for testing truth. As long as a treatment is clinically effective, it deserves great attention. The therapeutic effect on Ms. Yang in the article above is evident. She is living with tumors that have shrunk and living a life of markedly improved quality. Modern medicine tends to require tumor-free survival during treatment, which is actually difficult to achieve. To this end, whether cancer can be overcome depends not only on the means of treatment, but also on renewed thinking.

Hydrogen and Ovarian Cancer: Prevention Before Disease Onset

Treatment Before Disease Onset

The best time for cancer treatment or intervention is when cancer cells are not yet formed and just formed. As said in an ancient book (History of the Later Han Dynasty · A Biography of Ding Hong), "Check erroneous ideas at the outset. Be precautious beforehand. Caution against small matters. Destroy evils before they become menacing. A stitch in time saves nine."

Cancer cells are derived from normal cells in the body. Under the long-term influence of various internal and external factors, normal cells mutate. And mutations gradually accumulate, eventually forming cancer cells. This is a long and gradual multi-stage process in which dysplasia occurs and then precancerous lesions are formed. Afterwards, the lesions develop into carcinomas in situ, early-stage carcinoma, infiltrating carcinomas, and eventually metastasis to lymph nodes and distant tissues and organs successively. The entire process usually takes 10, 20 or 30 years, or even longer.

Prevention of recurrence and metastasis matters the most for patients who have cancer and have received effective treatments. Recurrent cancer might be derived from residual cancer cells, circulating cancer cells, pre-existing dormant cancer cells (especially cancer stem cells),

or newly grown cancer cells. The process of cancer recurrence depends on different sources and can be as short as several months or as long as several years or longer.

As early as 2,000 years ago, Inner Canon of the Yellow Emperor, an ancient Chinese medical classic, proposed that "The supreme doctor cures the disease to come; the good doctor cures disease that is about to break out; and the least able tries to cure fully manifested disease." Inhibiting occurrence of abnormal hyperplasia and precancerous lesions; preventing formation of primary tumors or recurrence of treated cancer, and suppressing residual or dormant cancer cells are measures for treating the disease to come.

> *Picture 1. Occurrence and development of cancer* (stages: Normal tissues → Pre-cancer lesion → Early-stage cancer → Terminal cancer → Cancer metastasis)

> *Picture 2. Source and process of cancer recurrence* (Surgical stress / Anesthesia acting on the Tumor; Surgical resection leading to Residual cancer cells, Circulating cancer cells and Pre-existing dormant cancer cells (cancer stem cells); Cancer cell "awakening" → Lymph node metastasis and Distant metastasis → Cancer progression)

A Lady with "Disease to Come"

Over a month ago, Prof. L called me and said: "My child is in trouble." He asked me for help. Prof. L is a well-known professor at the affiliated hospital of a prestigious university in Guangzhou. He is a friend that I have known for many years. I will definitely help him.

A few days later, Prof. L came to our hospital. The child he mentioned was Ms. Y, his daughter-in-law. Ms. Y looked as if there were mountains of worries and oceans of depression.

With left ovarian cancer, Ms. Y received an operation that removed the uterus and uterine adnexa in December 2017. Postoperative diagnosis was Stage Ia ovarian adenocarcinoma. In the past 3 months, she had three tumor marker tests successively, and all the results indicated tumor marker increase. According to the latest examination findings, her blood CEA and CA19-9 levels were 13.7 μg/L and 47.4 units /L respectively, which were 3 times and 27% higher than the upper limits of the normal ranges respectively.

Certain substances generated and released by tumor cells often exist in the host's body fluids in the form of metabolites such as antigens and enzymes. These tumor markers can be measured to diagnose tumors or understand tumor progression status. CEA is short for carcino-embryonic antigen, and CA19-9 refers to carbohydrate antigen 19-9, both of which help detect a variety of cancers. For people with cancer, an increase in these indicators suggests recurrence or imminent recurrence.

Ms. Y's tumor markers increased, which might be a result of normal variation. But for cancer patients, tumor marker rise should be regarded as a sign indicating tumor recurrence, presence of residual cancer cells (especially pre-existing dormant cancer cells) in the body, or growth of another type of cancer cells. Before the operation, Ms. Y's CEA and CA19-9 were at normal levels. Rise in the two markers is generally found in digestive system cancers such as intestinal and pancreatic cancers, and also in ovarian cancer. Ms. Y had examinations including PET-CT scan and gastrointestinal endoscopy, none of which showed any sign of tumor. However, tumor markers can be at abnormal levels in the first half year or even years before the tumor is clinically detected. Can they probably be indicators of cancer about to occur? This is undoubtedly a time bomb in the body.

"The child is very miserable and mainly suffers mentally." Prof. L forced a smile and looked helpless. "She doesn't want to have chemotherapy. It is such a dilemma."

I said: "Let's wait and do follow-up." I was also helpless. I knew that such "standard" answer would certainly not satisfy them. After all, Prof. L could have treated his daughter-in-law in the well-known hospital where he himself worked.

As expected, Prof. L was not happy with my "standard" reply, saying: "You are my old friend. Today, we came here to get the truth from you." He must have read my book Truth To Cancer Patients.

"Prof. Xu, look at yourself. You are in such good spirit. You must have some tricks. Your Daily Talk About Rehabilitation is seen by many as the Bible." said Ms. Y full of expectations. "Can I try hydrogen that you talked about?"

What she said reminded me that hydrogen might be helpful for her case. At least, it would do no harm.

I have been taking hydrogen for two years. I started with drinking hydrogen-rich water. Later, I began hydrogen inhalation. Now, I seem to be dependent on hydrogen.

Prof. L said: "The child wants to follow you to fight cancer." He smiled and said that he had read my other book Follow Me to Fight Against Cancer. I said: "Strictly speaking, we don't fight cancer but control cancer. Cancer control is not only aimed at eliminating cancer cells but is more focused on transforming cancer cells. To keep cancer cells from growing or running around to metastasize, the first measure is to transform cancer microenvironment."

Many studies have shown that hydrogen molecules can transform cancer microenvironment. If chemotherapy is the "right path" to treat Ms. Y's "disease to come", hydrogen can be understood at the "tortuous path" or "side path". We can always "overtake on a bend" or "change lanes to overtake".

I suggested to Ms. Y that she inhale hydrogen and that she can do it while watching TV or playing with the mobile phone, just for treating the "disease to come".

I recommended hydrogen inhalation, because inhalation delivers fast effects compared to other methods of administration. It has been reported that after people under 60kg inhale 2% hydrogen, hydrogen saturation capacity within 24 hours is 104 times or more than that from oral intake of saturated hydrogen-rich water. Within a certain range, the concentration of hydrogen in the blood is dose-dependent. That is, the more hydrogen inhaled, the higher the concentration in the blood. And more hydrogen enters tissues in the body.

Studies have revealed that the biological effects of hydrogen molecules are positively correlated with hydrogen concentration in tissues. Hydrogen volume reaches the highest level in the plasma approximately 30 minutes after inhalation; and returns to the baseline level around 60 minutes after the inhalation is stopped. Therefore, the concentration of hydrogen inhaled should be high. In China, a hydrogen-oxygen nebulizer that rapidly produces 66.7% H_2 and 33.3% O_2 mixture has been invented and been approved by the country as an innovative product. The machine is very safe and can be used continuously. I asked Ms. Y to inhale hydrogen for 2 to 4 hours a day for the following reason: According to other patients' experiences, the inhibitory effects of hydrogen on cancer seem to have a critical point, which is 80 to 120 hours after cumulative hydrogen inhalation.

Initial Disappearance of the "Disease to Come"

As a cancer patient myself, I have always regarded cancer patients as companions who fight cancer in the same trench. In the over half a month after Ms. Y and Prof. L left, their worries and helpless expressions flashed in my mind from time to time. I found myself often worried about whether hydrogen was effective for Ms. Y.

At around 9pm on May 25, 2018 when I just finished a meeting in Beijing, my mobile phone buzzed. It was a WeChat message from Ms. Y: "I have been inhaling hydrogen for half a month. I feel very good. Yesterday's test results showed that everything is normal. I didn't expect to see such remarkable effects... Thank you very much." Then, I received three photos of the test sheets, showing: CEA 3.3 μg/L, CA19-9 18.8 units/L. The results were completely normal.

> *Picture 3. WeChat message Ms Y*

That night, I was not able to fall asleep, which was not common for me. According to the schedule, I was going to teach a class of doctoral students at Peking University the next morning. As preparation for the class, I gathered several experts and friends in Beijing in the afternoon, and we discussed about what is innovative thinking. Everyone present mentioned hydrogen. We used an example from a paper published in the famous journal Nature Medicine in 2007. The paper reported the surprising therapeutic effects of molecular hydrogen on rat models of brain infarction. Then, we reflected on the following together: H_2 is an element in the upper-left corner of the periodic table of elements and is the smallest of all the elements. For a long time, hydrogen had been used only for hydrogen balls. Later, only Japanese professor Shigeo Ohta studied hydrogen and discovered its extensive biological significance. His discovery attracted worldwide attention. Isn't that innovation?

I was very happy for Ms. Y. If hydrogen therapy is an innovation, the therapeutic effect demonstrated in Ms. Y is the result of innovation.

Two days later, I sent Ms. Y a WeChat message, asking her to verify the effect of hydrogen. As a doctor, I always want to get the truest result. One month later, Ms. Y sent me her follow-up examination results: CEA 1.9 μg/L; CA19-9 10.3 units/L. Both indicators further decreased.

> *Picture 5. Changes in Ms. Y's blood tumor markers*

Date	CEA (μg/L)	CA19-9 (U/L)
2018.1.3	0.8	6.6
2018.3.20	14.2	49.9
2018.4.28	13.7	47.4
2018.5.22	3.3	18.8
2018.6.25	1.9	10.3

Ms. Y showed unexpected results from the treatment of "disease to come". Although she represents only an individual case, and further verification is needed, my interviews with patients with other types of cancers and literatures indicate that the treatment effect on Ms. Y is both unexpected and expected.

Myraculous Hydrogen

Hydrogen is indeed a gas with magical effects. Broadly speaking, hydrogen powers the fusion of the sun. From a narrow point of view, hydrogen molecules constitute almost a part of every molecule of the body: DNA, protein, sugar, and fat. Hydrogen in the human body mainly binds with carbon, oxygen and nitrogen. It is the key to the production of ATP in mitochondria. The present biological research on hydrogen is still in infancy stage. But rigorous scientific research tells us that hydrogen has the capacity to not only function at the cellular level but also to pass through the blood brain barrier and enter mitochondria before entering cell nuclei.

Hydrogen molecules inhibit cancer cells generally because of their antioxidant and anti-inflammatory effects. The most powerful and toxic oxidants in the body are •OH and $ONOO^-$, which act on nucleic acids, lipids and proteins; and cause DNA damage, lipid peroxidation and albuminous degeneration. Smoking, air pollution, chemicals, mental stress, and inflammation facilitate cell mutation by inducing these oxidants and eventually cause formation of cancer cells. H_2 is a specific scavenger of the two oxidants.

> *Picture 6. Formation and harms of free radicals (damaging DNA)*

Some in Japan stated that hydrogen is effective against 67 diseases. A U.S. scholar once wrote that hydrogen molecules can improve at least 170 poor health conditions and diseases. Another American expert said that hydrogen has mitigative effects on the top ten causes of death (except suicide and traffic accidents) in the United States.

Academician Zhong Nanshan, a distinguished respiratory expert, said that the application of hydrogen moves up the treatment of diseases and is of significance to etiological treatment. What he said is about treating the disease to come. Ms. Y is a real-world example of the magical effect.

Prof. Sun Xuejun, a well-known hydrogen researcher, believes that hydrogen has a wide range of application prospects. In his view, hydrogen research may cover all the diseases and symptoms for which hydrogen is potentially effective. He once said without exaggeration: "how can it be justifiable if an affordable, safe and effective treatment like hydrogen is not promoted to have worldwide influence?"

— *Xu Kecheng*

> **Comments from Tang Zhaoyou**
> I highly agree with Prof. Xu on the cancer control principles that he advocates: the ABC Principles. ABC stand for applicable (effective), brief (simple) and costless (cheap) respectively. In 2008, one of my doctoral students found that aspirin reduced the spread of residual cancer after radiotherapy. I suggest that relevant professors conduct clinical randomized controlled trials. Perhaps because the drug was too common, no one showed any interest. In 2015, A Cancer Journal for Clinicians, a medical journal with the highest impact factor, published a paper, stating that aspirin helps reduce the death rate of prostate cancer that has high risks of recurrence. Although everybody acknowledges that practice is the only criterion for testing truth, accidental events like hydrogen inhalation are highly likely to be ignored in today's general pursuit of high-tech, sophisticated and advanced treatments.

Hydrogen and Esophagus Cancer: Trying Hydrogen

"Old Friend" and Cancer Patient Friend

On the evening of November 17, 2018, I arrived in Beijing for a meeting. In the early morning the next day, I thought about visiting Wang and wondered how he was.

In the morning, I was visiting exhibition booths one by one in the huge exhibition hall of the International Conference Center in Beijing. Suddenly, someone patted me on the shoulder. I turned and saw Wang. A friend is like a soul-stirring song. I thought of him in the morning, and then he showed up in front of me. Seeing each other, Wang and I shook hands, hugged and exchanged greetings. Wang said: "Old friend, I miss you."

"Everything good?" I asked about his health.

"What do you think?" He patted me heavily on the back and held my hand tightly. "I'm very well. I have meetings, play balls and inhale hydrogen every day."

Wang was glowing with health and radiating vitality. With gray hair, he had ruddy and slightly dark cheeks as well as eyes full of wisdom. Then, we had a small arm-wrestling contest, during which I felt his firm skin and strong muscles. I joked, "You are so strong, congratulations, old friend."

In fact, I came to know this "old friend" only half a year ago. Some people say that friends are destined to meet and that it has little to do with time. I became friends with Wang, mainly because we were "cancer patient friends" who battled against cancer in the same trench. It felt like we covered for each other and fought together in a short-lived battle of life and death. We are "old friends" also because we use one protective weapon: hydrogen. My "cancer age" is much greater than Wang. But he has firm faith in the effects of hydrogen and has inhaled hydrogen for a longer time than me.

> *Picture 1. The author and Wang full of energy (left)*

A friend is like a book through which we open the whole world. It is Wang who urged me to delve into hydrogen control cancer with more confidence.

> *Picture 2. Medical reports before and after hydrogen inhalation*
> A-Endoscopy report after neoadjuvant chemotherapy + before hydrogen inhalation;
> B-Endoscopy report after neoadjuvant chemotherapy + after hydrogen inhalation;
> C-Ultrasonic endoscope report after neoadjuvant chemotherapy + before hydrogen inhalation
> D-Pathology of the esophageal section surgically resected

At the end of March 2018, a friend told me that there was a rehabilitation master in Beijing. This master was a great expert in cancer rehabilitation and was fascinated about hydrogen therapy. One night in a hotel in Guangzhou, my friend put me through to Wang. When the phone was connected, I heard a loud and cheerful voice on the other side of the phone. After short greetings, Wang said: "Hydrogen cancer control has a scientific basis. I have personal experience in that. You must study it properly and try to make achievements for our country."

Although I never met him, I guessed from the way he spoke that he might be a researcher, an expert, a cancer patient, and a long-term leader. From the volume and vigor of his speech, I knew that he must be a hydrogen beneficiary that had survived cancer well.

Later, I learned that Wang was indeed a high-ranking leader: the director of the a rehabilitation research center. Over a year ago, he received surgery for esophagus cancer.

Trying Hydrogen Himself

Two weeks after the phone call, I went to Beijing for a meeting. Shortly after checking in at the State Council Second Guesthouse, I heard a cheerful voice and laughter from the corridor. I quickly walked to the door and saw a man with medium height and figure. It was Wang, who was accompanied by a friend. He came up to me and held my hands. We walked into my hotel room and sat down on the couch. Wang took out a large pile of inpatient medical records and CT films from his handbag, and said, "You are a digestive disease expert. Please take a look. Is surgery worth it for me?"

I started reading the medical records. In February 2017, Wang did not feel well during eating and went for gastroscopy in a hospital in Beijing. A mass was found on the posterior wall of the esophagus 31cm to 35cm from the incisors. The mass exhibited errhysis and brittle texture; and was prone to bleeding. Endoscopic ultrasonography showed an ulcerative mass in the 31-35cm segment of the esophagus. The lesion was mainly located in the muscularis propria of the esophageal wall and had penetrated the outer membrane of the esophageal wall. CT scan indicated a mass in the lower part of the esophagus and potential lymph node metastasis in the left part of the stomach. Biopsy of the esophageal mass suggested squamous cell carcinoma. On March 12, 2017, Wang was admitted to a hospital for neoadjuvant chemotherapy using paclitaxel liposome and cisplatin. He had two cycles of chemotherapy, with 21 days in one cycle. At the same time, Wang inhaled hydrogen by himself (using a hydrogen-oxygen nebulizer) for at least 3 hours a day. His CT scan on May 11, 2017 showed no obvious space-occupying lesion or stenosis in the esophagus; and no apparent enlarged lymph node in the fixed gaps of the mediastinum.

Wang said: "I was very happy at that time. On the one hand, eating was no longer a problem for me. On the other hand, no mass was found on CT images. At least my 'large cancer' turned into 'small cancer'." Wang asked to continue with hydrogen inhalation and received another 2 to 4 cycles of chemotherapy.

However, Wang's family and supervisor did not agree with him. As mentioned above, Wang was a high-ranking leader at a senior level. His body and health were not entirely his own. His supervisor had a big say in what treatments he was about to receive.

On May 15 2017, Wang underwent radical surgery for esophageal cancer. The resected esophagus had a hard area about 2.5 × 1.0 × 0.8cm large. Postoperative pathological findings indicated Grade II squamous cell carcinoma and showed that cancer cells had retracted and invaded the fibrous membrane. The symptom was accompanied by lymphocytic response.

Based on the comparison of preoperative endoscopy and CT scan findings with postoperative pathological changes, it can be concluded that Wang's esophagus cancer changed from "large cancer" to "small cancer" before the surgery.

After reading the above report, I said: "Wang, I think surgery is good for you and is also necessary."

"Not necessarily. What If I continue to inhale hydrogen and take chemotherapy?" Wang looked very persistent and raised his voice, saying, "Look, my tumor was 5cm originally and reached deep into the esophageal outer membrane. With only two cycles of small-dose chemotherapy,

could the tumor be reduced by half? You are a digestive disease specialist. Have you seen a case like mine? This is definitely the effect of hydrogen."

Cases like Wang's are rare. It seems that hydrogen inhalation is effective for him. Viewing from the purpose of investigating the functions of hydrogen in cancer and emotionally speaking, I want to believe that hydrogen exerted positive effects in Wang's case. But as a doctor in the medical circle, I am expected to be highly rational and not to break away from traditions at will. As a researcher, I should be more rational and seek evidence instead of acting on assumptions. Admiring Wang for his courage and dedication, I took Wang by the hand and said to him: "But if I am your attending doctor, I will not dare to make such a choice for you."

"Why not? This is also an innovation. If we don't try, how can we achieve medical progress? To be honest, at that time, I just wanted to test the effect of hydrogen in me. But then, I had no choice..." Wang gave a helpless smile. I understood him. I learned that Wang accepted the surgery under the order of his supervisor.

He was Full of Confidence

The next day, Wang took me and my colleagues to visit the Green Rehabilitation Studio. He passed the retirement age. But as a rehabilitation expert, he still wanted to serve patients through another platform. In the studio, there were a variety of rehabilitation equipment, including hydrogen-oxygen nebulizers. Wang said that running the studio is his way of giving back to society, because he himself is living well.

Wang summed up the benefits of hydrogen inhalation that he gained. First, the application of hydrogen inhalation together with chemotherapy before surgery makes surgery easier to perform. Wang hopes to further study whether hydrogen inhalation alone can achieve the result. Second, hydrogen inhalation reduces side effects from chemotherapy. Wang said that he barely had any adverse reaction during chemotherapy. Third, hydrogen inhalation promotes postoperative recovery. Wang resumed his work shortly after his surgery. Fourth, hydrogen inhalation prevents recurrence. Wang is confident that his esophagus cancer will not relapse if he continues with hydrogen inhalation.

The fourth benefit that Wang mentioned remains to be determined and requires follow-up verification. Considering the extensive biological effects of hydrogen molecules, and seeing Wang in good spirits, I know that the role of hydrogen can never be neglected.

More than three months after our meeting in Beijing, I wanted to know the effect of hydrogen inhalation in Wang. So, I sent him a WeChat message and asked how he evaluated the contribution of hydrogen in his case. I received immediate reply from him saying that his treatment effect was directly related to hydrogen inhalation.

— Xu Kecheng

> *Picture 3. WeChat message from Wang on June 2, 2018*

> **Comments from Tang Zhaoyou**
> It can be sure that hydrogen delivered positive effects on the patient above. His case raises a very interesting question, that is, hydrogen may have a role before, during and after treatment. It can be used as a neoadjuvant therapy, whose purposes are to shrink the tumor and create conditions for surgery. In The Art of War, Sun Tzu advised against strong attacks. This applies to cancer treatment. When the tumor is big and patient is weak, strong

> attacks at the tumor may not lead to good results. I once used the following example: A cancer patient is like a drowning person desperate to survive. The moment when the patient is only one foot off the shore and about to be drowned, he can be saved if someone offers a hand with a gentle pull. Such a pulling hand must not be underestimated. Hydrogen inhalation is exactly such a "pull."

Hydrogen and Gastric Cancer: Living Freely

Writer with Worries About Gastric Cancer

The treatment of gastric cancer is simple. In the early stage of gastric cancer, tumors are confined to the gastric mucosa or submucosa, and are easy to find by experienced digestive disease doctors through gastroscopy. After surgery or endoscopic resection of the tumors, the 5-year survival rate can be above 80%; and no other treatment is needed generally. However, in the case of progressive gastric cancer, treatment is complicated. In particular, treatment is highly challenging for those with gastric cancer metastasis.

In recent months, I cannot forget the expecting looks of a writer. I remember that day when I was at the outpatient clinic, a middle-aged emaciated man with pale complexions came to me and solemnly handed me two books with his autographs. He was a writer from north China's Shandong Province. Based on the introduction in the books' title pages, I knew that he was great writer. He said that he had just returned from treatment in Taiwan and Hong Kong. He had gastric cancer with ascites and extensive metastases in the abdominal cavity. He took my hand and said, "Prof. Xu, you are my last savior." Holding his bony hands, I had trembling in the heart.

I often feel worried, sad and perplexed about the treatment for patients in similar cases. The purpose of cancer treatment is to prolong patients' lifetime. Everyone will die, but nobody wants to die prematurely or live a miserable life. This writer expected me to be his savior, which is a role that I dare not accept. But as a doctor, I consider saving people as my bounden duty.

I imagined that I could find a panacea for the writer. I was thrilled by an interview in Taipei on March 6, 2018. Perhaps, hydrogen could be the panacea for the writer's adjuvant therapy.

"Young" Old Lady from Taipei

At the invitation of Mr. Lin, a Taiwanese entrepreneur engaged in hydrogen molecular research, I visited Taiwan on an inspection tour. Before going to Taiwan, I checked the weather forecast, which said that Taipei had certain air pollution. It was the second day of the tour. I saw a blue sky and white clouds. The temperature was a little higher than 20 degrees Celsius, which was neither too hot nor too cold. In such pleasant weather, all of us were in good mood.

The hydrogen experience center stood next to the National Taiwan University Hospital. Soon after I entered and sat down on a white workbench, an old lady showed up. Of medium build and with flushing complexions and soft eyes, the old lady was wearing traditional Chinese clothes, radiating kindness and sincerity. I learned that she was Lin Yang Yueying from Taoyuan, Taiwan. I held her hand and asked: "How old are you this year?"

> *Picture 1. Gastroscopy on May 21, 2014 showed progressive gastric cancer under the cardia (pointed by circles). Biopsy indicated adenocarcinoma.*

> *Picture 2. CT scan on May 22, 2014 showed thickening (pointed by arrow) of local gastric wall on the right wall of the higher gastric body, suggesting gastric cancer.*

> *Picture 3. Follow-up gastroscopy on October 26, 2014 showed that the original tumor disappeared and shallow ulcers and erosions were formed (pointed by circles).*

> *Picture 4. CT scan on March 22, 2016 showed that the gastric wall in the higher gastric body was thinner than before, and the original area of thickening was narrowed (pointed by arrow), suggesting that the tumor was under control.*

Looking at me, she smiled. The young man who accompanied her immediately answered: "I'm sorry, my grandmother doesn't understand Mandarin Chinese. She is 86 this year."

I couldn't believe my eyes. I thought she was only a little over 70 and younger than me. She was really a 'young old lady.'

Progressive Gastric Cancer

Her grandson took out a pile of medical records and CDs from his handbag. The old lady had carcinoma under gastric cardia. After the Spring Festival in 2014, she started having difficulty swallowing food. Later, she often vomited after eating dry food. On May 21 that year, she received gastroscopy at Taipei × × Memorial Hospital. A 3-4cm bulging mass was found under the cardia and exhibited erosive bleeding. Biopsy indicated poorly differentiated adenocarcinoma. Abdominal CT scan showed that a new neoplasm was present under the cardia and reached the serosal layer, suggesting progressive gastric cancer. The blood tumor marker CA19-9 was at a level 1450 units, nearly 40 times higher than normal.

The old lady was advised by her doctor to take surgery, radiotherapy and chemotherapy. She refused, not because of financial problems. Besides, people in Taiwan are covered by national health insurance. Her grandson said: "Grandma is afraid of surgery. In fact, our family does not agree either, because she still has a pacemaker in her heart. The whole family only hopes that she will live in peace and without pain for a few years."

> *Picture 5. Blood CA19-9 decreased gradually after treatment to a normal level.* (CA19-9: 1450 units in May 2014 → 307 in August 2014 → 18 by May 2017)

"Hydrogen+" Treatment

Following the advice from a friend, the old lady started hydrogen inhalation for two hours each morning and afternoon respectively. Meanwhile, she took Namixun orally, a type of rice preparations, and a mixed fruit medicinal extract. The measure was called "hydrogen+" therapy.

Hydrogen is produced by an instrument called hydrogen-oxygen nebulizer. The machine is equipped with a special electrode that decomposes pure water and generates 66.6% hydrogen and 33.4% oxygen, which are colorless and odorless. The instrument automatically controls the gas flow rate. When inhaled through a nasal tube, the flow rate is generally 3 liters per minute. Hydrogen inhalation is very convenient, just like oxygen inhalation in a hospital. The old lady inhales hydrogen at home while watching TV, which is quite leisurely.

> *Picture 6. The author was performing a simple examination for the old lady.*

Miraculous Recovery

One month after hydrogen inhalation, the old lady's swallowing difficulty was alleviated. In August 2014, her CA19-9 level dropped to 307 units. The doctor recommended oral capecitabine. On October 26, 2014, the old lady received gastroscopy. According to the findings, the original tumor had disappeared, leaving shallow ulcers and erosions behind. She used xeloda for a total of 9 months until May 2015 and discontinued it due to adverse effects. But she continued with hydrogen inhalation, and had a CT scan every half year. Recent CT scans indicated that the cardia was unobstructed and original tumor signs had disappeared. CT scans and other tests showed no sign of cancer metastasis.

> *Picture 7. The old lady was sharing her experience of cancer.*

More specifically, the old lady's blood CA19-9 gradually decreased to a normal level: In August 2014, two months after the treatment "hydrogen+2", CA19-9 fell to 307 units. By February 2015, it further decreased to 40.62 units, which was close to a normal level (less than 37 units). Thereafter, CA19-9 continued to go down. In May 2017, three years after disease onset, CA19-9 level stood at 17.68 units. CA19-9 is a tumor marker and is very sensitive, reflecting the presence and recurrence of tumors.

I asked about the old lady's daily life. Her grandson said: "She cooks, does laundry and cleans the house every day. She's quite busy." I asked the old lady to weigh her weight. It was 64kg. According to her grandson, she weighed 58kg at the onset of disease 4 years ago. I gave the old lady a simple physical examination and did not detect any abdominal mass or enlarged supraclavicular lymph node. I took the old lady's hand and shook it, feeling that she had strong strength of the hand.

Having been engaged in the clinical work of digestive diseases for more than 40 years, I had never seen a patient with progressive gastric cancer who lived comfortably for 4 years without surgery and still in good systemic conditions. I advised the old lady to take another gastroscopy, but she refused. She said that she could eat and sleep well and felt good, thinking that the cancer had gone.

The old lady once took xeloda, a pro-drug of fluorouracil. Mainly used for treating metastatic colorectal cancer and breast cancer, the pro-drug has limited effects on gastric cancer. Therefore, the old lady's therapeutic results could not have been achieved by xeloda.

So, is it the effect of "hydrogen+"? Hydrogen molecules have been proven to have inhibitory effects on cancer cells. Although the credit cannot be fully given to hydrogen, at least hydrogen plays an important or synergistic role. If further proof can be obtained, "hydrogen + low-dose chemotherapy" can be a potential treatment option in the future for patients with gastric cancer (including other cancers) who are inoperable or unwilling to take surgery.

Cancer Control by Hydrogen Molecules

That night after visiting the old lady, I could not fall asleep. I got out of bed, searched online and found more than 20 articles about the clinical application of hydrogen molecules. According to the articles, hydrogen has therapeutic effects on dozens of diseases. Its effects on cancer were described as follows in one of the articles: Molecular hydrogen inhibits cancer

cell growth and invasion; and causes no harm to normal cell growth. Its mechanisms of inhibiting cancer are as follows:

(1) Molecular hydrogen reduces excessive reactive oxygen free radicals (oxidative stress), down-regulates cell division-related key growth factors (ERK), and suppresses the vascular endothelial growth factor, thereby inhibiting tumor progression and division.

(2) MMP proteins are involved multiple cellular functions (cell proliferation, apoptosis, and angiogenesis). Cancer cells overexpress the MMP gene, resulting in tumor invasion and angiogenesis. Hydrogen molecules reduce overexpression of the MMP gene and therefore help inhibit tumor invasion and growth.

(3) Hydrogen molecules shorten cancer cell telomerase ("end cap" of DNA), thereby promoting cancer cell apoptosis.

(4) Hydrogen molecules boost the body's immune function, thereby strengthening the body's immune surveillance of cancer cells.

(5) For cancer patients with radiotherapy and chemotherapy, hydrogen inhalation reduces side effects, prevents blood leukocytes from decreasing, and improves quality of life.

After discovering the results above, I sent a WeChat message to the Shandong writer that night, telling him that I found a supplementary way to help treat his progressive gastric cancer. I received an immediately reply from his: "Thank you. I will go to Guangzhou as soon as possible."

— *Xu Kecheng*

> **Comments from Tang Zhaoyou**
> Coincidentally, one of my relatives had non-differentiated gastric cancer many years ago, and received chemotherapy after surgery. The relative passed away over a year later. I was very surprised to read about the case in the article above. Three years after treatment, the patient even gained weight. And her gastric tumor disappeared and tumor marker returned to a normal level. There was nothing wrong with her diagnosis; and her therapeutic effect was evident. Then, I read carefully and wanted to know what treatment she received. It turned out to be "hydrogen + low-dose chemotherapy." In modern medicine, questions will be raised as to whether there is evidence-based medicine proof; what exactly exerts the effect; whether the effect is attributed to hydrogen, oxygen, chemotherapy or a combination of all methods. As the saying goes, practice is the sole criterion for testing truth. If a method is effective, it must have its merits. This reminds me of a saying by Sun Tzu: "Any victory in warfare should be acquired by frontal assault and supplemented with surprise strategy." Currently, "frontal assaults" (standard diagnosis and treatments) are much emphasized, which is correct. But without "surprise strategy" to secure treatment success, there will be limited medical advancement.

Hydrogen and Thymic Cancer: Yielding to Hydrogen

Residual Thymic Cancer Yielded to Hydrogen?

Mr. Z has been both very busy and happy in the past six months. He is busy with his medical equipment business. He is happy because he seems to have waved goodbye to death when going back to his business again, which was unexpected at the beginning.

On July 24, 2018, I met Mr. Z for the first time in a ward on the third floor of xx Cancer Hospital in Shandong. With flushing complexions and bright piercing eyes, Mr. Z was tall, spoke with a heavy Shandong accent loudly and went straight to the point, which were typical among uniquely frank and open men from Shandong.

"You never thought that I could come back here alive?" Mr. Z was hospitalized in this ward and was familiar with the medical staff here. After hearing that I was here to see patients, Mr. Z came specially today. He said: "I went to Beijing for surgery after I was found to have a thymic tumor in this hospital. But the tumor was too big to be surgically removed." He handed me a discharge diagnosis report from Peking University xx Hospital The report stated:

> *On June 29, 2017, the patient went to a cancer hospital for CT scan due to unexplained chest tightness. The scan showed irregular nodules and masses in the anterior mediastinum, and their maximum cross-section measured 101 × 38 mm. Multiple nodular masses were detected in the left pleura. A cord-like shadow was found in the lower lobe of the left lung. On July 5, 2017, the patient received thoracoscopic biopsy on the mediastinal mass under general anesthesia. During the operation, multiple hard white nodules were found on the pleural surface of the visceral layer. Moreover, a 6 × 5 × 5cm mass was seen in the anterior mediastinum and was closely adhered to the left lung tissues. During the operation, surgeons first removed the pleural surface nodules locally on the visceral layer. Biopsy results indicated thymoma (mainly B2 type) and the possibility of thymic cancer in local foci. Then, wedge-shaped excision was applied with a linear cut stapler to the pleural surface nodules of the visceral layer in the lower lobe of the left lung. Since radical excision was impossible due to the presence of thymoma and extensive multiple implantation metastases in the pleural cavity, surgeons ended the operation…*

After the operation, Mr. Z received chemotherapy. The first chemotherapy was still tolerable. After the second one, Mr. Z had weakness of limbs, poor sleep, no appetite and enormous fatigue. He lost more than 30kg weight in a short time. Unwilling to endure the suffering again, Mr. Z asked around in an attempt to find a non-painful treatment. He went to a certain treatment cancer and spent tens of thousands of RMB yuan on stem cell immunotherapy.

"Can stem cells treat cancer?" I asked surprisingly. "I was desperate at that time, trying to grasp at any straw. I knew that I had multiple metastases in the chest, and that chemotherapy would be useless. I had no choice but to look everywhere for any good method out there." said Mr. Z.

"Did you continue with chemotherapy? Only three sessions are not even enough for one course of treatment." I said.

"Yes, my doctor told me the same. Fortunately, I met the right person," said Mr. Z, looking at Mr. S standing next to him. "It is him who sent me a hydrogen-oxygen nebulizer. I myself work in the medical equipment industry. I saw that the nebulizer was an innovative medical product to be approved by the country. So, I used it."

Mr. S, a big fan of hydrogen research, added: "We are friends. Seeing him so weak and considering that he did not want to take chemotherapy again, I suggested that he inhale hydrogen during the period of chemotherapy, because I read from literatures that hydrogen reduced the side effects of chemotherapy."

"It's amazing! Hydrogen has helped me a lot. On October 1st, 2017, before the fourth chemotherapy, I started to inhale hydrogen." said Mr. S while looking at me. "He wanted me to inhale hydrogen for 4 hours a day, but I did it for 6 hours a day. A few days later, I had better

sleep, appetite and spirits. During the fourth chemotherapy, I did not experience side effects." My attending doctor also wondered why my white blood cells showed no decrease.

"Did your doctor know that you were inhaling hydrogen?" I was curious.

"No. At that time, the doctors did not know that hydrogen can treat diseases." said Mr. Z with a smile.

"Why did you receive only four sessions of chemotherapy?" I asked.

"I checked online. Chemotherapy is not effective for thymic cancer. The doctor who treated me said this too."

Mr. Z went to the Proton Heavy Ion Treatment Center in Shanghai, where the doctor checked his discharge report from a Beijing hospital, hurriedly went through the CT films, and told him that he was not suitable for proton heavy ion treatment.

In November 2017, Mr. Z went to Linkou × × Hospital in Taiwan and consulted Prof. Liu.

"Did Prof. Liu think it was good for you to take surgery again?" I asked. Prof. Liu is a thoracoscopy and thoracic disease expert with a distinguished reputation on both sides of the Taiwan Strait.

Mr. Z looked at me and said: "Prof. Liu was careful and repeatedly checked my medical imaging films. He also gave me a physical examination on the spot. He said he could open the thoracic cavity to check the inside but was not sure that the tumor could be removed by surgery. Hearing the words from such a well-known doctor, I was disappointed with modern medicine." said Mr. Z bitterly.

After that, Mr. Z just stopped thinking about the tumor in his body. In the following week, he traveled by train and car around Taiwan island for more than 2,000 kilometers. Strangely, he was energetic without feeling tired and uncomfortable. He attributed this to hydrogen inhalation.

> *Picture. Mr. Z's chest and abdomen CT scan showed: The thymic tumor is stable and is smaller by 2cm compared with the CT scan findings 4 months ago. The abdomen including the liver displays no sign of metastasis.*

Mr. Z went on excitedly. He took out the report and images from his latest CT scan. The report showed that the tumor shrank by 2 cm; and effusion in the left pleural cavity was in a small volume and significantly less than before.

"Mr. Z, do you think hydrogen inhalation really works for you?" I asked with a very heavy tone. I always raise the same question to every patient who has inhaled hydrogen. As a hydrogen medicine enthusiast and emotionally, I hope to get positive and affirmative answers from patients. But as a researcher, science and truth are the most important. In particular, as a clinician, I am responsible to patients. Therefore, I want to hear the truth and rationally, hope to hear negative voices.

"It is surely effective. I have no doubt. Prof. Xu, I came here to see you today just to tell you this fact. I hope other people will also benefit from hydrogen." said Mr. Z seriously. He continued: "A few days after hydrogen inhalation, I felt more energetic in the body. During my fourth chemotherapy, I did not experience the same side effects as my previous ones. If this were not because of hydrogen inhalation, what could it be? After 4 months of hydrogen

inhalation, I took a CT scan. It showed that the tumor shrank to the size of 0.1 × 0.3cm and became at least stable. My pleural effusion had also decreased. In that 4 months, I did not receive any other treatment."

What Is the Next Treatment?

Thymic epithelial tumors include a group of anatomical, clinical, histological, and molecular heterogeneous tumors that occur to the thymus. According to classification by the WHO, thymoma is categorized into subtypes A, AB, B1, B2, and B3 based on thymic epithelium forms. Thymic carcinoma mainly manifests as squamous-cell epithelioma, for which surgery is the only means of cure.

Prognosis is closely related to disease staging. Around 50% to 60% cases of Type B2 thymic cancer and 60% to 80% cases of Type B3 thymic cancer belong to Stage III to IV. Thymic tumors that cannot be surgically removed are usually progressive and difficult to treat. Radiotherapy and chemotherapy are the main palliative means.

Mr. Z's thymic tumor fell under Subtype B2 in pathology. Though having not been fully confirmed as thymic carcinoma by frozen biopsy, his tumor was surely malignant based on the biological behavior that multiple metastases were present.

"Prof. Xu, what other treatments are there?" Mr. Z asked me, looking full of expectations.

"You can take chemotherapy repeatedly. Chemotherapy is suitable in around 50% to 70% cases of thymoma recurrence. Paclitaxel and pemetrexed are less toxic and can be applied." I turned on my computer and found a literature of retrospective study by Dr. Song from Zhejiang Cancer Hospital. In the study, a total of 86 patients with progressive thymic cancer were treated from 2000 to 2012. The objective response rate in the patients after chemotherapy was 47.7%, and median progression-free survival was 6.5 months. There was little difference between different chemotherapy regimens (Clinics [Sao Paulo], 2015).

Suddenly, everyone present fell into silence. I noticed that the literature I showed might be too pessimistic. Mr. Z looked downcast. "Maybe his confidence was hurt." I thought to myself, feeling a little apologetic.

"But, look at this literature. It talks about prognostic factors, systemic conditions, histological grades and liver metastasis. This is the most relevant. Mr. Z is now in such good conditions and looks healthier than a healthy person. His liver is clear of any metastasis. His case will definitely be positive." To change the atmosphere, I asked several doctors present to read the literature and compare Mr. Z's CT scan images. Eventually, everyone laughed.

"therapy be applied?" asked the president of the xx Cancer Hospital, who was present at the scene. "This disease is not commonly seen, and we are not familiar with it. Is there any literature report about its treatment?"

I looked online and found an article published in 2013 European Respiratory Review. The article reported two Phase II trials, in which thymic cancer was treated with imatinib, a tyrosine kinase inhibitor targeted at KIT. No beneficial effect was achieved. In another Phase II trial, bevacizumab was used in combination with erlotinib in 11 cases of thymoma and 7 cases of thymic carcinoma. And no tumor response was observed.

"Can he take radiotherapy?" I asked Dean Wang, a professor at a medical school and a radiologist.

Dean Wang shook his head and said: "The original lesion was too extensive. Radiotherapy is not likely to be effective. Instead, I hope that he will continue to inhale hydrogen. After the lesion is further controlled, radiotherapy can be considered."

Afterword

I agree with Dean Wang that Mr. Z continues with hydrogen inhalation. Having been inhaling hydrogen for 19 months, Mr. Z said that he will not stop inhaling hydrogen regardless of other available new treatments.

While at a loss in front of some patient cases, clinicians often think of alternative treatments. Hydrogen inhalation is an alternative option.

Has the residual thymic tumor of Mr. Z really yielded to small hydrogen molecules? Mr. Z himself is not worried, because he believes that hydrogen is harmless and easy to inhale. He does not see a reason for discontinuing it.

In terms of clinical research, we seem to care more about data. Moreover, further research is required on the dose of hydrogen, the course of treatment, and how hydrogen works with other treatments.

The 21st Century Cures Act, promulgated by the United States in 2016, proposed the research method Real World Evidence (RWE). Based on clinical practice and effects, RWE adopts non-interventional and observational studies; and presents treatment safety, effects and models. Why don't we apply RWE to studying hydrogen inhalation, a natural treatment?

— *Xu Kecheng*

> **Comments from Tang Zhaoyou**
> Cancer transformation therapies (such as hydrogen inhalation in this case) are usually not taken seriously, for the simple reason that all efforts have been targeted at killing tumors since Rudolf Virchow proposed the theory on cancer origin. Transformation therapies have no immediate effect on killing tumors. To bring attention to the combined use of cancer killing and transformation treatments, it is necessary to start from the source, which is to renew the perception of the nature of cancer. In fact, cancer is different from infectious diseases, which are caused by invasion of "foreign enemies" (bacteria and viruses). Instead, cancer is an in vivo disorder caused by an imbalance between the internal and external environment, because cancer cells are derived from normal cells. Just like dealing with crimes, mere death penalty is not enough. There must be imprisonment. Imprisonment is assessed not by the same criteria as evaluating death penalty. This makes it necessary to recognize that tumor-bearing survival with good quality of life is another end point for cancer treatment.

Hydrogen and Pancreatic Cancer: Miracle in Daren (1)

Talking Him Out of Euthanasia

Celebrities are celebrities after all. Although fans of celebrities are mainly young people, I, in my eighties, found myself admiring Fu Daren from Taiwan like a fan. He was a well-known figure on both sides of the Taiwan Strait. In addition, he had pancreatic cancer and was prepared for euthanasia. More importantly, he was said to be inhaling hydrogen, which is relevant to my profession and research.

Recently, an idea keeps circling in my mind: Maybe hydrogen inhalation will be effective for Fu Daren? Maybe I can talk him out of euthanasia?

An Absolute Celebrity

Fu Daren was absolutely famous on both sides of the strait. He was a basketball master and coach in Taiwan. He had won a silver medal at Asian Games, reported more than 10,000 events including NBA competitions, hosted Taiwan TV variety show Da Jia Le, and won the Golden Bell Awards for the fine variety show. At the 1990 Beijing Asian Games, Fu Daren and Song Shixiong, a famous sports anchor in Chinese mainland, broadcasted the event as commentators. On the New Year's Eve in 1991, Fu Daren performed the show Truth About Shandong at CCTV Spring Festival Gala together with his wife. His ingenious way of speech in the show enchanted countless fans. Fu Daren was famous also because he was the descendant of a hero in the anti-Japanese war. His father Fu Zhonggui was a native of Changqing, Shandong province and a major general of the National Revolutionary Army. He was killed beside the Yellow River Shandong section in the war against Japanese aggressors in 1938. Fu daren's mother passed away very early. As a result, Fu Daren became an orphan from childhood. Later, he was adopted by Song Meiling and went to Taiwan at the age of 15.

> *Picture 1. Fu Daren hosting an NBA event live and attracting numerous fans (picture from public media)*

> *Picture 2. Fu Daren's sketch drawn in Switzerland, recording the last journey of his life (picture from public media)*

> *Picture 3. Fu Daren and his son Fu Junhao in the same show (picture from public media)*

> *Picture 4. Fu Daren hosting his son's wedding (picture from public media)*

Suffering from Advanced Pancreatic Cancer and Wanting Euthanasia

In recent years, Fu Daren became a legendary figure in Taiwan, because he wanted eagerly to practice euthanasia on himself but was somehow returned by euthanasia.

In May 2017, 84-year-old Fu Daren was found to have pancreatic cancer. Within three months, his weight dropped from 74kg to 58kg. He petitioned to the leader of Taiwan, asking for establishing a euthanasia act for those with incurable diseases. He even said: "If the act is enacted, I myself will be the first to execute euthanasia." He also intended to turn the original book launch conference into a Farewell to Life memorial service.

He found Dignitas, the world's only institution that provides euthanasia internationally, and became a qualified member of the organization. In November 2017, he went to Switzerland and obtained a euthanasia "green card" under Swiss euthanasia legislation. He was prepared to have euthanasia in Switzerland after a few months.

On November 19, 2017, Taiwan's Lianhe Zaobao published an article titled Fu Daren "About to Go Home" After Getting Euthanasia Permission. The article was as follows:

> *In the terminal stage of pancreatic cancer, a 84-year-old Taiwanese senior sports anchor Fu Daren has only two months to live and actively advocates a dignified and comfortable end of life. In the near future, he will go to Switzerland with his family to seek euthanasia. So far, he has obtained the green light to euthanasia, which can be executed at any time. The whole process is expected to be recorded on Facebook. Yesterday, Fu Daren posted a*

> *photo of him and his son on Facebook and expressed that his family has mixed feelings despite the fact that he had obtained the green light to a comfortable death. He wrote: "It's hard to part with my family. It's also hard not to. Crying is not enough to express the farewell of the century. Tears are not enough to shed in front of such huge sorrow." At the end of the post, Fu Daren revealed that he was "about to go home". He planned to use every minute possible to make a painting about the father-son hug and hold a poetry, painting and calligraphy exhibition in Taipei, calling for support for the euthanasia end of life act.*

The following was also reported in Taiwan media:

> *Fu Daren and his son Fu Junhao went to a TV show together. In the show, Fu Daren sat next to his son, with his hands on the son's shoulders, and asked gently: "I am leaving the world, will you be fine?" For a long time, the son could not speak: "Dad, I love you." Fu Daren looked emotional and responded: "Son, I love you."*

Postponing Visit to Switzerland

On January 27, 2018, Taiwanese media published the following article:

> *Fu Daren, a 84-year-old Taiwanese senior sports anchor, is in terminal stage of pancreatic cancer and went to Switzerland not long ago to seek euthanasia. So far, he has obtained the green light to euthanasia. He is scheduled to fly to Switzerland to receive euthanasia at the end of this month. He has also chosen to wear a red suit to welcome the "happy event". But on the 26th, he was told by his son: "Dad, Yanru and I decided to get married before the Spring Festival. Go to Switzerland later." Fu Daren decided to postpone his trip and stay in Taiwan for his son's wedding. He said he was speechless.*

Fu Daren said that he has 2.5 marriages. He has a daughter with the first wife. The couple divorced long time ago. His first wife is currently living in the United States with their daughter. Fu Daren's second wife was the widow of a lieutenant general of the Taiwan Navy and an anchor of the Taiwan radio station. The "0.5 marriage" is with Ms. Chen, who is 36 years younger than Fu Daren. Despite that she did not register marriage with Fu Daren, Ms. Chen has been staying together with Fu Daren for a long time like a confidante. In the year they met, 17-year-old Ms. Chen admired Fu Daren for his talent, fell in love with him, and gave birth to a child. The child is 26-year-old Fu Junhao, a 180cm-tall and handsome man. Having a son in his old age, Fu Daren loves the son beyond words. The second wife, who did not have any child with Fu Daren, also treats Junhao as her own. Junhao often said that he is very happy to have two mothers who love him.

Fu Daren decided to go to Switzerland after his son's wedding.

Visiting Fu Daren

On March 6, 2018, the second day after arriving in Taipei on Hydrogen Tour in Taiwan, the car drove the five of us through the Sun Yat-sen Memorial Hall. We saw the Taiwan TV tower, the biggest media center in Taiwan, in the distance. After passing a few old streets, the car stopped in front of an ordinary apartment building at the 3rd section of Bade Road. Mr. Lin, who was very familiar with the place, led us to a small elevator and then a room on the third floor. The door was open. A tall and thin old man stood by the door and shook hands with us, saying: "I am Fu Daren from Taiwan."

He was Fu Daren. With a tall and well-built figure and gray nice long hair, Fu Daren was wearing black traditional Chinese clothes and a brand-name scarf printed with white flowers.

In spite of his serious disease, Fu Daren kept the demeanor as a media star famous at home and abroad.

Then, we sat down on the couch in the living room. The living room was so simple that we could not believe it was the home of a media celebrity. On the wall of the living room displayed numerous awards, medals, trophies, and photos with Taiwanese celebrities, indicating that the owner had a profound cultural background and high social status. A big photo showing basketball star Jeremy Lin's shooting at the basket stood beside the front wall of the living room. The photo showed the owner's love for basketball.

Fu Daren and I stood face to face and looked at each other eye to eye. He uttered many of my titles and awards without stammering; and said with a smile: "I was very happy to hear that you are from the mainland. I immediately searched the internet for information about you. We are similarly afflicted with disease."

Mr. Fu took my hand and said, "I am planning to go to Switzerland at the end of this month. You are here. Maybe you are the last batch of mainland friends I will receive." He said that he had finished two things: He hosted the wedding for his son and split his property completely. With everything arranged, he no longer had any concern in the world.

He took out a basketball with his own signature as well as a photo and signature of Jeremy Lin; and solemnly handed the basketball to me and said, "I am about to leave this world. I have nothing valuable here. Only this basketball is precious to me. I want to give it to you, a master." Then, He picked up a newly published book, signed the book, and posed himself seriously, asking for a photo be taken of us.

He spoke unceasingly about his disease. He said that he had pancreatic cancer and suffered a lot of hardships. He didn't want his family to suffer and didn't want to consume social resources. Therefore, he decided to take euthanasia.

"Actually, I hate to part with my son and family. I was happy to host a wedding for my son after returning from Switzerland." Fu Daren laughed. He said: "Strangely, I seem to become better except abdominal pain and diarrhea."

I gave him a simple physical examination, and found no enlargement of the liver & spleen and no ascites or mass. He felt pressing plain when I applied deep pressure to the abdomen. He said that he had abdominal metastases according to his doctor. However, judging by my experience, Fu Daren's disease was not in a state beyond remedy. This gave me confidence to treat him. Surely, I needed to understand the history of his past treatment. He said that the key turnaround happened in the last two months.

> *Picture 5. Fu Daren presented an autographed basketball and book to the author. The book was titled Fighter with a Comfortable End of Life: Fu Daren's Poetry, Calligraphy and Painting Works Collection.*

I asked him what treatment he had received after he was diagnosed with pancreatic cancer. According to him, the tumor was in the head of the pancreas and pressed against the common bile duct, causing jaundice. The doctor placed a stent in the common bile duct guided by endoscopy. Afterwards, Fu Daren took morphine tablets every day to ease pain and did not receive any other special treatment.

He said: "Pancreatic cancer is too violent to be operated on. Luciano Pavarotti, a world-renowned operatic tenor, had pancreatic cancer and received surgery and chemotherapy in

the United States. He suffered tremendously but survived for only 13 months. I believe that people should work hard to move forward when they are young, and be happy to say goodbye to the world when they are old. So, I want to be the first person to have euthanasia in Taiwan. In this way, I want to tell other people that death should be comfortable."

"But you are currently in stable conditions." It was hard for me to understand. In general cases, pancreatic cancer develops progressively. For Fu Daren, it had been one year since the onset in May 2017. His case seemed tremendously against the "established patterns".

He thought about it and seemed to suddenly realize something. He said: "I inhale hydrogen. Starting from October 17, 2017, I inhale hydrogen for 6 hours a day." He looked at Mr. Lin, who was present, and said: "Lin sent a hydrogen apparatus to me from the mainland. We have been friends for years." He took Mr. Lin's hand and said, "Thank you."

> *Picture 6. Fu Daren signed on his book A Biography of Daren and gave the book to the author.*

Hydrogen is Effective?

Mr. Lin said: "Because there was no other way for Mr. Fu. I have studied the role of hydrogen in the human body for seven years. I discovered that in the process of making animal models of tumors, the models could not be successfully made if the animals were given hydrogen inhalation. During cell culture, I also found that tumor cell movement slowed down and cell division decreased in a hydrogen-containing environment."

My study of Chinese way of cancer control in recent years has given me the faith not to give up easily on patients. Academician Tang Zhaoyou advocates cancer control rather than cancer elimination. Cancer cells are not invading "foreign enemies" but represent "internal disorder". They are "traitors" generated by cumulative gene mutations in normal cells under the influence of adverse environment. In the case that "foreign enemies" such as disease-causing bacteria and viruses invade, measures are required to kill and eradicate them. But cancer cells generated as a result of "internal disorder" can only be controlled. In summary, not only cancer killing therapies but also cancer transformation measures are needed in cancer treatment.

> *Picture 7. The author introduced to Mr. Fu the newly published book Practice Chinese Way of Cancer Control and explained the application of strategies in Sun Tzu's The Art of War to cancer control.*

> *Picture 8. Fu Daren took a photo with us excitedly and said, "Go to Guangzhou, and take the road to survival." (from left: Dr. Wang Qiang, the author, Fu Daren, Mr. Lin)*

How to control cancer? Academician Tang Zhaoyou used Sun Tzu's The Art of War to explain cancer control strategies. For example, in the chapter Potential, Sun Tzu wrote: "Any victory in warfare should be acquired by frontal assaults and supplemented with surprise strategy." Frontal assault is the foundation, whereas surprise strategy is the soul. If prescribed routine is strictly followed, breakthroughs are difficult to achieve. The focus is to take surprise actions to defeat cancer. As written in the chapter Dispositions of The Art of War, "The general who is skilled in defense hides in the most secret recesses of the earth." If the tumor is fierce and cannot be eliminated according to estimation, it is necessary to "stay put and reserve energy",

launch moderate attacks at the tumor, and boost the body's immunity, thereby achieving peaceful coexistence with cancer cells and survival.

Hydrogen is a selective neutralizing agent for reactive oxygen such as toxic radicals and OH. Reactive oxygen has strong oxidizing properties and damages nuclear and mitochondrial DNAs in cells, causing cell mutations. Hydrogen can transform cancer cells and their microenvironment. Could Fu Daren's pancreatic cancer be controlled by H_2, a small natural substance?

I wanted to study Fu Daren's treatment and understand what actually happened. Moreover, for Fu Daren, a celebrity of courage and uprightness typical of the Chinese nation, I had only one idea in my mind: save him.

I also thought that if I further treat Fu Daren in compliance with strategies under Chinese way of cancer control, perhaps I will be successfully save him by standard treatments in combination with unconventional measures.

Inviting Him to Guangzhou

I proposed to invite Fu Daren to Guangzhou. I said, "Daren, it seems that we are brought together by fate. Come to Guangzhou. At least, I can take you to enjoy the food there."

Fu Daren stood up and said, "Really? I miss the mainland. I have many friends there. There are so many delicious foods in Guangzhou. I will be able to finally have a taste of them."

He took us by the hand and said: "Go to Guangzhou. Take the road to survival." As a media professional, he is indeed different from others in words and actions.

— *Xu Kecheng*

> **Comments from Tang Zhaoyou**
> The touching story above reminds me of a poem that reads: "Where the hills and streams end and there seems no way out, amidst shading willows and blooming flowers another village appears." Human beings cannot live forever but can live longer. Readers might ask: "Is it really because of the effect of hydrogen inhalation?" I think the question is difficult to answer for anyone. After all, the patient received multiple treatments. Remission of terminal pancreatic cancer is indeed a coincidence. However, there is often the inevitable in coincidences. Hydrogen inhalation is apparently not a treatment for mass destruction of tumors. But on the basis of cancer elimination therapies, hydrogen inhalation may achieve small victories as in guerrilla warfare. This case is once again a reminder of the importance to combine cancer elimination therapies with transformation methods.

Hydrogen and Pancreatic Cancer: Miracle in Daren (2)

"Reborn" in Guangzhou

At 7pm on April 5, 2018 in a private dining room on the fifth floor of Soluxe Hotel Guangzhou in Huangpu Avenue, Guangzhou, I treated Fu Daren to dinner to celebrate his examination results. He had a full day of barium meal radiography for the stomach and intestines. The findings showed no evidence of cancer metastasis in the abdominal cavity. At dinner, Mr. Fu ate sea cucumbers with good appetite. The sea cucumbers were imported from Brazil by my friend at my request. They were said to be natural, contain abundant arginine, and have anti-cancer effects. Suddenly, Mr. Fu's eyes were red, and the hand holding chopsticks shivered.

He said: "If I had not come to Guangzhou, I had already left the world for 48+8 hours." He sounded calm and sad.

His confidante Ms. Chen, sitting on his right side, had tears in her eyes. She said: "The day before yesterday, April 3, was Mr. Fu's birthday. It was also his planned death day…"

Mr. Fu went on to say, "I have pancreatic cancer. There is no treatment for me in Taiwan. With the disease certificate from the Taiwan hospital, our whole family went to Switzerland in November 2017. It is the only place in the world that is open to foreigners for euthanasia. After hosting the wedding for my son, I bought tickets for the whole family to go to Switzerland again on March 26 this year. It was scheduled for me to enter the death procedure at 11am on my birthday on April 3: First, I will take a liquid orally and fall asleep after 3 minutes. Then, I will be given an injection of a lethal agent and die in 10 seconds after the injection." He spoke very slowly and seemed to be talking about story of somebody else.

> *Picture 1. Mr. Fu, Ms. Chen and the author shook hands and said unanimously: "This is predestination."*

My eyes were covered by tears. I held Mr. Fu's hands all at once, and Ms. Chen joined us. The three pairs of hands clasped together, shaking and shaking. I said, "Congratulations to Daren on your rebirth."

"Yes. I have gone through the death day." Mr. Fu suddenly raised his voice, "Reborn in Guangzhou. I am Happy."

I am not sure whether there is such thing as rebirth. Some people say that from a quantum point of view, life probably runs in samsara, which is believed by Buddhists. But Mr. Fu's life was indeed reversed on the other side of the Taiwan Strait.

"If you hadn't come to Taiwan to see me on March 6, and if Mr. Lin hadn't brought me from Taiwan to Guangzhou on March 22, I would have flown to Switzerland. What a predestination!" Mr. Fu was excited and laughed.

Coming to Guangzhou

At 6pm on March 22, 2018, Fu Daren arrived in Guangzhou under the escort of Mr. Lin. In spite of abdominal pain from time to time and constant diarrhea with watery stool, Mr. Fu was excited to experience Guangzhou, a city which he had not visited for years.

Mr. Fu stayed in Room 501, a special room in the fifth ward of our hospital. The room was the closest to my office. Dr. Kong, a doctor with more than ten years of medical experience, was responsible for Mr. Fu's treatment. The nutrition room manager cooked every meal for him. Soon after hospitalization, Mr. Fu received a series of tests, including blood test, ultrasound, CT scan, chest X-ray and barium meal radiography.

Unexpected Results

The test results were both as expected and unexpected. CT scan findings showed that Mr. Fu's pancreatic cancer was still present in the head of the pancreas. The tumor measured around 7-8cm and had invaded the common bile duct, in which a stent was found. The pancreatic duct in the tail of the pancreas was dilated. There were multiple small lymph nodes around the pancreas and surrounding the blood vessels. The findings were not significantly different from those of the PET-CT scan in a Taiwan hospital 2 months ago. Amazingly, PET-

CT scan indicated that the metastases in the left hepatic lobe and close to the porta hepatis were gone.

> *Picture 2. Comparison of examination results*
> PET-CT scan in January 2018 showed a tumor in the pancreatic head.
> PET-CT scan in January 2018 showed metastasis in the liver (indicated by the white solid circle).
> CT scan (contrast-enhanced scan) on March 25, 2018 showed a tumor in the pancreatic head, which was similar to the tumor detected in the PET-CT scan described above.
> No metastasis was detected in the liver during the CT scan (contrast-enhanced scan) on March 25, 2018.

> *Picture 3. Barium meal radiography showed: The entire small and large intestines are supple and more actively. No sign of abdominal metastasis (mass) is noted in the abdominal cavity.*

It was the ninth month of Mr. Fu's pancreatic cancer. Generally, patients with inoperable pancreatic cancer have a median survival of 6 months. After half a year, pancreatic cancer metastasizes everywhere, especially to the liver. As a result, patients have their livers full of metastases. In addition, pancreatic cancer tends to metastasize to the lung and abdomen. Nevertheless, Mr. Fu had a clear lung field. He received gastrointestinal barium meal radiography. The director of the radiology department spent more than 8 hours observing the movement of barium from Mr. Fu's esophagus to rectum. Except that the sigmoid colon was found to be lengthy (probably congenital), Mr. Fu's intestinal canals showed good movement and no adhesion. No sign of metastatic mass was noted in his abdominal cavity.

Even more amazingly, Mr. Fu's level of circulating tumor cells was only 1/ml (within normal range). The various types of lymphocytes and cytokines in the blood, which reflected immune function, were in completely normal ranges.

The above examinations showed that Mr. Fu's pancreatic cancer was in a stable state. What was the magic medicine that tamed his severe cancer or at least slowed down cancer progression? Fu Daren himself found the new examination results hard to believe. In particular, he did not believe that the liver metastasis had disappeared. His doctors in Taiwan concluded that he needed no treatment based on the scale of metastases in the liver and abdomen. Mr. Fu asked the director of the radiology department to come to his ward and explain to him: Was the liver metastasis really gone?

Contribution of Hydrogen

Fu Daren firmly believed that it was hydrogen that saved him.

Right, what else could it be if it was not hydrogen? Mr. Fu did not receive any anti-cancer treatment, only an operation for placing a stent in the common bile duct. The operation was performed to relieve pressure on the common bile duct from the pancreatic tumor and to discharge bile. It did not have any anti-cancer effect.

On October 17, 2017, Mr. Lin sent Mr. Fu a hydrogen-oxygen nebulizer. As a Christian, Mr. Fu believed that it was a gift from God. He believed that God gives oxygen to keep people alive and God sends hydrogen to make people healthy.

Since the Japanese professor Shigeo Ohta first proposed the selective antioxidation of hydrogen in 2007, a large number of studies have been focused on the biological effects of

hydrogen molecules. It was found that hydrogen has the following unique advantages as a new type of antioxidant:

First, hydrogen has selective antioxidant effects. With weak reducibility, hydrogen does not react directly with the reactive oxygen that has weak oxidation. But it reacts directly with the reactive oxygen that possesses strong oxidation, such as hydroxyl radicals and nitrite anions. Second, hydrogen itself has a simple structure. And the products generated from hydrogen reaction with free radicals are also simple. Therefore, hydrogen leaves no residue and has no toxic side effects on the body. For example, it reacts with hydroxyl radicals to generate water, and excess hydrogen can be excreted out of the body through respiration. Third, with a low molecular weight, hydrogen can pass through the blood brain barrier and freely diffuse to any place within cells, including the nuclei and mitochondria.

Cancer cells are products of gene variation and mutations in normal cells under the influence of environmental factors. Factors such as smoking, chemicals, radiation, infections, inflammation, unhealthy emotions, and excessive exercise all facilitate the production of free radicals and gene mutations in cells. Therefore, in theory, the use of hydrogen to resist oxidation and eliminate free radicals evidently inhibits cancer cells and cancer progression. Surely, hydrogen molecules might have more effects. But existing experiments have proved that hydrogen has inhibitory effects on cancer cells.

Fu Daren's liver metastasis disappeared. Is it because of the inhaled hydrogen? I hope to obtain further verification. However, from a clinical perspective, hydrogen, which are simply convenient, affordable and non-toxic small molecules, is undoubtedly beneficial to transforming cancer cells or their microenvironment.

Enjoying Happiness

As a senior media professional, Fu Daren has extensive knowledge and rich experience. He wanted to enjoy the time in Guangzhou. He said: "From now on, all the time is a gift for me from God."

Once, I considered treating Fu Daren with percutaneous cryosurgery or irreversible electroporation (nanoknife) to ablate his pancreatic tumor. But for an 85-year-old patient like him, my biggest concern was safety even though the operation was minimally invasive.

For Mr. Fu, I asked the best acupuncturist in our hospital to take care; invited a famous pain treatment expert in Guangzhou for consultation; and applied painkillers, intestinal function regulating drugs, immunomodulators and traditional Chinese medicine.

Certainly, I encouraged him to continue with hydrogen inhalation. He was very experienced and inhaled hydrogen for 3 hours each morning and before going to sleep respectively. He used a face mask instead of a nasal tube for inhalation, and intentionally breathed in bigger amounts during inhalation. According to Mr. Fu, hydrogen molecules are too small and light. Therefore, it is necessary to make extra efforts to inhale hydrogen so that it spreads to every corner of the body.

The Art of War proposed that the supreme art of war is to subdue the enemy without fighting. The human body is a whole. And rehabilitation from diseases involves mood, nutrition, exercise and detoxing. The toxins to be detoxed are toxic active oxygen. Hydrogen bears this task, but its intensity might be insufficient. It is necessary to ensure that all the important

aspects of rehabilitation are at work before the optimal effects can be achieved with few resources and final victory can be attained.

I kept my promise to have Mr. Fu enjoy food in Guangzhou. In addition to the special diet for him in the hospital, I myself or my colleagues took him to restaurants outside every two or three days. So far, he has tasted Cantonese cuisine, Huaiyang cuisine, Chaozhou cuisine and Guangzhou traditional morning tea. He had very good appetite. Every time, I had to remind him to control food intake.

The earliest and most common symptom in patients with pancreatic cancer is anorexia. I was happy to see that Mr. Fu ate a lot. I had never seen a pancreatic cancer patient with such a good appetite. I was worried that the food was too much to digest, because pancreatic cancer patients are prone to pancreatic insufficiency.

Mr. Fu's pain attacks gradually decreased. His frequency of defecation changed from 10 times a day to 8 times, 6 times, 3 times…

Once I went to his ward room and happened to see Ms. Chen, with whom he claimed to have 0.5 marriage, leaning close to him, touching his face and pinching his cheeks. Noticing that I came in, Ms. Chen was a little embarrassed and said to me with a smile: "President Xu, look, he is more chubby. After coming to Guangzhou, he has gained 2kg weight."

A patient with terminal pancreatic cancer gained weight? Great news.

> *Picture 4. Mr. Fu was about to board a China Southern Airlines flight to Taipei and wanted to take this photo before leaving.*

> *Picture 5. WeChat message from Fu Daren on April 21, 2018. The lady standing on his right side is his step-daughter Lei Qian, a famous media professional in Taiwan.*

I As I just stepped into his room one morning, Fu Daren rose from the couch, stood upright, saluted me and said: "President, my stool today is in shape."

He seemed to be a simple and innocent boy.

He took my hand and said seriously: "President, I am very worried." Just when I was surprised and got worried, Mr. Fu laughed: "Now I have less abdominal pain. What if my abdominal pain is gone after I return to Taiwan and I am not used to it?"

He is such a big fun and really deserves the reputation as a popular TV presenter in Taiwan.

Returning to Taiwan with "Victory"

Fu Daren was going back to Taiwan. A nurse accompanied him to the former site of Huangpu Military Academy and Beijiao in Guangzhou. He cried and did not speak for a long time…

At 9:30am on April 21, Mr. Fu Daren and Ms. Chen boarded a flight of China Southern Airlines and flew to Taiwan.

After arriving in Taiwan, I received a WeChat message from Ms. Chen: "…My husband came back to life from the jaws of death. We are all grateful to you so much." At 7:14 pm, I received a WeChat message from Mr. Fu: "Because of you, I lived for so many more days and with so many blessings. It was unforgettable. Whenever I open or close my eyes, your smiles flash on my mind… Xiao Qian (Lei Qian, Fu Daren's stepdaughter, president of Taiwanese

Women's Association and former Kuomintang legislator). I am very grateful…" Later, Mr. Fu called me and said: "A month later, I will definitely go to Guangzhou again."

I said happily: "Looking forward to seeing you back, Daren."

— *Xu Kecheng*

> **Comments from Tang Zhaoyou**
> The second part of Mr. Fu Daren's story above is more touching. People cannot live forever but can live longer. However, currently, cancer patients live longer lives but are often suffering miserably. It is very rare for a patient with terminal pancreatic cancer to live a long life with high quality. I cannot help but think of the role of comprehensive treatment. In my view, Mao Zedong's On Protracted War has three main points: First, treat guerrilla warfare as a strategic move and refrain from underestimating small-scale actions. Second, be flexible. Advance and retreat as required by the situation. It is not advisable to only advance and never retreat. Third, take the building of "base areas" seriously. Dealing with cancer requires strengthening the body so that sickness is kept away.

Hydrogen and Pancreatic Cancer: Miracle in Daren (3)

Urgent Call from Shanghai

On the morning of June 1, 2018 when I just arrived at the office, my phone rang. Mr. Lin Xinyong called from Shanghai, saying: "President Xu, this is highly urgent. Fu Daren went to Switzerland."

"What happened? He promised to come to Guangzhou in the near future. We are prepared for his arrival." I could not believe it. "He promised me to cancel the trip to Switzerland."

"He was under pressure from Taiwan media. The media and public opinions in Taiwan are as fierce as tigers." Mr. Lin told me in an angry and helpless voice.

In the past six months, Fu Daren petitioned to Taiwan leaders several times and required a euthanasia act in Taiwan. He said that he wanted to be the first to have euthanasia under the act in Taiwan. He was covered in Taiwanese newspapers and on television almost every day. It was saddening to read some media articles. One of the headlines was Where Are Fu Daren and Li Ao in Their Death Progress?". In the article, the author addressed to Li Ao: "Master Li, see you in heaven."

Flying to Switzerland Overnight to Save Him from Death

A few minutes later, Mr. Lin called me again and said, "I am checking the ticket and will fly to Switzerland immediately."

> *Picture 1. Seeing Mr. Lin Xinyong, who rushed to Zurich, Switzerland, Fu Daren said: "You are so kind. I can only pay you back in my next life."*

> *Picture 2. Xu Honghui is feeding water melon to Fu Daren, who then said: "You came a long distance from China on behalf of your father. I am happy in this life."*

In a few minutes, my son Xu Honghui called me from Shanghai and said, "I will fly to Switzerland together with Mr. Lin, on your behalf."

In the early morning the next day, I received the following WeChat message from Honghui:

> *"At 2pm on June 2, I arrived in Zurich, Switzerland. A little earlier, Mr. Lin's son flew from Birmingham, England to Switzerland to join us. We rented a car and arrived at our hotel at 4 o'clock. But where exactly can we find Fu Daren in Zurich? Mr. Lin called his immediate family members, only to find that their mobile phones were all turned off. We asked Fu Daren's friends and the Zurich Taiwanese Business Association. None of them knew where Fu Daren was staying. It was late at night. We had to wait to find other ways the next day. At dinner, the three of us were silent…"*

At 10:30 the next night, I was supposed to go to bed according to my daily routine, but was not sleepy at all. I felt that something was keeping me in suspense. Sitting in the study, I checked my mobile phone and surfed the Internet on my computer, searching for information about euthanasia in Switzerland and hoping to help Mr. Lin far away. Suddenly, the phone rang. It was Mr. Lin. He said: "We found Fu Daren and are negotiating with him."

Later, I learned that Mr. Lin found Fu Daren's Facebook account and saw a street view photo that he posted. It was a sign from God. According to the road sign in the posted photo, Mr. Lin found a Marriott Hotel near the street. He went to the hotel reception and asked. He was told that Fu Junhao, Fu Daren's son, and his mother Ms. Chen were staying in the hotel. Later, hearing that Mr. Lin and his party arrived, Fu Daren was shocked and burst into crying.

Mr. Lin said: "I just want to tell you first so you can sleep well." He continued with confidence: "Rest assured, President Xu, we have the confidence to bring him back."

I could not sleep well? I immediately sent the following letter that I had prepared to Fu Daren:

> *Daren, we miss you. Come back soon.*
> *Four years ago, I also tried to persuade a patient with terminal malignant pancreatic tumors. The patient was Xiaofeng, a 17-year-old beautiful girl. Her pancreatic tumors spread to the periphery of the pancreas and the liver, causing a significant decrease in the levels of various cells in the blood. With pain and bleeding and unable to eat, Xiaofeng tried to commit suicide several times. After learning that, I went with my colleagues to her home by car for 4 hours and convinced her to come to our hospital for treatment. Surprisingly, Xiaofeng's conditions improved significantly. Now, she still has the tumor in her body and lives with cancer. But she has almost no symptom. She is currently working at a company in Guangzhou.*
> *Daren, you are older than Xiaofeng. There are more people who love you and care about you. You have created many miracles: First, your pancreatic tumor has stabilized and shown no significant progression in ten months. More importantly, the liver metastasis found in a Taiwan hospital has disappeared, which is unlikely to occur in general cases of pancreatic cancer. Second, your abdominal pain and diarrhea were not caused by the pancreatic tumor. They were triggered by functional disorders of the intestines, which are controllable. In a sense, there is no pain that cannot be controlled. Third, you have a good appetite and want to eat and taste good food. This is the best sign. Patients with terminal pancreatic cancer barely want to eat any food.*
> *Da Ren brother, come back and come to Guangzhou or Shanghai. We will do our best to treat you. You will survive and live happily.*
> *From Xu Kecheng at 6am in Guangzhou on June 2, 2018.*

I also sent an English letter to inform the loving euthanasia clinic in Switzerland the following:

As the attending doctor who had treated Mr. Fu Daren, I can say that he is yet to be eligible for euthanasia and he can survive. I hoped the clinic to spare Fu Daren.

Determined to Leave

However, Fu Daren was determined to leave the world. He said that he missed the mainland and his hometown in Shandong. He missed Guangzhou and the happiest one month in Guangzhou. He knew that his pancreatic cancer would not kill him immediately, and that he could survive. But he had promised to the public to be the first example of euthanasia. He wanted to show the patients who rely on several "tubes" to maintain life that they have a choice to get rid of pain and die a comfortable death.

He sent the last Facebook post and expressed the last sentence to friends from all walks of life: "If you have me on your mind, death is not farewell." He also joked that he would continue to argue with Li Ao in heaven. He accepted the last interview with the media and said: "I got off the train at the last stop."

At 5:33 on the morning of June 7, I received the following WeChat message from Fu Daren:

> *"My dear brother, Prof. Xu Cheng, President Xu: You have tried your best. Your kindness has moved me, and I will not be able to pay you back even with three lives. Man is destined to die once. As the old saying goes, although you escort a guest a thousand miles, yet the parting must come at last. At the call from God, I will extinguish my labor on the earth, return to heaven, and rest in peace. Goodbye, my brother. I will bless you will peace, joy, good health, longevity."*

Seeing the farewell message, I felt desperate. I wondered whether Fu Daren was crying, laughing, silent or sending goodbye messages at this moment. I imagined that my son, on behalf of me, followed Mr. Lin to save Fu Daren, only to return without results. He must be feeling guilty and sighing.

Subconsciously, I turned on the phone and sent Fu Daren a WeChat message: "Brother, have a nice journey. I wish you happy in heaven."

> *Picture 3. Media report about "farewell" of Fu Daren*

Farewell in Taiwan

On June 22, my secretary Rita and I flew to Taipei to meet Mr. Lin Xinyong, who arrived earlier. Then, we went directly to Fu Daren mourning hall, which was not far from Taiwan TV Tower. We were received by Zheng Yi (wife of Fu Daren), Ms. Chen, and Fu Junhao and his wife. Zheng Yi was once a radio anchor. Despite her age at 80, she looked like a middle-aged woman in terms of looks, demeanor and way of speaking. Zheng Yi solemnly conveyed what Fu Daren told her before leaving the world: First, I want to thank Mr. Lin Xinyong and Dr. Xu Kecheng, the two important people in the last days of my life. They provided me with the magical natural hydrogen and gave me meticulous care. Second, be sure to show the farewell video to Lin Xinyong and Xu Kecheng.

> *Picture 4. One hour before the euthanasia in Switzerland, Fu Daren broadcasted an NBA event as an anchor (left) and then sang the last song with his family (right). From the right: Zheng Yi (Mrs. Fu), Fu Daren, Fu Junhao, Ms. Chen.*

> *Picture 5. Remembering Fu Daren's happy days in one month of stay in Guangzhou. Front left: Ms. Chen, Lei Qian (Fu Daren's stepdaughter and the president of the Taiwanese Women's Association), Lei Jun (Fu Daren's stepson and the director of the Hawaii Office in Taiwan), the author, Ms. Zheng Yi, and Mr. Lin Xinyong.*

It was a 15-minute-long video about the last family gathering before Fu Daren had euthanasia. In the video, Fu Daren first talked about his own rough life experience. Then, he gave a vivid live performance of NBA game broadcasting. After that, he asked the relatives around him to sing a farewell song together. Finally, he said: "Work hard to always move forward when you are young. Say goodbye happily when you are old. Farewell. I love you all. Goodbye."

According to Ms. Zheng Yi, Fu Daren in his last days had no sorrow, posted on Facebook every day, communicated with the media, and was in high spirits. He was grateful to God for letting him live 86 years, 6 months and 6 days. He said that he had finished the extra round of life's game with honors. Asking all those present for cooperation, he hosted his last show: a comfortable end of life. Considering that his father was a general killed in the Anti-Japanese War in World War II, Fu Daren scheduled the euthanasia on June 6, a day when Douglas MacArthur led the allied forces to land in Normandy. But due to certain reason of the Swiss institution, the euthanasia was postponed to the next day. At 4:15pm local time on June 7, Fu Daren entered the room for euthanasia and embarked on a journey towards a safe, natural and painless end of life that he chose.

"Until the end of life, he kept comforting us. To make it less hard for us, he specially took us to a park that morning." said Ms. Zheng, her eyes wet with tears.

Fu Daren's ashes were placed in Jinshan Christian Peace Park in the northern suburbs of Taipei. The urns, tombstones, and burial site were all designed and selected by Fu Daren himself. The Peace Park was the final resting place for many cultural celebrities in Taiwan. Fu Daren said that he wanted to be with his friends after death. He was happy with them even if they quarreled sometimes.

Zheng Yi gently pulled me to her side. Watching the cinerary casket for Fu Daren being sent to the grave by his son, I suddenly had a sense of chill and reminiscence. I thought of the scene at the dinner in Soluxe Hotel Guangzhou on April 5, more than two months ago. That day, Daren accepted the evaluation results from whole abdomen radiography. The results showed no evidence of metastasis. Daren was very excited. With tears in his eyes, he held my hand tightly and said: "This is predestination."

At the burial site, a band played a low-pitched song. But lingering in my ears was the farewell song that Daren sang before euthanasia in Switzerland…

> *The farther he goes, the longer his parting grief grow, endless as vernal river flows.*

A group of birds suddenly flew over above us. They were not crows but yellow-red birds. They were twittering as if wanting to remind people: Fu Daren, a master in his generation, will always be the person who sings.

— Xu Kecheng

> *Picture 6. Ms. Lei Qian, the stepdaughter of Fu Daren and president of the Taiwanese Women's Association, expressed her sincere gratitude to us for our hospitality to Fu Daren in Guangzhou. This is her photo with the author's secretary Lu Tianyu (Rita, left).*

> **Comments from Tang Zhaoyou**
> It is a pity that the pancreatic cancer patient voluntarily applied for euthanasia, because his disease had not developed into the final stage. His pancreatic cancer was still stable; and liver metastasis was not even visible on CT images. I admire him for the fearlessness and courage that he showed for his ideal. Since the onset, his pancreatic cancer lasted more

> than 20 months. During this period, he did not receive anti-cancer treatment and had only hydrogen inhalation. Whether the effect was achieved by hydrogen required further research. But it is affirmative that his tumor stabilized.
> The patient was a typical example of "living with cancer." It is surely the best to kill cancer. However, in reality, it is almost impossible to eradicate cancer cells. Therefore, cancer cells should be treated also with methods for transforming cancer cells, the microenvironment and the body. In many cases, pancreatic cancer cannot be surgically removed at the time of diagnosis. Therefore, the primary treatment strategy is to keep patients living with cancer. Hydrogen can be one of the measures to achieve this end.

Hydrogen Protects the Lung

"Magic Responder"

At the hydrogen experience center on the right side of the National Taiwan University Hospital on March 5, 2018, I met Mr. Zeng, 84. He was of medium build and had a little hunchback. His weather-worn face was covered with wrinkles, leaving a deep trace of time. His deep-set eyes looked sparkling. He was silent and sat quietly in the chair. I shook hands with him. He looked at me and appeared shy. He said, "I come here every day to inhale hydrogen. They are nice to me." He pointed at the service lady.

Mr. Zeng was accompanied by his son, who told us that his father had a hard life and had been a coal miner for decades. Local mines had bad ventilation facilities. Although these mines were all closed in 2000, many miners found themselves with diseases. And almost all of them had pneumoconiosis. With serious pneumoconiosis, Mr. Zeng was hospitalized several times a year.

"During the hospital stay in 2017, he almost died." Mr. Zeng's son said, "He has survived to this day because of hydrogen inhalation." He was a little emotional and took out Mr. Zeng's chest X-ray films.

I checked Mr. Zeng's chest films. In spite of their poor quality, the images clearly showed plaques of different sizes in the upper, middle and lower areas of both lungs. The middle and lower parts of the right lung displayed more plaques, whose density level was Grade 3. Mr. Zeng could be diagnosed with Stage III pneumoconiosis. His son said: "This is from a recent X-ray. It was much more serious before. Both his lungs were fully covered with plaques. It was so scary."

> *Picture 1. Mr. Zeng's X-ray chest radiography: Both lungs were covered with plaque-like inhomogeneous shadows, indicating lung inflammation and fibrosis.*

> *Picture 2. The author examined Mr. Zeng and did not hear rales from the lung.*

When Mr. Zeng was hospitalized in 2017, he was out of breath and could not lie down horizontally. He had a heartbeat rate at 140-160 beats/minutes and purple lips. Even if he inhaled a high concentration of oxygen, his blood oxygen saturation was only 90. Mr. Zeng was diagnosed with cardiopulmonary failure and was given a critically ill notice. "At that time, our family members did not have much hope. The sad thing is that my dad has suffered hardships for a lifetime and barely had the chance to live in comfort. I don't want to see him dying like this." Mr. Zeng's son said.

Despite being separated for nearly 70 years, both the mainland and Taiwan share the same custom: The elderly who are about to die hope to die at home. Mr. Zeng was taken out of the hospital by his family. Under the introduction of a friend, he came to the hydrogen experience center to inhale hydrogen. In addition to hydrogen, oxygen was inhaled actually. Mr. Zeng used a hydrogen-oxygen nebulizer, which functioned according to the following principle: Electrolytic effect occurs to pure water under the action of a direct-current electric field and a small amount of food-grade catalyst, producing H_2 and O_2 molecules, which overflow from the cathode and anode respectively. The inhaled gas includes 34% oxygen and 66% hydrogen.

In the clinical trials of drugs, none of the tested subjects show positive effects usually. As a result, the trials end in failure. Occasionally, one or two cases demonstrate magical effects and are known as "magic responders." Mr. Zeng was a "magic responder." He responded not to a certain drug, but to hydrogen, the most common substance in the world. After inhaling hydrogen and oxygen, he had less asthma, lower heartbeat rate, and stabilized lung lesions.

I am happy to see the "magic response", which is good for Mr. Zeng. Moreover, his case seems to be an additional example of hydrogen as an effective treatment means to us. In our hospital, patients with chemo drug-related lung damage are generally treated with steroid hormones, which have limited therapeutic effects but serious side effects. Can hydrogen inhalation serve as an alternative treatment for lung disease?

Miserable Patient from Indonesia

I wrote about the painful experience of an Indonesian patient in my book Practice Chinese Way of Cancer Control published a year ago. The story was as follows:

> *The patient, male, 51 years old, was diagnosed with right lung cancer with multiple metastases in the lungs and bones in a local hospital in August 2015. His pathology indicated adenocarcinoma. Gene EGFR19 exon showed mutation. The patient was given combined chemotherapy using pemetrexed and carboplatin for a total of 6 cycles (one cycle every 3 weeks). His tumor progressed. He was given oral iressa for treatment instead for 3 months. But his conditions did not improve. Later, he took tarceva for 2 months. With the tumor continuing to progress, the patient stopped tarceva and took AZD9291 (osimertinib, a third-generation EGFR mutant inhibitor) orally. Due to difficulty in breathing, the patient was admitted to our hospital in an emergency on August 20, 2016. Examination results showed that neck lymph nodes were enlarged and pressed against the trachea. Tracheotomy was performed on the patient. His CT scan showed diffuse ground glass-like and reticular nodules on both sides of the lung. The patient was considered to have pulmonary infection and interstitial pneumonia caused by targeted drugs. He was given anti-infection, methylprednisolone and other symptomatic treatments, after which the symptoms were slightly relieved. However, the patient still could not live without the breathing machine. Seeing me, the patient took my hand with emotion and asked the nurse to seal the tracheal cannula orifice with a cotton swab. Holding breath, he said one word at a time: "Thank you. I want to live…" I managed to appear relaxed and said a few comforting words to him. To comfort is usually the biggest aching helplessness for a doctor.*

The reason why cancer is refractory mainly lies in the drugs currently used for systemic control. The drugs, be them chemo drugs or hugely popular molecular targeted drugs, are ineffective in most cancers and fail to deliver lasting maintenance effects. In addition, the drugs induce serious side effects by killing cancer cells and causing secondary damage to the body at the same time. The Indonesian patient above used the most advanced third-generation targeted drug. However, he did not respond well and experienced interstitial pneumonia, a fatal lung damage, from using the drug.

In cancer treatment, the incidence of lung damage caused by chemo drugs is 0.03% to 3%. Almost all types of anti-tumor drugs have been reported to cause lung damage in clinical trials and clinical applications. The drugs include early drugs such as bleomycin and alkylating agents for lung cancer; and recently applied drugs such as gemcitabine and taxol. The incidence of bleomycin-induced lung injury is 6% to 10%. One study showed that among 141 patients with Hodgkin's lymphoma treated with bleomycin-combined regimens, the incidence of lung damage was 18%, and that of death was 24%.

Targeted drugs may also cause lung damage. Of the 1,976 non-small cell lung cancer patients using gefitinib from 84 institutions, 70 (3.5%) developed interstitial pneumonia and 31 (1.6%) died. All the patients had history of chest radiotherapy. Lung damage was more likely to occur to men and those with history of smoking and interstitial pneumonia. A researcher conducted a statistical study of 92,821 patients who were treated with gefitinib outside of Japan. The overall incidence of interstitial lung disease was around 0.28%. In the 65,527 patients who used gefitinib in Japan, the incidence was about 1.7%.

In addition to chemotherapy and molecular targeted drugs, immunological checkpoint inhibitors represented by PD1/PDL1, which is known as a "milestone treatment", may also cause lung damage.

Tears of an Elderly Man from Beijing

There are certain great coincidences in the world. Recently, I received a PD-L1-positive patient at the outpatient clinic. He had liver cancer and developed metastases after liver cancer surgery. Seeing that he was able to afford the cost, I recommended him opdivo or keytruda. But he refused, saying that the drug had too big side effects. An hour later, an elderly man from Beijing came to me in a hurry and handed me CT scan films. He told me that he came to ask for help for his son, who was 40 with a Ph.D. degree and had lung cancer. A week ago, the son received injection of Keytruda. Later, the experienced explosive aggravation of disease and respiratory failure. I checked the CT images and saw diffuse plaque-like heterogeneous shadows on both sides of the lungs, which were typical of interstitial pneumonia.

It reminded me of a paper published in the journal Lancet. Titled Increased Reporting of Fatal Immune Checkpoint Inhibitor-associated Myocarditis, the paper warned that there was currently no effective treatment and that the mortality rate was as high as 46%. While commenting on the paper, one expert mentioned that this type of drug causes five serious complications, including immune pneumonia.

Acute interstitial pneumonia is the most serious pulmonary complication of anticancer drugs. It is a diffuse pulmonary interstitial disease with highly poor prognosis and a mortality rate of 72%. The aforementioned Indonesian patient died in the hospital eventually. Sadly, he did not die of his lung cancer but died of interstitial pneumonia, a complication of the targeted drug he used. It is a big pity.

The elderly man held my hand tightly and said with tears: "I have only one child. I beg you to save my son with no matter what method." I saw a copy of the doctor's medical order from the Beijing hospital where his son was treated. The medical order included administration of antibiotics, dexamethasone and prednisolone in large doses. Besides, the elderly man's son was already on breathing machine.

What panacea could I offer? I thought of Mr. Zeng, an old miner I visited in Taiwan. He suffered from pneumoconiosis, not lung cancer. But it was pneumoconiosis, an interstitial lung disease, that caused heart and lung failures to him. Nevertheless, he was eventually saved through hydrogen inhalation.

The elderly man seemed to be no older than me. Looking at his unkempt gray hair, deep-carved wrinkles on the face, and tired eyes with tears, I felt ache pounding my heart. We came across each other with no care of acquaintance or not. As a survivor who suffered from cancer, I found myself seeing the elderly man as a long-term friend in spite of our short meeting and my profession as a doctor. I took the elderly man by the hand and said, "Try hydrogen inhalation, maybe…"

Although I knew that my suggestion was not possible to be executed on his son who was far away in Beijing, I still could not help but made the suggestion. Moreover, I asked my secretary to take him to lunch and then experience hydrogen inhalation in my studio.

Enlightenment from Animal Experiments

The mechanism of action behind lung damage caused by chemotherapy and targeted drugs is unknown. One possibility is that the drugs cause direct damage to the alveolar Type I and Type II epithelium. Peroxidative free radicals are involved in this process.

Free radicals are generated endogenously and exogenously. Exogenous production of free radicals is induced by external factors, such as smoking, toxins, pollution, excessive exercise, radiation, and drugs. Both chemotherapeutic drugs and molecular targeted drugs facilitate the production of peroxidic free radicals in the body.

Free radicals are generally rapidly cleared from the body. If excessive free radicals are generated, or/and the antioxidant mechanism is insufficient, free radicals chemically react with DNA, proteins, lipids, and carbohydrates, damaging life molecules and causing cell damage.

Does hydrogen have therapeutic effects on drug-induced lung injury? Let's look at three animal experiments as follows:

Experiment 1: The experiment was designed to study the effects of hydrogen inhalation on experimental acute lung injury. Acute lung injury was made through intratracheal infusion of lipopolysaccharide (LPS). Sixty-four mice were randomly divided into 4 groups. Hydrogen inhalation started 1 hour and 6 hours respectively after the start of the experiment and lasted for 1 hour. All the mice that received intratracheal perfusion of lipopolysaccharide had acute lung injury. However, the animal group with hydrogen inhalation showed mild lung injury. Their manifestations included decreased alveolar epithelium apoptosis; less neutrophil aggregation and inflammation in the lungs; decline in the levels of some pro-inflammatory cytokines such as tumor necrosis factors and macrophage inflammatory proteins; and reduced interleukins 1 and 6. (From an article published in Shock, 2012, Vol. 37, No. 5).

Experiment 2: The experiment was designed to study the effects of drinking hydrogen water on experimental lung damage. According to findings of the experiment, the PQ + hydrogen group had markedly lighter lung injury than the PQ group, with less pleural effusion; less protein content and cells in airway secretions; and less histological changes in the lung. The animals given hydrogen water showed less pulmonary edema. And their indicators of cell lipid peroxidation and oxidative damage were also at significantly lower levels than those of the experimental group without drinking hydrogen water. The authors also measured the apoptotic

index and found that lung epithelial apoptosis was significantly reduced in the hydrogen water group. (From an article published in the Journal of Biomedicine and Biotechnology, 2011)

> *Picture 3. The picture shows that oxygen free radicals cause pulmonary inflammation and fibrosis through a variety of pathways, including by disrupting the balance between different genes; interfering with specific proteases and damaging DNA; and promoting alveolar epithelial cell apoptosis. p38MAPK indicates p38 mitogen-activated protein kinase; Smads indicates a substance targeted at special genes; FasL indicates Fas ligand; Fas indicates apoptosis-mediated surface antigen; FADD indicates Fas-related death domain; BAX indicates BCL-2-related protein; BCL-2 indicates B cell lymphoma.*

Experiment 3: The experiment was designed to examine the effects of hydrogen inhalation on mouse models of smoking-induced chronic obstructive pulmonary disease (COPD). The results showed that hydrogen inhalation significantly decreased inflammatory cells, α tumor necrosis factor, matrix metalloproteinase-12, caspase-3 and caspase-8 in bronchoalveolar lavage fluid; and reduced the expressions of interleukins 6, 17 and 23; but increased metalloproteinase-1 tissue inhibitors. The experiment also demonstrated that hydrogen inhalation improved lung pathology, lung functions and cardiovascular functions; and reduced the hypertrophy index for the right ventricle. Inhalation of 22% and 41.6% hydrogen achieved better results than that of 2% hydrogen.

(From an article published in the International Journal of Chronic Obstructive Pulmonary Disease, 2017)

Afterword

Mr. Zeng's experience and the above three experiments highly indicate that hydrogen protects the lung and relieves and prevents lung damage. The protective effects of hydrogen on the lung are characterized as follows:

First, hydrogen has a small molecular weight and easily penetrates the biofilm into the cytoplasm, mitochondria, and nuclei. The lung is the organ closest to external environment. And hydrogen can easily enter the lung.

Second, hydrogen molecules selectively react with free radical hydroxides and peroxynitrites without reducing other reactive oxygen species (such as O_2) associated with cellular signaling. Therefore, it does not affect normal metabolic oxidation-reduction reaction in the body or affect the gas exchange of the lung itself.

Third, hydrogen molecules have higher histocompatibility than other antioxidants, which is especially important for delicate tissues like lung tissues.

Fourth, hydrogen is highly safe.

Fifth, the lung has abundant blood flow. Regardless of being administered by inhalation or injection, hydrogen can quickly enter the whole body through lung tissues, which is very helpful for improving systemic conditions.

The above functions of hydrogen suggest that hydrogen removes or reduces the side effects of chemotherapy and targeted drugs, therefore playing a safeguarding role in mainstream treatments.

— *Xu Kecheng*

Comments from Tang Zhaoyou

> Reading about the protective effects of hydrogen on the lung, I have thoughts thronging my mind. Twenty years ago, my wife, who advocated "west learning from the east", applied Chinese medicine to treating a postoperative patient with early to mid-term breast cancer. The effect was good. However, since the medical cost of the Chinese medicine could not be reimbursed according to the insurance policy then, the patient went back to the hospital for "standard" chemotherapy. Three years later, we were surprised to learn that the patient died of pulmonary fibrosis as a result of chemotherapy. In 2016, The New England Journal of Medicine published an article titled Cardiovascular Toxic Effects of Targeted Cancer Therapies. My wife was deeply afflicted with the cardiovascular toxic effects described in the article during breast cancer treatment. Chemotherapy should be recognized for both its good points and shortcomings. Targeted therapy is no exception. Pointing out the negative points of a therapy is not for undercutting therapy but for strengthening it. Now that hydrogen is found to be protective of the lung. Isn't this a good example that gives us clues to strengthen therapeutic effects with cancer transformation methods?

Extra: Langshan in Nantong

> *(Picture by Xu Kangning)*

Langshan (or Wolf Hill) is a. hill in Nantong, Jiangsu Province. Wang Anshi, a poet of the Song Dynasty, dedicated a poem to Langshan as follows: "Who broke through the Kunlun Mountain thousands of miles away, leading to boundless waves rushing down from the sky? In the morning of spring on top of the mountain, clouds and fog spread, connecting islands. After warmth sets in, the spring thunder moves, waking up the dormant fish and dragons. Where to find the immortal in this fairyland on earth? When will the messenger come back from heaven by raft? I have traveled halfway in the rivers and lakes. Only today I catch such an eye-opening view." On the north bank of the vast and boundless Yangtze River, Langshan, a 100-meter-high hill, appears exquisite, beautiful and unique. It has the reputation as the "No.1 mountain in the rivers and seas". Zhang Jian, a great figure with remarkable achievements in modern Chinese history, built Nantong at the foot of Langshan Mountain. His spirit that "I will try to achieve success but need not to get the credit." has motivated countless students in Nantong.

Langshan in Nantong: A Dialogue with the Deity

Zhong Zimei

> Because of you, the deity;
> Langshan became the first mountain in the mountains and seas;
> The mountain is sacred because of you;
> Your reputation spreads far because of the mountain;
> I came because of you;
> I came out of confusion;
> I came, hoping to unfold the mystery of life;
> A pilgrim with poetic quality;
> I am just as common as Langshan bandits;
> Except that tens of thousands of sick people are on my mind;
> Except that I have their sufferings and pain on my shoulder;
> To dispel clouds and give sunshine back to them;
> To lead them to the spring flower field;
> God offers me the altar;
> *Encountering your poems—*

> **A healthy** physical and mental encounter.

PART 6 — Hydrogen Cancer Control in the "Third Eye"

Surprise: Her indicators Returned to Normal 1 Month After Hydrogen Inhalation – A Recurrent Cancer Patient with Multiple Metastases

Ms. Quan, a 60-year-old teacher, had ovarian cancer recurrence with multiple metastases. Her tumor marker CA125 became normal after chemotherapy interruption and regular hydrogen inhalation for a month. On the afternoon of August 6, 2018, a series of lectures themed Hydrogen Molecule Medicine and Health hosted by Xu Kecheng Care for Health Studio were taking place. At the event, Ms. Quan shared her gratifying therapeutic effects with the audience. Her radiant and energetic appearance left a deep impression on the people present.

Ms. Quan is very idle. Her children studied and settled down in the UK. Ms. Quan is also very busy. In addition to inhaling hydrogen every day in Xu Kecheng Care for Health Studio, she often happily shares her positive changes and feelings after hydrogen inhalation with other experiencers. Nowadays, she has become the studio's "hydrogen messenger" who promotes the concept of hydrogen and health. She is living an enriching and happy life. But when talking about her disease 5 years ago, Ms. Quan still has a lingering fear. In October 2013, Ms. Quan had bloating, abdominal distension, systemic weakness, short of breath during walking, and a big belly. She went to a hospital for routine examination, but no pathogen was detected. She was told by the doctor that she gained weight and should do more exercise to lose weight.

After a while, Ms. Quan's symptoms persisted. Her belly grew bigger with each passing day. She looked like a woman pregnant for 7 or 8 months. Feeling strange, she went to the hospital again for examination and was found to have a large amount of fluid in the abdominal cavity. Her tumor marker CA125 was high and doubled, reaching 2000 at the highest level.

After being diagnosed with Stage IIIc ovarian cancer with metastasis, Ms. Quan could not sleep and lamented that life was not fair to her. Two days later, she gradually let go of all bad thoughts and started to actively seek treatment.

Ms. Quan took four courses of chemotherapy successively. After chemotherapy, she received an operation that completely removed the uterus, ovaries and fallopian tubes. Thereafter, she had a follow-up examination once every 3 months. All indicators were found to be normal. At the beginning of 2017, her CA125 began to rise again; and she had waist pain. She took a CT scan, which revealed that her cancer relapsed and abdominal cavity was full of tumors. After tumor reductive surgery twice, Ms. Quan took chemotherapy. She said: "I had chemotherapy 5 times. Every time after chemotherapy, my white blood cells decreased to around 1; absolute neutrophils dropped 0; and platelets fell to around 20. That means, every chemotherapy means a brush past death for me."

Chemotherapy failed to control Ms. Quan's disease. Her CA125 did not decline significantly during the first five sessions of chemotherapy. At the same time, she developed herpes zoster due to low immunity caused by chemotherapy.

"The doctor said that I was resistant to chemotherapy drugs and wanted to switch the drugs. Then, I heard that Xu Kecheng Care for Health Studio offered hydrogen inhalation for free.

The selective oxidation resistance of hydrogen removes redundant and harmful free radicals; and helps control cancer." Later, Ms. Quan started hydrogen inhalation on June 30, 2018. Five days later, she took a blood test for tumor marker examination. It was found that her tumor marker decreased by more than 70 points, which was almost 50%.

Ms. Quan was surprised with joy; and made a bold decision to stop chemotherapy and continue to inhale hydrogen for a month to check whether her cancer can be controlled.

On August 6, Ms. Quan got up early and went to the hospital for a blood test. Results showed that her CA125 decreased more than 50 points to the normal range. She was so excited that she immediately shared the good news in the hydrogen service WeChat group for the studio. Other members of the group were delighted to hear the news and were sincerely happy for Ms. Quan.

"Hydrogen inhalation gave me a big surprise. I used to feel tired after walking a dozen meters. After hydrogen inhalation, my conditions became better with each passing day. Half a month after hydrogen inhalation, I walked several kilometers without feeling tired. Recently, I went to the UK for my son's graduation ceremony. It was a long flight, but I did not feel tired at all."

Since then, Ms. Quan kept inhaling hydrogen. Meanwhile, she always enjoyed sharing her personal experience with other cancer patients and explaining them the effects of hydrogen molecules on cancer control. She became a special volunteer for Xu Kecheng Care for Health Studio.

Ms. Quan: Best Gift of Teacher's Day

Ms. Quan had Stage IIIc ovarian cancer. After the cancer recurred, she inhaled hydrogen for 5 days. Her CA125 (related tumor marker) decreased by almost 50%. After one month of hydrogen inhalation, her CA125 dropped to the normal level of 29.41. On September 10, 2018, two months after hydrogen inhalation, the CA125 declined to 17.22, which was the lowest level for Ms. Quan in the past 5 years of disease. She said that the result was the best Teacher's Day gift for her as a teacher.

On September 10, 2018, Ms. Quan shared the good news in the hydrogen inhalation experience group on WeChat. She shared all the test results before and after hydrogen inhalation (see Pictures 1 to 4), and recounted her story as follows:

> *When my cancer was first discovered in December 2013, I already had ascites and sesame-sized metastases in the entire abdominal cavity and peritoneum. The normal level of CA125 is 35. But my CA125 level at that time reached 2252. I received the first exploration operation on December 25, 2013, followed by chemotherapy 4 times. In June 2014, I had cytoreduction, followed by another four sessions of chemotherapy. My CA125 returned to a normal level in January 2015.*
> *But 2 years later, at the beginning of 2017, I felt unwell. My tumor marker, which was examined once very 3 months, went up gradually. I took a CT scan. The findings showed metastases of different sizes in the S7 of the liver and abdominal cavity. The tumors were observed until December 27, 2017 when I received laparotomy and cytoreduction again. It was exactly 4 years after my first exploratory operation.*
> *Why was the operation called cytoreduction every time? Since my tumors had spread to the entire abdomen, the doctor could only remove the tumors visible to the naked eye and had no way to clear the invisible ones. After another cytoreduction, I was given chemotherapy again. Since I had already received chemotherapy 8 times, my body was greatly harmed; and the subsequent chemotherapy was unbearable for me. Each time after chemotherapy, my white blood cells decreased to a little more than 1; absolute neutrophil dropped to 0; and*

> *platelets declined to over 20. That means I almost had no immune function. Any accidental infection might kill me. A blood platelet level below 10 might cause massive haemorrhage and even death. Therefore, every time I had chemotherapy, I passed by death. On June 4, 2018, I received the fifth chemotherapy. Due to low immunity, I developed herpes zoster unfortunately.*
> *With what I had been through, I was very afraid of chemotherapy. By accident, I heard that Xu Kecheng Care for Health Studio offered hydrogen inhalation for free. So, I went to the studio to have an experience on June 30, 2018. Five days later, I had a tumor marker test. I was surprised to see the results: CA125 decreased by more than 70 points and was almost 50% lower than the initial level. This gave me great hope, because every session of my previous chemotherapy failed to effectively reduce the tumor marker. Probably, I had drug resistance. In addition, I had fear for chemotherapy. All this made me decide to stop chemotherapy and continue with hydrogen inhalation for one month to check whether it would be effective.*
> *On August 6, which was one month after hydrogen inhalation, I went to the hospital with a restless mood for a blood test early in the morning. I waited until 12 noon time when the results came out: CA125 dropped by more than 50 points and was at a normal level. I could not help but thinking that hydrogen inhalation was miraculous. On September 7, after hydrogen inhalation for one month, I went for a follow-up examination. The results showed: CA125 (normal level: 35) decreased from the previous 29.41 to 17.22, which was the lowest level in the past 5 years of my disease. The tumor marker decline testified to the magical effects of hydrogen inhalation.*
> *Since I inhaled hydrogen, I felt increasingly better mentally. Originally, I had very weak physical strength. Every time I saw a doctor, the doctor asked me what was wrong with me. Every time I answered: "I have no strength, even for walking and going up and down the stairs." But now, I walk faster than young people without panting for breath.*
> *As I said, the 5-year survival rate of patients with Stage 3 and 4 ovarian cancer is only 20% to 30%. December 2018 will be exactly 5 years since I had cancer. Can I make it to beyond 5 years? I inhale hydrogen every day. It depends on whether hydrogen inhalation will show a magic effect again and help me pass the milestone year. Good luck to me.*

— Lin Hong

> *Picture 1. Test results on May 23, 2018: Before hydrogen inhalation, CA125 level was 163.*

> *Picture 2. Test results on July 5, 2018: Five days after hydrogen inhalation, CA125 level was 85.63.*

> *Picture 3. Test results on August 6, 2018: One month after hydrogen inhalation, CA125 level was 29.41, which was in the normal range.*

> *Picture 4. Test results on September 7, 2018: Two months after hydrogen inhalation, CA125 level was 17.22, which was the lowest level since patient was diagnosed.*

> **Comments from Sun Xuejun**
> I have met Ms. Quan. Truly, she does not look like a typical patient who has been through multiple chemotherapy sessions. Her disease is highly relevant to the index CA125, which was at the level of 2000 before surgery. After surgical resection, the index returned to normal. After cancer recurrence, her CA125 rose again, and did not drop to a normal level after multiple chemotherapy sessions. After a short period of hydrogen inhalation, her CA125 unexpectedly decreased rapidly. This undoubtedly can be attributed to the effects of hydrogen.
> So far, there has been no conclusive assertion that hydrogen controls tumors. At present, it is difficult to analyze and judge its effect in a rigorous and accurate way. Based on my understanding of the biological effects of hydrogen and knowledge of Ms. Quan's

> conditions, I personally believe that hydrogen did exert positive effects on her. However, the effects are still short after all. Long-term effects must be observed. Surely, I hope that she will recover and restore good health. The effects of hydrogen shown in Ms. Quan were achieved probably by the following mechanism: After inhalation of a large amount of hydrogen, the antioxidative and anti-inflammatory effects of hydrogen help repair immune function deterioration caused by chemotherapy and tumor factors. The strong immune function effectively attacks tumor cells, keeping Ms. Quan in relatively good health. It cannot be ruled out that hydrogen is directly involved in the anti-tumor effect. Of course, this needs to be verified with more research in the future.

"Hydrogen Sister" with Controlling Cancer Walks 20,000 Steps a Day Easily

Ms. Quan, whose cancer recurred with metastases after ovarian cancer surgery, started to inhale hydrogen at the end of June 2018. One month later, her tumor index became normal. Two months after hydrogen inhalation, her tumor index dropped again, which was very encouraging. Her story was spread as a good example among cancer patients. And she became known as the "hydrogen sister". Five months have passed. What is Ms. Quan's condition now? Is her tumor marker normal?

On December 1, 2018, Ms. Quan showed up in Wuyi Mountain, radiating with vigor. She was there to participate in a celebration event for cancer survivors across the country. At the event, Ms. Quan completed the 2.5km journey under the program Walk with Thousands of People. She walked additional steps by herself. At the end of the day, she walked nearly 20,000 steps a day, an action that told all those who cared about her that she was very well.

Brisk Walk in Wuyi Mountain

On the afternoon of November 30, 2018, Ms. Quan arrived at Wuyishan Airport. Stepping out of the cabin, she saw blue sky and green hills, with fresh wind brushing the face. She felt as if it was spring time with twittering birds and fragrant flowers. "This is truly a natural oxygen bar." She was excited and took a picture in front of the airport terminal.

According to Ms. Quan, she went to a hospital for magnetic resonance (MRI) and blood test after returning from a family visit in the U.K. in middle November. Blood test results showed that the tumor marker remained within the normal range. Surprisingly, MRI findings indicated that the suspicious liver metastasis as detected in July MRI had disappeared.

On the trip to Wuyi Mountain this time, Ms. Quan felt particularly relaxed. She believed that hydrogen gave her such good mood and good state.

The Wuyishan Square on December 1 was very lively. The opening ceremony of My Exercise, My Health, Meet at the 2022 Beijing Winter Olympics for the first China Wuyishan Health, Fitness and Tourism Season was held on Wuyishan Square. Nearly 1,000 members of cancer rehabilitation associations from all over the country gathered together for the event.

At the event, Ms. Quan was busy greeting old friends and meeting new friends. "So, you are the 'hydrogen sister'." Many people had heard about Ms. Quan's cancer recovery and were happy to meet her in person.

After the opening ceremony, the program Walk with Thousands of People was initiated. Prof. Xu and Ms. Quan, hand in hand, strode forward in high spirits.

They also happened to meet Miaoxuan from Shenzhen Light of Life branch. Miao Xuan suffered from cholangiocarcinoma 7 years ago. She received three operations successively, followed by chemotherapy. After chemotherapy, her cancer recurred. She was given another chemotherapy, after which the cancer relapsed again. Miaoxuan was miserable beyond words. With the help of Prof. Xu Kecheng, she had a series of rehabilitation treatments. "In the past two years, my cancer indicators are in normal ranges. My disease is very well controlled."

Prof. Xu Kecheng said that both Ms. Quan and Miaoxuan are beneficiaries of Chinese-style cancer control. He is very pleased to see that both of them are in good conditions now.

While walking, everyone talked and laughed. Soon, they completed the 2.5km journey without even realizing it. Feeling happy, Ms. Quan decided to continue with the walk. While breathing fresh air, she enjoyed the unique natural beauty of Wuyi Mountain and tasted the famous local black tea and food. On that day, she walked nearly 20,000 steps. She attributed her physical fitness to hydrogen inhalation.

A Shining Star In and Outside the Conference Hall

Academician Tang Zhaoyou, a distinguished oncologist, proposed the concept "Chinese way of cancer control", which calls for combined use of cancer elimination and transformation treatments. As a student of academician Tang, Prof. Xu Kecheng has been practicing the concept in clinical practice.

"Ms. Quan is a beneficiary of the concept. Ms. Quan, could you please stand up?"

Prof. Xu Kecheng pointed at Ms. Quan's direction in the audience. Then he introduced how Ms. Quan suffered terribly from chemotherapy but the tumor marker showed no significant decrease; and how her tumor marker returned to a normal level after hydrogen inhalation.

Ms. Quan, glowing with vigor, stood up and waved to the audience as self-introduction. She received a warm applause.

Everyone showed great interest in Ms. Quan's experience of hydrogen cancer control. Before the forum concluded, many cancer patients present moved to where Ms. Quan was seated, asking her questions and asking for her WeChat account. Ms. Quan patiently answered the questions one by one and set up a hydrogen inhalation WeChat group on the spot. That night, a patient took taxi to the hotel where Ms. Quan stayed, asking for advice on cancer treatment. Ms. Quan was busy but happy.

Ms. Quan revealed that her daughter would have annual vocation in December and insisted on taking her to Thailand for travel. She appreciated her daughter's offer but found herself in a dilemma. On the one hand, she would like to travel outside considering that her disease is well controlled and physical conditions are good. On the other hand, she cannot live without hydrogen inhalation. If she travels to Thailand, she might have to stop inhaling hydrogen for a while.

— Lin Hong

Good Luck or Hydrogen? Another Ovarian Cancer Patient with Normal Tumor Marker After Hydrogen Inhalation

On October 8, 2018, Pan received test results from the hospital. That moment was probably the happiest one for her in the past 7 years of fight with cancer. In the 7 years, she experienced cancer recurrence 3 times, received two major operations, and took 24 sessions of chemotherapy.

Her tumor marker level was like a roller coaster. It dropped immediately after chemotherapy and went up rapidly soon. Without knowing what to do, she resorted to hydrogen. Two months after inhaling hydrogen, Pan's tumor marker returned to a normal level and remained stable.

One day in December 2010, Pan received a call from the street management office: "It's the end of the year. Have you done gynecological examination this year?" Pan answered: "I forgot. I will do it tomorrow."

The next day, Pan hurried to Haizhu Maternal and Child Health Hospital and was given B-ultrasound. "Perhaps because it was a weekend, there were not many patients. The doctor checked very carefully. Suddenly, she said seriously: 'Something is wrong. There is a mass in the ovary. You need further examination'."

Then, she was found to have Stage III ovarian cancer. It was so shocking for Pan that she was not psychologically prepared at all. In January 2011, she underwent surgery that completely removed both ovaries. Then, she took six chemotherapy sessions. In the following four years, she had regular checkups every year. Nothing particular was detected.

In January 2015, Pan felt that her private parts were sticky. She went to the hospital for examination. The results indicated tumor marker increase. She had further examination in a cancer hospital and was diagnosed with ovarian cancer recurrence. Afterwards, she received six courses of chemotherapy in total for a period of six months. In July 2016, one year after the end of the last chemotherapy, the cancer relapsed again. Pan had another six courses of chemotherapy. In August 2017, around one year after the previous chemotherapy, cancer recurred to Pan for a third time. This time, the doctor recommended 3 sessions of chemotherapy to reduce the tumor and then surgery. On March 12, 2018, Pan underwent surgery again, followed by 3 sessions of chemotherapy.

In the last 7 years, Pan experienced cancer recurrence 3 times, underwent two major operations, and received chemotherapy 24 times successively in total. Cancer occurred to her again and again, which was devastating to her both physically and mentally. Speaking of her tough history of fighting cancer, Pan was filled with emotions.

But the restless cancer cells in Pan's body did not let go of her. In April this year, Pan took an examination in the hospital. Results showed that the tumor marker CA153 increased to 28.23 (normal level: 0 to 25), indicating the presence of cancer or precancerous lesion. None of the signs was good.

Should Pan continue with chemotherapy? She had suffered enough from chemotherapy over the years. But what other options did she have except another chemotherapy? She could not find a way out.

> *Picture 1. Before hydrogen inhalation, the level of tumor marker CA153 was 28.23.*

> *Picture 2. One month after hydrogen inhalation, the level of tumor marker CA153 dropped to 25.39, which was close to a normal level.*

> *Picture 2. Two months after hydrogen inhalation, the level of tumor marker CA153 fell to 21.38, which was within the normal range.*

"Go to Prof. Xu Kecheng's health studio to try hydrogen." Ms. Quan, an ovarian cancer patient who had free experience in the studio, suggested to Pan. As a patient with postoperative ovarian cancer recurrence, Ms. Quan inhaled hydrogen for a month in Xu Kecheng Care for Health Studio. After that, she took an examination; and results showed that her tumor marker became normal. She continued with hydrogen inhalation. And her tumor marker showed further decline.

Wanting to give it a try, Pan inhaled hydrogen in the studio every day starting from July 22, 2018. On September 7, she went to a hospital for examination. Results showed that the tumor marker CA153 dropped to 25.39, which was almost a normal level. During the period, Pan did not receive any other treatment. Was it because of good luck or hydrogen?

She decided to continue to inhale hydrogen for one more month to see if hydrogen was really that magical. On October 8, she obtained the latest examination result: 21.38. The level was finally normal. Pan was overwhelmed with joy.

Pan said that in addition to the unexpectedly good blood test results, hydrogen also brought her many delighting changes: a) Good sleep till dawn; b) good appetite for any food; c) Good physical strength, with the ability to walk tens of thousands of steps every day. She said that she does not feel tired after walking in Xiaogang Park for an hour and a half every day.

— Lin Hong

> *Picture 4. Comparison of experimental results*

Hydrogen has a miraculous effect on ovarian cancer?

Ovarian malignant tumor is one of the common malignant tumors of female reproductive organs. Due to the hidden location of the disease and absence of apparent early symptoms, patients are usually in terminal stage of cancer at the time when they start seeking treatment. Although the treatment of ovarian cancer has witnessed great progress in recent years, the overall cure rate is only around 30%. Therefore, it is extremely urgent to develop effective drugs for ovarian cancer. Prof. Ma Xuemei from Beijing University of Technology published the paper Therapeutic Potential of Molecular Hydrogen in Ovarian Cancer in the journal Translational Cancer Research in July 2018.

The paper pointed out that in mouse models of ovarian cancer, the mice with continuous breathing of hydrogen for 6 weeks showed significant decrease in tumor volume (32.3%) and considerable decline in the expression of Ki67 (30.0%) and CD34 (74.0%).

In vitro cell experiments have demonstrated that hydrogen molecules significantly inhibit the proliferation, infiltration, migration and clone formation of ovarian cancer cells; and weaken the cells' capability of tumor ball formation. Experimental results suggest that hydrogen molecules can play an anti-cancer role as potential drug molecules for treating ovarian cancer.

How to Alleviate the Sequelae of Radiotherapy? Answer from a Patient with Nasopharynx Cancer: Hydrogen Inhalation

Radiotherapy is currently the most effective treatment for nasopharynx cancer. However, effective treatment is often followed by different levels of radiotherapy sequelae. In serious cases, patients suffer from significantly lower quality of life and tremendous misery. Zhou was one of the patients.

Zhou, 64, worked at medical equipment company in Guangdong before retirement. One day in December 2001, her nose was bleeding. It happened that she had to go to Guangdong Hospital of Traditional Chinese Medicine due to the requirement of work that day. Her colleagues kindly reminded her to do a physical examination while they were at the hospital.

Zhou's examination results indicated stage I nasopharynx cancer. She was admitted to the hospital shortly and received three courses radiotherapy for a total of 36 times successively. After that, she had a follow-up examination every year. No tumor was detected. Zhou went on about her life as usual and gradually walked out of the shadow of cancer.

In the tenth year after cancer was detected, something unexpected happened. Zhou began to have rotten teeth. Then, her lingual base gradually atrophied, resulting in unclear articulation, hearing loss, swallowing difficulty, and impaired sense of smell. The doctor told Zhou that those were the sequelae of radiotherapy for nasopharynx cancer. There was no particularly good treatment available. The symptoms could be controlled only through massage and rehabilitation exercises.

"The pain after radiotherapy is far more afflicting than cancer itself." said Zhou, with tears running down her face. In spite of efforts to keep rehabilitation exercises, Zhou's symptoms were not alleviated. Because of hindered speech, Zhou became a Grade 1 language disabled person. Her hearing continued to deteriorate, with pus often discharged from the left ear. With increasingly weaker hearing, Zhou was unable to hear anything eventually.

Fortunately, Zhou's willpower was not undermined by the disease. She was very passionate about calligraphy and painting. In spite of strong pain, she often went to nursing homes to perform painting and calligraphy for the elderly, and voluntarily taught painting to children in remote schools. As the person in charge of nasopharynx disease rehabilitation work of Guangdong Light of Life Cancer Rehabilitation Association, Zhou was active in organizing and participating in various activities of the Association and helping other patients.

In the eyes of outsiders, Zhou was strong and optimistic, with a passion for charity activities. What others did not know was that the aftereffects of radiotherapy never receded because of Zhou's strong willpower. Trying to look for effective treatments, Zhou had tried acupuncture, physiotherapy and Chinese herbal medicine, but to little avail.

On October 18, 2018, she came to Xu Kecheng Care for Health Studio. She had hydrogen inhalation for free for four consecutive days. The result was surprising. "At the turn of seasons when there was temperature difference in daytime in the past, I always had nasal obstruction and tinnitus. After a few days of hydrogen inhalation, the symptoms were much relieved even when I went from outside to a place with air-conditioning. Besides, less pus came out of my ear."

Therefore, Zhou canceled all her social activities, and dedicated her time to hydrogen inhalation in the studio. A month later, she found that her ear was without any pus at all; nose breathing became smooth; and quality of life improved greatly.

> *Picture Zhou gave Prof. Xu Kecheng two folding fans with her handwriting.*

A few days ago, she made a special trip to send her calligraphy and painting works to the studio as a way of expressing her gratitude to Prof. Xu Kecheng and the studio staff. She said: "Prof. Xu Kecheng is the president of our Light of Life association. Influenced by his great benevolence, I learned to see life with the right attitude. The hydrogen cancer control he has been promoting has benefited me immensely. I am really grateful to him."

— Lin Hong

Clinical studies of hydrogen on reducing the side effects of radiotherapy

Prof. Xu Kecheng often says that rehabilitation is supplementary to the mainstream treatments of cancer. The purpose of cancer treatment is to enable patients not only to survive but also to live with good quality. Take Ms. Zhou for example. She suffered tremendously from the complications of radiotherapy. However, the late complications of radiotherapy are not artificial, but are determined by the inherent characteristics of radiotherapy. Therefore, patients cannot complain but have to bear it.

Despite the numerous achievements of modern medical research, complications, which are seemingly simple, generally can only be alleviated for a while with symptomatic treatment. Ms. Zhou had severe radiotherapy-induced complications, which were significantly relieved after hydrogen inhalation. Perhaps, she never expected that, nor did the doctor.

There have been clinical studies of hydrogen in reducing the side effects of radiotherapy and chemotherapy. In 2011, a clinical study from the University of Pittsburgh provided evidence. In the study, liver cancer patients were given radiotherapy and meanwhile, inhaled hydrogen for 6 weeks. The results showed that patients taking hydrogen had markedly higher quality of life scores during radiotherapy than the placebo group without hydrogen intake. There was no difference in the effect of radiotherapy between the two groups. The patients with hydrogen intake had less reactive oxygen products in the blood than in the placebo group.

Prof. Sun Xuejun from Shanghai Second Military Medical University and other scholars found that hydrogen can reduce animal mortality caused by radiation. Of the mice receiving lethal doses of radiation, 90% died two weeks later. But of the mice with both lethal doses of radiation and hydrogen exposure, only 20% died after 2 weeks. The difference between the two groups was significant. The authors believe that hydrogen protects animals against overall damage caused by radiation.

According to Prof. Xu Kecheng, the fact that Ms. Zhou's complications from radiotherapy were relieved after hydrogen inhalation can be explained theoretically.

The mechanisms of cancer radiotherapy are as follows: First, radioactive rays work directly on the DNA molecular chain, causing single-strand and double-strand breaks. Second, radioactive rays exert ionization effects on water molecules, producing reactive oxygen free radicals. Free radicals interact with the biological macromolecule (RH) and then act on DNA chains. In other words, free radicals can kill cancer cells. But radiotherapy does not target only at cancer tissues. It almost always causes damage to adjacent normal tissues, especially in

the case of nasopharynx cancer that involves complex local tissue structure. For this reason, collateral damage is inevitable. In the tenth year after radiotherapy, Ms. Zhou had rotten teeth, atrophy of the lingual base, loss of smell, unclear articulation, and pus in the ear, all of which were the manifestations of collateral damage. The large number of toxic reactive oxygen species caused by radiation acts on local tissues continuously, resulting in chronic inflammation and tissue destruction that promote the production of reactive oxygen species. As a result, the vicious circle repeats. That is why Ms. Zhou's complications persisted for a long time.

Hydrogen selectively neutralizes toxic active oxygen. Rehabilitation with hydrogen inhalation is a big gift from God to Ms. Zhou. Being strong, enthusiastic and caring, she deserves the gift. I wish Ms. Zhou the best.

His Metastatic Tumor Shrank Unexpectedly: Thanks to Hydrogen

On Wednesday, November 14, 2018, Prof. Xu worked at the outpatient clinic and received patients who visited for consultation. An old couple hurried in when Prof. Xu was about to get off work. The couple were very excited to meet Prof. Xu. The wife said that they came for medical treatment (of drug-induced mandibular necrosis), and also to thank Prof. Xu. Because of hydrogen, her husband's lung metastasis was reduced significantly. "The doctor even thought that my husband had surgery before." The couple looked very happy.

> *Picture 1. Mr. and Mrs. Tan with Prof. Xu Kecheng*

The 64-year-old Mr. Tan was a faculty member of a school in Guangzhou before retirement. During a medical checkup organized by the school at the beginning of 2011, Mr. Tan was unexpectedly found to have kidney cancer. In March the same year, he received radical nephrectomy on the right kidney. Postoperative pathology indicated a tendency to Grade II clear cell carcinoma, which had invaded the renal capsule. In the following few years, Mr. Tan had repeated medical checkups, and no abnormality was found.

At the beginning of 2017, Mr. Tan had pain in the right knee joint and weakness in the lower limbs. After being admitted to hospital, he was given a PET-CT scan and biopsy. Then, he was diagnosed with lung cancer with bone metastasis. He started taking sunitinib to control his condition.

Starting from February 2018, Mr. Tan felt increasing pain in the lower front teeth. He had teeth extraction in an oral hospital. After that, the pain persisted; and signs of mandibular necrosis appeared. It hurt so much that Mr. Lin could not swallow food and stop tears rolling down. After several twists and turns in an attempt to seek medical treatment, Mr. Lin found out that his symptom was caused by the long-term use of zometa. Zometa can be used to treat solid tumors with bone metastasis like Mr. Tan's. But it also causes different degrees of side effects. Mr. Tan joked with a bitter smile, "I was hit by the lottery prize of zometa." In desperation, he discontinued the medicine.

The severe side effects brought Mr. Lin endless grievances. In addition, his lung metastasis was not optimistic. At that time, a cancer patient from Liwan station of Guangdong Light of Life Cancer Rehabilitation Association told Mr. Lin that Prof. Xu Kecheng, a well-known oncologist

and the president of the Association, applied hydrogen to cancer control and had brought benefits to many cancer patients.

In June 2018, Mr. Tan, at the suggestion of Prof. Xu, started hydrogen inhalation using a hydrogen-oxygen nebulizer. He inhaled hydrogen for around 5 hours at home every day.

He said that he didn't feel anything during hydrogen inhalation. But he was very surprised to see the results of a examination he took on August 31, which was more than 2 months after hydrogen inhalation.

"At that time, the doctor checked my imaging examination results and said: 'Your condition looks much better. Of course, this is because you just had surgery (on lung metastasis).'" Mr. Tan was stupefied for a moment and then said, "Doctor, I haven't received any surgery." "How is it possible?" The doctor could not believe it. "It's true. I have been inhaling hydrogen for more than two months at home." After saying this, Mr. Tan felt very happy. "So, hydrogen actually achieved similar effects as surgery. This is unbelievable."

On this visit, the Tan couple came to not only seek treatment but also to tell the good news. After hearing their story, Prof. Xu laughed: "The doctor's greatest joy is to know that patients have benefited." Since the time he applied hydrogen to cancer control, Prof. Xu has been receiving joy every day.

— Lin Hong

> *Picture 2. T cells express PD-1 molecules during activation. There are corresponding ligands PD-L1 on the surfaces of tumor cells. PD-1 binds with PD-L1 to prevent T cells from attacking cancer cells. T cells themselves are depleted and stop proliferation, therefore losing the ability to produce cytokines and activate immunity. That is, T cells fail to attack cancer cells.*

> **Comments from Xu Kecheng: cancer control from the perspective of regulating immunity**
> As important immune killer cells, CD8+ cytotoxic T cells play an important role in the immune control of tumors. However, their immune functions are closely related to cell surface molecules. Programmed death 1 (PD-1), an important immunosuppressive molecule, is expressed during T cell activation and binds with its ligand PD-L1 and CD28. Under continuous stimulation by tumor antigens, the PD-1 on the surfaces of CD8+ T cells bind with the PD-L1 of cancer cells. As a result, CD8+ T cells are depleted, stop proliferation, and lose the ability to produce cytokines and activate immunity. In other words, CD8+ T cells become unable to attack cancer cells. PD-1 is a marker of T cells in depletion and is abundantly expressed in cancer patients' CD8+ T cell circulation and tumor-infiltrated lymphocytes. It is associated with the poor prognosis of various cancers (including breast, pancreatic and gastric cancers).
> Japanese scholars Akagi J and Baba H discovered that the level of PD-1+ CD8+ T cells in peripheral blood was correlated with progression-free survival (PFS) and overall survival (OS) in cancer patients. A total of 55 patients with Stage IV colorectal cancer were studied. The patients inhaled hydrogen 3 hours every day for a total of 3 months. CD8+ T cells were separated from peripheral blood and analyzed for their phenotypes using flow cytometry. The results showed that patients with high levels of depleted PD-1+ CD8+ T cells in peripheral blood had short progression-free survival (PFS) and overall survival (OS). After hydrogen inhalation, depleted PD-1+ CD8+ T cells decreased; active PD-1-CD8+ T cells increased; and PFS and OS were prolonged accordingly. These results indicated that hydrogen restored the functions of depleted anaphase PD-1+ CD8+ T cells and turned them into active anaphase PD-1-CD8+ T cells, achieving a balance between PD-1+ and PD-1-CD8+T cells. As a result, patients' anti-tumor immune functions were boosted, leading to better prognosis.

> The mechanism by which hydrogen restores depleted T cell functions remains unclear. Depleted tumor infiltration and circulating CD8+ T cells are known for their characteristics of insufficient metabolism accompanied by mitochondrial function and quality decline as well as loss of mitochondrial metabolism, PGC-1α (peroxisome proliferator-activated receptor γ coactivator 1α (PGC-1α). As a positive regulator of mitochondrial metabolism, PGC-1α enhances insulin sensitivity, increases mitochondrial respiration, promotes mitochondrial biosynthesis, and facilitates b-oxidation of fatty acids. It is currently believed that mitochondrial dysfunction caused by inactivation of PGC-1α results in the depletion and immune function loss of CD8+ T cells. Hydrogen molecules activate PGC-1α and save CD8+ T cells that are depleted due to mitochondrial dysfunction, thereby restoring the immune activity of CD8+ T cells.

Brave Woman with 23 Years of Cancer Found A New Magic Weapon to Control Cancer

No one can tell that Huang Xueqiong is a patient with breast cancer for 23 years. She has survived the ups and downs in her fight with cancer like a brave warrior. A few years ago, she was found with breast cancer recurrence, followed by the appearance of a ground glass-like nodule in the left lung. Recently, she found hydrogen, a new magic weapon for cancer control. After hydrogen inhalation, Ms. Huang's breast cancer stabilized; sleep became better; and the annoying ground glass-like nodule shrank.

The 64-year-old Ms. Huang is a little plump, friendly, talkative, and cheerful. She looks no different from a healthy person. Few could have guessed that she is a patient with 23 years of breast cancer.

During a shower one day in October 1995, Ms. Huang noticed seepage from the right nipple. She immediately realized that it was serious. Without delay, she went to a hospital for examination and was later diagnosed with ductal papillary carcinoma. Not long afterwards, she received an operation that removed the entire right breast. Without taking chemotherapy, she took endocrine drugs for half a year.

In the next 15 years, Ms. Huang worked and lived a life like any other healthy people. She walked out of the darkness of breast cancer long time ago. Until in the 19th year at the end of March 2014, Ms. Huang felt a hard mass at her previous surgical site. Thinking back, she had illness and fever frequently recently. It occurred to her that her cancer might have recurred.

> *Picture 1. The CT scan report in May showed a ground glass-like nodule as large as 8 × 6mm in the lung.*

> *Picture 2. The CT scan report in November showed that the ground glass-like nodule in the lung measured 5×6mm.*

> *Picture 3. Ms. Huang and Prof. Shi Jianguo, deputy director of the Cancer Research Institute of The Fourth Military Medical University*

"From the images, it might not be good." At the doctor's suggestion, Ms. Huang underwent surgical resection of the mass. Three days later, the pathological results came out, confirming the doctor's inference that her breast cancer recurred.

At that moment, Ms. Huang could not believe that her cancer came back after nearly 20 years. She quickly accepted the reality and adjusted her mentality. She received six sessions of chemotherapy and continued to take endocrine drugs.

During an examination in 2015, a round glass-like nodule of around 7.5 × 6mm was detected in Ms. Huang's lower-left lung. Was it lung cancer? Ms. Huang did not know what to do. Since the nodule was likely to be benign or a sign of pre-cancerous lesion or early-stage lung cancer, doctors suggested that her condition be continuously monitored and follow-ups be done regularly.

One year later, the ground glass-like nodule of in Ms. Huang's lower-left lung grew bigger to 8mm. The doctor advised her to have an operation. But she wanted to wait and began to take hyperthermia in a hospital near her home. After half a year, the nodule shrank to 8mm. "I had hyperthermia regularly in the last 2 years, but the size of the nodule was always 8mm without further reduction." said Ms. Huang helplessly.

In October 2018, with the help of Guangdong Light of Life Cancer Rehabilitation Association, of which Ms. Wang was a member, Ms. Huang had hydrogen inhalation in Xu Kecheng Care for Health Studio. After half of month of inhalation, she felt that her sleep improved significantly, and was able to sleep for 8 hours instead of 5 or 6 hours before.

The bigger surprise came later. One month after hydrogen inhalation, Ms. Huang's CT scan report showed no sign of breast recurrence, and that the ground glass-like nodule in the lung shrank to 5x6mm from 8x6mm.

With hydrogen inhalation, Ms. Huang is very happy with hydrogen as a miraculous means of cancer control. She said: "I have stopped hyperthermia and just inhale hydrogen in the studio."

— Lin Hong

> **Comments**
> Ground glass-like nodules in the lung are displayed as shadows or blurs CT images. Ground glass-like changes are induced by many factors, such as inflammation, fibrosis, atypical adenomatoid hyperplasia, adenocarcinoma in situ, and alveolar hemorrhage. Senior age, history of smoking, over five-year-long history of extracranial malignancy, a nodule with burr on the edge, a large-diameter nodule, and a nodule located in the upper pulmonary lobe all represent risk factors for malignancy.
> The bigger the nodule, the greater the possibility of malignancy. If a nodule measures over 8mm in diameter or continues to grow, the probability of the nodule being a malignant lesion increases significantly. Generally, such a nodule is treated with surgical resection or ablation. Ms. Huang had a 8mm ground glass-like nodule in the lung and suffered from breast cancer earlier. Her case should be handled with surgery or ablation according to standard practice. However, she chose to inhale hydrogen first. A month later, the nodule shrank, probably because of the following: First, the nodule was a non-cancerous lesion, such as inflammation. Hydrogen has anti-inflammatory effects. Second, the nodule is an early-stage carcinoma. Hydrogen has the effect of inhibiting cancer cell growth.

Brain Metastasis Disappeared Because of Hydrogen?

When receiving her latest brain MR report, Ms. He couldn't believe her eyes. According to the report, the metastatic lesion in the parietal lobe on the left side of the brain had disappeared. Such a result is good enough to make a patient with Stage IV breast cancer with multiple bone

metastases and brain metastasis cry with joy. Ms. He wondered what made her brain metastasis disappear. It might be hydrogen.

In May 2017, Ms. He went to a hospital for examination due to a mass in the right breast. To her surprise, she was diagnosed with Stage IV breast cancer with multiple bone metastases. Then, she received seven sessions of chemotherapy and took lapatinib for treatment. In November the same year, she underwent a modified radical operation of the right breast cancer. On December 8, she completed the eighth session of chemotherapy. Starting from February 7, 2018, she had radiotherapy 25 times. In July the same year, she took xeloda.

> *Picture 1. Anteroposterior (left) and lateral (right) CT images during the contrast-enhanced MR scan of the head on July 27 (the metastatic tumor is indicated by the red circle).*

> *Picture 2. During the contrast-enhanced MR scan of the head on November 8, anteroposterior (left) and lateral (right) CT images showed that the brain metastatic tumor disappeared.*

In spite of active treatment, Ms. He's cancer progressed. According to the contrast-enhanced MR scan of the head on July 27, 2018, the left occipital lobe parenchyma and pia-arachnoid displayed a 8x5mm flaky abnormal signal with clear boundary and regular form. The diagnosis was intracranial metastasis.

For the brain metastasis, the doctor recommended Ms. He to receive brain surgery or radiotherapy and take xeloda at the same time. Considering the effect and adverse reaction, Ms. He was hesitant. At that time, she heard that Xu Kecheng Care for Health Studio offered hydrogen inhalation experience, which might help control cancer. She decided not to take brain surgery or radiotherapy. Starting from August, she inhaled hydrogen in the studio for more 3 months and meanwhile, took oral xeloda.

On November 8, Ms. He had a contrast-enhanced MR scan of the head. The results were unexpectedly good: Brain parenchyma showed no abnormal signal; the gray and white matters exhibited clear boundary; contrast-enhanced scan showed no abnormal enhancement; no abnormal signal in the skull; the original lesion in the left occipital lobe disappeared.

Ms. He recalled that after being diagnosed with brain metastasis on June 20, she did not receive any other treatment except oral intake of chemo drug xeloda. As far as she knew, xeloda was not very effective in controlling brain metastasis. Was it hydrogen that relieved her disease? She was overjoyed.

Good effects were also shown in Ms. He's husband, another cancer patient. Mr. He was diagnosed with nasopharynx cancer in 2013 and received radiotherapy 32 times. He said that there was often dirt in the nasal cavity after radiotherapy, causing a strong sense of obstruction. In particular, the nasal obstruction had serious impact on his sleep. "I had headache, neck pain, and poor spirits. It was very miserable for me." To improve the situation, he took lots of nutrients, but to little avail.

"Previously, I could not fall asleep. But now, I can sometimes sleep for 10 hours." After inhaling hydrogen together with his wife, Mr. He said, "My nose is unobstructed. I have better spirits, appetite and sleep than before. I have also gained weight. I weigh 62kg now. My target is 65kg. There is a small gap. I will make it soon."

— Lin Hong

Comments
According to a review published in the July 2018 issue of Clinical Oncology, most cytotoxic drugs cannot pass through the blood brain barrier; and almost all large-scale clinical trials have excluded patients with brain metastases. Therefore, controversy remains as to whether chemotherapy can be applied to the treatment of brain metastases. So far, there has been no standard systemic treatment for breast cancer with brain metastasis. In recent years, studies have shown that some chemotherapy drugs can pass the blood brain barrier and reach certain effective levels of concentration for brain metastases, thereby killing tumor cells. The U.S. National Comprehensive Cancer Network (NCCN) guidelines recommend treating breast cancer with brain metastasis (recommendation level: 2A) by using capecitabine (xeloda), high-dose methotrexate, cisplatin plus etoposide, and temozolomide monotherapy.
But there is no evidence that single use of capecitabine is effective for brain metastases. Ms. He's brain metastatic tumor disappeared 3 months after hydrogen inhalation, probably because hydrogen alone exerted a positive effect or hydrogen together with her oral chemo drug created a synergistic effect.

She Walked into the Sports Ground for Her First Rehabilitation Sports Meeting: Thanks to Hydrogen

On October 30, 2018, a special sports meeting was held at Liwan Stadium in Guangzhou. Participants of the sports meeting came from different places, with different life experience and professional background. Ranging from teenagers to the elderly in their eighties, the participants shared a common family: Guangdong Light of Life Cancer Rehabilitation Association.

This was the sixth sports meeting organized by the Association. It was also the first sports meeting for Ms. Cao after joining the Association. She was particularly excited. In the past few years, she had been afflicted with severe side effects of chemotherapy, including numbness of the hands and feet; unsteady walking; and frequent dizziness. Thanks to hydrogen, her knee joint pain was eased significantly. She walked into the stadium for the sports meeting for the first time.

In 2012, Ms. Cao was unfortunately found to have breast infiltrating ductal carcinoma, which was triple-negative (negative in ER, PR and Her-2). It meant that endocrine therapy and targeted therapy against Her-2 would not be effective in her case. In the same year, she underwent mastectomy of the right breast, followed by eight courses of chemotherapy.

Chemotherapy impaired Ms. Cao's peripheral nerves, giving her a sense of piecing pain like needles punching in the hands and feet. Moreover, due to strong pain in the knee joint, she could not stand for a long time and had to walk slowly with the support of a wall while going upstairs and downstairs.

"I joined Light of Life Wuyang station several years ago. I have never participated in a sports meeting. This year, I was finally in. I walked during the whole march-in ceremony and also stood for nearly one hour." said Ms. Cao excitedly. Such an experience was beyond her wildest dream before.

Why was there such a big change? She said that her knee pain was significantly eased because of hydrogen inhalation.

In middle September 2018, she heard that hydrogen can improve the side effects of radiotherapy and chemotherapy; and is helpful for cancer rehabilitation. Prof. Xu Kecheng, a famous oncologist and the president of Guangdong Light of Life Cancer Rehabilitation Association, opened a hydrogen inhalation experience center in his public welfare studio and provided hydrogen inhalation to the public for free. Ms. Cao took her husband Mr. Jiang to experience hydrogen inhalation together in the studio.

According to Ms. Cao, her husband suffered from chronic nephritis for more than 25 years and had been taking drugs such as Corbrin Capsule (Bailing Jiaonang) and uremic clearance granules to control the disease. After Ms. Cao became ill, her husband took care of her wholeheartedly. In 2013, she found that her husband had a lot of bubbles in the urine and urged him to take an examination in the hospital. Results showed a high level of white blood cells. Worried that the kidney could not bear it, the doctor did not prescribe any anti-inflammatory drug for him. Mr. Jiang had an examination once every six months. And his white blood cell count was founded to be higher than the upper limit each time.

During the Spring Festival in February 2018, Mr. Jiang had severe pain in the chest and was admitted to a hospital for emergency treatment. He was diagnosed with acute osmotic pleurisy and multiple parts of inflammation in both lungs. After half a month of hospitalization, his condition stabilized, and he was discharged. "At the time, the doctor called me to his office and said that my husband's pleural effusion was turbid and difficult to absorb."

After being discharged from the hospital, Mr. Jiang took anti-inflammatory drugs for more than a month. The results of follow-up examinations in both May and August 2018 showed that his white blood cells exceeded the normal level. After learning that hydrogen helped eliminate inflammation, Ms. Cao asked her husband to give it a try. "He had concerns at the beginning and refused to come. Eventually, he tried on September 22 the first time. After that, he felt that he could breathe more smoothly and was more relaxed. Then, he inhaled hydrogen regularly."

After inhaling hydrogen for half a month, the couple once went to take care of their grandson. "Previously, he was terribly out of breath after going upstairs for two floors. But now, he can go up to the sixth floor without a break and still have the physical strength to hold our grandson." The couple were overjoyed.

In November 2018, Ms. Cao accompanied her husband for a follow-up examination. The results were pleasantly surprising. Mr. Jiang's white blood cells, which had been beyond the normal level for years, fell into the normal range for the first time; and the pleural effusion had been basically absorbed.

Because of hydrogen inhalation, the couple feel stronger in the legs and are no longer bothered by shortness of breath. Nowadays, they often walk to the area near Haizhu Lake. They can easily walk more than 10,000 steps at a time.

— *Lin Hong*

> **Comments**
> Hydrogen does help cure arthritis. In Lourdes, France, it is said that on February 11, 1852, Virgin Mary made her presence that drove a 14-year-old shepherd girl to take water from Benat River. Later, people discovered that the spring water cured diseases and was particularly effective for paralysis. Many patients visited the place; were cured by the spring water; and then threw away their wheelchairs and crutches that supported them before. Later, these wheelchairs and crutches became a unique local scenery. There was a 30-meter-long underground tunnel in the Nordenau mine pit in Germany. Spring water emerged

> from the tunnel's rock wall, which measured around 100 square meters. By 2003, more than 2 million people worldwide had visited the place and used the spring water. The spring water cured 18 types of diseases in the visitors, with the top disease being joint pain, followed by bronchial pneumonia and kidney disease.
> In view of the antioxidant and anti-inflammatory effects of hydrogen molecules, hydrogen has been applied to sports medicine and proven to be effective in relieving rheumatoid, traumatic and gouty arthritis (International Journal of Sports Medicine, 2015).

Almost a Revolutionary Discovery: Patient with Late stage Gallbladder Cancer Were Brought Back to Life

Gallbladder cancer is no less malignant than pancreatic cancer. A large literature review published in European Journal of Surgical Oncology in October 2018 reported a study of around 474 patients. The patients with unresectable progressive gallbladder cancer had an overall survival of only 5 to 10.8 months. Overall survival was even shorter for patients with terminal gallbladder cancer accompanied by metastases.

The following is only an individual case which comes from the real world.

The patient has absolutely terminal gallbladder cancer with the following symptoms:

- *Huge gallbladder cancer that invaded the duodenum and caused intestinal obstruction;*
- *Multiple huge liver metastases that occupied at least half of the liver;*
- *Extreme anemia, with the level of hemoglobin at only 4g;*
- *Rheumatic heart disease with history of mitral valve replacement surgery and heart failure;*
- *72 years old; body-consuming hospitalization for 6 times in 3 years.*

All the symptoms above manifested in one person. Any one of the symptoms was enough to cause death.

What treatment reversed the situation and pulled the patient back from the verge of death?

- *Surgery? It was impossible for the patient.*
- *Chemotherapy? It was intolerable for the patient.*
- *Targeted therapy? The patient showed no sign for effective targeted therapy.*
- *Immunotherapy? The patient showed no sign for effective immunotherapy.*

The patient had no choice but to wait…

However, every cloud has a silver lining.

Persevere, wisdom, and innovation created a miracle in the strong patient, an elderly lady.

Medical History

Ms. Pan, 72 years old and a native of Fujian Province, suffered from terminal gallbladder adenocarcinoma with metastases in multiple lymph nodes in the porta hepatis and pancreatic head. In addition, she had severe anemia, heart failure and Type 2 diabetes; and had received mitral valve replacement surgery for rheumatic heart disease. After surgery, the tumor pressed against the duodenum, causing obstruction. As a result, Ms. Pan was unable to take in food normally and was given intubation for food intake. The tube stayed in her body for 51 days.

If not handled carefully, each of the above-mentioned symptoms might cause death to Ms. Pan immediately. She was not eligible for surgery and could not tolerate chemotherapy. Moreover, she showed significant side effects from and had to stop using an immune drug, which is hugely popular and costly at present. Except symptomatic treatment, everything else seemed to be ineffective in Ms. Pan's case. Despite the fact that she was on the verge of death, her condition made a significant turnaround eventually: gallbladder tumor was reduced by 2/3; liver metastasis became smaller; tumor marker returned to a normal level. With stable vital signs; the ability to eat and walk normally; and high spirits, Ms. Pan was recently discharged from the hospital. Her case was almost a medical miracle.

What brought her back to life? So far, all the evidence seems to point to hydrogen. If it is really hydrogen that turned around Ms. Pan's condition, the case can be a revolutionary discovery in the medical field.

Perseverance

"Save her. No matter how difficult it is and no matter how much it costs, we all try our best to save our mother." September 21, 2018 was probably the most difficult and heartbreaking day for the five brothers of the Huang family. But every one of them was determined to save their mother.

Since her illness, Ms. Pan had been admitted to the hospital six times. Her five sons took turns to take care of her and covered all the cost without any complaint. On September 21, Ms. Pan was admitted to the hospital for the sixth time and was sent to the ICU the next day. The doctor repeatedly told her family: "She might not survive long. You need to be mentally prepared."

At the time of the sixth hospitalization, Ms. Pan had severe anemia, with hemoglobin level at only 37g/L; serum total protein 53.3g/L; albumin 25.6g/L; red blood cell count 1.49 × 10^{12}/L; and hematocrit 0.14%. Her CA 199, SFP and CEA levels were 88.18 U/ml, 14.11 IU/ml, and 39.68 ng/ml respectively. Moreover, she had cardiac insufficiency; intestinal fistula caused by tumors compressing the duodenum; and metastatic tumors covering half of the liver. Ms. Pan was in extremely critical conditions. But her five sons were not willing to give up on her, even if there was only a glimmer of hope. For them, they have only one mother in this life.

> *Picture 1. Ms. Pan's biopsy pathology: gallbladder cancer; adenocarcinoma; moderate differentiation.*

According to the five sons, their mother suffered hardships for a lifetime. Ms. Pan was a peasant in the early years, and later worked as a textile worker in Hong Kong to earn a living. In the factory, she did the hardest work at the lowest level. Fortunately, her children had promising prospects. The elder four sons were smart and started their own business, which turned out to be booming later. The youngest son was brilliant in study and worked at a large company in Hongkong after graduation. The sons were always caring towards their mother. They often invited Ms. Pan to their houses for eating.

But Ms. Pan's happy time came to an abrupt end in 2017. In November 2017, she went to the hospital for examination due to pain in the upper-right abdomen. She was highly suspected to have gallbladder cancer, with metastases in the liver, porta hepatis and pancreatic head. Over a month later, her biopsy results came out, indicating Stage IIIa gallbladder adenocarcinoma of moderate differentiation. She was treated with nano-knife ablation and then took oral tegafur, gimeracil and oteracil potassium. But the effect was not satisfactory. Ms. Pan had anemia repeatedly and was hospitalized several times.

Every time Ms. Pan was hospitalized, she was accompanied and looked after by one or two of her sons. The other sons would visit her in Guangzhou once they had time. The Huang brothers' filial piety and perseverance deeply touched Pan Zhongbao, Ms. Pan's attending doctor, and Qianwei, the director of the inpatient department of Ward 3 of Fuda Cancer Hospital. The attending doctor said: "We will spare no effort to ease the patient's symptoms and prolong her lifetime." After being transferred to the ICU, Ms. Pan was given intubation in the stomach and rescued by the medical staff, who later carefully managed to maintain Ms. Pan's nutrition and treat her symptoms. Every staff member treated Ms. Pan as if treading on thin ice. But deep down, they knew that Ms. Pan was about to die soon unless there would be a miracle.

Miracle

While Ms. Pan was being rescued in the hospital, the Huang brothers were busy finding other ways to turn the situation around and save their beloved mother. At the beginning of October 2018, the youngest son in Hong Kong accidentally read a news report about Prof. Xu Kecheng using hydrogen to control cancer. He immediately contacted Shanghai Asclepius Meditec and obtained a hydrogen-oxygen nebulizer. While in conscious state, Ms. Pan inhaled hydrogen-oxygen mixture for 2 hours a day at the beginning, then 4 hours a day, and 8 hours a day eventually. She had a break for half an hour after 2 hours of inhalation, and then continued for another 2 hours. In this way, she inhaled hydrogen for 8 hours in total per day.

Ms. Pan's spirits became better with each passing day. On the 51st day of hospitalization, her stomach tube was removed. And she was able to eat and walk normally again. Her five sons were thrilled with joy. In order to make the situation more hopeful, they decided to give Ms. Pan a blind test of anti-PD-1 drugs. But after only one injection of the drug, Ms. Pan showed serious side effects, including fatigue, diarrhea, itchy skin and hypokalemia. She had to stop taking it.

"Except symptomatic treatment, hydrogen inhalation is the only way for her." The Huang brothers never expected that this seemingly desperate decision brought hope of survival to their mother. On January 8, 2019, Ms. Pan's blood tests showed that red blood cells, hemoglobin, and albumin increased to normal levels. More importantly, her three tumor markers returned to normal levels.

The more encouraging news was the CT scan findings on January 12, 2019, which showed that the gallbladder tumor shrank by 2/3 and all the liver metastases were smaller compared with the examination results on November 27, 2018.

Prof. Xu found it unbelievable. A few months ago, Ms. Pan was given a critically ill notice several times. But now, she looks refreshed with flushing complexion and full of energy. She can eat and walk normally, and visits the coffee shop every day. Seeing Prof. Xu, Ms. Pan stopped hydrogen inhalation and immediately got out of the bed. She followed Prof. Xu and Director Qian Wei to the coffee shop.

"It's a miracle." "I can't believe it!" Knowing about Ms. Pan's recovery, Prof. Xu Kecheng, accompanied by Director Qian Wei, visited Ms. Pan on January 17.

The liver and gallbladder are interconnected. Gallbladder cancer, like liver cancer, is highly malignant, easily relapses, and has poor prognosis. It is frightening for patients. In October 2018, the first issue of European Journal of Surgical Oncology published a literature review, which included a total of eight articles covering the chemotherapy results in 474 cases of

progressive gallbladder cancer. In the literature review, it was concluded that only 1/3 of the patients benefit from chemotherapy; and that the patients not eligible for radical surgery have an overall survival of 5 to 10.8 months. So far, it has not been reported in existing literatures that anti-PD-1 drugs benefit patients with gallbladder cancer.

Prof. Xu Kecheng was full of emotion after coming to know Ms. Pan's examination results. He held Ms. Pan's sons by the hand and said, "Thank you. Thanks to the five of you. You mother has survived exactly because of your perseverance and wisdom."

Prof. Xu continued: "There has been experimental evidence on hydrogen's effects on cancer control. But an old lady like Ms. Pan with so severe cancer demonstrated so positive results from only symptomatic treatments and hydrogen inhalation (hydrogen-oxygen mixture). The results are nearly revolutionary."

Prof. Xu Kecheng emphasized that Ms. Pan is an encouraging individual case that gives us hope. After reading Ms. Pan's medical records, Prof. Shi Jianguo, a doctoral tutor from the Air Force Medical University (formerly the Fourth Military Medical University) and also an oncologist, said that her case was indeed encouraging and hoped to see more similar cases.

— Lin Hong

> *Picture 2. Serum albumin change curve for Ms. Pan before and after hydrogen inhalation*

Date	2018.09.20	2018.10.08	2018.12.10	2019.01.08
Albumin (g/L)	25.6	32.2	37.8	41.8

> *Picture 3. Blood routine test index (red blood cell and hemoglobin) change curve for Ms. Pan before and after hydrogen inhalation*

Date	2018.09.20	2018.10.08	2018.12.10	2019.01.08
Red blood cell ($\times 10^{12}$/L)	1.49	1.43	3.03	5.99
Hematocrit (%)	0.14	0.14	0.28	0.29
Hemoglobin (g/L)	27	41	88	91

> *Picture 4. Tumor marker change curve for Ms. Pan before and after hydrogen inhalation*

Date	2018.09.20	2018.10.08	2018.12.10	2019.01.08
CA19-9 (U/mL)	88.18	61.34	93.84	15.47
CEA (ng/mL)	39.68	27.7	34.55	8.07
AFP (IU/mL)	14.21	11.25	22.85	2.09

> *Picture 5. Comparison of CT scan findings before hydrogen inhalation (left) and after hydrogen inhalation (right)*
> Primary tumors of gallbladder cancer (indicated by the red circles)
> Liver metastases (indicated by the red circle)
> Liver metastases (indicated by the red circle)

> *Picture 6. Ms. Pan walks every day.*

> *Picture 7. Xu Kecheng was full of surprise when checking Ms. Pan's CT images.*

Surprises from Hydrogen Inhalation: She Would Not Exchange Hydrogen Even for Gold

"If I had to choose between big villa and the only one hydrogen-oxygen nebulizer in the world, I would definitely choose the nebulizer." said Ms. Yang, a breast cancer patient from Jining, Shandong, after one month of hydrogen inhalation. Her tumor marker dropped to a normal level. She was excited to see many gratifying changes in her body after hydrogen inhalation.

August 22, 2018 was an unforgettable day for Ms. Yang. Just the day before, she flew from Shandong to Guangzhou with her younger daughter, who was only 8 years old. On the morning of August 22, she left her daughter sleeping in the hotel and went to see Prof. Xu in the outpatient clinic at Fuda Cancer Hospital according to her appointment.

Ms. Yang is a well-known local lactagogue practitioner (a profession for promoting lactation) in Jining, Shandong. As dictated by her occupation, she usually cleared mammary ducts for others. One day in 2016, she applied lactagogue to herself out of whim. Surprisingly, reddish yellow water came out of her right breast. Realizing that something was wrong with her breasts, Ms. Yang went to a hospital for examination. Later, she was diagnosed with benign papilloma, and underwent an operation that removed the papilloma. She remembered that on the day after the operation was completed, there was a hard mass in the affected area. She asked the doctor and was told that the mass would disappear probably in three months.

However, after 3 months, 6 months and then a year, the hard mass was still there. In November 2017, Ms. Yang took mammography and was diagnosed with breast cancer. The pathological results indicated Stage 0 ductal carcinoma in situ. In December 2017, Ms. Yang received an operation that removed the entire right breast in a large hospital in Beijing.

On August 19, 2018, Ms. Yang's cancer antigen CA125 was found to have increased to 75.39 (normal value: 0-35). She was only told by the doctor to keep observing the condition.

At that time, she happened to read a news about Prof. Xu Kecheng treating cancer with hydrogen. She immediately inquired about Fuda Cancer Hospital where Prof. Xu Kecheng worked, and made an appointment as an outpatient. However, her relatives and friends discouraged her: "Treat disease with hydrogen? Isn't that crazy? It's impossible." Fortunately, Ms. Yang's husband fully supported her.

On August 22, Ms. Yang left her younger daughter in the hotel and took a taxi to Fuda Cancer Hospital to meet Prof. Xu Kecheng. After patiently listening to her statement and carefully reading her pathological data, Prof. Xu said: "The increase in CA125 means that there is cancer or a precancerous lesion. You can observe your condition for a while."

"The doctor from my hometown told me the same. But I am worried. What if the cancer spreads?" Ms. Yang sounded anxious. She was also a little disappointed, because she heard from others that Prof. Xu dared to challenge the most difficult cancer. However, he did not challenge her case.

She kept silent and looked at the kind old doctor. "He will definitely find a solution for me." She thought to herself. Then, Prof. Xu said: "Try to inhale hydrogen. There is a member from our

cancer rehabilitation association. She had ovarian cancer. Her CA125 dropped after hydrogen inhalation."

Soon after Prof. Xu finished, Ms. Yang stood up and held Prof. Xu's hands, saying: "Professor, I came here exactly for your hydrogen (treatment). I have been your fan for a long time and want to buy your book. But it is out of stock." Prof. Xu rose from the chair, took his new book Practice Chinese Way of Cancer Control, and signed it before handing it to Ms. Yang. "I didn't expect that he is such a famous expert and still so easy-going." Ms. Yang was grateful.

After leaving the hospital, Ms. Yang went straight to Xu Kecheng Care for Health Studio and inhaled hydrogen there for one and half hour. The gas from the hydrogen-oxygen nebulizer passed through the nasal cannula and entered her body. After returning to the hotel that day, Ms. Yang had unprecedented fatigue and slept soundly at night. The next day, she went to the studio to inhale hydrogen again. After completing inhalation, she felt much relaxed, and her complexion improved. However, after all, she lived in a place far away from the studio and could not come for hydrogen inhalation every day. As a result, Ms. Yang bought a hydrogen-oxygen nebulizer and inhaled hydrogen at home for 4 to 6 hours per day. She said: "As soon as I get home, I turn the machine on and inhale hydrogen while watching TV and even sleeping."

Significant Changes in Her Body

1. For many years, Ms. Yang had adenomyosis and menstruation with large blood volumes, pain and irregular period. One month after hydrogen inhalation, she did not experience any discomfort during the menstrual period. And the menstrual blood volume and period became normal.
2. Originally, Ms. Yang had many dark spots on the skin. She used many products and methods to remove the dark spots, but to no avail. Since hydrogen inhalation, the dark spots on the face were lighter and smaller; and vanished eventually. Gradually, she found herself with more luster, ruddy expression and smooth skin on the face.
3. In dry days in autumn, Ms. Yang had dry and bitter mouth in the morning previously. But she does not have the problem nowadays.
4. With thin and fine hair, Ms. Yang bought five wigs successively before. After hydrogen inhalation, her hair became a lot thicker and shiny black. Her eyebrows thickened and eyelashes were longer. Her nails grew faster.

During the National Day holiday of 2018, Ms. Yang went to the hospital for a follow-up examination. According to the results, the liver cyst disappeared; and tumor marker CA125 decreased to 24.62, which was a normal level. Ms. Yang was surprised with joy.

Her parents in-law also benefited from hydrogen inhalation. It turned out that during a National Day holiday, Ms. Yang's parents in-law came to visit her and tried to inhale hydrogen at her home. "My mother-in-law had arthritis, which caused so much pain that she had to take painkillers. After a few days of hydrogen inhalation, she said that she had much less pain and no longer needed painkillers. My father-in-law had cerebral thrombosis. After hydrogen inhalation, his dizziness was gone and appetite was better."

Based on her personal experience, Ms. Yang concluded that hydrogen inhalation has a dose effect. The longer the inhalation, the more significant the effect. "Now I can't live without this baby (hydrogen-oxygen nebulizer). I take it with me wherever I go." Every weekend, Ms. Yang drives her younger daughter from Jining to a county to meet her husband and older daughter. She always brings the hydrogen-oxygen nebulizer with her. "My mother-in-law is afraid of damaging the machine and specifically covered it with a cloth."

Ms. Yang said with emotion: "I feel that my greatest luck came from accepting this new thing. If I had to choose between a big villa and the only one hydrogen-oxygen nebulizer in the world, I would definitely choose the nebulizer."

Sharing Her Anti-cancer Experience

Both Ms. Yang's parents died of cancer, with her father from colon cancer and mother bladder cancer. Over a year after her mother's death, Ms. Yang was found to have cancer. She thought that her cancer may have a lot to do with depression.

Ms. Yang's cancer was not serious. Seeing that cancer took the lives of her loved ones and so many young people suffering from cancer, she knew how hard it is to fight cancer. She is very willing to share her experience of hydrogen inhalation, hoping to help more other cancer patients in suffering.

In their book Practice Chinese Way of Cancer Control, academician Tang Zhaoyou and Prof. Xu mentioned the effect of swimming. According to Tang Zhaoyou, moderate swimming promotes the secretion of dopamine, which inhibits tumors. Nowadays, Ms. Yang swims 1,000 meters in The Grand Canal almost every day. Swimming has boosted her immunity, while hydrogen inhalation has improved the environment in her body.

— Lin Hong

Unbelievable Test Results

On October 25, 2018, Ms. He came to Prof. Xu with the test results she just received. Despite that the test sheet clearly showed that the two most important tumor markers CEA and CA19-9 were at normal levels, Ms. He still found it hard to believe and wanted to get confirmation from an authoritative expert.

"Your results are very good. Congratulations!" said Prof. Xu Kecheng. Ms. He was very delighted and said thanks again and again.

"What treatment did you have during this period?" Prof. Xu Kecheng asked.

"A month ago, you suggested that I try hydrogen. Do you still remember?"

"Apart from hydrogen inhalation, did you get any other treatment?" Dr. Wen Weifeng and the nurse asked at the same time.

"No, not really." Ms. He raised her voice to confirm.

Ms. He, 66, is a retired college teacher. In March 2015, she had discomfort in the stomach and went to a hospital for gastroscopy, in which abnormality was detected. Then, Ms. He was diagnosed with Stage I gastric adenocarcinoma (including signet ring cells) of poor differentiation. During the treatment of gastric cancer in the hospital, she was accidentally found to have a 0.3cm ground glass-like nodule in the left lung.

The CT scan in January 2018 showed that the ground glass-like nodule in the left lung was 0.8cm long and contained some solid tissues. At the doctor's suggestion, Ms. He received surgical resection of the nodule. Biopsy results indicated stage I lung adenocarcinoma, for which chemotherapy was not needed. Further genetic test results suggested mutation of No.19 gene. Therefore, targeted drugs could be applied.

The test results in March 2018 were normal. But in July 2018, Ms. He's tumor markers CEA and CA19-9 increased to 17.64 (normal value: 0 to 5) and 36.7 (normal value: 0 to 35) respectively. Increase in these markers indicated that the tumor had recurred or was about to relapse. Ms. He's CT scan and gastroscopy showed no sign of lesion. It was undoubtedly a "time bomb" in the body. Ms. He was advised to take chemotherapy or targeted drugs.

Was there any other better option? Ms. He was at a loss what to do. She made an appointment with Prof. Xu Kecheng. Prof. Xu also suggested that she take chemotherapy or at least targeted drugs. She refused and hoped that Prof. Xu recommended certain rehabilitation measures, especially hydrogen inhalation.

On September 17, 2018, Ms. He started to inhale hydrogen in Xu Kecheng Care for Health Studio. On October 15, after less than one month of hydrogen inhalation, Ms. He went to the Cancer Hospital of Sun Yat-sen University for a follow-up examination. After receiving the test results, she couldn't believe her eyes: CEA 3.71; CA19-9 32.77. They indicators were back to normal.

In addition, Ms. He experienced other significant changes in her body: Previously, she had difficulty falling asleep and always woke up at 4 in the morning. Now, she sleeps well and wakes up only at dawn. Previously, she had loose stool. Because of good digestive functions now, her stool is in shape. Ms. He is very happy and will continue to inhale hydrogen to maintain the good effect.

— *Lin Hong*

> **Comments from Xu Kecheng**
> CEA and CA19-9 are tumor markers that increase in the cases of many cancers. For Ms. He, both the markers increased. Was it due to her stomach or lung cancer? Both are possible.
> Increase in tumor markers for stomach or lung cancer indicates one abnormality or several simultaneous abnormalities. Tumor marker increase does not occur to every patient but must be regarded as an important warning of cancer recurrence for particularly the patients with history of cancer. Cancerous tumors cannot be detected even by endoscopy, ultrasound, CT or other means. That is because the period from tumor marker increase to detection of cancerous tumors can be a few months or even years. The status in this period is known as "disease to come".
> In the case of such "disease to come", it is a difficult for both patients and doctors to make treatment choices. Chemotherapy is generally required but brings side effects that are usually difficult for patients to accept. Ms. He was exactly concerned about this.
> As a senior doctor who has received traditional medicine training, I am a member of the "circle" and therefore speaks the language specific to the "circle". On the other hand, I am a cancer survivor and may understand patients' thinking better than those in the "circle".
> The best doctor presents disease. We should aspire to be the best doctors.
> If chemotherapy is the "right path" to treat "disease to come", hydrogen can be understood as the "tortuous path" or "side path". We can always "overtake on a bend" or "change lanes to overtake". Many studies have shown that hydrogen molecules can transform the microenvironment. Other studies reveal that the biological effects of hydrogen molecules are positively correlated with hydrogen concentration in tissues.
> Hydrogen molecules can inhibit cancer cells, which is generally attributed to their antioxidant and anti-inflammatory effects. The most powerful and toxic oxidants in the body are •OH and $ONOO^-$. They act on nucleic acids, lipids and proteins, causing DNA damage, lipid peroxidation and protein denaturation. Smoking, air pollution, chemicals, mental stress, and inflammation facilitate cell mutation by inducing these oxidants and eventually cause formation of cancer cells. Hydrogen molecules are specific scavengers of the two oxidants.

> Recent studies have shown that hydrogen molecules also exert controlling effects on cancer by maintaining mitochondria and regulating immunity.

Because of Hydrogen, He Gave a Deep Bow to Xu Kecheng on the Stage

On October 22, 2018, a lecture titled Hydrogen Molecules and Health was held at Soluxe Hotel Guangzhou. The speaker was Prof. Xu Kecheng, a distinguished oncologist. In the past year, he has been focused on one thing: studying hydrogen for cancer treatment. He read a large number of literatures, conducted a Hydrogen Tour, and set up a hydrogen inhalation experience area in his own studio.

At the sharing session following the lecture, several experiencers who had inhaled hydrogen shared their feelings. At the end, an old man suddenly went to the stage and gave a deep bow to Prof. Xu Kecheng and then to volunteers of the health studio. The old man's move surprised everyone present.

Who was he? Why did he do it?

The old man was Mr. Sun, a villager from the First Village of Happiness in China. Led by the village chief, Mr. Sun went to a lecture about hydrogen medicine by Prof. Xu Kecheng and Sun Xuejun on August 6, 2018. At that time, several hydrogen-oxygen nebulizers were placed on both sides of the venue for everyone to experience for free. Throughout the lecture, Mr. Sun was carefully inhaling hydrogen and listening to the lecture at the same time. When the lecture ended 2 hours later, Mr. Shen, who was sitting next to Mr. Sun and also inhaling hydrogen, looked at Ms. Sun and said: "Hey, your cheeks have become rosy."

"Oh? Really so amazing? I need to try this again." But he was only thinking about it. Considering that there were many people who went to Prof. Xu's studio for hydrogen inhalation, Mr. Sun guessed that there might not be an available spot for him. Afterwards, he did not take the initiative to visit the studio for hydrogen inhalation.

> *Picture 1. Paper published by Prof. Sun Wushuang and other scholars*

> *Picture 2. Experimental data (1)*

On September 15, a celebration was held by the First Village of Happiness in China in Soluxe Hotel Guangzhou. At the event, Mr. Sun met the studio's volunteer Deng Yingzi. With the latter's help, he inhaled hydrogen in the studio every day starting from September 16.

It turned out that Mr. Sun was a heavy smoker with more than 30 years of smoking history. "I was more self-disciplined when serving the air force as a soldier. Then, I smoked two packages of cigarettes every day." Starting from 2010, Mr. Sun had terrible shortness of breath. He went to a hospital for examination. Results indicated Stage III chronic obstructive pulmonary disease (COPD).

Patients with COPD experience increasingly obstructed respiratory flow, which not only affects the respiratory system, but also involves multiple organs such as skeletal muscles and the heart. Gradually, the symptom develops into pulmonary heart disease, eventually leading to respiratory failure, systemic organ failure, and even death.

The doctor ordered Mr. Sun to quit smoking immediately; or else he would "be finished soon". Realizing the harm of COPD, Mr. Sun, who was once a soldier, endured all the discomfort and managed to quit smoking in one week. However, there was no specific drug or cure for COPD. Every acute exacerbation was a huge blow to Mr. Sun's respiratory function. The only thing that could be done was to slow down COPD development.

> *Picture 3. Experimental data (2)*

> *Picture 4. Paper published by Prof. Zheng Zeguang's research team*

"I had no strength in the hand to even carry a mineral water bottle. I couldn't breathe after walking a few steps up and down the stairs. I couldn't fall asleep at night." The effects of COPD on Mr. Sun's life were self-evident. Besides, the disease was sometimes less and sometimes exacerbated, making Mr. Sun very miserable.

By coincidence, Mr. Sun listened to the hydrogen medicine lecture on August 6. Afterwards, he started to be interested in hydrogen. He inhaled hydrogen consecutively for a month with the help of a volunteer from Xu Kecheng Care for Health Studio.

"Unbelievable. Hydrogen is really magical." After hydrogen inhalation, Mr. Sun had better appetite and sleep. He felt sleepy at 10pm. He also became physically stronger and could take things that weighed dozens of kilograms from the food market to home. Even the spots on his skin were lighter than before.

Hydrogen significantly relieved Mr. Sun's symptoms. He was full of gratitude to Prof. Xu Kecheng and volunteers of his studio. That was why he went up to the stage and gave a deep bow to Prof. Xu.

— Lin Hong

Scientific evidence of hydrogen's protection of the lung

In China, chronic obstructive pulmonary disease (COPD) features high incidence, high disability, high mortality and high disease burden. It has become a very serious public health concern. Smoking is the most common risk factor of COPD. There are strong correlations between smoking, oxidative stress and COPD symptoms.

In May 2017, Prof. Sun Suzhuang from Hebei Medical University and other scholars published a medical paper titled Hydrogen Coadministration Slows the Development of COPD-like Lung Disease in a Cigarette Smoke-induced Rat Model in the International Journal of COPD. The study investigated the beneficial effects of hydrogen at different concentration levels on COPD and the potential mechanisms. It was found that in rat models, hydrogen inhalation slowed down the occurrence of lung disease in rats induced by cigarette smoke; and that inhalation of 22% and 41.6% hydrogen led to better results than inhalation of 2% hydrogen.

Researchers established rat COPD models through smoke exposure. With nebulizers manufactured by Shanghai Asclepius Meditec, the rats were given inhalation of hydrogen at different concentration levels as an intervention measure. Then, the rats' daily life and weight changes were observed. Their lung functions and right ventricular hypertrophy indexes were evaluated; and white blood cell counts in bronchoalveolar lavage fluid were assessed. Pathological changes of lung tissues were analyzed using light microscopy and electron microscopy. The changes of cardiovascular structure and pulmonary artery pressure were

monitored through ultrasonography. Immunohistochemistry, Westernblot, and real-time polymerase chain reaction were applied to determining the levels of tumor necrosis factor alpha; interleukins IL-6, IL-17 and IL-23; matrix metalloproteinase-12; metalloproteinase-1 tissue inhibitor (TIMP-1); and mRNA in caspase-3 protein, caspase-8 proteins and lung tissues.

The experimental results showed that inhaled hydrogen significantly decreased inflammatory cells in bronchoalveolar lavage fluid and reduced the levels of tumor necrosis factor TNF-α, IL-6, IL-17, IL-23, metalloproteinase-12, and mRNA and protein expression of caspase-3 and caspase-8. However, the hydrogen increased the mRNA and protein expression of metalloproteinase-1 tissue inhibitor (TIMP-1). In addition, hydrogen inhalation improved lung pathology, lung function, and cardiovascular function, thereby lowering the right ventricular hypertrophy index.

In May 2018, Prof. Zheng Zeguang from the Institute of Respiratory Diseases of Guangzhou Medical University published a research paper in the Journal of Thoracic Disease, stating that inhalation of high concentrations of hydrogen relieved smoking-induced emphysema, chronic bronchitis, small airway remodeling and lung function deterioration in mice. This protective effect is attributed to the anti-inflammatory and antioxidant effects of hydrogen. Inhibition of ERK1/2 and NF-κB signaling pathways might be related to the anti-inflammatory effects of hydrogen.

Wild with Joy , Just for a Bad Smell

"Do you believe it? I lost my sense of smell over 20 years ago. Just because I inhaled hydrogen for an hour and a half, it came back to me miraculously." Zhang Ming'en, a Chinese American, was delighted to share her story as a "magic responder" to hydrogen.

Born in Hongkong, Ms. Zhang emigrated to Washington, D.C. with her family when she was young. It was not a happy thing for her to live in Washington, where there were four distinct seasons and lots of flowers and trees. She started a protracted confrontation with pollen allergy. In springs and summers when pollen flew in the air, as well as in midsummers and autumns when reeds flourished, Ms. Zhang had constant sneezes, a running nose and watery eyes. Many people with pollen allergy have nasal discharge running out of the nasal cavity. But Ms. Zhang's nasal discharge flew inversely. Before this, she once suffered from bronchial pneumonia 4 times in one year and almost lost her life.

To control her condition, Ms. Zhang had to take steroids and antibiotics repeatedly. However, the side effects of the drug were also immediate. Due to excessive burden on the regulatory mechanism, the originally precise endocrine system was disturbed; normal intestinal flora equilibrium was broken; liver and kidney functions deteriorated; and the immune function declined. It was miserable for her.

In addition to the suffering, Ms. Zhang's sense of smell disappeared without her realizing it. "One day when I was about 20 years old, I found myself unable to smell the fragrance of nice roses and stinky smell from garbage piles. I realized that my sense of smell is gone."

Ten years ago, Ms. Zhang received some Brazilian green propolis from a friend. By dripping green propolis into the nose in the morning and evening every day, her protracted pollen

allergy was finally under effective control. However, she did not restore the sense of smell. She felt very sad and helpless.

At the beginning of 2018, Ms. Zhang accompanied her friend Amy, who was suffering from breast cancer, to Fuda cancer Hospital for treatment. Thanks to the help of Prof. Xu Kecheng and other experts, Amy's condition was effectively controlled. Ms. Zhang specially brought her solo album and gave it to Prof. Xu Kecheng as a token of gratitude.

> *Picture 1. Zhang Ming'en gave her solo album to Prof. Xu Kecheng as a token of gratitude.*

On July 9, 2018, Xu Kecheng invited Zhong Zimei, a famous writer from Hongkong, and Amy to visit Xu Kecheng Care for Health Studio in Room 708 of Soluxe Hotel Guangzhou. As Amy's good friend, Ms. Zhang also went to the studio that day.

According to Prof. Xu Kecheng's introduction, the occurrence and progression of more than 200 diseases of the human body are associated with free radicals. Hydrogen is an ideal antioxidant reducing agent for selective elimination of free radicals. It helps treat various diseases and has positive regulating effects on many functions of the human body. On the spot, Prof. Xu invited them to experience hydrogen inhalation.

Can hydrogen really cure diseases? Ms. Zhang thought it was unbelievable. "Hydrogen seems to be flammable and explosive. Is it dangerous?" She raised her concerns. Volunteers in the studio told her that hydrogen only burns and explodes in an environment with excessive pressure or temperature. In a big room with normal temperature and ventilation, the small amount of hydrogen generated by nebulizers will not cause an explosion. Moreover, the hydrogen-oxygen nebulizers used in the studio are provided by Shanghai Asclepius Meditec. There is no hydrogen storage space in the studio. All the hydrogen-oxygen mixture generated via water electrolysis is directly sent to the human body through inhalation. Therefore, it is very safe to inhale hydrogen.

Without further concerns, Ms. Zhang inhaled hydrogen according to the instructions of a volunteer in the studio while going through popular science readings about hydrogen.

After one and half hour, Ms. Zhang and her companion finished hydrogen inhalation and stepped into an elevator to leave. In the elevator, she suddenly smelled smoke. "How can there be smoke?" She muttered. Immediately after saying that, she realized that her sense of smell was back. "I want to go back." Looking ecstatic, she returned to Xu Kecheng Care for Health Studio, held the volunteer by hand excitedly, and thanked her.

She felt that every blood vessel and every cell in her nose were slowly waking up, just like plants awaken in early spring gradually and radiate vitality. It was a wonderful feeling.

More surprisingly, Ms. Zhang's recovery of smell was not transient. Every day in the future, she can use her nose to perceive the world. She feels happier.

On July 30, Ms. Zhang got up early and went from Hong Kong to Xu Kecheng Care for Health Studio to inhale hydrogen again. To this end, she deliberately changed the original ticket to the United States from August 1 to August 2. Before leaving, she bought a mango-flavor cake and a few packages of hand-made cookies from an old and famous shop in Hong Kong and gave them to Prof. Xu Kecheng personally. "I can smell the mango in this cake. It smells very sweet!" She was full of gratitude.

— Lin Hong

Comments from Wei Youzhen
In addition to rays of light in the natural world, the biosphere air layer is filled with gas molecules. These chemical substances guide organisms to discover and undoubtedly represent the gas navigation for organisms. After hundreds of millions of years of evolution, many of human beings' animal partners, such as dogs and hawks, have retrogressive smell but keep a keen sense of smell, on which they rely to identify dangers. However, human beings experienced high development of the neocortex, which covered the rhinencephalon as the old brain. As a result, intelligence was generated, and the smell function retrograded, making human beings smell-blunt animals. Nevertheless, for human beings, the sense of smell is still a basic ability to discover the world. It helps us identify things and further influences the high emotions of the brain. After wall, any good smell boosts the emotion.
However, in the real world, there are some people who cannot tell odor from fragrance, only because the olfactory cells in the nasal mucosa are disabled. This phenomenon is allergy or inflammation.
Hydrogen can restore olfactory cells and enable them to accept all stimulations in the air. Simulations from good and bad smell pass through the olfactory bulb and bundle to the brain. As a result, the brain does not feel lonely or end up in dementia.

Weak Lady Refreshed after One Experience of Hydrogen Inhalation

Xu Kecheng Care for Health Studio has two regular visitors, a loving couple who have been together for more than 30 years. The wife Ms. Chen was a patient with stomach cancer, while her husband had high blood pressure and sleep problems. Ms. Chen showed surprising response after an accidental experience of hydrogen inhalation. Since then, she continued with hydrogen inhalation, as a result of which her symptoms were relieved significantly. The husband, who often accompanied Ms. Chen to the studio, took the chance to inhale hydrogen and has benefited tremendously.

Picture Ms. Chen and her husband are experiencing hydrogen inhalation.

Before hydrogen inhalation, Ms. Chen was as weak as a willow catkin. She received eight sessions of chemotherapy. On a family visit in Canada in July 2017, Ms. Chen felt pain in the upper abdomen but did not take it seriously. After returning to China in September 2017, she had a physical examination and was later diagnosed with gastric antrum cancer. In October 2017, she underwent an operation that removed a major part of the stomach. After surgery, she was given chemotherapy.

Ms. Chen suffered significantly from chemotherapy: She had very poor quality of sleep. Often, she went to bed at 9pm, fell asleep at 12pm, and woke up every one or two hours at night. Her weight plummeted and facial complexion became dark. She spoke in a faint voice. In addition, she often felt very dizzy and was so weak that she had to stay in bed a lot. Her husband Mr. Huang cooked every day and took care of her without complaint. When Ms. Chen was in a bad mood, Mr. Huang patiently appeased her and guided her out of negative emotions. He was both his wife's personal caregiver and moral support in cancer treatment.

One day in early August 2018, Ms. Chen was attracted by a message shared in a WeChat group of cancer patients: On the afternoon of August 6, Xu Kecheng Care for Health Studio will hold a lecture on hydrogen and cancer rehabilitation in Soluxe Hotel Guangzhou. Hydrogen cures cancer and promotes recovery? In spite of her suspicion, Ms. Chen decided to attend the lecture out of Xu Kecheng's reputation.

At that time, going outside was a luxury for Ms. Chen, who was extremely weak. She panted for breath after only a few steps of walk. Her husband had to carry a small bench with him so that she could sit when feeling tired. From their home to Soluxe Hotel Guangzhou, Ms. Chen sat on the small bench for rest after every few steps. Eventually, they arrived at the venue for the lecture.

Not long after the lecture started, Ms. Chen felt physically weak but still listened carefully. At the time Prof. Xu made a speech, Ms. Chen was dizzy and almost fainted. She left the spot with support by her husband. Volunteer Deng Yingzi took her to the neighboring room 708, the place for Xu Kecheng Care for Health Studio, for a break. Deng Yingzi turned on a hydrogen-oxygen nebulizer and asked Ms. Chen to try. According to the expiration and inspiration instructions from Deng Yingzi, Ms. Chen started inhaling hydrogen while feeling the constant flow of hydrogen and oxygen into the body. After one and half hour, Ms. Chen recovered certain strength and felt a little more relaxed.

The real amazing change occurred after Ms. Chen went to sleep that day: She sweat so heavily before midnight that her clothes were wet with sweat. In the next half of the night, she fell into sound sleep until dawn. She woke up with a fresh mind. How did it happen? Ms. Chen wondered if it was. Why is this happening? She wondered if it might be detoxification.

The surprising effect of detoxification drove Ms. Chento visit Xu Kecheng Care for Health Studio again. Volunteer Peng Ximei received her and added her to the hydrogen inhalation service group. Since then, she went to the studio for hydrogen inhalation in the company of her husband regardless of weather conditions.

The changes shown in Ms. Chen after hydrogen inhalation were self-evident. After three days of hydrogen inhalation, she was able to walk a short distance without using the small bench for rest. Previously, she struggled after walking only a few steps. Half a month later, she was able to go home alone without support by her husband. Moreover, she had much better sleep and often slept without waking up until dawn.

Ms. Chen's husband was also a beneficiary of hydrogen inhalation. He had high blood pressure and sleep problems. After hydrogen inhalation, he slept well without waking up at night; and his blood pressure stabilized. They said that they themselves knew the changes in their bodies the best. Nowadays, the couple have good spirits, enough physical strength, and good mood. In terms of the changes shown by Ms. Chen, the author specifically spoke to a few professionals. The conversations were as follows:

Author: Ms. Chen recovered her physical strength after hydrogen inhalation for one and half hour. How can it be effective so fast?

Dr. Kong Xiaofeng:

According to the cases we observed, the patients with existing and serious symptoms showed rapid improvement after hydrogen inhalation. As Ms. Chen said, she had toxins in her body. Such toxins are exactly reactive oxygen species, mainly hydroxy •OH.

She is a patient with gastric cancer and has received surgery and chemotherapy. The formation, progression, and invasion of cancer cells are all related to reactive oxygen species. But cancer cells themselves are where toxic reactive oxygen species are produced. Surgical trauma and particularly chemotherapy facilitate the production of abundant reactive oxygen species in the body. These reactive oxygen species may destroy the function of mitochondria,

which are the energy factories of the body. When we humans walk, move, and think, we need energy. Once the energy is insufficient, we have difficulty walking and tend to be tired. Hydrogen specifically neutralizes toxic reactive oxygen species; and this action occurs very quickly. Moreover, hydrogen protects mitochondria. Therefore, it is understandable that Ms. Chen felt stronger after hydrogen inhalation.

Author: On the day after her first hydrogen inhalation, Ms. Chen sweat heavily in the middle of the night. How to explain that? The biggest feature of hydrogen is that it is very safe. Hydrogen is generated in our intestines. Is Ms. Chen's sweating a side effect of hydrogen inhalation?

Prof. Wei Youzhen:

Ms. Chen's heavy sweating in midnight is like night sweat in the case of tuberculosis. It is a sign showing that the body's immune function is in play. It seems that hydrogen can be used to mobilize the body's immune function. The sweat gland is a skin gland that secretes sweat and is regulated by autonomic nerves (vegetative nerves). Under normal circumstances, sweat glands play a role of secretion and excretion through the feedback and the influence of the central nervous system. For example, if the ambient temperature is high, people naturally perspire to lower body temperature and facilitate the body's self-protection. If a person is emotional, excited or nervous, perspiration is controlled through the regulation of autonomic nerves. If vegetative nerves dysfunction or are disturbed by toxins, sweat gland secretion and excretion are interrupted. As a result, sweating becomes a disorder. Conditions such as hyperthyroidism, diabetes sequelae, tuberculosis and menopausal syndrome all cause involuntary, unconscious, uncontrolled, and rapid sweating, which is night sweat.

Due to disease intrusion, Ms. Chen's immune system was inhibited and metabolism was low. After hydrogen inhalation, call-back of the body's immune system was probably triggered in Ms. Chen. If the call-back reaches the level of sweating, it is considered to be a good sign.

— Lin Hong

Advice from a Beijing Professor with Allergic Rhinitis

After the beginning of winter, Beijing is rampant with cold north wind, giving people chills. The visit of friends from the south brings a different warmth to the north cold.

On November 16 to 18, 2018, Prof. Xu Kecheng arrived in Beijing for the Second Annual Conference of the Chinese Association of Rehabilitation Medicine. He also had an important activity on the agenda: to meet Ms. Wang, an old friend and a long-time patient who he had been thinking about.

Ms. Wang was once the leader of a big organization and a bureau-level official. Her husband, once a major leader of certain provincial government and later a member of the central government, was referred to as the "old secretary".

Ms. Wang suffered from ureter cancer of the right renal pelvis. She had atrophy of the left kidney for other reasons many years ago, rendering the left kidney in poor function. If her right kidney was removed, she would need lifelong blood dialysis. Ms. Wang refused to take nephrectomy. Two years ago, accompanied by her husband, she came to Fuda Cancer Hospital in Guangzhou and took minimally invasive ablation and other treatments. Her right

kidney was saved and tumor was basically controlled. But the trouble was that Ms. Wang often felt tired. In particular, after receiving a more complicated ablation in December 2017, she was weak all over and unwilling to talk or move.

As described by herself, raising hands or lifting legs felt as if her hands or legs were filled with lead. Fatigue does not seem to matter much. But cancerous fatigue in cancer patients is very tricky. To treat Ms. Wang's fatigue, Prof. Xu Kecheng tried a lot of methods, including: discontinuing relevant drugs; improving nutrition; increasing calorie in the body; infusion of energy drugs; psychological support; and taking Chinese medicine prescriptions for strengthening the spleen, nourishing the kidney and replenishing vital essence. Ms. Wang also orally took a ginseng essence product, which the U.S. FDA declared was dedicated to cancer-related fatigue. However, none of the methods achieved satisfactory results.

Later, Prof. Xu managed to get a small hydrogen-oxygen nebulizer for Ms. Wang. She was instructed to inhale a mixture of 66.6% hydrogen and 33.4% oxygen for 2 to 4 hours a day. She took the nebulizer with her when going back to Beijing. In June 2018, Prof. Xu went to Beijing, where he saw Ms. Wang full of energy and with good appetite. Was this the result of hydrogen inhalation? Prof. Xu once asked Ms. Wang to think about what treatment for fatigue was the most effective for her. Ms. Wang smiled and said: "Frankly, the hydrogen you recommended is the most effective. So far, I have been inhaling hydrogen. It is really effective. It is a good thing."

Ms. Wang looked happy. She was about to go to the northwest, where she worked hard for half a lifetime and shared the fruits of the reform and opening up policy. But after all, she was in her eighties. As a result of environment changes in addition to her advanced age, Ms. Wang developed dermatomyositis accompanied by severe muscle weakness, difficulty in movement and edema of the hands. She was in great misery. Later, she went back to Beijing and was hospitalized immediately. After use of steroids, her condition was controlled.

On the afternoon of November 17, 2018, Prof. Xu Kecheng specially invited Prof. Ge from Beijing and his wife; and the three of them went to visit Ms. Wang together.

Prof. Ge is a cardiovascular expert who has been engaged in clinical research for nearly 40 years in a military hospital. He came from the same native place as Prof. Xu. So, the two have a lot in common. On the way to Ms. Wang's home, they naturally talked about the air and traffic in Beijing, and also about hydrogen.

"Xu, your hydrogen is really magical." A few months ago, Prof. Xu sent Prof. Ge a hydrogen-oxygen nebulizer. On the one hand, he hoped that Prof. Ge, a cardiologist, would test hydrogen for cardiovascular disease. There had been quite some reports about it in Japan. He believed that China should keep up. He wanted Prof. Ge to be his "hydrogen partner". On the other hand, he believed that hydrogen would boost health for Prof. Ge at his currently advanced age.

At the beginning, Prof. Ge did not take the nebulizer seriously and left it in idle. A month later when Prof. Xu met Prof. Ge and asked him how he was doing with hydrogen inhalation, Prof. Ge smiled and responded in ambiguity. Hydrogen medicine is too young a subject. Even the doctors who do not care about it will not understand it. Moreover, hydrogen molecules are very simple. Are they really helpful for health? Although feeling somehow helpless, Prof. Xu hoped to see the question answered with time. Now that Prof. Ge finally showed a positive attitude towards hydrogen, Prof. Xu was happy.

"I tried it last month and felt that my nose was not so obstructed. Then, I inhaled hydrogen for more than ten days. My nasal obstruction and sneezing basically disappeared. Prof. Xu, you made a point." Prof. Ge said very seriously. "I am sorry that I did not take it seriously." "Previously, he always applied dexamethasone and various oils such as salad oil and sesame oil to his nose before going to sleep." Mrs. Ge said, "My husband is now without nasal obstruction and does not sneeze anymore. He tells whoever he meets that hydrogen is good."

> *Picture Influence of hydrogen inhalation on nasal mucosal inflammation. (A) Control group; (B) Allergic rhinitis (AR) group; (C) Control group + H_2 inhalation group; (D) AR + H_2 inhalation group; (E) control group (control) + helium Inhalation (He) group; (F) AR + helium (He) group. Yellow arrows indicate eosinophils and black arrows indicate lymphocytes. The rhinitis hydrogen group had significantly less inflammatory cells than the control group and helium inhalation group.*

There were a lot of cars on the road during the rush hours. Two hours later, the car stopped in front of a purple-red building. It was the "ministerial court" that ordinary people talked about. The building looked like a common residential building, except with guards at the gate. Ms. Wang's husband was waiting. Seeing us, he came close. The "old secretary" dressed simply and behaved in a modest and courteous manner. After short greetings, he led us inside the house.

With guests visiting from the south part of the country, Ms. Wang, who had difficulty in moving, specifically moved from the bed to a wheelchair and came to meet us in the living room. Her edema on the face and arms was significantly reduced. Prof. Xu Kecheng carefully read her medical reports and then concluded that her condition stabilized and that hydrogen exerted positive effects. But Ms. Wang also showed the side effects of steroids. Prof. Ge stood up, went close to Ms Wang and said: "Ms. Wang, make sure to continue with hydrogen inhalation. Hydrogen eliminates these side effects. Look at me. My original allergic rhinitis has disappeared." Then, based on his personal experience, Prof. Ge shared his story of hydrogen and its magical effects on him.

In the evening, everyone talked about hydrogen when they were eating at a restaurant near Ms. Wang's home. At dinner, Prof. Ge was very happy, both because he met his good friend visiting from afar and the hydrogen brought by his good friend gave him great physical comfort.

Prof. Xu was also very happy that Prof. Ge had finally become his "hydrogen partner." Prof. Xu turned on the computer that he took with him and showed Prof. Ge a paper on hydrogen treatment of rhinitis. This paper was written by Chinese scholars and published in Experimental and Therapeutic Medicine.

In the study, researchers used ovalbumin (OVA) - sensitized female BALB/c mice, and established animal models of allergic rhinitis (AR) induced by intranasal ovalbumin stimulation. Then, the mice were exposed to hydrogen and helium, an inert gas, at different frequencies and for different periods of time. The mice's weight and frequency of sneezing and scratching were recorded. The effects of hydrogen on ovalbumin-induced acute rhinitis were assessed through histological analysis and various cytokine assays. Results showed that the hydrogen-inhaling group (AR+H_2) had significantly lower frequency of nasal scratching and sneezing than the control group and helium-inhaling group. The hydrogen-inhaling group showed the most decrease in nasal scratching and sneezing of all the groups. Moreover, the longer and higher frequency of hydrogen inhalation, the more significant the improvement.

From the study, an interesting discovery was that the structure of the nasal mucosa changed after hydrogen inhalation, which manifested as significant decline in inflammatory cell infiltration.

Reading about the above experiment, Prof. Ge smiled and said: "It is a pity. If I took a nasal mucosa biopsy before hydrogen inhalation and another biopsy now, I would be an experimental mouse."

— *Lin Hong*

Words from A Sub-healthy Person After 30 Days of Hydrogen Inhalation

What is sub-health status?

According to western medicine, sub-health status refers to a state in which the body is between health and disease. With the use of modern medicine means, significant changes in disease indicators cannot be detected. However, those in sub-health status show lower vitality and decline in adaptive capacities to varying degrees. They also complain of some uncomfortable physiological conditions.

A few thousands of years ago, Inner Canon of the Yellow Emperor, an ancient Chinese medical classic, proposed to focus on preventing "disease to come", which refers to certain degrees of yin-yang imbalance, Qi-blood disorder, visceral dysfunction or other issues. However, the disease is not yet in its pathological state, and is between disease and health.

The best time for the treatment or intervention of disease is when it is not formed and just formed. As said in Book of the Later Han: Biography of Ding Hong: In all matters, if everything is dutifully performed and hidden dangers are eliminated in the bud, all the unlucky and unfavorable can be prevented, therefore ensuring safety and happiness.

Many studies have shown that hydrogen molecules exert anti-inflammatory and selective antioxidant effects; transform the human micro-environment; and therefore are good for treating "disease to come".

Mr. Shen, who suffered from rhinitis for many years and often had discomfort in the liver, was a typical person in sub-health status. He inhaled hydrogen using a hydrogen-oxygen nebulizer in Xu Kecheng Care for Health Studio for 30 days. As a result, his physical conditions improved significantly. He was thrilled and kept detailed records of his changes and feelings in the past 30 days. The records are as follows:

> *By chance, I attended the lecture Hydrogen and Health organized by Xu Kecheng Care for Health Studio. I remember that at the time of the meeting, an old man slowly walked towards my direction and sat down next to a hydrogen inhalation machine placed in the conference hall. He was in low spirits with gray complexion. He put the nasal tube in his nose and slumped on the chair. Well, he was a patient. I did not pay much attention to him but listened to the lecture carefully. At the end of the lecture 2 hours later, I unconsciously turned to look at the old man near me. I was surprised to see him with glowing face and brightened eyes. I wondered what happened to him just now. Finally, I couldn't help but wanted to experience hydrogen inhalation myself. With the help of Ms. Deng from the studio, I was offered 30-day-long experience with the hydrogen-oxygen nebulizer. That was the start of my wonderful journey towards good health.*

> *Before going to Dr. Xu Kecheng's lecture on hydrogen inhalation, I actually felt that I was in very bad conditions. I had apparent discomfort in the liver area. While walking, I had heavy steps and mild headache. Moreover, I had rhinitis for years. Since hydrogen inhalation was safe, I might as well try it.*
> *After inhaling hydrogen for 1.5 hour a day and two days in a row, I felt that my steps were much lighter and energy grew. Learning about my rhinitis, the studio staff kindly reminded me to increase the mist from the nebulizer in order to enhance the conditioning effects. On the fifth day of hydrogen inhalation, the discomfort in my liver area was significantly alleviated. Interestingly, it seemed that my cravings for cigarettes, which I had smoked for many years, weakened. And I had no strong desire to smoke randomly.*
> *On the 15th day of hydrogen inhalation, I felt that my mental state became completely normal. The rhinitis, which had been plaguing me for years, seemed to have disappeared because of the relief of nasal obstruction. Later, I added selenium-rich food to my daily diet. I had at least one selenium-rich egg every day and tried to drink selenium-rich tea. On the 20th day of hydrogen inhalation, my discomfort in the liver basically disappeared. Also, I have to mention one more surprise. Due to long-term use of mobile phones, I had muscae volitantes, which was relieved after more than 20 days of hydrogen inhalation. It is really amazing.*
> *Looking back at my nearly 30 days of hydrogen inhalation, I found my body gradually returning to a level of comfort. I also want to share the following experience with hydrogen inhalation:*
> *1. Sitting position: Keep the whole body relaxed. Sit with your back in parallel with the back of the hydrogen-inhaling chair. Put your legs on the seat extension board and place your hands naturally on both armrests of the seat. Keep your eyes naturally closed or slightly closed.*
> *2. Breathing: Close your mouth; inhale with your nostrils; inhale evenly; try to prolong the inhalation time of the same breath; and then open your mouth slightly and gently exhale.*
> *3. Recommended cycle: Hydrogen inhalation must be continuous. It is better to keep high-quality hydrogen inhalation for more than one hour every day. First timers are recommended to inhale hydrogen for 7 to 15 days consecutively. Once the signal that the body feels significantly better appears, stop hydrogen inhalation for one to two days. Then, continue to inhale hydrogen according to the previous cycle of days. If your conditions allow, inhale hydrogen for a long time.*
> *4. Suggestions about supplementary measures: Hydrogen inhalation should be an integral part of the body's conditioning. Therefore, medicine should still be taken if necessary. Sub-healthy people are advised to adjust their diet, follow a healthy daily routine, and properly increase the intake of trace elements.*
> *5. Other: Make sure to drink a certain amount of water before and during hydrogen inhalation respectively.*
> *At last, I would like to thank Prof. Xu Kecheng and his studio. I am grateful to Ms. Deng for her kind help. I also want to thank the two female employees in the studio for their meticulous attention and contribution to our health. Surely, I want to acknowledge Shanghai Asclepius Meditec for its hydrogen-oxygen nebulizer. High-concentration medical-level hydrogen is a guarantee for fast recovery.*
> *It is also important to keep nursing health and take the right medicine. I wish all those on hydrogen inhalation better health.*

Prof. Xu Kecheng has a very easy-to-understand description of good health with the following features: good complexion, great vigor, good appetite, gaining weight, unobstructed bowel movement, and smooth urination. Any physical status inconsistent with the description is sub-health. Prof. Xu believes that sub-health is actually a problem with energy metabolism.

Prof. Erwin Schrödinger, winner of the Nobel Prize in Physics and one of the founders of quantum mechanics, pointed out that life is an energy system with an orderly and regular mechanism. All activities of the human body require energy, just as machines need power to

operate. But the effective utilization of human energy is much higher than existing artificial machines. Mitochondria are power plants that produce energy for human cells. The generator set burns food and produces energy-bearing ATP, thereby establishing the order of the living body and maintaining the body's structure and function. If mitochondria dysfunction, energy supply and consumption are insufficient. And the living body falls into a state of disorder. Consequently, the human body generates discomforts or small problems, that is, sub-health conditions. Difficult to identify clinically, the discomforts or small problems greatly compromise the quality of life.

Mitochondrial damage is associated with excessive production of toxic reactive oxygen species in the body. Hydrogen molecules selectively scavenge toxic reactive oxygen species such as hydroxyl groups, maintain mitochondrial function, and correct abnormal energy metabolism. This might be the main mechanism by which hydrogen relieves sub-health conditions.

— Lin Hong

PART 7 — Collection of Published Papers

Hydrogen gas therapy induced shrinkage of metastatic gallbladder cancer: A case report

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Informed consent statement: This clinical trial was approved by the Ethics Committee of Fuda Cancer Hospital, Jinan University and was conducted in accordance with the Declaration of Helsinki. The patient provided written informed consent for publication of this case report and any accompanying images.

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Abstract

BACKGROUND

We present the case of a 72-year-old female patient with gallbladder cancer (GBC) who developed in situ recurrence and liver metastases 9 mo after irreversible electroporation ablation and oral tegafur (a fluoropyrimidine derivative) chemotherapy, which failed to control the progression of the disease. The patient further developed metastases in the lymph nodes around the head of the pancreas. The patient had severe anemia, requiring weekly blood transfusions. The gallbladder tumor invaded the descending part of the duodenum, causing intestinal leakage and hepatic colonic adhesion.

CASE SUMMARY

The patient refused other treatments and began daily hydrogen inhalation therapy. After 1 mo of treatment, the gallbladder and liver tumors continued to progress, and intestinal obstruction occurred. After continuous hydrogen therapy and symptomatic treatments including gastrointestinal decompression and intravenous nutrition support, the intestinal obstruction was gradually relieved. Three months after hydrogen therapy, the metastases in the abdominal cavity gradually reduced in size, her anemia and hypoalbuminemia were corrected, and the patient was able to resume normal life.

CONCLUSION

This is the first report of an efficacy and safety study about hydrogen therapy in patient with metastatic GBC and a critical general condition, who has remained stable for more than 4 months.

Key words: Hydrogen gas; Metastatic gallbladder cancer; Case report

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Core tip: This is the first report of an efficacy and safety study about hydrogen therapy in patient with metastatic gallbladder cancer and a critical general condition, who has remained stable for more than 4 mo.

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INTRODUCTION

Gallbladder cancer (GBC) is the sixth most common gastrointestinal cancer and the sixth highest cause of cancer-related deaths in China. Despite improvements in the diagnosis and treatment of GBC, the majority of the patients are usually confirmed as having metastatic GBC at diagnosis. Due to the high incidence of lymph node or distant metastasis, the 5-year overall survival (OS) rate for GBC is only 5%. In mGBC patients who are not candidates for surgery, the National Comprehensive Cancer Network (NCCN) guidelines recommend the use of biliary drainage, chemotherapy, clinical trials, and the best supportive care available. At present, there are few alternative options, and new and more effective methods are urgently required.

In patients with cancer, the treatment strategy, outcome, and prognosis are strongly dependent on immune status. $CD8^+$ T cells become exhausted by persistent stimulation by tumor antigens, resulting in cessation of proliferation, cytokine production, and immune function. The preventive and therapeutic effects of hydrogen in various diseases, including cancer, have been investigated in several studies. Hydrogen was recently reported to stimulate peroxisome proliferator-activated receptor γ coactivator 1α, which enhances mitochondrial function and may rescue exhausted $CD8^+$ T cells. In an advanced study of the effects of inhaled hydrogen gas in 55 patients with stage IV colorectal cancer, Akagi et al identified accumulation of terminal programmed cell death 1 (PD1) $CD8^+$ T cells as a significant index for poor prognosis, with reduction in terminal $PD1^+$ $CD8^+$ T cells following hydrogen gas treatment found to be significantly associated with improved progression-free survival and OS.

Here, we present the case of a 72-year old Chinese woman with stage IIIA GBC who was treated with irreversible electroporation (IRE) and oral tegafur (a fluoropyrimidine derivative) chemotherapy. Primary recurrence and liver metastatic lesions was observed with continuous application of a hydrogen oxygen nebulizer, and a clinically objective response lasting for more than 4 mo was achieved. The patient remained disease-free at the time of writing this report.

CASE PRESENTATION

Chief complaints

We present the case of a 72-year-old female GBC patient, who was admitted to our hospital due to chest tightness in September 2018.

History of present illness

After examination, GBC was found to recur in situ, invading the duodenal descending part and causing intestinal fistula and hepatic colon adhesion. In addition, multiple metastases of the liver, lymph node metastasis around the head of the pancreas, severe anemia requiring weekly blood transfusion, and symptoms of heart failure were noted.

History of past illness

In December 2017, the 72-year old Chinese patient underwent IRE following diagnosis as GBC (T3N1M0 stage IIIA) based on computed tomography (CT) imaging and pathological findings of GBC (8.0 cm × 3.9 cm) with multiple hilar lymph node invasion (max 4.7 cm × 3.6 cm). She had elevated levels of a variety of serum tumor markers, including CA19-9 (2556 U/mL) and CEA (607.6 ng/mL).

Personal and family history

After IRE ablation of the gallbladder and hilar lymph nodes, oral tegafur (20 mg bid) chemotherapy was administered for 9 mo. CT re-examination in April 2018 showed liquefaction necrosis at the ablation site. During the treatment period, the patient received intermittent red blood cell (RBC) infusions as supportive care for repeated episodes of anemia.

Physical examination upon admission

Accompanied by a progressive increase in pain in the upper right abdomen, a large area of adhesion between the gallbladder and the descending and horizontal segments of the duodenum was found in addition to compression of the inferior vena cava (October 6, 2018).

Laboratory examinations

She had elevated levels of a variety of serum tumor markers, including CA19-9 (88.18 U/mL), AFP (14.11 IU/mL), and CEA (39.68 ng/mL) in September, 2018.

Imaging examinations

After 2 wk of blood transfusions and anti-infective treatment, CT examination again revealed a bladder tumor (6.3 cm × 4.9 cm), with an adjacent descending duodenal fistula, multiple spotted high-density lesions in the liver parenchyma, and dilatation and gas accumulation in the intrahepatic and extrahepatic bile ducts. Enlarged lymph nodes (2.7 cm × 2.1 cm) were visible around the pancreatic head.

FINAL DIAGNOSIS

Metastatic GBC, severe anemia, infection, and gallbladder-duodenal fistula.

TREATMENT

Hydrogen gas treatment

The hydrogen oxygen nebulizer (AMS-H-01, Asclepius Meditec, Shanghai, China) generates 3 L/min hydrogen gas by water electrolysis. As measured by gas chromatography, the gas generated consisted of 67% hydrogen and 33% oxygen. Using a special mask, the patient continued to inhale hydrogen for 3-6 h a day at rest, with no interruption even after the obvious relief of symptoms.

Best supportive care

Due to the long-term difficulty in reversing the anemia in the early stage of hydrogen inhalation therapy, the patient received weekly (1-2) blood infusions, and her RBC and hemoglobin levels were tested regularly. Daily anti-infection treatment was administered for the gallbladder-duodenal fistula, and white blood cell counts were measured regularly. After 1 mo of hydrogen aspiration therapy, the patient presented with intestinal obstruction, which was treated by gastrointestinal decompression and intravenous nutrition. Analgesics and sedatives were prescribed for pain and insomnia, respectively.

Therapeutic procedure

Due to the lack of currently available conventional treatments combined with the patient's poor condition requiring continuous supportive care, hydrogen gas monotherapy was started on October 24, 2018 (Table 1). At the time of writing this report, the patient still continues to receive daily hydrogen therapy.

Table 1. Clinical details and therapeutic procedure

Date	Therapy
2017 December	(A) Symptoms of upper right quadrant distension pain and discomfort were aggravated; (B) Color doppler ultrasound: gallbladder tumor, local liver invasion, and multiple enlarged lymph nodes; (C) Gallbladder tumor aspiration biopsy: gallbladder cancer
2018 January 5	(A) Irreversible electroporation ablation of the gallbladder and hilar lymph nodes; (B) Started oral S-FU (20 mg bid); (C) Infusion of red blood cells for anemia
April	CT examination: gallbladder tumors and hilar lymph nodes showed liquefactive necrosis after treatment
September 20–October 8	(A) Routine blood tests and tumor markers; (B) CT examination: gallbladder tumor invaded the descending

Date	Therapy
	duodenum, inferior vena cava was compressed; (C) Infusion of red blood cells for anemia
October 24	(A) CT examination: gallbladder tumor enlarged, multiple spotted metastases found in the liver, and multiple lymph nodes around the pancreatic head; (B) Started hydrogen gas therapy
November 26	(A) CT examination: gallbladder and intrahepatic tumors enlarged, duodenal bowel obstruction; (B) Gastrointestinal decompression and intravenous nutrition support
December 10–24	(A) Routine blood tests and tumor markers; (B) Gastric tube removed, the patient gradually began taking semi-liquid food, and her spirit, appetite, and sleep were good
2019 January 8–11	(A) Routine blood tests and tumor markers; (B) CT examination: gallbladder and intrahepatic tumors, lymph nodes around the pancreatic head all shrank significantly, and obstruction was markedly relieved

CT: Computed tomography.

Adverse events

According to the Common Terminology Criteria for Adverse Events version 3.0 (National Cancer Institute, Bethesda, MD, United States), adverse events were classified and graded every week for at least 2 mo after the treatment was started. Liver function was evaluated based on the levels of alanine transaminase, aspartate transaminase, total bilirubin, and gamma-glutamyl transpeptidase on multiple occasions during hydrogen treatment. Bone marrow hematopoietic function was evaluated based on peripheral blood erythrocyte counts and hemoglobin levels. Inflammation caused by infection was assessed based on white blood cell counts.

Curative evaluation

The curative effects of the treatment were evaluated according to several aspects of the patient's general condition. Since patients cannot eat normally, serum total protein and albumin levels were monitored after intravenous nutrition. Due to visceral adhesion, intestinal fistula and obstruction, degrees of pain and relief in the upper abdomen were recorded continuously. Absolute lymphocyte counts and serum tumor markers were measured to reflect changes in immune function and tumor activity.

CT imaging changes

Changes in CT tumor imaging were monitored to evaluate the curative effect of hydrogen gas therapy. According to the RECIST 1.1 guidelines, therapeutic effects are categorized as a complete response (CR), characterized as disappearance of tumor detection in all target lesions; partial response (PR), total reduction in the diameter of the target lesions ≥ 30%; stable disease (SD), tumor regression failing to reach PR or progressive disease (PD); or PD, defined as total progression of the tumor diameter ≥ 20%.

OUTCOME AND FOLLOW-UP

Hydrogen gas inhalation was started on October 24, 2018. Within 1 mo of treatment, the patient's condition was relatively stable, and her blood indexes, infection status, and degree of pain gradually improved. Subsequently, intestinal obstruction occurred at the adhesion site between the gallbladder and duodenum; this was relieved after 1 mo of gastrointestinal

decompression and intravenous nutrition. The patient insisted on regular hydrogen therapy throughout the course of this treatment.

Adverse events

The most common adverse reactions recorded included drowsiness or agitation, and no other adverse events were observed. The patient's transaminase levels remained normal throughout the treatment. Occasionally her bilirubin levels exceeded the reference range, but returned to normal after short-term use of hepatoprotective drugs. Despite regular blood transfusions, there was little change in the patient's red blood cell counts and hemoglobin levels during the two weeks before hydrogen treatment was initiated. Both indexes continued to increase after hydrogen therapy and blood transfusions, and both reached the lower limit of the reference range after 2.5 mo of treatment (Figure 1A). The frequency of transfusions was decreased gradually after hydrogen therapy and stopped completely after 2 mo of treatment.

Within 1 mo prior to treatment, the patient had a high inflammatory response to infection and a high white blood cell count. After anti-infection treatment, the white blood cell count was restored to the upper limit of the reference range (9.8×10^9/L), with no further reduction. After 1.5 mo of hydrogen treatment (with no further anti-infection treatment), the leukocyte count decreased to 4.7×10^9/L (Figure 1C left).

Curative evaluation

Serum total protein levels returned to normal shortly after the start of hydrogen therapy and continued to rise. Because of the intestinal obstruction, the patient received intravenous nutrition for 1-2 mo after treatment. Within 2 wk of the removal of the gastric tube, the patient returned to a normal diet and the total protein remained at the upper limit of the reference range. Serum albumin levels returned to normal after 2 mo of hydrogen therapy and continued to rise (Figure 1B).

Prior to hydrogen therapy, extensive adhesion and compression of gallbladder tumors caused severe pain in the upper abdomen that required daily pain medication. This was stopped when the pain gradually diminished after 2 wk of hydrogen treatment. Subsequent pain-induced administration of analgesics was not necessary, even during ileus.

T and NK cells are the major populations of lymphocytes that mediate anti-tumor immunity; therefore, the total number of lymphocytes is positively correlated with the immune function of patients. Before tumor recurrence and metastasis, the total number of lymphocytes continued to rise (1.2×10^9/L), indicating an increase in immune function. However, the total number of lymphocytes declined rapidly with tumor progression, to significantly below the reference range (0.84×10^9/L), reaching a minimum (0.7×10^9/L) during intestinal obstruction (1.5 mo after hydrogen therapy). The lymphocyte numbers then rebounded, reaching 1×10^9/L after 2.5 mo of hydrogen treatment (Figure 1C right).

Before the detection of tumor recurrence (September 20, 2018), all three tumor markers were significantly higher than the reference range, but all showed a slight decline, which may be related to the compensation of immune function before treatment failure. After identification of tumor recurrence and metastasis, all three markers began to increase rapidly. This was consistent with the rapid growth of the tumor, which caused obstruction. After 1.5 mo of hydrogen therapy, the levels of all three markers began to decrease rapidly, falling into the reference ranges after 2.5 mo (Figure 2).

Figure 1. Blood parameters of patients before and after hydrogen treatment. A: Red blood cell counts and hemoglobin levels; the red line in the figures represents the lower limit of the reference range; B: Total protein and albumin concentrations; the red lines in the figures represent the reference range; C: White blood cell and lymphocyte counts; the red lines in the figures represent the reference range.

CT imaging changes

Before hydrogen gas treatment, the sum of the tumor diameters was 12.2 cm; 1 mo after treatment, the sum was 13.1 cm (37% increase, PD); and 2.5 mo after treatment, the sum was 7.5 cm (6% increase, SD) (Figure 3 and Table 2). At the end of February 2019, no tumor recurrence was detected (very similar to the CT examination at 2.5 mo; images not shown). To date, the patient appears completely well (Karnofsky score 100) and has resumed a normal diet and exercise.

DISCUSSION

In GBC patients who are not candidates for surgery, the NCCN guidelines recommend the use of chemotherapy and best supportive care. Due to the large size of tumor in this patient (stage IIIA), the gallbladder function was completely lost with widespread adhesions to the hilum of the liver. Therefore, it was necessary to choose an ablation method that causes minimal damage to the extrahepatic bile duct system in reducing the tumor size. IRE is a novel ablation technology that utilizes short pulses of high voltage electrical energy to induce tissue necrosis. This technique has many advantages, including short ablation time, preservation of the internal structure of vital organs, and lack of the heat/cold-sink effect. We have previously verified the safety and efficacy of gallbladder ablation in a rabbit model. Although not yet included in the NCCN guidelines, IRE has the potential to be a superior alternative to other ablation techniques for the removal of tumors situated near the gallbladder.

IRE, combined with oral chemotherapy, effectively controlled the tumors in the gallbladder and the hilar part of the liver, and re-examination after 3 mo of treatment showed that the tumor was basically necrotic. However, for this patient with advanced GBC, palliative treatment could only delay the disease, with continued slow progression of the tumor. In the re-examination after 9 mo of treatment, recurrence of the gallbladder tumor, duodenal fistula, liver metastases, and continued slow metastases around the head of pancreas as well as severe anemia were detected, which indicated that the disease had entered a stage of rapid and life-threatening progression in a short period of time. Best supportive care was the only option at this timepoint, although in general, it is not possible to reverse the progression of such tumors.

Molecular hydrogen, or H_2, can selectively neutralize hydroxyl radicals, but not other reactive oxygen species (ROS). Hydrogen has been used to treat various states associated with oxidative stress, including trauma, neurodegenerative disease, inflammatory disease, metabolic syndrome, adverse reactions to chemotherapy, and radiation injury. In addition to the apparent improvement in the immune system, there is much evidence that hydrogen directly kills different types of tumors, including cutaneous squamous cell carcinoma, leukemia, tongue cancer, and colon cancer. Since there has been no report on the upper limit of hydrogen use, this therapy might be a very safe method.

At the same time as the delivery of optimal supportive care, the patient was enrolled in this clinical study (24 October 2018) with the intention of improving clinical symptoms. One month after the start of hydrogen therapy, the disease continued to progress, and the levels of tumor markers (including CA19-9, AFP, and CEA) continued to rise, although the patient's general

status and blood indexes (including total protein, albumin, multiple blood cells count, hemoglobin content, etc.) showed continuous improvement, so the patient persisted with the hydrogen treatment. Gradually, the levels of multiple tumor markers began to decline, and multiple hematological indicators continued to improve. The patient underwent gastric tube removal on December 24, and gradually began eating semi-liquid food, with significant improvement in spirit, appetite, and sleep. Subsequent CT examination also confirmed relief of the obstruction, with significant shrinkage of tumors at multiple sites. The reason for the patient's extraordinary recovery, apart from symptomatic treatment, is probably because of the persistence of hydrogen therapy; this therapy not only mediates rapid improvement in the physical condition of the patient, but also significantly reduces tumor marker levels, increases lymphocyte counts, and even induces tumor shrinkage.

> *Figure 2. Changes in tumor markers before and after hydrogen treatment. The red lines in the figures represent the upper limit of the reference ranges.*

At present, there is only a temporal connection between hydrogen and improvement of this patient's cancer. The proposed mechanism underlying the anti-cancer properties of hydrogen may involve: (1) Elimination of the high levels of ROS produced by cancer cells; (2) Downregulation of the proliferation, invasion, and migration of cancer cells; (2) Downregulation of inflammatory factors, elimination of chronic inflammation, and modification of the microenvironment; (3) Protection of mitochondria, which maintains energy generation and help correct hypoxia, thereby transforming the cardiovascular, endocrine, and nervous systems; and (4) Restoration of the function of exhausted cytotoxic T cells, and enhancement of systemic anti-cancer effects. The detailed mechanism remains to be fully elucidated in further follow-up studies, and indications for advanced cancer therapy require verification in studies with greater numbers of patients.

> *Table 2. Tumor sizes before and after hydrogen gas treatment*

	Gallbladder	Liver metastases	Lymph node around the pancreas head	Sum of the tumor diameters
Pretreatment	6.3 cm × 4.9 cm	—	2.7 cm × 2.1 cm	12.2 cm
1 mo after treatment	6.7 cm × 6.3 cm	3.2 cm × 2.6 cm	2.2 cm × 1.8 cm	16.7 cm
2.5 mo after treatment	3.7 cm × 3.6 cm	4.2 cm × 4.0 cm (large), 3.6 cm × 3.2 cm (small), 4.0 cm × 3.3 cm (large), 3.2 cm × 2.8 cm (small)	2.0 cm × 1.6 cm	12.9 cm

CONCLUSION

This is the first report on the rehabilitation of an advanced GBC patient with a critical general condition, whose disease has gradually improved and has survived for more than 4 mo.

> *Figure 3. Computed tomography imaging findings at different time points before and after hydrogen treatment. In the first line of figures, the triangle and the contour lines represent the location and range of the gallbladder. In the second line of figures, the contour lines represent the location and range of liver metastases. In the third line of figures, the arrows represent the location of the duodenal fistula or obstruction.* (Columns: 06/10/2018 half mo before; 24/10/2018 d0 of treatment; 27/11/2018 1 mo after; 12/01/2019 2.5 mo after. Rows: Gallbladder tumor; Liver metastases; Duodenal fistula and obstruction.)

References

1. Chen W, Zheng R, Baade PD, Zhang S, Zeng H, Bray F, Jemal A, Yu XQ, He J. Cancer statistics in China, 2015. CA Cancer J Clin 2016; 66: 115-132
2. Shi JS, Wang JS, Liu G, Ye YL, Lu Y, Jiao XY, Yang YJ, Li GC, Han Y. Early diagnosis of primary gallbladder carcinoma. Hepatobiliary Pancreat Dis Int 2002; 1: 273-275
3. Hensel MT, Vollmer CM, Pawlik TM. Evolving treatment strategies for gallbladder cancer. Ann Surg Oncol 2009; 16: 2101-2115
4. Hundal R, Shaffer EA. Gallbladder cancer: epidemiology and outcome. Clin Epidemiol 2014; 6: 99-109
5. Barber DL, Wherry EJ, Masopust D, Zhu B, Allison JP, Sharpe AH, Freeman GJ, Ahmed R. Restoring function in exhausted CD8 T cells during chronic viral infection. Nature 2006; 439: 682-687
6. Wherry EJ. T cell exhaustion. Nat Immunol 2011; 12: 492-499
7. Ge L, Yang M, Yang NN, Yin XX, Song WG. Molecular hydrogen: a preventive and therapeutic medical gas for various diseases. Oncotarget 2017; 8: 102653-102673
8. Runtuwene J, Amitani H, Amitani M, Asakawa A, Cheng KC, Inui A. Hydrogen-water enhances 5-fluorouracil-induced inhibition of colon cancer. PeerJ 2015; 3: e859
9. Wang D, Wang L, Zhang Y, Zhao Y, Chen G. Hydrogen gas inhibits lung cancer progression through targeting SMC3. Biomed Pharmacother 2018; 104: 788-797
10. Kamimura N, Ichimiya H, Iuchi K, Ohta S. Molecular hydrogen stimulates the gene expression of transcriptional coactivator PGC-1α to enhance fatty acid metabolism. NPJ Aging Mech Dis 2016; 2: 16008
11. Handschin C, Spiegelman BM. Peroxisome proliferator-activated receptor gamma coactivator 1 coactivators, energy homeostasis, and metabolism. Endocr Rev 2006; 27: 728-735
12. Akagi J, Baba H. Hydrogen gas restores exhausted CD8+ T cells in patients with advanced colorectal cancer to improve prognosis. Oncol Rep 2019; 41: 301-311
13. Basch E, Reeve BB, Mitchell SA, Clauser SB, Minasian LM, Dueck AC, Mendoza TR, Hay J, Atkinson TM, Abernethy AP, Bruner DW, Cleeland CS, Sloan JA, Chilukuri R, Baumgartner P, Denicoff A, St Germain D, O'Mara AM, Chen A, Kelaghan J, Bennett AV, Sit L, Rogak L, Barz A, Paul DB, Schrag D. Development of the National Cancer Institute's patient-reported outcomes version of the common terminology criteria for adverse events (PRO-CTCAE). J Natl Cancer Inst 2014; 106
14. Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, Dancey J, Arbuck S, Gwyther S, Mooney M, Rubinstein L, Shankar L, Dodd L, Kaplan R, Lacombe D, Verweij J. New response evaluation criteria in solid tumours: revised RECIST guideline (version 1.1). Eur J Cancer 2009; 45: 228-247
15. Wagstaff PG, Buijs M, van den Bos W, de Bruin DM, Zondervan PJ, de la Rosette JJ, Laguna Pes MP. Irreversible electroporation: state of the art. Onco Targets Ther 2016; 9: 2437-2446
16. Zeng J, Qin Z, Zhou L, Fang G, Chen J, Niu L, Liang B, Xu K. Comparison between cryoablation and irreversible electroporation of rabbit liver at a location close to the gallbladder. Radiol Oncol 2017; 51: 40-46
17. Chen X, Ren Z, Zhu T, Zhang X, Peng Z, Xie H, Zuo L, Liang B, Xu K. Electric Ablation with Irreversible Electroporation (IRE) in Vital Hepatic Structures and Follow-up Investigation. Sci Rep 2015; 5: 16233
18. Herwald SE, Chen JH, Arellano RS. Irreversible Electroporation for Treatment of Hepatocellular Carcinoma Adjacent to the Gallbladder. J Vasc Interv Radiol 2017
19. Siddiqui IA, Kirks RC, Latouche EL, DeWitt MR, Swet JH, Baker EL, Vrochides D, Iannitti DA, Davalos RV, McKillop IH. High-Frequency Irreversible Electroporation: Safety and Efficacy of Next-Generation Irreversible Electroporation Adjacent to Critical Hepatic Structures. Surg Innov 2017; 24: 276-283
20. Ohsawa I, Ishikawa M, Takahashi K, Watanabe M, Nishijima K, Yamaguchi K, Katsura K, Katayama Y, Asoh S, Ohta S. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med 2007; 13: 688-694

21. Chen T, Tao Y, Yan W, et al. Protective effects of hydrogen-rich saline against traumatic brain injury via reducing oxidative stress. J Surg Res 2012; 178: e9-16
22. Ren J, Ma H, Hou J, Xiao W, Jin WH, Wu J, Fan XH, et al. Hydrogen-rich saline as a novel antioxidant in a rat model of inflammation. (J Surg Res / Mediators Inflamm)
23. Nakao A, Toyoda Y, Sharma P, Evans M, Guthrie N. Effectiveness of hydrogen rich water on antioxidant status of subjects with potential metabolic syndrome — an open label pilot study. J Clin Biochem Nutr 2010; 46: 140-149
24. Kikkawa S, Nakagawa Y, Taniguchi M, et al. Hydrogen-rich saline protects against ischemic injury induced free radicals. Neurosci Lett 2014; 579: 125-129
25. Watanabe S, Fujita M, Ishihara M, Tachibana S, Yamamoto Y, Kaji T, Kawauchi T, Kanatani Y. Protective effect of inhalation of hydrogen gas on radiation-induced dermatitis and skin injury in rats. J Radiat Res 2014; 55: 1107-1113
26. Dole M, Wilson FR, Fife WP. Hyperbaric hydrogen therapy: a possible treatment for cancer. Science 1975; 190: 152-154
27. Roberts BJ, Fife WP, Corbett TH, Schabel FM. Response of five established solid transplantable mouse tumors and one mouse leukemia to hyperbaric hydrogen. Cancer Treat Rep 1978; 62: 1077-1079
28. Saitoh Y, Okayasu H, Xiao L, Harata Y, Miwa N. Neutral pH hydrogen-enriched electrolyzed water achieves tumor-preferential clonal growth inhibition over normal cells and tumor invasion inhibition concurrently with intracellular oxidant repression. Oncol Res 2008; 17: 247-255
29. Saitoh Y, Yoshimura Y, Nakano K, Miwa N. Platinum nanocolloid-supplemented hydrogendissolved water inhibits growth of human tongue carcinoma cells preferentially over normal cells. Exp Oncol 2009; 31: 156-162
30. Yang J, Zhu Y, et al. Anti-inflammatory and antitumor action of hydrogen via reactive oxygen species. Oncol Lett 2018; 16: 2771-2776
31. Huang L. Molecular hydrogen: a therapeutic antioxidant and beyond. Med Gas Res 2016; 6: 219-222
32. Hu Z, Wu B, Meng F, Zhou Z, Lu H, Zhao H. Impact of molecular hydrogen treatments on the innate immune activity and survival of zebrafish (Danio rerio) challenged with Aeromonas hydrophila. Fish Shellfish Immunol 2017; 67: 554-560
33. (final reference)

"Real world survey" of hydrogen-controlled cancer: a follow-up report of 82 advanced cancer patients

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Abstract

Advanced cancer treatment is a huge challenge and new ideas and strategies are required. Hydrogen exerts antioxidant and anti-inflammatory effects that may be exploited to control cancer, the occurrence and progression of which is closely related to peroxidation and inflammation. We conducted a prospective follow-up study of 82 patients with stage III and IV cancer treated with hydrogen inhalation using the "real world evidence" method. After 3-46 months of follow-up, 12 patients died in stage III or IV. After 4 weeks of hydrogen inhalation, patients reported significant improvements in fatigue, insomnia, anorexia and pain. Furthermore, 41.5% of patients had improved physical status, with the best effect achieved in lung cancer patients and the poorest in patients with pancreatic and gynecologic cancers. Of the 58 cases with one or more abnormal tumor markers, the markers were decreased at 13-45 days (median 23 days) after hydrogen inhalation in 36.2%. The greatest marker decrease was in achieved lung cancer and the lowest in pancreatic and gynecologic cancers. Of the 80 cases with tumors visible in imaging, the total disease control rate was 57.5%, with complete and partial remission appearing at 21-80 days (median 55 days) after hydrogen inhalation. The disease control rate was significantly higher in stage III patients than in stage IV patients (83.0% and 47.7% respectively), with the lowest disease control rate in pancreatic cancer patients. No hematological toxicity was observed although improved adverse reactions that resolved spontaneously were seen in individual cases. Hydrogen inhalation is a simple, low-cost treatment with few adverse reactions that may improve patients' quality-of-life and control cancer progression. Hydrogen inhalation has potential as a strategy for clinical rehabilitation of patients with advanced cancer. The study protocol received ethical approval from the Ethics Committee of Fuda Cancer Hospital of Jinan University on December 7, 2018 (approval number: Fuda20181207).

Key words: hydrogen inhalation therapy; advanced cancer; real world survey; disease control rate; physical status; tumor marker; tumor stage; life quality; clinical rehabilitation

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INTRODUCTION

Cancer is the leading cause of death worldwide, and its incidence is still rising. Over the past half century, there has been significant progress in basic and clinical research on cancer, with a decline in the incidence and mortality of certain cancers. This is largely due to primary prevention (e.g., smoking cessation) and early cancer screening, rather than the effectiveness of any particular drug. The current treatments including radiotherapy, chemotherapy, fine-needle puncture and surgery can be a double-edged sword, because their use can increase circulating tumor cells, and promote cancer progression and distant metastasis. Precision treatment strategies based on molecular detection have not shown benefit in most cancer patients, with the development of precision drugs available for only 3-13% of patients. Studies have shown that targeted therapy induces high proteome secretion, leading to cancer metastasis. Thus, alternative cancer treatments are required to avoid these challenges.

Molecular hydrogen biology and hydrogen therapy are new and rapidly developing areas of research. Hydrogen has been shown to exert antioxidant and anti-inflammatory effects, and since the occurrence and progression of cancer are closely related to peroxidation and inflammation, hydrogen may play a role in the control of cancer. Using the "real world evidence" method, we conducted a retrospective and prospective follow-up study on advanced cancer patients recovering from hydrogen therapy.

SUBJECTS AND METHODS

Subjects

We analyzed two sets of patients: (1) Retrospective investigation of patients (n = 14) treated with hydrogen inhalation at home from March 2014 to April 2018, mainly through home visits and hospital medical records. (2) Prospective study of 68 patients enrolled after May 2018. Hydrogen inhalation was carried out mainly in the Guangdong Xukecheng Health Care Studio, China. Specialized doctors provided care and guidance to the volunteers, reviewed relevant materials as required, and carried out "prospective" observational studies. The study protocol received ethical approval from the Ethics Committee of Fuda Cancer Hospital of Jinan University on December 7, 2018 (approval number: Fuda20181207) and conformed to the specifications of the World Medical Association's Declaration of Helsinki. Written informed consent was obtained from each participant. This study followed the Reporting of OBservational studies in Epidemiology (STROBE) Statement.

All patients who met the following inclusion criteria were included: (1) advanced cancer patient according to definite pathological and imaging diagnosis; (2) the patient failed to respond to conventional treatments, could not receive conventional treatment due to systemic conditions and diseases, or rejected the conventional treatments; (3) the patient had cancer symptoms, such as poor appetite, fatigue, insomnia and pain; (4) patient with existing tumors or/or abnormal tumor markers; (5) aged 20-80 years, without obvious cardiac, cerebral, pulmonary and renal failure and mental abnormalities, able to describe symptoms.

Hydrogen inhalation method

Hydrogen was produced by a hydrogen-oxygen nebulizer (AMS-H-01, Shanghai Asclepius Meditec Co., Ltd., Shanghai, China). The patient remained seated or recumbent, and inhaled a mixture of hydrogen (66.7%) and oxygen (33.3%) at low rate of 3000 mL/min. The hydrogen inhalation should be continued for > 3 hours per day for at least 3 consecutive months.

Imaging evaluation

Responses were evaluated according to the RECIST (Response Evaluation Criteria in Solid Tumors) criteria on the basis of changes in tumor size detected by computed tomography or magnetic resonance imaging as follows: complete response (CR), all the target lesions disappeared, no new lesions appeared, and the tumor markers were normal for at least 4 weeks; partial response (PR), reduction in the maximum diameter of the target lesion by 30% for at least 4 weeks; stable disease (SD), the sum of the maximum diameter of the target lesion did not decrease by PR or increase by progressive disease; progressive disease, the maximum diameter of the target lesion increased by at least 20%, or new lesions appeared. The disease control rate (DCR) was calculated as CR + PR + SD as a proportion of the total number of patients treated.

Quality-of-life score

The treatment quality-of-life was evaluated using the Quality of Life Questionnaire Core 30 (QLQ-C30) scale of the European Organization for Research and Treatment of Cancer. The QLQ-C30 scale has a total of 30 items including 5 functional subscales (physical function, role function, cognitive function, emotional function and social function); one general health/quality-of-life subscale; and 3 symptom subscales (fatigue, pain, nausea/vomiting), 6 individual measures and 1 overall quality-of-life scale. Evaluation was conducted every other week before and after hydrogen inhalation. The evaluation was performed by a clinician who was blinded to the disease condition.

Physical condition score

The physical condition of patients was evaluated before and after treatment using the five-point Zubrod-ECOG-WHO score system of the United States' Eastern Cooperative Oncology Group (ECOG). Physical condition was classified as follows: improvement, score decrease of 1 point or more; deterioration, score increase of 1 point or more; stability, no change in score.

Detection of tumor markers

Serum tumor markers were measured using an automatic electrochemiluminescence system (e411; Roche, Basel, Switzerland) for quantitative determination of markers including alpha fetal protein, carcinoembryonic antigen (No. 11731629, Roche), carbohydrate antigen (CA)19-9 (No. 11776193, Roche), CA125 (No. 11776223, Roche) and CA153 (No. 03045838, Roche). The selection of these markers was based on abnormal indicators reported in the medical records, with each patient having a range of elevated markers (0, 1 or multiple) at the time of diagnosis.

Statistical analysis

Paired t-tests were used to analyze differences in quality-of-life scores between groups, and chi-squared tests were used to analyze differences in physical fitness, tumor markers and imaging evaluation between groups. Significant differences were indicated by $P < 0.05$. All analyses were conducted using GraphPad software (GraphPad Software, San Diego, CA, USA).

RESULTS

Clinical data of 82 advanced cancer patients treated with hydrogen inhalation rehabilitation

A total of 82 patients were followed up (14 at home and 68 patients in our clinic). The clinicopathological data of patients who received hydrogen inhalation therapy are shown in Table 1.

Follow-up time and survival of 82 advanced cancer patients treated with hydrogen inhalation rehabilitation

All patients were followed up for 3-46 months, with a median time of 6 months. The follow-up continued for 3-6 months in 45 cases (55%), for 7-12 months in 33 cases (40%), and only died (all in stage IV) in 1 case each for 13, 14, 26 and 46 months. Twelve patients died (all tumor progression): two cases of liver cancer (liver failure and infection for one case), two cases of pancreatic cancer (programmed cell death protein-1 (PD-1) antibody-associated pneumonia), three cases of gynecologic malignancy (one case each of intestinal obstruction, abdominal infection, and tumor progression), one case of gastrointestinal malignancy (massive upper gastrointestinal bleeding), and one other case (infection).

Quality-of-life

Thirty patients were prospectively evaluated by the QLQ-C30 score. After 2 weeks of hydrogen inhalation, patients reported a reduction in dyspnea and an increase in appetite, as well as significant improvements in their physical, role, and emotional functions, with reduced fatigue, nausea, vomiting, pain, and insomnia. After 4 weeks of treatment, cognitive function, pain, appetite, constipation and diarrhea were all significantly improved (Table 2).

Physical fitness evaluation

The physical fitness of patients was evaluated using the Zubrod-ECOG-WHO score system. After 3 months of hydrogen inhalation therapy, 42% of the patients showed improvement in physical condition, 34% showed stability, and 24% showed deterioration. The improvement rate in patients treated with hydrogen inhalation alone was 32%, and that of patients who received hydrogen inhalation therapy combined with other treatments was 44%; and there was no significant difference between the two groups (P = 0.295). Compared with before treatment, the improvement rate of stage III patients was 57%, and that of stage IV patients was 36%, with no significant difference between the groups (P = 0.572). There was variation in the improvements in physical fitness among patients with different tumor types. The greatest improvement in physical fitness was observed in lung cancer patients (68%), while pancreatic cancer patients (0%) and patients with gynecological tumors (12%) showed the least improvement. The improvement of patients with lung cancer was significantly greater than that of patients with gynecological and pancreatic cancers (Table 3).

Changes in tumor markers of 82 advanced cancer patients treated with hydrogen inhalation rehabilitation

Before hydrogen treatment, a total of 58 patients had elevated tumor markers. Three months after hydrogen treatment, tumor markers were decreased in 36% of patients, 16% showed no changes and 48% showed increase. Typical cases with decreased tumor markers are shown in Figure 1. The initial decline in tumor markers occurred over the period of 13-45 days (median 23 days) after treatment. Compared with the expression levels before treatment, the decrease in tumor marker expression in the patients who received hydrogen inhalation therapy alone and those who received combined treatments was 22% and 42%, respectively, with no significant difference between the two groups of patients (P = 0.3131). No elevation of tumor markers was observed after hydrogen inhalation therapy in patients with normal tumor markers before treatment. Fifty-eight patients with elevated biomarkers were analyzed according to their different tumor types. After 3 months of hydrogen inhalation therapy, the greatest decrease in tumor markers was observed in patients with lung cancer (75%), while no decrease was observed in patients with pancreatic and liver cancers (Table 4). There were significant differences in the changes in tumor markers post-treatment between patients with lung cancer and those with gynecologic, liver cancer and pancreatic cancer.

Tumor response of 82 advanced cancer patients treated with hydrogen inhalation rehabilitation

Of the 82 patients who participated in the study, 80 had tumors before enrollment, and the other two patients only had elevated tumor markers. Among the 80 patients with tumors, one (1%) showed CR, 15 (19%) showed PR, 30 (38%) showed SD, and 34 (42%) showed progressive disease after 3 months of hydrogen inhalation treatment, with a DCR of 57.5%. The typical case of tumor reduction is shown in Figure 2. CR and PR occurred within 21-80

days, with a median time of 55 days. The DCR in the patients who received hydrogen inhalation therapy only was 54%, and 59% in the patients who received hydrogen inhalation therapy combined with other treatments, with no significant difference in the DCR between the two groups (P = 0.5917). From the perspective of tumor staging, the DCR of stage III patients (83%) was significantly higher than that of stage IV patients (48%) (Table 5). The higher DCR was achieved for patients with lung cancer (79%), while pancreatic cancer patients showed the lowest DCR (20%) and was the only group to show a significant difference in DCR when compared with lung cancer patients (P = 0.0161).

Side-effects of 82 advanced cancer patients treated with hydrogen inhalation rehabilitation

Of the 29 patients treated with hydrogen inhalation alone, no hematological toxicity was found (data not shown for blood routine test), 1 patient developed stomach discomfort and chills, 1 patient developed occasional dizziness (which disappeared spontaneously after several days). Of the 53 patients treated with hydrogen inhalation combination treatments, 1 patient developed headache (which disappeared spontaneously within 3-5 days), 1 developed occasional epistaxis (minor bleeding and gradually resolved), and 1 patient reported dryness of the nasal cavity, which disappeared a few hours after the inhalation stopped.

Table 1. Clinicopathological data of advanced cancer patients who received hydrogen inhalation therapy

Parameters	Data
Sex — Female	29 (35)
Sex — Male	53 (65)
Age (year) — 20–40	26 (32)
Age (year) — 41–60	8 (10)
Age (year) — 61–85	48 (58)
Cancer type — Non-small cell lung cancer	26 (32)
Cancer type — Hepatocellular cancer	19 (23)
Cancer type — Gynecologic malignancy	11 (13)
Cancer type — Pancreatic cancer	16 (20)
Cancer type — Breast cancer	10 (12)
Cancer type — Gastrointestinal malignancy	6 (7)
Cancer type — Urinary system malignancy	6 (7)
Cancer type — Others	6 (7)
TNM stage — III	8 (10)
TNM stage — IV	21 (26)
Concomitant therapy — None	61 (74)
Concomitant therapy — Chemotherapy, targeted therapy or endocrine therapy	28 (34)

Note: Data represent as n (percent), and analyzed by chi-squared test. TNM: tumor, lymph node, and metastasis.

Table 2. Quality-of-life scores of advanced cancer patients who received hydrogen inhalation therapy

Evaluation indicator	Pre-treatment score	Two weeks post-treatment	P-value (2 wk vs. pre)	Four weeks post-treatment	P-value (4 wk vs. pre)
Physical function	55.06±23.64	63.98±20.30	0.0017	68.06±22.93	0.0010

Evaluation indicator	Pre-treatment score	Two weeks post-treatment	P-value (2 wk vs. pre)	Four weeks post-treatment	P-value (4 wk vs. pre)
Role function	59.14±33.57	69.89±28.68	0.0045	76.88±30.63	0.0004
Cognitive function	52.15±26.78	62.22±29.40	0.2850	65.05±29.61	0.0004
Emotional function	51.08±33.32	62.63±30.42	0.0004	65.59±29.56	0.0035
Fatigue	35.48±34.89	43.37±27.42	0.0002	37.99±29.65	0.0024
Pain	61.65±30.20	32.80±31.47	0.3617	21.50±26.59	0.0001
Nausea/vomiting	23.12±32.68	11.29±23.33	0.0019	10.75±22.99	0.0009
Dyspnea	30.11±29.00	19.35±24.00	0.0390	18.28±24.10	0.0374
Insomnia	65.59±29.17	51.61±28.34	0.0003	41.93±28.50	0.0463
Anorexia	48.39±38.34	37.63±36.25	0.0305	26.88±33.80	0.0012
Constipation	36.56±34.81	32.26±31.60	0.3542	23.65±28.79	0.0024
Diarrhea	19.35±28.25	15.05±22.51	0.2547	9.68±17.62	0.0435

Note: Data represent the mean ± standard deviation (n = 82), and analyzed by paired t-test.

> *Table 3. Physical status of advanced cancer patients with different cancer types* (columns: Cancer type | Patients (n) | Improvement (%) | Stabilization (%) | Deterioration (%) | P-value)

Cancer type	Patients (n)	Improvement (%)	Stabilization (%)	Deterioration (%)
Non-small cell lung cancer	19	68	11	21
Hepatocellular cancer	11	36	36	27
Gynecologic malignancy	16	12	56	31
Pancreatic cancer	6	0	75	25
Breast cancer	10	25	50	25
Gastrointestinal malignancy	6	33	50	17
Urinary system malignancy	6	33	50	17
Others	8	25	67	—

Note: Data represent percent, and analyzed by chi-squared test. (P-value column omitted — values 0.2178, 0.0039, 0.2159, 0.0020, 0.1037, 0.1166 are printed but their row alignment was not legible enough to assign reliably.)

> *Table 4. Changes in tumor markers in advanced cancer patients with different tumor types* (columns: Cancer type | Patients (n) | Decrease (%) | No change (%) | Increase (%) | P-value)

Cancer type	Patients (n)	Decrease (%)	No change (%)	Increase (%)
Non-small cell lung cancer	16	75	—	—
Gynecologic malignancy	16	—	—	—
Pancreatic cancer	8	15	6	—
Hepatocellular cancer	7	0	15	19
Gastrointestinal malignancy	5	0	29	—
Breast cancer	4	40	25	69
Others	2	25	0	71

Note: Data represent percent, and analyzed by chi-squared test. (Several cells and the P-value column — values 0.0059, 0.0041, 0.0025, 0.1939, 0.0680, 0.5886 — are printed but were not legible enough to assign reliably; a sharper close-up of Tables 1, 3 and 4 would confirm the dense numeric cells.)

> *Figure 1. The tumor markers were decreased in the typical advanced cancer cases treated with hydrogen inhalation rehabilitation. (A, B) Miss Y, 28 years old. The uterus and attachments were removed because of the left ovarian cancer in December 2017. After 3 months, the tumor under examination was elevated twice, and there were no symptoms on May 3, 2018, 3-4 hours daily. After 2 weeks, the tumor markers CA19-9 (A) and carcinoembryonic antigen (CEA) (B) were reviewed and completely reduced to normal (dotted lines in the figure). Until the beginning of 2019, there was no recurrence.*

> *Table 5. Tumor response of stage III and IV advanced cancer patients with hydrogen inhalation rehabilitation*

Stage	Patients (n)	CR (%)	PR (%)	SD (%)	PD (%)	DCR (%)	P-value
III	19	5	21	58	16	84	0.0167
IV	61	0	18	31	51	49	

Note: Data are analyzed by chi-squared test. CR: Complete response; PR: partial response; SD: stable disease; PD: progressive disease; DCR: disease control rate, which was calculated as CR + PR + SD.

> *Figure 2. The tumor reduction in the typical advanced cancer cases treated with hydrogen inhalation rehabilitation. (A) Miss Z, 52-year-old, had irritable cough and was diagnosed with positron emission tomography (PET)-computed tomography (CT) scanning in October 27, 2016, showed right middle lung mass (4.1 cm × 3.9 cm) with hilar invasion. (B, C) Bronchial endoscopy showed that a hemorrhagic-erosive lesion was located at the right main bronchus (white arrow), and biopsy showed poorly-differentiated squamous cell carcinoma (C). (D) The patient refused chemotherapy and inhaled hydrogen from November 4, 2016, at least 4 hours a day. After 1 week, the cough was reduced and the breathing became smooth. After 2.5 months, on January 13, 2017, PET-CT was performed and the original lesions in the lungs were not seen. So far, the patient has disease-free survival up to now.*

DISCUSSION

Investigations of the potential of hydrogen to treat tumors have a long history and a great deal of evidence. As early as 1975, Dole et al reported that hydrogen inhibited cancer. Nude mice with cutaneous squamous cell carcinoma were administered a mixture of 97.5% hydrogen and 2.5% oxygen (at a pressure of 8 atmospheres), and the tumor shrank significantly after 2 weeks, while the control group receiving hyperbaric oxygen-helium showed no such changes. In 2008, Saitoh et al reported that neutral pH hydrogen-enriched electrolytic water inhibited the colony formation efficiency and colony size of human HSC-4 tongue cancer cells while having no significant inhibitory effect on normal tongue epithelial cells and also inhibited the growth and migration ability of human HT-1080 fibrosarcoma cells. In 2009, Saitoh et al also reported that enhanced platinum-colloid hydrogen water-containing water had an inhibitory effect on the colony formation and size of human HSC-4 tongue cancer cells. In 2011, Zhao et al found that hydrogen prevented the occurrence of radiation-induced lymphoma. In 2015, Runtuwene et al reported that hydrogen increased the survival rate of colon 26 carcinoma tumor-bearing mice, induced apoptosis of cancer cells, and enhanced the sensitivity of colon cancer cells to 5-fluorouracil. Our observations of 82 patients with advanced cancers who

received hydrogen inhalation therapy with spontaneous breathing suggest that hydrogen therapy acts in four ways.

First, hydrogen inhalation therapy improves the quality-of-life of patients. After hydrogen inhalation for 2 weeks, there was significant improvement in breathing, appetite, fatigue and insomnia and after 4 weeks, significant improvements in pain, constipation and diarrhea were observed. Second, hydrogen inhalation therapy can improve physical fitness. After 3 months, lung cancer patients showed the most significant physical improvement, whereas the poorest effect was observed for patients with gynecological and pancreatic cancers. Third, hydrogen inhalation therapy can reduce tumor markers. After 3 months, lung cancer patients showed the most significant reduction in markers, whereas the poorest effect was observed for patients with gynecological, liver and pancreatic cancers. Fourth, hydrogen inhalation therapy can control cancer progression. After 3 months, the tumor control rate of stage III patients was much higher than that of stage IV patients, with the best effect achieved in patients with lung cancer and the worst in those with pancreatic cancer.

The treatment of advanced cancer is a major challenge. Of the cancer drugs approved by the US and European drug administrations, 67% and 57% showed no evidence of long-term survival and improved quality-of-life, respectively. Liang et al found that the tumor microenvironment associated with chemo- and radiotherapy enhanced the malignancy of cancer cells leading to a stem-like phenotype. In a study of 413 patients with brain metastasis of different causes, Amelot et al found that anticancer drugs, especially paclitaxel and its derivatives, promoted brain metastasis and reduce disease-free survival of patients. A recent review suggested that all commonly used treatments, including radiotherapy, chemotherapy, fine-needle puncture, and surgery, can cause an increase in circulating tumor cells and promote cancer progression and distant metastasis. Thus, the positive effects of hydrogen inhalation on cancer patients are very encouraging.

According to our follow-up observations, the effect of hydrogen on the control of cancer is not only related to its type, but also to the mode of delivery. In this study, hydrogen was inhaled at 66.7%, with a flow rate of 3000 mL/min. Since several researchers reported in 2007 that inhaling 2% hydrogen can attenuate cerebral ischemia/reperfusion injury, most researchers have used 1-4% hydrogen to study the molecular biological effects of hydrogen. Reports of the effects of high pressure or high concentration hydrogen are rare, although Dole et al reported that 97.5% hydrogen successfully inhibited skin squamous cell carcinoma. In addition, the extremely high diffusivity of hydrogen provides a higher requirement for daily and total inhalation time. Yamamoto et al studied the distribution of hydrogen in rats after continuous inhalation. The hydrogen concentration in rats after a single dose gradually increased with time for the first 20 minutes, reaching the highest concentration in liver and the lowest in kidney. The inhalation time in this study exceeded 3 hours per day, with some patients receiving treatment for 8 hours continuously. It can be speculated that this may account for the fall in tumor markers at 13-45 days (median 23 days) and the appearance of CR and PR at 21-80 days (median 55 days).

This is the first report of hydrogen inhalation therapy resulting in decreased tumor markers and tumor shrinkage. Furthermore, oxygen, not pure hydrogen. Hypoxia can induce metabolic modification and promote the progression of malignant tumor progression. Conversely, improving hypoxia may inhibit cancer progression. Thus, our findings demonstrate that inhaling oxygen along with hydrogen helps control cancer progression.

Finally, hydrogen can be delivered through inhalation, drinking hydrogen-rich water and injecting hydrogen-rich saline. Hydrogen is an inert gas and its solubility in water is very low, so it is difficult to ensure that the required concentration in tissues is achieved by drinking hydrogen-rich water. It has been reported that hydrogen in the brain increases detectable after drinking hydrogen-rich water. Although hydrogen-rich saline injection can rapidly increase the concentration of hydrogen in the blood, continuous infusion of large amounts of saline will cause water-salt retention. Hydrogen inhalation is the simplest method of delivery and inhaling 66.7% hydrogen can achieve rapid rises in hydrogen concentration in tissues, generally reaching a peak of 520 μM within 30 minutes. Sustained 600 μM hydrogen, which inhibits cancer cells growth in vitro. The efficiency of hydrogen inhalation is easily affected by the inhalation tool (the efficiency of masks is higher than that of a nasal catheter), the evenness and stability of respiration, and the state of the basic lung function.

In 2014, Hanahan criticized the use of the term "cancer war," which increased the use of toxic drugs or overly aggressive treatment for incurable cancer. This term resulted in many patients with advanced cancer receiving highly-recommended treatments with severe side-effects rather than being offered access to palliative care. Haines proposed that the time has come to change the term "cancer war." The selective antioxidant, anti-inflammatory, and signal-conditioning effects of hydrogen may play a major role in improving the cancer microenvironment. More recently, molecular hydrogen has been also been associated with immune function, reviving $CD8^+$ T cells in an exhausted state by maintaining mitochondrial function and restoring their anti-tumor function, thus improving the prognosis of patients with advanced cancer. Hydrogen application is simple, with low cost and few adverse reactions, which is an important advantage for patient-centered, family-based home rehabilitation.

This follow-up observation study is the first to show that inhaling high concentrations hydrogen can improve quality-of-life and physical fitness and control cancer progression in patients with advanced cancer. To ensure therapeutic effectiveness, the inhaled hydrogen should be delivered at sufficiently high concentrations to rapid allow entry into tissues, and with enough time to produce a dose-accumulation effect. In this report, we emphasize the importance of rehabilitation and auxiliary intervention, rather than treatment in the general sense. However, more patients of each kind of cancers are required to validate this approach. Further investigations will also clarify issues such as the optimal treatment regimen (dose, duration, mode of delivery and effectiveness of hydrogen rehabilitation combined with mainstream treatment) and the mechanism by which of hydrogen treatment controls cancer (such as removal of reactive oxygen radicals and control of inflammation). This information is important for the development of effective hydrogen therapy for patients with advanced cancer.

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Institutional review board statement: The study protocol received ethical approval from the Ethics Committee of Fuda Cancer Hospital of Jinan University on December 7, 2018 (approval number: Fuda20181207) and conformed to the specifications of the World Medical Association's Declaration of Helsinki. Written informed consent was obtained from each participant.

Informed consent statement: The authors certify that they have obtained all appropriate patient consent forms. In the form the patients or their legal guardians have given their consent for the patient's images and other clinical information to be reported in the journal. The patients or their legal guardians understand that their names and initials will not be published and due efforts will be made to conceal their identity.

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Data sharing statement: Individual participant data that underlie the results reported in this article, after deidentification (text, tables, figures, and appendices). Study protocol and informed consent form will be available immediately following publication, without end date. Results will be disseminated through presentations at scientific meetings and/or by publication in a peer-reviewed journal. Anonymized trial data will be available indefinitely at www.figshare.com.

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References

1. Siegel RL, Jemal A, Wender RC, Gansler T, Ma J, Brawley OW. An assessment of progress in cancer control. CA Cancer J Clin. 2018;68:329-339.
2. Neeman E, Ben-Eliyahu S. Surgery and stress promote cancer metastasis: new outlooks on perioperative mediating mechanisms and immune involvement. Brain Behav Immun. 2013;30 Suppl:S32-40.
3. Karagiannis GS, Pastoriza JM, Wang Y, et al. Neoadjuvant chemotherapy induces breast cancer metastasis through a TMEM-mediated mechanism. Sci Transl Med. 2017;9:eaan0026.
4. Zenitani M, Nojiri T, Hosoda H, et al. Chemotherapy can promote liver metastasis by enhancing metastatic niche formation in mice. J Surg Res. 2018;224:50-57.
5. Li T, Zeng ZC, Wang L, et al. Radiation enhances long-term metastasis potential of residual hepatocellular carcinoma in nude mice through TMPRSS4-induced epithelial-mesenchymal transition. Cancer Gene Ther. 2011;18:617-626.
6. Lapchak PA, Zhang H. Data standardization and quality management. Transl Stroke Res. 2018;9:4-8.
7. Hodson R. Precision medicine. Nature. 2016;537:S49.
8. Tannock IF, Hickman JA. Limits to personalized cancer medicine. N Engl J Med. 2016;375:1289-1294.
9. Obenauf AC, Zou Y, Ji AL, et al. Therapy-induced tumour secretomes promote resistance and tumour progression. Nature. 2015;520:368-372.
10. Sano M, Suzuki M, Homma K, et al. Promising novel therapy with hydrogen gas for emergency and critical care medicine. Acute Med Surg. 2018;5:113-118.
11. Ge L, Yang M, Yang NN, Yin XX, Song WG. Molecular hydrogen: a preventive and therapeutic medical gas for various diseases. Oncotarget. 2017;8:102653-102673.
12. Ohta S. Molecular hydrogen as a preventive and therapeutic medical gas: initiation, development and potential of hydrogen medicine. Pharmacol Ther. 2014;144:1-11.

13. Booth CM, Karim S, Mackillop WJ. Real-world data: towards achieving the achievable in cancer care. Nat Rev Clin Oncol. 2019;16:312-325.
14. Krause JH, Saver RS. Real-world evidence in the real world: beyond the FDA. Am J Law Med. 2018;44:161-179.
15. Xu H, Ping Y, Lin H, He Z, Li W, Dai H. Antiplatelet strategies and outcomes in patients with noncardioembolic ischemic stroke from a real-world study with a five-year follow-up. Transl Stroke Res. 2017;8:228-233.
16. Clarke LP, Sriram RD, Schilling LB. Imaging as a biomarker: standards for change measurements in therapy workshop summary. Acad Radiol. 2008;15:501-530.
17. Bjordal K, de Graeff A, Fayers PM, et al. A 12 country field study of the EORTC QLQ-C30 (version 3.0) and the head and neck cancer specific module (EORTC QLQ-H&N35) in head and neck patients. EORTC Quality of Life Group. Eur J Cancer 2000;36:1796-1807.
18. Girard R, Zeineddine HA, Fam MD, et al. Plasma biomarkers of inflammation reflect seizures and hemorrhagic activity of cerebral cavernous malformations. Transl Stroke Res. 2018;9:34-43.
19. Dole M, Wilson FR, Fife WP. Hyperbaric hydrogen therapy: a possible treatment for cancer. Science. 1975;190:152-154.
20. Saitoh Y, Okayasu H, Xiao L, Harata Y, Miwa N. Neutral pH hydrogen-enriched electrolyzed water achieves tumor-preferential clonal growth inhibition over normal cells and tumor invasion inhibition concurrently with intracellular oxidant repression. Oncol Res. 2008;17:247-255.
21. Saitoh Y, Yoshimura Y, Nakano K, Miwa N. Platinum nanocolloid-supplemented hydrogendissolved water inhibits growth of human tongue carcinoma cells preferentially over normal cells. Exp Oncol. 2009;31:156-162.
22. Zhao L, Zhou C, Zhang J, et al. Hydrogen protects mice from radiation induced thymic lymphoma in BALB/c mice. Int J Biol Sci. 2011;7:297-300.
23. Runtuwene J, Amitani H, Amitani M, Asakawa A, Cheng KC, Inui A. Hydrogen-water enhances 5-fluorouracil-induced inhibition of colon cancer. PeerJ. 2015;3:e859.
24. Prasad V. Do cancer drugs improve survival or quality of life? BMJ. 2017;359:j4528.
25. Prasad V, De Jesús K, Mailankody S. A further strategy to combat the high price of anticancer drugs. Nat Rev Clin Oncol. 2017;14:629.
26. Liang Y, Zhong Z, Huang Y, et al. Stem-like cancer cells are inducible by increasing genomic instability in cancer cells. J Biol Chem. 2010;285:4931-4940.
27. Amelot A, Terrier LM, Mathon B, et al. Can anticancer chemotherapy promote the progression of brain metastases? Med Oncol. 2018;35:35.
28. Martin OA, Anderson RL. Editorial: Therapy-induced metastasis. Clin Exp Metastasis. 2018;35:219-221.
29. Ohsawa I, Ishikawa M, Takahashi K, et al. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med. 2007;13:688-694.
30. Yamamoto R, Homma K, Suzuki M, Sano M, Sasaki J. Hydrogen gas distribution in organs after inhalation: Real-time monitoring of tissue hydrogen concentration in rat. Sci Rep. 2019;9:1255.
31. Kakkad S, Krishnamachary B, Jacob D, et al. Molecular and functional imaging insights into the role of hypoxia in cancer aggression. Cancer Metastasis Rev. 2019;38:51-64.
32. Al Tameemi W, Dale TP, Al-Jumaily RMK, Forsyth NR. Hypoxia-modified cancer cell metabolism. Front Cell Dev Biol. 2019;7:4.
33. Fujita K, Seike T, Yutsudo N, et al. Hydrogen in drinking water reduces dopaminergic neuronal loss in the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine mouse model of Parkinson's disease. PLoS One. 2009;4:e7247.
34. Ono H, Nishijima Y, Adachi N, et al. A basic study on molecular hydrogen (H_2) inhalation in acute cerebral ischemia patients for safety check with physiological parameters and measurement of blood H_2 level. Med Gas Res. 2012;2:21.
35. Hanahan D. Rethinking the war on cancer. Lancet. 2014;383:558-563.
36. Haines I. The war on cancer: time for a new terminology. Lancet. 2014;383:1883.
37. Iida A, Nosaka N, Yumoto T, et al. The clinical application of hydrogen as a medical treatment. Acta Med Okayama. 2016;70:331-337.

38. Akagi J, Baba H. Hydrogen gas restores exhausted CD8+ T cells in patients with advanced colorectal cancer to improve prognosis. Oncol Rep. 2019;41:301-311.
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Hydrogen-oxygen therapy can alleviate radiotherapy-induced hearing loss in patients with nasopharyngeal cancer

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Abstract

Three patients with nasopharyngeal carcinoma developed binaural secretory otitis media 12, 2, and 0.5 years after radiotherapy, respectively. The secretions subsided after conventional drug and drainage treatments, but hearing continued to deteriorate until severe loss was documented in both ears. After examination of the eardrum and tympanum, patients were enrolled in a clinical trial in the first half of 2019 (ClinicalTrials.gov: NCT03818347). After 0.5, 1 and 2 months of continuous hydrogen-oxygen therapy, our first three patients reported different levels of improvement in binaural hearing. This is the first report to show that, after treatment for nasopharyngeal carcinoma, hearing loss can be alleviated using hydrogen-oxygen therapy.

Keywords: Nasopharyngeal carcinoma; radiotherapy; secretory otitis media; hydrogen-oxygen therapy; tone audiogram

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Introduction

Nasopharyngeal carcinoma is one of the most common cancers of the head and neck in Asian populations. In southern China, the incidence of nasopharyngeal carcinoma is 30-50 per 100,000 population. Radiotherapy is the main treatment for nasopharyngeal carcinoma. However, patients often experience different degrees of adverse reactions after radiotherapy, such as dry mouth, difficulty swallowing, secretory otitis media (SOM), sinusitis, hearing loss or brain injury. Many adverse reactions are permanent and worsen progressively, which can reduce the quality of life of patients severely. Studies on whether radiotherapy-induced deafness can be alleviated are lacking. We treated three patients who suffered SOM after radiotherapy for nasopharyngeal carcinoma. Their hearing was normal before radiotherapy, and SOM appeared after radiotherapy. The secretions subsided after drug treatment, but the extent of hearing loss continued to increase, and a hearing aid was needed for 6-15 years.

After 1-2 months of treatment with a hydrogen-oxygen nebulizer, the binaural hearing of the three patients showed different levels of improvement, and one of the patients no longer needs a hearing aid.

Case presentation

Case 1

A 63-year-old woman was diagnosed with a poorly differentiated, stage-1, nasopharyngeal squamous cell carcinoma in December 2000. The radiotherapy dose was 72 Gy given in 36 fractions. This tumor was diagnosed as being in complete remission after treatment.

In March 2012, tongue-muscle atrophy, unclear speech, reduced hearing, reduced sense of smell, and SOM in both ears began to appear. After symptomatic treatments, ear fluid and pus disappeared, but hearing loss in both ears was severe, and she started to use a hearing aid.

A pre-treatment check for hydrogen-oxygen therapy (HOT) was undertaken in May 2019. Pure-tone audiometry using an audiometer (AT235; Interacoustics, Copenhagen, Denmark) was done (Figure 1A) and revealed the decibel (dB) number she could hear under a sound frequency of 500, 1,000, 2,000 and 4,000 Hz. The mean hearing threshold (in dB) of bone conduction and air conduction was 53.75 and 90, and that of the left ear was 55 and 72.5, respectively, which denoted severe hearing loss in both ears.

A tympanogram at 226 Hz using a tympanometer (AT22t; Interacoustics) showed a tympanic B-type map for binaural hearing, which suggested effusion and perforation of the tympanic membrane.

Otoscopy revealed thickening of the right tympanic membrane without a cone of light, healing of scars of bilateral tympanic membranes, and no obvious secretions in the external auditory canal (Figure 1B).

The patient underwent treatment with a hydrogen-oxygen nebulizer (AMS-H-03; Asclepius Meditec, Shanghai, China) for 4 h each day. This machine generated 3 L/min hydrogen gas by the electrolysis of water (67% hydrogen and 33% oxygen). On day-16 of treatment, she began to hear sounds in her left ear. After 1.5 months of treatment, she reported that her binaural hearing had improved considerably. The audiogram showed improvements: the mean result (in dB) for bone conduction and air conduction in the right ear was 51.25 and 80, and that of the left ear was 47.5 and 71.25, respectively (Figure 1C), but no change was observed after tympanometry and otoscopy (Figure 1D). Detection of a pure tone in both ears improved, but it continues to take longer to treat that that for a normal hearing range in humans (Figure 1E).

> *Figure 1. Pure-tone audiograms for Case 1. (A) Hearing tests before enrollment and treatment. The ordinate shows the hearing threshold at each field frequency (FF). The abscissa shows the different sound frequencies. The upper polyline represents bone conduction and the lower represents air conduction. (B) Hearing tests 1.5 months after hydrogen-oxygen treatment. (C) Comparison of the hearing threshold of our patient and normal hearing range. The red line represents the normal hearing range (0-25 dB). (D) Otoscopy results before hydrogen treatment. (E) Otoscopy results 1.5 months after hydrogen treatment.*

Case 2

A 64-year-old women was diagnosed as having a stage-II, undifferentiated, non-keratinized nasopharyngeal carcinoma in October 2010. She received radiotherapy (70 Gy in 36 fractions), and the tumor was judged to be in complete remission. Headache, insomnia, severe dry mouth and bilateral SOM appeared after radiotherapy.

After symptomatic treatments, ear fluid and pus disappeared. However, hearing loss in both ears was severe, and a hearing aid was started in February 2013. A pre-treatment check for hydrogen therapy was undertaken in May 2019.

A pure-tone audiogram is shown as Figure 2A, and reveals the dB number she could hear under a sound frequency of 500, 1,000, 2,000 and 4,000 Hz. The mean hearing threshold (in dB) of bone conduction and air conduction in the right ear was 67.5 and 101.25, and that of the left ear was 65 and 110, respectively, which denoted extremely severe hearing loss in both ears.

A tympanogram at 226 Hz showed a tympanic B-type map for binaural hearing, suggesting effusion and perforation of the tympanic membrane. Otoscopy revealed that both sides of the tympanic membrane were turbid and invaginated, and that the cone was deformed (Figure 2B).

The patient underwent treatment with a hydrogen-oxygen nebulizer for 4 h each day. After 1 month of treatment, she reported that her binaural hearing had improved considerably. The audiogram showed improvements: the mean value (in dB) for bone conduction and air conduction in the right ear was 42.5 and 78.75, and that of the left ear was 37.5 and 65, respectively (Figure 2C), but no change was observed after tympanometry and otoscopy (Figure 2D). Detection of a pure tone for both ears improved, but it continues to take longer to treat that that for a normal hearing range in humans (Figure 2E).

> *Figure 2. Pure-tone audiograms for Case 2. (A) Hearing tests before enrollment and treatment. The ordinate shows the hearing threshold at each field frequency (FF). The abscissa shows the different sound frequencies. The upper polyline represents bone conduction and the lower represents air conduction. (B) Hearing test 1 month after hydrogen-oxygen treatment. (C) Comparison of the hearing threshold of patient and normal hearing range. The red line represents the normal hearing range (0-25 dB). (D) Otoscopy results before hydrogen treatment. (E) Otoscopy results 1 month after hydrogen treatment.*

Case 3

A 63-year-old man was diagnosed as having a stage-II, poorly differentiated, nasopharyngeal squamous cell carcinoma in January 2000. The radiotherapy dose was 72 Gy given in 40 fractions. This tumor was considered to be in complete remission after treatment.

Hearing loss along with ear pus was noticed first in September 2001, and he underwent ear drainage. Hearing loss increased gradually, and hearing aids have been used since June 2016.

A pre-treatment check for HOT was undertaken in May 2019. A pure-tone audiogram is shown as Figure 3A, and reveals the dB number he could hear under a sound frequency of 500, 1,000, 2,000 and 4,000 Hz. The mean hearing threshold (in dB) of bone conduction and air conduction in the right ear was 22.5 and 62.5, and that in the left ear was 25 and 58.75, respectively, which suggested moderate hearing loss in both ears.

A tympanogram at 226 Hz showed a B-type map for left tympanum and C-type map for the right tympanum. These data suggested eustachian-tube dysfunction in the right ear, and effusion/large perforation of the tympanic membrane in the left ear. Otoscopy revealed turbidity and thickening of bilateral tympanic membranes (Figure 3B).

We underwent treatment with a hydrogen-oxygen nebulizer for 4 h each day. After 2 months of treatment, he reported that binaural hearing had improved considerably. The audiogram showed improvements: the mean bone conduction and air conduction (in dB) in the right ear was 12.5 and 41.25, and that of the left ear was 12.5 and 42.5, respectively (Figure 3C), but no change was observed after tympanometry and otoscopy (Figure 3D). The bone conduction hearing of both ears remained normal, and air conduction hearing continued to improve, so the patient stopped using a hearing aid (Figure 3E).

> *Figure 3. Pure-tone audiograms for Case 3. (A) Hearing test before enrollment and treatment. The ordinate shows the hearing threshold at each field frequency (FF). The abscissa shows the different sound frequencies. The upper polyline represents bone conduction and the lower represents air conduction. (B) Hearing test 2 months after hydrogen-oxygen treatment. (C) Comparison of the hearing threshold of patient and normal hearing range. The red line represents the normal hearing range (0-25 dB). (D) Otoscopy results before hydrogen treatment. (E) Otoscopy results 2 months after hydrogen treatment.*

Discussion

A nasopharyngeal carcinoma, and its main treatment, radiotherapy, can cause conductive hearing loss due to external- and middle-ear damage and sensorineural hearing loss due to injury to the cochlea and auditory nerves. The main parts of the middle ear to be damaged are the tympanic membrane, tympanic mucosa, and the eustachian tube, and the clinical manifestation is SOM. Otitis media caused by tumor compression can usually be alleviated completely after radiotherapy without causing sustained hearing loss. Usually, inflammation caused by radiotherapy does not appear immediately after treatment, but instead appears suddenly after many years, and causes hearing loss (and even deafness) rapidly. About half of patients with a nasopharyngeal carcinoma have SOM by 5 years following radiotherapy completion.

Our three patients developed SOM suddenly 12, 2, and 0.5 years after radiotherapy. After conventional treatment, hearing loss disappeared gradually, which suggested post-irradiation hearing loss. There is no specific treatment for inner-ear injury and hearing loss caused by radiotherapy for nasopharyngeal carcinoma. We reported, for the first time, that the hearing loss caused by radiotherapy for nasopharyngeal carcinoma can be alleviated, and that use of hearing aids can be stopped.

The three patients reported here were the first to show improvement in clinical features and pure-tone audiometry. Although the time to hearing loss after radiotherapy was very different (12, 2, and 0.5 years, respectively), the time to considerable improvement was similar (1.5, 1, and 2 months, respectively), indicating that there was no correlation between these two factors. The degree of hearing loss in the high-frequency region (4,000 Hz) that Honore and co-workers described in 2002 was correlated positively with the radiation dose given to the cochlea, the age of the patient, and hearing status before radiotherapy. The factors mentioned above were similar for our three patients. Hence, the reason why hydrogen therapy could alleviate hearing loss may be related to these factors. Interestingly, our three patients with self-reported hearing improvement had only improved pure-tone audiograms, whereas the

tympanograms and otoscopy data did not change, suggesting that this improvement was functional rather than organic. Therefore, whether such improvement can be maintained for a long-time merits further study. Case 3 was able to stop using hearing aids for normal communication, and we postulate that the pure-tone audiogram can be used as an independent prognostic factor to evaluate treatment outcomes.

In 2007, Ohsawa et al. were the first to report that hydrogen has antioxidant effects, and its biologic effects have received increasing attention since then. Studies have shown that hydrogen has: (I) selective anti-oxidation, anti-inflammatory and anti-apoptotic effects in 70 disease models; (II) good results in clinical trials; (III) achieved good results in oxidative stress, the inflammatory response and local-tissue hypoxia.

In our three patients, after HOT, the pus in the ear disappeared, and the threshold of air conduction and bone conduction decreased considerably, which may have been related to the anti-oxidation and anti-inflammatory effects of the hydrogen-oxygen mixture. Studies have suggested that the levels of hypoxia-inducible transcription factors increase during hypoxia, then increase the expression of multiple signaling pathways and molecules, and neutrophil activation increases levels of superoxide anions and reactive oxygen species. Hydrogen can relieve chronic inflammation by antagonizing these mechanisms. Studies have demonstrated that hyperbaric oxygen can improve the therapeutic effect of sudden deafness and promote hearing recovery. Therefore, treatment with a mixture of hydrogen and oxygen can improve local hypoxia, reduce inflammation, reduce oxidative stress, and aid cell repair.

The hearing threshold of a patient during pure-tone audiometry reflects the condition of the cochlea. The hearing threshold of our patients was reduced considerably. Improvement in hearing upon HOT could be related to improvement of injury to the cochlea. One advantage of HOT at sites in the head and neck compared with those at other sites is the short distance needed for its diffusion, which leads to high concentrations during HOT.

Conclusions

The radiotherapy-induced hearing loss in the three patients in our study was relieved after HOT, but the mechanism of action is not clear. Further research is needed to provide safe, economical and efficacious treatment for patients with hearing loss after radiotherapy for nasopharyngeal carcinoma.

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References

1. Li C, Wang Y. Factors associated with early diagnosis in pediatric vs adult nasopharyngeal carcinoma. Acta Otolaryngol 2018;138:56-9.
2. Sun D, Zhao M, Yin J, et al. Vestibular function disorders and potential mechanisms in irradiation nasopharyngeal carcinoma patients. Acta Otolaryngol 2016;136:759-63.
3. Tian YM, Huang WZ, Yuan X, et al. The challenge in treating locally recurrent T3-4 nasopharyngeal carcinoma: the survival benefit and severe late toxicities of re-irradiation with intensity-modulated radiotherapy. Oncotarget 2017;8:43450-7.

4. Kong F, Zhou J, Du C, et al. Long-term survival and late complications of intensity-modulated radiotherapy for recurrent nasopharyngeal carcinoma. BMC Cancer 2018;18:1139.
5. Hsin CH, Tseng HC, Lin HP, et al. Post-irradiation otitis media, rhinosinusitis, and their interrelationship in nasopharyngeal carcinoma patients treated by IMRT. Eur Arch Otorhinolaryngol 2016;273:471-7.
6. Honoré HB, Bentzen SM, Møller K, et al. Sensori-neural hearing loss after radiotherapy for nasopharyngeal carcinoma: individualized risk estimation. Radiother Oncol 2002;65:9-16.
7. Ohsawa I, Ishikawa M, Takahashi K, et al. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med 2007;13:688-94.
8. Ohta S. Molecular hydrogen as a novel antioxidant: overview of the advantages of hydrogen for medical applications. Methods Enzymol 2015;555:289-317.
9. Nishiguchi M, Ono S, Iseda K, et al. Effect of vasodilation by milrinone, a phosphodiesterase III inhibitor, on vasospastic arteries after a subarachnoid hemorrhage in vitro and in vivo: effectiveness of cisternal injection of milrinone. Neurosurgery 2010;66:158-64; discussion 64.
10. Thompson AA, Binham J, Plant T, et al. Hypoxia, the HIF pathway and neutrophilic inflammatory responses. Biol Chem 2013;394:471-7.
11. Wang YZ, Li TT, Cao HL, et al. Recent advances in the neuroprotective effects of medical gases. Med Gas Res 2019;9:80-7.
12. Gaitanou K, Fildissis G, Vavasis P, et al. Management of sudden hearing loss with hyperbaric oxygen therapy. Undersea Hyperb Med 2014;41:363-70.

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Brain Metastases Completely Disappear in Non-Small Cell Lung Cancer Using Hydrogen Gas Inhalation: A Case Report

Case Report — OncoTargets and Therapy (Dovepress). OncoTargets and Therapy 2019:12 11145-11151.

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Abstract

Lung cancer is the most common type of tumor, prone to contralateral lung, bone and brain metastasis. We report a 44-year-old woman diagnosed with lung cancer with multiple brain metastases in November 2015. Oral targeted drugs were initiated after the removal of brain metastases, and most lesions remained stable for 28 months. In March 2018, intracranial multiple metastases, as well as hydrocephalus accumulation in the third ventricle and lateral ventricles, and metastases in bone, adrenal gland, liver were noted. Hydrogen-gas monotherapy was started to control the tumor a month later. After 4 months, the size of multiple brain tumors was reduced significantly, and the amount of hydrocephalus in the third ventricle and lateral ventricles reduced significantly. After 1 year, all brain tumors had disappeared, and there were no significant changes in metastases in the liver and lung. These data show that, after standard treatments had failed, hydrogen-gas monotherapy elicited significant effective control of tumors (especially those in the brain), and survival time was lengthened.

Keywords: hydrogen gas, lung cancer, brain metastasis, extrapulmonary metastasis

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Introduction

Non-small-cell lung cancer (NSCLC) is the second most frequently diagnosed tumor worldwide, and is the leading cause of cancer-related deaths. With respect to metastasis of NSCLC, there are some preferential sites (bone, 34.3%; lung, 32.1%; brain, 28.4%; adrenal glands, 16.7%; liver, 13.4%). NSCLC metastasizes to the brain in 10-25% of all cases. This type of spread can lead to significant morbidity and mortality, and 5-year survival has been reported to be 3.6%. No significant changes in 5-year survival for patients with NSCLC metastases to the brain have been observed in the last three decades. The short-term survival of NSCLC patients with brain metastasis is due to the lack of efficacious methods. Standard chemotherapy is ineffective because of the blood-brain barrier. Radiotherapy and neurosurgery are recognized as locoregional methods; molecularly targeted therapies and immunotherapies are recognized as systemic treatments.

In 2018, Wang et al reported on delivery of different concentrations of hydrogen gas into two culture systems for lung cancer cells (A549 and H1975). They found that the ability of cells to divide, migrate and infiltrate into tissue was inhibited significantly, and that apoptosis was accelerated. Also, lung cancer tumor-bearing mice were treated with hydrogen gas for 4 weeks, and the tumor volume was reduced by 25%. The reason for these phenomena was that hydrogen can reduce the expression of structural maintenance of chromosomes 3 (SMC3), SMC5, SMC6 in lung cancer cells.

Also in 2018, Akagi and Baba reported the results of hydrogen gas monotherapy for patients with stage-IV colon cancer. They found that the proportion of programmed death (PD)-1^{+}CD8^{+} T-cell subsets in the blood of patients was reduced upon continuous inhalation of hydrogen, and was reduced by ~60% after several months. After 3 years of follow-up, progression-free survival and overall survival were prolonged significantly.

Here, we report a 44-year-old woman diagnosed with NSCLC and multiple metastases. After resection of an intracranial metastasis and targeted drug therapy, the outcome was good, but NSCLC metastasized (brain, cerebellum, lung, liver, bones and adrenal glands) after 28 months of treatment. In view of the literature stated above, she underwent hydrogen inhalation, and brain metastases disappeared after 4 months. Intracranial recurrence was not found after 1 year of treatment, and other metastatic sites remained stable.

Case Presentation

In November 2015, a 44-year-old woman was admitted to our hospital complaining of dizziness and instability upon standing. Through chest computed tomography (CT) and cranial magnetic resonance imaging (MRI) examination, we found multiple tumors in the left cranial cavity and right lung. After resection of an intracranial tumor, pathology studies suggested metastatic lung adenocarcinoma. Genetic testing suggested mutation of the epidermal growth factor receptor (EGFR) gene at exon 19. She started taking gefitinib tablets (Iressa™ 250 mg, once daily). The intracranial and lung tumors were controlled well until June 2017 (Figure 1).

When a review was conducted in September 2017, the disease had progressed. A combination of pemetrexed and erlotinib was initiated, and the tumors were controlled again. The intracranial and lung tumors progressed again in March 2018, and the targeted treatment regimen was adjusted to icotinib hydrochloride (125 mg, t.d.s.). After 2 weeks, the patient suddenly complained of dyspnea, and difficulties in speaking. Enhanced MR imaging of the head revealed multiple nodular metastases in the left cranial cavity; the third ventricle and lateral ventricles accumulated hydrocephalus, and the largest lesion was in the frontal lobe (1.9 × 1.4 cm) (Figure 2A). Computed tomography showed that the lesion on the upper lobe of the right lung was enlarged (1.2 × 0.6 cm), and new metastases were found at the lower lobe of the right lung, mediastinum, lung hilum (1.7 × 1 cm), seventh thoracic spine and left adrenal gland. The patient refused radiotherapy and surgery of brain, signed informed consent of hydrogen therapy and agreed on the publication of the case.

From April 2018, the patient began hydrogen inhalation and received no other treatments during the time period. The hydrogen oxygen nebulizer (AMS-H-03; Asclepius Meditec, Shanghai, China) generates 3 L/min hydrogen gas by hydrocephalus electrolysis. As measured by gas chromatography, the gas generated consisted of 67% hydrogen and 33% oxygen. Using a special mask, the patient continued to inhale hydrogen for 3-6 hrs a day at rest, with no interruption even after the obvious relief of symptoms. After 4 months, most brain metastases disappeared and the amount of hydrocephalus in the third ventricle and lateral ventricles were both reduced (Figure 2B). After 1 year, all visible brain tumors had disappeared, and there were no obvious changes in metastases in the liver and lung (Figure 2C).

There were increased levels of carcinoembryonic antigen (CEA, 29.44 ng/mL), carbohydrate antigen-125 (CA125, 153 U/mL) and cytokeratin fragment antigen21-1 (CYFRA21-1, 12.1 ng/mL) before the patient started hydrogen monotherapy. Serum levels of these three markers continued to decrease after 4 months of treatment (21.6 ng/mL, 83 U/mL and 8.5 ng/mL, respectively), and were close to (but remained above) the reference range after 1 year of treatment (12.3 ng/mL, 61 U/mL and 5.9 ng/mL, respectively) (Figure 3).

> *Figure 1. Post-contrast brain MR imaging of the head before and after treatment. In November 2015, the patient was found to have lung cancer with brain (left, 4.4×3.1 cm) and cerebellar (right, 4.0×3.1 cm) metastasis. After resection and targeted drug therapy, there was no recurrence of metastases after 19 months. Solid arrow = brain metastasis; hollow arrow = cerebellar metastasis.*

Discussion

In general, NSCLC cells tend to metastasize to the brain, bones and adrenal glands, adenocarcinomas metastasize to the brain, and squamous cell carcinomas metastasize to bones. The exact homing mechanisms and how cancer cells communicate with this new stroma are not known.

Brain metastasis of NSCLC cells is closely related to various signaling pathways, several of which involve the participation of hydrogen gas. In 2013, Bleckmann et al reported that, within the Wnt pathway, T-cell factor (TCF)/lymphoid enhancer factor (LEF-1) act independently of β-catenin in human lung adenocarcinomas that metastasize to the cerebrum. In 2014, Kafka et al reported altered expression of dishevelled (DVL)-1, DVL-3, E-cadherin, and β-catenin in brain metastases of adenocarcinomas, which again suggested the importance of Wnt

signaling. Lin et al were the first (in 2016) to demonstrate that hydrogen gas suppresses abnormal activation of Wnt/β-catenin signaling.

Crosstalk between EGFR and MET, the hepatocyte growth factor (HGF) receptor, has been reported in adenocarcinomas that have metastasized to the brain. This is not a direct interaction but instead signaling via activation of mitogen-activated protein kinases (MAPKs). In 2017, Yang et al found that hydrogen-containing saline can decrease phosphorylation of p38 MAPK and Smad2/3 in a rat model of cardiac fibrosis. In 2019, Guan et al reported that hydrogen gas can ameliorate chronic intermittent hypoxia-induced kidney injury by inhibiting oxidative stress-dependent activation of p38 and c-Jun N-terminal kinase (JNK). Similar results were demonstrated by Zhang and colleagues in experiments on the proliferation and migration of vascular smooth muscle cells. Han et al showed that sirtuin (SIRT)1 is highly expressed in brain metastasis, and several scholars have demonstrated that hydrogen can suppress SIRT1 signaling in different models. Those studies suggest that hydrogen gas has a unique therapeutic effect on the brain metastasis of NSCLC.

Several interesting studies have focused on the comparison of genomic alterations between primary lung carcinomas versus metastases to the brain and bone. One hypothesis is that there might be clonal diversity between these two. In our patient, there was a considerable difference between brain metastasis and other extrapulmonary metastases. This was most likely due to the clonal diversity of different metastatic sites, which resulted in significant differences in sensitivity to hydrogen inhalation.

> *Figure 2. Post-contrast brain MRI of the head before and after hydrogen inhalation. (A) In April 2018, the patient was found to have multiple metastases (white arrows) in the brain (left, 2.5×3.0 cm and 2.4×2.8 cm for two bigger tumors) and cerebellum (right, 1.3×1.8 cm and 1.2×0.7 cm for two bigger tumors). Also, the third ventricle and lateral ventricles were expanded and had accumulated hydrocephalus. (B) After 4 months of hydrogen inhalation, all tumors visible to the naked eye had disappeared, and the shape of ventricles had returned to normal. (C) After 1 year of inhalation (April 2019), recurrence had not occurred.*

As with many biomarker studies focusing on single genes/proteins, a selection bias or overinterpretation can occur. Shen et al found higher levels of metastasis-associated lung adenocarcinoma transcript-1 (MALAT1) in brain metastases compared with those in other extrapulmonary sites. A similar investigation searched for brain-metastasis genes and discovered a regulator of epithelial-mesenchymal transition (EMT); pre-B cell leukemia homeobox (PBX)-regulating protein-1 (PREP1) overexpression triggered EMT, whereas PREP1 downregulation inhibited EMT induction in response to transforming growth factor-β expression. PREP1 modulates sensitivity to SMAD3 and induces expression of Fos-related antigen (FRA)-1. FRA-1 and PBX1 are required for the mesenchymal changes triggered by PREP1 in lung tumor cells. PREP1-induced EMT correlates with increased lung colonization, and PREP1 accumulation has been found in human brain metastases. When investigating adenocarcinomas with rearrangement of activin receptor-like kinase, amplification of the fibroblast growth factor receptor (FGFR1) gene correlated significantly with brain metastases. Although in those cases there were more visceral metastases, FGFR1 amplification in the brain metastases of adenocarcinomas were fivefold more frequent than that in the primary tumors.

Also, C-X-C motif chemokine receptor (CXCR4) seems to have a role in brain metastasis. CXCR4 protein has been shown to be highly overexpressed in patients with brain-specific metastasis but significantly less in NSCLC patients with metastases to other organs and in

patients without metastases. Also, a disintegrin and metallopeptidase domain 9 (ADAM9) expression have been reported to be relatively higher in brain metastases than that in primary lung tumors. ADAM9 regulates metastasis of lung cancer cells to the brain by facilitating tissue plasminogen activator-mediated cleavage of CUB domain-containing protein-1.

We are the first to report the efficacy of hydrogen on brain metastases from NSCLC. Our patient has survived for >1 year with hydrogen inhalation, and indicators we explored the possible reason for hydrogen to inhibited NSCLC brain metastasis in this patient (e.g. TCF/LEF-1 and DVL-1/3 of Wnt/β-catenin signaling, MAPK, and SIRT1), and proposed differences in the expression of signaling molecules between brain metastasis and other extrapulmonary metastases (e.g. MALAT1, EMT, PREP1, FGFR1, CXCR4 and ADAM9).

Hydrogen inhalation may have had a role in the treatment of our patient in different ways. For example, hydrogen exerts neuroprotective effects by reducing cyclooxygenase-2 activity or activating expression of anti-apoptotic protein kinase B. Also, hydrogen can inhibit expression of pro-apoptotic factors such as JNK and caspase-3. Hydrogen inhalation can down-regulate the expression of various pro-inflammatory cytokines, including interleukin (IL)-1β, IL-6, tumor necrosis factor-α, intracellular adhesion molecule-1, high mobility group box-1, nuclear factor-kappa B, and prostaglandin-E2. Whether the mechanisms mentioned above are related to the therapeutic effect of hydrogen must be confirmed by further research.

> *Figure 3. Detection of serum tumor markers before and after hydrogen inhalation. The red line and the lower area in each figure represent the normal range. Abbreviations: CEA, carcinoembryonic antigen; CA125, carbohydrate antigen 125; CYFRA21-1, cytokeratin fragment antigen 21-1.*

Ethics Approval and Consent for Publication

This research was approved by the research ethics committee of Fuda Cancer Hospital of Jinan University, and written informed consent for publication of the clinical details and images was obtained from the patient.

Author Contributions

All authors contributed to data analysis, drafting and revising the article, gave final approval of the version to be published, and agree to be accountable for all aspects of the work.

Disclosure

The authors report no conflicts of interest regarding this work.

References

1. Siegel RL, Miller KD, Jemal A. Cancer statistics, 2019. CA Cancer J Clin. 2019;69(1):7-34.
2. Tamura T, Kurishima K, Nakazawa K, et al. Specific organ metastases and survival in metastatic non-small-cell lung cancer. Mol Clin Oncol. 2015;3(1):217-221.
3. Ulahannan D, Khalifa J, Faivre-Finn C, et al. Emerging treatment paradigms for brain metastasis in non-small-cell lung cancer: an overview of the current landscape and challenges ahead. Ann Oncol. 2017;28(12):2923-2931.
4. Goldstraw P, Chansky K, Crowley J, et al. The IASLC lung cancer staging project: proposals for revision of the TNM stage groupings in the forthcoming (Eighth) edition of the TNM classification for lung cancer. J Thorac Oncol. 2016;11(1):39-51.
5. Sperduto PW, Yang TJ, Beal K, et al. Estimating survival in patients with lung cancer and brain metastases: an update of the graded prognostic assessment for lung cancer using molecular markers (lung-molGPA). JAMA Oncol. 2017;3(6):827-831.

6. Metro G, Chiari R, Ricciuti B, et al. Pharmacotherapeutic options for treating brain metastases in non-small cell lung cancer. Expert Opin Pharmacother. 2015;16(17):2601-2613.
7. Wang D, Wang L, Zhang Y, et al. Hydrogen gas inhibits lung cancer progression through targeting SMC3. Biomed Pharmacother. 2018;104:788-797.
8. Akagi J, Baba H. Hydrogen gas restores exhausted CD8+ T cells in patients with advanced colorectal cancer to improve prognosis. Oncol Rep. 2019;41(1):301-311.
9. Bleckmann A, Siam L, Klemm F, et al. Nuclear LEF1/TCF4 correlate with poor prognosis but not with nuclear beta-catenin in cerebral metastasis of lung adenocarcinomas. Clin Exp Metastasis. 2013;30(4):471-482.
10. Kafka A, Tomas D, Beros V, et al. Brain metastases from lung cancer show increased expression of DVL1, DVL3 and beta-catenin and down-regulation of E-cadherin. Int J Mol Sci. 2014;15(6):10635-10651.
11. Lin Y, Ohkawara B, Ito M, et al. Molecular hydrogen suppresses activated Wnt/beta-catenin signaling. Sci Rep. 2016;6:31986.
12. Breindel JL, Haskins JW, Cowell EP, et al. EGF receptor activates MET through MAPK to enhance non-small cell lung carcinoma invasion and brain metastasis. Cancer Res. 2013;73(16):5053-5065.
13. Yang J, Wu S, Zhu L, et al. Hydrogen-containing saline alleviates pressure overload-induced interstitial fibrosis and cardiac dysfunction in rats. Mol Med Rep. 2017;16(2):1771-1778.
14. Guan P, Sun ZM, Luo LF, et al. Hydrogen protects against chronic intermittent hypoxia induced renal dysfunction by promoting autophagy and alleviating apoptosis. Life Sci. 2019;225:46-54.
15. Zhang YX, Xu JT, You XC, et al. Inhibitory effects of hydrogen on proliferation and migration of vascular smooth muscle cells via down-regulation of mitogen/activated protein kinase and ezrin-radixin-moesin signaling pathways. Chin J Physiol. 2016;59(1):46-55.
16. Han L, Liang XH, Chen LX, et al. SIRT1 is highly expressed in brain metastasis issues of non-small cell lung cancer (NSCLC) and in positive regulation of NSCLC cell migration. Int J Clin Exp Pathol. 2013;6(11):2357-2365.
17. Xing Z, Pan W, Zhang J, et al. Hydrogen rich hydrocephalus attenuates renal injury and fibrosis by regulation transforming growth factor-beta induced Sirt1. Biol Pharm Bull. 2017;40(5):610-615.
18. Sun Q, Han W, Hu H, et al. Hydrogen alleviates hyperoxic acute lung injury related endoplasmic reticulum stress in rats through upregulation of SIRT1. Free Radic Res. 2017;51(6):622-632.
19. Li S, Fujino M, Ichimaru N, et al. Molecular hydrogen protects against ischemia-reperfusion injury in a mouse fatty liver model via regulating HO-1 and Sirt1 expression. Sci Rep. 2018;8(1):14019.
20. Shen L, Chen L, Wang Y, et al. Long noncoding RNA MALAT1 promotes brain metastasis by inducing epithelial-mesenchymal transition in lung cancer. J Neurooncol. 2015;121(1):101-108.
21. Li Q, Yang J, Yu Q, et al. Associations between single-nucleotide polymorphisms in the PI3K-PTEN-AKT-mTOR pathway and increased risk of brain metastasis in patients with non-small cell lung cancer. Clin Cancer Res. 2013;19(22):6252-6260.
22. Preusser M, Berghoff AS, Berger W, et al. High rate of FGFR1 amplifications in brain metastases of squamous and non-squamous lung cancer. Lung Cancer. 2014;83(1):83-89.
23. Wang L, Wang Z, Liu X, et al. High-level C-X-C chemokine receptor type 4 expression correlates with brain-specific metastasis following complete resection of non-small cell lung cancer. Oncol Lett. 2014;7(6):1871-1876.
24. Lin CY, Chen HJ, Huang CC, et al. ADAM9 promotes lung cancer metastases to brain by a plasminogen activator-based pathway. Cancer Res. 2014;74(18):5229-5243.
25. Yang Y, Zhu Y, Xi X. Anti-inflammatory and antitumor action of hydrogen via reactive oxygen species. Oncol Lett. 2018;16(3):2771-2776.
26. Hou Z, Luo W, Sun X, et al. Hydrogen-rich saline protects against oxidative damage and cognitive deficits after mild traumatic brain injury. Brain Res Bull. 2012;88(6):560-565.

27. Sun H, Chen L, Zhou W, et al. The protective role of hydrogen-rich saline in experimental liver injury in mice. J Hepatol. 2011;54(3):471-480.
28. Zhang JY, Liu C, Zhou L, et al. A review of hydrogen as a new medical therapy. Hepato-Gastroenterology. 2012;59(116):1026-1032.

A Gallbladder Carcinoma Patient With Pseudo-Progressive Remission After Hydrogen Inhalation

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Abstract

Background: Hydrogen therapy has been reported to convert exhausted programmed death (PD-1)+CD8+ T cells to PD-1-CD8+ T cells, in advanced colorectal cancer patients, which is associated with significantly prolonged survival.

Case presentation: A 72-year-old female patient presented with metastatic gallbladder cancer and underwent symptomatic treatment combined with hydrogen therapy. The tumors were initially enlarged and displayed increased tumor marker expression following hydrogen inhalation therapy, after which they continued to remit, similar to the pseudo-progression that occurs after anti-PD-1 treatment. During one month of hydrogen therapy, the patient's gallbladder and liver tumors continued to progress, and intestinal obstruction occurred. The intestinal obstruction was gradually relieved after symptomatic treatment, and the anemia were corrected, and both the lymphocyte and tumor marker levels returned to normal. The patient was able to resume normal life two and a half months after hydrogen inhalation and survived over 10 months.

Conclusion: This is the first report of pseudo-progression followed by sustained remission after hydrogen inhalation. This phenomenon is similar to the pseudo-progression-remission pattern that occurs following PD-1 antibody treatment. These findings suggest that hydrogen may have an inhibitory effect on PD-1 expression.

Keywords: hydrogen gas, metastatic gallbladder cancer, pseudo-progression, programmed cell death receptor-1

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Introduction

Hydrogen gas is a type of "physiological gas" that is continuously produced by human intestinal bacteria, and has been confirmed to have selective antioxidant and anti-

inflammatory effects. Several studies have shown that hydrogen can inhibit the formation, migration, and invasion of cancer cells through its anti-oxidative effects. Due to technical challenges, the ability of hydrogen to directly kill cancer cells in vivo or lead to tumor shrinkage has been difficult to confirm. In 2019, Akagi et al first reported a long-term follow-up study involving 55 participants and found that continued hydrogen inhalation reversed the exhausted state of PD-1+CD8+ T cells to a CD8+PD-1- phenotype, and could extend the survival time from 18 to 46 months. To study the effect of hydrogen therapy on ovarian cancer, we report a case for whom the disease first worsened and then continued to improve along with changes in CD8+ T cell immune functionality.

Case Presentation

A 72-year-old female patient developed a gallbladder tumor in December 2017 due to pain in the right upper quadrant. The biopsy showed poorly differentiated adenocarcinoma. She was diagnosed as stage IIIA gallbladder carcinoma (T3N1M0) because of lymph node metastases at hepatic hilar and pancreatic head by positron emission tomography (PET) scan. Since the patient had been previously diagnosed with rheumatic heart disease and diabetes, irreversible electroporation ablation was performed. The patient's abdominal pain was relieved after ablation and she then received oral tegafur (a fluoropyrimidine derivative) chemotherapy. In September 2018, the patient developed severe pain in her upper right abdomen, with vomiting associated with the inability to eat, and a subsequent rapid reduction in body weight. After being admitted to our hospital, the patient was diagnosed with heart failure (class IV heart function), severe anemia, and hypoproteinemia. A computed tomography (CT) examination revealed gallbladder tumors (6.3 × 4.9 cm), multiple spotted high-density lesions in the liver parenchyma, tumor invasion of the duodenum, accompanied by gallbladder-duodenal descending fistula, multiple enlarged lymph nodes around the pancreatic head (maximum 2.7 × 2.1 cm), and compression of the inferior vena cava. Moreover, the gallbladder cancer had progressed to stage IV (T4N1aM0). On the second day after admission, the patient entered the Intensive Care Unit and received both symptomatic and systemic supportive treatments, including parenteral nutrition, an infusion of albumin, red blood cells, insulin, cardiotonic diuretics, and antibiotics, as well as gastrointestinal catheter drainage.

This patient was enrolled in a hydrogen therapy clinical trial (NCT03818347) on October 24, 2018. The inhalation of hydrogen gas via a hydrogen oxygen atomizer (AMS-H-03; Asclepius Meditec, Shanghai, China) was initiated simultaneously with symptomatic treatments. Initially, the patient underwent hydrogen inhalation for 2 h/day (3 L/min), and was gradually increased to 6 h/day. In addition to continuing to administer the above symptomatic and systemic supportive treatments, no routine anti-cancer therapy was provided.

Post-treatment evaluations included: 1) adverse reactions: referred to in the Common Terminology Standard Version 5.0 (CTCAE 5.0). Adverse events were classified and scored weekly after treatment initiation. Laboratory indicators included peripheral blood cell subsets, serum alanine aminotransferase (ALT), aspartate aminotransferase (AST), total bilirubin (TB), and gamma-glutamyl transpeptidase (GGT), and expression of tumor markers; 2) clinical symptoms and PS scores; 3) imaging changes: the tumor response was measured using computed tomography (CT) according to the RECIST 1.1 guidelines, and the tumor response was classified as complete remission (CR), partial remission (PR), stable disease (SD), or progressive disease (PD).

Following hydrogen inhalation, the patient's daily agitation gradually decreased and disappeared completely after two weeks. In addition, the patient's sleep time was extended

from 4-5 h/day to 8-10 h/day within two weeks. No adverse reactions related to hydrogen inhalation were observed during the first three months. The patient's upper abdominal pain persisted, requiring the daily use of analgesics prior to hydrogen treatment; however, after two weeks of hydrogen inhalation, the pain gradually decreased and painkillers were no longer required. The PS score gradually decreased from 4 points prior to hydrogen treatment to 1 point after two and a half months. By the end of April 2019, the patient had a PS score of 0 and had resumed normal diet and activity. The disease has been continuously relieved for 10 months and has exhibited increased improvement.

During the entire period of hydrogen treatment, serum levels of ALT, AST, TB, and GGT exhibited no abnormalities; the red blood cell count and hemoglobin levels stopped decreasing; and the interval between blood transfusions was extended from one week before hydrogen inhalation to two weeks. Moreover, these two indexes began to increase continuously one month later (Figure 1A), and the blood transfusion was completely stopped after two months. The level of serum total protein and albumin began to rise after supportive treatments and continued to increase after hydrogen inhalation, reaching a normal range after two and a half months, at which time, supplements were no longer required (Figure 1B). In the two weeks before hydrogen treatment, the peripheral blood leukocyte count was significantly increased due to the intestinal fistula. The leukocyte count decreased to the upper limit of the reference range after antibiotic treatment and continued to decrease after hydrogen inhalation, falling to normal levels after 1.5 months (Figure 1C left). The total number of lymphocytes was low at the time of admission, and the number began to rebound after one and a half months of hydrogen inhalation, returning to normal after two and a half months (Figure 1C right).

> *Figure 1. Blood parameters before and after hydrogen treatment. (A) Red blood cell counts and hemoglobin levels; the red line in the figures represent the reference range. (B) Total protein and albumin concentrations; the red lines in the figures represent the reference range. (C) White blood cell and lymphocyte counts; the red lines in the figures represent the lower limit of the reference range.*

Serum CA19-9, AFP, and CEA were all elevated at the time of admission (2, 1.8, and 8 times the upper limit of the reference values, respectively). Following hydrogen inhalation, the three markers did not decline and the AFP even exceeded the level at the time of admission; however, these levels rapidly declined afterwards, and all fell to within the normal reference range after two and a half months (Figure 2).

Abdominal CT images were analyzed at different time points before and after hydrogen treatment (Figure 3A). Before hydrogen treatment, the sum of the diameter of multiple tumors was 12.2 cm. After hydrogen treatment for: 1) one month, the sum was 16.7 cm (37% increase compared with pretreatment, PD); 2) two and a half months, the sum was 12.9 cm (6% increase compared with pretreatment, SD); and 3) six months, the total was 8.1 cm (33% decrease compared with pretreatment, PR) (Figure 3B).

The patient was tested for immunological function before hydrogen treatment, as well as at six and nine months after treatment. In the terminally differentiated CD8+ T cells, the proportion of exhausted cells was 46.8% at two weeks before treatment, 31.3% at six months after treatment, and 27.6% after nine months (Figure 4).

> *Figure 2. Changes in the tumor markers before and after hydrogen treatment. The red lines in the figures represent the upper limit of the reference ranges.*

Discussion

The treatment for gallbladder cancer is difficult, primarily due to the development of early intrahepatic and hepatic portal metastases. There is no evidence that minimally invasive ablation and chemotherapy can be used to obtain survival gains. In this patient, intrahepatic and hepatic portal metastases were present at the time of diagnosis. Although irreversible electroporation ablation and chemotherapy were performed, the lesion progressed further after nine months, invading the duodenum and appearing around the head of the pancreas. Multiple lymph node metastases involving the inferior vena cava was indicative of metastatic cancer (T4N1aM0). In addition to the patient's original rheumatic heart disease with mitral valve replacement, the patient was 72 years old with diabetes (fasting blood glucose up to 22 mmol/L) and severe anemia (hemoglobin only 37 g/L), making it impossible for her to receive routine anti-tumor treatments.

The patient's disease status changed following hydrogen inhalation. One month after hydrogen therapy, the patient's general condition and blood parameters (including total protein, albumin, red blood cell count, and hemoglobin levels) gradually improved. After two and a half months of therapy, her duodenal obstruction was relieved and subsequently disappeared, the gastric tube was removed, the level of tumor marker expression began to decline, the CT examination showed that the tumors in the gallbladder and liver had gradually reduced, and the PS score was significantly improved. The patient has undergone hydrogen treatment for 10 months to date, and her condition has further improved. However, in this case, although the symptoms and systemic conditions improved within one month of hydrogen inhalation, the tumor size and tumor marker expression increased and subsequently decreased following continued treatment. Clearly, the initial changes observed regarding the tumors and associated markers are false indicators of progression. Moreover, these findings are highly similar to the pseudo-remission-response patterns that occur during immunotherapy with PD-1 inhibitors.

Pseudo-progression refers to an increase in the original lesions at the beginning of treatment or the emergence of new lesions, which is reduced following continued treatment. This response appears to be "exclusive" to anti-PD-1/PD-L1 immunotherapy, first observed in melanoma patients, and has subsequently been reported in other cancers. The mechanism for this phenomenon may be that during immunotherapy, immunocytes (i.e., cytotoxic T cells) infiltrate the tumor, causing edema and necrosis. PD-1 is a typical marker of senescent apoptotic CD8+ T cells. Recent studies have shown that hydrogen can inhibit the expression of PD-1 on T cells in vivo.

> *Figure 3. (A) CT imaging findings at different time-points before and after hydrogen treatment. In the first line of figures, the triangle and the contour lines represent the location and range of the gallbladder. In the second line of figures, the contour lines represent the location and range of the liver metastases. In the third line of figures, the arrows represent the location of the duodenal fistula or obstruction. (B) Evaluation of the effect of hydrogen treatment at different time-points according to CT imaging and RECIST 1.1. Abbreviations: PD, progressed disease; SD, stable disease; PR, partial remission; CT, computed tomography.* (Columns: 06/10/2018 half month before; 24/10/2018 d0 of treatment; 27/11/2018 1 month after; 12/01/2019 2.5 month after; 15/04/2019 6 month after. Rows: Gallbladder tumor; Liver metastases; Duodenal fistula and obstruction.)

> *Table (Figure 3B). Tumor sizes and effect evaluation*

From D0 of hydrogen inhalation	Gallbladder tumor	Liver metastases	Pancreas head/lymph node	Sum of the tumor main diameters	Effect evaluation
D0 of hydrogen inhalation	6.3 × 4.9 cm	—	2.7 × 2.1 cm	12.2 cm	—
1 month after	6.7 × 6.3 cm	3.2 × 2.6 cm	2.2 × 1.8 cm	16.7 cm	PD
2.5 month after	3.7 × 3.6 cm	4.2 × 4.0 cm (large), 3.6 × 3.2 cm (small)	2.0 × 1.8 cm	12.9 cm	SD
6 month after	2.8 × 2.6 cm	4.0 × 3.3 cm (large), 3.2 × 2.6 cm (small), 2.5 × 2.4 cm (large)	2.8 × 2.4 cm	8.1 cm	PR

Figure 4. In terminally differentiated CD8+ T cells, the proportion of exhausted cells varied with the treatment time. The results were tested by flow cytometry, in which the terminally differentiated CD8+ T cells were labeled CD3+CD8+CD27-, in which PD-1+ was considered a marker of exhaustion and non-exhausted cells were PD-1-.

Moreover, Akagi et al reported that after the hydrogen treatment of 55 patients with stage IV colorectal cancer, PD-1+CD8+ T cells decreased and PD-1-CD8+ T cells increased in the peripheral blood. As the hydrogen inhalation time increases, the ratio of the two cells is further reduced. Additionally, the greater the reduction in PD-1+CD8+ T cells, the longer the progression-free survival and overall survival of the patients. Indeed, this study showed that hydrogen therapy restored exhausted CD8+ T cell activity by inhibiting PD-1 expression. We observed a similar phenomenon in this case, which supports the evidence that hydrogen therapy may exhibit an effect similar to the pseudo-progression phenomenon observed following anti-PD-1 treatment.

It has been found that an increased number of CD8+ T cells expressing PD-1 in the peripheral blood of cancer patients represents a serious prognostic marker. In addition, PD-1+CD8+ T cells are functionally exhausted and have lost their anti-tumor functionality. Further studies have found that mitochondrial dysfunction results in an insufficient energy supply to such T cells, which leads to functional impairment. Hydrogen can stimulate peroxisome proliferator-activated receptor gamma cofactor 1 alpha (PGC1α), which positively regulates several metabolic processes, including mitochondrial biosynthesis, respiration, adaptive caloric production, and gluconeogenesis, thereby enhancing and improving mitochondrial activity and exerting immunomodulatory effects.

Conclusion

Here, we reported a case of progressive gallbladder cancer that received only hydrogen inhalation therapy in addition to symptomatic treatment. Pseudo-progression and subsequent remission after hydrogen inhalation exhibited a similar response pattern following anti-PD-1 immunotherapy. We found a significant decrease in the proportion of exhausted CD8+PD-1+ T cells by immunological function tests before and after hydrogen inhalation, which suggests that hydrogen can revitalize senescent apoptotic CD8+ T cells through mitochondrial rescue. However, the process of tumor killing and shrinking may induce pseudo-progression. The potential clinical use of this therapy still needs validation with additional large-scale trials.

Ethics Approval And Consent For Publication

This research was approved by the research ethics committee of Fuda Cancer Hospital of Jinan University, and written informed consent for publication of the clinical details and images was obtained from the patient.

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Author Contributions

All listed authors took part in the design of the study, performed the experiment and acquired the data, performed the data analysis and interpretation, drafted and revised the article, approved the version to be published, and agreed to be accountable for all aspects of this work.

Disclosure

The authors report no conflicts of interest in this work.

References

1. Levitt MD. Production and excretion of hydrogen gas in man. N Engl J Med. 1969;281(3):122-127.
2. Levitt MD. Intestinal gas production-recent advances in flatology. N Engl J Med. 1980;302(26):1474-1475.
3. Ohsawa I, Ishikawa M, Takahashi K, et al. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med. 2007;13(6):688-694.
4. Yang Y, Zhu Y, Xi X. Anti-inflammatory and antitumor action of hydrogen via reactive oxygen species. Oncol Lett. 2018;16(3):2771-2776.
5. Wang D, Wang L, Zhang Y, Zhao Y, Chen G. Hydrogen gas inhibits lung cancer progression through targeting SMC3. Biomed Pharmacother. 2018;104:788-797.
6. Liu MY, Xie F, Zhang Y, et al. Molecular hydrogen suppresses glioblastoma growth via inducing the glioma stem-like cell differentiation. Stem Cell Res Ther. 2019;10(1):145.
7. Akagi J, Baba H. Hydrogen gas restores exhausted CD8+ T cells in patients with advanced colorectal cancer to improve prognosis. Oncol Rep. 2019;41(1):301-311.
8. Rakić M, Patrlj L, Kopljar M, et al. Gallbladder cancer. Hepatobiliary Surg Nutr. 2014;3(5):221-226.
9. Shukla SK, Singh G, Shahi KS, Pant P. Staging, treatment, and future approaches of gallbladder carcinoma. J Gastrointest Cancer. 2018;49(1):9-15.
10. Kao C, McNamara M, Alley C, et al. A complete response after pseudo-progression: pembrolizumab for metastatic Squamous Cell Carcinoma (SCC) of the bladder. Clin Genitourin Cancer. 2019;17(3):e672-e677.
11. Furubayashi N, Negishi T, Uozumi T, et al. Isolated adrenocorticotropic hormone deficiency potentially induced by nivolumab following pseudo-progression in clear cell renal cell carcinoma: a case report. Mol Clin Oncol. 2019;10(2):304-308.
12. Atrash S, Makhoul I, Mizell JS, Hutchins L, Mahmood F. Response of metastatic mucosal melanoma to immunotherapy: it can get worse before it gets better. J Oncol Pharm Pract. 2017;23(3):215-219.
13. Markovic SN, Galli F, Suman VJ, et al. Non-invasive visualization of tumor infiltrating lymphocytes in patients with metastatic melanoma undergoing immune checkpoint inhibitor therapy: a pilot study. Oncotarget. 2018;9(54):30268-30278.
14. Saada-Bouzid E, Defaucheux C, Karabajakian A, et al. Hyperprogression during anti-PD-1/PD-L1 therapy in patients with recurrent and/or metastatic head and neck squamous cell carcinoma. Ann Oncol. 2017;28(7):1605-1611.

15. Kazandjian D, Keegan P, Suzman DL, Pazdur R, Blumenthal GM. Characterization of outcomes in patients with metastatic non-small cell lung cancer treated with programmed cell death protein 1 inhibitors past RECIST version 1.1-defined disease progression in clinical trials. Semin Oncol. 2017;44(1):3-7.
16. Cohen JV, Alomari AK, Vortmeyer AO, et al. Melanoma brain metastasis pseudoprogression after pembrolizumab treatment. Cancer Immunol Res. 2016;4(3):179-182.
17. Knaus HA, Berglund S, Hackl H, et al. Signatures of CD8+ T cell dysfunction in AML patients and their reversibility with response to chemotherapy. JCI Insight. 2018;3(21).
18. Narayanan S, Kawaguchi T, Yan L, Peng X, Qi Q, Takabe K. Cytolytic activity score to assess anticancer immunity in colorectal cancer. Ann Surg Oncol. 2018;25(8):2323-2331.
19. Ahmadzadeh M, Johnson LA, Heemskerk B, et al. Tumor antigen-specific CD8 T cells infiltrating the tumor express high levels of PD-1 and are functionally impaired. Blood. 2009;114(8):1537-1544.
20. Sun S, Fei X, Mao Y, et al. PD-1(+) immune cell infiltration inversely correlates with survival of operable breast cancer patients. Cancer Immunol Immunother. 2014;63(4):395-406.
21. Zarour HM. Reversing T-cell dysfunction and exhaustion in cancer. Clin Cancer Res. 2016;22(8):1856-1864.
22. Ogando J, Sáez ME, Santos J, et al. PD-1 signaling affects cristae morphology and leads to mitochondrial dysfunction in human CD8(+) T lymphocytes. J Immunother Cancer. 2019;7(1):151.
23. Sobue S, Inoue C, Hori F, Qiao S, Murate T, Ichihara M. Molecular hydrogen modulates gene expression via histone modification and induces the mitochondrial unfolded protein response. Biochem Biophys Res Commun. 2017;493(1):318-324.
24. Kamimura N, Ichimiya H, Iuchi K, Ohta S. Molecular hydrogen stimulates the gene expression of transcriptional coactivator PGC-1alpha to enhance fatty acid metabolism. NPJ Aging Mech Dis. 2016;2:16008.
25. Handschin C, Spiegelman BM. Peroxisome proliferator-activated receptor gamma coactivator 1 coactivators, energy homeostasis, and metabolism. Endocr Rev. 2006;27(7):728-735.

Hydrogen therapy can be used to control tumor progression and alleviate the adverse events of medications in patients with advanced non-small cell lung cancer

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Abstract

Chemotherapy, targeted therapy, and immunotherapy are used against advanced non-small cell lung cancer. A clinically efficacious method for relieving the adverse events associated of such therapies is lacking. Fifty-eight patients who refused drug treatment were assigned equally and randomly to a hydrogen (H_2)-only group and a control group. According to the results of tumor-gene mutations and drug-sensitivity tests, 10, 18, and 10 patients were enrolled into chemotherapy, targeted therapy, and immunotherapy groups in which these therapies were combined with H_2-therapy, respectively. Patients underwent H_2 inhalation for

4-5 hours per day for 5 months or stopped when cancer recurrence. Before study initiation, the demographics (except for tumor-mutation genes) and pulmonary symptoms (except for moderate cough) of the five groups showed no significant difference. During the first 5 months of treatment, the prevalence of symptoms of the control group increased gradually, whereas that of the four treatment groups decreased gradually. After 16 months of follow-up, progression-free survival of the control group was lower than that of the H_2-only group, and significantly lower than that of H_2 + chemotherapy, H_2 + targeted therapy, and H_2 + immunotherapy groups. In the combined-therapy groups, most drug-associated adverse events decreased gradually or even disappeared. H_2 inhalation was first discovered in the clinic that can be used to control tumor progression and alleviate the adverse events of medications for patients with advanced non-small cell lung cancer. This study was approved by the Ethics Committee of Fuda Cancer Hospital of Jinan University on December 7, 2018 (approval No. Fuda20181207), and was registered at ClinicalTrials.gov (Identifier: NCT03818347) on January 28, 2019.

Key words: adverse event; chemotherapy; hydrogen; immunotherapy; non-small-cell lung cancer; NSCLC; PFS; progression-free survival; targeted drug

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INTRODUCTION

Lung cancer is the most prevalent form of cancer-related disease worldwide, of which non-small-cell lung cancer (NSCLC) accounts for ~85%. Many patients are at an advanced stage of lung cancer when the diagnosis is made, and progression-free survival (PFS) is usually < 6 months. In these cases, the tumors cannot be resected; therefore, chemotherapy, targeted therapy and immunotherapy are common treatment options.

Each treatment has different indications, and also produces different adverse events. Serious adverse events (e.g., severe granulocytopenia, thrombocytopenia, abnormal liver function) often lead to significant changes in vital signs and patients are forced to stop or change medications.

Hydrogen (H_2) is an anti-inflammatory, antioxidant, and antiapoptotic molecule. It can diffuse into mitochondria, neutralize reactive oxygen species selectively, and restore cell viability by regulating expression of various genes. In animal experiments, H_2 has been demonstrated to alleviate the serious adverse events caused by chemotherapy, such as targeted therapy. H_2 has been used in clinical trials of multiple non-neoplastic diseases, indicating the safety of H_2 gas inhalation.

In the present study, H_2 therapy was used to control cancer progression and alleviate the adverse events of multiple standard therapies in patients with advanced NSCLC.

SUBJECTS AND METHODS

The inclusion and exclusion criteria

This clinical trial was registered at ClinicalTrials.gov (Identifier: NCT03818347) on January 28, 2019. The enrolled patients were divided into five groups according to the precise medical test results of tumors. The inclusion criteria were patients with stage-III or -IV NSCLC diagnosed by imaging and pathology with specialist doctor; tumor number 1-6; maximum tumor length <

2 cm; Karnofsky performance status (KPS) score ≥ 70; expected survival time > 6 months; platelet count ≥ 80 × 10^9/L; white blood cell count ≥ 3 × 10^9/L; neutrophil count ≥ 2 × 10^9/L; hemoglobin ≥ 80 g/L. The exclusion criteria were patients with: a cardiac pacemaker; brain metastasis; grade-3 hypertension or diabetic complications; severe cardiac and pulmonary dysfunction. This study protocol received ethical approval from the Ethics Committee of Fuda Cancer Hospital of Jinan University on December 7, 2018 (approval No. Fuda20181207). Written informed consent was obtained from each patient.

Subjects

Between June and September 2019, 58 patients with advanced NSCLC at Fuda Cancer Hospital of Jinan University met the inclusion criteria mentioned above and were enrolled in the study. Thirty-four patients were in advance stage when diagnosis, and Immunohistochemical assays of the ratio of programmed cell death-1, programmed cell death-1 ligands, tumor mutation burden, and microsatellite instability in tumor specimens were undertaken. Based on the results, 10 patients were administered a drug based on antibodies against programmed cell death-1 (Nivolumab [Opdivo®, Bristol-Myers Squibb, New York, NY, USA] or Pembrolizumab [Keytruda®, Merck, Kennyworth, NJ, USA]). Through detection of gene mutations in tumor specimens, 18 patients were selected to be given targeted therapy. Patients with a mutation in the epidermal growth factor receptor gene were administered Osimertinib (Tagrisso®, AstraZeneca, London, UK), Gefitinib (Iressa®, AstraZeneca); patients with a mutation in the anaplastic lymphoma kinase gene or receptor tyrosine kinase-1 gene were administered Crizotinib (Xalkori®, Pfizer, New York, NY, USA). Based on the drug-sensitivity tests, 10 patients were administered chemotherapy (Cisplatin or Carboplatin [both from Qilu Pharmaceutical, Haikou, Hainan Province, China]).

The remaining 20 patients who were not sensitive to common drugs or who failed to respond to treatment by common drugs were distributed evenly in the H_2-monotherapy group or control group. Three groups of patients who had H_2 treatment combined with another treatment (immunotherapy, targeted therapy or chemotherapy) started therapy before enrollment of our clinical trial. Most of those patients experienced significant effects on cancer-related lung symptoms after taking combination therapy, but new drug-related adverse events emerged. Lung symptoms or drug-related adverse events were compared before and after H_2 treatment. The comparison of tumor or drug-related symptoms in each group after hydrogen treatment and the changes of treatment are shown in Figure 1. This study followed the Transparent Reporting of Evaluations with Nonrandomized Designs (TREND) Statement.

> *Figure 1. Symptom comparison of patients in each group before and after hydrogen treatment. Note: Chemo: Chemotherapy; H_2: hydrogen; Immuno: immunotherapy; NSCLC: non-small-cell lung cancer; Target: targeted therapy.*

H_2 inhalation

H_2 was produced by a H_2-oxygen nebulizer (H_2 66.7% and oxygen 33.3%; maximum gas flow, 3 L/min; AMS-H-03, Shanghai Asclepius Meditec, Shanghai, China). The control group underwent a sham procedure (H_2 0% and oxygen 33.3%; maximum gas flow 3 L/min; Shanghai Asclepius Meditec). Enrolled patients inhaled the gas mixture for 4-6 hours every day and underwent imaging examination every month until the existing lesions progressed significantly or new metastases appeared. PFS was calculated from the start of H_2 inhalation to tumor progression. All patients received computed tomography or magnetic resonance

examination every month. If the existing tumors grew up significantly or new metastases appeared, it is considered as tumor progression.

Pulmonary symptoms and drug-associated adverse events

The respiratory function of enrolled patients before H_2 therapy was assessed by very experienced respiratory physicians using a pulmonary function tester (Autospiro AS-507; Minato Medical Science, Tokyo, Japan). Pulmonary tumor-related symptoms and the KPS score of all patients before H_2 inhalation were evaluated. The adverse events of chemotherapy, targeted therapy or immunotherapy were assessed by the same respiratory physicians according to Common Terminology Criteria for Adverse Events 5.0.

Statistical analyses

Before H_2 treatment, the demographics and tumor-associated symptoms of patients were compared using chi-squared and Fisher's exact tests; respiratory function and the KPS score were compared using one-way analysis of variance and Bonferroni's multiple comparison test. After H_2 treatment, each tumor-associated symptom and drug-associated adverse event was compared using linear regression analysis; the PFS of each group was compared by one-way analysis of variance and Bonferroni's multiple comparison test. $P < 0.05$ was considered significant difference. Analyses were done using Prism 5.0 (GraphPad, San Diego, CA, USA).

RESULTS

Clinical data of advanced non-small cell lung cancer patients with H_2 inhalation treatment

Patients underwent H_2 inhalation per day for 5 months or stopped the inhalation when cancer relapsed. There was no significant difference in most patient characteristics (e.g., sex) in each group. However, the targeted therapy-H_2-therapy group had a higher proportion of tumor-gene mutations than the other four groups (P = 0.005; Table 1).

> *Table 1. Patient demographics of advanced non-small cell lung cancer patients in different groups* (columns: Control (n = 10); H_2 (n = 10); Immuno-H_2 (n = 10); Target-H_2 (n = 18); Chemo-H_2 (n = 10); P-value. Rows cover Sex, Age (yr), Pathology, TNM stage, Tumor number, tumor location (lung/mediastinum and pleura, brain, bone, others), and Tumor-gene mutation (EGFR, ALK, ROS1, Not found). Note: Data are expressed as number (percent), and analyzed by chi-squared and Fisher's exact tests. EGFR: epidermal growth factor receptor; ALK: anaplastic lymphoma kinase; ROS: receptor tyrosine kinase; TNM: tumor-node-metastasis.)*

Pulmonary signs and symptoms of advanced non-small cell lung cancer patients with H_2 inhalation treatment

Before the start of H_2 therapy, there was no significant difference in pulmonary function or the KPS score among the five groups of patients (Table 2). The prevalence of most pulmonary symptoms (e.g., mild dyspnea, non-cardiac chest pain, pleural effusion, and hemoptysis) was similar among groups. The prevalence of most pulmonary symptoms in the control group, H_2-monotherapy group, and immunotherapy-H_2-therapy group was higher than that in the target therapy-H_2-therapy group and chemotherapy-H_2-therapy group (P = 0.0137).

> *Table 2. Pulmonary signs and symptoms before hydrogen therapy of advanced non-small cell lung cancer patients with H_2 inhalation treatment* (columns: Control (n = 10); H_2 only (n = 10); Immuno-H_2 (n = 10); Target-H_2 (n = 18); Chemo-H_2 (n = 10); P-value. Rows: Respiratory function (FEV1 (L), FVC (L)), KPS score, and Tumor-associated symptoms

(Moderate cough, Mild dyspnea, Non-cardiac chest pain, Mild pleural effusion, Mild hemoptysis). Note: Data are expressed as mean ± SD or number. Respiratory function and the KPS score were compared using one-way analysis of variance and Bonferroni's multiple comparison tests. Tumor-associated symptoms were compared using chi-squared and Fisher's exact tests. FEV1: forced expiratory volume in one second; FVC: forced vital capacity; KPS: Karnofsky performance status.)*

Tumor-associated symptoms of advanced non-small cell lung cancer patients with H_2 inhalation treatment

At the beginning of H_2 treatment, the prevalence of tumor-associated symptoms in the control group and H_2-monotherapy group was similar (P = 0.9994). With prolongation of the treatment time, in the control group, the prevalence of moderate cough (P = 0.0023), mild dyspnea (P = 0.0019), mild non-cardiac chest pain (P = 0.0006), mild pleural effusion (P = 0.0023), and mild hemoptysis (P = 0.0028) increased significantly (Figure 2A). In the H_2-monotherapy group, the prevalence of moderate cough (P = 0.0014), mild dyspnea (P = 0.0247), mild non-cardiac chest pain (P = 0.0015), mild pleural effusion (P = 0.0136), and mild hemoptysis (P = 0.0048) decreased significantly (Figure 2B).

Figure 2. Tumor-associated symptoms varied with the inhalation time of hydrogen (H_2). Note: (A) Control group (inhalation of 33.3% oxygen gas and no H_2) (n = 10). (B) H_2-only group (inhalation of 66.7% H_2 + 33.3% O_2) (n = 10). Each tumor-associated symptom was compared using linear regression analysis.

Drug-associated adverse events of advanced non-small cell lung cancer patients with H_2 inhalation treatment

At the beginning of H_2 treatment, the prevalence of tumor-associated symptoms in the three groups was similar (P ≥ 0.5120), but drug-associated symptoms in the three groups were quite different (Figure 3). With the prolongation of the treatment time, the prevalence of cough and non-cardiac chest pain (P = 0.0013), maculopapular rash (P = 0.0064), hepatobiliary disease (P = 0.0001), and dizziness and headache (P = 0.0111) decreased significantly, but diarrhea did not (P = 0.4144) (Figure 3A). In the target therapy-H_2-therapy group, the prevalence febrile granulocytopenia (P = 0.0051) and vomiting (P = 0.0026), nausea, insomnia (P = 0.0144), maculopapular rash (P < 0.0001), and oral mucositis (P = 0.0007) decreased significantly (Figure 3B). In the chemotherapy-H_2-therapy group, anemia and febrile granulocytopenia (P = 0.0086), thrombocytopenia (P = 0.0009), constipation and diarrhea (P = 0.0129) decreased significantly, but nausea and anorexia (P = 0.0720; Figure 3C) did not.

Figure 3. Drug-associated symptoms varied with the inhalation time of hydrogen (H_2). Note: (A) Immunotherapy-H_2 group (n = 10). (B) Target-H_2 therapy group (n = 18). (C) Chemotherapy-H_2 therapy group (n = 10). Drug-associated adverse event was compared using linear regression analysis.

PFS of advanced non-small cell lung cancer patients with H_2 inhalation treatment

After 16 months of follow-up, all 58 patients developed tumor progression. PFS for the control group was 4.4 ± 1.2 months, whereas that for the H_2-only group was 7.9 ± 2.2 months, H_2-immunotherapy group was 10.1 ± 2.6 months, H_2-targeted therapy group was 9.4 ± 3.1 months, and H_2-chemotherapy group was 8.5 ± 3.0 months. PFS of the four treatment groups was longer than that of the control group, and that of the three H_2 therapy-combination groups was prolonged significantly (Figure 4).

DISCUSSION

Molecular H_2 has been used to treat pulmonary symptoms in animal models of acute lung injury, asthma and chronic obstructive pulmonary disease. The principle of H_2 therapy includes inhibition of secretion of cytokines such as interleukin-4, interleukin-13, interleukin-6 and tumor necrosis factor-α. H_2 therapy can alleviate pulmonary inflammation without impairing anti-tumor effects. Therefore, H_2 gas can be adopted as adjuvant therapy to suppress these symptoms.

> *Figure 4. Comparison of progression-free survival (PFS) between groups after hydrogen (H_2) treatment. Note: PFS of each group was compared by one-way analysis of variance and Bonferroni's multiple comparison test.* \P < 0.05, \\P < 0.01, *P < 0.001.*

Chemotherapy, targeted therapy, and immunotherapy are first-line treatments against advanced NSCLC. The vastly increased generation of reactive oxygen species during treatment is believed to contribute to adverse events, resulting in oxidative stress, inflammation and apoptosis. In the present study, H_2 therapy was shown to alleviate drug-related adverse events, most of which have been reported in animal models, including lung injury caused by various factors, hepatobiliary diseases, maculopapular rash, diarrhea and constipation, nausea and vomiting, oral mucositis, anemia, thrombocytopenia, and anorexia. We found that the prevalence of insomnia, dizziness and headache could be reduced significantly after H_2 inhalation, which could be related to relief of diseases of the central nervous system, such as cerebral hemorrhage, Parkinson's disease and Alzheimer's disease. The mechanism of action observed in animal experiments could be used as a reference for clinical research. Surprisingly, H_2 therapy, which is non-toxic and can alleviate adverse events in multiple organs simultaneously, has been used rarely.

We found that H_2 monotherapy could prolong the PFS of patients with advanced NSCLC from 4.4 ± 1.2 months to 7.9 ± 2.2 months, suggesting that H_2 could inhibit the growth of lung cancer cells independently. This hypothesis has been bolstered by data from an in vitro and in vivo study, which confirmed that H_2 can inhibit the proliferation, migration, and invasion of the lung-cancer cell lines and tumor growth in mouse model. These data suggested that H_2 could serve as new therapy against lung cancer. However, for patients eligible for first-line treatment, drugs will produce more pronounced effects upon tumor control. Whether a combination of drug therapy and H_2 therapy can elicit better tumor control must be studied further, but gradual reduction of most drug-associated adverse events after H_2 inhalation is clear.

Several detective methods of molecular H_2 are available and convenient: inhalation, drinking H_2-dissolved water, injection with H_2-saturated saline, and taking a H_2 bath. H_2 is non-toxic, inexpensive, can be administered readily, can diffuse into tissues and cells and cross the blood-brain barrier. Hence, H_2 could be used to treat tumors of the head, neck and chest.

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Informed consent statement: The authors certify that they have obtained all appropriate patient consent forms. In the form the patients have given their consent for the images and other clinical information to be reported in the journal. The patients understand that their names and initials will not be published and due efforts will be made to conceal their identity.

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References

1. Siegel RL, Miller KD, Jemal A. Cancer statistics, 2019. CA Cancer J Clin. 2019;69:7-34.
2. Reinke LF, Feemster LC, Backhus LM, Gylys-Colwell I, Au DH. Assessment and management of symptoms for outpatients newly diagnosed with lung cancer. Am J Hosp Palliat Care. 2016;33:178-183.

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A narrative review of hydrogen oncology: from real world survey to real world evidence

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Abstract

The use of hydrogen for cancer control has made great progress in cytology and animal experiments. With the increasing number of hydrogen products on the market, larger numbers of advanced cancer patients have participated in clinical trials or received treatment at home after purchase. Our study reported a real world survey from 82 patients with good cancer control using hydrogen products, including many patients who received traditional treatment. Two typical cases were reported herein. Subsequently, we included studies on the mechanism of hydrogen oncology. The mechanism of cancer control using hydrogen includes the inhibition of tumor cells and the activation of exhausted lymphocytes. Large-scale real world evidence has shown clinical value, and yet remains to be further developed and researched.

Key words: cancer; hydrogen; lymphocyte; mechanism; real world evidence; real world survey; RWE; RWS

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INTRODUCTION

Hydrogen gas or molecular hydrogen has been confirmed as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. A TV program called "The Truth of Holy Water" was shown on the Discovery Truth channel in Japan on June 13, 1998. In summary, there is an abandoned mine pit in Nordennau, a well-known small town located in the northwestern part of Düsseldorf, Germany. Every year, thousands of people with a variety of diseases, including cancer, come to Nordennau aiming to find "holy water" from the local mine diggings to cure their illnesses. In April 1986, a group of children who suffered from leukemia resulting from the Chernobyl nuclear disaster were brought by the owner (Theo Tom) of the mine to drink the holy water. A few months later, most of the children presented with significant symptomatic improvement, among them one case showed complete recovery. These results caused a mass uproar in the media. Subsequently, it was discovered that the large amount of hydrogen gas contained in the holy water. Since a real world survey of 82 patients with advanced cancer from China was reported, "hydrogen oncology" obtained the real world evidence for the first time.

The articles used in this oncology review were retrieved by replicating the search terms of Chen et al. An electronic search of the Medline database for literature describing clinical hydrogen therapy for cancer patients from 2000 to 2019. The results were further screened by title and abstract to only cancer treatment, not Complications treatment. The main cited article is from the latest Science Citation Index paper report in 2019.

REAL WORLD SURVEY OF HYDROGEN ONCOLOGY

Approximately 40 years ago, Dole et al reported that hydrogen exhibited effects on inhibiting squamous cell carcinoma cells. However, despite being published in the journal Science, the report did not attract much attention, possibly because hydrogen is simple and ordinary.

We have conducted a voluntary field survey in patients diagnosed with cancer who inhaled hydrogen at home or at rehabilitation centers from 2014 to 2019. Follow-up was performed on 82 patients with advanced (stage III and IV) cancer. The patients used the Hydrogen-oxygen Atomizer (AMS-H-03, Shanghai Asclepius Meditec Co., Ltd., Shanghai, China), which provided 66.7% hydrogen with mixed 33.3% oxygen with gas flow at 3000 mL/min.

REAL WORLD EVIDENCE OF HYDROGEN ONCOLOGY

Primary results of clinical observation

Follow-up time was between 3-46 months, with a median of 6 months. During the follow-up period, 12 patients died of disease progression and complications with stage IV cancer. The patient quality of life was prospectively evaluated by quality of life questionnaire core 30 (QLQ-C30). All quality of life areas improved after four weeks of hydrogen inhalation, with the most significant improvements in fatigue, insomnia, loss of appetite, and pain. A performance status assessment was completed on all patients according to Zubrod-ECOG-WHO (ZPS, 5-point method), and 41.5% of patients showed improved performance status, 34.1% of patients were stable, and 24.4% of patients declined. Lung cancer patients had the highest improvement rate, and pancreatic cancer and gynecologic cancer patients had the lowest improvement rate. Tumor markers, including alpha-fetoprotein, carcinoembryonic antigen, CA19-9, CA125, CA153, and CA724, were observed. A total of 58 cases exhibited one or more abnormal tumor markers. A decrease of tumor markers was observed in 36.2% of patients, 15.5% of patients had stable expression of tumor markers, and 48.3% of patients showed a decline in tumor markers and (13-45 days (median 23 days)) following hydrogen inhalation. The highest rate of tumor marker decrease was seen in lung cancer and the lowest in pancreatic and hepatic malignancies.

A total of 80 cases had tumors that could be evaluated by imaging analysis. Evaluation of oncologic response according to the Response Evaluation Criteria in Solid Tumors criteria showed complete response in 1 case (1.3%), partial response in 15 cases (18.8%), stable disease in 30 cases (37.5%), and progressive disease in 34 cases (42.5%) after hydrogen inhalation, with a total disease control rate of 57.5%. Complete response and partial response appeared between 21-80 days with a median time of 55 days after hydrogen inhalation. The disease control rate was 83.0% in stage III and 47.7% in stage IV patients ($P < 0.05$), respectively. No hematological toxicity was observed. Stomach upset, dizziness, headache, and nasal mucus at the beginning of hydrogen inhalation were observed in individual cases but quickly disappeared spontaneously.

Typical clinical cases

The hydrogen control of cancer confirmed by the real world survey is clearly illustrated in the following two cases.

Case 1: A 72-year-old female patient was hospitalized on September 20, 2018 with stage IV gallbladder cancer with massive intrahepatic and the lymph node metastases in the hilar area and around the head of the pancreas. There was involvement of the duodenum that caused upper gastrointestinal obstruction. Biopsy showed poorly differentiated adenocarcinoma. The patient was extremely exhausted, accompanied by severe anemia with hemoglobin as low as 37 g/L and severe hypoalbuminemia with albumin as low as 23 g/L. The patient had a history of rheumatic heart disease with recent cardiac failure, and fasting blood glucose had been elevated (up to 22 mM) for a long time. Blood tumor markers CA19-9, alpha-fetoprotein, and carcinoembryonic antigen were increased. In addition to receiving symptomatic and supportive care, any conventional "anti-cancer" treatment could not be used. Beginning on October 23, 2018 the patient volunteered to inhale hydrogen-oxygen mixed gas only through a nasal cannula, two hours per day initially, and gradually increased to 6 hours per day. After one month of hydrogen inhalation, the patient showed obvious pseudo-progression of the disease and quickly relieved after symptomatic treatments. After 2 months of hydrogen

inhalation, the patient's performance status showed significant improvement (decreased from 5 to 3). After 3 months of hydrogen inhalation, tumor markers were reduced to the normal reference range, and CT showed more than a 50% reduction in gallbladder tumors and multiple intrahepatic and hilar metastases (defined as partial response). The patient's blood sugar dropped to normal when insulin was no longer used. The patient was discharged on February 1, 2019. Currently, the patient continues to inhale hydrogen every day at home and is in the process of further improvement (recent performance status of 0).

Case 2: A 44-year-old woman diagnosed with lung cancer with multiple metastases in November 2015. Oral targeted drugs were initiated after removal of brain metastases, and most lesions remained stable for 28 months. In March 2018, intracranial multiple metastases, as well as hydrocephalus accumulation in the third ventricle and lateral ventricles, and metastases in bone, adrenal gland, liver were noted. Hydrogen monotherapy was started to control the tumor a month later. After 4 months, the size of multiple brain tumors was reduced significantly, and the amount of hydrocephalus in the third ventricle and lateral ventricles reduced significantly. After 1 year, all brain tumors had disappeared, and there were no significant changes in metastases in the liver and lung. These data show that, after standard treatments had failed, hydrogen-gas monotherapy elicited significant effective control of tumors (especially those in the brain), and survival time was lengthened.

Case 3: Three patients with nasopharyngeal carcinoma developed binaural secretory otitis media 0.5, 2, and 12 years after radiotherapy, respectively. The secretions subsided after conventional drug and drainage treatments, but hearing continued to deteriorate until severe loss was documented in both ears. After examination of the eardrum and tympanum, patients were enrolled in the first half of 2019. After 0.5, 1 and 2 months of continuous hydrogen-oxygen therapy, our first three patients reported different levels of improvement in binaural hearing. This is the first report to show that, after treatment for nasopharyngeal carcinoma, hearing loss can be alleviated using hydrogen-oxygen therapy.

MECHANISMS OF HYDROGEN CANCER CONTROL

Hydrogen gas suppresses tumor formation and growth

Hydrogen cancer control has been supported by serial experimental studies in cancer cells and tumor models, which have demonstrated the inhibitory effects of hydrogen on cancer cell formation as well as proliferation, viability, cell cycle, migration, and invasion of kidney and liver cancer cells. Hydrogen treatment has been shown to decrease the expression of CD34, demonstrating its anti-angiogenesis effects in ovarian cancer. A previous finding showed that molecular hydrogen could markedly inhibit the sphere-forming ability of ovarian cancer cells, cervical cancer cells, and glioma cells. There is evidence to suggest that hydrogen inhibited lung cancer progression through the down-regulation structural maintenance of chromosomes protein 3, a regulator for chromosome condensation. In human esophageal squamous cell carcinoma (KYSE-70) cells, hydrogen exerted its anticancer effects via inducing hydrogen peroxide accumulation, cell cycle arrest, and apoptosis induction mediated by mitochondrial apoptotic pathways. In a rat orthotopic glioma model, hydrogen gas was shown to inhibit several markers involved in stemness, resulting in the suppression of sphere formation, cell migration, invasion, and colony formation of glioma cells.

Hydrogen gas restores exhausted CD8+ T cells

Hydrogen has been shown to modulate immune function. A previous report showed that inhalation of hydrogen gas decreased the abundance of exhausted terminal programmed cell

death protein 1 $(PD\text{-}1)^+$ $CD8^+$ T cells, increased active terminal PD-1- $CD8^+$ T cells, and restored exhausted $CD8^+$ T cells to improve progression free survival and overall survival in patients with stage IV colorectal cancer. Hydrogen gas was recently reported to activate peroxisome proliferator-activated receptor γ coactivator-1α, inactivation of which induces mitochondrial dysfunction. Thus, increasing evidence has shown that hydrogen gas can directly inhibit the growth of certain cancer cells and can restore the vitality of exhausted cytotoxic T lymphocytes (Figure 1).

Hydrogen gas reverses adaptive and innate immune system senescence

Twenty advanced non-small cell lung cancer (NSCLC) patients were enrolled to evaluate the immunosenescence of peripheral blood lymphocyte subsets before and after 2 weeks of hydrogen inhalation. Before treatment, the abnormally higher indexes included exhausted cytotoxic T cells, senescent cytotoxic T cells, and killer Vδ1 cells. After two weeks of hydrogen therapy, the number of exhausted and senescent cytotoxic T cells decreased to within the normal range, and there was an increase in killer Vδ1 cells. The abnormally lower indexes included functional helper and cytotoxic T cells, Th1, total NKT cells, killer NK, and Vδ2 cells. All six cell subsets recovered to within the normal range after treatment. The current data indicate that the immunosenescence of advanced NSCLC involves nearly all lymphocyte subsets, and 2 weeks of hydrogen treatment can significantly improve most of these indexes.

Hydrogen gas controls tumor progression and alleviate the adverse events of medications

Fifty-eight adult patients were enrolled to relieve pulmonary symptoms (e.g., moderate cough, mild dyspnea, mild non-cardiac chest pain, mild pleural effusion and mild hemoptysis) or the adverse events of drugs (e.g., severe granulocytopenia, thrombocytopenia and abnormal liver function). During the first 5 months of treatment, the prevalence of pulmonary symptoms of the control group (oxygen inhalation) increased gradually, whereas that of the four treatment groups (hydrogen inhalation only and combination with chemotherapy, target therapy and immunotherapy) decreased gradually. After 16 months of follow-up, progression-free survival of the control group was significantly shorter than that of the four treatment groups. In the three combination-therapy groups, most drug-associated adverse events decreased gradually or even disappeared.

> *Figure 1. Two main mechanisms of hydrogen (H_2) cancer control. Note: PD-1: Programmed cell death protein 1.*

CHALLENGES AND QUESTIONS

Hydrogen medicine is new and using hydrogen to control cancer is unconventional for most people, including professional medical staff. According to our real world survey, inhaling hydrogen can improve quality-of-life and control cancer progression in patients with advanced cancer, and could prolong life expectancy. Professor Zhao-You Tang, a highly respected Chinese oncologist from Hepatoma Institute of Fudan University (China), stated the following: "I was very dubious about the use of hydrogen in cancer control when the first patient case showed to me. But when I was shown nearly 50 cases successively, I became convinced."

Cancer treatment remains a huge challenge. As a simple, cheap, and safe way to control cancer, hydrogen can be used as a complimentary option to conventional therapies for cancer.

We plan to continue to perform real world survey and show real world evidence toward establishing "hydrogen oncology."

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References

1. Ohta S. Molecular hydrogen as a novel antioxidant: overview of the advantages of hydrogen for medical applications. Methods Enzymol. 2015;555:289-317.
2. Chen JB, Kong XF, Lv YY, et al. "Real world survey" of hydrogen-controlled cancer: a follow-up report of 82 advanced cancer patients. Med Gas Res. 2019;9:115-121.
3. Dole M, Wilson FR, Fife WP. Hyperbaric hydrogen therapy: a possible treatment for cancer. Science. 1975;190:152-154.
4. Bjordal K, de Graeff A, Fayers PM, et al. A 12 country field study of the EORTC QLQ-C30 (version 3.0) and the head and neck cancer specific module (EORTC QLQ-H&N35) in head and neck patients. EORTC Quality of Life Group. Eur J Cancer. 2000;36:1796-1807.
5. Chow R, Chiu N, Bruera E, et al. Inter-rater reliability in performance status assessment among health care professionals: a systematic review. Ann Palliat Med. 2016;5:83-92.
6. Chen J, Mu F, Lu T, Ma Y, Du D, Xu K. A gallbladder carcinoma patient with pseudo-progressive remission after hydrogen inhalation. Onco Targets Ther. 2019;12:8645-8651.
7. Chen J, Pan ZB, Du DM, et al. Hydrogen gas therapy induced shrinkage of metastatic gallbladder cancer: A case report. World J Clin Cases. 2019;7:2065-2074.
8. Chen J, Mu F, Lu T, Du D, Xu K. Brain metastases completely disappear in non-small cell lung cancer using hydrogen gas inhalation: a case report. Onco Targets Ther. 2019;12:11145-11151.
9. Chen J, Kong X, Mu F, Lu T, Du D, Xu K. Hydrogen therapy can alleviate radiotherapy-induced hearing loss in patients with nasopharyngeal cancer. Ann Palliat Med. 2019;8:746-751.
10. Li FY, Zhu SX, Wang ZP, Wang H, Zhao Y, Chen GP. Consumption of hydrogen-rich water protects against ferric nitrilotriacetate-induced nephrotoxicity and early tumor promotional events in rats. Food Chem Toxicol. 2013;61:248-254.
11. Kawai D, Takaki A, Nakatsuka A, et al. Hydrogen-rich water prevents progression of nonalcoholic steatohepatitis and accompanying hepatocarcinogenesis in mice. Hepatology. 2012;56:912-921.
12. Shang L, Xie F, Li J, et al. Therapeutic potential of molecular hydrogen in ovarian cancer. Transl Cancer Res. 2018;7:988-995.
13. Kissebah AH, Sonnenberg GE, Myklebust J, et al. Quantitative trait loci on chromosomes 3 and 17 influence phenotypes of the metabolic syndrome. Proc Natl Acad Sci U S A. 2000;97:14478-14483.
14. Wang D, Wang L, Zhang Y, Zhao Y, Chen G. Hydrogen gas inhibits lung cancer progression through targeting SMC3. Biomed Pharmacother. 2018;104:788-797.

15. Liu MY, Xie F, Zhang Y, et al. Molecular hydrogen suppresses glioblastoma growth via inducing the glioma stem-like cell differentiation. Stem Cell Res Ther. 2019;10:145.
16. Ohta N, Miwa N. Influence of hydrogen-occluding-silica on migration and apoptosis in human esophageal cells in vitro. Med Gas Res. 2017;7:76-85.
17. Akagi J, Baba H. Hydrogen gas restores exhausted CD8$^+$ T cells in patients with advanced colorectal cancer to improve prognosis. Oncol Rep. 2019;41:301-311.
18. Chen JB, Kong XF, Qian W, et al. Two weeks of hydrogen inhalation can significantly reverse adaptive and innate immune system senescence patients with advanced non-small cell lung cancer. Med Gas Res. 2020;10:72-79.
19. Chen JB, Kong XF, Mu F, Lu TY, Lu YY, Xu KC. Hydrogen therapy can be used to control tumor progression and alleviate the adverse events of medications in patients with advanced non-small cell lung cancer. Med Gas Res. 2020;10:75-80.
20. Xu KC, Chen JB. Hydrogen Oncology. Singapore: World Scientific Publishing Co. Pte. Ltd. 2020.

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Two weeks of hydrogen inhalation can significantly reverse adaptive and innate immune system senescence patients with advanced non-small cell lung cancer: a self-controlled study

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Abstract

Following standard treatments, the traditional model for enhancing anti-tumor immunity involves performing immune reconstitution (e.g., adoptive immune cell therapies or immunoenhancing drugs) to prevent recurrence. For patients with advanced non-small cell lung cancer, we report here on two objectives, the immunosenescence of peripheral blood lymphocyte subsets, and hydrogen gas inhalation for immune reconstitution. From July 1st to September 25th, 2019, 20 non-small cell lung cancer patients were enrolled to evaluate the immunosenescence of peripheral blood lymphocyte subsets, including T cell, natural killer/natural killer T cell and gamma delta T cell. Two weeks of hydrogen inhalation was performed during the waiting period for treatment-related examination. All patients inhaled a mixture of hydrogen (66.7%) and oxygen (33.3%) with gas flow rate of 3 L/min for 4 hours each day. None of the patients received any standard treatment during the hydrogen inhalation period. Before pretreatment testing, major indexes of patients included exhausted cytotoxic T cells, senescent cytotoxic T cells, and killer Vδ1 cells. After 2 weeks of hydrogen therapy, the number of exhausted and senescent cytotoxic T cells decreased to within the normal range, and there was an increase in killer Vδ1 cells. The abnormally lower indexes included functional helper and cytotoxic T cells, Th1, total natural killer T cells, natural killer, and Vδ2 cells. After

2 weeks of hydrogen therapy, all six cell subsets increased to within the normal range. The current data indicate that the immunosenescence of advanced non-small cell lung cancer involves nearly all lymphocyte subsets, and 2 weeks of hydrogen treatment can significantly improve most of these indexes. The study was approved by the Ethics Committee of Fuda Cancer Hospital, Jinan University in China (approval No. Fuda20181207) on December 7th, 2018, and was registered on ClinicalTrials.gov (ID: NCT03818347) on January 24th, 2019.

Key words: adaptive and innate immune system; gamma delta T cell; hydrogen inhalation; immunosenescence; natural killer cell; natural killer T cell; non-small cell lung cancer; T cell

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INTRODUCTION

The recurrence and metastasis of malignant tumors are difficult to control, which can be attributed to three main reasons: 1) immunosenescence; 2) tumor-associated factors; and 3) cancer therapy itself. Immunosenescence is characterized by the gradual deterioration of the immune system during the natural ageing process. A long-term tumor-bearing state will also lead to the exhaustion and senescence of cytotoxic lymphocytes, including T cells, natural killer (NK) cells, NKT cells, and gamma delta (γδ) T cells. Tumor factors are also associated with an excessive tumor burden and severely inhibited immune function, including increased $CD4^+CD25^+$ regulatory T cells. Currently, a variety of conventional treatments have been shown to accelerate the recurrence and metastasis of residual tumors, including surgery, radiation, chemotherapy, and even general anesthesia.

Of all cancers, lung cancer is associated with the highest incidence throughout the world, of which non-small cell lung cancer (NSCLC) accounts for approximately 85% of cases. Due to the lack of early symptoms for lung cancer, many patients are in an advanced stage at the time of diagnosis, and survival time is typically with 1 year. Studies have shown that nearly 27% of patients who underwent surgery and chemotherapy died from recurrence and metastasis. Thus, immune reconstitution, both before or after standard therapies, is a necessary method for prolonging the survival of cancer patients who are in an advanced stage. The current strategies of immune reconstitution primarily include adoptive immune cell supplementation, specific or non-specific immune activators, cancer vaccines, and exercise. As an antioxidant gas, hydrogen can diffuse into the mitochondria of lymphocytes and selectively neutralize oxygen free radicals. Moreover, hydrogen inhalation has been verified to reduce PD-1 expression, a marker of cell exhaustion, on the surface of cytotoxic T cells in patients with advanced colorectal cancer, which prolongs progression-free survival and overall survival. In this study, immunosenescence in advanced NSCLC patients was tested, and hydrogen inhalation was used to reverse the immunosenescence while patients were awaiting treatment.

SUBJECTS AND METHODS

Subjects

Between July and September 2019, 20 advanced NSCLC patients who hospitalized at Fuda Cancer Hospital and needed wait 2 weeks for treatment were enrolled in this self-controlled study. According to the registered clinical trial (ClinicalTrials.gov, ID: NCT03818347;

Registration Date: January 24, 2019), the inclusion criteria consisted of: stage III or IV NSCLC diagnosed by imaging and pathology; tumor number 1-6; maximum tumor length < 2 cm; Karnofsky performance status (KPS) score ≥ 70%; expected survival time > 6 months; platelet count ≥ 80 × 10^9/L; white blood cell count ≥ 3 × 10^9/L; neutrophil count ≥ 2 × 10^9/L; and hemoglobin ≥ 80 g/L. The exclusion criteria consisted of patients with a cardiac pacemaker, brain metastasis, grade 3 hypertension or diabetic complication, severe cardiac and pulmonary dysfunction. None of the patients received any standard treatment during the hydrogen inhalation period.

Ethical statement

The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. The study protocol received ethical approval from the Ethics Committee of Fuda Cancer Hospital, Jinan University in China (approval No. Fuda20181207) on December 7, 2018. Written informed consent was obtained from each participant in accordance with the Declaration of Helsinki.

Hydrogen inhalation method

Hydrogen was produced by a hydrogen-oxygen nebulizer (AMS-H-03, Shanghai Asclepius Meditec Co., Ltd., Shanghai, China). The patients remained seated or recumbent, and inhaled a mixture of hydrogen (66.7%) and oxygen (33.3%) via a nasal tube or mask with a gas flow rate of 3 L/min. The hydrogen inhalation was continued for 4 hours per day for 2 weeks in a specialized treatment facility.

Pulmonary symptoms and KPS score

Before and after hydrogen therapy, the respiratory function was assessed by very experienced respiratory physicians using a pulmonary function tester (Autospiro AS-507; Minato Medical Science, Tokyo, Japan), pulmonary tumor-related symptoms and the KPS score were evaluated by resident doctors in charge.

Immunophenotype evaluation

Approximately 5 mL of peripheral blood was extracted from elbow of all enrolled patients before hydrogen inhalation, as well as at one and two weeks after hydrogen inhalation. Peripheral blood monocyte cells were isolated using a Ficoll solution and labeled with fluorescent antibodies (BD Biosciences, San Jose, CA, USA). T lymphocytes, NK, NKT, and γδ T cells were analyzed using flow cytometry (FACSanto II; BD Biosciences, San Jose, CA, USA) by a professional third-party inspection center (Shuangzhi Purui Medical Laboratory Co., Ltd., Wuhan, Hubei Province, China).

Statistical analysis

The calculation method of sample size is based on MedSci Sample Size tools (MSST) software (MedSci, Shanghai, China). Pulmonary symptoms before and after hydrogen inhalation were compared using paired t-test. Respiratory function, KPS score, and immunoassays were compared by repeated measures analysis of variance and Bonferroni's multiple comparison test. Statistical differences were indicated by $P < 0.05$. All analyses and figures were produced using GraphPad Prism 5.0 (GraphPad software, San Diego, CA, USA).

RESULTS

Clinical data of NSCLC patients

A total of 20 consecutive patients admitted to the hospital participated in this study. The clinicopathological data are listed in Table 1.

Table 1. Patient demographics information

Parameters	Data
Sex — Female	12 (60)
Sex — Male	8 (40)
Age (yr) — 41–60	9 (45)
Age (yr) — 61–70	5 (25)
Age (yr) — 71–80	6 (30)
Pathological type — Adenocarcinoma	9 (45)
Pathological type — Squamous cell carcinoma	8 (40)
Pathological type — Large cell lung cancer	3 (15)
Cancer stage — III	7 (35)
Cancer stage — IV	13 (65)
Metastasis site — Lung, mediastinum and pleura	14 (70)
Metastasis site — Liver	7 (35)
Metastasis site — Bone	9 (45)
Metastasis site — Others	5 (25)

Note: Data are expressed as number (percentage).

Changes in the KPS score and pulmonary symptoms of NSCLC patients

All patients underwent hydrogen inhalation for 2 weeks, with no treatment-related discomfort or side effects. The results regarding respiratory function, KPS score, and pulmonary symptoms before and 2 weeks after hydrogen therapy are presented in Table 2. At the end of hydrogen treatment, fewer people with different lung symptoms, with a statistical reduction in those with moderate cough and mild dyspnea.

Changes in T cell subsets of NSCLC patients undergoing hydrogen therapy

This portion of the test consisted of three parts: $CD4^+$ T cells, $CD8^+$ T cells, and cytokine-secreting helper T (Th) cells. Before the initiation of hydrogen therapy, six primary immunosenescence indexes were identified. Among these indexes, the abnormally lower indexes included a percentage of functional helper and cytotoxic T cells, Th1, and follicular Th cells; the abnormally higher indexes were associated with the percentage of exhausted and senescent cytotoxic T cells (Figure 1). For the $CD4^+$ subsets, functional Th cells increased to within the normal range after one week of hydrogen inhalation and increased significantly after 2 weeks ($P < 0.05$). Both exhausted Th cell and regulatory T cells decreased gradually (both $P < 0.05$) after 2 weeks of hydrogen therapy (Figure 1A). For the $CD8^+$ subsets, functional cytotoxic T cells increased to within the normal range after 2 weeks of hydrogen inhalation ($P < 0.05$); both exhausted and senescent cytotoxic T cells decreased gradually after 2 weeks of hydrogen inhalation ($P < 0.05$) (Figure 1B). For the cytokine-secreting Th subsets, only Th1 cells increased gradually near normal range after 2 weeks of hydrogen inhalation therapy ($P < 0.05$; Figure 1C). No other cells changed significantly within 2 weeks of hydrogen inhalation.

Table 2. Clinical findings in 20 non-small cell lung cancer patients undergoing hydrogen therapy.

Index	Before treatment	2 wk after treatment	P-value
Forced expiratory volume in one second (L)	—	—	< 0.05
Forced vital capacity (L)	1.56±0.59	—	< 0.05
Karnofsky performance status score	1.93±0.57	2.13±0.48	< 0.05
Lung symptoms	—	—	—
Moderate cough	75±6	2.38±0.53	< 0.05
Mild dyspnea	—	88±7	—
Mild haemoptysis	13	—	—
Chest pain	11	5	0.0248
Mild pleural effusion	9	4	0.0484
Lung bubble	8	5	0.3203
—	6	4	0.3008
—	4	4	0.7164
—	4	2	0.6614

Note: Data in the forced expiratory volume in one second, forced vital capacity and Karnofsky performance status score are expressed as the mean ± SD, and were analyzed by repeated measures analysis of variance followed by Bonferroni's multiple comparison test. Data in the lung symptoms are expressed as number and analyzed by t-test.

Figure 1. Immunoassay of the T cell subsets before and after hydrogen inhalation in non-small cell lung cancer patients. Note: (A) Test results of the CD4⁺ subsets. (B) Test results of the CD8⁺ subsets. (C) Test results of the cytokine-secreting Th subsets. The parallel red long lines in the figure represent the normal range, the black short lines represent the average value at each time point, and the pink red line names represent the abnormal indicators before hydrogen treatment. Data were analyzed by repeated measures analysis of variance followed by Bonferroni's multiple comparison test. \P < 0.05, \\P < 0.01, *P < 0.001. CXCR5, C-X-C chemokine receptor type 5; Th, helper T; Tfh, follicular helper T.*

Changes in NKT and NK cells of NSCLC patients undergoing hydrogen therapy

Before the start of hydrogen therapy, the percentage of total NKT cell, activated NK and killer NK subsets was all below the normal range (Figure 2). One week after hydrogen inhalation, the activated NK cell subset increased to within the normal range (Figure 2C). Two weeks later, both the total NKT and killer NK subsets were higher than the percentage before treatment (both $P < 0.05$; Figure 2A and D), and activated NK cells were significantly elevated ($P < 0.01$; Figure 2C). Total NK cells did not change significantly within 2 weeks of hydrogen inhalation (Figure 2B).

Figure 2. Immunoassay of natural killer (NKT and NK) cells before and after hydrogen inhalation in non-small cell lung cancer patients. Note: (A) Changes in the number of NKT cells. (B-D) Test results of the NK subsets. The parallel red long lines in the figure represent the normal range, the black short lines represent the average value at each time point, and the pink red line names represent the abnormal indicators before hydrogen treatment. Data were analyzed by repeated measures analysis of variance followed by Bonferroni's multiple comparison test. \P < 0.05, \\P < 0.01.*

Changes in γδ T cell subsets of NSCLC patients undergoing hydrogen therapy

Before the start of hydrogen therapy, the percentage of both Vδ1 cell and killer Vδ1 cell was higher than the normal range, and the percentage of Vδ2 cell was below the normal range (Figure 3). One week after hydrogen inhalation, the percentage of Vδ2 cell in γδ T cells rose to within the normal range (Figure 3C). Two weeks later, the total γδ T cells and Vδ2 cells was higher than the percentage before treatment (both $P < 0.05$; Figure 3A and C), exhausted Vδ2 cell was lower than that before treatment ($P < 0.05$; Figure 3C); exhausted Vδ1 cell decreased significantly ($P < 0.05$; Figure 3C); and killer Vδ2 cell increased significantly ($P < 0.001$; Figure 3B). No other cells changed significantly ($P < 0.01$; Figure 3C) within 2 weeks of hydrogen inhalation.

> *Figure 3. Immunoassay of the gamma delta (γδ) T cell subsets before and after hydrogen inhalation in non-small cell lung cancer patients. Note: (A) Change in the number of γδ T cells. (B) Test results of the Vδ1 subsets. (C) Test results of the Vδ2 subsets. The parallel red long lines in the figure represent the normal range, the black short lines represent the average value at each time point, and the pink red line names represent the abnormal indicators before hydrogen treatment. Data were analyzed by repeated measures analysis of variance followed by Bonferroni's multiple comparison test.* \P < 0.05, \\P < 0.01, *P < 0.001. NK, Natural killer; PD-1, programmed cell death protein 1.*

Discussion

Although a connection between immunodeficiency and oncogenesis has been proposed, a detailed immune profile in cancer has not yet been described in sufficient detail. It is well-known that the immune system deteriorates with cancer progression as a consequence of decreasing levels of functional T cells, downregulation of costimulatory molecules, and expansion of memory T cells. Chen et al showed that there were differences in the immune profiles between cancer patients and normal subjects; in particular, a decreased proportion of $CD4^+$ and $CD8^+$ T cell subsets was observed in cancer patients, especially in patients who were associated with a more advanced disease stage and those who had received conventional chemotherapy treatment. In addition, Saavedra et al reported that cancer and chemotherapy influenced various lymphocyte subsets in NSCLC patients, in which B cells decreased with cancer disease progression, and $CD28^-$ subpopulations (exhausted or senescent T cells) increased following chemotherapy. Our study found similar results in the adaptive immune system, which was found to be primarily due to an increase in the proportion of exhausted and senescent T cell subsets, as well as a decreased proportion of functional subsets. Moreover, we also found evidence for the weak ability of Th1 and follicular Th cells on cytokine secretion. In the detection of the innate immune response (NK, NKT, and γδ T cell), we discovered that there was a decreased proportion of NKT, activated NK and killer NK cells, and an insufficient content of Vδ2 subsets among γδ T cells. Thus, it is highly likely that the adaptive and innate immune systems jointly contribute to the progression of NSCLC.

Advanced NSCLC has many characteristics that are distinct from other cancers, including that over 65% of patients are often over 70 years old and in most cases, the disease has progressed from early stage cancer and they have been exposed to long-term chronic antigenic stimulation. Therefore, the immunosenescence characteristics obtained in advanced NSCLC can only be used as a reference for other cancer types. Moreover, there may be huge differences among different cancer types, which require further investigation.

Immune cell senescence focuses on the phenotypic characteristics of individual lymphocytes; however, the function does not necessarily decrease during aging. In contrast, immune cell exhaustion is due to the presence of excessive free radicals in the mitochondria, which results

in a loss of function and induces programmed apoptosis. Senescence and exhaustion is not simply the result of an inevitable and progressive decline of all immune functions, but rather represent a highly dynamic process of remodeling and adaptation. Thus, we attempted to rescue the function of exhausted and senescent lymphocytes with hydrogen gas, which was reported to protect the mitochondrial function of human lymphocytes. By comparing the immune function before and after hydrogen inhalation, we found that a period of two weeks was sufficient to reverse multiple senescent or exhausted cell subsets into functional subsets, which had no effect on the percentage of $CD4^+$ or $CD8^+$ T cells. In addition to adaptive immune cells, two natural killer-associated Th1 cytokines and innate immune cells (i.e., NK, NKT, total γδ T cell, Vδ1 cell, and Vδ2 cell) have been found to be significantly increased following hydrogen inhalation. Surprisingly, after hydrogen therapy, the proportion of regulatory T cells in the blood decreased significantly, demonstrating that hydrogen can control tumor progress with different mechanisms. Although the improvement in the number and function of these cells does not necessarily reduce the size of the tumor, it may alter the systemic immunosuppressive state and the tumor microenvironment, as well as reduce tumor invasiveness. Improvements in the systemic signs and pulmonary symptoms after hydrogen inhalation in some patients are likely to validate this hypothesis.

Since most cancer patients need time to complete the admission check and wait for a specific treatment program, we should try to turn this "wasted time" into a "prime time," in an effort to restore the immune system prior to hospital treatment. This study reports primary evidence that two weeks of hydrogen inhalation can significantly reverse the senescence of both the adaptive and innate immune system. The limitations of this paper are the small number of participants and the short time of hydrogen treatment, so the following issues remain unknown: 1) whether the duration of prolonged hydrogen inhalation can continue to improve immune function; 2) the optimal level of immune improvement; 3) whether the therapeutic effects rebound after the cessation of hydrogen inhalation; and 4) whether improving immune function before treatment will reduce tumor recurrence and metastasis. Such questions should be further investigated in future research.

Author Contributions

Design of the study: KCX and YYL; data collection: XFK and WQ; data analysis: FM and TYL; and manuscript writing: JBC. All authors read and approved the final version of the manuscript for publication.

Conflicts of Interest

The authors report no conflicts of interest in this work.

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Institutional Review Board Statement

This study protocol received ethical approval from the Ethics Committee of Fuda Cancer Hospital of Jinan University on December 7, 2018 (approval No. Fuda20181207) and conformed to the specifications of the World Medical Association's *Declaration of Helsinki*.

Informed Consent Statement

The authors certify that they have obtained all appropriate patient consent forms. In the forms, the patients or their legal guardians have given their consent for patients' images and other clinical information to be reported in the journal. The patients or their legal guardians

understand that their names and initials will not be published and due efforts will be made to conceal their identity.

Biostatistics Statement

The statistical methods of this study were conducted and reviewed by the epidemiologist of Fuda Cancer Hospital of Jinan University.

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Data Sharing Statement

Individual participant data that underlie the results reported in this article, after deidentification (text, tables, figures, and appendices), immediately following publication, without end date. Study protocol and informed consent form will be available immediately following publication, without end date. Results will be disseminated through presentations at scientific meetings and/or by publication in a peer-reviewed journal. Anonymized trial data will be available indefinitely at www.figshare.com.

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References

1. Manjili MH. Tumor dormancy and relapse: from a natural byproduct of evolution to a disease state. Cancer Res. 2017;77:2564-2569.
2. Pawelec G. Immunosenescence and cancer. Biogerontology. 2017;18:717-721.
3. Tarazona R, Sanchez-Correa B, Casas-Avilés I, et al. Immunosenescence: limitations of natural killer cell-based cancer immunotherapy. Cancer Immunol Immunother. 2017;66:233-245.
4. Singer K, Cheng W, Kreutz M, Ho PC, Siska PJ. Immunometabolism in cancer at a glance. Dis Model Mech. 2018;11:dmm034272.
5. Tohme S, Simmons RL, Tsung A. Surgery for cancer: a trigger for metastases. Cancer Res. 2017;77:1548-1552.
6. Chen X, Zhang L, Jiang Y, et al. Radiotherapy-induced cell death activates paracrine HMGB1-TLR2 signaling and accelerates pancreatic carcinoma metastasis. J Exp Clin Cancer Res. 2018;37:77.
7. Ramani VC, Vlodavsky I, Ng M, et al. Chemotherapy induces expression and release of heparanase leading to changes associated with an aggressive tumor phenotype. Matrix Biol. 2016;55:22-34.
8. Sekandarzad MW, van Zundert AAJ, Lirk PB, Doornebal CW, Hollmann MW. Perioperative Anesthesia Care and Tumor Progression. Anesth Analg. 2017;124:1697-1708.
9. Siegel RL, Miller KD, Jemal A. Cancer statistics, 2019. CA Cancer J Clin. 2019;69:7-34.
10. Expert Panel on Thoracic Imaging, de Groot PM, Chung JH, et al. ACR Appropriateness Criteria® noninvasive clinical staging of primary lung cancer. J Am Coll Radiol. 2019;16:S184-S195.

11. Yan X, Jiao SC, Zhang GQ, Guan Y, Wang JL. Tumor-associated immune factors are associated with recurrence and metastasis in non-small cell lung cancer. Cancer Gene Ther. 2017;24:57-63.
12. Finn OJ. Immuno-oncology: understanding the function and dysfunction of the immune system in cancer. Ann Oncol. 2012;23 Suppl 8:viii6-viii9.
13. Moore SC, Lee IM, Weiderpass E, et al. Association of leisure-time physical activity with risk of 26 types of cancer in 1.44 million adults. JAMA Intern Med. 2016;176:816-825.
14. Kelso GF, Porteous CM, Coulter CV, et al. Selective targeting of a redox-active ubiquinone to mitochondria within cells: antioxidant and antiapoptotic properties. J Biol Chem. 2001;276:4588-4596.
15. Forrer MD, Sureda A, Tauler P, Palacin C, Tur JA, Pons A. Impaired lymphocyte mitochondrial antioxidant defenses in variate porphyria are accompanied by more inducible reactive oxygen species production and DNA damage. Br J Haematol. 2010;149:759-767.
16. Kamphorst AO, Wieland A, Nasti T, et al. Rescue of exhausted CD8 T cells by PD-1-targeted therapies is CD28-dependent. Science. 2017;355:1423-1427.
17. Akagi J, Baba H. Hydrogen gas restores exhausted $CD8^+$ T cells in patients with advanced colorectal cancer to improve prognosis. Oncol Rep. 2019;41:301-311.
18. Osmani L, Askin F, Gabrielson E, Li QK. Current WHO guidelines and the critical role of immunohistochemical markers in the subclassification of non-small cell lung carcinoma (NSCLC): Moving from targeted therapy to immunotherapy. Semin Cancer Biol. 2018;52:103-109.
19. Thuluvath PJ, Thuluvath AJ, Savva Y. Karnofsky performance status before and after liver transplantation predicts graft and patient survival. J Hepatol. 2018;69:818-825.
20. Klebanoff CA, Gattinoni L, Restifo NP. $CD8^+$ T-cell memory in tumor immunology and immunotherapy. Immunol Rev. 2006;211:214-224.
21. Klebanoff CA, Gattinoni L, Torabi-Parizi P, et al. Central memory self/tumor-reactive $CD8^+$ T cells confer superior antitumor immunity compared with effector memory T cells. Proc Natl Acad Sci U S A. 2005;102:9571-9576.
22. Chen IH, Lai YL, Wu CL, et al. Immune impairment in patients with terminal cancers: influence of cancer treatments and cytomegalovirus infection. Cancer Immunol Immunother. 2010;59:323-334.
23. Saavedra D, García B, Lorenzo-Luaces P, et al. Biomarkers related to immunosenescence: relationships with therapy and survival in lung cancer patients. Cancer Immunol Immunother. 2016;65:37-45.
24. Owonikoko TK, Ragin CC, Belani CP, et al. Lung cancer in elderly patients: an analysis of the surveillance, epidemiology, and end results database. J Clin Oncol. 2007;25:5570-5577.
25. Franceschi C, Valensin S, Fagnoni F, Barbi C, Bonafè M. Biomarkers of immunosenescence within an evolutionary perspective: the challenge of heterogeneity and the role of antigenic load. Exp Gerontol. 1999;34:911-921.
26. Lanna A, Henson SM, Escors D, Akbar AN. The kinase p38 activated by the metabolic regulator AMPK and scaffold TAB1 drives the senescence of human T cells. Nat Immunol. 2014;15:965-972.
27. Henson SM, Lanna A, Riddell NE, et al. p38 signaling inhibits mTORC1-independent autophagy in senescent human $CD8^+$ T cells. J Clin Invest. 2014;124:4004-4016.
28. Akbar AN, Henson SM. Are senescence and exhaustion intertwined or unrelated processes that compromise immunity? Nat Rev Immunol. 2011;11:289-295.
29. Garg AD, Agostinis P. Cell death and immunity in cancer: From danger signals to mimicry of pathogen defense responses. Immunol Rev. 2017;280:126-148.
30. Falop T, Kotb R, Fortin CF, Pawelec G, de Angelis F, Larbi A. Potential role of immunosenescence in cancer development. Ann N Y Acad Sci. 2010;1197:158-165.
31. Yang Y, Li B, Liu C, et al. Hydrogen-rich saline protects immunocytes from radiation-induced apoptosis. Med Sci Monit. 2012;18:Br144-148.
32. Yang Y, Gao F, Zhang H, et al. Molecular hydrogen protects human lymphocyte AHH-1 cells against $^{12}C^{6+}$ heavy ion radiation. Int J Radiat Biol. 2013;89:1003-1008.

33. De La Cruz LM, Nocera NF, Czerniecki BJ. Restoring anti-oncodriver Th1 responses with dendritic cell vaccines in HER2/neu-positive breast cancer: progress and potential. Immunotherapy. 2016;8:1219-1232.
34. Sun C, Sun HY, Xiao WH, Zhang C, Tian ZG. Natural killer cell dysfunction in hepatocellular carcinoma and NK cell-based immunotherapy. Acta Pharmacol Sin. 2015;36:1191-1199.
35. Taniguchi M, Harada M, Dashtsoodol N, Kojo S. Discovery of NKT cells and development of NKT cell-targeted anti-tumor immunotherapy. Proc Jpn Acad Ser B Phys Biol Sci. 2015;91:292-304.
36. Mao TL, Miao CH, Liao YJ, Chen YJ, Yeh CY, Liu CL. Ex Vivo Expanded Human Vγ9Vδ2 T-Cells Can Suppress Epithelial Ovarian Cancer Cell Growth. Int J Mol Sci. 2019;20:1139.
37. Davey MS, Willcox CR, Joyce SP, et al. Clonal selection in the human Vδ1 T cell repertoire indicates γδ TCR-dependent adaptive immune surveillance. Nat Commun. 2017;8:14760.
38. Fisher JPH, Yan M, Heuijerjans J, et al. Neuroblastoma killing properties of Vδ2 and Vδ2-negative γδ T cells following expansion by artificial antigen-presenting cells. Clin Cancer Res. 2014;20:5720-5732.
39. Ohue Y, Nishikawa H. Regulatory T (Treg) cells in cancer: Can Treg cells be a new therapeutic target? Cancer Sci. 2019;110:2080-2089.

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Effect of Hydrogen Inhalation Therapy on Hearing Loss of Patients With Nasopharyngeal Carcinoma After Radiotherapy

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Abstract

Objective: To evaluate the clinical efficacy and safety of hydrogen inhalation in improving hearing loss in patients with long-term survival of nasopharyngeal carcinoma after radiotherapy.
Methods: The eustachian tube dysfunction score, pure tone air conduction threshold, bone conduction threshold, the score of tympanogram and otoscope were prospectively observed in patients with deafness after radiotherapy only or combined radiotherapy and chemotherapy for nasopharyngeal carcinoma. Paired t test and one-way analysis of variance were used to analyze the data before and after treatment.
Results: A total of 17 patients were observed. The median time from radiotherapy to now was 228 months, and the median time from the diagnose of deafness to now was 92 months. After 4 weeks of hydrogen inhalation, the score of eustachian tube dysfunction, air conduction and bone conduction hearing thresholds were significantly reduced, P values were 0.0293, 0.0027, 0.0404, respectively. The mean air-bone gap, the score of

otoendoscopy and tympanogram were also decreased, but the differences were not significant (P = 0.2079, P = 0.0536, P = 0.1056). Patients with radiotherapy alone and concurrent chemo-radiotherapy had significantly lower air conduction hearing threshold after hydrogen absorption (P = 0.0142, P = 0.0495). The air and bone hearing threshold improved 4 and 12 weeks after hydrogen inhalation. The results of air and bone hearing thresholds before hydrogen inhalation showed a descending trend. The air and bone hearing thresholds before hydrogen inhalation were 74.69 ± 27.03 dB and 45.70 ± 21.58 dB, respectively. At the 12th week, the mean values of air and bone hearing thresholds were the lowest, which were 66.88 ± 20.88 dB and 40.94 ± 18.93 dB, respectively, but there was no significant difference in the score of air and bone among all groups (P = 0.6755, P = 0.7712). After hydrogen inhalation treatment, no adverse reactions such as nosebleed, chest pain, dyspnea, nausea, vomiting, dizziness, earache and allergic reaction were observed.

Conclusion: This is the first prospective study on the effect of hydrogen on hearing improvement in patients with deafness after radiotherapy/chemotherapy for nasopharyngeal carcinoma, suggesting that continuous hydrogen inhalation may be an alternative rehabilitation therapy for these patients.

Keywords: hydrogen inhalation, hydrogen oxygen inhalation, nasopharyngeal carcinoma, radiotherapy, chemotherapy, hearing loss, hearing impairment

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Introduction

As one of the most common malignant tumors in the head and neck, nasopharyngeal carcinoma (NPC) is primarily treated with radiotherapy alone or radiotherapy combined with chemotherapy. Patients' 5-year survival rate can be 75-87.4%. Due to the fact that the entire auditory system including the middle ear, cochlea, auditory nerves, brainstem and auditory cortex are all located at or close to the irradiation area for nasopharyngeal cancer treatment, the radiation from radiotherapy inevitably implicates the aforementioned anatomical structure of the nasopharynx. Radiotherapy is usually followed by such complications as secretory otitis media, ossicular chain necrosis, and cochlea & auditory nerve damage. Hearing impairment is the most common late-stage adverse reaction. As patients with nasopharyngeal carcinoma radiotherapy survive longer, this complication becomes progressively worse and seriously compromises the quality of life. So far, there has been no special treatment, making tumor rehabilitation a major challenge.

Hydrogen (H_2) is the lightest and smallest gas in nature. Dole et al. first reported the therapeutic effect of high-pressure and high-concentration hydrogen on mouse models with squamous cell carcinoma in 1975. In 2001, Gharib et al. proved that inhalation of high-pressure hydrogen alleviates the inflammatory response caused by liver parasite infection. In 2007, Ohsawa et al. found that low-concentration hydrogen selectively neutralizes hydroxyl radicals (OH) and peroxynitrite (ONOO-) and possesses antioxidant properties. In recent years, a large number of preclinical studies and a few clinical trials have observed the

preventive and therapeutic effects of this gas on different system diseases, including the effects on inhibiting cancer cell growth, invasion and metastasis, and in reducing adverse reactions of radiotherapy/chemotherapy. In respect of the hearing system, some studies have revealed that molecular hydrogen protects hair cells of the hearing system through antioxidant effects, and ameliorates hearing damage.

This study observes the changes of hearing-related indexes before and after hydrogen inhalation by 17 patients with 5-year-long or longer hearing impairment after nasopharyngeal carcinoma treatment, with a focus on assessing the effectiveness and safety of hydrogen in improving hearing. As far as we know, this is the first report of research findings in this domain. With the hope to reduce hearing loss after radiotherapy/chemotherapy, we wish to provide a new, safe and effective treatment and rehabilitation option for patients with nasopharyngeal carcinoma.

Subjects and Methods

Subjects

From May 2019 to October 2020, a total of 34 nasopharyngeal carcinoma patients with post-radiotherapy hearing loss from the Nasopharyngeal Carcinoma Group of the Light of Life Cancer Rehabilitation Association of Guangdong Province were enrolled in this study after signing informed consent. According to the registered clinical trial (ClinicalTrials.gov, ID: NCT03818347. Registration Date: January 24, 2019), all the enrolled patients met the following conditions: (1) Diagnosed with Stage I to IVb nasopharyngeal carcinoma after pathological examinations; (2) Received conventional radiotherapy or radiotherapy combined with conventional chemotherapy (chemoradiotherapy) 5 years ago; (3) Hearing and hearing test results before and after treatment were normal; (4) Experienced hearing loss at the age of 60 or lower, which excluded the probability of presbycusis; (5) Having inhaled hydrogen for at least 4 weeks. Patients with the following conditions were excluded from this study: (1) Severe hypertension, diabetic autoimmune disease, history of head trauma, and experience of long-time working in noisy environment; (2) Recurrence or metastasis of existing tumor(s); (3) Middle ear disease or hearing impairment developed before anti-tumor treatment; (4) Head and neck radiotherapy due to other reasons; (5) Middle ear effusion as evidenced by otoscopy.

The three patients who did not keep inhaling hydrogen daily and 14 patients who failed to complete regular relevant examinations were excluded from analysis. A total of 17 patients (34 ears) completed the study eventually.

The 17 patients, including seven males and 10 females, aged from 47 to 67 years old, with a median of 58 years old. According to AJCC (American Joint Committee on Cancer) tumor staging (7th edition), 10 cases were in Stage I, 4 cases in Stage II, 1 case in Stage III, and 2 cases in Stage IV. Eleven of the cases had received radical radiotherapy alone; and 6 cases radiotherapy combined with chemotherapy. During radiotherapy, the radiation dose of 68-72 Gy was applied to nasopharyngeal carcinoma and draining lymph node area; 70.0 Gy to positive cervical lymph nodes; and 50.0-60.0 Gy to negative cervical lymph nodes. The patients were exposed to radiation 34 to 38 times in total once a day each time. Concurrent chemotherapy regimen 40 mg/m2 cisplatin (DDP) given through intravenous drips once a week. It has been 100 to 355 months (a median of 259 months) since the aforementioned treatment was completed. Hearing loss occurred 24 to 289 months after the completion of radiotherapy or chemoradiotherapy. It persisted for 30-186 months, with a median of 90 months (Table 1). The patients received hearing loss treatments including: glucocorticoid

treatment in 12 cases, acupuncture in seven cases, Chinese medicine treatment in 16 cases, and hyperbaric oxygen treatment in five cases. None of the patients showed improvement from the treatments.

Table 1. Basic information of the 17 patients with hearing loss after nasopharyngeal carcinoma radiotherapy/chemoradiotherapy.

Case no.	Age (Y)	Gender	Staging at time of treatment	Tumor treatment	End of treatment till now (month)	End of treatment to discovery of hearing loss (month)	End of treatment to receiving hydrogen treatment (month)	Hydrogen inhalation duration (week)

Hydrogen Inhalation Method

The subjects inhaled hydrogen-oxygen mixture, which contained 2.0 L/min hydrogen and 1.0 L/min oxygen (produced by hydrogen-oxygen nebulizer AMS-H-03, Shanghai Asclepius Meditec Co., Ltd.), through nasal tubes in quite conditions. The inhalation lasted 3-6 h for 4-12 weeks, with eight cases for 12 weeks, six cases for 8 weeks and three cases for 4 weeks. The test period was divided into three periods for assessment: before hydrogen treatment, after 4 weeks of hydrogen treatment, and after 12 weeks of hydrogen treatment.

Eustachian Tube Dysfunction Questionnaire-7 (ETDQ-7) Survey

Eustachian tube dysfunction questionnaire-7 (ETDQ-7) survey was adopted. The scale included seven items, representing seven levels (from 1 to 7 scores) according to the severity of symptoms. The higher the score, the more severe the symptoms. Patients were given face-to-face questionnaire surveys 1-3 days before hydrogen inhalation and 1-3 days after 4 weeks of hydrogen treatment.

Tympanic Membrane Detection

Scoring was performed according to otoscope-based examination: 1 score: tympanic membrane turbidity and thickening; 2 scores: tympanic membrane congestion and indentation as well as disappearance of light cone; 3 scores: tympanic membrane effusion; and 4 scores: tympanic membrane perforation.

Tympanogram Changes

Acoustic impedance tester was used to assess the changes in calm breathing tympanogram. Tympanogram scoring rules were: 3 scores for type Ad; 2 scores for type A or As; 1 score for type C; and 0 score for type B.

Pure-Tone Hearing Threshold Testing

Pure-tone hearing threshold testing (with AT235 from a Danish international hearing health care company) was carried out in a standard sound-proof chamber. With acoustic standard set at GB/T16296 and reverberation time at 0.3 ± 0.15s, pure-tone hearing thresholds in conditions of 0.5, 1, 2, 4, and 8 kHz were tested in sequence. The average air conduction threshold, bone-conduction threshold, and air-bone gap under 0.5-4 kHz were calculated. The average pure tone hearing threshold (PTA) of four frequencies divides hearing loss into the following levels: normal hearing is defined as the mean of pure tone hearing threshold ≤ 25 dB, 26-40 dB was mild hearing loss, 41-55 dB HL was moderate hearing loss, 56-70 dB HL was moderately severe hearing loss, 71-90 dB HL was severe hearing loss, and >91 dB HL

was very severe hearing loss. Pure tone hearing threshold test by hearing detection professional testing.

Safety Assessment

According to Common Terminology Criteria for Adverse Events (CTCAE 4.0 version) defined by the National Cancer Institute of U.S. Department of Health and Human Services, symptoms or side effects are graded from Grade 1-5. Safety evaluation was conducted once every 4 weeks after hydrogen inhalation, covering symptoms of nose bleeding, cough, chest pain, dyspnea, nausea, vomiting, dizziness, earache and allergic reaction.

Statistical Analysis

Measurement data were expressed as x ± s. The data before and after hydrogen inhalation were compared by paired t test. Different hydrogen uptake durations were compared using one-way analysis of variance. P≤0.05 indicated that the difference is statistically significant. All analyses and figures were produced using GraphPad Prism 5.0 (GraphPad software, San Diego, CA, USA).

Results

Eustachian Tube Function

A total of 17 cases received the survey using 7-item questionnaires about eustachian tube dysfunction. 34 questionnaires were returned back, with 100% effective response rate. The score before hydrogen inhalation was 3.25 ± 2.00, and that after 4 weeks of hydrogen inhalation was 2.59 ± 1.65. The difference between the two groups was statistically significant (P < 0.05) (Table 2).

Scoring of Tympanic Membrane Under Otoscope and Tympanogram

Tympanic membrane perforation occurred in five ears before hydrogen inhalation and persisted after treatment. After 4 weeks of hydrogen inhalation, the tympanic membrane score based on otoscopy stabilized in 10 cases, improved in 6 cases, and worsened in one case. However, the difference before hydrogen inhalation was not statistically significant (P > 0.05). Tympanogram scores before and after hydrogen inhalation showed no statistically significant difference (P > 0.05) (Table 2).

> *Table 2. Eustachian tube dysfunction 7-item score, otoscope score and tympanogram score.*

	Number of ears	Before hydrogen-oxygen inhalation ($\bar{x}$ ± s)	4 weeks after hydrogen-oxygen inhalation ($\bar{x}$ ± s)	P value
Eustachian tube dysfunction 7-item score	34	3.25 ± 2.00	2.59 ± 1.65	0.0293*
Otoscope score	34	1.76 ± 1.26	1.44 ± 1.24	0.0536
Tympanogram score	34	0.66 ± 0.98	0.97 ± 1.06	0.1056

Note: *P < 0.05.

Hearing Changes

Hearing Changes After 4 Weeks of Hydrogen Inhalation

The average air conduction, bone conduction and air-bone gap before hydrogen inhalation in 17 patients (34 ears) were 77.46 ± 28.12, 46.76 ± 20.73, and 30.70 ± 11.12 dB, respectively. After 4 weeks of hydrogen inhalation treatment, both air conduction threshold and bone conduction threshold were significantly lower than those before hydrogen inhalation, being 73.35 ± 28.20 dB (P = 0.0027) and 44.56 ± 19.50 dB (P = 0.0404), respectively (Figures 1A,B). Among them, the threshold of air conductivity was significantly improved, with a decrease in 64.71% of the patients (Table 3). The mean air bone gap (ABG) decreased from that before treatment (28.78 ± 13.44 dB), but the difference was not statistically significant (P = 0.2079) (Figure 1C).

> *Figure 1. Pure tone hearing threshold tests result before and after hydrogen-oxygen mixed gas inhalation on hearing loss patients with long-term survival after combined modality treatment of nasopharyngeal carcinoma. (A) Test results of the air conduction hearing threshold. (B) Test results of the bone conduction hearing threshold. (C) Test results of the air bone gap (ABG) hearing threshold. The parallel red long lines in the figure represent the normal range, the black short lines represent the average value at each time point. Data are analyzed by paired t test.* \P < 0.05, \\P < 0.01.*

> *Table 3. Air conductance threshold improvement before and 4 weeks after hydrogen and oxygen inhalation therapy.*

Air-conduction hearing threshold improvement decibel (dB)	Number of ears	Percentage (%)
≤0	12	35.29
0-10	18	52.94
11-20	3	8.82
21-30	1	2.94

Hearing Improvement in Patients With Different Initial Treatment Regimens for Tumors

According to the initial methods of tumor treatment, 17 patients (34 ears) were separated to radiotherapy group (22 ears) and concurrent chemoradiotherapy group (12 ears). The average air-conduction threshold, bone-conduction threshold and air-bone gap of the radiotherapy group before hydrogen inhalation were 76.70 ± 28.26, 47.05 ± 22.30, and 29.66 ± 9.86 dB, respectively. After 4 weeks of hydrogen inhalation, the air-conduction threshold showed decline with significant difference (73.98 ± 30.29 dB, P = 0.0142, Figure 2A); and bone-conduction and air-bone gap hearing thresholds showed decrease without significant difference (P = 0.6137 and P = 0.1699, respectively, Figures 2C,E). Before hydrogen inhalation, the average air conduction threshold, bone-conduction threshold and air-bone gap of patients with concurrent chemoradiotherapy were 78.85 ± 28.55, 47.05 ± 17.72, and 31.81 ± 14.63 dB, respectively. After hydrogen inhalation, the thresholds of air conduction and bone conduction were significantly reduced, being 72.19 ± 25.13 dB and 39.04 ± 12.61 dB, respectively (Figures 2B,D). P values were 0.0495 and 0.0134, respectively. The average air-bone gap between the two groups was increased (33.15 ± 18.48 dB) compared to before treatment, but the difference was not statistically significant (P = 0.7283, Figure 2F).

Hearing Threshold Changes in Patients With Different Hydrogen Inhalation Durations

Eight of the deaf patients (16 ears) were treated with hydrogen inhalation for 12 weeks. The results of air conduction threshold and bone conduction threshold were compared before, 4 weeks and 12 weeks after hydrogen inhalation, showing a downward trend. Before hydrogen

inhalation, air and bone hearing thresholds were 74.69 ± 27.03 dB and 45.70 ± 21.58 dB, respectively. At 12 weeks of treatment, the mean values of air conduction threshold and bone conduction threshold were the lowest, which were 66.88 ± 20.88 dB and 40.94 ± 18.93 dB, respectively. However, there was no significant difference in air and bone hearing threshold among all groups (P = 0.6755, P = 0.7712, Figures 3A,B).

Patients' Hearing Changes After Suspension of Hydrogen-Oxygen Inhalation

Five patients (10 ears) were given hearing checkups 6-9 months after they stopped hydrogen-oxygen gas inhalation. According to the checkup results, the bone- and air-conduction hearing thresholds of three ears were stable or showed continuous improvement; and the hearing thresholds of seven ears increased in decibels. Average decibels of both air- and bone-conduction hearing thresholds for the affected ears were higher than before, but no significant difference was observed with P value of 0.0596 and 0.3473 respectively (see Figures 4A,B).

> *Figure 2. Pure tone hearing threshold tests result before and after hydrogen-oxygen mixed gas inhalation on hearing loss patients with long-term survival after radiotherapy only and concurrent chemoradiotherapy of nasopharyngeal carcinoma. (A) Test results of the air conduction hearing threshold in RT patients. (B) Test results of the air conduction hearing threshold in CCRT patients. (C) Test results of the bone conduction hearing threshold in RT patients. (D) Test results of the bone conduction hearing threshold in CCRT patients. (E) Test results of the air bone gap (ABG) hearing threshold in RT patients. (F) Test results of the air bone gap (ABG) hearing threshold in CCRT patients. The parallel red long lines in the figure represent the normal range. Data are analyzed by paired t test.* \P < 0.05.*

> *Figure 3. Patients' hearing changes at different time points of hydrogen-oxygen inhalation. (A) Diagram of changes of air conduction hearing threshold at different time point after hydrogen-oxygen gas inhalation. (B) Diagram of changes of bone conduction hearing threshold at different time point after hydrogen-oxygen gas inhalation. One-way analysis of variance was used for data analysis.*

Adverse Reactions Related to Hydrogen-Oxygen Inhalation

No such common adverse reaction as nose bleeding, chest pain, dyspnea, nausea, vomiting, dizziness, earache or skin allergy was observed after hydrogen-oxygen inhalation. Ten patients (58.8%) complained of mild cough before hydrogen inhalation, and 11 patients (64.7%) complained of cough 4 weeks after hydrogen inhalation. Earache occurred before hydrogen inhalation in one of the cases and no change of earache after the treatment. As time passed, the aforementioned symptoms did not worsen (Table 4).

Discussion

Hearing Impairment After Radiotherapy

Nasopharyngeal carcinoma (NPC) is the most common malignant tumor of the head and neck, with more than half of the world's cases found in China. And such cases are mostly seen in southern and southeastern coastal areas of China. Currently, radiotherapy is the most effective treatment for NPC. As radiotherapy technology advances, the efficacy has improved gradually. Early-stage NPC patients' 5-year survival rate may reach as high as 95%. As the most common adverse reaction after radiotherapy, hearing impairment takes place in the following three circumstances: (1) Hearing impairment occurs directly after radiotherapy and is usually temporary. Patients recover from it after several weeks or months. (2) Sudden deafness or sudden hearing loss occurs without any apparent cause at a certain time point after the end of radiotherapy. (3) Hearing loss occurs gradually after radiotherapy or

chemoradiotherapy after a "latency period" of varying lengths. This is the case with the group of patients included in this study. The median interval (shortest at 100 months) from the completion of radiotherapy or chemoradiotherapy till now is 259 months. Hearing loss occurred at the median time of 117 months (ranging from 24 months to 285 month) after the completion of radiotherapy or chemoradiotherapy. It persisted for a median time of 90 months (shortest at 30 months). In these cases, there is neither evidence of the existence or recurrence of any primary tumor, nor other factor that could have caused hearing impairment.

Hearing loss is a major late complication for long-term surviving patients after radiotherapy or radiotherapy combined with chemotherapy. As a patient survives longer, the incidence of hearing loss increases, and hearing loss becomes more serious. The hearing loss rate of patients who survive more than 5 years can reach 60.5-94%. Due to the lack of targeted treatments, existing treatments are usually based on symptomatic treatments, with limited effects. It has been reported that hearing improved after sudden deafness was treated with Intratympanic steroids. All the patients in this study group received a variety of treatments, including oral steroids or ear drops, acupuncture and hyperbaric oxygen. But the treatments failed to relieve hearing damage or prevent the aggravation.

Observation on Hydrogen-Based Treatment of Hearing Loss

In 2020, we reported about three cases of NPC patients with hearing loss after radiotherapy. The patients had significant improvement in hearing after hydrogen inhalation. In a real-world evidence survey of cancer patients who took hydrogen inhalation voluntarily as a means of rehabilitation, a 63-year-old male patient with nasopharyngeal carcinoma radiotherapy 19 years ago was observed. After radiotherapy, the patient experienced hearing loss, which showed no substantial improvement in spite of symptomatic treatments, and had to use hearing aid for smooth communication. On January 17, 2019, he started to inhale hydrogen on a daily basis. After March 2019, his hearing improved tremendously, ridding him of the hearing aid. The patient's experience led us to the unexpected discovery that hydrogen might be effective in treating hearing loss. Therefore, with a request and support from the Nasopharyngeal Cancer Group of Light of Life Cancer Rehabilitation Association in the patient's place of living, we conducted this prospective study. The 17 patients in this study had clear history of nasopharyngeal carcinoma and radiotherapy or chemoradiotherapy. In quiet environment, the patients inhaled hydrogen (mixture of hydrogen and oxygen) through nasal tubes at least 3 h a day for more than 4 weeks. Results indicated that the hearing was improved in most cases. After 4 weeks of hydrogen inhalation, the patients' eustachian tube function scale scores were significantly lower than before treatment; and both their air- and bone-conduction hearing thresholds of the affected ears declined notably. As the treatment continued, the patients' hearing decibels of the affected ears decreased more than before. After hydrogen inhalation was stopped, hearing in some of the cases continued to improve. For subgroups with different initial treatments, the air-conduction hearing thresholds of ears affected by radiotherapy alone and air- and bone-conduction hearing thresholds of ears affected by concurrent radiotherapy and chemotherapy were significantly improved.

It is worth noting that the hydrogen inhalation is very safe. Few patients in the study experienced hydrogen-specific adverse reactions.

Hydrogen inhalation by the 17 patients in this study group was carried out under strict medical supervision and was not given any other medical intervention during the entire period.

Therefore, it can be concluded that the hearing improvement found in these patients is undoubtedly attributable to hydrogen inhalation.

> *Figure 4. Hearing changes in patients after hydrogen and oxygen inhalation therapy was discontinued. (A) Diagram of changes in air conduction hearing threshold after stopping hydrogen-oxygen inhalation therapy for 24 weeks. (B) Figure of changes in bone conduction hearing threshold after stopping hydrogen-oxygen inhalation therapy for 24 weeks. Paired t test was used for data analysis.*

> *Table 4. Adverse reactions at different time points after hydrogen inhalation.*

Adverse reaction	Before hydrogen inhalation (17 patients)	4 weeks after hydrogen inhalation (17 patients)	8 weeks after hydrogen inhalation (8 patients)	12 weeks after hydrogen inhalation (8 patients)
Nose bleeding	—	—	—	—
Cough	10 (58.80%)	11 (64.70%)	7 (87.5%)	1 (12.5%)
Chest pain	—	—	—	—
Dyspnea	—	—	—	—
Nausea	—	—	—	—
Vomiting	—	—	—	—
Dizziness	—	—	—	—
Earache	1 (5.88%)	—	—	—
Allergic reaction	—	—	—	—

Mechanism of Hydrogen in Improving Hearing Loss

The mechanism by which hydrogen improves hearing loss in patients with nasopharyngeal carcinoma after radiotherapy or chemoradiotherapy remains unclear. However, if we connect the biological effect of radiation (or chemotherapy drugs) with the mechanism of action of hydrogen, it is not difficult to note that hydrogen represents a "tit-to-tat" "pathogenesis treatment" for post-radiation hearing loss.

First of all, hydrogen has anti-oxidant and anti-inflammatory effects that resist oxidative stress and inflammation caused by radiotherapy. In the process of radiotherapy, reactive oxygen species (ROS), especially hydroxyl radicals (OH-) and peroxy-nitrosamine, are generated in large volume. Excessive levels of ROS destroy the composition of mitochondrial electron transport chains; trigger imbalance of intracellular redox systems; and cause lipid peroxidation, protein misfolding and DNA strand breakage. In the meantime, they activate the JNK signaling system; up-regulate pro-oxidant genes; and inhibit antioxidants related to nuclear factor-e2 related factor (Nrf2), thereby causing oxidative stress and meanwhile inducing matrix metalloproteinases (MMP); triggering secretion of inflammatory chemokines including tumor necrosis factors (TNFs), interleukin (IL)-1, IL-6 and IL-8; and precipitating inflammation. Peroxidation and inflammation promote cell apoptosis. Subsequently, pro-fibrotic cytokines such as platelet-derived growth factors (PDGFs), insulin-like growth factors (IGFs) and basic fibroblast growth factors (FGFs) are released, promoting differentiation of monocytes into M2 macrophages; strengthening fibroblasts to proliferate and differentiate into myofibroblasts; and then amplifying inflammation and fibrosis formation. Radiation can also induce the expression of several miRNAs including miRNA-1 and miRNA-21. And the up-regulation of miRNA-21 expression is related to fibrosis.

The dose of radical radiotherapy for nasopharyngeal carcinoma is far higher than the normal tissue tolerance dose. Therefore, oxidative stress, inflammation and fibrosis that accompany

radiotherapy inevitably damage the auditory system in the radiation field, leading to middle ear, inner ear and auditory nerve cell damages. Eventually, conductive, sensorineural or mixed hearing loss occurs. Although intensity-modulated radiotherapy helps reduce the dose to surrounding normal tissues, symptoms are still unavoidable for nasopharyngeal cancer survivors. The hearing loss of patients receiving radiotherapy combined with chemotherapy is usually more severe than that of patients with radiotherapy alone. This might be attributed to the fact that cisplatin, which is commonly used in chemotherapy, has a strong effect on inducing ROS generation.

Hydrogen molecule is a weak reducing agent with very low molecular weight. It can quickly diffuse and pass through cell membranes and lipid bilayers; reach hair cell nuclei and mitochondria where abundant invasive ROS gather; and selectively neutralize highly reactive toxic ROS (such as •OH) directly. Further studies have revealed that hydrogen regulates the Nrf2 pathway. Nrf2 is considered to be an important regulator of electrophilic/antioxidant homeostasis, and is especially capable of maintaining the functional integrity of cells under oxidative stress conditions. Hydrogen helps activate the Nrf2-Keap1 system; induce activation of antioxidant response elements (AREs); and promote expression of antioxidant cytoprotective proteins such as glutathione, catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase and heme-1 oxygenase; activate transcription factor FoxO1; reduce damage of OH to mitochondria; and inhibit overproduction of ROS. Moreover, hydrogen inhibits infiltration of phagocytes to sites of inflammation and subsequent release of reactive substances; and down-regulates various pro-inflammatory and inflammatory cytokines including interleukin (IL)-1β, IL-6, TNF-α and intracellular adhesion molecules (ICAM)-1, thereby achieving anti-inflammatory effect. Hydrogen also weakens abnormal expression of miRNA induced by radiation and reduces fibrotic damage.

According to the findings of existing studies, hydrogen treatment helps significantly mitigate hearing loss caused by cisplatin; reduce Organ of Corti hair cell damage from cisplatin; improve levels of malondialdehyde (MDA) and isoprostane F2α (8-iso-PGF2α) in serum and cochlea tissues; significantly increase the number of residual acoustic hair cells in the cochlea; and reduce formation of hydroxyl radicals in the cochlea. These facts support the aforementioned mechanism.

Second, hydrogen has cytoprotective activity that improves cell apoptosis induced by radiotherapy. Apoptosis plays an important role in progression of radiation injury. Hydrogen significantly inhibits ectopic expression of death promoter Bcl-2 related X protein (bax) and expression of caspase-3, and meanwhile promotes expression of the anti-apoptotic protein Bcl-2, thereby achieving cytoprotective activity.

Third, hydrogen improves blood perfusion and alleviates vascular damage as a result of radiotherapy. Vascular injury and endothelial dysfunction play key roles in development of radiation injury. Within a few minutes of exposure to ionization radiation (IR) and as ROS is generated in excessive amount, vascular protectant nitric oxide (NO) is eliminated, triggering nitrosylation of protein tyrosine residues and lipid peroxidation. Ultimately, vasomotor response is weakened, and vascular stenosis appears. After radiation, NADPH oxidases (NOXs), especially NOX2 and NOX4 that are abundantly expressed in vascular endothelial cells, are up-regulated. This promotes excessive production of ROS; changes calcium homeostasis and calcium metabolism disorders; and triggers antifibrinolysis-coagulation cascade action, leading to blood clotting and vascular occlusion. Radiation stimulates migration of monocytes to inner membranes, induces expression of inflammatory adhesion

molecules, enhances adhesive capacity of monocytes, absorbs monocyte chemotactic protein-1 into inner membranes, recruits low-density lipoproteins, and promotes arteriosclerosis, which eventually leads to arterial stenosis and blood perfusion. Since the neck is usually included in the irradiation scope of radiotherapy for patients with nasopharyngeal cancer, carotid artery damage and cochlear vascular damage are inevitable, causing damage to outer hair cells and spiral ganglion cells of the cochlea.

Evidence suggests that hydrogen protects damaged blood vessels and improves blood perfusion, including reducing degradation of cyclic guanosine monophosphate (cGMP) through phosphodiesterase, increasing cGMP levels and promoting protein kinase activation. It can also increase intracellular calcium levels and stimulate vascular endothelial growth factors, thereby increasing production of nitric oxide. Moreover, hydrogen functions to open the potassium channel sensitive to adenosine triphosphate and activate downstream mitogen-activated protein kinase pathways, thus promoting angiogenesis. Further experimental studies indicate that hydrogen prevents arterial intimal hyperplasia and atherosclerosis by inhibiting ROS and TNF-α/NF-κB pathways; inhibits macrophage-derived foam cell apoptosis; stabilizes atherosclerotic plaques; and promotes formation of vascular collaterals using the Flk1-Notch signal stimulated by paracrine VEGFs, thereby improving local microcirculation.

Discussion on Hydrogen Therapy

There are multiple ways to introduce hydrogen into the body, including oral and intravenous injection of hydrogen water. However, since hydrogen has short half-life in the body and cannot be retained in the body's tissues for a long time, non-inhalation methods may not deliver satisfactory results. The therapeutic effects of this study group appeared at least 2 weeks after hydrogen inhalation in all the cases. From patients with long-term hydrogen inhalation, it has been observed that as hydrogen inhalation continued for longer time, the patients' air- and bone-conduction hearing thresholds decreased more significantly. Fransson AE and his team also found repeated administration of H_2 inhalation may further improve the therapeutic effect. After hydrogen inhalation was stopped, the patients' bone- and air-conduction hearing thresholds rose. Given that hearing loss in this study lasted for months or even years, it is speculated that the disease had "aged". The effect of hydrogen depends on sufficient dose and time. It seems that only inhalation of hydrogen can meet these requirements.

The concentration level and flow rate of inhaled hydrogen are also important. The hydrogen inhaled by this study group has a raw rate of 3L/min and concentration level at 67%. These are the maximum levels that can be provided by the only hydrogen inhalation equipment approved by Chinese authority.

So far, there has been no evidence as to whether inhalation of pure hydrogen is better or inhalation of hydrogen-oxygen mixture is. If high-concentration pure hydrogen is to be inhaled, oxygen intake is inevitably affected. Inhaling a mixture of hydrogen and oxygen helps at least prevent hypoxia. Studies have revealed that during inhalation of hydrogen-oxygen mixture, hydrogen molecules, which are extremely small and permeable, can "carry" larger molecules of oxygen to the deep areas of tissues and improve oxygen supply. The mixture inhaled by this study group contains 33% oxygen, which exceeds the oxygen level in normal air.

Conclusion

Hearing loss is a common and persisting adverse reaction for nasopharyngeal carcinoma patients with radiotherapy. The hearing loss of such patients is usually long-lasting, chronical

and gradual, thus affecting the quality of life in a serious manner. Unfortunately, there has been no special treatment available so far. Hydrogen inhalation can improve the hearing of long-term surviving patients with nasopharyngeal carcinoma after radiotherapy, and achieves more significant efficacy as the duration of treatment is extended. Its mechanism of action might be associated with hydrogen's functions in controlling oxidative stress and inflammation; protecting cells; and improving blood perfusion. This study represents the first effort to adopt hydrogen inhalation as an independent measure for improving hearing loss in the patients with long-term survival of nasopharyngeal cancer. If this study would be supported by more experiments and verification in the future, hydrogen inhalation would be proven a safe and effective means of rehabilitation for the challenge in the medical world.

Data Availability Statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Ethics Statement

The studies involving human participants were reviewed and approved by the Ethics Committee of Fuda Cancer Hospital, Jinan University, China. The patients/participants provided their written informed consent to participate in this study.

Author Contributions

KX developed the idea for and was involved in the design of this study. XK and TL reviewed available data sources and drafted the manuscript. Y-YL, ZY, and KX critically revised the manuscript. All authors read and approved the final manuscript.

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References

1. Lin C, Lin SW, Weng SF, Lin YS. Risk of developing sudden sensorineural hearing loss in patients with nasopharyngeal carcinoma: a nationwide population-based cohort study. Head Neck. 2014;36:203-208.
2. Lee AW, Sze WM, Au JS, Leung SF, Leung TW, Chua DT, et al. Treatment results for nasopharyngeal carcinoma in the modern era: the Hong Kong experience. Int J Radiat Oncol Biol Phys. 2005;61:1107-1116.
3. Langendijk JA, Leemans CR, Buter J, Berkhof J, Slotman BJ. The additional value of chemotherapy to radiotherapy in locally advanced nasopharyngeal carcinoma: a meta-analysis of the published literature. J Clin Oncol. 2004;22:4604-4612.
4. Zhang MX, Li J, Shen GP, Zou X, Xu JI, Jiang R, et al. Intensity-modulated radiotherapy prolongs the survival of patients with nasopharyngeal carcinoma compared with conventional two-dimensional radiotherapy: a 10-year experience with a large cohort and long follow-up. Eur J Cancer. 2015;51:2587-2595.
5. Lee AW, Lin JC, Ng WT. Current management of nasopharyngeal cancer. Semin Radiat Oncol. 2012;22:233-244.
6. Wei Y, Zhou Y, Zhu J, Zhang Y, Sun M, Ding X, et al. Long-term outcome of sensorineural hearing loss in nasopharyngeal carcinoma patients: comparison between treatment with radiotherapy alone and chemoradiotherapy. Cell Biochem Biophys. 2014;69:433-437.
7. Zhu W, Chen F, Li J, Wang W, Zhang H, Yang G, et al. Dosimetric parameters associated with conductive or sensorineural hearing loss 5 years after intensity-modulated radiation therapy in nasopharyngeal carcinoma. Acta Otolaryngol. 2019;139:263-268.
8. Dole M, Wilson FR, Fife WP. Hyperbaric hydrogen therapy: a possible treatment for cancer. Science. 1975;190:152-154.

9. Gharib B, Hanna S, Abdallahi OM, Lepidi H, Gardette B, De Reggi M. Anti-inflammatory properties of molecular hydrogen: investigation on parasite-induced liver inflammation. C R Acad Sci III. 2001;324:719-724.
10. Ohsawa I, Ishikawa M, Takahashi K, Watanabe M, Nishimaki K, Yamagata K, et al. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med. 2007;13:688-694.
11. Ge L, Yang M, Yang NN, Yin XX, Song WG. Molecular hydrogen: a preventive and therapeutic medical gas for various diseases. Oncotarget. 2017;8:102653-102673.
12. Wang D, Wang L, Zhang Y, Zhao Y, Chen G. Hydrogen gas inhibits lung cancer progression through targeting SMC3. Biomed Pharmacother. 2018;104:788-797.
13. Yang Y, Liu PY, Bao W, Chen SJ, Wu FS, Zhu PY. Hydrogen inhibits endometrial cancer growth via a ROS/NLRP3/caspase-1/GSDMD-mediated pyroptotic pathway. BMC Cancer. 2020;20:28.
14. Li S, Liao R, Sheng X, Luo X, Zhang X, Wen X, et al. Hydrogen gas in cancer treatment. Front Oncol. 2019;9:696.
15. Kikkawa YS, Nakagawa T, Horie RT, Ito J. Hydrogen protects auditory hair cells from free radicals. Neuroreport. 2009;20:689-694.
16. Lin Y, Kashio A, Sakamoto T, Suzukawa K, Kakigi A, Yamasoba T. Hydrogen in drinking water attenuates noise-induced hearing loss in guinea pigs. Neurosci Lett. 2011;487:12-16.
17. Zhou Y, Zheng H, Ruan F, Chen X, Zheng G, Kang M, et al. Hydrogen-rich saline alleviates experimental noise-induced hearing loss in guinea pigs. Neuroscience. 2012;209:47-53.
18. Chen L, Yu N, Lu Y, Wu L, Chen D, Guo W, et al. Hydrogen-saturated saline protects intensive narrow band noise-induced hearing loss in guinea pigs through an antioxidant effect. PLoS ONE. 2014;9:e100774.
19. McCool ED, Anand VK, Christos PJ. Validating the clinical assessment of eustachian tube dysfunction: the Eustachian tube dysfunction questionnaire (ETDQ-7). Laryngoscope. 2012;122:1137-1141.
20. Tarakchi M, Poe DS, Noguera JF. Eustachian tube redefined. Otolaryngol Clin North Am. 2016;49:XVII-XX.
21. US Department of Health and Human Services, National Institutes of Health (NIH) and National Cancer Institute (NCI). Common terminology criteria for adverse events (CTCAE) version 4.0 [EB/OL]. Bethesda: NIH Publication.
22. Au KH, Ngan RKC, Ng AWY, Poon DMC, Ng WT, Yuen KT, et al. Treatment outcomes of nasopharyngeal carcinoma in modern era after intensity-modulated radiotherapy (IMRT) in Hong Kong: a report of 3328 patients (HKNPCSG 1301 study). Oral Oncol. 2018;77:16-21.
23. Low WK, Toh ST, Wee J, Fook-Chong SM, Wang DY. Sensorineural hearing loss after radiotherapy and chemoradiotherapy: a single, blinded, randomized study. J Clin Oncol. 2006;24:1904-1909.
24. Young YH, Lou PJ. Post-irradiation sudden deafness. J Laryngol Otol. 1999;113:815-817.
25. Yilmaz YP, Aytas FI, Akdogan O, Sari K, Savas ZG, Titiz A, et al. Sensorineural hearing loss after radiotherapy for head and neck tumors: a prospective study of the effect of radiation. Otol Neurotol. 2008;29:461-463.
26. Zheng Y, Han F, Xiao W, Xiang Y, Lu L, Deng X, et al. Analysis of late toxicity in nasopharyngeal carcinoma patients treated with intensity-modulated radiation therapy. Radiat Oncol. 2015;10:17.
27. McDowell LJ, Rock K, Xu W, Chan B, Waldron J, Lu L, et al. Long-term late toxicity, quality of life, and emotional distress in patients with nasopharyngeal carcinoma treated with intensity-modulated radiation therapy. Int J Radiat Oncol Biol Phys. 2018;102:340-352.
28. Huang TL, Chien CY, Tsai WL, Liao KC, Chou SY, Lin HC, et al. Long-term late toxicities and quality of life for survivors of nasopharyngeal carcinoma treated with intensity-modulated radiotherapy versus non-intensity-modulated radiotherapy. Head Neck. 2016;38:E1026-E1032.
29. Lan XH, Tang AZ, Zhou Y. The audiologic study of the long-term survivors with nasopharyngeal carcinoma after radiotherapy. J Speech-Lang Pathol Audiol. 2012;20:336-338.

30. Michels TC, Duffy MT, Rogers DJ. Hearing Loss in Adults: Differential Diagnosis and Treatment. Am Fam Physician. 2019;100:98-108.
31. Peng Y, Xiong S, Cheng Y, Qi YF, Yang Y. Clinical investigation of different routes of administration of dexamethasone on sudden deafness. Lin Chuang Er Bi Yan Hou Tou Jing Wai Ke Za Zhi. 2008;22:442-445.
32. Chen L, Kong X, Ma F, Lu T, Du D, Xu K. Hydrogen-oxygen therapy can alleviate radiotherapy-induced hearing loss in patients with nasopharyngeal cancer. Ann Palliat Med. 2019;8:746-751.
33. Rani V, Banati RB. Effects of ionizing radiation on mitochondria. Free Radic Biol Med. 2013;65:607-619.
34. Faraonio R, Vergara P, Di Marzo D, Pierantoni MG, Napolitano M, Russo T, et al. p53 suppresses the Nrf2-dependent transcription of antioxidant response genes. J Biol Chem. 2006;281:39776-39784.
35. Hua MM, Young TH, Lu YC, Liu HL. Eustachian tube function of patients with nasopharyngeal carcinoma. Ann Otol Rhinol Laryngol. 1995;104:453-457.
36. Low WK, Toh ST, Wang DY, et al. Effect of Radiotherapy on Retro-Cochlear Auditory Pathways. Laryngoscope. 2005;115:1823-1826.
37. Wang SZ, Chen ZC, McManar JP, Wang SY, Cheng QF. The relationship between the protective effect of amifostine and decreased intracellular adhesion molecule expression. Am J Otolaryngol. 2005;26:118-122.
38. Xiao J, Luo ZQ, Shi JD, Huang YT, Tan YF. Short-term results of biological change of nasopharyngeal carcinoma patient after radiotherapy and chemotherapy. Zhonghua Er Bi Yan Hou Tou Jing Wai Ke Za Zhi. 2008;43:906-909.
39. Lee N, Xia P, Quivey JM, Sultanem K, Poon I, Akazawa C, et al. Intensity-modulated radiotherapy in the treatment of nasopharyngeal carcinoma: an update of the UCSF experience. Int J Radiat Oncol Biol Phys. 2002;53:12-22.
40. Hwang CH, Chen TH, Young YH, Lin WS. Comparison of otologic complications between intensity-modulated and two-dimensional radiotherapies in treatment of nasopharyngeal carcinoma patients. Otolaryngol Head Neck Surg. 2010;143:662-668.
41. Young YH, Ko JY, Sheen TS. Postirradiation dizziness in nasopharyngeal carcinoma survivors. Otol Neurotol. 2004;25:366-370.
42. Chen YH, Young YH. Inner ear deficits in irradiated nasopharyngeal carcinoma survivors. Laryngoscope. 2015;125:2565-2571.
43. Hwang CH, Chen TH, Liang KL, Young YH, Liu WS. Postirradiation otitis media with effusion in nasopharyngeal carcinoma patients treated by intensity-modulated radiotherapy. Laryngoscope. 2013;123:2148-2153.
44. LeBaron TW, Kura B, Kalocayova B, Tribulova N, Slezak J. A new approach for the prevention and treatment of cardiovascular disorders: molecular hydrogen significantly reduces the effects of oxidative stress. Molecules. 2019;24:2076.
45. Bertoncello KT, ... Inhaled molecular hydrogen attenuates intense acute exercise-induced hippocampal inflammation in sedentary rats. (authors and journal not fully legible)
46. Nogueira JE, de Deus JL, Amorim MR, Batalhão ME, Leão RM, Carnio EC, et al. Inhaled molecular hydrogen attenuates intense acute exercise-induced hippocampal inflammation in sedentary rats. Neurosci Lett. 2018;715:134577.
47. Nogueira JE, Passaglia P, Mota CMD, Santos BM, Batalhão ME, Carnio EC, et al. Molecular hydrogen reduces acute exercise-induced inflammatory and oxidative stress status. Free Radic Biol Med. 2018;129:186-193.
48. Liu H, Wang L, Mi W, Xie X, Qiu J. Inhalation of hydrogen gas attenuates cisplatin-induced ototoxicity via reducing oxidative stress. Int J Pediatr Otorhinolaryngol. 2012;76:111-115.
49. Kikkawa YS, Nakagawa T, Taniguchi M, Ito J. Hydrogen protects auditory hair cells from cisplatin-induced free radicals. Neurosci Lett. 2014;579:125-129.
50. Zhao YS, An JR, Yang S, Guan P, Yu FY, Li W, et al. Hydrogen and oxygen mixture to improve cardiac dysfunction and myocardial pathological changes induced by intermittent hypoxia in rats. Oxid Med Cell Longev. 2019;2019:7415212.

51. Mei XY, Li XM, She CS, Liu XQ, Xiao CG, Wang SH, et al. Hydrogen-rich saline protects rats from oxygen glucose deprivation and reperfusion-induced apoptosis through VDAC1 via Bcl-2. Brain Res. 2019;1706:110-118.
52. Wang B, Wang H, Zhang M, Ji R, Wei J, Xin Y, et al. Radiation-induced myocardial fibrosis: mechanisms underlying its pathogenesis and therapeutic strategies. J Cell Mol Med. 2020;24:7717-7729.
53. Venkatesulu BP, Mahadevan LS, Aliru ML, Yang X, Bodd MH, Singh PK, et al. Radiation-induced endothelial vascular injury: a review of possible mechanisms. JACC Basic Transl Sci. 2018;3:563-572.
54. Wang Z, Yang B, Chen X, Zhou Q, Li H, Chen S, et al. Nrf2 regulates the ROS/ADMA/DDAH/eNOS/NO pathway and alleviates radiation-induced injury caused by iron overload. Biol Trace Elem Res. 2020.
55. Zhang Y, Murugesan P, Huang K, Cai H. NADPH oxidases and oxidative stress in cardiovascular diseases: novel therapeutic targets. Nat Rev Cardiol. 2020;17:170-194.
56. Ping Z, Peng Y, Lang H, Xinyong C, Zhiyi X, Xiaocheng W, et al. Oxidative stress in radiation-induced cardiotoxicity. Oxid Med Cell Longev. 2020;2020:3579143.
57. Farhood B, Goradel NH, Mortezaee K, Khanlarkhani N, Salehi E, Nashtaei MS, et al. Intercellular communications-redox interactions in radiation therapy; potential targets for radiation mitigation. J Cell Commun Signal. 2019;13:3-16.
58. Stewart FA, Hoving S, Russell NS. Vascular damage as an underlying mechanism of cardiac and cerebral toxicity in irradiated cancer patients. Radiat Res. 2010;174:865-869.
59. Li CS, Schmieke U, Tan TY. Extracranial carotid artery disease in nasopharyngeal carcinoma patients with post-irradiation ischemic stroke. Clin Neurol Neurosurg. 2010;112:682-686.
60. Young YH, Sheen TS. Preservation of tubal function in patients with nasopharyngeal carcinoma after radiation. Acta Otolaryngol. 1998;118:280-283.
61. Xu KC. Hydrogen-Oxygen Inhalation for Treatment of COVID-19. World Scientific; 2020. p. 23-25.
62. Qin ZX, Yu P, Qian DH, Song MB, Tan H, Yu Y, et al. Hydrogen-rich saline prevents neointima formation after carotid balloon injury by suppressing ROS and the TNF-α/NF-κB pathway. Atherosclerosis. 2012;220:343-350.
63. Ohsawa I, Nishimaki K, Yamagata K, Ishikawa M, Ohta S. Consumption of hydrogen water prevents atherosclerosis in apolipoprotein E knockout mice. Biochem Biophys Res Commun. 2008;377:1195-1198.
64. Song G, Zong C, Zhang Z, Yu Y, Yao S, Jiao P, et al. Molecular hydrogen stabilizes atherosclerotic plaque in low-density lipoprotein receptor-knockout mice. Free Radic Biol Med. 2015;87:58-68.
65. Fransson AE, Videhult Pierre P, Risling M, Laurell GFE. Inhalation of molecular hydrogen, a rescue treatment for noise-induced hearing loss. Front Cell Neurosci. 2021;15:658263.
66. Zhou ZQ, Zhong CH, Su ZQ, Li XY, Chen Y, Chen XB, et al. Hydrogen-oxygen mixture decreases inspiratory effort in patients with tracheal stenosis. Respiration. 2019;97:42-51.

Conflict of Interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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Explosive Progression of Brain Metastases from Lung Adenocarcinoma after Radiotherapy and Significant Reversal after Inhalation of Hydrogen-Oxygen Gas: A Case Report

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Abstract

Rationale: For multiple brain metastases complicated by Non-Small Cell Lung Cancer (NSCLC), Whole Brain Irradiation Therapy (WBRT) is the first choice for treatment. But little is known about the explosive progression of brain metastases caused by radiotherapy and the reversal of hydrogen gas inhalation.
Patient concerns: A 51-year-old female patient was diagnosed with NSCLC with multiple brain metastases. After receiving Gefitinib and Osimertinib, she received 28 days of WBRT.
Diagnosis: One week after the end of WBRT, the patient's general condition suddenly worsened, with severe headache, vomiting, confusion, and incontinence. MRI showed that the number and size of brain metastases increased exponentially.
Interventions and outcomes: In the absence of any special "anti-cancer" treatment available, the patient only receives hydrogen gas inhalation therapy, inhaling a mixture of hydrogen and oxygen (66% H_2 and 34% O_2), 3000 ml/min, at least 6 hours a day. The patient's condition began to improve after a week, and markedly improved after one month. A reexamination of MRI showed that more than 90% of brain metastases disappeared.
Lessons: Radiotherapy is a double-edged sword. Doctors should pay attention to the adverse effects of radiotherapy, especially in patients with lung cancer. Molecular hydrogen is a therapeutic gas that can selectively scavenge toxic free radicals; its role deserves further research.
Keywords: Lung cancer; Radiotherapy; Hydrogen gas; Inhalation

Introduction

Non Small Cell Lung Cancer …

Case Presentation

The patient began taking Gefitinib (250 mg/d) since December 25, 2019. In April 2020, nearly three-month treatment later, the patient complained of worsening headaches, reevaluation showed progression in both lung and brain lesions. Repeated genetic sequencing of tumors performed twice in different centers showed that the original EGFR exon 21 L858R mutations turned negative.

> *Figure 1. Chest Computed Tomography (CT) showed irregular solid mass lesions in the left lower lung, with small cavities, multiple small lesions in both lungs, with mediastinal lymph node metastasis.*

> *Figure 2. Head Magnetic Resonance Imaging (MRI) showed multiple abnormal nodular enhancements in the two cerebral hemispheres.*

From April 23, the patient stopped taking Gefitinib, changed to take Osimertinib. From May 2 until May 29, the patient received WBRT with an intensity of D50Gy/25f, at a frequency of 5 days per week.

One week after the radiotherapy, the patient suddenly had difficulty walking, severe headache, vomiting followed by confusion and incontinence. Re-examination of MRI on June 3 showed that the number and size of metastases in the brain increased explosively, covering the entire brain (Figure 3). In the following month, the patient did not receive any special "anti-cancer" treatment except for symptomatic and supportive treatment. On July 16, the patient began to receive hydrogen inhalation therapy by using a hydrogen-oxygen nebulizer (AMS-H-01, Asclepius, Shanghai, China, SFDA registration No. 20203080066) and inhaling a mixture of hydrogen and oxygen (66% H_2 and 34% O_2) with a gas flow rate of 3,000 ml/min through a nasal cannula for a total of at least 6 hours per day, intermittently or continuously inhaling. After half a month inhalation, the patient's self-feeling began to improve. One month later, the patient's headache disappeared, she was able to walk on the ground, and her eating, consciousness, urination and bowel movement return to normal. Re-examination of MRI showed that more than 90% of the brain metastases disappeared (Figure 4).

Since then, the patient successfully received treatments such as Sintilimab (PD-1 inhibitor), Carboplatin, and Bevacizumab. On October 22, 2021, 15 months after the start of hydrogen inhalation, the follow-up found that the patient felt good about herself and was able to perform light physical labor, the lung lesions were further improved, and the brain metastases were still stable and in further improvement (Figure 5).

> *Figure 3. Re-examination of MRI after WBRT showed that the number and size of metastases in the brain increased explosively, covering the entire brain.*

> *Figure 4. MRI was rechecked at 2 months after the start of hydrogen inhalation, and more than 90% of the patient's brain metastases had disappeared.*

> *Figure 5. Follow-up 15 months after the start of hydrogen inhalation, the patient's brain metastases are still stable.*

Discussion

The NSCLC patient stated above has very special and thought-provoking clinical features. First, in view of the EGFR mutation, the patient first received the first-generation TKI Gefitinib treatment, but there was no clinical response and the recheck genetic sequencing showed that the original EGFR exon 21 L858R mutations disappeared. The patient changed to receive the third-generation TKI Osimertinib, which did not show clinical effects, but progressed to lung lesion and brain metastasis. Secondly, after receiving whole brain irradiation for BMs, the whole body condition deteriorated sharply, life was in a critical state, and brain metastases, both in number and size, showed an explosive increase. Third, given that no special treatment was available, the patient received hydrogen (mixed hydrogen and oxygen) inhalation. The magic is that only relying on this simple therapy, the patient's general state and brain metastasis have been significantly improved and reversed. It has been 15 months since

hydrogen inhalation, and the patient's lung damage and brain metastasis are still stable and in further improvement.

BMs are the main complication of EGFR-positive NSCLC. Chemotherapy-based treatment is generally not used to treat BM, mainly because the blood-brain barrier prevents the penetration of chemotherapy drugs into the brain, resulting in limited efficacy. The first-generation TKI has a low brain penetration rate and a low response rate to the treatment of BMs. Compared with the first-generation TKI, Osimertinib has become the first-line TKI of choice for the treatment of NSCLC with BMs due to its tolerability, prolonged Progression-Free Survival (PFS) and higher brain penetration. A group of multi-center clinical trials showed that the clinical response rate and disease control rate of Osimertinib in the treatment of NSCLC with BMs reached 91% and 95%, respectively. Unfortunately, the patient reported now, neither the Gefitinib nor the Osimertinib have been able to prevent disease progression. This may be related to the heterogeneity of lung cancer and genetic instability. The original EGFR mutation of this patient turned negative during the reexamination, indicating that the genotype of the tumor has changed, which may be the cause of the failure of the above-mentioned TKI treatment.

The patient's systemic state and brain metastasis progression showed a direct correlation in time with WBRT. As mentioned earlier, radiotherapy is a double-edged sword. On the one hand, radiation is an important local control method for cancer treatment. On the other hand, radiation can promote the growth and invasion of cancer cells, especially metastasis. A large amount of evidence from in vivo and in vitro experiments indicates that ionizing radiation applied to cancer cells or host cells, tumors, the entire host, or putative tumor development sites may stimulate the metastasis process.

More than 60 years ago, Kaplan and Murphy showed the local roentgen irradiation increased frequency of pulmonary metastasis in a transplantable mouse carcinoma. In 1988, Milas et al reported that most tumors growing in irradiated tissues have an increased propensity to metastasize, which is linked to their manifestation of Tumor Bed Effect (TBE). Some randomized and non-randomized clinical studies have reported that radiotherapy promotes metastasis in hepatocellular carcinoma or head and neck cancer. Martin et al identified increased Circulating Tumor Cells (CTC) numbers singly and in clumps in large numbers in patients treated with palliative or curative-intent radiotherapy.

The mechanisms of pro-metastatic effect of radiation are not fully understood. Cui et al showed that radiation up-regulate the expression of Granulocyte-Colony-Stimulating Factor (G-CSF) and transducing its intracellular signal transduction JAK/STAT3 (Janus kinase/signal transduction), trigger the Epithelial-Mesenchymal Cell Transformation (EMT) of NSCLC. Cancer cells that undergo EMT have been shown to acquire stemness and undergo metabolic changes, promoting metastasis and invasion. Further research shows that radiation can increase the production of Reactive Oxygen Species (ROS), which can mediate most of the biological effects of radiation itself, and is closely related to tumorigenesis and tumor progression. There is evidence that ROS have been implicated in radiation-induced EMT, via activation of several EMT transcription factors.

Based on the role of ROS in promoting metastasis by radiation, it is not difficult to understand that hydrogen inhalation can reverse the progression of brain metastasis in this case. In fact, the ability of hydrogen to control cancer should be traced back to 1975, when Dole et al reported the successful treatment of transplanted skin squamous epithelial carcinoma in mice

by inhalation of a mixture of hydrogen and oxygen (97.5% H_2 and 2.5% O_2) at 8 atmospheres. But this major discovery seemed to have been forgotten until 2007, when Ohsawa et al found that hydrogen could ameliorate cerebral ischemia-reperfusion injury by selectively reducing cytotoxic ROS, including hydroxyl radical (•OH) and peroxynitrite (ONOO-), provoking a worldwide attention. Now it has known that hydrogen gas has great value as a therapeutic agent for a variety of illnesses, including cancer.

In 2019, our team took the lead in reporting a retrospective and prospective follow-up study of 82 patients with stage III and IV cancers undergoing hydrogen inhalation therapy, including 22 cases of NSCLC with or without brain metastases. The results showed that inhaled hydrogen (66% H_2 and 34% O_2) can improve the quality of life of patients and control cancer progression. According to our research results, hydrogen inhalation seems to have a unique effect on NSCLC with brain metastases. There is a 44-year-old female patient with NSCLC who has multiple intracranial metastases, and has hydrocephalus in the third and lateral ventricles, as well as bone, adrenal glands, and liver metastases. In the case of failure of conventional treatment, the patient only receives hydrogen and oxygen inhalation therapy. Four months later, the brain tumors significantly shrunk, and the amount of hydrocephalus in the third and lateral ventricles was significantly reduced. One year later, all brain tumors disappeared, liver, lung and other organs metastasis were stable. Tumor markers that were increased before treatment returned to normal range.

The mechanism of hydrogen inhibiting cancer has been studied in a series of experiments, and it has been found that hydrogen molecules can inhibit the viability, migration and invasion of cancer cells, and catalyzed cell apoptosis and induced cancer cells G2/M arrest. Liu et al suggested that H_2 can promote lung cancer cell apoptosis via inhibiting the activation of STAT3/Bcl2 signaling. Meng et al found that H_2 treatment resulted in a decrease in the expression levels of CD47 and cell division control protein 42 (CDC42) in a dose-dependent manner. CD47 is a transmembrane glycoprotein that mediates 'self/don't eat me' signal by inhibiting macrophage phagocytosis, and is frequently up-regulated in a variety of cancers, including NSCLC. Wang et al showed that H_2 down-regulates the expression of a regulator for chromosome condensation, that is, structural maintenance of chromosomes (SMC), SMC, and reduces the expression of cyclin D1, CDK4 and CDK6, thereby inhibiting lung cancer progression.

In addition, the better effect of hydrogen on brain metastasis may also be related to its physical properties. Hydrogen molecules are the smallest and highly dispersed, which makes it easy to penetrate the blood-brain barrier into the brain.

The inhibitory effect of hydrogen on NSCLC may also be related to its immune regulation. Akagi and baba have confirmed that hydrogen gas inhalation in NSCLC patients can decrease the abundance of exhausted terminal PD1+CD8+ T cells, increased that of active terminal PD1+CD8+ T cells, maintain the balance between terminal PD1+ and PD1+CD8+ T cells. Our research showed that two weeks of hydrogen inhalation in 20 NSCLC patients induced decrease of number of exhausted and aging cytotoxic T cells, and the abnormally decreased lymphocyte subsets, including functional helper and cytotoxic T cells, natural killer T cells, and γδ T2 cells, tended to be normal. It is particularly important that we found that the patient of metastatic gallbladder cancer who only received hydrogen inhalation therapy. Before significant improvement was achieved, the tumor and tumor markers not only did not improve, but "worsened" and showed "false progress". Pseudo progression is considered to be a special remission mode that occurs after PD-1 antibody treatment, and it may be that cytotoxic

T cells infiltrate the tumor and cause edema and necrosis, suggesting immune activation. The similar reaction of hydrogen shows that it has the effect of activating anti-tumor immunity. Unfortunately, we did not follow up the immune status of the NSCLC patient reported now.

In the treatment of cancer with hydrogen, there is no controlled study as to whether it is advisable to inhale pure hydrogen or a mixture of hydrogen and oxygen. There is no evidence that oxygen itself inhibits cancer, but hypoxia will definitely promote the invasion and metastasis of cancer cells. The smallest hydrogen molecules can carry larger molecules of oxygen into the deep tissues. It has been reported that hydrogen gas can improve the flexibility of red blood cells and enable them to transport oxygen into the capillaries in the final organs, thereby improving the tumor hypoxia and reducing the cancer promotion effect caused by hypoxia.

Conclusion

We report a case of NSCLC with multiple metastases with a very special clinical course. After whole-brain irradiation, brain metastasis increased explosively, and miraculously improved after inhalation of hydrogen (hydrogen and oxygen) only. Although it is difficult to be sure that hydrogen can completely control the disease, at least when the patient is in a critical state, it reverses the condition and provides an opportunity for further treatment.

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Author Contribution

Project administration: Kecheng Xu; Clinical Supervision: Wei Qian, Juan Zhen; Data collection: Tianyu Lu; Writing - original draft: Kecheng Xu; Co-author: Juanjuan Shi; Review & editing: Xiaofeng Kong.

References

1. Ulahannan D, Khalifa J, Faivre-Finn C, Lee SM. Emerging treatment paradigms for brain metastasis in non-small-cell lung cancer: an overview of the current landscape and challenges ahead. Ann Oncol. 2017;28(12):2923-2931.
2. Ernani V, Stinchcombe TE. Management of brain metastases in non-small-cell lung cancer. J Oncol Pract. 2019;15(11):563-570.
3. Errico A. Radiotherapy: A double-edged sword for NSCLC? Nat Rev Clin Oncol. 2014;11(12):684.
4. Brown PD, Ballman KV, Cerhan JH, Anderson SK, Carrero XW, Whitton AC, et al. Postoperative stereotactic radiosurgery compared with whole brain radiotherapy for resected metastatic brain disease (NCCTG N107C/CEC.3): a multicentre, randomised, controlled phase 3 trial. Lancet Oncol. 2017;18(8):1049-1060.
5. Ernani V, Stinchcombe TE. Management of brain metastases in non-small-cell lung cancer. J Oncol Pract. 2019;15(11):563-570.
6. Ballard P, Yates JW, Yang Z, Kim DW, Yang JCH, Cantarini M, et al. Preclinical comparison of osimertinib with other EGFR-TKIs in EGFR-mutant NSCLC brain metastases models and early evidence of clinical brain metastases activity. Clin Cancer Res. 2016;23(20):5130-5140.
7. Soria JC, Ohe Y, Vansteenkiste J, Reungwetwattana T, Chewaskulyong B, Lee KH, et al. Osimertinib in untreated EGFR-mutated advanced non-small-cell lung cancer. N Engl J Med. 2018;378(2):113-125.
8. de Bruin EC, McGranahan N, Mitter R, Salm M, Wedge D, Yates L, et al. Spatial and temporal diversity in genomic instability processes defines lung cancer evolution. Science. 2014;346(6206):251-256.

9. Sundahl N, Duprez F, Ost P, De Neve W, Mareel M. Effects of radiation on the metastatic process. Mol Med. 2018;24(1):1-16.
10. Kaplan HS, Murphy ED. The effect of local roentgen irradiation on the biological behavior of a transplantable mouse carcinoma; increased frequency of pulmonary metastasis. J Natl Cancer Inst. 1949;9(5-6):407-413.
11. Milas L, Hirata H, Hunter N, Peters LJ. Effect of radiation-induced injury of tumor bed stroma on metastatic spread of murine sarcomas and carcinomas. Cancer Res. 1988;48(8):2116-2120.
12. Chang VL, Jian JI, Cheng SH, Tsai SYC, Chuang VP, Soong T, et al. Sublethal irradiation induces vascular endothelial growth factor and promotes growth of hepatoma cells: implications for radiotherapy of hepatocellular carcinoma. Clin Cancer Res. 2006;12(9):2706-2715.
13. Cheng TC, Chuang VP, Cheng SH, Lin YM, Cheng TJ, et al. Unresectable hepatocellular carcinoma treated with radiotherapy and/or chemoembolization. Int J Cancer. 2001;96(4):243-252.
14. Nozue M, Isaka N, Fukao K. Over-expression of vascular endothelial growth factor after preoperative radiation therapy for rectal cancer. Oncol Rep. 2001;8(6):1247-1249.
15. Schantz SP, Peters LJ. Patterns of recurrence from head and neck cancer. An immunologic perspective. Am J Clin Oncol. 1987;10(6):469-474.
16. Strong MS, Vaughan CW, Kayne HL, Aral IM, Ucmakli A, Feldman M, et al. A randomized trial of preoperative radiotherapy in cancer of the oropharynx and hypopharynx. Am J Surg. 1978;136(4):494-500.
17. Martin OA, Anderson RL, Russell PA, Cox RA, Ivashkevich A, Swierczak A, et al. Mobilization of viable tumor cells into the circulation during radiation therapy. Int J Radiat Oncol Biol Phys. 2014;88(2):395-403.
18. Cui YH, Suh Y, Lee YJ, Yoo KC, Eddin N, Jeong YJ, et al. Radiation promotes invasiveness of non-small-cell lung cancer cells through granulocyte-colony-stimulating factor. Oncogene. 2015;34(42):5372-5382.
19. Dave B, Mittal V, Tan NM, Chang JC. Epithelial-mesenchymal transition, cancer stem cells and treatment resistance. Breast Cancer Res. 2012;14(1):202.
20. Marie-Egyptienne DT, Lohse I, Hill RP. Cancer stem cells, the epithelial to mesenchymal transition (EMT) and radioresistance: potential role of hypoxia. Cancer Lett. 2013;341(1):63-72.
21. Chang L, Graham PH, Hao J, Bucci J, Cozzi PJ, Kearsley JH, et al. Emerging roles of radioresistance in prostate cancer metastasis and radiation therapy. Cancer Metastasis Rev. 2014;33(2-3):469-496.
22. Kam WW, Banati RB. Effects of ionizing radiation on mitochondria. Free Radic Biol Med. 2013;65:607-619.
23. Leach JK, Van Tuyle G, Lin PS, Schmidt-Ullrich R, Mikkelsen RB. Ionizing radiation-induced, mitochondria-dependent generation of reactive oxygen/nitrogen. Cancer Res. 2001;61(10):3894-3901.
24. Tulard A, Hoffschir F, de Boisferon FH, Luccioni C, Bravard A. Persistent oxidative stress after ionizing radiation is involved in inherited radiosensitivity. Free Radic Biol Med. 2003;35(1):68-77.
25. Lee SY, Jeong EK, Ju MK, Jeon HM, Kim MY, Kim CH, et al. Induction of metastasis, cancer stem cell phenotype, and oncogenic metabolism in cancer cells by ionizing radiation. Mol Cancer. 2017;16(1):10.
26. Ohsawa I, Ishikawa M, Takahashi K, Watanabe M, Nishimaki K, Yamagata K, et al. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med. 2007;13(6):688-694.
27. Dole M, Wilson FR, Fife WP. Hyperbaric hydrogen therapy: a possible treatment for cancer. Science. 1975;190(4210):152-154.
28. Ohsawa I, Ishikawa M, Takahashi K, Watanabe M, Nishimaki K, Yamagata K, et al. Hydrogen acts as a therapeutic antioxidant by selectively reducing cytotoxic oxygen radicals. Nat Med. 2007;13(6):688-694.

29. Ohno K, Ito M, Ichihara M, Ito M. Molecular hydrogen as an emerging therapeutic medical gas for neurodegenerative and other diseases. Oxid Med Cell Longev. 2012;2012:353152.
30. Iida A, Nosaka N, Yumoto T, Knaup E, Naito H, Nishiyama C, et al. The clinical application of hydrogen as a medical treatment. Acta Med Okayama. 2016;70(5):331-337.
31. Liu MY, Xie F, Zhang Y, Wang TT, Ma SN, Zhao PX, et al. Molecular hydrogen suppresses glioblastoma growth via inducing the glioma stem-like cell differentiation. Stem Cell Res Ther. 2019;10(1):145.
32. Chen JB, Kong XF, Lv YY, Qin SC, Sun XJ, Mu F, et al. Real world survey of hydrogen-controlled cancer: a follow-up report of 82 advanced cancer patients. Med Gas Res. 2019;9(3):115-121.
33. Chen J, Mu F, Lu T, Ma Y, Du D, Xu K. A gallbladder carcinoma patient with pseudo-progressive remission after hydrogen inhalation. Onco Targets Ther. 2019;12:11145-11151.
34. Liu J, Yan Z, Wang Y, Meng J, Chen G. Suppression of autophagy facilitates hydrogen gas-mediated lung cancer cell apoptosis. Oncol Lett. 2020;20(4):112.
35. Wu P, Yuan M, Song J, Chen X, Yang H. Hydrogen Gas from Inflammation Treatment to Cancer Therapy. ACS Nano. 2019;13(8):8505-8511.
36. Yang Y, Liu PY, Bao W, Chen SJ, Wu FS, Zhu PY. Hydrogen inhibits endometrial cancer growth via a ROS/NLRP3/caspase-1/GSDMD-mediated pyroptotic pathway. BMC Cancer. 2020;20(1):28.
37. Meng J, Liu L, Wang D, Yan Z, Chen G. Hydrogen gas represses the progression of lung cancer via down-regulating CD47. Biosci Rep. 2020;40(4):BSR20192761.
38. Wang D, Wang L, Zhang Y, Zhao Y, Chen G. Hydrogen gas inhibits lung cancer progression through targeting SMC3. Biomed Pharmacother. 2018;104:788-797.
39. Akagi J, Baba H. Hydrogen gas activates coenzyme Q10 to restore exhausted CD8+ T cells, especially PD-1+Tim3+terminal CD8+ T cells, leading to better nutritional outcomes in patients with lung cancer. Oncol Lett. 2020;20(5):258.
40. Chen JB, Kong XF, Mu F, Lu TY, Lu YY, et al. Two weeks of hydrogen inhalation can significantly reverse adaptive and innate immune system senescence in patients with advanced non-small cell lung cancer: a self-controlled study. Med Gas Res. 2020;10(4):149-154.
41. Chen J, Mu F, Lu T, Ma Y, Du D, Xu K. A gallbladder carcinoma patient with pseudo-progressive remission after hydrogen inhalation. Onco Targets Ther. 2019;12:11145-11151.
42. Cohen JV, Alomari AK, Vortmeyer AO, Jilaveanu LB, Goldberg SB, Mahajan A, et al. Melanoma brain metastasis pseudoprogression after pembrolizumab treatment. Cancer Immunol Res. 2016;4(3):179-182.
43. Zhou ZQ, Zhong CH, Su ZQ, Li XY, Chen Y, Chen XB, et al. Breathing hydrogen-oxygen mixture decreases inspiratory effort in patients with tracheal stenosis. Respiration. 2019;97(12):42-51.
44. Oser A, Arslan M, Çomu FM, Küçük A, Mardin B, Koçak B, et al. The effect of hydrogen-rich saline solution on erythrocyte deformability in power limb ischemia reperfusion injury in rats. GMJ. 2021;32(2):170-172.