Original Research Article

Efficacy and safety of hydrogen gas versus standard therapy in Chinese patients with cerebral infarction: A pilot study

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Abstract

Purpose: To evaluate the efficacy and safety of hydrogen (H_2) gas versus standard therapy in Chinese patients with acute cerebral infarction (ACI)

Method: Chinese patients with ACI who had National Institutes of Health stroke scale (NIHSS) scores of 2 - 6 were enrolled and randomized to receive hydrogen gas through inhalation route or standard therapy. Hydrogen gas (3 %) was administered twice daily for 1 hour each over a period of 2 weeks using a facial mask (non-rebreathing). Standard treatment included edaravone 30 mg (iv every 12 h + antiplatelet drugs for 2 weeks. Efficacy endpoints were change in vital signs, change in NIHSS scores from baseline, change in magnetic resonance imaging (MRI) signal intensity, and improvement in rehabilitation index. Safety was also assessed.

Results: Data for 200 patients were analyzed. Patients treated with H_2 gas had significantly greater improvement in NIHSS score than patients treated with standard therapy. This indicates that the neurological improvement was significantly greater in patients treated with H_2 gas than those that received standard therapy for all the days tested (p < 0.05). The onset of NIHSS score reduction was faster in patients treated with H_2 gas when compared to standard therapy. Furthermore, H_2 gas demonstrated significant improvement in MRI signal Intensity score, which indicates that H_2 gas was effective in recovery of infraction site.

Conclusion: The findings of this study show that H_2 gas administered via the inhalation route is effective and safe in Chinese patients with acute cerebral infarction, and is therefore, superior to standard therapy.

Keywords: Acute cerebral infarction, Hydrogen gas, Rehabilitation index, MRI, NIHSS

INTRODUCTION

Molecular hydrogen (H_2) has antioxidant properties with therapeutic value. It is generally being used in gas cylinder to avert the decompression and sickness induced by nitrogen [1]. In animals, H_2 is instinctively formed by duodenal microorganisms by anaerobic metabolism, that produces energy [2,3]. It is being metabolized enzymatically using hydrogenases enzyme to produce electrons [2-4].

In 1975, the role of hyperbaric H_2 in rodent model of carcinoma was reported, where it caused noticeable regression of tumours. However, hyperbaric H_2 was not available in clinical use as it was not a clinically possible option as it may interact with oxygen gas in cells and may causes harmful effects, misbalancing risk-benefits ration [3-7]. In contrast, H_2 is a physiologically inert gas so it may not interact with oxygen gas in cells [3,4]. In 2007, it has been reported that H_2 gas has anti-oxidant properties that has ability to protect brain against the ischemic attack and stroke by neutralizing free radicals namely OH and ONOO, however, H_2 gas is not effective against O^{2-}, H_2O_2 and NO [5-8]. These reports generated interest in scientific community to explore its effect in several studies. The effect of H_2 gas have been explored in several biomedical research and data showed that H_2 gas is one of the key pathophysiological controlling factors with protective effect on cells and organs using its antiapoptotic and anti-inflammatory properties [3,4]. From patient compliance and convenience perspective, H_2 is highly convenient and have been administered as inhalation, injectables and H_2 rich drink [6].

Preclinical evidence report suggested that H_2 gas administered (up to 4%) as inhalation techniques has been noticeably effective in improving cerebral infarction in rodent model. In addition, treatment with H_2 gas showed significantly greater improvement of neurological outcomes in rodent model of cardiac arrest [7-9]. These findings have been endorsed by several lines of pre-clinical and pilot clinical studies. In addition, efficacy and safety of H_2 gas in various models was evaluated in subjects who had acute cerebral ischemia or post-cardiac arrest [10-12]. Ono et al., reported that H_2 gas treatment was effective and safe in Japanese patients with cerebral infarction [12]. Considering inter-ethnic difference plays a very important role in determine the effect of drug in diseases conditions [10-12]. Thus, it is very important to evaluate efficacy and safety of H_2 gas in China before recommending the use of H_2 gas in Chinese patients with cerebral infarction.

In China, efficacy and safety of hydrogen gas versus standard therapy in Chinese patients with acute cerebral infarction (ACI) is not well established. Therefore, the present preliminary investigation designed to evaluate efficacy and safety of hydrogen gas versus standard therapy in Chinese patients with ACI.

METHODS

Patients and ethics

This pilot study enrolled patients who visited Department of Neurology, Kunshan Hospital of Jiangsu University, Suzhou, China after meeting eligibility criteria. Study duration was March 2021 to Sep 2021. All the eligible patients were randomized in either H_2 gas (3 %) or Standard treatment in allocation ratio of 1:1. The key inclusion criteria were: 1) Chinese patients with ACI who had NIHSS scores of 2 – 6; 2) Patients who had MRI from 0.5 - 3.0 cm lesion for any of image slices inside the region of single major cerebral artery perfusion. Patients were not enrolled if patients had history of severe renal impairment, liver disease, lung disease, and thyroid disease were excluded. Moreover, patients with any other pathology likely to affect the outcome of study, and patients who received concomitant and contra-indicated medications, as well as patients undergoing any other form of surgery. Hydrogen gas was administered twice daily for 1 hour for 2 week using facial mask (non-rebreathing). A blood sample was taken on day 2 to measure H_2 gas using chromatography. Standard treatment includes edaravone 30 mg (IV every 12 hours + antiplatelet drugs for 2 weeks (14 days).

The study was initiated after getting ethical approvals from the institutional ethics committee (ethics committee approval reference number: KHJU/20-21/IEC-KJ-231/23-06), and it was implemented in line with the ethical principles laid down in the Helsinki Declaration and its later amendments.

Treatment and procedure

Subjects who met eligibility criteria were enrolled and received either H_2 gas (3%) or Standard treatment in allocation ratio of 1:1. Standard treatment includes edaravone 30 mg (IV every 12 hours + antiplatelet drugs for 2 weeks (14 days). Patients of H_2 gas group received 3% of H_2 gas for 1 h twice daily for 1 week (7 days) using facial mask (non-rebreathing mask).

Hydrogen gas concentration was measured using gas chromatography Blood sample was taken to measure H_2 gas level in blood using chromatography technique to ensure adequate supply of H_2 gas.

Efficacy and safety assessment

Vital signs such as BP, pulse rate, oxygen levels and body temperature was measured thrice daily. NIHSS score was recorded from each subject before and after treatment. Rehabilitation index using BI, BRS and mRS tool was measured before and after the study drug administration. Blood samples were taken on day 1, 7 and 14 for blood chemistry. Also, MRI of brain taken before and after treatment at day 3, 5, 7, 10 and 14. Infarction site was evaluated using hyperintensity area. Abnormality in MRI scan was observed with size and severity using MRI intensity. Efficacy endpoints includes change in vital signs, change in NIHSS scores from baseline, blood chemistry, change in MRI signal intensity, and improvement in rehabilitation index. Safety was also assessed throughout the study period.

Statistical analysis

Since the present investigation was designed as a pilot study. Thus, there was no formal calculation of sample size. Data for at least 200 evaluable patients were projected for analysis. Numerical category data showing bell shaped curve were analyzed using unpaired t-test, while numerical category data with non-bell shape characteristics were analyzed using Mann Whitney test after normality assessment. Quantitative data are presented as mean ± SD, while categorical data are presented as percentage/proportion of patients, and were analyzed using Fisher exact test or chi-square test based on size of data. Numerical data were subjected for normality test to assess whether data is normal or non-normal. $P < 0.05$ was considered statistically significant.

RESULTS

A total of 215 patients were screened. Of these, a total of 200 patients completed the study and data of these patients were subjected in statistical analysis. Demography and baseline characteristic is presented in Table 1. Demography and baseline characteristic was found to be similar between both the groups.

The changes in NIHSS score (neurological improvement) between both the treatment group is presented in Figure 1. The patients treated

with H2 gas had significantly greater improvement in NIHSS score as compared to patients treated with standard therapy at all days tested except Day 3.

Table 1: Demography and baseline characteristics

Variable	H_2 gas (N=100)	Standard (N=100)	P-value
Age (years)	62.2±14.1	63.2±13.1	0.871
BMI (kg/m²)	26.2±11.6	27.6±13.1	0.55
Gender (M/F)	82/18	78/22	0.46
SBP (mmHg)	121.3±15.3	123.24±14.2	0.32
DBP (mmHg)	89.2±14.12	84.1±14.3	0.88
NIHSS	3.32±12.1	3.42±13.2	0.76
Intensity of MRI	234±23.4	242.3±43.3	0.693
HbA1c	6.62±1.4	6.23±1.3	0.817
Cholesterol	198.2±11.3	206.3±21.3	0.764
Stroke subtype, (%)			
Atherosclerosis	14 (14)	15 (15)	0.23
Embolism	23 (23)	24 (24)	
Occlusion	30 (30)	12 (12)	
Others	12 (12)	14 (14)	
***History*, %**			
Stroke	21 (21)	13 (13)	0.82
Hypertension	12 (12)	15 (15)	
Bleeding	18 (18)	23 (23)	
MI	25 (25)	27 (27)	
***Current treatment* %**			
Statin	21 (21)	20 (20)	0.64
High blood pressure	19 (19)	18 (18)	
Anti-coagulant	11 (11)	12 (12)	
Diabetes	19 (19)	15 (15)	

Values expressed as mean ± SD for numerical variable; percent of patients reported for categorical variables

On day 3, H_2 gas demonstrated numerically higher change in NIHSS score as compared to standard, however, difference was not statistically significant. Change in NIHSS score was significantly higher in patients treated with H_2 gas as compared to standard therapy on Day 5, 7, 9, 11 and 14.
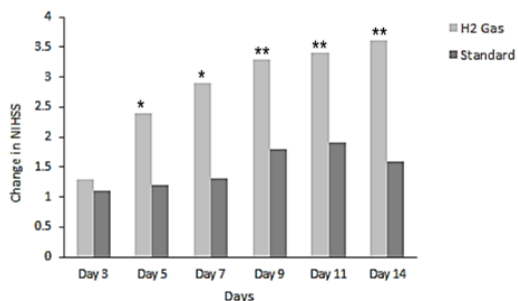
Figure 1: Change in NIHSS. **$P < 0.01$ compared to Standard; *$p < 0.05$ compared to Standard. Otherwise, no statistical significant difference. P value was calculated using Mann-Whitney statistical test

The changes in MRI signal Intensity score between both the treatment group is presented in Figure 2. The patients treated with H_2 gas had significantly greater improvement in MRI signal Intensity score as compared to patients treated with standard therapy at all days tested except Day 1.
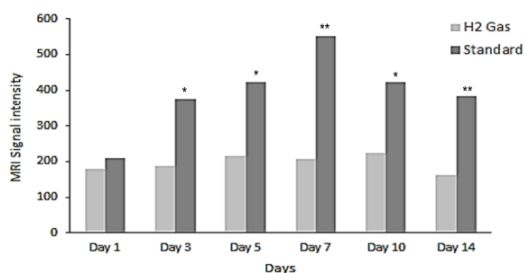

Figure 2: MRI signal Intensity. **$P < 0.01$ compared to Standard; *$p<0.05$ compared to Standard. Otherwise, no statistically significant difference
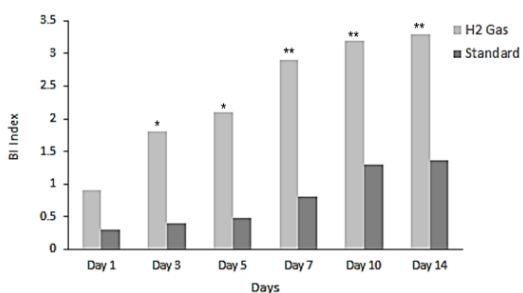

Figure 3: Changes in BI rehabilitation index. **$P < 0.01$ compared to Standard; *$p < 0.05$ compared to Standard. Otherwise, no statistically significant difference. P value was calculated using Mann-Whitney statistical test

On day 1, H_2 gas demonstrated numerically higher change in MRI signal Intensity score as compared to standard, however, difference was not statistically significant. Change in MRI signal Intensity score was significantly higher in patients

treated with H_2 gas as compared to standard therapy on Day 3, 5, 7, 10 and 14.

The changes in BI rehabilitation index is presented in Figure 3. The patients treated with H_2 gas had significantly greater improvement in BI rehabilitation index as compared to the patients treated with standard therapy at all days tested, except Day 1.

On day 1, H_2 gas demonstrated numerically higher change in BI rehabilitation index as compared to standard, however, difference was not statistically significant. Change in BI rehabilitation index was significantly higher in patients treated with H_2 gas as compared to standard therapy on Day 3, 5, 7, 10 and 14. Overall, H_2 gas demonstrated significantly greater improvement in BI rehabilitation index as compared to standard treatment. This indicates that H_2 gas could be a better alternative in the treatment of ACI among Chinese patients.

The changes in BRS rehabilitation index between both the treatment group is presented in Figure 4. The patients treated with H_2 gas had significantly greater improvement in BRS rehabilitation index as compared to the patients treated with standard therapy at all days tested except Day 1.
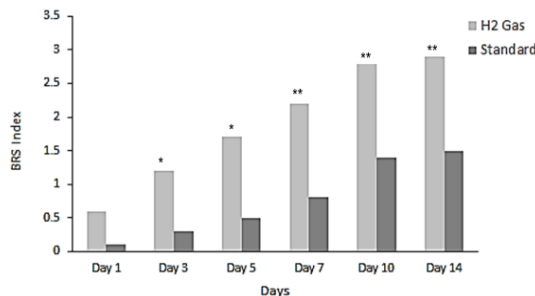

Figure 4: Changes in BRS index. **$P < 0.01$ compared to Standard; *$p < 0.05$ compared to Standard. Otherwise, no statistically significant difference. P value was calculated using Mann-Whitney statistical test

On day 1, H_2 gas demonstrated numerically higher change in BRS rehabilitation index as compared to standard, however, difference was not statistically significant. Change in BRS rehabilitation index was significantly higher in patients treated with H_2 gas as compared to standard therapy on Day 3, 5, 7, 10 and 14. Overall, H_2 gas demonstrated significantly greater improvement in BRS rehabilitation index as compared to standard treatment. This indicates that H_2 gas could be a better alternative in the treatment of ACI among Chinese patients.

The changes in mRS rehabilitation index between both the treatment group is presented in Figure 5. The patients treated with H_2 gas had significantly greater improvement in mRS rehabilitation index as compared to the patients treated with standard therapy at only Day 7, 10 and 14.
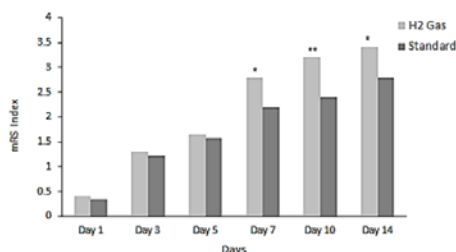

Figure 5: Changes in mRS rehabilitation index. **p<0.01 compared to Standard; *p < 0.05 compared to Standard. Otherwise, no statistically significant difference. *P*-value was calculated using Mann-Whitney statistical test

On day 1, H_2 gas demonstrated numerically higher change in mRS rehabilitation index as compared to standard, however, difference was not statistically significant. Also, similar trend of numerically greater improvement in mRS rehabilitation index was observed on day 3 and day 5. Change in mRS rehabilitation index was significantly higher in patients treated with H_2 gas as compared to standard therapy on Day 7, 10 and 14. Overall, H_2 gas demonstrated significantly greater improvement in mRS rehabilitation index as compared to standard treatment.

The changes in FIM rehabilitation index from baseline between both the treatment group is presented in **Figure 6**. The patients treated with H_2 gas had significantly greater improvement in FIM rehabilitation index as compared to the patients treated with standard therapy at only Day 7, 10 and 14.
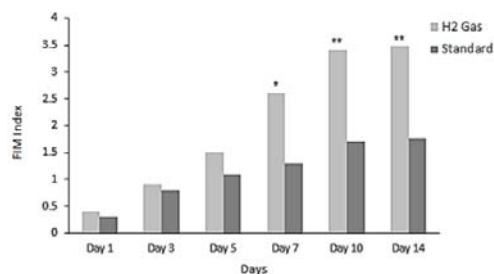

Figure 6: Changes in FIM rehabilitation index from baseline. **p<0.01 compared to Standard; *p<0.05 compared to Standard. Otherwise, no statistically significant difference. P value was calculated using Mann-Whitney statistical test

On day 1, H_2 gas demonstrated numerically higher change in FIM rehabilitation index as compared to standard, however, difference was not statistically significant. Also, similar trend of numerically greater improvement in FIM rehabilitation index was observed on day 3 and day 5. Change in FIM rehabilitation index was significantly higher in patients treated with H_2 gas as compared to standard therapy on Day 7, 10 and 14. Overall, H_2 gas demonstrated significantly greater improvement in FIM rehabilitation index as compared to standard treatment. This indicates that H_2 gas could be a better alternative in the treatment of ACI among Chinese patients.

Vital signs and biochemistry investigation after treatment showed no clinical abnormality in either group. Both the groups were comparable for vital signs parameters except for oxygen saturation level, which was significantly higher in patients treated with H_2 gas as compared to standard therapy.

The oxygen saturation level between both the treatment group is presented in **Figure 7.** The patients treated with H2 gas had significantly greater improvement in oxygen saturation level as compared to the patients treated with standard therapy at all days except Day 1.
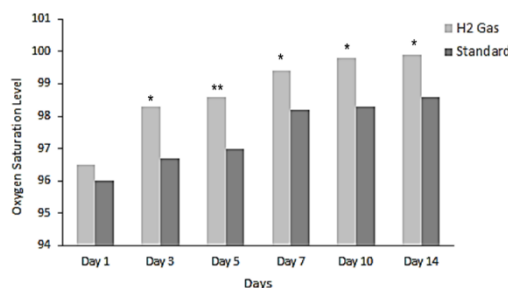

Figure 7: Oxygen saturation level. **P < 0.01 compared to Standard; *p < 0.05 compared to Standard. Otherwise, no statistically significant difference

On day 1, H_2 gas demonstrated numerically higher oxygen saturation level as compared to standard, however, difference was not statistically significant. Change in oxygen saturation level was significantly higher in patients treated with H2 gas as compared to standard therapy on Day 3, 5, 7, 10 and 14. Overall, H_2 gas demonstrated significantly greater improvement in oxygen saturation level as compared to standard treatment.

Both the study drugs were found well tolerated and generally safe. The most common adverse events of H_2 gas were cough and chest distress, that does not require any treatment.

DISCUSSION

This is the first study evaluating efficacy and safety of hydrogen gas versus standard therapy in Chinese patients with ACI. The results of present study showed that the patients treated with H_2 gas had significantly greater improvement in NIHSS score as compared to patients treated with standard therapy. This indicates that the neurological improvement was significantly greater in patients treated with H_2 gas as compared to standard therapy for all days tested. Within group comparison also showed significant improvement in neurological outcome as compared to baseline score. The onset of NIHSS score reduction was faster in patients treated with H_2 gas as compared to standard therapy. Also, H_2 gas demonstrated significant improvement in MRI signal Intensity score, which indicates that H_2 gas administered as inhalation technique was found effective in recovery of infraction site. Since H_2 gas rapidly absorbs so rapid onset of action was observed in recovering the infraction site.

The finding of present study show that recovery started from Day 3 in patients treated with H_2 gas as reduction of vasogenic edema was observed as early as on Day 3. In contrast, the patients treated with standard therapy have noted meaningful effect in reduction of infraction size by day 7. The MRI data suggested that H_2 gas further improves the core etiology of the cerebral infarction. Therefore, it is recommended to initiate H_2 gas treatment during or well before starting intravascular therapy. The MRI results of present study was consistent with the previous studies [10-12]. The possible reason for faster onset of action was due to inhalation technical of H2 gas administration, which absorbs faster and elicit its effect in improving the MRI score.

In the present study, both the treatment showed significant improvement in rehabilitation index as compared to baseline score. However, the patients treated with H_2 gas had significantly greater improvement in rehabilitation index as compared to the patients treated with standard therapy. In this study, rehabilitation index was measured using BI, BRS, mRS and FIM tools. On day 1, H_2 gas demonstrated numerically higher change in rehabilitation index as compared to standard, however, difference was not statistically significant. Change in rehabilitation index was significantly higher in patients treated with H_2 gas as compared to standard therapy on Day 3, 5, 7, 10 and 14. Overall, H_2 gas demonstrated significantly greater improvement in rehabilitation index as compared to standard treatment. This indicates that H_2 gas could be a better alternative in the treatment of ACI among Chinese patients. Moreover, both the groups were comparable for vital signs parameters except for oxygen saturation level, which was significantly higher in patients treated with H_2 gas as compared to standard therapy. In this study, H_2 gas demonstrated numerically higher oxygen saturation level as compared to standard immediately after start of treatment, however, difference was not statistically significant. Change in oxygen saturation level was significantly higher in patients treated with H_2 gas as compared to standard therapy on Day 3, 5, 7, 10 and 14. Overall, H_2 gas demonstrated significantly greater improvement in oxygen saturation level as compared to standard treatment. This indicates that H_2 gas could be a better alternative in the treatment of ACI among Chinese patients.

Overall, the findings of present study showed that H_2 gas is effective and safe as compared to standard therapy in Chinese patients with acute cerebral infarction (ACI). The finding of present study may benefit to scientific community and helps to design large clinical trial to evaluate safety and clinical outcomes of H_2 gas in Chinese patients with acute cerebral infarction (ACI) across globe. Since the present trial was conducted at a single hospital in China, thus, the findings of the present trial can not to be generalized to the Chinese population. Due to lower sample size, the power of trial was less, thus, a large clinical trial with appropriate sample size is needed to confirm the present findings.

CONCLUSION

The findings of this study indicate that H_2 gas administered by inhalation is effective and safe for Chinese patients with acute cerebral infarction. Therefore, H_2 gas therapy has a potential role in the management of acute cerebral infarction, and is considered superior to standard therapy in the management of acute cerebral infarction.

DECLARATIONS

Acknowledgement

Conflict of interest

No conflict of interest is associated with this work.

Contribution of authors

We declare that this work was done by the author(s) named in this article and all liabilities pertaining to claims relating to the content of this article will be borne by the authors. All authors have made substantial contributions to the conception or design of the work; or the acquisition, analysis, or interpretation of data for the work; AND drafted the work or revised it critically for important intellectual content; AND gave final approval of the version to be published; AND agreed to be 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. Wenjuan Yao and Likui Shen contributed to this work equally.

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