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**Article Title:** NO DOSE-RESPONSE EFFECT OF CARBOHYDRATE MOUTH RINSE CONCENTRATION ON 5 KM RUNNING PERFORMANCE IN RECREATIONAL ATHLETES

Running Head: Carbohydrate mouth rinse concentration and 5 km running

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# ABSTRACT

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3	Oral carbohydrate rinsing has been demonstrated to provide beneficial effects on
4	exercise performance of durations of up to one hour, albeit predominately in a
5	laboratory setting. The aim of the present study was to investigate the effects of
6	different concentrations of carbohydrate solution mouth-rinse on 5 km running
7	performance. Fifteen healthy men (n=9; mean±SD age: 42±10 years; height:
8	177.6±6.1 cm; body mass: 73.9±8.9 kg) and women ( <i>n</i> =6; mean±SD age: 43±9 years;
9	height: 166.5±4.1 cm; body mass: 65.7±6.8 kg) performed a 5 km running time trial
10	on a track on four separate occasions. Immediately before starting the time trial and
11	then after each 1 km, subjects rinsed 25 mL of either 0, 3, 6, or 12% maltodextrin for
12	10 s. Mouth-rinsing with 0, 3, 6 or 12% maltodextrin did not have a significant effect
13	on the time to complete the time trial (0%: 26:34±4:07 min:sec; 3%: 27:17±4:33
14	min:sec; 6%: 27:05±3:52 min:sec; 12%: 26:47±4.31 min:sec; $P=0.071$ ; • $^2_P=0.15$ ).
15	heart rate ( $P$ =0.095; $^{\bullet}_{P}$ =0.16), rating of perceived exertion (RPE) ( $P$ =0.195;
16	• $^2_p$ =0.11), blood glucose ( $P$ =0.920; • $^2_p$ =0.01) and blood lactate concentration
17	( $P$ =0.831; $^{\bullet}_{p}$ =0.02), with only non-significant trivial to small differences between
18	concentrations. Results of this study suggest that carbohydrate mouth-rinsing provides
19	no ergogenic advantage over that of an acaloric placebo (0%), and that there is no
20	dose-response relationship between carbohydrate solution concentration and 5 km
21	track running performance.

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Key Words: Maltodextrin, Oral receptors, Field-based

# 222 INTRODUCTION

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Oral rinsing of a carbohydrate solution prior to, and during, exercise can improve performance without altering metabolic responses (e.g. 3,16,23,24). The underlying mechanism is believed to relate to the presence of carbohydrate within the mouth inducing increased brain activity within the orbitofrontal cortex (8). Chambers, et al. (4) reported that, independent of sweetness, carbohydrate can activate similar brain regions related to reward and motor control, possibly through non-sweet taste receptors found in the mouth. In addition, Gant, et al. (13) demonstrated that carbohydrate ingestion during fatiguing isometric elbow flexion can immediately affect performance by increasing corticomotor excitability through non-sweet receptors in the oral cavity area which can activate parts of the brainstem able to counteract the decreasing motor activity.

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Several 30-min to 1-hour time trial (TT) studies exist, with many reporting positive effects of mouth-rinsing on cycling (4,18,21) and running (22,23,24) performance. However, studies investigating running time trials have reported contradictory results. The first study using a running protocol showed no change in performance when mouth-rinsing a 6% maltodextrin solution during a 45 min time trial following 15 min at 65% maximal oxygen uptake ( $\sqrt[3]{O}_{2max}$ ) (26). In contrast, these observations were not supported by Rollo, et al. (23) where, during a 30 min running trial at a rating of perceived exertion [(RPE) 6-20] of level 15, mouth-rinsing a 6.4% concentration of carbohydrate drink throughout exercise significantly improved performance. The difference in findings between the two studies could be explained by the fact that the  $\boxed{2}$ 

studies utilized different types of motorized treadmill. Rollo, et al. (23) used an automated treadmill, whereas Whitham and McKinney (26) used a manually controlled treadmill. Automated treadmills are thought to be a more sensitive performance measure compared to the 'traditional' treadmill, as they do not require subjects to manually change speed (17). However, another possible explanation for the differences is the runners' nutritional status with subjects arriving at the laboratory after an overnight fasting (23) or a standardized diet 4 hours before the experimental protocol (26). Therefore, the effects of carbohydrate rinsing appear more profound after an overnight fast, although are still evident after ingestion of a meal (15).

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The majority of previous studies (e.g. 9,23,26) have used carbohydrate mouth-rinse solutions with concentrations of 6 to 6.4%, with a few exceptions. Fraga et al. (11) demonstrated that an 8% carbohydrate solution increased time to exhaustion on a treadmill. Lane, et al. (18) reported that a 10% carbohydrate mouth rinse improved a 60-min simulated cycling TT performance to a greater extent in a fasted state compared with a fed state, although optimal performance was achieved in a fed state with the addition of a carbohydrate mouth rinse. Kasper, et al. (16) demonstrated rinsing a 10% carbohydrate solution improved high-intensity interval running, albeit in a reduced glycogen state. Furthermore, Rollo, et al. (24) reported that self-selected jogging pace and repeated sprint performance was increased when rinsing a 10% carbohydrate solution. In contrast, rinsing a 6.4% maltodextrin solution was reported to have no benefit on repeated sprint running during a similar protocol (9). Therefore, in line with the occupancy theory (5), the greater the concentration of carbohydrate the more receptors within the buccal cavity may be activated, and consequently contribute to improved performance. However, only one previous study (14) to date

??? has attempted to ascertain whether a potential dose-response relationship exists ??? between the concentration of the carbohydrate mouth-rinse solution and performance, albeit in cycling. Therefore, the aim of the present study was to investigate the effects ?!?!? of differing concentrations of carbohydrate mouth-rinse on 5 km running performance overland outdoors. ???

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#### **METHODS**

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## **Experimental Approach to the Problem**

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The investigation was a single-blind randomized, placebo-controlled cross-over experiment. Methods were approved by the local Ethics Committee and subjects were made fully aware of the procedures, including any risks and benefits of participation in the study, before providing written informed consent. Procedures were undertaken in accordance with the Declaration of Helsinki. The study consisted of a total of four time trials after an initial familiarization trial where unflavored water was rinsed and conducted at the same outdoor grass running track, 500 m in circumference measured out on a college sports field. This allowed subjects to be accustomed to the experimental procedures and ameliorate a learning effect. Subjects performed four time trials with a minimum of 48 h recovery between trials and in the same clothing and trainers. In order to avoid potentially confounding effects, subjects refrained from strenuous exercise and consumed a standardized diet 24 h before each trial, details of which were recorded within a 24-hour food diary, which was adhered to for subsequent trials. Subjects arrived at the running track slot between 17:00 and 18:30

h following a five hour fast, during which they were instructed to avoid consumption of food, caffeine, tobacco or alcohol but were permitted to drink water *ad libitum* prior to the first trial, which was replicated for subsequent trials. Only non-significant differences were observed for ambient temperature (mean:  $19.4\pm0.5^{\circ}$ C;  $F_{3,42}=0.662$ ; P=0.580;  $\eta_P{}^2=0.05$ ), relative humidity (mean:  $64.0\pm0.8\%$ ;  $F_{3,42}=0.178$ ; P=0.911;  $\eta_P{}^2=0.01$ ) and wind speed (mean:  $1.3\pm0.2~\text{m}\cdot\text{s}^{-1}$ ;  $F_{3,42}=1.255$ ; P=0.302;  $\eta_P{}^2=0.08$ ) between conditions. Upon arrival subjects were weighed and fitted with a heart rate monitor before undertaking a standardized warm-up prior to the exercise trial. The warm up consisted of low to moderate aerobic exercise (jogging) for 5 min followed by 5 min during which the subjects could undertake their own stretching protocol and were instructed to reproduce the same preparation for each trial. Before commencing each track run subjects were encouraged verbally to give maximal effort to complete the 5 km running TT in the shortest time possible.

## **Subjects**

Fifteen healthy men (*n*=9; mean±SD age: 42±10 years; height: 177.6±6.1 cm; body mass: 73.9±8.9 kg) and pre-menopausal women (*n*=6; mean±SD age: 43±9 years; height: 166.5±4.1 cm; body mass: 65.7±6.8 kg) volunteered to take part to in the study. Subjects were recreational runners and members of the same running club and had consistently trained on average 3±1 times, covering a total of 17±7 miles, per week for the past two years and were familiar with running 5 km as part of their training and competition schedule. Subjects were required to complete a general health questionnaire (PAR-Q) to exclude any history of diabetes, cardiovascular or respiratory diseases.

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### **Familiarization**

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As familiarization, subjects completed the experimental protocol whilst mouth rinsing unflavored water at least 5 days prior to the first experimental trial. In order to establish any learning effect, following completion of the four experimental trials, 5 km time to completion was compared between the familiarization trial and placebo trial of the main experimental using a paired samples t-test. No significant difference between trials was observed (Familiarization:  $26:11\pm4:33$  min:sec, Placebo:  $26:56\pm4:08$  min min:sec; d=0.14; P=0.634).

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## **Mouth Rinse Solution and Procedure**

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The mouth-rinse solutions used were 0, 3, 6, or 12% maltodextrin (Myprotein, Cheshire, England) with water and energy-free sweetener (Vimto, Nichols plc., Merseyside, England). The sweetener was adjusted in volume at each trial by approximately 5% to match for taste and viscosity. Solutions were matched for flavor and color to make them indistinguishable and 25 mL solution was divided into polystyrene cups using a volumetric syringe. Five cups were prepared per subject, making a total volume of 125 mL of mouth-rinse solution per subject per trial.

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Subjects were required to mouth-rinse on five occasions, immediately before starting the TT and then after two completed laps (i.e. at 1, 2, 3, and 4 km). Consequently, the mean time between rinses was 5:21±0:50 min:sec. Subjects were informed every two laps (1000 m) that they had a total of 15 s (which was individually timed by one of the

investigators) to complete the rinse procedure i.e. to collect the cup, rinse for 10 s and expectorate. The "rinse-zone" was 15 m before the start/finish point of the track with signs and colored cones were used to direct subjects to pick up a polystyrene cup from a table set back 50 cm from the inside of the track. These cups contained the set bolus ???? (25 mL) of mouth-rinse solution. Subjects rinsed 25 mL of the solution around their ???? mouth for 10 s according to Sinclair, et al. (25) whilst running. The solution was then ???? expectorated and measured using electronic scales (Model no. 951, Salter Housewares Ltd., Kent, United Kingdom) to ensure that subjects did not ingest any of the solution. ???? After completing all trials subjects were questioned whether they could differentiate between the four different solutions in terms of taste or texture, and if they had ???? experienced any gastro-intestinal symptoms during the trials. For practical reasons, ???? the study was single-blinded, leaving potential for experimenter bias. However, no ???? subjects successfully identified 100% of the solutions, with a 23% success rate and only two subjects correctly identifying the placebo. ????

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# **Procedures**

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Subjects were fitted on arrival with a heart monitor, which consisted of a chest strap and receiver (Polar RS400, Polar Electro, Kempele, Finland). Subjects' heart rate (HR) was recorded at rest (5 min before starting the warm-up), at the end of every lap (500 m) and at completion of the TT. Maximum heart rate (HRmax) had previously been measured using the Yo-Yo endurance test. Before the warm-up and immediately after completion of the TT, blood lactate (Lactate Pro, Panasonic, Osaka, Japan) and glucose concentrations (Contour blood glucose monitor, Bayer Health Care, Mishawaka, IN) were measured with fingertip capillary blood samples. The rating of perceived exertion (2) was individually determined every 500 m of the TT. This scale was presented to the subjects on large signs positioned round the outside of the track.

# **Statistical Analysis**

Data are reported as the mean  $\pm$  the standard deviation (SD). All variables, with the exception of performance times were assessed using a two-way (condition x km) analysis of variance (ANOVA) with repeated measures. Performance times were analyzed using a one-way ANOVA with repeated measures. Sphericity was analyzed by Mauchly's test of sphericity followed by the Greenhouse-Geisser adjustment where required. Where any differences were identified, post-hoc pairwise comparisons with Bonferroni correction were conducted. All statistical procedures were conducted using IBM SPSS Statistics for Windows, Version 22.0 (Armonk, NY: IBM Corp.) and an alpha level of P < 0.05 was considered statistically significant. Furthermore, effect sizes using partial eta squared ( $\bullet_{\frac{3}{p}}$ ) were calculated, which were defined as trivial (0-0.19), small (0.20-0.49), moderate (0.50-0.79) or large ( $\bullet$ 0.80) (6).

## RESULTS

There was no significant effect of carbohydrate concentration on mean 5 km TT performance for men (0%: 27:02±4:02 min:sec; 3%: 27:49±4:34 min:sec; 6%: 27:47±3:59 min:sec; 12%: 27:25±4.29 min:sec;  $F_{3,24}$ =2.544; P=0.080;  $\bullet_P^2$ =0.24) or women (0%: 25:50±4:31 min:sec; 3%: 26:29±4:49 min:sec; 6%: 26:02±3:46 min:sec; 12%: 25:50±4.49 min:sec;  $F_{3,15}$ =0.925; P=0.453;  $\bullet_P^2$ =0.16). Furthermore, there was

no significant difference in 5 km TT performance time between men and women 197  $(F_{1,13}=0.416; P=0.530; \bullet_{p}^{2}=0.03)$ . In addition, there was a non-significant interaction 198 between sex and 5 km TT performance time ( $F_{3.39}=0.424$ ; P=0.737;  $^{\bullet}_{p}=0.03$ ). As a 199 200 consequence, the results are subsequently presented as a single group (n=15)201 No significant differences in the time taken to complete the 5 km TT performance 202 were observed between experimental conditions (F<sub>3,42</sub>=2.513; P=0.071;  $^{\bullet}_{P}$ =0.15; 203 Figure 1 and Figure 2). In addition, no significant order effect was observed 204 205  $(F_{3,42}=0.776; P=0.514; \bullet_p^2=0.05)$ . No significant differences were observed in mean heart rate ( $F_{2,25}$ =2.648; P=0.095;  $^{\bullet}_{p}$ =0.16; Table 1) and relative heart rate (%max) 206  $(F_{2.25}=2.457; P=0.111; \bullet_{p}^{2}=0.15; Table 1)$  and during the 5 km TT. Rating of 207 perceived exertion during the 5 km TT was also similar for all conditions 208  $(F_{3,42}=1.639; P=0.195; \bullet_p^2=0.11; Table 1)$ . Blood lactate (Table 1) increased to by a 209 large extent as a consequence of completing the time trial ( $F_{1,14}$ =43.351; P<0.001; 210 •  $^{2}_{p}$  =0.76), but there were no significant differences between conditions (F<sub>2.29</sub>=0.292; 211 P=0.831;  $^{\circ}_{p}=0.02$ ). Similarly, blood glucose (Table 1) increased by a moderate 212 213 extent during the time trial  $(F_{1.14}=11.112; P=0.005; \bullet_p^2=0.44)$ , but again, there were no significant differences between conditions (F<sub>3,42</sub>=0.163; P=0.920;  $^{\bullet}_{p}$ =0.01). The 214 mean volume of expectorate for the 0%, 3%, 6% and 12% trials was 24±2 mL, 24±1 215 mL, 24±2 mL and 24±1 mL, respectively. Thus, the difference between the volume 216 rinsed and expectorated was 1±2 mL in the 0% trial, 1±1 mL in the 3% trial, 1±2 mL 217 in the 6% trial, and 1±1 mL in the 12% trial. Furthermore, no subjects reported any 218 gastro-intestinal symptoms during the trials. 219

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### **DISCUSSION**

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The primary aim of the present study was to determine the effect of mouth-rinsing ????? different concentrations of carbohydrate solution on 5 km track running TT ???? performance in recreational athletes. The effect of mouth-rinsing carbohydrate ???? solutions on both running and cycling performance has been studied previously ???? (9,11,14,23,24,26). However, this is the first study to investigate the effects of ???? differing carbohydrate concentration on 5 km track running performance. The main ???? ???? finding of the present study was that mouth-rinsing with 3, 6 or 12% carbohydrate solutions for 10 s approximately every 5 min did not have a significant effect on 5 ???? km performance, subjects' heart rate, RPE, blood glucose and blood lactate ???? ???? concentrations during 5 km running compared to the placebo solution (0%). Furthermore, figure 2 reveals that the responses to the different concentrations are ???? individual and with no clear pattern. The results of the present study also support ???? ???? those of Ispoglou, et al. (14) and suggest that there is no dose-response relationship between carbohydrate concentration and performance when mouth-rinsing during ???? ???? exercise.

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The finding that only non-significant trivial differences between the four conditions is consistent with those of Whitham and McKinney (26), who concluded that mouth-rinsing a 6% carbohydrate solution had no significant effect on distance covered during a 45 min running time trial. However, in contrast, Rollo, et al. (22, 23) reported beneficial effects of carbohydrate rinsing during running-based protocols.

The present study also sought to address the limitations of the study reported by Whitham and McKinney (26), by conducting it in the field in order to allow subjects to change speed naturally, and be more representative of competitive situations. It has been suggested that carbohydrate mouth-rinsing affects the central nervous system, resulting in improved performance, thus manually changing speeds during treadmill performance could have masked the potential unconscious effects of the carbohydrate mouth-rinse (15,17). In addition, in the current study, mouth-rinsing lasted for 10 s instead of 5 s. This increase in time taken to rinse has been found to have a greater positive effect on performance (25). However, despite the longer time for mouth rinsing (10 s) and apparent optimum frequency of approximately every 5 min (10,23,25), the present study failed to reproduce results reported in the laboratory. Furthermore, 10 s may not be practical whilst running due to interrupting the breathing cycle, as subjects must either hold their breath or breathe through the nose while the solution is rinsed in the mouth, resulting in decrease efficiency and a possible increase in time to completion (12).

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It has been suggested that carbohydrate mouth-rinsing activates regions in the brain related to motor output and pleasure/reward (4). Similarly, De Pauw, et al. (8) reported that the presence of carbohydrate within the mouth sends signals that activate the reward centers of the brain, due to a direct link between the buccal mucosa and the brain (19). Thus, exercise performed by an athlete might be perceived as 'easier' when carbohydrate is mouth-rinsed compared to a placebo. This neural mechanism could explain why although studies have found increased performance with carbohydrate mouth-rinsing, no change or a decrease in RPE, suggesting that carbohydrate mouth-rinsing may allow increased exertion whilst the perception of

fatigue remains stable. However, in the current study, RPE remained relatively constant between conditions and performance did not improve, suggesting that carbohydrate mouth-rinsing did not sufficiently stimulate the reward and motor output brain regions sufficiently to improve 5 km performance. Furthermore, as RPE has been shown to be comparable at different percentages of maximal oxygen uptake in amateur and professional cyclists (20) and at lactate threshold in trained and untrained runners (7), similar responses to those seen in the present study may be observed in athletes, although this is only speculation at present.

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The majority of previous studies that have reported performance gains from carbohydrate mouth-rinsing when compared to a placebo have produced marginal performance gains of approximately 2-3% (15), especially during cycling events. Furthermore, Gam, et al. (12) reported the act of repeatedly rinsing the mouth during a cycle time trial had a detrimental effect on performance, although the addition of carbohydrate to the rinse solution reduced the decrease in performance associated with repeated mouth rinsing. Therefore, it is possible that the act of rinsing the mouth during the time trials caused a loss of attention and focus on the task resulting in these transient declines in performance (12), as well as efficiency, which when repeated cause an overall decrease in performance. Consequently, the findings in the present study may be attributed to a slowing in the running pace in order to mouth rinse. Therefore, future studies should include a "no-rinse" control condition in order to ascertain the true effect of carbohydrate rinsing.

This study is not without limitations. Some subjects ingested a small amount of the solutions (approximately 1 mL) during the rinse procedure, which could have be confounded by saliva output, although this volume was likely to be trivial in the time allowed for rinsing. However, no effect on blood glucose or performance was observed, most likely due to the small amount of carbohydrate ingested (less than 1 g over the duration of the trial). In addition, large standard deviations are evident for the majority of variables. The reason for this is primarily attributed to the variability of athletic standards amongst the subjects, which had implications for all recorded measures, such as heart rate or TT performance, which ranged from the fastest 21:21 min:sec to the slowest 36:13 min:sec across the four trials. Ideally, a more homogeneous population would have been recruited thus avoiding a large range in characteristics and abilities which can result in a greater increase in 'noise' within the data. Also for practical reasons, the study was single-blinded, leaving potential for experimenter bias, however as no subjects could correctly guess the solutions, this would seem unlikely. Furthermore, the use of a 500 m track on grass did allow for a standardized distance between rinses, it may have contributed to the variability between trials. However, the grass was in good condition and trials took place on sunny days, so the surface was consistent. Finally, although trivial and not significant, the familiarization session trial was performed 2% faster was than the placebo trial. Although no obvious explanation for this occurrence, Chambers, et al (4) reported that areas of the brain, such as the anterior cingulate cortex and ventral striatum, that were unresponsive to artificial sweetener however, Arnaoutis, et al (1) suggested that water may activate pharyngeal receptors and thus improve exercise performance. However, this is only speculation and further research is required to substantiate this suggestion.

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### PRACTICAL APPLICATIONS

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The results of the present study suggest that compared to an acaloric solution (0%), mouth-rinsing with solutions containing, 3, 6 or 12% carbohydrate did not improve 5 km track performance in recreational runners. Therefore, coaches, practitioners and athletes may wish to evaluate the effectiveness of carbohydrate rinsing against a "norinse" condition before consideration. Furthermore, a personalized diet designed to meet carbohydrate and fluid requirements may be of greater benefit. However, in situations such as where individuals suffer from gastrointestinal distress or are undertaking exercise for weight management purposes, and the exercise duration is less than 60 m, then carbohydrate mouth-rinsing may be a useful strategy.

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In conclusion, the results of the present study suggest that there is not a dose-response relationship and mouth-rinsing with a carbohydrate solution might not be as effective as previous studies suggest during running lasting less than 30 min and performed outdoors. Furthermore, future mouth rinsing studies should include a "no-rinse" trial as a control.

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<ul><li>????</li><li>????</li><li>????</li></ul>	(13)	mouth rinse solution opposes the detrimental effect of mouth rinsing during cycling time trials. <i>Int J Sport Nutr Exerc Metab</i> 3: 48-56, 2013.  Gant, N, Stinear, C, and Byblow, W. Carbohydrate in the mouth immediately facilitates motor output. <i>Brain Res</i> 1350: 151-158, 2010.
<ul><li>????</li><li>????</li><li>????</li><li>????</li><li>????</li></ul>	(13)	mouth rinse solution opposes the detrimental effect of mouth rinsing during cycling time trials. <i>Int J Sport Nutr Exerc Metab</i> 3: 48-56, 2013.  Gant, N, Stinear, C, and Byblow, W. Carbohydrate in the mouth immediately facilitates motor output. <i>Brain Res</i> 1350: 151-158, 2010.  Ispoglou, T, O'Kelly, D, Angelopoulou, A, Bargh, M, O'Hara, J, and
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# **Figure legends**

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Figure 1: Mean ( $\pm$ SD) time taken (min) to complete 5 km time trial. n=15

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Figure 2: Individual male (♂) and female (♥) time taken (min) to complete 5 km

2222 time trial.

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**Table 1:** Mean ( $\pm$ SD) heart rate, RPE, lactate and glucose concentrations during 5 km time trials (n=15).

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	Solution				
Variable	0%	3%	6%	12%	
Heart Rate (beats·min <sup>-1</sup> )	160±9	154±12	155±13	153±11	
Heart Rate (%max)	90±4	87±5	87±6	86±5	
RPE	14±2	13±2	13±1	13±2	
Pre-lactate (mmol·L <sup>-1</sup> )	2.31±1.38	2.16±1.70	2.66±1.47	2.08±1.17	
Post-lactate (mmol·L <sup>-1</sup> )	8.78±4.00	10.22±7.09	8.96±6.39	8.68±5.93	
Pre-glucose (mmol·L <sup>-1</sup> )	4.48±0.95	4.31±0.73	4.21±0.63	4.21±0.83	
Post-glucose (mmol·L <sup>-1</sup> )	5.97±1.69	5.96±1.86	5.90±2.45	6.02±2.08	

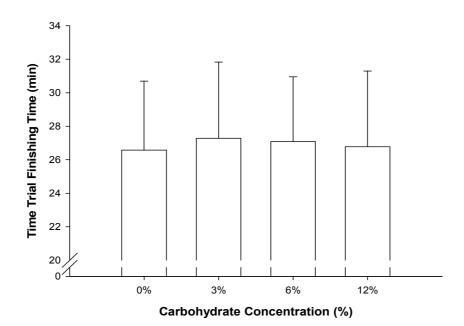
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