Effects of 7-day intake of hydrogen-rich water on physical performance of trained and untrained subjects

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ABSTRACT: Hydrogen-rich water (HRW) is used as a supplement to improve performance and reduce fatigue in athletes. However, the potentially beneficial effects of HRW intake could be mediated by the training status of athletes. The purpose of the study was to analyse the ergogenic effect of intake of HRW for one week on aerobic and anaerobic performance, both in trained and untrained individuals. Thirty-seven volunteers participated in the study and were divided into two experimental groups: trained cyclists and untrained subjects. A double-blind crossover design was performed in which all subjects took a placebo (PW) and nano-bubble HRW (pH: 7.5; hydrogen concentration: 1.9 ppm; oxidation-reduction potential (ORP): -600 mV). At the end of 7-day intake, performance was assessed by an incremental V02max test and by a maximum anaerobic test. After HRW intake, only trained cyclists improved their performance in the anaerobic test with an increase in peak power (from 766.2 ± 125.6 to 826.5 ± 143.4 W; d = .51) and mean power (from 350.0 ± 53.5 to 380.2 ± 71.3 W; d = .51), and a decrease in the fatigue index (from 77.6 ± 5.8 to 75.1 ± 5.9%; d = .45). The findings demonstrate that the ergogenic effect of HRW is mediated by the training status, and that 7-day intake of HRW would be an effective strategy for improving anaerobic performance in trained cyclists.

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INTRODUCTION

It is well known that high intensity exercise causes an increase in reactive oxygen species (ROS), altering the redox balance and increasing oxidative stress [1, 2]. This increase in ROS can cause damage to the cell membrane and dysfunctions in the cellular mitochondria [3], which are associated with increased fatigue and loss of performance [4, 5].

Many athletes ingest buffer substances to increase the antioxidant response and improve the performance of aerobic and anaerobic metabolism. The hydrogen acts as an efficient antioxidant by rapid diffusion into living tissues and cells [6, 7]. Most antioxidant supplements are limited in their cellular distributions, but hydrogen has the ability to effectively penetrate biomembranes and infiltrate into the mitochondria and the nucleus [8, 9]. Hydrogen can be administered by different methods (inhalation of H2 gas or injectable saline solution) but the easiest and safest way is the intake of hydrogen-rich water (HRW), without adding a colour change or water flavour [10].

Recently, some studies have concluded that molecular hydrogen might have healthy effects on the body, inducing antioxidant and anti-inflammatory responses, and limiting the metabolic acidosis [11–13]. HRW supplementation has been shown to have a positive effect on repeated-sprint ability performance, muscle fatigue and ventilatory response [14–17]. Similarly, the inhalation of 4% gaseous hydrogen during a week improved the peak running velocity of physically active men and women [18].

Currently there are different devices on the market that produce HRW, and even bags/bottles of different sizes that contain supposedly hydrogenated water are sold. The electrolysis technology used to enrich the water determines the pH level, the hydrogen concentration and the oxidation-reduction potential (ORP), decisively influencing these factors in terms of the ability to eliminate ROS. Another determining factor to promote the absorption of hydrogen by the cell membrane is the size of the hydrogen bubble. In the same vein, it has been suggested that the antioxidant activity of nano-bubble HRW (≤ 717 ± 387-nm diameter) is superior to normal hydrogen water containing a similar or lower level of dissolved hydrogen but not nano-bubbles [19].

On the other hand, the antioxidant response against the attack of ROS is more effective in trained individuals than in non-trained individuals, since they have a greater antioxidant capacity and a better immune response to exercise [17, 20]. These training-induced adaptations could also influence the ergogenic effect of HRW intake. To the best of our knowledge, there is only one previous study [17]...
that has tried to analyse the positive effect of HRW intake taking into account the athlete performance level, although it was conducted with a small sample size and with an acute intake that was only performed for 24 hours.

Therefore, the main aim of the study was to analyze the ergogenic effect of 7-day intake of nano-bubble HRW on aerobic and anaerobic performance, both in trained amateur cyclists and in untrained subjects. We hypothesized that the intake of HRW may be beneficial for athletes, reducing fatigue and increasing the aerobic and anaerobic performance.

MATERIALS AND METHODS

Participants

A total of 37 volunteers participated in the study. The information of the project was publicized through the bulletin board and the social networks of the research group. The sample size was calculated a priori to obtain a statistical power of 0.8, with a significance level of 0.05, and a mean difference of 5% between groups. Two experimental groups were formed: a group of untrained individuals (n = 15; age: 26.3 ± 5.9 years, body weight: 69.8 ± 11.4 kg, body height: 169.3 ± 7.1 cm, body fat: 24.5 ± 6.5%), who were identified as moderate active individuals according to the International Questionnaire of Physical Activity [21], but who did not perform regular or controlled sports training; and a group of trained amateur cyclists (n = 12; age: 25.5 ± 5.5 years, body weight: 70.9 ± 8.5 kg, body height: 177.3 ± 6.6 cm, body fat: 17.9 ± 5.8%) who met the criteria of having a training background of 1–3 years, with a training duration of 60–120 minutes and a frequency training of 2–3 days/week [22]. All participants were free of musculoskeletal injuries in the lower limbs and they could not ingest any dietary supplement/medicine while the experiment lasted. The purpose of the study and the experimental protocols were explained and written informed consent was obtained from the participants before starting the research. The study was approved by the Ethics Committee of the university and met the requirements of the Declaration of Helsinki (Reg: 108/2018).

Experimental design

A double-blind design crossover was proposed to evaluate the ergogenic effect of 7-day intake of HRW in two different experimental groups: trained amateur cyclists and untrained subjects. The study lasted for a total of 18 days, in which during the first week, all participants ingested a type of water (placebo water [PW] or HRW, according to the randomization), and during the second week, they ingested the other type of water as appropriate according to the crossover design, without a washout period between the two interventions. After each week of water intake, the following two days were used to assess the performance achieved by the participants in an incremental VO₂ max test and in a maximum anaerobic test, separated one test from the other for a period of 24 hours (see Figure 1). Tests were always performed at the same time of the day (± 1 h) and without having performed strenuous lower limb exercises in the preceding 48 h. All tests were carried out under similar atmospheric condition (21–24°C and 45–55% relative humidity).

Administration of HRW

A portable drinking bottle of 320 ml capacity was used to produce nano-bubble HRW (HL-A1, H2Life, Chuanghui electronics, China). This device is able to generate very high level HRW using a proton exchange membrane (PEM) electrolysis technology, with a thick platinum-coated electrolysis plate. In a time period of 3 minutes, this water ionizer allows one to achieve nano-bubble HRW starting from mineral water. HRW is safe for the human body and has no side-effect [23], and this device is certified and in compliance with CE (Europe) and FCC (North America) regulations.

During the study, a portable drinking bottle was provided to all participants. They were instructed to drink water ranging between 1920 and 2240 ml per day (6–7 bottles of PW or HRW, according to the crossover double-blind design), having to drink all the water immediately after the hydrogenation process to maintain the level of hydrogen concentration and avoid deterioration of the ORP. During the PW intake, the portable drinking bottle was similar to the HRW intake, with bubbles but without an electrolysis plate. Moreover, participants could not distinguish between HRW and PW, because HRW was colourless, odourless, and tasteless. The characteristics of the water were as follows: PW (pH: 7.4; hydrogen concentration: 0 ppm and ORP: +241 mV) and HRW (pH: 7.5; hydrogen concentration: 1.9 ppm and ORP: -600 mV). The concentration of hydrogen in water was measured by oxidimetric determination using methylene blue-platinum colloid reagent [24]. Participants were also asked to
minimize the intake of other types of beverages (alcohol, soft drinks, fruit juices, etc).

**Measurements**

Anthropometric measurements were taken at the beginning of the first testing session to characterize the participants. Body height was measured using a portable stadiometer (Seca 213, Germany). Body weight and % body fat were evaluated using a bioelectrical impedance analyser (BF-350, Tanita Europe BV, The Netherlands). Hydration prior to body composition testing was not allowed.

Maximal oxygen uptake (VO\textsubscript{2max}), percentage of VO\textsubscript{2}max in the ventilatory anaerobic threshold (VT2 %VO\textsubscript{2max}), maximal work rate (Wmax), time to exhaustion (TTE) and maximum heart rate (HRmax) were determined using an incremental test to exhaustion on an electromagnetically braked bike potentiometer (Cycle Ops400 pro; Saris Cycling Group; USA). Ventilatory and gas exchange responses were measured continuously using a portable high resolution gas analysis system with breath-by-breath technology (Metalyzer 3b; CORTEX Biophysik GmbH, Germany). After a warm-up of 5 minutes at 50 W, participants started cycling at 100 W, and the work rate was increased by 30 W every 2 minutes until exhaustion. Subjects were required to maintain a cadence of 60–70 rpm. Test ended when the pedalling frequency could not be maintained under a work load. VO\textsubscript{2}max was reached when the oxygen consumption reached its plateau (defined from 1-min mean VO\textsubscript{2} values) [25] and the ventilatory threshold was determined according to the triphasic model developed by Skinner and McLeLLan [26]. Wmax was recorded from the last completed work stage. HRmax was recorded with a heart rate monitor (V800, Polar Electro Oy, Finland).

An anaerobic test adapted from the Quebec 90-second test [27] was also performed to determine peak power (PP), mean power (MP), and fatigue index (FI). Participants performed a maximum test of 90 seconds on an electromagnetically braked bike potentiometer (Cycle Ops400 pro; Saris Cycling Group; USA). Firstly, participants performed a warm-up of 3 minutes at 100 W with a cadence of 60–70 rpm. Subsequently, just before starting the 90-second test, participants were instructed to increase the pedalling frequency progressively, and the resistance of the potentiometer was adjusted (by setting twice the work rate reached by each participant in the incremental VO\textsubscript{2}max test). During the first 20 seconds of the test, the subjects had to maintain a cadence greater than 130 rpm. After these initial seconds, if the subject was not able to maintain a high pedalling cadence (above 100 rpm), the resistance of the potentiometer was gradually lowered (from 30 W to 30 W), in order to maintain the highest power possible throughout the test. PP was determined based on the maximum watts achieved during the first 10 seconds of effort. MP was calculated as the average value of watts recorded throughout the test. FI was defined as the difference between the peak power and minimum power, and expressed as a percentage of the peak power). Heart rate was monitored with a heart rate monitor (V800, Polar Electro Oy, Finland) throughout the test, and HRmax was recorded.

Capillary blood lactate and rated perceived exertion (RPE) were analysed after both tests. Lactate concentration was measured 1 minute after the end of the test, with a fast and reliable portable analyser (Lactate Scout+, SensLab GmbH, Germany) that uses an enzymatic–amperometric detection method and that only requires 0.5 µL of blood. RPE was assessed by a scale reporting options between 1 (extremely easy) and 10 (extremely hard). Thirty minutes after finishing the tests, a copy of this scale was given to the participants and they rated how hard the session had been.

**Statistical analysis**

Statistical analyses were performed using IBM SPSS Statistics for Windows, version 23.0. (IBM Corp., Armonk, USA). The Shapiro-Wilk test was applied in order to verify a normal distribution of data and Levene’s test was used to assess the homogeneity of variance. A two-way ANOVA was used to investigate the main effects and the interaction between the Protocol factor (PW vs HW) and the Training factor (Trained vs Untrained). The significance of simple effect in groups required the use of the SPSS syntax command. Reliability was determined according to the ICC with 95% confidence intervals between the two testing sessions. The effect size (ES) was calculated for all dependent variables using Cohen’s d. The magnitude of effect was classified as trivial (0.25), small (between 0.25 and 0.50), moderate (between 0.50 and 1.0) and large (> 1.0) [28]. The significance level was set at p ≤ 0.05, with a confidence level of 95%. Means and standard deviations (SD) were used as descriptive statistics.

**RESULTS**

All participants completed the study and no person reported any adverse effects that could be related to HRW intake.

Table 1 shows the results obtained in the incremental VO\textsubscript{2}max test by both amateur trained cyclists and untrained subjects. The trained cyclists achieved significantly higher values of VO\textsubscript{2}max, VT2 %VO\textsubscript{2}max, Wmax and TTE in both the PW and HW protocol. An interaction effect (p = .043) on the lactate concentration was observed in trained cyclists after the intake of HRW, although with a small effect size (d = .33). No significant effect was observed in untrained subjects.

Table 2 shows the results obtained in the maximum anaerobic test by participants. The trained cyclists had better values of PP, MP and FI in both PW and HW protocols. It was observed that HRW had an interaction effect with the Training factor, observing a moderate positive effect on PP (d = .51), MP (d = .51) and FI (d = 0.45) in trained cyclists. However, HRW compared with PW did not have a clear effect in untrained subjects.

**DISCUSSION**

The findings of the study show that HRW intake for one week had a positive effect on the performance (PP, MP and FI) of trained cyclists during the anaerobic test, but no significant effect on untrained cyclists.
subjects. Previous studies have shown that the antioxidant capacity of the organism can be influenced by the status training [20, 29], so the magnitude of the effect of the HRW could also be influenced by this variable.

Recently Botek, Krejčí [17] concluded that the ergogenic effect of acute HRW intake on performance in a 4.2-km up-hill race is determined by the athlete performance level, noting that performance improved in the 4 slowest athletes, and remained without significant improvement in the 4 fastest athletes. This result is totally contrary to those obtained in our study, in which we observed that the intake of HRW only had an ergogenic effect in the trained subjects (with a VO\textsubscript{2max} greater than 60 ml/min*kg). It is difficult to contrast the results between the two studies because the protocol used and the HRW were very different. In the aforementioned study, an acute intake of HRW was carried out for a short period of 24 hours before the race (in the present research, a 7-day intake was used), and most decisively, a bottled HRW without nano- or micro-bubbles with an H\textsubscript{2} concentration of only 0.9 ppm was used (in our study, HRW nano-bubbles with a concentration of 1.9 ppm were used). In this vein, the effectiveness of hydrogenated water will depend on the type of HRW used, the way of administration and the duration of the intervention.

The performance improvement in trained cyclists after HRW ingestion occurred with a moderate magnitude of the effect during the

| Subjects | Protocol | Mean ± SD | 95%CI | ICC | Effect size (d) | Main effect Protocol (p-value) | Main effect Training (p-value) | Interaction effect Training x Protocol (p-value) |
|----------|----------|-----------|-------|-----|----------------|-------------------------------|-------------------------------|---------------------------------|
| VO\textsubscript{2max} (ml/min/Kg) | Trained | PW | 61.1 ± 3.8 | 58.1–63.8 | .97 | .18 | .742 | .001 | .794 |
| | | HW | 60.5 ± 3.2 | 57.3–62.2 | .98 | .13 | | | |
| | Trained | PW | 39.7 ± 7.6 | 35.4–43.9 | .82 | .41 | .807 | .001 | .854 |
| | | HW | 40.7 ± 8.3 | 36.4–45.1 | | | | | |
| VT2 %VO\textsubscript{2max} | Trained | PW | 82.9 ± 3.7 | 80.9–84.9 | .69 | .35 | | | |
| | | HW | 81.3 ± 4.4 | 78.2–84.1 | | | | | |
| Wmax (Watts) | Trained | PW | 290.0 ± 51.9 | 260.0–323.3 | .96 | .04 | .557 | .001 | .957 |
| | | HW | 300.0 ± 51.9 | 270.0–333.3 | | | | | |
| | | PW | 223.2 ± 49.6 | 196.9–250.0 | | | | | |
| | | HW | 224.6 ± 50.2 | 199.2–254.6 | | | | | |
| HRmax (bpm) | Trained | PW | 185.4 ± 12.0 | 177.8–192.4 | .98 | .21 | .886 | .928 | .653 |
| | | HW | 186.2 ± 8.6 | 181.0–191.5 | | | | | |
| | | PW | 184.6 ± 15.7 | 176.3–192.1 | | | | | |
| | | HW | 183.4 ± 15.2 | 175.8–191.8 | | | | | |
| TTE (s) | Trained | PW | 890.0 ± 212.1 | 770.0–1030.0 | .98 | .10 | .859 | .01 | .861 |
| | | HW | 880.0 ± 207.8 | 760.0–1013.3 | | | | | |
| | | PW | 567.6 ± 163.6 | 484.6–660.0 | | | | | |
| | | HW | 583.8 ± 176.8 | 491.5–685.3 | | | | | |
| Lactate (mmol/L) | Trained | PW | 12.2 ± 3.2 | 10.3–14.2 | .96 | .33 | .511 | .495 | .043 |
| | | HW | 11.1 ± 3.9* | 8.8–13.5 | | | | | |
| | | PW | 12.9 ± 3.0 | 11.3–14.5 | | | | | |
| | | HW | 12.4 ± 3.8 | 10.5–14.5 | | | | | |
| RPE | Trained | PW | 8.2 ± 0.9 | 7.6–8.8 | .47 | .26 | .833 | .500 | .339 |
| | | HW | 8.4 ± 0.7 | 8.0–8.9 | | | | | |
| | | PW | 8.3 ± 0.6 | 8.0–8.6 | | | | | |
| | | HW | 8.0 ± 1.0 | 7.6–8.4 | | | | | |

Note: * Significance of simple effect between PW and HW in the group of trained cyclists. PW: Placebo water; HW: Hydrogen rich water; VT2 %VO\textsubscript{2max}: Percentage of maximal oxygen uptake in the ventilatory anaerobic threshold; HR: Heart rate; TTE: Time to exhaustion; RPE: Rated perceived exertion.
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TABLE 2. Values obtained during the maximal anaerobic test

| Variables | Training status | Protocol | Mean ± SD | 95% CI | ICC | Effect size (d) | Main effect Protocol (p-value) | Main effect Training (p-value) | Interaction effect Training x Protocol (p-value) |
|-----------|-----------------|----------|-----------|--------|-----|----------------|-------------------------------|-------------------------------|---------------------------------|
| Peak Power (Watts) | Trained | PW | 766.2 ± 125.6 | 691.4–845.2 | .97 | .51 | .484 | .001 | .046 |
| | Trained | HW | 826.5 ± 143.4* | 743.2–916.3 | .77 | .08 | | | |
| | Untrained | PW | 625.6 ± 145.9 | 549.9–706.9 | | | | | |
| | Untrained | HW | 613.6 ± 154.4 | 529.7–696.7 | | | | | |
| Mean Power (Watts) | Trained | PW | 350.0 ± 53.5 | 319.2–383.6 | .94 | .51 | .303 | .001 | .047 |
| | Trained | HW | 380.2 ± 71.3* | 342.3–426.9 | | | | | |
| | Untrained | PW | 272.6 ± 78.3 | 233.5–313.5 | .68 | .08 | | | |
| | Untrained | HW | 277.0 ± 53.7 | 247.7–307.7 | | | | | |
| FI (%) | Trained | PW | 77.6 ± 5.8 | 73.8–80.9 | .92 | .45 | .732 | .018 | .049 |
| | Trained | HW | 75.1 ± 5.9* | 71.4–78.5 | | | | | |
| | Untrained | PW | 80.6 ± 8.9 | 76.3–85.1 | .91 | .10 | | | |
| | Untrained | HW | 81.4 ± 9.6 | 76.9–86.5 | | | | | |
| HRmax (bpm) | Trained | PW | 179.6 ± 10.7 | 172.2–185.6 | .93 | .00 | .866 | .631 | .703 |
| | Trained | HW | 179.6 ± 12.0 | 171.8–186.8 | | | | | |
| | Untrained | PW | 175.9 ± 26.0 | 159.8–186.8 | .91 | .13 | | | |
| | Untrained | HW | 172.5 ± 39.4 | 148.1–190.0 | | | | | |
| Lactate (mmol/L) | Trained | PW | 13.0 ± 3.4 | 10.8–15.0 | .91 | .22 | .773 | .852 | .265 |
| | Trained | HW | 12.2 ± 4.4 | 9.4–14.7 | | | | | |
| | Untrained | PW | 12.6 ± 2.4 | 11.3–13.8 | .49 | .25 | | | |
| | Untrained | HW | 12.1 ± 1.7 | 11.2–13.0 | | | | | |
| RPE | Trained | PW | 8.3 ± 1.4 | 7.5–9.2 | .69 | .43 | .264 | .227 | .178 |
| | Trained | HW | 8.8 ± 0.9 | 8.4–9.3 | | | | | |
| | Untrained | PW | 8.6 ± 1.0 | 8.1–9.1 | .49 | .24 | | | |
| | Untrained | HW | 8.8 ± 0.7 | 8.4–9.2 | | | | | |

Note: *. Significance of simple effect between PW and HW in the group of trained cyclists. PW: Placebo water; HW: Hydrogen rich water; FI: Fatigue index; HR: Heart rate; RPE: Rated perceived exertion.
The beneficial effects of HRW are mediated by the training status of athletes. Seven-day intake of nano-bubble HRW improved anaerobic performance of trained cyclists, but had no effect on untrained subjects. Therefore, the HRW intake seems to be an effective hydration strategy, although a greater number of studies are necessary to define precisely the duration of intake, the amount of water and the most effective concentration of hydrogen to individually optimize the ergogenic effects of HRW.

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**Conflict of interest declaration**

All authors declare no conflicts of interest.

**REFERENCES**

1. Tanskanen M, Atalay M, Usitalo A. Altered oxidative stress in overtrained athletes. J Sports Sci. 2010; 28(3):309–317. doi:10.1080/02640410903473844

2. Magherini F, Fiaschi T, Marzocchini R, Mannelli M, Gamberi T, Modesti P, et al. Oxidative stress in exercise training: the involvement of inflammation and peripheral signals. Free Radic Res. 2019; 53(11–12):1155–1165. doi:10.1080/10717544.2019.1697438

3. Carri MT, Valle C, Bozzo F, Cozzolino M. Oxidative stress and mitochondrial damage: importance in non-SOD1 ALS. Front Cell Neurosci. 2015;9:41. doi:10.3389/fncel.2015.00041

4. Finaud J, Lac G, Filarei E. Oxidative stress – Relationship with exercise and training. Sports Med. 2006; 36(4):327–358. doi:10.2165/00007256-200636040-00004

5. Powers SK, Jackson MJ. Exercise-induced oxidative stress: cellular mechanisms and impact on muscle force production. Physiol Rev. 2008; 88(4):1243–76. doi:10.1152/physrev.00031.2007

6. Ishibashi T. Molecular hydrogen: new antioxidant and anti-inflammatory therapy for rheumatoid arthritis and related diseases. Curr Pharm Des. 2013; 19(35):6375–81. doi:10.2174/138161213800000057

7. 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. doi:10.1038/nm1577

8. Ohta S. Molecular hydrogen as a novel antioxidant: overview of the advantages of hydrogen for medical applications. Methods Enzymol. 2015;555:289–317. doi:10.1016/bs.mie.2014.11.038

9. Nicolson G, De Mattos G, Settineri R, Costa C, Elithorpe R, Rosenblatt S, et al. Clinical effects of hydrogen administration: from animal and human diseases to exercise medicine. Int J Clin Med. 2016;7(1):32–76.

10. Ohta S. Recent Progress Toward Hydrogen Medicine: Potential of Molecular Hydrogen for Preventive and Therapeutic Applications. Curr Pharm Des. 2011;17(22):2241–2252. doi:10.2174/138161211797052664

11. Ishibashi T, Sato B, Rikitake M, Seo T, Kurokawa R, Hara Y, et al. Consumption of water containing a high concentration of molecular hydrogen reduces oxidative stress and disease activity in patients with rheumatoid arthritis: an open-label pilot study. Med Gas Res. 2012; 2(1):27. doi:10.1186/2045-9912-2-27

12. LeBaron T, Singh R, Fatima G, Kartikey K, Sharma J, Ostojic S, et al. The Effects of 24-Week, High-Concentration Hydrogen-Rich Water on Body Composition, Blood Lipid Profiles and Inflammation Biomarkers in Men and Women with Metabolic Syndrome: A Randomized Controlled Trial. Diabetes Metab Syndr Obes. 2020;13:889–896. doi:10.2147/DMSO.S240122

13. Ostojic S, Stojanovic M. Hydrogen-Rich Water Affect ed Blood Alkalinity in Physically Active Men. Res Sports Med. 2014;22(1):49–60. doi:10.1080/15438627.2013.852092

14. Aoki K, Nakao A, Adachi T, Matsui Y, Miyakawa S. Pilot study: Effects of drinking hydrogen-rich water on muscle fatigue caused by acute exercise in elite athletes. Med Gas Res. 2012; 2:12. doi:10.1186/2045-9912-2-12

15. Da Ponte A, Giovanelli N, Nigris D, Lazzer S. Effects of hydrogen rich water on prolonged intermittent exercise. J Sport Med Phys Fit. 2018; 58(5):612–621. doi:10.23736/ S0022-4707.17.06883-9

16. Botek M, Krejci J, McKune A, Sladeckova B, Naumovski N. Hydrogen Rich Water Improved Ventilatory, Perceptual and Lactate Responses to Exercise. Int J Sports Med. 2019; 40(14):879–885. doi:10.1055/a-0991-0268

17. Botek M, Krejci J, McKune AJ, Sladečková B. Hydrogen-Rich Water Supplementation and Up-Hill Running Performance: Effect of Athlete Performance Level. Int J Sports Physiol Perform. 2020;1–4. doi:10.1123/ijpp.2019-0507

18. Javorac D, Stajer V, Ratgeber L, Betlehem J, Ostojic S. Short-term H2 inhalation improves running performance and torso strength in healthy adults. Biol Sport. 2019;36(4):333–339. doi:10.5114/biolsp.2019.88756

19. Kato S, Matsuoka D, Miwa N. Antioxidant activities of nano-bubble hydrogen-dissolved water assessed by ESR and 2,2’-bipyridyl methods. Mater Sci Eng C Mater Biol Appl. 2015; 53:7–10. doi:10.1016/j.msec. 2015.03.064
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20. Koltai E, Bori Z, OsATH P, Ihasz F, Peter S, Toth G, et al. Master athletes have higher miR-7, SIRT3 and SOD2 expression in skeletal muscle than age-matched sedentary controls. Redox Biol. 2018;19:46–51. doi:10.1016/j.redox.2018.07.022
21. Craig CL, Marshall AL, Sjöström M, Bauman AE, Booth ML, Ainsworth BE, et al. International physical activity questionnaire: 12-country reliability and validity. Med Sci Sports Exerc. 2003;35(8):1381–95. doi:10.1249/01.MSS.0000078924.61453.FB
22. Jeukendrup AE, Craig NP, Hawley JA. The bioenergetics of World Class Cycling. J Sci Med Sport. 2000;3(4):414–33. doi:10.1016/s1440-2440(00)80008-0
23. Saitoh Y, Harata Y, Mizuhashi F, Nakajima M, Miwa N. Biological safety of neutral-pH hydrogen-enriched electrolyzed water upon mutagenicity, genotoxicity and subchronic oral toxicity. Toxicol Ind Health. 2010; 26(4):203–216. doi:10.1177/0748233710362989
24. Seo T, Kurokawa R, Sato B. A convenient method for determining the concentration of hydrogen in water: use of methylene blue with colloidal platinum. Med Gas Res. 2012;2:1. doi:10.1186/2045-9912-2-1
25. Lucia A, Rabadam M, Hoyos J, Hernandez-Capillaz M, Perez M, San Juan A, et al. Frequency of the VO2max plateau phenomenon in world-class cyclists. Int J Sports Med. 2006;27(12):984–992. doi:10.1055/s-2006-923833
26. Skinner JS, McLellan TM, McLellan TH. The transition from aerobic to anaerobic metabolism. Res Q Exerc Sport. 1980;51(1):234–48. doi:10.1080/02701367.1980.10609285
27. Simoneau JA, Lortie G, Bouray MR, Bouchard C. Tests of anaerobic alactacid and lactacid capacities: description and reliability. Can J Appl Sport Sci. 1983;8(4):266–70.
28. Rhea MR. Determining the magnitude of treatment effects in strength training research through the use of the effect size. J Strength Cond Res. 2004;18(4):918–920. doi: 10.1519/14403.1
29. Clarkson P, Thompson H. Antioxidants: what role do they play in physical activity and health?. Am J Clin Nutr. 2000;72(2):637S-646S. doi:10.1093/ajcn/72.2.637s
30. Vidal K, Robinson N, Ives SJ. Exercise performance and physiological responses: the potential role of redox imbalance. Physiol Rep Apr 2017; 5(7):doi:10.14814/phy2.13225
31. Dobashi S, Takeuchi K, Koyama K. Hydrogen-rich water suppresses the reduction in blood total antioxidant capacity induced by 3 consecutive days of severe exercise in physically active males. Med Gas Res. 2020; 10(1):21–26. doi:10.4103/2045-9912.279979
32. Chen H, Xie K, Han H, Li Y, Liu L, Yang T, et al. Molecular hydrogen protects mice against polymicrobial sepsis by ameliorating endothelial dysfunction via an Nrf2/HO-1 signaling pathway. Int Immunopharmacol. 2015;28:643–654. doi: 10.1016/j.intimp.2015.07.034
33. Ostojic SM. Does H2 alter mitochondrial bioenergetics via GHS-R1α activation? Theranostics. 2017;7:1330–1332. doi:10.7150/thno.18745
34. McLellan CT, McLellan CT. Does H2 alter mitochondrial bioenergetics via GHS-R1α activation? Theranostics. 2017;7:1330–1332. doi:10.7150/thno.18745
35. Ostojic SM. Does H2 alter mitochondrial bioenergetics via GHS-R1α activation? Theranostics. 2017;7:1330–1332. doi:10.7150/thno.18745
36. Sondergaard E, Gormsen LC, Nellermann B, Vestergaard ET, Christiansen JS, Nielsen S. Visceral fat mass is a strong predictor of circulating ghrelin levels in premenopausal women. Eur J Endocrinol. 2009;160(3):375–379. doi: 10.1530/EJE-08-0735.