

James Madison University
JMU Scholarly Commons

Masters Theses

The Graduate School

Spring 2017

Impact of pre-exercise feedings with a low or high glycemic index on the ergogenic effects of carbohydrate mouth-rinsing during cycling

Nikolai J. Hladick
James Madison University

Follow this and additional works at: <https://commons.lib.jmu.edu/master201019>

 Part of the [Exercise Science Commons](#)

Recommended Citation

Hladick, Nikolai J., "Impact of pre-exercise feedings with a low or high glycemic index on the ergogenic effects of carbohydrate mouth-rinsing during cycling" (2017). *Masters Theses*. 479.
<https://commons.lib.jmu.edu/master201019/479>

This Thesis is brought to you for free and open access by the The Graduate School at JMU Scholarly Commons. It has been accepted for inclusion in Masters Theses by an authorized administrator of JMU Scholarly Commons. For more information, please contact dc_admin@jmu.edu.

Impact of Pre-Exercise Feedings with a Low or High Glycemic Index
on the Ergogenic Effects of Carbohydrate Mouth-Rinsing during Cycling

Nikolai J. Hladick

A thesis submitted to the Graduate Faculty of

JAMES MADISON UNIVERSITY

In

Partial Fulfillment of the Requirements

for the degree of

Master of Science

Department of Kinesiology

May 2017

FACULTY COMMITTEE:

Committee Chair: Michael J. Saunders, Ph.D.

Committee Members/ Readers:

Nicholas D. Luden, Ph.D.

Christopher J. Womack, Ph.D

Acknowledgements

I would like to thank Dr. Michael Saunders for guiding me through this project as my thesis advisor. Your patience, knowledge, and support were greatly appreciated and vital in the completion of this thesis. I would also like to thank Dr. Nicholas Luden and Dr. Christopher Womack for their input, suggestions, and enthusiasm for this project. I am fortunate in having had the opportunity to work with such a group of knowledgeable individuals.

I would like to thank my research partner, Kevin Decker, as well. Without your knowledge of lab equipment, willingness to teach me, and patience throughout data collection, this project would not have been realized. Thanks also to Kendall Clark, for making the majority of the pre-exercise drinks and assisting with data collection. I would also like to thank Megan Pedersen and Josh Forrest for making pre-exercise drinks and assisting with data collection. Thanks to any other graduate students and interns who assisted me in the lab during data collection and answered any questions I had throughout this process.

Finally, I would like to thank my parents and sister for supporting me throughout my educational career and encouraging me to pursue my interests. I would not have made it this far without their guidance, support, and encouragement.

This project was supported by a CHBS Faculty Research Grant (M. Saunders).

Table of Contents

Acknowledgements.....	ii
Table of Contents.....	iii
List of Tables.....	iv
List of Figures.....	v
Abstract.....	vi
I. Introduction.....	1
II. Methods.....	11
III. Manuscript.....	18
IV. Appendices.....	51
V. References.....	66

List of Tables

Table 1: Treatment Conditions.....	12 & 24
Table 2: Satiety Ratings Before and After Beverage Consumption.....	31
Table 3: Physiological Responses during Constant-Load Exercise.....	35
Table 4: Physiological Responses during 30-km Time Trial.....	36
Table 5: Performance Times and Treatment Differences.....	37

List of Figures

Figure 1: Experimental Protocol.....	14 & 27
Figure 2: Effects of Treatment Beverages on Blood Glucose Responses.....	32

Abstract

Purpose: Carbohydrate (CHO) ingestion during exercise enhances performance in short endurance events (~ 1 hr) due to neural influences, as demonstrated by the efficacy of CHO mouth-rinsing during cycling. However, the magnitude of these neural effects may be blunted following pre-exercise CHO feedings. This study examined whether the glycemic index (GI) of a pre-exercise meal affected time-trial (TT) performance in cyclists using a CHO mouth-rinse during exercise. **Methods:** Eight cyclists (age: 24 ± 6 yr; VO_{2max} : 61 ± 8 ml·kg⁻¹·min⁻¹) completed 4 exercise trials, consisting of 15 min of constant-load cycling followed by a simulated 30-km TT. Treatments were: a) L-CHO: low GI CHO beverage pre-exercise (1.5 g·kg⁻¹ CHO, 120 min prior), CHO mouth rinsing during exercise (6.4% maltodextrin solution), b) H-CHO: high GI CHO beverage (1.5 g·kg⁻¹ CHO) pre-exercise, CHO mouth rinsing during-exercise, c) PL-CHO: placebo beverage pre-exercise, CHO mouth rinsing during exercise, and d) PL-PL: placebo beverage pre-exercise, placebo mouth rinsing during exercise. Blood glucose was measured before beverage consumption and at 30 and 120 min following ingestion. Physiological measurements (VO_2 , V_E , RER, HR, RPE, glucose, lactate, and gastrointestinal distress) were assessed during constant-load cycling and the TT. Magnitude-based qualitative inferences were used to assess differences in responses between trials. **Results:** Blood glucose differed among treatments 30 min post-feeding (H-CHO > L-CHO > PL-CHO = PL-PL), and was lower in H-CHO versus PL-CHO and PL-PL during subsequent exercise. Compared to PL-CHO, TT performance was faster in both L-CHO (-0.5 ± 0.8 min; “likely” beneficial) and H-CHO (-0.7 ± 0.7 min; “likely” beneficial), with no systematic differences between L-CHO and H-CHO. However, none

of the 3 mouth rinse trials were clearly different from the PL-PL trial. **Conclusions:** When using a CHO mouth rinse during exercise, CHO ingestion 2 hr prior to cycling enhanced TT performance versus exercise in the fasted state. The GI of the pre-exercise feeding did not systematically affect TT performance in cyclists using a CHO mouth-rinse. However, the impact of these findings is confounded by the lack of performance differences versus a control trial without CHO before or during exercise.

Chapter 1

Introduction

Carbohydrate Consumption and Performance

The ergogenic effects of carbohydrate consumed before exercise, during exercise, and in various combinations of both time periods have been extensively investigated.

Pre-exercise carbohydrate ingestion can enhance endurance performance across various modes, intensities, and durations of exercise (18,50,51,60). Ergogenic effects have been reported in exercise bouts with feedings close in proximity (15 minutes) to exercise (60) and feedings 2-3 hr prior to exercise (50,65,67). Tokmakidis et al. (60) demonstrated improved running performance following carbohydrate ingestion in comparison to placebo, which is consistent with other data gathered on cyclists (18,50,51).

Carbohydrate ingestion *during* exercise also positively impacts endurance performance. Coyle et al. (13,14) reported improved cycling endurance with carbohydrate feedings at 20 minute intervals throughout exercise, with other studies reporting similar outcomes (29,33,44). Additionally, carbohydrate feeding both before and during exercise, a common practical strategy in endurance events, improves performance over feeding exclusively before or during exercise (11,12). The ergogenic effects of carbohydrate have primarily been attributed to increased carbohydrate oxidation rates, resulting in greater energy availability in the late-stages of prolonged exercise. Carbohydrate oxidation rates increase as the dose of ingested carbohydrate increases, with peak oxidation rates attained at $\sim 60\text{-}70 \text{ g}\cdot\text{hr}^{-1}$ from single-carbohydrate sources (34,35). As a result, the ergogenic effects of carbohydrate during prolonged exercise appear to be dose-dependent. For example, Smith and colleagues fed subjects

15, 30, or 60 g of carbohydrate per hr during a 2 hr steady state bout of cycling preceding a performance test (54). Performance improved as the carbohydrate dose increased, with the 60 g·hr⁻¹ dose producing the best performance times (54).

Metabolic and Neural Effects of Carbohydrate Consumption

As described above, the ergogenic effects of carbohydrate consumption during prolonged exercise (> 90 min) have been predominantly attributed to increased carbohydrate oxidation. However, this is not the case in shorter bouts of intense aerobic exercise (≤ 60 min). Carter et al. (8) concluded that exogenous carbohydrate contributes minimally to overall carbohydrate oxidation during intense aerobic exercise, as only 9 g of ~ 60 g of intravenously infused glucose were oxidized during the last quarter of a 1 hr exercise trial. Similarly, McConnell et al. (42) measured only 22 g of 84 g of ingested carbohydrate in the circulation of subjects cycling intensely within the same timeframe. Despite low glucose oxidation and appearance rates in exercise lasting 1 hr, performance has reportedly been improved with carbohydrate consumption during exercise of this duration (2,33).

Recently, neural factors related to oral sensing of carbohydrate have been cited as a possible mechanism by which carbohydrate enhances performance during shorter bouts of exercise. Multiple studies have begun to elucidate the link between carbohydrate consumption and neural stimulation leading to enhanced motor performance. Gant and colleagues (25) observed increases in maximal voluntary force production with ingestion of glucose. Force increased before glucose entered the blood stream (25). At the same time, corticomotor regions of the brain were activated, leading the authors to hypothesize that chemoreceptors in the mouth sense the presence of carbohydrate and calories, and

subsequently stimulate neural pathways related to motor performance (25). Interestingly, the degree to which maximal voluntary force production was enhanced was not correlated with blood glucose levels (25).

Other studies investigating oral carbohydrate sensing and neural activation have concluded that neural activation is facilitated by the presence of carbohydrate, regardless of sweetness. In one study, sucrose and a non-caloric sucralose placebo were matched for sweetness and administered to subjects (23). Sucrose ingestion activated more brain areas related to taste compared to ingestion of the non-caloric sucralose (23). Specifically, sucrose ingestion activated the primary gustatory cortex, which includes the frontal operculum and the anterior insula, in addition to activating the striatum, the prefrontal cortex, and the anterior cingulate cortex (23). Sucrose activated brain areas related to reward and led to a different neuro-physiological response in the brain compared to sucralose ingestion, though subjects could not distinguish between the two solutions (23).

In a similar study, Chambers and colleagues (9) measured brain activity upon mouth rinsing a 6.4% maltodextrin solution, a glucose solution, and a non-caloric sweetened placebo. Both carbohydrate solutions activated brain areas related to reward processing and motor output (9). The placebo did not activate the same brain regions, leading the researchers to propose that yet unidentified oral sensors in the human mouth detect carbohydrate regardless of sweetness (9).

Carbohydrate Mouth Rinsing and Exercise Performance

Carter and colleagues (7) were the first to investigate the effects of oral-pharyngeal carbohydrate exposure without ingestion on exercise performance. Subjects

improved 1 hr cycling performance compared to a placebo trial when they mouth rinsed for five seconds with 25 ml of a 6.4% maltodextrin solution at regular intervals throughout exercise (7). The authors proposed that the activation of brain regions related to reward and pleasure may have made higher exercise intensities more tolerable, as the rating of perceived exertion remained the same between trials despite higher power output during the mouth rinse trial (7).

Subsequent studies have produced conflicting results, with some (3,9,16,19,40,41,46,47,48,49,52) but not all (1,5,28,30,39,63) confirming the ergogenic effect of carbohydrate mouth rinsing. Other studies reporting improved exercise performance have also reported no change in rating of perceived exertion during mouth rinse trials that produced faster performance times and higher work rates (9,24,46), with one study reporting faster self-selected running paces at the same rating of perceived exertion with a carbohydrate mouth rinse compared to a placebo mouth rinse (48). Pottier et al. (46) compared a carbohydrate mouth rinse protocol to a carbohydrate ingestion protocol and found the mouth rinse trials produced superior performances to the ingestion trials, though another study reported no performance differences between carbohydrate rinsing and ingestion (22). While the majority of mouth rinse studies have been conducted using cycling protocols, multiple studies have reported improvements in running performance with carbohydrate mouth rinsing (47,49).

Using mouth rinse protocols, researchers have manipulated the length of mouth rinse time, the concentration of the carbohydrate solution, and the intensity of exercise, and have observed the subsequent effects of each variable on performance. Most studies have reported an improvement in performance with a 5 second rinse time (7,16,46,47,49),

though a 10 second rinse time also enhanced performance (3,9,40). In addition to rinse duration, multiple studies have investigated the effects of varying the concentration of the carbohydrate solution. Devenney et al. (16) reported improved cycling performance with 6% and 16% carbohydrate mouth rinse solutions, with no difference between the two experimental trials. Other studies have reported no differences in performance between carbohydrate solutions varying widely in concentration, though carbohydrate rinses did not enhance performance relative to placebo rinses in those studies (28,39). Intensity of exercise may also play a role in the efficacy of carbohydrate mouth rinsing, as one study reported an improvement in cycling performance at 80% of subjects' respiratory compensation point, but no improvement at 110% of peak power output when rinsing with a carbohydrate solution (3).

Pre-Exercise Feeding in Combination with Mouth Rinsing

In addition to the above factors, the effects of feeding or fasting prior to exercise can influence performance when using a carbohydrate mouth rinse during exercise. Most studies in which carbohydrate mouth rinsing enhanced high-intensity endurance performance (~ 1 hr) entailed exercising in the fasted state (9,22,47,49), leading researchers to speculate that pre-exercise carbohydrate feedings may blunt the effects of mouth rinsing during exercise. However, mouth rinse studies in which subjects consumed carbohydrate roughly 2-3 hr before exercise have reported both an improvement in performance (16,46) and no change in performance (5,28). Similarly, studies in which subjects fasted before trials have also provided inconsistent findings, with some reporting an improvement in performance with carbohydrate mouth rinsing (9,22,47,49), and others reporting no change in performance (1,22,39).

Some studies have directly compared the influence of pre-exercise feeding or fasting on the ergogenic effects of mouth rinsing. In one study, subjects either fasted or consumed a standardized breakfast 2 hr before completing as much work as possible in 1 hr on a cycle ergometer while mouth rinsing with a 10% maltodextrin solution or a placebo solution (40). The carbohydrate mouth rinse enhanced performance in both states, though the improvement was greater when subjects fasted; power output improved by 1.8% and 3.4% in the fed and fasted state respectively, compared to rinsing with a placebo solution in both states (40). Overall performance, however, was best during trials in which subjects consumed carbohydrate before exercise and utilized a carbohydrate mouth rinse during exercise (40). Fares et al. (19) reported similar results, concluding that the effect of carbohydrate mouth rinsing was more pronounced when subjects were fasted. However, studies done by Trommelen et al. (61) and Beelen et al. (5) found no influence of pre-exercise feedings on the effects of a carbohydrate mouth rinse, as performance remained unchanged between prandial states when subjects rinsed with a placebo solution or a carbohydrate solution.

The presumed attenuation of the ergogenic effects of mouth rinsing in the fed state may be mediated by neurological responses to the body's metabolic environment. There is evidence to suggest that satiety prior to carbohydrate feeding affects neurological responses to the feeding (26,53). Overall neural activation and activation of brain areas related to reward processing are attenuated when consuming carbohydrate in the fed state compared to the fasted state (26). Furthermore, Smeets et al. (53) reported a dose-response relationship between amount of glucose consumed and attenuation of hypothalamic activity. The decrease in hypothalamic activity occurred prior to a

substantial increase in blood glucose, while the time-course of further decreases suggested that hypothalamic activation was related to blood glucose and insulin levels (53). These data suggest that changes in blood glucose and/or insulin levels following pre-exercise carbohydrate feedings could at least partially influence the neural response to carbohydrate mouth rinsing during exercise.

Glycemic Index of Pre-Exercise Meals

Based on evidence from Lane et al. (40), the combination of pre-exercise feeding and the use of a carbohydrate mouth rinse during exercise may provide the optimal performance advantage compared to using one of these strategies alone. However, as pre-exercise feeding has been shown to diminish the efficacy of carbohydrate mouth rinsing, a strategy that maximizes the effects of both feeding and rinsing is of value to athletes. Manipulating the glycemic index of the pre-exercise meal could be a useful strategy in this regard.

The glycemic index (GI) was created by Jenkins et al. (31) to quantify the glycemic responses of different foods. Foods with a low GI value produce lower blood glucose and insulin responses than foods with a high GI value (31). As a result, pre-exercise meals with different GI values produce different metabolic effects during exercise. Compared to high GI pre-exercise meals of similar carbohydrate content, low GI pre-exercise meals lead to a reduced insulin response (57,59), higher plasma glucose levels late in exercise (59), greater concentrations of plasma free fatty acids during exercise (57,59,66), and increased rates of fat oxidation during exercise (57,66), though not all studies matched nutrients and calories across meals (59). Moreover, Thomas et al. (59) reported an inverse relationship between post-exercise plasma glucose and insulin

concentrations and the GI value of the pre-exercise meal, in addition to a positive correlation between the area under the curve for RER during exercise and the GI value of the pre-exercise meal. These effects have been observed when meals were consumed shortly before exercise (~ 1 hr prior) and further from exercise (~ 3 hr prior), and during exercise bouts ranging from 60 min to 90 min in duration (57,59,66).

The different metabolic effects of high and low GI carbohydrates seem to elicit different neural responses as well; one study reported reduced activity in brain areas that process reward recognition and appetite regulation after consumption of a high GI carbohydrate (glucose) compared to a low GI carbohydrate (fructose) (45). This response occurred immediately after glucose ingestion and continued for 1 hr (45). Consuming a low GI meal versus a high GI meal pre-exercise seems to create a more favorable metabolic and neural environment for optimizing the ergogenic effects of a carbohydrate mouth rinse protocol, as low GI meals keep blood glucose and insulin levels comparatively low. This may keep brain areas related to reward processing and motor output sensitive enough to be sufficiently activated by the centrally-mediated effects of oral carbohydrate sensing. Conversely, the metabolic and neural effects of consuming a high GI carbohydrate may desensitize these same brain areas, which could diminish the centrally-mediated effects of oral carbohydrate sensing.

The GI value of pre-exercise meals also seems to affect performance in bouts of exercise lasting longer than 1 hr. The first study investigating the effects of pre-exercise meals of different glycemic indexes on endurance performance was performed by Thomas et al. (58) in 1991. Eight trained cyclists consumed $1 \text{ g} \cdot \text{kgBW}^{-1}$ carbohydrate from lentils (low GI) and potatoes (high GI) 1 hr prior to exercise (58). Cycling time to

exhaustion at 67-68% VO_{2max} was enhanced following lentil consumption, but not potato consumption (58). Studies following this initial investigation have produced varied results. Multiple studies have shown improvements in cycling performance (15,37,38,43) and running performance (65,67) following consumption of a low GI meal compared to a high GI meal, while other studies have reported no significant performance effects in cycling trials (6,17,20,21,32,36,37,55,56) or running trials (10,62,64) following consumption of a low GI meal compared to a high GI meal. Methodological differences between studies related to the type of food consumed, the exercise test used, and the timing of pre-exercise feedings may help to explain these discrepant findings.

Considering the metabolic and neural effects of a low GI vs high GI pre-exercise meal, and the possible ergogenic effects of a low GI pre-exercise meal, consuming carbohydrate with a low GI before using a carbohydrate mouth rinse during exercise may confer the greatest performance benefit. To the knowledge of the authors, no studies have been conducted investigating the effect of the glycemic index of a pre-exercise meal on the effects of carbohydrate mouth rinsing during exercise.

Purpose and Hypothesis

The purpose of this study was to investigate the influence of a low GI pre-exercise meal (versus a high GI meal) on the ergogenic effects of a carbohydrate mouth rinse protocol used during exercise. We hypothesized that consuming a low GI pre-exercise meal in combination with a carbohydrate mouth rinse would improve exercise performance over consuming a high GI pre-exercise meal.

Assumptions, Limitations, Delimitations

During this study, it was assumed that subjects gave maximal efforts during all performance trials. The researchers also assumed that subjects adhered to behavioral and dietary protocols and instructions before trials, and all experimental protocols during trials. Accuracy of measurement instruments and competency of all researchers and assistants involved was assumed. Due to the homogeneity of the subject group, the results of this study can only be applied to similarly trained subjects, between the ages of 18 and 45 years old. Trials were performed on cycle ergometers in an exercise laboratory; as such, the practical applications of the findings are limited when applying the same feeding strategies in outdoor competitions or with different modes of exercise.

Chapter 2

Methods

Subjects

Eight well-trained cyclists between the ages of 18 and 45 were recruited from James Madison University and the Harrisonburg, VA community. Subjects had at least 2 years of experience in endurance cycling events; a $\text{VO}_{2\text{max}} \geq 50 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$; consistently trained over the past 2 months, defined as cycling an average of ≥ 3 days $\cdot\text{week}^{-1}$; and had completed at least 4 training sessions ≥ 2 hr in duration over the previous 2 months. Subjects gave written informed consent and were free of any disease or health complication that could have caused adverse effects during exercise or exercise testing. All protocols were approved by the James Madison University Institutional Review Board.

Study Design

We used a randomized double-blind placebo-controlled crossover design. Subjects performed 4 experimental trials, each consisting of a 15-min constant-load phase followed by a simulated 30-km cycling time trial on a Velotron cycle ergometer (Racermate, Inc., Seattle, WA) in the Human Performance Laboratory at James Madison University. The 4 trials were identical, other than the pre-exercise beverage and/or during-exercise mouth-rinse used, as shown in Table 1. Trial order was randomly counterbalanced across subjects and separated by ≥ 7 days each.

Table 1: Treatment Conditions

Condition	Pre-exercise beverage	During-exercise mouth-rinse
PL-PL	PL	PL
PL-CHO	PL	CHO
L-CHO	LGI	CHO
H-CHO	HGI	CHO

PL = placebo; LGI = low glycemic index; HGI = high glycemic index; CHO = carbohydrate

Preliminary Testing and Familiarization

Height and weight were measured on subjects' first visit to the laboratory. An incremental cycling test to volitional fatigue was performed to determine subjects' VO_{2max} . The test began with a 5 min warmup at 100 W on the aforementioned cycle ergometer, after which the subjects selected a workload that was subjectively sustainable for ~ 1 hr. Every 2 min the workload was increased by 25 W until volitional fatigue. A Moxus Modular Metabolic System (AEI Technologies, Pittsburgh, PA) was used to measure and record oxygen uptake (VO_2) and carbon dioxide production throughout the test, and to assess maximal oxygen consumption (VO_{2max}) and respiratory exchange ratio. The VO_{2max} value was recorded as the highest 30 second average VO_2 value during the test. Heart rate was measured throughout the test using a Polar heart rate monitor.

Subjects completed a familiarization trial prior to the experimental trials. During this trial, subjects completed a 15-min constant-load phase followed by a 30-km time trial, as described below, with the exceptions that a) no fingerstick blood samples were obtained, b) no pre-exercise beverage was provided, and c) subjects rinsed with water instead of a carbohydrate solution during exercise. A familiarization trial was used so subjects could learn the testing protocol and become comfortable using a cycle ergometer in a laboratory setting, thereby minimizing any learning effects that had the potential to confound results.

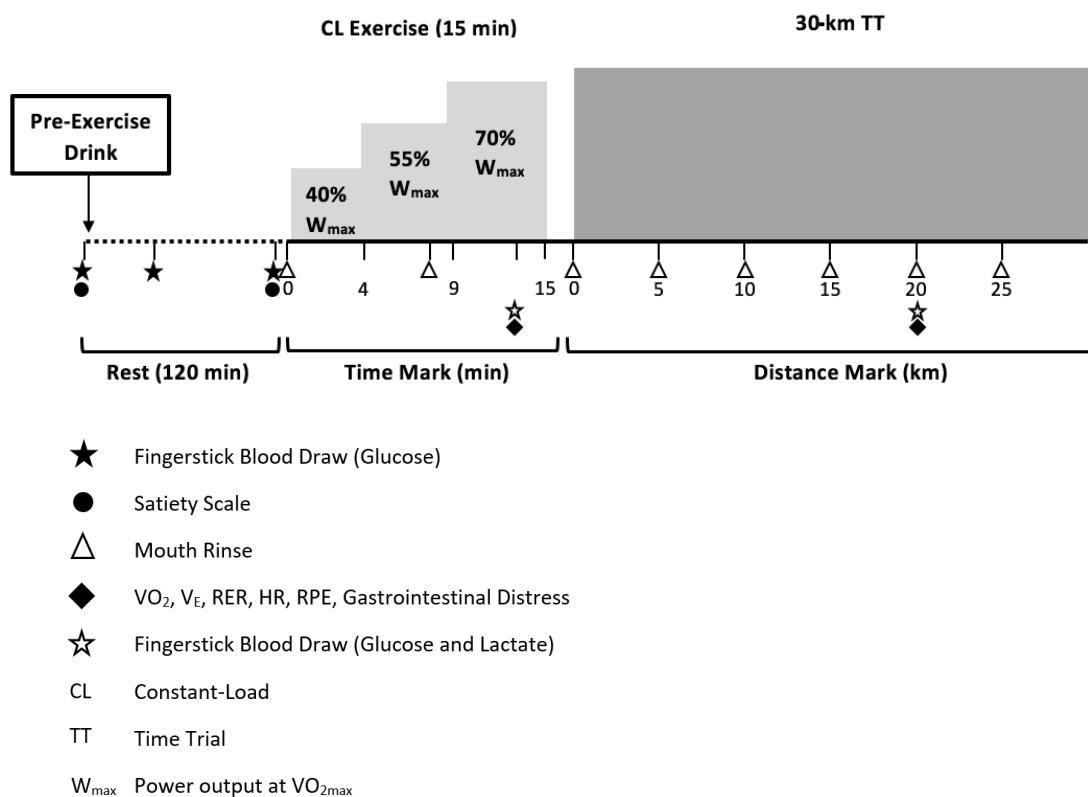
Experimental Protocol

The experimental protocol is displayed in Figure 1. Subjects arrived at the laboratory after an overnight fast. A fingerstick blood sample (~0.25 ml) was obtained and subjects consumed the pre-exercise beverage at a relatively constant rate over the course of ~ 2 min. Two more fingerstick blood samples were obtained, one at 30 min and one at 120 min after consumption of the pre-exercise beverage. Two hr after consuming the pre-exercise beverage, subjects began the constant-load phase. The first 4 min of this phase were performed at 40% of the workload corresponding to subjects' VO_{2max} (W_{max}), the second 5 min were performed at 55% W_{max} , and the final 6 min were performed at 70% W_{max} . The constant-load phase provided subjects with a progressive warmup prior to the time trial and allowed for comparison of physiological measurements at the same workload and time point between trials (i.e. independent of potential differences in pacing). The simulated 30-km time trial began about 2-3 min after the constant-load phase. Subjects were asked to give a maximal effort to complete the distance in the shortest possible amount of time, and to treat each trial as a competitive event. Other than distance completed, no feedback was provided to subjects during performance trials.

During the constant-load phase, subjects rinsed their mouths with 25 ml of either a carbohydrate solution or an artificially sweetened, non-caloric placebo at minute 0 and minute 7.5. During the 30-km performance trial, subjects rinsed every 5 km beginning at 0 km. Subjects rinsed a total of 8 times throughout each trial: twice during the constant-load phase and six times during the time trial. Subjects rinsed the solution around their

entire mouth for 5 seconds, before expectorating the solution back into the cup provided. Subjects were instructed to avoid swallowing any of the mouth rinse solution.

Figure 1: Experimental Protocol



Pre-Exercise Beverages and Mouth Rinse Solutions

The pre-exercise beverages consisted of a low GI carbohydrate solution (LGI_{Pre}), a high GI carbohydrate solution (HGI_{Pre}), and a non-caloric placebo (PL_{Pre}). LGI_{Pre} consisted of $10 \text{ ml}\cdot\text{kg}^{-1}$ of a slow-releasing high molecular weight 15% modified starch solution (UCAN Co., Woodbridge CT) providing $1.5 \text{ g CHO}\cdot\text{kg}^{-1}$ body weight. HGI_{Pre} consisted of $10 \text{ ml}\cdot\text{kg}^{-1}$ of a 15% maltodextrin solution providing $1.5 \text{ g CHO}\cdot\text{kg}^{-1}$ body weight. Both beverages were made by mixing powdered forms of each carbohydrate with water, creating virtually tasteless solutions that were uniformly flavored with a non-

caloric sweetener. PL_{Pre} consisted of 10 ml·kg⁻¹ of water flavored with the same non-caloric sweetener. All solutions were matched for taste and sweetness.

The mouth rinse solutions consisted of a maltodextrin solution (CHO_{Ex}) and a non-caloric placebo (PL_{Ex}). The CHO_{Ex} solution consisted of a 6.4% maltodextrin solution, while PL_{Ex} consisted of water flavored with a non-caloric artificial sweetener. Both solutions were prepared as previously described.

Dependent Measurements

Performance: Performance was assessed by recording the time to complete the 30-km time trial.

Oxygen Consumption (VO₂), Ventilation (VE), and Respiratory Exchange Ratio (RER): Expired gas samples were measured with a metabolic cart (described previously) from minute 9 to minute 15 of the constant-load phase and for 5 min at 20 km of the time trial, after subjects had mouth rinsed. VO₂, VE, and RER were calculated from gas samples at each time-point via automated software. Values were averaged over the final 3 min of data collection, following 2-3 min of breathing equilibration.

Heart Rate: Heart rate was measured continuously throughout trials with an automated heart rate monitor consisting of a chest strap and wrist receiver, held by the researchers.

Ratings of Perceived Exertion (RPE), Gastrointestinal Discomfort, and Satiety: RPE was obtained at minute 13 of the constant-load phase and at 20 km of the time trial using Borg's 6-20 scale. Subjects rated gastrointestinal discomfort using ten 1-10 scales at the same time points. These scales assessed the following: stomach problems, gastrointestinal cramping, bloated feeling, diarrhea, nausea, dizziness, headache,

belching, vomiting, and the urge to urinate or defecate. A score of 1 indicated absence of the symptom, while higher scores indicated that subjects were experiencing the symptom to varying degrees. Satiety was assessed upon arrival to the laboratory, before subjects consumed the pre-exercise beverage, and again immediately before beginning the constant-load phase. Satiety was assessed with a 1-100 mm visual analog scale.

Blood Glucose and Lactate Concentrations: Blood was collected via fingerstick blood samples immediately prior to consuming the pre-exercise beverage, 30 min after beverage consumption, immediately prior to the exercise trial, at minute 13 of the constant-load phase, and at 20 km of the time trial. Blood glucose and lactate were measured using automated instrumentation (YSI 2300 STAT glucose/lactate analyzer; YSI Life Sciences, Yellow Springs, OH), and required ~ 0.125 ml of blood at each time point measured.

Dietary and Exercise Control

Subjects recorded their dietary intake over the 24 hr before the first experimental time trial and were asked to replicate this 24 hr diet prior to each time trial. In addition, consistent dietary patterns 72 hr before each trial were requested. Subjects were asked to refrain from heavy exercise 48 hr pre-trial, alcohol and tobacco 24 hr pre-trial, caffeine 12 hr pre-trial, and were asked to fast after consuming a standardized beverage after their last meal of the day the night before each trial. Subjects were also asked to maintain consistent exercise habits between trials, and to record physical activity 72 hr before trials. Exercise and dietary logs were analyzed to assess consistency between trials.

Statistical Analyses

Magnitude-based inferences were used to compare effects between the four treatment trials for all dependent measures, using methods described by Batterham and Hopkins (4). A threshold for ‘meaningful’ change was determined for each measure. A meaningful change in performance time was defined as $0.3 \times$ the coefficient of variation between repeated time-trials in cyclists, with $CV = 1.3\%$, as reported by Hopkins (27). Meaningful changes in other variables were defined as $0.2 \times$ standard deviation of the variable in the sample under control conditions. Using a published spreadsheet, the percent likelihood that treatments caused ‘meaningful’ changes in dependent measures in the population was determined; results are also reported using 90% confidence intervals (4,27). Semantic inferences are provided for observed effects based on the degree to which they are beneficial or harmful to performance or whether they are likely or not likely to have changed. Semantic inferences are listed as follows: $< 1\%$ = almost certainly no chance, $1-5\%$ = very unlikely, $5-25\%$ = unlikely, $25-75\%$ = possible, $75-95\%$ = likely, $95-99\%$ = very likely, and $> 99\%$ = almost certain. If the 90% confidence interval exceeded minimum thresholds for both a negative change and a positive change, the effect is classified as “unclear”.

Chapter 3
Manuscript

**Impact of Pre-Exercise Feedings with a Low or High Glycemic Index
on the Ergogenic Effects of Carbohydrate Mouth-Rinsing during Cycling**

Authors: Nikolai J. Hladick, Kevin P. Decker, Nicholas D. Luden, Christopher J. Womack, Michael J. Saunders

Institution: James Madison University, Harrisonburg, Virginia, 22802

Contacts: Nikolai J. Hladick, hladicnj@dukes.jmu.edu
Kevin P. Decker, deckerkp@dukes.jmu.edu
Nicholas D. Luden, ludennd@jmu.edu
Christopher J. Womack, womackcx@jmu.edu
Michael J. Saunders, saudemj@jmu.edu

Address of Correspondence:

Michael J. Saunders, Ph.D.
Department of Kinesiology
James Madison University
Harrisonburg, VA 22807

Phone:

Email: saudemj@jmu.edu

Abstract

Purpose: Carbohydrate (CHO) ingestion during exercise enhances performance in short endurance events (~ 1 hr) due to neural influences, as demonstrated by the efficacy of CHO mouth-rinsing during cycling. However, the magnitude of these neural effects may be blunted following pre-exercise CHO feedings. This study examined whether the glycemic index (GI) of a pre-exercise meal affected time-trial (TT) performance in cyclists using a CHO mouth-rinse during exercise. **Methods:** Eight cyclists (age: 24 ± 6 yr; $\text{VO}_{2\text{max}}$: $61 \pm 8 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$) completed 4 exercise trials, consisting of 15 min of constant-load cycling followed by a simulated 30-km TT. Treatments were: a) L-CHO: low GI CHO beverage pre-exercise ($1.5 \text{ g}\cdot\text{kg}^{-1}$ CHO, 120 min prior), CHO mouth rinsing during exercise (6.4% maltodextrin solution), b) H-CHO: high GI CHO beverage ($1.5 \text{ g}\cdot\text{kg}^{-1}$ CHO) pre-exercise, CHO mouth rinsing during-exercise, c) PL-CHO: placebo beverage pre-exercise, CHO mouth rinsing during exercise, and d) PL-PL: placebo beverage pre-exercise, placebo mouth rinsing during exercise. Blood glucose was measured before beverage consumption and at 30 and 120 min following ingestion. Physiological measurements (VO_2 , V_E , RER, HR, RPE, glucose, lactate, and gastrointestinal distress) were assessed during constant-load cycling and the TT. Magnitude-based qualitative inferences were used to assess differences in responses between trials. **Results:** Blood glucose differed among treatments 30 min post-feeding ($\text{H-CHO} > \text{L-CHO} > \text{PL-CHO} = \text{PL-PL}$), and was lower in H-CHO versus PL-CHO and PL-PL during subsequent exercise. Compared to PL-CHO, TT performance was faster in both L-CHO (-0.5 ± 0.8 min; “likely” beneficial) and H-CHO (-0.7 ± 0.7 min; “likely” beneficial), with no systematic differences between L-CHO and H-CHO. However, none

of the 3 mouth rinse trials were clearly different from the PL-PL trial. **Conclusions:** When using a CHO mouth rinse during exercise, CHO ingestion 2 hr prior to cycling enhanced TT performance versus exercise in the fasted state. The GI of the pre-exercise feeding did not systematically affect TT performance in cyclists using a CHO mouth-rinse. However, the impact of these findings is confounded by the lack of performance differences versus a control trial without CHO before or during exercise.

Introduction

Carbohydrate consumption during prolonged endurance exercise (≥ 2 hr) improves performance by increasing carbohydrate availability and oxidation (12,13,26,29,30,31,39,49). Carbohydrate consumption also improves performance in shorter, high intensity bouts of exercise (~ 1 hr, $> 80\%$ VO_{2max}) (2,29), but contributes minimally to carbohydrate oxidation under these conditions (9,37). It is believed that these ergogenic effects are the result of neural influences, as oral-pharyngeal receptors can detect the presence of carbohydrate, and activate brain areas related to reward and pleasure, thereby facilitating motor output (8,10,21). Accordingly, a number of studies have reported performance enhancement with carbohydrate mouth rinsing (without ingestion) during short, high-intensity bouts of exercise (3,10,15,18,35,36,41,42,43,44,47).

Pre-exercise feeding seems to blunt the ergogenic effect of carbohydrate mouth rinsing. Lane and colleagues reported that carbohydrate mouth rinsing improved cycling power output (versus placebo) by 3.4% after an overnight fast, but only 1.8% in the fed state (35). Overall performance, however, was best during trials in which subjects consumed carbohydrate before exercise and utilized a carbohydrate mouth rinse during exercise (35). The attenuation of the ergogenic effects of mouth rinsing in the fed state may be mediated by neurological responses to the body's metabolic environment. There is evidence to suggest that satiety prior to carbohydrate feeding affects neurological responses to the feeding, decreasing hypothalamic activation (23,48).

Based on this evidence, a strategy that maximizes the effects of both pre-exercise feeding and carbohydrate rinsing during exercise may be of value to athletes.

Manipulating the glycemic index (GI) of the pre-exercise meal could be a useful strategy in this regard. Compared to high GI meals of similar carbohydrate content, low GI meals prior to exercise result in reduced insulin responses (52,54), higher plasma glucose levels late in exercise (54), elevated plasma free fatty acid concentrations during exercise (52,54,60), and increased rates of fat oxidation during exercise (52,60). As such, low GI feedings prior to exercise have improved endurance performance over high GI feedings in some studies (14,33,34,38,53,59,61). Additionally, consuming high GI carbohydrates reduces activity in brain areas that process reward recognition and appetite regulation compared to consuming low GI carbohydrate (40), which may downregulate responsiveness to subsequent carbohydrate exposure.

Considering the metabolic and neural effects of low GI feedings, and the possible ergogenic effects of low GI pre-exercise meals, consuming carbohydrate with a low GI before using a carbohydrate mouth rinse during exercise may confer the greatest performance benefit. The purpose of this study was to investigate the influence of a low GI pre-exercise meal on the ergogenic effects of a carbohydrate mouth rinse protocol used during exercise. We hypothesize that consuming a low GI pre-exercise meal in combination with a carbohydrate mouth rinse will improve exercise performance over consuming a high GI pre-exercise meal under the same conditions.

Methods

Subjects

Eight well-trained cyclists between the ages of 18 and 45 were recruited from James Madison University and the Harrisonburg, VA community. Subjects had at least 2 years of experience in endurance cycling events; a $VO_{2max} \geq 50 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$; consistently trained over the past 2 months, defined as cycling an average of ≥ 3 days $\cdot\text{week}^{-1}$; and had completed at least 4 training sessions ≥ 2 hr in duration over the previous 2 months. Subjects gave written informed consent and were free of any disease or health complication that could have caused adverse effects during exercise or exercise testing. All protocols were approved by the James Madison University Institutional Review Board.

Study Design

We used a randomized double-blind placebo-controlled crossover design. Subjects performed 4 experimental trials, each consisting of a 15-min constant-load phase followed by a simulated 30-km cycling time trial on a Velotron cycle ergometer (Racermate, Inc., Seattle, WA) in the Human Performance Laboratory at James Madison University. The 4 trials were identical, other than the pre-exercise beverage and/or during-exercise mouth-rinse used, as shown in Table 1. Trial order was randomly counterbalanced across subjects and separated by ≥ 7 days each.

Table 1: Treatment Conditions

Condition	Pre-exercise beverage	During-exercise mouth-rinse
PL-PL	PL	PL
PL-CHO	PL	CHO
L-CHO	LGI	CHO
H-CHO	HGI	CHO

PL = placebo; LGI = low glycemic index; HGI = high glycemic index; CHO = carbohydrate

Preliminary Testing and Familiarization

Height and weight were measured on subjects' first visit to the laboratory. An incremental cycling test to volitional fatigue was performed to determine $\text{VO}_{2\text{max}}$. The test began with a 5 min warmup at 100 W on a Velotron ergometer, after which the subjects selected a workload that was subjectively sustainable for ~ 1 hr. Every 2 min the workload was increased by 25 W until volitional fatigue. A Moxus Modular Metabolic System (AEI Technologies, Pittsburgh, PA) was used to measure and record oxygen uptake (VO_2) and carbon dioxide production throughout the test. The $\text{VO}_{2\text{max}}$ value was recorded as the highest 30 second average VO_2 value during the test. Heart rate was measured throughout the test using a Polar heart rate monitor.

Subjects completed a familiarization trial prior to the experimental trials. During this trial, subjects completed a 15-min constant-load phase followed by a 30-km time trial, as described below, with the exceptions that a) no fingerstick blood samples were obtained, b) no pre-exercise beverage was provided, and c) subjects rinsed with water instead of a carbohydrate solution during exercise. A familiarization trial was used so subjects could learn the testing protocol and become comfortable using a cycle ergometer in a laboratory setting, thereby minimizing any learning effects that had the potential to confound results.

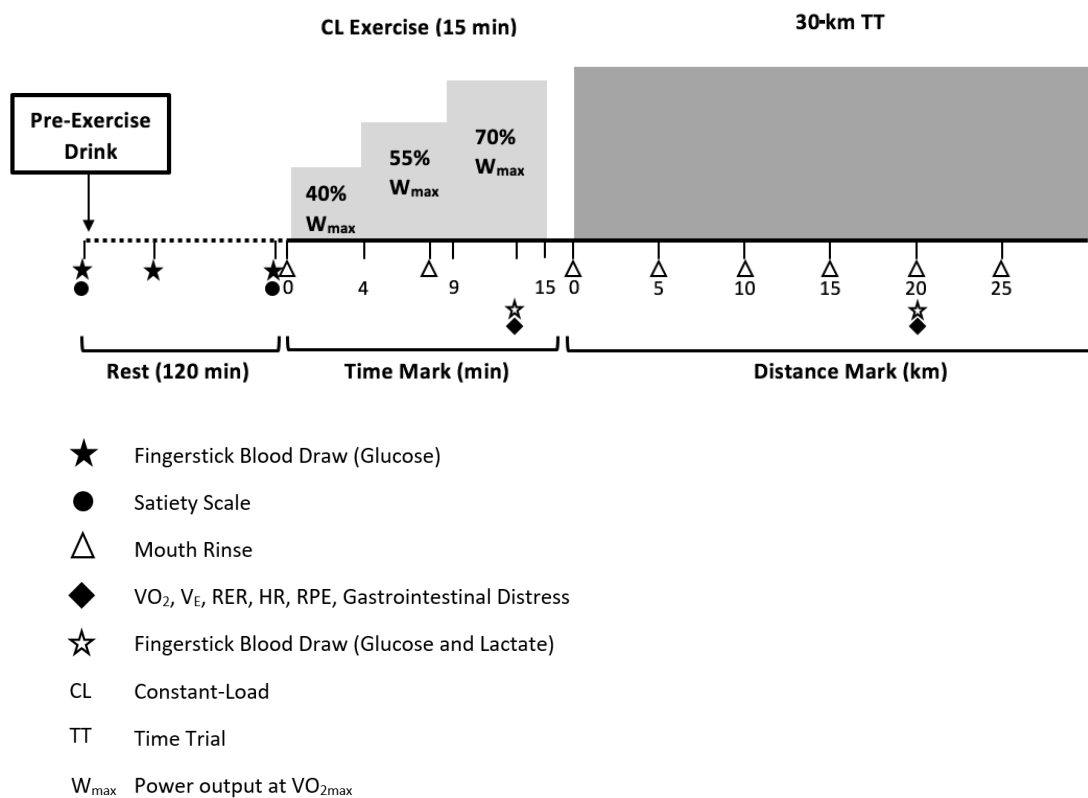
Experimental Protocol

The experimental protocol is displayed in Figure 1. Subjects arrived at the laboratory after an overnight fast. A fingerstick blood sample (~ 0.25 ml) was obtained and subjects consumed the pre-exercise beverage at a relatively constant rate over the

course of ~ 2 min. Two more fingerstick blood samples were obtained, one at 30 min and one at 120 min after consumption of the pre-exercise beverage. Two hr after consuming the pre-exercise beverage, subjects began the constant-load phase. The first 4 min of this phase were performed at 40% of the workload corresponding to subjects' $\text{VO}_{2\text{max}}$ (W_{max}), the second 5 min were performed at 55% W_{max} , and the final 6 min were performed at 70% W_{max} . The constant-load phase provided subjects with a progressive warmup prior to the time trial and allowed for comparison of physiological measurements at the same workload and time point between trials (i.e. independent of potential differences in pacing). The simulated 30-km time trial began about 2-3 min after the constant-load phase. Subjects were asked to give a maximal effort to complete the distance in the shortest possible amount of time, and to treat each trial as a competitive event. Other than distance completed, no feedback was provided to subjects during performance trials.

During the constant-load phase, subjects rinsed their mouths with 25 ml of either a carbohydrate solution or an artificially sweetened, non-caloric placebo at minute 0 and minute 7.5. During the 30-km performance trial, subjects rinsed every 5 km beginning at 0 km. Subjects rinsed a total of 8 times throughout each trial: twice during the constant-load phase and six times during the time trial. Subjects rinsed the solution around their entire mouth for 5 seconds, before expectorating the solution back into the cup provided. Subjects were instructed to avoid swallowing any of the mouth rinse solution.

Figure 1: Experimental Protocol



Pre-Exercise Beverages and Mouth Rinse Solutions

The pre-exercise beverages consisted of a low GI carbohydrate solution (LGI_{Pre}), a high GI carbohydrate solution (HGI_{Pre}), and a non-caloric placebo (PL_{Pre}). LGI_{Pre} consisted of 10 ml·kg⁻¹ of a slow-releasing high molecular weight 15% modified starch solution (UCAN Co., Woodbridge CT) providing 1.5 g CHO·kg⁻¹ body weight. HGI_{Pre} consisted of 10 ml·kg⁻¹ of a 15% maltodextrin solution providing 1.5 g CHO·kg⁻¹ body weight. Both beverages were made by mixing powdered forms of each carbohydrate with water, creating virtually tasteless solutions that were uniformly flavored with a non-caloric sweetener. PL_{Pre} consisted of 10 ml·kg⁻¹ of water flavored with the same non-caloric sweetener. All solutions were matched for taste and sweetness.

The mouth rinse solutions consisted of a maltodextrin solution (CHO_{Ex}) and a non-caloric placebo (PL_{Ex}). The CHO_{Ex} solution consisted of a 6.4% maltodextrin solution, while PL_{Ex} consisted of water flavored with a non-caloric artificial sweetener. Both solutions were prepared as previously described.

Dependent Measurements

Performance: Performance was assessed by recording the time to complete the 30-km time trial.

Oxygen Consumption (VO_2), Ventilation (V_E), and Respiratory Exchange Ratio (RER): Expired gas samples were measured with a metabolic cart (described previously) from minute 9 to minute 15 of the constant-load phase and for 5 min at 20 km of the time trial, after subjects had mouth rinsed. VO_2 , V_E , and RER were calculated from gas samples at each time-point via automated software. Values were averaged over the final 3 min of data collection, following 2-3 min of breathing equilibration.

Heart Rate: Heart rate was measured continuously throughout trials with an automated heart rate monitor consisting of a chest strap and wrist receiver, held by the researchers.

Ratings of Perceived Exertion (RPE), Gastrointestinal Discomfort, and Satiety: RPE was obtained at minute 13 of the constant-load phase and at 20 km of the time trial using Borg's 6-20 scale. Subjects rated gastrointestinal discomfort using ten 1-10 scales at the same time points. These scales assessed the following: stomach problems, gastrointestinal cramping, bloated feeling, diarrhea, nausea, dizziness, headache, belching, vomiting, and the urge to urinate or defecate. A score of 1 indicated absence of the symptom, while higher scores indicated that subjects were experiencing the symptom to varying degrees. Satiety was assessed upon arrival to the laboratory, before subjects

consumed the pre-exercise beverage, and again immediately before beginning the constant-load phase. Satiety was assessed with a 1-100 mm visual analog scale.

Blood Glucose and Lactate Concentrations: Blood was collected via fingerstick blood samples immediately prior to consuming the pre-exercise beverage, 30 min after beverage consumption, immediately prior to the exercise trial, at minute 13 of the constant-load phase, and at 20 km of the time trial. Blood glucose and lactate were measured using automated instrumentation (YSI 2300 STAT glucose/lactate analyzer; YSI Life Sciences, Yellow Springs, OH), and required ~ 0.125 ml of blood at each time point measured.

Dietary and Exercise Controls

Subjects recorded their dietary intake over the 24 hr before the first experimental time trial and were asked to replicate this 24 hr diet prior to each time trial. In addition, consistent dietary patterns 72 hr before each trial were requested. Subjects were asked to refrain from heavy exercise 48 hr pre-trial, alcohol and tobacco 24 hr pre-trial, caffeine 12 hr pre-trial, and were asked to fast after consuming a standardized beverage after their last meal of the day the night before each trial. Subjects were also asked to maintain consistent exercise habits between trials, and to record physical activity 72 hr before trials. Exercise and dietary logs were analyzed to assess consistency between trials.

Statistical Analyses

Magnitude-based inferences were used to compare effects between the four treatment trials for all dependent measures, using methods described by Batterham and Hopkins (4). A threshold for 'meaningful' change was determined for each measure. A meaningful change in performance time was defined as 0.3 x the coefficient of variation

between repeated time-trials in cyclists, with $CV = 1.3\%$, as reported by Hopkins (25). Meaningful changes in other variables were defined as $0.2 \times$ standard deviation of the variable in the sample under control conditions. Using a published spreadsheet, the percent likelihood that treatments caused ‘meaningful’ changes in dependent measures in the population was determined; results are also reported using 90% confidence intervals (4,25). Semantic inferences are provided for observed effects based on the degree to which they are beneficial or harmful to performance or whether they are likely or not likely to have changed. Semantic inferences are listed as follows: $< 1\%$ = almost certainly no chance, $1-5\%$ = very unlikely, $5-25\%$ = unlikely, $25-75\%$ = possible, $75-95\%$ = likely, $95-99\%$ = very likely, and $> 99\%$ = almost certain. If the 90% confidence interval exceeded minimum thresholds for both a negative change and a positive change, the effect is classified as “unclear”.

Results

Subject Demographics

Eleven trained cyclists were recruited from James Madison University and the Harrisonburg, VA community. Two individuals withdrew before completing all trials due to issues unrelated to the study and one individual was dismissed for not adhering to study protocols, resulting in complete data from eight subjects. Subject demographics were as follows: two females, six males; age, 24 ± 6 yr; $\text{VO}_{2\text{max}}$, $61 \pm 8 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$; height, 176 ± 6 cm; weight, 75 ± 12 kg.

Responses Following Pre-Exercise Feeding

Blood Glucose: Pre-exercise blood glucose responses are displayed in Figure 2. Blood glucose 30 min post-feeding was different between treatments as follows: H-CHO > L-CHO > PL-CHO = PL-PL (inferences shown in figure legend). At 120 min, blood glucose was higher in L-CHO versus PL-CHO and PL-PL, with no clear effects between other treatments.

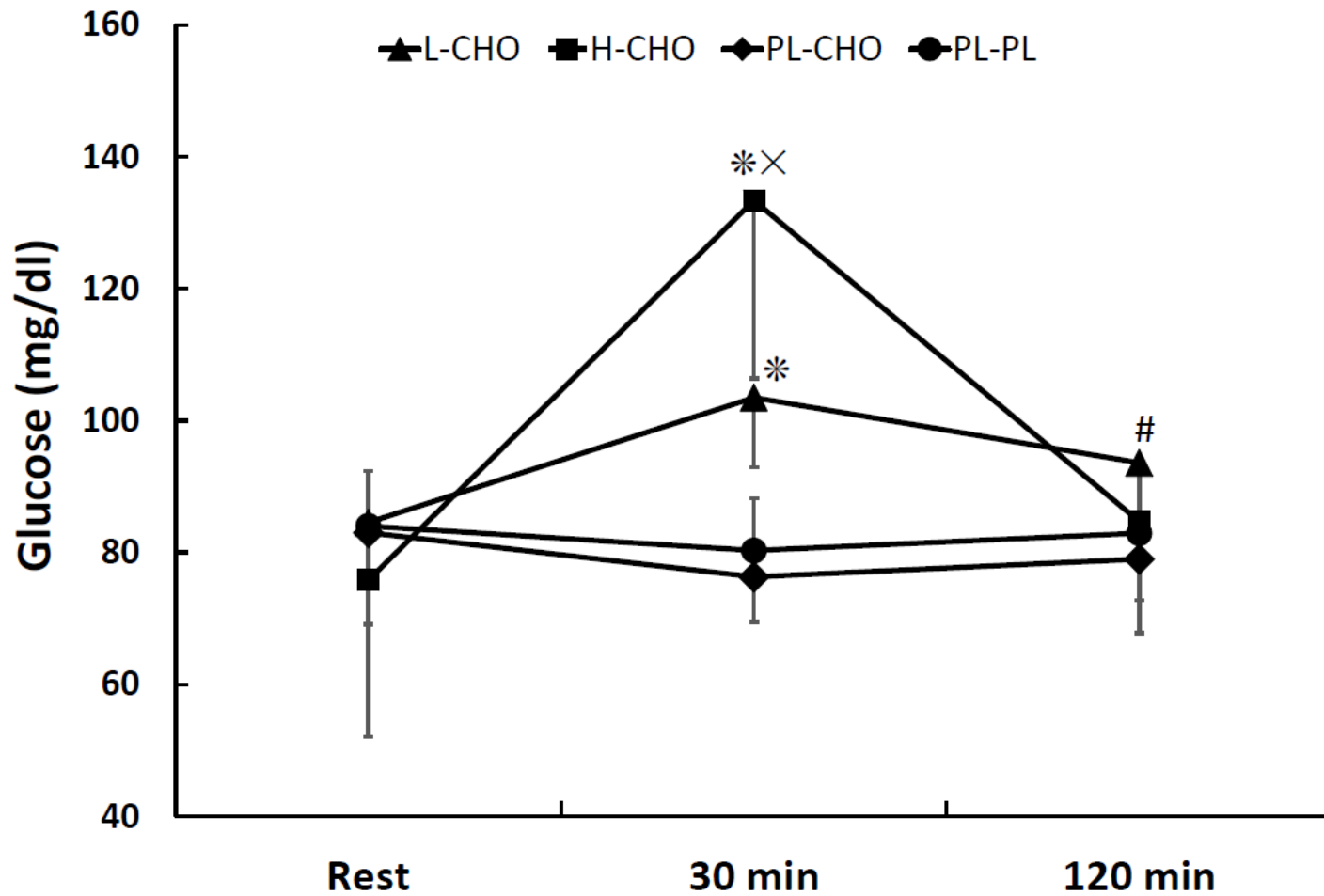
Satiety: Satiety responses are displayed in Table 2. Changes in satiety scores between all trials were “unclear”.

Table 2: Satiety Ratings Before and After Beverage Consumption

Treatment	Pre	120 min
L-CHO	52 ± 22	65 ± 20
H-CHO	48 ± 22	60 ± 22
PL-CHO	65 ± 17	74 ± 19
PL-PL	47 ± 20	67 ± 14

Data are presented as mean \pm SD. All treatment effects were “unclear”, arbitrary units.

Figure 2: Effects of Treatment Beverages on Blood Glucose Responses



Data are presented as mean and SD. * = "most likely" higher than PL-CHO and PL-PL, X = "very likely" higher than L-CHO, # = "very likely" higher than PL-CHO and "likely" higher than PL-PL.

Physiological Responses during Constant-Load Exercise

Constant-load physiological data are displayed in Table 3, and summarized in the text below.

Metabolic Measurements: VO_2 in L-CHO was “possibly” lower compared to both H-CHO and PL-PL. V_E was “possibly” higher in H-CHO than L-CHO. RER was “possibly” higher in both H-CHO and L-CHO compared to PL-PL. All other treatment comparisons in metabolic measures were “trivial/unclear”.

HR and RPE: Compared to PL-CHO, HR was “possibly” and “likely” higher in H-CHO and L-CHO respectively, while other HR comparisons were “unclear”. RPE was “likely” higher in H-CHO than PL-CHO, while all other RPE comparisons were “unclear”.

Blood Glucose and Lactate: Blood glucose was “most likely” and “likely” lower in H-CHO versus PL-CHO and PL-PL respectively. Blood glucose was “likely” lower in L-CHO than PL-CHO and was “unclear” between L-CHO and PL-PL. Glucose responses between H-CHO and L-CHO were “unclear”, while glucose was “likely” lower in PL-PL compared to PL-CHO. All lactate comparisons were “unclear”.

Physiological Responses during Time Trial

Time trial physiological data are displayed in Table 4, and summarized in the text below.

Metabolic Measurements: VO_2 was “possibly” lower in H-CHO compared to L-CHO and PL-PL and “possibly” higher in L-CHO compared to PL-CHO. RER was “likely” higher in L-CHO versus PL-CHO and PL-PL, and in H-CHO versus PL-PL. Comparisons of all other metabolic responses between treatments were “unclear”.

HR and RPE: HR was “likely” lower in H-CHO compared to L-CHO and “possibly” lower in H-CHO than PL-CHO. All RPE comparisons were “unclear”.

Blood Glucose and Lactate: Blood glucose was “possibly” lower in H-CHO compared to both L-CHO and PL-PL, with other glucose comparisons being “unclear”. All lactate comparisons were “unclear”.

Performance Time: Performance times and treatment differences in performance are displayed in Table 5. Due to methodological issues in two trials, the number of subjects included in the analyses differed between specific treatment comparisons (as indicated in the table). Differences in performance time were “unclear” between L-CHO and H-CHO. L-CHO and H-CHO were “likely” faster than PL-CHO. Comparisons between L-CHO/H-CHO/PL-CHO and PL-PL were unclear.

Ratings of Perceived Gastrointestinal Distress

There were no systematic differences in ratings of perceived gastrointestinal distress between trials. Mean gastrointestinal distress scores in each category were ≤ 2 out of 10. Only one subject reported a moderate/severe score for upper gastrointestinal distress symptoms with a 5 out of 10 for the “belching” category; this occurred at the 20 km mark of the L-CHO time trial

Table 3: Physiological Responses during Constant-Load Exercise

Measure	L-CHO	H-CHO	PL-CHO	PL-PL
VO ₂ (ml·kg ⁻¹ ·min ⁻¹)	42.8 ± 5.1 ^(P-P)	44.2 ± 7.3 ^(P-L)	43.8 ± 5.2	44.4 ± 5.7
V _E (L·min ⁻¹)	90.2 ± 14.8 ^(P-H)	93.0 ± 19.8	92.0 ± 15.7	91.5 ± 15.9
RER	0.95 ± 0.04 ^(P-P)	0.94 ± 0.06 ^(P-P)	0.93 ± 0.08	0.93 ± 0.06
HR (bpm)	159 ± 11 ^(L-C)	156 ± 14 ^(P-C)	151 ± 12	158 ± 13
RPE	13 ± 2	13 ± 1 ^(L-C)	12 ± 2	13 ± 1
Glucose (mg·dl ⁻¹)	69.0 ± 10.8 ^(L-C)	61.1 ± 8.3 ^{(ML-C)(L-P)}	79.8 ± 9.5 ^(L-P)	73.3 ± 13.6
Lactate (mmol·l ⁻¹)	2.76 ± 1.14	2.57 ± 1.19	2.33 ± 1.35	2.52 ± 1.32

Data are presented as mean ± SD. ^{P-P} = “possibly” different than PL-PL, ^{P-L} = “possibly” different than L-CHO, ^{P-H} = “possibly” different than H-CHO, ^{L-C} = “likely” different than PL-CHO, ^{P-C} = “possibly” different than PL-CHO, ^{ML-C} = “most likely” different than PL-CHO, ^{L-P} = “likely” different than PL-PL; all other comparisons were “unclear” or “trivial”.

Table 4: Physiological Responses during 30-km Time Trial

Measure	L-CHO	H-CHO	PL-CHO	PL-PL
VO ₂ (ml·kg ⁻¹ ·min ⁻¹)	40.6 ± 5.45 ^(P-C)	39.7 ± 4.30 ^{(P-L)(P-P)}	39.8 ± 5.74	41.3 ± 6.63
V _E (L·min ⁻¹)	78.6 ± 20.46	80.8 ± 11.40	78.7 ± 9.07	81.5 ± 11.30
RER	0.86 ± 0.04 ^{(L-C)(L-P)}	0.85 ± 0.04 ^(L-P)	0.83 ± 0.04	0.83 ± 0.03
HR (bpm)	163 ± 16 ^(L-H)	156 ± 17 ^(P-C)	162 ± 14	161 ± 13
RPE	16 ± 2	15 ± 1	16 ± 2	16 ± 1
Glucose (mg·dl ⁻¹)	72.7 ± 8.4 ^(P-H)	66.9 ± 7.7 ^(P-P)	74.9 ± 20.0	74.0 ± 18.1
Lactate (mmol·l ⁻¹)	2.53 ± 1.25	2.37 ± 1.22	2.30 ± 0.65	2.27 ± 1.13

Data are presented as mean ± SD. ^{P-C} = “possibly” different than PL-CHO, ^{P-L} = “possibly” different than L-CHO, ^{P-P} = “possibly” different than PL-PL, ^{L-C} = “likely” different than PL-CHO, ^{L-P} = “likely” different than PL-PL, ^{L-H} = “likely” different than H-CHO, ^{P-H} = “possibly” different than H-CHO; all other comparisons were “unclear” or “trivial”.

Table 5: Performance Times and Treatment Differences

PL-PL v L-CHO (N = 7)	PL-PL v H-CHO (N = 7)	PL-PL v PL-CHO (N = 6)
55.9 ± 3.1; 55.9 ± 3.4	55.9 ± 3.1; 55.7 ± 3.0	55.3 ± 3.0; 56.0 ± 3.0
0.0 (0.9)	0.1 (0.9)	-0.7 (1.2)
31/35/34	23/35/41	77/14/9
unclear	unclear	unclear

H-CHO v L-CHO (N = 8)	PL-CHO v L-CHO (N = 7)	PL-CHO v H-CHO (N = 7)
55.9 ± 2.9; 56.1 ± 3.2	56.5 ± 3.0; 55.9 ± 3.4	56.5 ± 3.0; 55.8 ± 3.1
-0.2 (0.5)	0.5 (0.8)	0.7 (0.7)
45/45/10	5/17/78	2/9/90
unclear	likely harmful	likely harmful

Performance times are presented as mean ± SD; treatment differences in performance time (min) are presented as mean difference (± 90% confidence interval), % beneficial/trivial/harmful, and qualitative inference.

Discussion

The purpose of this study was to examine the effects of pre-exercise carbohydrate ingestion, and the GI of this feeding, on 30-km cycling performance when using a carbohydrate mouth rinse protocol during exercise. We found that consuming carbohydrate 2 hr prior to a cycling time trial that included carbohydrate mouth rinsing resulted in better performance than fasting before exercise in the same situation. We also found that the GI of the pre-exercise feeding had no systematic effect on exercise performance when mouth rinsing during a cycling time trial lasting ~ 1 hr. The outcome of our PL-PL trial (i.e. similar performance to fasted/carbohydrate mouth rinse and fed/carbohydrate mouth rinse trials) is difficult to explain, and may be an anomalous finding. The comparatively fast performance time in this trial could be a result of measurement error or researcher error. For example, subject weight was incorrectly entered into the cycle ergometer software in two trials, in a manner which would have enhanced the average times for this trial. The data included has removed these subjects from analysis, and the resultant decrease in statistical power could have influenced this result. There may also be other, unknown reasons for the unexpected outcome, such as inconsistencies in subject behaviors which could have favored the PL-PL trial. For this reason, further discussion of our results will focus mainly on data from the other three trials, which all included carbohydrate mouth rinses.

There were no systematic differences in performance between the low and high GI pre-exercise feeding trials. In accordance with this outcome, other studies have reported no performance benefits from manipulating the GI of a pre-exercise feeding (7,11,16,19,20,28,32,33,50,51,57,58), though these studies did not include carbohydrate

mouth rinsing. However, other studies (also without carbohydrate mouth rinsing protocols) have reported enhanced performance following low GI pre-exercise feedings (14,33,34,38,53,59,61). The varied outcomes between studies may be related to differences in feeding protocols. The amount of carbohydrate consumed before exercise in glycemic index studies ranged from 0.8 g·kg⁻¹ carbohydrate to 2 g·kg⁻¹ carbohydrate, with some studies feeding a fixed amount of carbohydrate to subjects, regardless of bodyweight. The time between feeding and exercise has also varied widely, as rest periods predominantly ranged from 30 min to 3 hr, with one study (16) feeding only 15 g of carbohydrate immediately before exercise. Additionally, the composition of pre-exercise meals has included both whole foods and modified starches dissolved in water.

Multiple factors may explain the ergogenic effects of low GI pre-exercise feedings observed in some prior studies. One factor is more stable blood glucose levels prior to- and during-exercise, compared to high GI feedings (52,54,60). High GI meals result in large increases in blood glucose compared to low GI meals or fasting, with levels peaking around 15-30 min post-prandially (54). During exercise, blood glucose levels after high GI feedings may drop below those seen after low GI feedings, and may remain low throughout exercise (54,60). This outcome was observed in the present study, as blood glucose levels were highest in the H-CHO trial 30 min post-feeding, but dropped to levels lower than the other treatments during exercise. Another factor that could influence performance is increased fat oxidation after low GI feedings versus high GI feedings (52,54,60). This shift in substrate oxidation relative to pre-exercise meal GI may spare muscle glycogen, allowing for higher intensity efforts late in exercise, or for a longer duration of exercise. Accordingly, Thomas et al. (54) observed increased blood

glucose late in exercise in low GI versus high GI trials, in addition to increased free fatty acid concentrations in the blood, suggesting glycogen was spared earlier in exercise.

Glycogen sparing is presumably beneficial during exercise bouts in which glycogen reserves are the limiting factor, which is the case in longer bouts of exercise. Glycogen depletion is not usually a limiting factor in exercise bouts lasting ~ 1 hr. Indeed, the shortest bout of exercise in studies that reported performance improvement with a low GI pre-exercise meal was about 90 min, when glycogen reserves may begin to affect performance (22). Additionally, multiple studies that reported no performance improvement with a low GI pre-exercise meal had exercise durations less than 1 hr (28,50,51). Thus, the duration of the exercise bout in our study may explain why we observed no ergogenic effect of a low GI pre-exercise feeding.

Based on previous studies (23,40,48), we hypothesized that the rapid and large increase in blood glucose caused by a high GI feeding would desensitize brain areas that are responsive to carbohydrate mouth rinsing, attenuating the ergogenic effect of the rinse. If this were true, then performance would have been faster in L-CHO than H-CHO. Performance differences between these trials, however, were unclear, suggesting that either 1) these brain areas are not desensitized when blood glucose increases, or 2) the 2 hr postprandial period was enough time for these brain areas to regain sensitivity. Moreover, we may have lacked sufficient statistical power to detect any changes between these conditions, if they are in fact different. Further studies with more subjects and brain imaging would help elucidate the relationship between pre-exercise blood glucose and the efficacy of carbohydrate mouth rinsing.

Our finding that pre-exercise feedings enhance performance when using a carbohydrate mouth rinse during exercise is in agreement with previous studies that used similar protocols. Lane et al. (35) had subjects complete as much work as possible in 60 min on a cycle ergometer while mouth rinsing with a carbohydrate solution. Subjects either fasted or consumed $2.5 \text{ g}\cdot\text{kg}^{-1}$ carbohydrate 2 hr before exercise (35). Relative to a placebo trial, subjects completed more work in the mouth rinse trials; the most work (i.e. the best performance) was completed in the trial that entailed feeding before mouth rinsing during exercise (35). Fares et al. (18) reported similar results in a cycling time to exhaustion test, as their subjects performed best when rinsing during exercise after consuming a pre-exercise meal. Though outcomes in these studies and the current study were similar, the protocols used differed slightly in pre-exercise meal composition and pre-exercise meal timing in relation to exercise.

The beneficial effects of feeding before exercise of this duration may be related to endogenous carbohydrate availability during subsequent exercise. Ali and colleagues (1) showed this by using a glycogen depletion protocol the night before a cycling test in which subjects completed a set amount of work in as little time as possible while fasted (1). Throughout the performance test, subjects either rinsed with or ingested a carbohydrate solution (1). Power output was significantly higher, and subjects rode $\sim 5\%$ faster, in the carbohydrate ingestion trial compared to the rinse and placebo trials, suggesting that carbohydrate availability (i.e. from carbohydrate ingested before or during exercise) may be an important factor in realizing the potential neural benefits of carbohydrate (1).

Contrary to our findings and those of Lane et al. (35) and Fares et al. (18), Trommelen and colleagues (56) reported no overall difference in performance when mouth rinsing in a fed versus fasted state. Trommelen et al. (56) hypothesized that the magnitude of signaling responses from carbohydrate sensors in the mouth may be lessened with higher liver glycogen stores, brought about by pre-exercise feeding. Beelen and colleagues (6) reported similar results, though they investigated performance with a carbohydrate mouth rinse in the post-prandial state only. Subjects in that study performed no differently when mouth rinsing with a carbohydrate solution or a placebo solution after consuming $2.36 \text{ g}\cdot\text{kg}^{-1}$ carbohydrate 2 hr before completing a set amount of work on a cycle ergometer (6).

Our finding that pre-exercise carbohydrate ingestion enhances endurance performance amongst the trials with a carbohydrate mouth rinse is generally consistent with previous studies that have investigated the effects of pre-exercise feeding with no mouth rinsing during exercise. Most pre-exercise carbohydrate feeding studies reported performance improvements in exercise bouts longer than 2 hr. For example, Sherman and colleagues (46) reported improved cycling time trial performance after a 90 min steady state ride when subjects consumed carbohydrate 1 hr prior to cycling. Schabert et al. (45) also observed improved performance in a cycling time to exhaustion test 1 hr after carbohydrate consumption, with other researchers similarly confirming the ergogenic effects of carbohydrate consumption before prolonged exercise (17,55). Performance enhancement with carbohydrate consumption before exercise lasting ≥ 2 hr is likely due to greater substrate availability during exercise. Our results and a few others

(17,18,35) suggest that pre-exercise carbohydrate consumption can improve performance in exercise bouts lasting ~ 1 hr as well.

One potential explanation for the varied outcomes amongst the studies described above is the limited sensitivity of ergometer-based endurance tests to detect changes in performance due to treatments, as compared to changes due to normal physiological variability. In high level athletes, day-to-day variability in cycling time-trial performance is ~1-3% (25), and is presumably higher in athletes with less training and experience. Because the presumed effects of carbohydrate mouth rinsing on performance are relatively small (1-4% in most studies), and the available sample sizes for performance studies are also quite small, it is logical that performance differences are not detected in all studies due to low statistical power. It is possible that low statistical power affected the outcome of our study, as we detected no differences between the PL-PL and PL-CHO trials.

As mentioned previously, the composition of the pre-exercise meal has varied throughout studies investigating the performance effects of pre-exercise feeding, with some studies using whole foods and others using liquid feedings. Solid and liquid meals may elicit different metabolic and physiological responses upon ingestion, with solid meals producing greater increases in metabolic rate than liquid meals (24). Solid and liquid meals may also differ slightly in some of the cardiovascular responses they elicit upon ingestion (24). Additionally, subjects cannot be blinded to the fact that they consumed calories prior to exercise when whole foods were used, whereas subjects in our study did not know when they were consuming calories prior to exercise. Thus, our data add to the prior literature by demonstrating that pre-exercise carbohydrate intake *per se*

can have ergogenic effects during subsequent cycling when using a carbohydrate mouth rinse.

In the present study, we used a modified starch that has been associated with gastrointestinal distress during exercise (5). Though only one subject in our study reported a moderate/severe upper gastrointestinal symptom during exercise (belching), this symptom was reported in the modified starch trial. Baur et al. (5) had subjects consume 60 g of the same modified starch 30 min before exercise, followed by either 30 g·hr⁻¹ or 60 g·hr⁻¹ of the starch during exercise. After modified starch consumption, subjects reported nausea and abdominal cramping, which may have negatively affected exercise performance in a repeated sprint cycling protocol in the 30 g·hr⁻¹ trial (5). Our subjects consumed 1.5 g·kg⁻¹ of the modified starch 2 hr prior to exercise, with no further ingestion during exercise. Considering the collective results of Baur et al. (5) and the present study, it seems that gastrointestinal distress may be less likely if the modified starch is consumed ≥ 30 min prior to exercise, and none is consumed during exercise. However, because CHO ingestion is desirable during longer endurance events, future research should examine the optimal combinations of pre- and during-exercise carbohydrate sources with respect to gastrointestinal tolerance.

In summary, we found that carbohydrate consumption 2 hr prior to exercise was ergogenic during subsequent cycling exercise that included carbohydrate mouth rinsing, in comparison to exercising in a fasted state under the same conditions. These and other data suggest that carbohydrate consumption before exercise may be a desirable strategy to enhance performance in shorter endurance events (~ 1 hr), even when carbohydrate mouth rinsing is performed during exercise. Additionally, we observed no systematic

difference in performance between pre-exercise feedings with either a high or low GI. Although this suggests that the GI of pre-exercise feedings is not particularly important prior to short endurance events, this finding should not be generalized to longer endurance events, as minor metabolic differences existed between L-CHO and H-CHO that may have become meaningful if exercise had continued beyond ~ 1 hr.

Manuscript References

1. Ali A, Yoo MJ, Moss C, Breier BH. Carbohydrate mouth rinsing has no effect on power output during cycling in a glycogen-reduced state. *J Int Soc Sports Nutr.* 2016; 13:19.
2. Anantaraman R, Carmines AA, Gaesser GA, Weltman A. Effects of carbohydrate supplementation on performance during 1 hour of high-intensity exercise. *Int J Sports Med.* 1995; 16(7):461-5.
3. Bastos-Silva VJ, Melo Ade A, Lima-Silva AE, Moura FA, Bertuzzi R, de Araujo GG. Carbohydrate mouth rinse maintains muscle electromyographic activity and increases time to exhaustion during moderate but not high-intensity cycling exercise. *Nutrients.* 2016; 8(3):49.
4. Batterham AM, Hopkins WG. Making meaningful inferences about magnitudes. *Int J Sports Physiol Perform.* 2006; 1(1):50-7.
5. Baur DA, Vargas FDCS, Bach CW, Garvey JA, Ormsbee MJ. Slow-absorbing modified starch before and during prolonged cycling increases fat oxidation and gastrointestinal distress without changing performance. *Nutrients.* 2016; 8:392.
6. Beelen M, Berghuis J, Bonaparte B, Ballak SB, Jeukendrup AE, van Loon LJ. Carbohydrate mouth rinsing in the fed state: lack of enhancement of time-trial performance. *Int J Sport Nutr Exerc Metab.* 2009; 19(4):400-9.
7. Burke LM, Claassen A, Hawley JA, Noakes TD. Carbohydrate intake during prolonged cycling minimizes effect of glycemic index of preexercise meal. *J Appl Physiol.* 1998; 85(6):2220-6.
8. Carter JM, Jeukendrup AE, Jones DA. The effect of carbohydrate mouth rinse on 1-h cycle time trial performance. *Med Sci Sports Exerc.* 2004; 36(12):2107-11.
9. Carter JM, Jeukendrup AE, Mann CH, Jones DA. The effect of glucose infusion on glucose kinetics during a 1-h time trial. *Med Sci Sports Exerc.* 2004; 36(9):1543-50.
10. Chambers ES, Bridge MW, Jones DA. Carbohydrate sensing in the human mouth: effects on exercise performance and brain activity. *J Physiol.* 2009; 587(Pt 8):1779-94.
11. Chen YJ, Wong SHS, Chan COW, Wong CK, Lam CW, Siu PMF. Original paper: Effects of glycemic index meal and CHO-electrolyte drink on cytokine response and run performance in endurance athletes. *J Sci Med Sport.* 2009; 12:697-703.

12. Coyle EF, Coggan AR, Hemmert MK, Ivy JL. Muscle glycogen utilization during prolonged strenuous exercise when fed carbohydrate. *J Appl Physiol.* 1986; 61(1):165.
13. Coyle EF, Hagberg JM, Hurley BF, Martin WH, Ehsani AA, Holloszy JO. Carbohydrate feeding during prolonged strenuous exercise can delay fatigue. *J Appl Physiol Respir Environ Exerc Physiol.* 1983; 55(1):230.
14. DeMarco HM, Sucher KP, Cisar CJ, Butterfield GE. Pre-exercise carbohydrate meals: application of glycemic index. *Med Sci Sports Exerc.* 1999; 31(1):164-70.
15. Devenney S, Collins K, Shortall M. Effects of various concentrations of carbohydrate mouth rinse on cycling performance in a fed state. *Eur J Sport Sci.* 2016:1-6.
16. Earnest CP, Lancaster SL, Rasmussen CJ, et al. Low Vs. High Glycemic Index Carbohydrate Gel Ingestion during Simulated 64-km Cycling Time Trial Performance. *J Strength Cond Res.* 2004; 18(3):466-72.
17. El-Sayed M, Balmer J, Rattu AJM. Carbohydrate ingestion improves endurance performance during a 1 h simulated cycling time trial. *J Sports Sci.* 1997; 15(2):223-30.
18. Fares EJ, Kayser B. Carbohydrate mouth rinse effects on exercise capacity in pre- and postprandial States. *J Nutr Metab.* 2011; 2011:385962.
19. Febbraio MA, Stewart KL. CHO feeding before prolonged exercise: effect of glycemic index on muscle glycogenolysis and exercise performance. *J Appl Physiol.* 1996; 81(3):1115-20.
20. Febbraio MA. Effects of carbohydrate ingestion before and during exercise on glucose kinetics and performance. *J Appl Physiol.* 2000; 89(6):2220.
21. Gant N, Stinear CM, Byblow WD. Carbohydrate in the mouth immediately facilitates motor output. *Brain Res.* 2010; 1350:151-8.
22. Gollnick PD, Piehl K, Saltin B. Selective glycogen depletion pattern in human muscle fibers after exercise of varying intensity and at varying pedaling rates. *J Physiol.* 1974; 241:45-57.
23. Haase L, Cerf-Ducastel B, Murphy C. Cortical activation in response to pure taste stimuli during the physiological states of hunger and satiety. *Neuroimage.* 2009; 44(3):1008-21.
24. Habas ME, Macdonald IA. Metabolic and cardiovascular responses to liquid and solid test meals. *Br J Nutr.* 1998; 79:241-47.

25. Hopkins WG. How to interpret changes in an athletic performance test. *Sports Science*. 2004; 8:1-7.
26. Ivy JL, Milley W, Dover V, et al. Endurance improved by ingestion of a glucose polymer supplement. *Med Sci Sports Exere*. 1983; 15(6):466-71.
27. Jenkins DA, Wolever TMS, Taylo RH, et al. Glycemic index of foods: a physiological basis for carbohydrate exchange. *Am J Clin Nutr* 1981; 34:362-366.
28. Jentjens R, Jeukendrup A. Effects of pre-exercise ingestion of trehalose, galactose and glucose on subsequent metabolism and cycling performance. *Eur J Appl Physiol*. 2003; 88(5):459-65.
29. Jeukendrup A, Brouns F, Wagenmakers AJM, Saris WHM. Carbohydrate-electrolyte feedings improve 1 h time trial cycling performance. *Int J Sports Med*. 1997; 18(2):125-9.
30. Jeukendrup AE, Wagenmakers AJM, Stegen JH, et al. Carbohydrate ingestion can completely suppress endogenous glucose production during exercise. *Am J Physiol (Endocrinol Metab 39)* 1999; E672-E683.
31. Jeukendrup AE. Carbohydrate feeding during exercise. *Eur J Sport Sci*. 2008; 8(2):77-86.
32. Kern M, Heslin CJ, Rezende RS. Metabolic and Performance Effects of Raisins Versus Sports Gel as Pre-Exercise Feedings in Cyclists. *J Strength Con Res*. 2007; 21(4):1204-7.
33. Kirwan JP, Cyr-Campbell D, Campbell WW, Scheiber J, Evans WJ. Effects of moderate and high glycemic index meals on metabolism and exercise performance. *Metabolism*. 2001; 50(7):849-55.
34. Kirwan JP, O'Gorman D, Evans WJ. A moderate glycemic meal before endurance exercise can enhance performance. *J Appl Physiol*. 1998; 84(1):53-9.
35. Lane SC, Bird SR, Burke LM, Hawley JA. Effect of a carbohydrate mouth rinse on simulated cycling time-trial performance commenced in a fed or fasted state. *Appl Physiol Nutr Metab*. 2013; 38(2):134-9.
36. Luden ND, Saunders MJ, D'Lugos AC, et al. Carbohydrate mouth rinsing enhances high intensity time trial performance following prolonged cycling. *Nutrients*. 2016; 8(9):576.
37. McConell GK, Canny BJ, Daddo MC, Nance MJ, Snow RJ. Effect of carbohydrate ingestion on glucose kinetics and muscle metabolism during intense endurance exercise. *J Appl Physiol (1985)*. 2000; 89(5):1690-8.

38. Moore LJS, Midgley AW, Thurlow S, Thomas G, Mc Naughton LR. Original paper: Effect of the glycaemic index of a pre-exercise meal on metabolism and cycling time trial performance. *J Sci Med Sport*. 2010; 13:182-8.
39. Murray R, Paul GL, Seifert JG, Eddy DE. Responses to varying rates of carbohydrate ingestion during exercise. *Med Sci Sports Exerc*. 1991; 23(6):713-8.
40. Page KA, Chan O, Arora J, et al. Effects of fructose vs glucose on regional cerebral blood flow in brain regions involved with appetite and reward pathways. *JAMA*. 2013; 309(1):63-70.
41. Pottier A, Bouckaert J, Gilis W, Roels T, Derave W. Mouth rinse but not ingestion of a carbohydrate solution improves 1-h cycle time trial performance. *Scand J Med Sci Sports*. 2010; 20(1):105-11.
42. Rollo I, Cole M, Miller R, Williams C. Influence of mouth rinsing a carbohydrate solution on 1-h running performance. *Med Sci Sports Exerc*. 2010; 42(4):798-804.
43. Rollo I, Williams C, Gant N, Nute M. The influence of carbohydrate mouth rinse on self-selected speeds during a 30-min treadmill run. *Int J Sport Nutr Exerc Metab*. 2008; 18(6):585-600.
44. Rollo I, Williams C, Nevill M. Influence of ingesting versus mouth rinsing a carbohydrate solution during a 1-h run. *Med Sci Sports Exerc*. 2011; 43(3):468-75.
45. Schabort EJ, Bosch AN, Weltan SM, Noakes TD. The effect of a preexercise meal on time to fatigue during prolonged cycling exercise. *Med Sci Sports Exerc*. 1999; 31(3):464,471 8p.
46. Sherman WM, Peden MC, Wright DA. Carbohydrate feedings 1 h before exercise improves cycling performance. *Am J Clin Nutr*. 1991; 54(5):866-70.
47. Sinclair J, Bottoms L, Flynn C, et al. The effect of different durations of carbohydrate mouth rinse on cycling performance. *Eur J Sport Sci*. 2014; 14(3):259-64.
48. Smeets PA, de Graaf C, Stafleu A, van Osch MJ, van der Grond J. Functional MRI of human hypothalamic responses following glucose ingestion. *Neuroimage*. 2005; 24(2):363-8.
49. Smith JW, Zachwieja JJ, Péronnet F, et al. Fuel selection and cycling endurance performance with ingestion of [¹³C]glucose: evidence for a carbohydrate dose response. *J Appl Physiol (1985)*. 2010; 108(6):1520-9.
50. Sparks MJ, Selig SS, Febbraio MA. Pre-exercise carbohydrate ingestion: effect of the glycemic index on endurance exercise performance. *Med Sci Sports Exerc*. 1998; 30(6):844-849.

51. Stannard SR, Thompson MW, Brand Miller JC. The effect of glycemic index on plasma glucose and lactate levels during incremental exercise. *Int J Sport Nutr Exerc Metab.* 2000; 10:51-61.
52. Stevenson EJ, Williams C, Mash LE, Phillips B, Nute ML. Influence of high-carbohydrate mixed meals with different glycemic indexes on substrate utilization during subsequent exercise in women. *Am J Clin Nutr.* 2006; 84(2):354,360 7p.
53. Thomas DE, Brotherhood JR, Brand JC. Carbohydrate feeding before exercise: effect of glycemic index. *Int J Sports Med.* 1991; 12(2):180-6.
54. Thomas DE, Brotherhood JR, Miller JB. Plasma glucose levels after prolonged strenuous exercise correlate inversely with glycemic response to food consumed before exercise. *Int J Sport Nutr.* 1994; 4(4):361,373 13p.
55. Tokmakidis SP, Karamanolis IA. Effects of carbohydrate ingestion 15 min before exercise on endurance running capacity. *Appl Physiol Nutr Metab.* 2008; 33(3):441-9.
56. Trommelen J, Beelen M, Mullers M, Gibala MJ, van Loon LJ, Cermak NM. A Sucrose Mouth Rinse Does Not Improve 1-hr Cycle Time Trial Performance When Performed in the Fasted or Fed State. *Int J Sport Nutr Exerc Metab.* 2015; 25(6):576-83.
57. Wee S, Williams C, Gray S, Horabin J. Influence of high and low glycemic index meals on endurance running capacity. *Med Sci Sports Exerc.* 1999; 31(3):393,399 7p.
58. Wong SHS, Oi WC, Ya JC, Heng LH, Ching WL, Pak KC. Effect of Preexercise Glycemic-Index Meal on Running When CHO-Electrolyte Solution Is Consumed During Exercise. *Int J Sport Nutr Exerc Metab.* 2009; 19(3):222-42.
59. Wong SHS, Siu PM, Lok A, Chen YJ, Morris J, Lam CW. Effect of the glycaemic index of pre-exercise carbohydrate meals on running performance. *Eur J Sport Sci.* 2008; 8(1):23-33.
60. Wu C, Nicholas C, Williams C, Took A, Hardy L. The influence of high-carbohydrate meals with different glycaemic indices on substrate utilisation during subsequent exercise. *Br J Nutr.* 2003; 90(6):1049,1056 8p.
61. Wu CL, Williams C. A low glycemic index meal before exercise improves endurance running capacity in men. *Int J Sport Nutr Exerc Metab* 2006; 16:510-527.

Appendices

James Madison University – Department of Kinesiology Informed Consent

Purpose

You are being asked to volunteer for a research project conducted by Nikolai Hladick, Dr. Nick Luden, Dr. Mike Saunders, and Dr. Christopher Womack from James Madison University titled *Impact of the glycemic index of a pre-exercise feeding on the ergogenic effects of carbohydrate mouth-rinsing during cycling*.

The primary goal of this study is to determine the effect that pre-exercise beverages of differing glycemic indexes have on high intensity cycling performance when a carbohydrate mouth rinse is used during exercise.

Experimental Procedures

You will be asked to report to James Madison University's Human Performance Laboratory (Godwin 209) on 6 occasions, each separated by at least 7 days. These include one initial testing session, one familiarization trial, and four experimental exercise trials. The initial testing session will last approximately 1 hour and the familiarization trial will last approximately 1 hour and 30 minutes. Each experimental exercise trial will require approximately 3.5 hours. The total time commitment will be approximately 16 hours and 30 minutes.

Initial Exercise Testing Session – Visit 1 – 1 hour

You will be asked to complete short questionnaires related to your health history and exercise training, to determine whether you meet the criteria for participation and to rule out any health-related risk factors that would prevent you from participating in this study. During this process, you will be asked to share information concerning your lifestyle, training habits, and general health with the researchers. If you meet the participation criteria, your height and body weight will be measured and your maximal oxygen consumption (VO_{2max}) will be assessed with a test on a cycle ergometer. You will begin this test by cycling at a moderate intensity, after which the workload will be increased by 25 watts every 2 minutes until you are unable to continue due to fatigue (~10-20 min). Throughout the trial, you will breathe through a mouthpiece that is connected to a metabolic cart, in order to measure your oxygen consumption and other variables during exercise. Heart rate will be also be monitored continuously by a wearable heart rate monitor on your chest.

Familiarization Trial – Visit 2 – 1 hour and 30 minutes

During the familiarization trial you will be asked to complete a simulated 40 km cycling time trial on a cycle ergometer (~ 60 min). During the time trial you will be asked to rinse your mouth with water for 5 seconds every 5 km without swallowing. On two occasions during the trial (5 km and 30 km), you will have your oxygen consumption measured for 5 minutes, by wearing the mouthpiece described above. You will also be asked to rate your perceived effort and gastrointestinal discomfort (using a scale provided

by the researchers) at these time-points. Heart rate will be measured continuously via a wearable heart rate monitor on your chest.

Experimental Trials – Visits 3 through 6 – 3.5 hours each

You will report to the laboratory after an overnight fast (no food after dinner the night prior to the trial), and provide a small (5 ml) blood sample from a vein in your arm. Following the blood sample, you will consume a sports beverage, and then rest for two hours. Following the two hours of rest, a second 5 ml blood sample will be obtained, and then a simulated 40 km cycling time trial will be completed, as described above. You will be asked to give a maximal effort during each time trial and to treat it as a competitive event. During each trial you will also be asked to rinse your mouth with a sports drink for 5 seconds every 5 km without swallowing. You will receive all of the measurements described in the Familiarization Trial above (oxygen consumption, heart rate, perceived effort and gastrointestinal discomfort). You will also receive fingersticks at the same two time-points to obtain small blood samples (0.5 ml) from your finger. Each of the four experimental trials will include a different pre-exercise sports drink and a different combination of pre-exercise beverage and sports drink mouth rinse during exercise. The order in which you receive the different beverages and mouth rinses during the experimental trials will be randomly assigned.

Dietary and Exercise Controls

You will be asked to record your food intake for 24 hours prior to each experimental visit. After bringing the initial dietary record to the Human Performance Laboratory, you will be given a copy, and will be asked to replicate your food intake for the 24 hours before each subsequent visit. You will also be asked to record your physical activity/exercise during the 72 hours prior to each experimental trial and to maintain consistent physical activity/exercise patterns between trials. You will be asked to refrain from heavy exercise 48 hours pre-trial, alcohol and tobacco 24 hours pre-trial, caffeine 12 hours pre-trial, and will be asked to fast the night before each experimental visit (no food after dinner).

Risks

The risks associated with maximal exercise and maximal exercise testing are minimal in individuals who are considered healthy and at low risk for cardiovascular disease and cardiac events according to the American College of Sports Medicine. In order to participate in this study, you must be considered low risk after initial assessment via health history questionnaires. You are expected to be honest when filling out questionnaires and identifying any risk factors you may have. In the case of a cardiac or emergency event during exercise, an emergency plan is in place, including access to a phone to contact emergency personnel. At least one investigator at each testing session will be CPR certified, and an AED is present in the laboratory.

The cycling time trials may induce muscle fatigue and soreness both immediately after the trial and for 1-2 days following the visit. Gastrointestinal distress is a possibility when consuming sports drinks before intense exercise. However, this poses no threat to your health or safety, and will at most cause mild discomfort. In addition, you may stop

exercising at any point throughout the trials. The risks of blood sampling include slight discomfort, temporary minor bleeding, possibility for infection, and the possible transfer of blood-borne pathogens. Risks during blood sampling are considered to be minimal and OSHA safety protocols will be followed when handling blood samples. The researchers have completed JMU blood-borne pathogen training. In addition, the total amount of blood obtained throughout the study is very small [~ 11 ml per trial = 44 ml or 1.5 fluid ounces, which is $<10\%$ of the amount given when donating blood in a single session (approximately 1 pint, or 473 ml)].

Benefits

Participating in this study includes receiving a free assessment of maximal oxygen consumption (which typically cost $> \$100$ at commercial testing facilities). You will also be contributing to the first study investigating the interaction between pre-exercise meal glycemic index and the efficacy of a carbohydrate mouth rinse during high intensity exercise. In addition, participants will receive a monetary incentive of \$150 for completion of the study. Participants who do not complete the entire study will receive a prorated payment of \$35 for each of the experimental time-trials completed (i.e. trials 3-6 above).

Inquiries

If you have any questions or concerns, please contact Dr. Mike Saunders at saundemj@jmu.edu and (540) 568-8121 or Dr. Nicholas Luden at ludennd@jmu.edu and (540) 568-4069.

Questions about Your Rights as a Research Subject

Dr. David Cockley
Chair, Institutional Review Board
James Madison University
(540) 568-2834
cocklede@jmu.edu

Confidentiality

Data obtained in this study will be kept confidential and your name will not be identified with individual data. An identification code will be assigned to each participant in order to avoid identifying participant names with data, which will be kept in a locked cabinet. Once the study has been completed, any information connecting participants to their information/data will be destroyed. The researchers retain the right to use and publish non-identifiable data. Final aggregate results will be made available to you upon request.

Freedom of Consent

Your participation is entirely voluntary. You are free to choose not to participate. Should you choose to participate, you can withdraw at any time without consequences of any kind.

I have read this consent form and I understand what is being requested of me as a participant in this study. I freely consent to participate. I have been given satisfactory answers to my questions. The investigator provided me with a copy of this form. I certify that I am at least 18 years of age.

Name of Subject (Printed)

Name of Researcher (Printed)

Name of Subject (Signed)

Name of Researcher (Signed)

Date

Date

CYCLISTS WANTED FOR PERFORMANCE NUTRITION STUDY

The Human Performance Laboratory at JMU will be conducting a study examining the effects of a carbohydrate sports drink and mouth rinse protocol on cycling performance.

Who Are We Looking For?

- 18-45 year old male or female
- Trained cyclists (at least 2 years of cycling experience)

What Will You Be Asked To Do?

- Complete preliminary paperwork/screening
- Participate in 6 exercise sessions:
 - One 10-20 min fitness test to assess peak cardiorespiratory fitness
 - One familiarization trial (~60 min)
 - Four 1-hour simulated cycling time trials
- Receive laboratory assessments before and during exercise sessions (including measurement of oxygen consumption and small blood samples)

What Are the Benefits and Incentives of Participation?

- Free assessment of aerobic capacity (VO_{2max})
- \$150 for study completion

For more information, please contact Dr. Mike Saunders at saundemj@jmu.edu (540-568-8121), or Nikolai Hladick at hladicnj@dukes.jmu.edu (609-577-8499)

Subject Prescreening Information**Subject #:** _____**Age:** _____ **Sex:** _____**Height:** _____ **Weight:** _____**Exercise Habits over the Past 2 Months:****Avg. # of days of aerobic exercise per week:** _____**Avg. # of days of cycling per week:** _____**Total # of bike rides > 2 hrs:** _____**Describe your training/cycling history (i.e. how long, and at what level have you been training/cycling):** _____**Allergies:** _____**Food allergies/sensitivities:** _____**Medications used:** _____

AHA/ACSM Health/Fitness Facility Pre-Participation Screening Questionnaire

Assess your health status by marking all *true* statements

History

You have had:

- a heart attack
- heart surgery
- cardiac catheterization
- coronary angioplasty (PTCA)
- pacemaker/implantable cardiac defibrillator/rhythm disturbance
- heart valve disease
- heart failure
- heart transplantation
- congenital heart disease

If you marked any of these statements in this section, consult your physician or other appropriate health care provider before engaging in exercise. You may need to use a facility with a **medically qualified staff**.

Symptoms

- You experience chest discomfort with exertion
- You experience unreasonable breathlessness
- You experience dizziness, fainting, or blackouts
- You experience ankle swelling
- You experience unpleasant awareness of a forceful or rapid heart rate
- You take heart medications

Other Health Issues

- You have diabetes
- You have asthma or other lung disease
- You have burning or cramping sensation in your lower legs when walking short distances
- You have musculoskeletal problems that limit your physical activity
- You have concerns about the safety of exercise
- You take prescription medication(s)
- You are pregnant

If you marked two or more of the statements in this section, you should consult your physician or other appropriate health care as part of good medical care and progress gradually with your exercise program. You might benefit from using a facility with a **professionally qualified exercise staff** to guide your exercise program.

Cardiovascular risk factors

- You are a man ≥ 45 yr
- You are a woman ≥ 55 yr
- You smoke, or quit smoking within the previous 6 months
- Your blood pressure is $> 140/90$ mmHg
- You do not know your blood pressure
- You take blood pressure medication
- Your blood cholesterol level is > 200 mg/dl
- You do not know your cholesterol level
- You have a close blood relative who had a heart attack or heart surgery before age 55 (father or brother) or age 65 (mother or sister)
- You are physically inactive (i.e. you get < 30 min of physical activity on at least 3 d per week)
- You have a body mass index ≥ 30 kg/m²
- You have prediabetes
- You do not know if you have prediabetes
- None of the above

You should be able to exercise safely without consulting your physician or other appropriate health care provider in a self-guide program or almost any facility that meets your exercise program needs.

Rating of Perceived Gastrointestinal Distress

Stomach Problems

1	2	3	4	5	6	7	8	9	10
None		Slight		Moderate		Significant		Very much	Severe

GI Cramping

1	2	3	4	5	6	7	8	9	10
None		Slight		Moderate		Significant		Very much	Severe

Bloated Feeling

1	2	3	4	5	6	7	8	9	10
None		Slight		Moderate		Significant		Very much	Severe

Diarrhea

1	2	3	4	5	6	7	8	9	10
None		Slight		Moderate		Significant		Very much	Severe

Nausea

1	2	3	4	5	6	7	8	9	10
None		Slight		Moderate		Significant		Very much	Severe

Dizziness

1	2	3	4	5	6	7	8	9	10
None		Slight		Moderate		Significant		Very much	Severe

Headache

1	2	3	4	5	6	7	8	9	10
None		Slight		Moderate		Significant		Very much	Severe

Belching

1	2	3	4	5	6	7	8	9	10
None		Slight		Moderate		Significant		Very much	Severe

Vomiting

1	2	3	4	5	6	7	8	9	10
None		Slight		Moderate		Significant		Very much	Severe

Urge to Urinate or Defecate

1	2	3	4	5	6	7	8	9	10
None		Slight		Moderate		Significant		Very much	Severe

Attachment 5
24-HOUR DIET RECORD

Subject number _____

Date _____

Day of Week _____

Time	Food and/or Drink	Method of Preparation	Quantity Consumed	Brand Name

Adapted From: Lee RD, Nieman DC. *Nutritional Assessment*. 2nd ed. United States of America: Mosby; 1996

INSTRUCTIONS FOR KEEPING YOUR 24-HOUR FOOD RECORD

Keep your record for three days per trial. You will include the day before, the day of, and the day after each trial. Include all meals, snacks, nibbling, and beverages including water and cocktails

1. Fill out the date and day of the week at the top of food record sheet
2. Record the time you consumed your food and/or drink. To be most accurate, fill out the food record as soon as you finish eating.
3. List the first food and/or drink you consumed when you began your day and continue to record until you consume your last food and/or drink of your day (usually before bedtime)

4. List each food and/or drink on a separate line

Example: cereal with milk, cereal and milk should each be on separate lines
spaghetti, noodles, and sauce should each be on separate lines

Combination foods:

List parts of food on separate lines

Include preparation method, quantity, and brand name of each food

Example: Sandwich (4 oz healthy choice turkey, 2 slices Sara Lee wheat bread, 1 tbsp Hellman's light mayo, 2 oz Kraft American cheese, 1 slice of red fresh tomato)

5. Record the method of preparation

Example: fried, baked, grilled
salt, oil (olive, canola, corn, other) butter or margarine, spices, etc.

6. Record quantity consumed

Do not record any food not eaten

Example: made two cups of vegetables but ate half so you would record one cup

Quantity of food and/or drink

Example: cups, ounces, liters, grams, each, or other unit of measure

Example: 1 cup of vegetables, 4 ounces of meat, one medium apple

7. Record brand name

Example: fast food chain name and/or package name

Example: Wendy's, Betty Crocker, Lean Cuisine, Gatorade, Thomas Bagel

8. Place any helpful food labels in manila envelope that is attached to folder

USE THE FOLLOWING TO HELP DETERMINE PORTION SIZES AND TYPES OF FOODS

PLEASE SPECIFY	
Beverages	Sugar or creamer? Regular or sugar-free? Alcohol content? Name of drink and ingredients (if mixed drink)
Breads	Butter or margarine added?
Cereal/Milk	Milk, sugar, or fruit added? The type of milk? (skim, 1%, 2%, whole) Cereal: dry or cooked measure?
Dairy	Is yogurt fruited or plain? % fat of milk or yogurt? Indicate brand name of cheese substitute and/or nondairy creamer.
Desserts	Whipped topping added? Frosting? Fat modified (i.e., reduced)? Sugar-free?
Eggs	Preparation method (scrambled, hard-boiled, etc)? Fat used in cooking?
Fast Food	What restaurant? If not a national fast food chain, describe food in detail Size order of fries? Super-size? Extra toppings on sandwich?
Fats/Oils	Regular or salt-free? Stick, tub, or liquid margarine? Reduced calorie or diet product?
Fish	Water or oil packed (fresh or canned)? Baked or fried (With batter or without)?

	Type of fat added? Raw or cooked weight?
Fruit	Sweetened or unsweetened? Fresh, canned, or frozen? With or without skin?
Meats	Visible fat removed? Light or dark meat? Raw or cooked?
Sugars and Sweets	Regular or reduced-calorie? Don't forget hard candy as well as chocolate.
Vegetables	Raw or cooked? Fresh, frozen, or canned? Low-sodium or regular? Added fat or sauce?

Helpful Hints with Portion Sizes

- 1 teaspoon (5 ml)
 - about the size of the top half / tip of your thumb
- 1oz (28g)
 - approximately inch cube of cheese
 - volume of four stacked dice
 - slice of cheese is about the size of a 3 1/2 inch computer disk
 - chunk of cheese is about as thick as 2 dominoes
 - 1 handful (palm) of nuts
- 2 ounces (57 g)
 - 1 small chicken leg or thigh
 - 1/2 cup of cottage cheese or tuna
- 3 ounces (85 g)
 - serving of meat is about the size of a deck of playing cards (3 exchanges)
 - the size of the palm of your hand
 - 1/2 of whole chicken breast
 - 1 medium pork chop
 - 1 small hamburger
 - unbreaded fish fillet
- 1/2 cup (118 ml)
 - fruit or vegetables can fit in the palm of your hand
 - about the volume of a tennis ball
- 1 cup (236 ml)
 - about the size of a woman's fist
 - breakfast cereal goes halfway up the side of a standard cereal bowl
 - broccoli is about the size of a light bulb
- 1 medium apple = A tennis ball

References

1. Ali A, Yoo MJ, Moss C, Breier BH. Carbohydrate mouth rinsing has no effect on power output during cycling in a glycogen-reduced state. *J Int Soc Sports Nutr.* 2016; 13:19.
2. Anantaraman R, Carmines AA, Gaesser GA, Weltman A. Effects of carbohydrate supplementation on performance during 1 hour of high-intensity exercise. *Int J Sports Med.* 1995; 16(7):461-5.
3. Bastos-Silva VJ, Melo Ade A, Lima-Silva AE, Moura FA, Bertuzzi R, de Araujo GG. Carbohydrate mouth rinse maintains muscle electromyographic activity and increases time to exhaustion during moderate but not high-intensity cycling exercise. *Nutrients.* 2016; 8(3):49.
4. Batterham AM, Hopkins WG. Making meaningful inferences about magnitudes. *Int J Sports Physiol Perform.* 2006; 1(1):50-7.
5. Beelen M, Berghuis J, Bonaparte B, Ballak SB, Jeukendrup AE, van Loon LJ. Carbohydrate mouth rinsing in the fed state: lack of enhancement of time-trial performance. *Int J Sport Nutr Exerc Metab.* 2009; 19(4):400-9.
6. Burke LM, Claassen A, Hawley JA, Noakes TD. Carbohydrate intake during prolonged cycling minimizes effect of glycemic index of preexercise meal. *J Appl Physiol.* 1998; 85(6):2220-6.
7. Carter JM, Jeukendrup AE, Jones DA. The effect of carbohydrate mouth rinse on 1-h cycle time trial performance. *Med Sci Sports Exerc.* 2004; 36(12):2107-11.
8. Carter JM, Jeukendrup AE, Mann CH, Jones DA. The effect of glucose infusion on glucose kinetics during a 1-h time trial. *Med Sci Sports Exerc.* 2004; 36(9):1543-50.
9. Chambers ES, Bridge MW, Jones DA. Carbohydrate sensing in the human mouth: effects on exercise performance and brain activity. *J Physiol.* 2009; 587(Pt 8):1779-94.
10. Chen YJ, Wong SHS, Chan COW, Wong CK, Lam CW, Siu PMF. Original paper: Effects of glycemic index meal and CHO-electrolyte drink on cytokine response and run performance in endurance athletes. *J Sci Med Sport.* 2009; 12:697-703.
11. Chryssanthopoulos C, Williams C, Nowitz A, Kotsiopolou C, Vleck V. The Effect of a High Carbohydrate Meal on Endurance Running Capacity. *Int J Sport Nutr Exerc Metab.* 2002; 12(2):157.

12. Chryssanthopoulos C, Williams C. Pre-exercise carbohydrate meal and endurance running capacity when carbohydrates are ingested during exercise. *Int J Sports Med.* 1997; 18(7):543-8.
13. Coyle EF, Coggan AR, Hemmert MK, Ivy JL. Muscle glycogen utilization during prolonged strenuous exercise when fed carbohydrate. *J Appl Physiol.* 1986; 61(1):165.
14. Coyle EF, Hagberg JM, Hurley BF, Martin WH, Ehsani AA, Holloszy JO. Carbohydrate feeding during prolonged strenuous exercise can delay fatigue. *J Appl Physiol Respir Environ Exerc Physiol.* 1983; 55(1):230.
15. DeMarco HM, Sucher KP, Cisar CJ, Butterfield GE. Pre-exercise carbohydrate meals: application of glycemic index. *Med Sci Sports Exerc.* 1999; 31(1):164-70.
16. Devenney S, Collins K, Shortall M. Effects of various concentrations of carbohydrate mouth rinse on cycling performance in a fed state. *Eur J Sport Sci.* 2016:1-6.
17. Earnest CP, Lancaster SL, Rasmussen CJ, et al. Low Vs. High Glycemic Index Carbohydrate Gel Ingestion during Simulated 64-km Cycling Time Trial Performance. *J Strength Cond Res.* 2004; 18(3):466-72.
18. El-Sayed M, Balmer J, Rattu AJM. Carbohydrate ingestion improves endurance performance during a 1 h simulated cycling time trial. *J Sports Sci.* 1997; 15(2):223-30.
19. Fares EJ, Kayser B. Carbohydrate mouth rinse effects on exercise capacity in pre- and postprandial States. *J Nutr Metab.* 2011; 2011:385962.
20. Febbraio MA, Stewart KL. CHO feeding before prolonged exercise: effect of glycemic index on muscle glycogenolysis and exercise performance. *J Appl Physiol.* 1996; 81(3):1115-20.
21. Febbraio MA. Effects of carbohydrate ingestion before and during exercise on glucose kinetics and performance. *J Appl Physiol.* 2000; 89(6):2220.
22. Fraga C, Velasques B, Koch AJ, et al. Carbohydrate mouth rinse enhances time to exhaustion during treadmill exercise. *Clin Physiol Funct Imaging.* 2015.
23. Frank GK, Oberndorfer TA, Simmons AN, et al. Sucrose activates human taste pathways differently from artificial sweetener. *Neuroimage.* 2008; 39(4):1559-69.
24. Gam S, Guelfi KJ, Fournier PA. Opposition of carbohydrate in a mouth-rinse solution to the detrimental effect of mouth rinsing during cycling time trials. *Int J Sport Nutr Exerc Metab.* 2013; 23(1):48-56.

25. Gant N, Stinear CM, Byblow WD. Carbohydrate in the mouth immediately facilitates motor output. *Brain Res.* 2010; 1350:151-8.
26. Haase L, Cerf-Ducastel B, Murphy C. Cortical activation in response to pure taste stimuli during the physiological states of hunger and satiety. *Neuroimage.* 2009; 44(3):1008-21.
27. Hopkins WG. How to interpret changes in an athletic performance test. *Sportscience.* 2004; 8:1-7.
28. Ispoglou T, O'Kelly D, Angelopoulou A, Bargh M, O'Hara JP, Duckworth LC. Mouth Rinsing With Carbohydrate Solutions at the Postprandial State Fail to Improve Performance During Simulated Cycling Time Trials. *J Strength Cond Res.* 2015; 29(8):2316-25.
29. Ivy JL, Milley W, Dover V, et al. Endurance improved by ingestion of a glucose polymer supplement. *Med Sci Sports Exere.* 1983; 15(6):466-71.
30. Jeffers R, Shave R, Ross E, Stevenson EJ, Goodall S. The effect of a carbohydrate mouth-rinse on neuromuscular fatigue following cycling exercise. *Appl Physiol Nutr Metab.* 2015; 40(6):557-64.
31. Jenkins DA, Wolever TMS, Taylo RH, et al. Glycemic index of foods: a physiological basis for carbohydrate exchange. *Am J Clin Nutr* 1981; 34:362-366.
32. Jentjens R, Jeukendrup A. Effects of pre-exercise ingestion of trehalose, galactose and glucose on subsequent metabolism and cycling performance. *Eur J Appl Physiol.* 2003; 88(5):459-65.
33. Jeukendrup A, Brouns F, Wagenmakers AJM, Saris WHM. Carbohydrate-electrolyte feedings improve 1 h time trial cycling performance. *Int J Sports Med.* 1997; 18(2):125-9.
34. Jeukendrup AE, Wagenmakers AJM, Stegen JH, et al. Carbohydrate ingestion can completely suppress endogenous glucose production during exercise. *Am J Physiol (Endocrinol Metab 39)* 1999; E672-E683.
35. Jeukendrup AE. Carbohydrate feeding during exercise. *Eur J Sport Sci.* 2008; 8(2):77-86.
36. Kern M, Heslin CJ, Rezende RS. Metabolic and Performance Effects of Raisins Versus Sports Gel as Pre-Exercise Feedings in Cyclists. *J Strength Con Res.* 2007; 21(4):1204-7.
37. Kirwan JP, Cyr-Campbell D, Campbell WW, Scheiber J, Evans WJ. Effects of moderate and high glycemic index meals on metabolism and exercise performance. *Metabolism.* 2001; 50(7):849-55.

38. Kirwan JP, O'Gorman D, Evans WJ. A moderate glycemic meal before endurance exercise can enhance performance. *J Appl Physiol*. 1998; 84(1):53-9.
39. Kulaksiz TN, Kosar SN, Bulut S, et al. Mouth Rinsing with Maltodextrin Solutions Fails to Improve Time Trial Endurance Cycling Performance in Recreational Athletes. *Nutrients*. 2016; 8(5):10.3390/nu8050269.
40. Lane SC, Bird SR, Burke LM, Hawley JA. Effect of a carbohydrate mouth rinse on simulated cycling time-trial performance commenced in a fed or fasted state. *Appl Physiol Nutr Metab*. 2013; 38(2):134-9.
41. Luden ND, Saunders MJ, D'Lugos AC, et al. Carbohydrate mouth rinsing enhances high intensity time trial performance following prolonged cycling. *Nutrients*. 2016; 8(9):576.
42. McConell GK, Canny BJ, Daddo MC, Nance MJ, Snow RJ. Effect of carbohydrate ingestion on glucose kinetics and muscle metabolism during intense endurance exercise. *J Appl Physiol (1985)*. 2000; 89(5):1690-8.
43. Moore LJS, Midgley AW, Thurlow S, Thomas G, Mc Naughton LR. Original paper: Effect of the glycaemic index of a pre-exercise meal on metabolism and cycling time trial performance. *J Sci Med Sport*. 2010; 13:182-8.
44. Murray R, Paul GL, Seifert JG, Eddy DE. Responses to varying rates of carbohydrate ingestion during exercise. *Med Sci Sports Exerc*. 1991; 23(6):713-8.
45. Page KA, Chan O, Arora J, et al. Effects of fructose vs glucose on regional cerebral blood flow in brain regions involved with appetite and reward pathways. *JAMA*. 2013; 309(1):63-70.
46. Pottier A, Bouckaert J, Gilis W, Roels T, Derave W. Mouth rinse but not ingestion of a carbohydrate solution improves 1-h cycle time trial performance. *Scand J Med Sci Sports*. 2010; 20(1):105-11.
47. Rollo I, Cole M, Miller R, Williams C. Influence of mouth rinsing a carbohydrate solution on 1-h running performance. *Med Sci Sports Exerc*. 2010; 42(4):798-804.
48. Rollo I, Williams C, Gant N, Nute M. The influence of carbohydrate mouth rinse on self-selected speeds during a 30-min treadmill run. *Int J Sport Nutr Exerc Metab*. 2008; 18(6):585-600.
49. Rollo I, Williams C, Nevill M. Influence of ingesting versus mouth rinsing a carbohydrate solution during a 1-h run. *Med Sci Sports Exerc*. 2011; 43(3):468-75.
50. Schabort EJ, Bosch AN, Weltan SM, Noakes TD. The effect of a preexercise meal on time to fatigue during prolonged cycling exercise. *Med Sci Sports Exerc*. 1999; 31(3):464,471 8p.

51. Sherman WM, Peden MC, Wright DA. Carbohydrate feedings 1 h before exercise improves cycling performance. *Am J Clin Nutr.* 1991; 54(5):866-70.
52. Sinclair J, Bottoms L, Flynn C, et al. The effect of different durations of carbohydrate mouth rinse on cycling performance. *Eur J Sport Sci.* 2014; 14(3):259-64.
53. Smeets PA, de Graaf C, Stafleu A, van Osch MJ, van der Grond J. Functional MRI of human hypothalamic responses following glucose ingestion. *Neuroimage.* 2005; 24(2):363-8.
54. Smith JW, Zachwieja JJ, Péronnet F, et al. Fuel selection and cycling endurance performance with ingestion of [¹³C]glucose: evidence for a carbohydrate dose response. *J Appl Physiol (1985).* 2010; 108(6):1520-9.
55. Sparks MJ, Selig SS, Febbraio MA. Pre-exercise carbohydrate ingestion: effect of the glycemic index on endurance exercise performance. *Med Sci Sports Exerc.* 1998; 30(6):844-849.
56. Stannard SR, Thompson MW, Brand Miller JC. The effect of glycemic index on plasma glucose and lactate levels during incremental exercise. *Int J Sport Nutr Exerc Metab.* 2000; 10:51-61.
57. Stevenson EJ, Williams C, Mash LE, Phillips B, Nute ML. Influence of high-carbohydrate mixed meals with different glycemic indexes on substrate utilization during subsequent exercise in women. *Am J Clin Nutr.* 2006; 84(2):354,360 7p.
58. Thomas DE, Brotherhood JR, Brand JC. Carbohydrate feeding before exercise: effect of glycemic index. *Int J Sports Med.* 1991; 12(2):180-6.
59. Thomas DE, Brotherhood JR, Miller JB. Plasma glucose levels after prolonged strenuous exercise correlate inversely with glycemic response to food consumed before exercise. *Int J Sport Nutr.* 1994; 4(4):361,373 13p.
60. Tokmakidis SP, Karamanolis IA. Effects of carbohydrate ingestion 15 min before exercise on endurance running capacity. *Appl Physiol Nutr Metab.* 2008; 33(3):441-9.
61. Trommelen J, Beelen M, Mullers M, Gibala MJ, van Loon LJ, Cermak NM. A Sucrose Mouth Rinse Does Not Improve 1-hr Cycle Time Trial Performance When Performed in the Fasted or Fed State. *Int J Sport Nutr Exerc Metab.* 2015; 25(6):576-83.
62. Wee S, Williams C, Gray S, Horabin J. Influence of high and low glycemic index meals on endurance running capacity. *Med Sci Sports Exerc.* 1999; 31(3):393,399 7p.

63. Whitham M, McKinney J. Effect of a carbohydrate mouthwash on running time-trial performance. *J Sports Sci.* 2007; 25(12):1385-92.
64. Wong SHS, Oi WC, Ya JC, Heng LH, Ching WL, Pak KC. Effect of Preexercise Glycemic-Index Meal on Running When CHO-Electrolyte Solution Is Consumed During Exercise. *Int J Sport Nutr Exerc Metab.* 2009; 19(3):222-42.
65. Wong SHS, Siu PM, Lok A, Chen YJ, Morris J, Lam CW. Effect of the glycaemic index of pre-exercise carbohydrate meals on running performance. *Eur J Sport Sci.* 2008; 8(1):23-33.
66. Wu C, Nicholas C, Williams C, Took A, Hardy L. The influence of high-carbohydrate meals with different glycaemic indices on substrate utilisation during subsequent exercise. *Br J Nutr.* 2003; 90(6):1049,1056 8p.
67. Wu CL, Williams C. A low glycemic index meal before exercise improves endurance running capacity in men. *Int J Sport Nutr Exerc Metab* 2006; 16:510-527.