

Hydrogen-Rich Water Supplementation and Up-Hill Running Performance: Effect of Athlete Performance Level

Michal Botek, Jakub Krejčí, Andrew J. McKune, and Barbora Sládečková

Purpose: Hydrogen-rich water (HRW) has been shown to have an antifatigue effect. This study assessed up-hill running performance, as well as physiological and perceptual responses after supplementation with 1680 mL HRW between 24 h and 40 min before running, in athletes of heterogeneous running ability. **Methods:** Sixteen males (mean [SD] age 31.6 [8.6] y, VO_2max 57.2 [8.9] $\text{mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$, body fat 13.4% [4.4%]) participated in this study. Using a randomized, double-blind, placebo-controlled crossover design, participants consumed either HRW or placebo prior to performing two 4.2-km up-hill races separated by a week. Race time (RT), average race heart rate, and immediately postrace rating of perceived exertion were assessed. **Results:** After analysis of data for all runners, HRW effect was unclear (−10 to 7 s, 90% confidence interval) for RT, likely trivial for heart rate (−2 to 3 $\text{beats}\cdot\text{min}^{-1}$), and likely trivial for postrace rating of perceived exertion (−0.1 to 1.0). A possible negative correlation was found between RT differences and average RT ($r = -.79$ to $-.15$). HRW for the 4 slowest runners (RT = 1490 [91] s) likely improved the RT (−36 to −3 s), whereas for the 4 fastest runners (RT = 1069 [53] s) the performance effect of HRW was unclear (−10 to 26 s). **Conclusions:** HRW intake had an unclear antifatigue effect on performance in terms of mean group values. However, it appears that the magnitude of the antifatigue effect of HRW on performance depends on individual running ability.

Keywords: molecular hydrogen, antioxidant, antifatigue, exercise, field testing

During exercise, reactive oxygen species and reactive nitrogen species are produced by mitochondrial and nonmitochondrial sources, reflecting oxidative stress, that promote adaptation to exercise.¹ However, high levels of reactive oxygen species and reactive nitrogen species have been associated with mitochondrial dysfunction and cellular damage² that may contribute to fatigue and delayed recovery in athletes.³ Molecular hydrogen (H_2) has been shown to be a strong and selective antioxidant with high scavenger affinity toward cytotoxic hydroxyl free radicals, thus aiding in maintaining cellular redox balance,⁴ and has a stimulating effect on mitochondrial oxidative phosphorylation.⁵ Supplementation with hydrogen-rich water (HRW) before exercise has been shown to improve lactate, ventilatory, and perceptual response⁶ as well as have an antifatigue effect, particularly in endurance, strength, and repeated-sprint ability performance.^{7–9} However, athletes display better immunological responses to exercise,¹⁰ endogenous antioxidant capacity, more efficient mitochondrial function, and adenosine

triphosphate (ATP) production compared with sedentary or less-active individuals.¹¹ These training-induced adaptations may potentially alter performance advantages of HRW supplementation in athletes with different abilities. Therefore, the primary aim of this study was to assess physiological, perceptual, and performance responses to an up-hill running race after administration of HRW in a heterogeneous group of athletes.

Methods

Participants

A total of 16 male athletes (mean [SD]; age 31.6 [8.6] y, body mass 71.5 [8.8] kg, body height 177.0 [7.2] cm, body fat 13.4% [4.4%], VO_2max 57.2 [8.9] $\text{mL}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$) volunteered for this study. They followed instructions to avoid using dietary supplements (including sport drinks and coffee) and maintain the same individually prescribed training load from 1 week before the first run until completion of the second run, including during the washout period. Athletes performed 4 to 6 training sessions/week with training session duration ranging from 45 to 140 minutes. One day before each race, all runners trained; both session rating of perceived exertion (RPE; HRW: 11.9 [1.5], placebo: 11.8 [1.2], $P = .55$, Wilcoxon test) and duration (HRW: 47 [9] min, placebo: 48 [10] min, $P = .47$, Wilcoxon test) were not significantly different. Two days before each race, 5 runners trained with no difference in session RPE (HRW: 9.6 [1.7], placebo: 10.0 [1.4], $P = .63$) or duration (HRW: 44 [10] min, placebo: 43 [7] min, $P = .69$). Participants provided informed consent, and the study was approved by the ethics committee of Faculty of Physical Culture, Palacký University Olomouc.

Using a randomized, double-blind, placebo-controlled crossover design, participants consumed either HRW or placebo prior to performing two, 4.2-km up-hill races (same asphalt road, 215-m

© 2020 The Authors. Published by Human Kinetics, Inc. This is an Open Access article distributed under the terms of the Creative Commons Attribution-NonCommercial-NoDerivatives 4.0 International License, CC BY-NC-ND 4.0, which permits the copy and redistribution in any medium or format, provided it is not used for commercial purposes, no modifications are made, appropriate credit is given, and a link to the license is provided. See <http://creativecommons.org/licenses/by-nc-nd/4.0>. This license does not cover any third-party material that may appear with permission in the article. For commercial use, permission should be requested from Human Kinetics, Inc, through the Copyright Clearance Center (<http://www.copyright.com>).

Botek, Krejčí, and Sládečková are with the Faculty of Physical Culture, Palacký University Olomouc, Olomouc, Czech Republic. McKune is with the Faculty of Health, UC-Research Inst for Sport and Exercise, University of Canberra, Canberra, ACT, Australia, and the Discipline of Biokinetics, Exercise and Leisure Sciences, School of Health Sciences, University of KwaZulu-Natal, Durban, South Africa. Krejčí (jakub.krejci@upol.cz) is corresponding author.

elevation, race start at 5:00 PM, and environmental temperature 20–24°C). A total volume of 1680 mL of HRW (Aquistamina HRW; Nutristamina s.r.o., Ostrava, Czech Republic) or placebo (Aquistamina H₂ free; Nutristamina s.r.o.) was administered in four, 420-mL doses at 24 hours, 3 hours, 2 hours, and 40 minutes before the races that were separated by a 1-week washout period.⁷ Both drinks were served in visually identical plastic aluminum packages. Athletes could not distinguish between HRW and placebo, because H₂ is colorless, odorless, and tasteless. HRW/placebo characteristics were as follows: pH = 7.8/7.6 and dissolved H₂ = 0.9/0.0 ppm.

Athletes started each race separated by 2-minute intervals (order randomized), with race time (RT) measured manually using a digital timer (HS80; Casio, Shibuya, Japan). Average race heart rate (HR, V800; Polar Electro Oy, Kempele, Finland) and an immediately post-race RPE (6–20 points) were recorded.

Statistical Analysis

Data are expressed as mean (SD) unless otherwise stated. Differences between HRW and placebo values were checked for normality using the Kolmogorov–Smirnov test and changes in means were evaluated using a paired *t* test. Correlations between HRW and placebo differences and pooled values of RT, $(RT_{HRW} + RT_{Pla})/2$, were evaluated using Pearson correlation coefficient. Subgroups of the 4 slowest runners and 4 fastest runners were selected from the whole sample of 16 runners. The effect of HRW compared with placebo in each subgroup was evaluated again using the paired *t* test. The outcomes were interpreted using magnitude-based inference.¹² The smallest worthwhile change for RT and HR was set at 0.3 and 0.5, respectively, of the within-individual SD. The smallest worthwhile change for RPE was set to 1.0. Only a large (≥ 0.5) correlation coefficient was considered meaningful, so the smallest worthwhile change was set to 0.5.

Results

The data were normally distributed (RT: $P = .41$, HR: $P = .35$, RPE: $P = .28$). After analyzing the whole sample, HRW compared with placebo had an unclear effect on RT and a likely trivial effect on both HR and RPE (Table 1). However, a possible negative correlation was found between RT_{HRW} and RT_{Pla} differences and pooled RT (Figure 1). Correlation between HR_{HRW} and HR_{Pla} differences and pooled RT was possibly trivial ($r = .47$; 90% confidence interval, .05 to .75; $P = .07$; chances 44/56/0). Correlation between RPE_{HRW} and RPE_{Pla} differences and pooled RT was likely trivial ($r = -.14$; 90% confidence interval, $-.53$ to $.31$; $P = .61$, chances 1/92/7). Analysis of the subgroup of the 4 slowest runners (RT = 1490 [91] s) revealed that HRW likely improved RT, likely increased HR, and effect on RPE was likely trivial (Table 2). In the subgroup of the 4 fastest runners (RT = 1069 [53] s), the effect of HRW on both RT and HR was unclear and effect on RPE was likely trivial (Table 2).

Discussion

Although HRW has been associated with an antifatigue effect in various modes of exercise,^{7–9} we found an unclear effect of preexercise HRW intake on performance in terms of mean group values. However, the analyses suggest that the performance enhancing (antifatigue) effect of HRW may depend on the performance ability of athletes, because a possible negative correlation ($r = -.54$) between RT differences and pooled RT was found. Specifically, prerace hydration with 1680-mL HRW compared with placebo likely improved endurance running performance by 1.3% in the slowest runners, whereas the effect of HRW on race performance in the fastest runners was unclear (deterioration by 0.8%). Furthermore, the increase in performance was accompanied by a likely increase in average race HR by 3.8% in the slowest runners, whereas in the fastest runners there was an unclear change (0.1%). HRW administration in the slowest runners had a likely trivial effect on post-race RPE, suggesting that increased race intensity was not accompanied by increased perceived effort. These findings are in line with a recently published study in which perceptual strain at an exercise intensity of 4 W·kg⁻¹ for 8 minutes was lower after acute preexercise HRW administration compared with placebo.⁶

An antifatigue effect of HRW ingestion (2 L·d⁻¹ for 2-wk preexercise) during intermittent cycling was also reported by Da Ponte et al,⁹ who showed a 7.4% attenuation in the decline of peak power output from the sixth to the ninth of 10 sprints. Similarly, Aoki et al⁷ demonstrated an attenuated decrease (3.7%) in peak torque and postexercise lactate level after 20 isokinetic knee

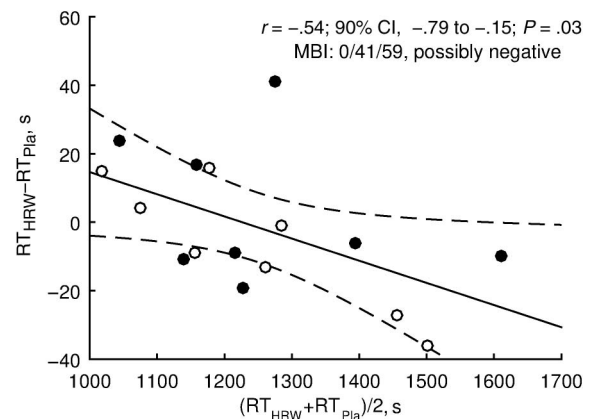


Figure 1 — Correlation analysis between RT differences and pooled RTs. CI indicates confidence interval; HRW, hydrogen-rich water; MBI, magnitude-based inference; RT, race time; RT_{HRW} , RT when HRW was administered; RT_{Pla} , RT when placebo was administered. Filled circles indicate runners who received HRW in the first race; open circles, runners who received HRW in the second race. Dashed lines denote 90% CI.

Table 1 Effect of HRW Compared With Placebo on Race Time, Heart Rate, and RPE in the Sample of 16 Runners

	HRW	Placebo	HRW–Placebo	90% CI	<i>P</i>	Chances ^a +/trivial/–	Inference
Race time, s	1249 (163)	1250 (173)	–1 (20)	–10 to 7	.77	8/73/19	Unclear
Heart rate, beats·min ⁻¹	178 (7)	178 (10)	0 (6)	–2 to 3	.80	5/93/2	Likely trivial
RPE, points	17.8 (1.2)	17.4 (1.3)	0.4 (1.3)	–0.1 to 1.0	.20	5/95/0	Likely trivial

Abbreviations: CI, confidence interval; HRW, hydrogen-rich water; RPE, rating of perceived exertion.

^a Chances that the true value of HRW effect is substantially positive, trivial, or substantially negative.

Table 2 Effect of HRW Compared With Placebo on Race Time, Heart Rate, and RPE in the 4 Fastest Runners and the 4 Slowest Runners

	Group	HRW–Placebo	90% CI	RC (90% CI)	P	Chances ^a +/trivial/–	Inference
Race time, s	Fastest	8 (15)	–10 to 26	0.8 (–0.9 to 2.5)	.37	66/24/10	Unclear
	Slowest	–20 (14)	–36 to –3	–1.3 (–2.4 to –0.2)	.07	2/4/94	Likely negative
Heart rate, beats·min ^{–1}	Fastest	0 (2)	–2 to 3	0.1 (–1.2 to 1.5)	.82	25/60/15	Unclear
	Slowest	6 (7)	–2 to 14	3.8 (–1.0 to 8.5)	.16	78/19/3	Likely positive
RPE, points	Fastest	0.3 (1.0)	–0.9 to 1.4	1.5 (–5.0 to 8.1)	.64	11/85/4	Likely trivial
	Slowest	0.0 (0.8)	–1.0 to 1.0	0.1 (–5.9 to 6.1)	>.99	5/90/5	Likely trivial

Abbreviations: CI, confidence interval; HRW, hydrogen-rich water; RC, relative change in percentage; RPE, rating of perceived exertion.

^a Chances that the true value of HRW effect is substantially positive, trivial, or substantially negative.

extensions following HRW ingestion (1.5-L HRW within 8-h preexercise). Because of the reduced exercise-induced muscle fatigue and lactate lowering effects of H₂, HRW ingestion was suggested to be suitable and beneficial for athletes.⁷ Unfortunately, there is still limited information about the dose–response as well as appropriate chemical characteristics of HRW, particularly of dissolved H₂ concentration and pH.

In an animal study, Ara et al,⁸ who used a comparable technology for HRW preparation to the present study, found that ad libitum HRW intake over 4 weeks, in chronically forced exercise mice, led to a 2.7-fold increase in swimming time to exhaustion compared with the placebo group. The authors suggested that HRW exerted antifatigue effects, mediated via enhanced metabolic coordination and immune redox balance, specifically through increased liver glycogen storage; lactate dehydrogenase and glutathione peroxidase activity; and reduction of interleukin-6, interleukin-17, and tumor necrosis factor- α .⁸ The present results suggest that the faster runners had reduced sensitivity to the performance-enhancing effects of HRW. We propose that faster runners compared with slower runners will probably exhibit lower or even negligible performance benefits from acute HRW administration, possibly due to existing training-induced upregulation of antioxidative,¹¹ metabolic,¹¹ and/or immune systems.¹⁰

Limitations of the study include that ventilatory and metabolic responses as well as changes in participant antioxidative capacity were not measured. These variables may have improved the understanding of mechanisms underlying our results. Other limitations are that the dose of H₂ was not adjusted for body mass and the low sample size.

Practical Applications

Athlete endurance ability determines the effectiveness of preperformance HRW supplementation and/or athlete individual sensitivity to H₂ exposure. Therefore, it is important to evaluate individual responsiveness. This finding should be considered when designing future studies assessing the HRW effect on athletes.

Conclusion

Preexercise HRW intake had an unclear antifatigue effect on performance in terms of mean group values. However, it appears that the magnitude of the effect of prerace HRW supplementation on up-hill running performance depended on individual running ability.

Acknowledgments

This work was supported by grant IGA_FTK_2019_002 from Palacký University Olomouc titled Effect of HRW on Body Response During Laboratory Tests in Elite Athletes. The authors would like to thank Robert Krupička for helping to collect data during testing.

References

- Merry TL, Ristow M. Do antioxidant supplements interfere with skeletal muscle adaptation to exercise training? *J Physiol*. 2016;594:5135–5147. PubMed ID: 26638792 doi:10.1113/JP270654
- 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
- Powers SK, Jackson MJ. Exercise-induced oxidative stress: cellular mechanisms and impact on muscle force production. *Physiol Rev*. 2008;88:1243–1276. PubMed ID: 18923182 doi:10.1152/physrev.00031.2007
- 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. PubMed ID: 17486089 doi:10.1038/nm1577
- Murakami Y, Ito M, Ohsawa I. Molecular hydrogen protects against oxidative stress-induced SH-SY5Y neuroblastoma cell death through the process of mitohormesis. *PLoS One*. 2017;12:e0176992. PubMed ID: 28467497 doi:10.1371/journal.pone.0176992
- Botek M, Krejčí J, McKune AJ, Sládečková B, Naumovski N. Hydrogen-rich water improved ventilatory, perceptual and lactate responses to exercise. *Int J Sports Med*. 2019;40(14):879–885. PubMed ID: 31574544 doi:10.1055/a-0991-0268
- 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. PubMed ID: 22520831 doi:10.1186/2045-9912-2-12
- Ara J, Fadriuela 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. PubMed ID: 29850492 doi:10.1155/2018/2571269
- Da Ponte A, Giovanelli N, Nigris D, Lazzar S. Effects of hydrogen rich water on prolonged intermittent exercise. *J Sports Med Phys Fitness*. 2018;58:612–621. PubMed ID: 28474871
- Alack K, Krüger K, Weiss A, et al. Aerobic endurance training status affects lymphocyte apoptosis sensitivity by induction of molecular genetic adaptations. *Brain Behav Immun*. 2019;75:251–257. PubMed ID: 30790541 doi:10.1016/j.bbi.2018.10.001

11. Koltai E, Bori Z, Osvath P, 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. PubMed ID: [30107294](https://pubmed.ncbi.nlm.nih.gov/30107294/) doi:[10.1016/j.redox.2018.07.022](https://doi.org/10.1016/j.redox.2018.07.022)
12. Batterham AM, Hopkins WG. Making meaningful inferences about magnitudes. *Int J Sports Physiol Perform.* 2006;1:50–57. PubMed ID: [19114737](https://pubmed.ncbi.nlm.nih.gov/19114737/) doi:[10.1123/ijsp.1.1.50](https://doi.org/10.1123/ijsp.1.1.50)