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CLINICAL TRIAL REPORT

Pilot Feasibility and Safety Study of Hydrogen Gas Inhalation in Locally Advanced Head and Neck Cancer Patients

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Purpose: Hydrogen (H_2) gas inhalation might alleviate acute radiotherapy toxicities by scavenging free radicals produced by ionizing radiation and anti-inflammatory properties. This study aimed to investigate the feasibility and safety of H_2 gas inhalation during concurrent chemoradiotherapy (CCRT) in patients with locally advanced head and neck cancer (LAHNC).

Patients and Methods: We designed a pilot prospective study combining CCRT with aerosol inhalation of H_2 gas. Each patient was scheduled to receive daily intensity-modulated radiotherapy (IMRT) in 33 fractions on a weekday and six cycles of weekly chemotherapy. All patients inhaled H_2 gas through a cannula or mask 1 hour per day, 1–2 hours before IMRT. The primary endpoint was the feasibility of H_2 inhalation. Eighty percent of the patients who completed at least 20 applications of H_2 gas inhalation were considered feasible. The secondary endpoints were safety profiles during H_2 gas inhalation (vital signs and symptoms related to H_2 gas inhalation) and acute toxicities during CCRT.

Results: We enrolled 10 patients with LAHNC between July 2023 and December 2023. All patients received 33 fractions of H_2 gas inhalation on the same day as the IMRT. Vital signs during and at the end of H_2 gas inhalation were stable in all patients. None of the 10 patients had hypertension or hypotension during any of the 33 inhalations. No adverse events related to H_2 gas inhalation, such as cough, nasal bleeding, dizziness, headache, nausea, or vomiting, were reported. Grade 3 leukopenia was found in two patients (20%) during the 5th week of CCRT. Grade 2 radiation dermatitis and pharyngitis were found in three patients (30%).

Conclusion: H_2 gas inhalation combined with CCRT is feasible and safe for patients with LAHNC.

Keywords: hydrogen gas, head and neck cancer, concurrent chemoradiotherapy

Introduction

Hydrogen (H2) has small molecular size and neutral charge. It can efficiently penetrate and reach target tissues. It has been suggested as a potential treatment for various diseases related to oxidative stress.[1] It is well known that free radical scavengers can decrease oxidative stress (OS) in mammalian cells via the indirect effect of ionizing radiation (IR). Reactive oxygen species (ROS) are the main cause of indirect effect of IR-induced cellular damage through the production of ROS by water radiolysis. Most IR-induced side effects and toxicity to normal tissue cells are caused by hydroxyl radicals ($^{\cdot}OH$), the most toxic ROS.[2] In this context, the combination of an antioxidant (such as $^{\cdot}OH$ scavengers) with IR might prevent normal tissue damage.

Numerous studies have confirmed that molecular hydrogen can be a novel antioxidant and therapeutic gas (non-toxic at concentrations less than 4%)[3–6] because of its ability to selectively react and reduce the strongest oxidants, such as $^{\cdot}OH$ and peroxynitrite ($ONOO^-$).[3–5] Several previous studies have indicated the preventive and therapeutic effects of H_2 gas inhalation, including anti-oxidation, anti-apoptotic, anti-inflammatory, and autophagy regulation, thereby decreasing OS

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in cells induced by IR.[1,7–9] Furthermore, the radioprotective effects of H_2 gas inhalation before irradiation were also demonstrated in the gastrointestinal tract and cardiovascular system in vitro and in vivo studies.[10–12]

These assumptions are supported by earlier researches[13,14] showing that H2, in various forms (H2-rich water, H2 gas inhalation) can reduce oxidative stress and improve QOL in patients undergoing irradiation without diminishing its therapeutic effects. The results further suggest that the radioprotective effects of H2 may not be solely due to direct scavenging of ·OH but also through the activation of endogenous protective mechanisms.

The latest review[15] suggests that H2's ability to selectively neutralize highly toxic reactive oxygen species (ROS) is likely the primary and essential mechanism behind its therapeutic effects. This selective scavenging helps reduce oxidative stress, which is crucial for its potential applications in treating various diseases, including cancer. Although reactive oxygen species like ·OH and peroxynitrite (ONOO–) are generated rapidly during radiation, the study suggests that H2 can be therapeutically effective when administered post-radiation due to its dual direct and indirect effects on oxidative stress. This finding aligns with many literatures demonstrating the radioprotective potential of H2 in animal studies.[16,17]

A previous study investigated the safety of H_2 gas inhalation in 9 healthy adults using a high-flow nasal cannula (15 L/min) for 24, 48, and 72 h.[18] They found that inhalation of 2.4% H_2 was well tolerated with no clinically significant adverse events and no clinically significant changes in vital signs, neurologic examination, pulmonary function testing, electrocardiography (ECG) changes, or any laboratory variables associated with up to 72 hours of H_2 inhalation.

According to the most recent GLOBOCAN data from 2020,[19] head and neck cancer (HNC) ranks as the seventh most common cancer worldwide, representing about 4.5% of all cancer diagnoses globally. The incidence and mortality rates of HNC show significant variation depending on the geographic region and population demographics. South and Southeast Asia have the highest incidence rates, largely due to the widespread use of the carcinogenic betel and areca nut in these areas. HNC in Thailand has an age-standardized incidence rate (ASR) of approximately 15.7 per 100,000 males and 10.7 per 100,000 females. It ranks among the top five most common cancers in the country, highlighting its significant public health burden.[20]

Concurrent chemoradiotherapy (CCRT) is a treatment modality for locally advanced head and neck cancer (LAHNC).[21] However, acute toxicities from treatment that occur in most patients directly affect quality of life.[22,23] Reporting of grade 3 toxicities from radiotherapy combined with chemotherapy was 77%, while radiotherapy alone accounted for only 34%.[24] Oral mucositis can affect swallowing musculature and result in fatigue, anorexia, dehydration, and malnutrition. Salivary glands are organs that can be damaged by radiotherapy, and xerostomia is related to dental caries followed by infection. Dermatitis is also a problem in that some patients withdraw from the treatment because they are unable to tolerate.

The rationale for this study is to investigate the potential of H2 gas inhalation as a radioprotective agent for LAHNC patients undergoing CCRT. The hypothesis is that H2 provides protection against radiation-induced damage by two mechanisms: directly neutralizing harmful reactive oxygen species (ROS) and activating the body's natural defense systems, enhancing antioxidant and anti-inflammatory responses. This dual action is expected to offer broad radioprotection by reducing oxidative stress and limiting radiation-related damage, while preserving the therapeutic benefits of CCRT.

Given the limited data on the use of H2 gas inhalation in HNC patients, the study was designed as a single-arm prospective pilot feasibility trial. The primary goal of the study was to assess the feasibility of administering H2 gas inhalation to LAHNC patients undergoing CCRT. Secondary objectives included evaluating the safety profile of H2 gas inhalation and monitoring acute toxicities during the course of CCRT treatment.

Methods

Patients

The eligibility criteria included patients with locally advanced stage of all sites of head and neck cancer (HNC), age ≥ 18–70, ECOG performance status 0–2, planned for curative CCRT. In our study, we did not exclude the patients with cranial nerve VII, IX, X, and XI palsy. Patients were excluded if they had recurrent or metastatic disease, prior head and neck irradiation, or previous surgery in the head and neck area (except incisional or excisional biopsy). Patients with contraindications to radiotherapy or chemotherapy and those who had a tracheostomy tube were also excluded. This study was approved by the Institutional Review Board of the Faculty of Medicine, Chiang Mai University and was registered with the Thai Clinical Trials Registry number TCTR20230627002. Written informed consent was obtained from all patients.

Sample Size

There are no universally established guidelines for determining the ideal sample size in pilot studies for healthcare study. While having a sample size justification is essential for pilot and feasibility studies, formal sample size calculations are not always deemed appropriate. As suggested in the literature, the sample size for pilot studies can be estimated based on recommendations for feasibility trials.[25,26] Following Stallard's approach,[27] it is advised that the sample size for a pilot study should be roughly 3% of the sample size planned for the larger, definitive trial. Based on this proposed, a sample size of 10 patients was chosen for our study.

Radiotherapy

All patients received intensity-modulated radiotherapy (IMRT) with a total dose of 69.96 Gy to the gross tumor volume (GTV) and high-risk area, 59.4 Gy to the intermediate-risk area, and 54 Gy to the low-risk area in 33 fractions.

Chemotherapy

Concurrent chemotherapy consisted of weekly cisplatin (40 mg/m^2) or weekly carboplatin with an area under the curve (AUC) of 2.[28] Chemotherapy regimens depended on the patient's renal function or on the discretion of the doctors.

H_2 Gas Inhalation

All patients inhaled H_2 gas through a cannula or mask 1 hour per day connected to a Hycellvator ET 100 (Helix Japan, Co., Ltd., Tokyo, Japan), 1–2 hours before irradiation. This machine produced 99.99% H_2 purity using an electrolysis system (66% H2 in air) with a flow rate of 1.2 L/min.

Assessment

Vital signs and symptoms related to H_2 gas inhalation (cough, nasal bleeding, dizziness, nausea, and vomiting) were recorded before, during, and immediately after the procedure. The acute toxicities of fatigue, anorexia, dry mouth, oral mucositis, dysphagia, nausea, vomiting, radiation dermatitis, and bone marrow were assessed weekly and graded according to the Common Terminology Criteria for Adverse Events (CTCAE) version 5.0.

Statistical Analysis

Feasibility was defined as 80% of the patients completing at least 20 applications of H_2 gas inhalation. Intent-to-treat analysis was performed for all enrolled patients who received at least one session of H_2 gas inhalation. Descriptive statistics were used for baseline characteristics. CCRT toxicity assessment was reported based on the frequency.

Results

Patients

The Details of 10 patients in the study are shown in Table 1. Ten patients with HNC scheduled to receive curative CCRT were enrolled in this pilot study. Ten patients with HNC scheduled to receive curative CCRT were enrolled in this pilot study between July 2023 and December 2023. Most patients were male (90%). Their ages ranged from to 30–68 years old with a median of 58 years. All were locally advanced stages (III–IVB AJCC 8^{th} edition). The most common primary site was nasopharynx in 4 patients followed by oropharynx. Concurrent weekly cisplatin was prescribed to 8 patients, while 2 patients received concurrent weekly carboplatin.

Compliance of CCRT

Compliance with CCRT is shown in Table 1. All patients completed 33 fractions of IMRT as planned. The median overall treatment time (OTT) was 49 days (range 47–63). One patient experienced delayed chemotherapy due to grade 3 leukopenia and another due to a combination of grade 2 radiation dermatitis, dysphagia, and leukopenia.

Table 1 Details of 10 Patients in the Study

Patient No.	Sex	Age	Primary Site	Stage	CCRT Regimen	Overall Treatment Time; OTT (Days)	No. of Cycles of Chemo therapy	Delayed of Chemo therapy	Follow Up Time (Months)	Status at the Last Follow Up
1	M	61	Nasopharynx	III	Cisplatin	54	6	No	14	NED
2	M	68	Oropharynx	IVA	Cisplatin	63	6	Yes	13	Lt Axillary lymph node metastasis
3	M	48	Larynx	IVA	Cisplatin	49	6	No	13	NED
4	M	56	Nasopharynx	IVA	Carboplatin	47	6	No	12	NED
5	M	43	Oropharynx	IVA	Cisplatin	47	6	No	12	NED
6	M	59	Oropharynx	IVA	Cisplatin	48	6	No	11	NED
7	M	59	Nasopharynx	III	Cisplatin	49	6	No	11	NED
8	F	60	Nasopharynx	IVA	Carboplatin	56	6	Yes	10	NED
9	M	58	Larynx	III	Cisplatin	59	6	No	10	NED
10	M	30	Hypopharynx	IVB	Cisplatin	49	6	No	6	Died from lung metastasis

Abbreviations: M, Male, F, Female; NED, No evidence of disease.

Feasibility and Safety of H_2 Gas Inhalation

All 10 patients completed 33 applications of H_2 gas inhalation on the same day as RT. According to the definition of feasibility in this study (80% of patients completing at least 20 applications), H_2 gas inhalation is feasible for HNC patients treated with CCRT. Regarding the safety of H_2 gas inhalation, we did not find any adverse events or symptoms related to H_2 gas, including cough, epistaxis, headache, dizziness, nausea and vomiting through 33 applications in all 10 patients. Blood pressure before, during, and immediately after H_2 gas inhalation were also normal.

Acute Toxicities During CCRT

We assessed weekly acute CCRT toxicities using the NCI CTC version 5.0. The maximum toxicity grades during CCRT for each patient are shown in Figure 1. Grade 3 leukopenia was observed in 1 patient (10%). The most common CCRT toxicities were radiation dermatitis, dysphagia, and dry mouth, which were found in 40% of the patients, and all were grade 2.

Tumor Response

With a mean follow up of 11.2 months (Range 6–14), 1 patient had lung metastasis and died after 6 months of CCRT completion, 1 patient had axillary lymph node metastasis. Other 8 patients had no evidence of disease.

Discussion

Although H_2 is an explosive gas, its dissolution in distilled water or physiological saline could make it non-explosive and safe.[9] H_2 is produced by bacteria in large bowels and circulates in the human body, although it is physiologically harmless to H_2-rich solution gas inhalation.[29] Several studies have explored the feasibility and safety of H_2 gas inhalation in healthy volunteers and in many types of patients. The safety of 1- session of prolonged H_2 gas inhalation from 4 to 72 h was studied in eight healthy adults. However, no serious adverse effects were reported. Moreover, changes in vital signs, organ function tests (pulmonary, neurology, cardiology), and serologic tests for abnormalities of bone marrow, renal, liver, pancreas, and cardiac organs during gas inhalation were not found compared to baseline.[18] This was a prospective study in the intensive care unit of 5 patients diagnosed with post-cardiac arrest syndrome (PCAS). All patients received 18-hour H_2 gas inhalation through a ventilator. No adverse side effects or events related to H_2 gas inhalation were observed. Four patients survived for more than 3 months and had good neurological function.[30]

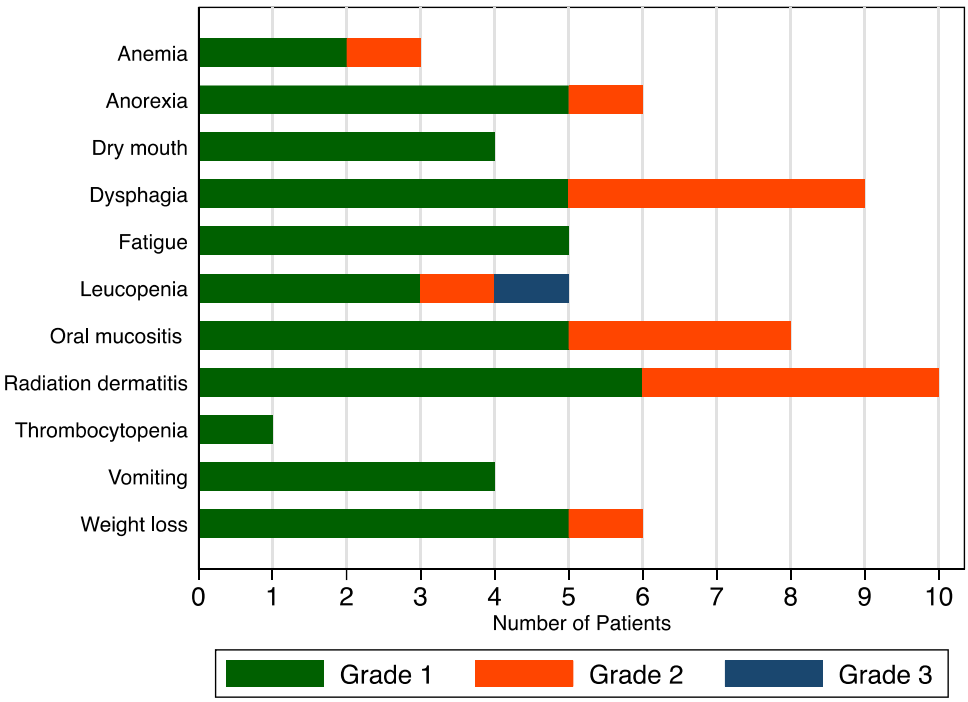

Figure 1 Maximum grade toxicities (NCI CTC version 5.0) during CCRT.

Chemotherapy and radiotherapy are commonly used approaches in cancer treatment; however, both can result in considerable oxidative stress and inflammation, leading to damage in healthy tissues.[15] In 2015, Meng et al[31] suggested that incorporating hydrogen (H2) as an adjuvant therapy may help alleviate these side effects by its antioxidant and anti-inflammatory properties.

H2 has been shown to reverse cisplatin-induced oxidative stress and restore the activity of antioxidant enzymes. In animal models, the administration of H2 gas notably increased the number of surviving auditory hair cells and provided protection to the cochlea from cisplatin-related toxicity.[32]

Several studies have indicated that inhaling H2 gas during RT can help minimize damage to the hematological and immune systems.[33,34]

Akagi et al explored the role of H_2 gas in improving the diagnosis of patients with stage IV colorectal carcinoma who received chemotherapy (XELOX regimen).[35] In this study, H_2 gas inhalation was scheduled for 3- hours daily for 3 months, and no side effects were observed in any of the 55 patients. Kong et al[36] designed a study on H2 inhalation in nasopharyngeal cancer patients with hearing loss after RT. The patients inhaled H2 gas for 3–6 hours per day for 4–12 weeks. No side effects related to H2 gas inhalation, such as epistaxis, allergic reaction, dyspnea, chest pain, dizziness, nausea, or vomiting, were reported. Another retrospective observational study was performed on patients with metastatic cancer undergoing radiotherapy.[1] The study compared patients who received daily H2 gas inhalation along with hyperbaric oxygen therapy to those who received only hyperbaric oxygen therapy for 30 minutes after each day of radiotherapy.[1] The results showed that H_2 gas inhalation significantly reduced the hematologic toxicities of radiotherapy and did not compromise the quality of life of patients. Our study was designed differently by repeating multiple sessions of a short course (1 h) of H_2 gas inhalation. All 10 LAHNC patients completed 33 sessions of 1-hour H_2 gas inhalation without interruptions or any side effects.

A critical aspect of non-hematologic acute toxicities resulting from CCRT in HNC is that conditions like mucositis, dermatitis, and dysphagia are major complications that significantly impact patients' quality of life. These toxicities pose a substantial burden, often leading to severe discomfort and difficulty in completing the full course of treatment.

We found that only one patient (10%) developed severe toxicity (grade 3 or higher), which was grade 3 anorexia. Grade 2 was commonly found in this study including dry mouth in four patients (40%), oral mucositis in three patients

(30%), dysphagia in four patients (40%), and radiation dermatitis in four patients (40%). The large cohort study of 576 hNC patients receiving IMRT[37] reported that 62.5% of the patients developed severe oral mucositis. The latest study from the DARS Trialist Group[38] reported grade 3 or higher acute toxicities during radiotherapy receiving standard IMRT as follows: anorexia, 47%; dry mouth, 33%; oral mucositis, 56%; and dysphagia, 58%.

Hematologic toxicities from CCRT pose a significant threat to the continuity and effectiveness of treatment in HNC patients. These toxicities increase the risk of treatment interruptions, chemotherapy dose reduction, delayed chemotherapy cycles, and prolonged overall radiotherapy treatment time. Addressing these toxicities promptly and appropriately is critical to maintaining the balance between managing side effects and achieving optimal therapeutic outcomes.

In our study, severe (grade 3) leukopenia was found in only one patient and caused a prolonged overall treatment time of 63 days. Grade 2 leukopenia also occurred in only one patient. Comparing to DARS study,[23] which had grade 3–4 leukopenia in 9%, grade 3 neutropenia in 4% and febrile neutropenia 2%.

Although these acute toxicities observed in our study were of lower grade and occurred at a lower percentage compared to other studies,[22,23] the findings suggest that H2 gas inhalation therapy holds promise as a novel therapeutic approach, particularly in acute care settings. While the potential benefits of H2 inhalation therapy are promising, incorporating it into current clinical protocols demands cautious consideration due to the preliminary nature of the studies conducted so far. Most of the available data comes from pilot or early-phase studies, which often involve small sample sizes and lack the robust evidence needed to establish definitive safety and efficacy profiles.

Previous animal studies[1,34,39,40] have demonstrated the radioprotective effects of H_2 by the mechanisms of direct effects on ·OH and indirect effects on ·OH via anti-inflammatory processes and host-mediated antioxidant activation. H_2 gas inhalation has a cytoprotective effect that improves RT-induced apoptosis, which plays a significant role in radiation-induced damage. Study by Zhao et al[41] found that H_2 can induce the expression of the anti-apoptotic protein Bcl-2 and inhibit the expression of the death promoter Bcl-2 related X protein. According to our study results, H2 gas inhalation may be a promising therapeutic strategy for patients with LAHNC receiving CCRT. Further research is needed to investigate the underlying mechanism involved in the protective role of H_2 gas inhalation against radiation-induced adverse effects in LAHNC patients.

Out of 677 articles reviewed,[42] despite variations in H2 administration methods, study designs, and cancer types, the outcomes remained consistent across the studies. The analysis concluded that H2 shows potential as both a standalone therapy and an adjuvant to existing cancer treatments, contributing to improved survival rates, enhanced quality of life, better blood parameters, and tumor reduction.

This trial, being the first prospective pilot study on H2 gas inhalation in patients with head and neck cancer undergoing CCRT, was designed to assess the feasibility of this intervention in a highly specific patient population. The strict inclusion and exclusion criteria, such as the exclusion of patients with tracheostomy tubes or secretion problems, limit the study's scope.

While the result offers valuable insights, several limitations must be acknowledged. The pilot nature of the research, along with its small sample size and highly selective patient criteria, restricts the generalizability of the findings. This narrow focus increases the potential for selection bias, where the outcomes observed in this specific group may not accurately represent the broader population.

To address these limitations, future studies should aim to include larger and more diverse HNC patient populations to ensure broader applicability of the results. Moreover, more comprehensive tracking of long-term outcomes is essential to gain a clearer understanding of the sustained efficacy and potential risks of H2 inhalation therapy. Large-scale randomized controlled trials (RCTs) are critical to confirm the initial results and determine the clinical utility of H2 inhalation. Additionally, identifying reliable biomarkers, such as oxidative stress or inflammation markers, would enhance the precision of H2 treatment protocols.

Conclusion

We conclude that H_2 gas inhalation is a safe and feasible procedure for patients with head and neck cancer who are treated with CCRT. We plan to conduct a prospective randomized study to explore metabolomic changes and evaluate the efficacy of this integrated and complimentary treatment combined with CCRT in terms of reducing acute toxicities.

Data Sharing Statement

All data generated or analyzed in this study are included in this manuscript.

Ethics Approval and Consent to Participate

This study *was* approved by the Ethical Committee of the Faculty of Medicine, Chiang Mai University, dated May 9, 2023, under approval number RAD-2565-09342. This study was registered in the Thai Clinical Trials Registry under the number TCTR20230627002. All procedures performed in this study involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 helsinki Declaration and its later amendments or comparable ethical standards. Informed consent was obtained from all the participants before participating in the study.

Author Contributions

All authors made a significant contribution to the work reported in all these areas; took part in drafting, revising or critically reviewing the article; gave final approval of the version to be published; have agreed on the journal to which the article has been submitted; and agree to be accountable for all aspects of the work.

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Disclosure

All authors declare that they have no competing interest.

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