Investigating The Effect Of Carbohydrate Doses On Exogenous Carbohydrate Oxidation Rates

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Abstract

Carbohydrate supplementation during endurance exercise has been extensively studied, but despite the presence of fueling guidelines, there is insufficient evidence to personalise strategies based on individual factors such as oxidation rates, body size or sex. The potential to personalise carbohydrate intakes based on exogenous carbohydrate oxidation rates has been suggested, but remains unexplored. This thesis aims to evaluate the feasibility of personalising carbohydrate intakes to optimise exogenous oxidation rates during endurance exercise. The primary objective was to assess whether a high, fixed dose of glucose (90 g/h) during exercise could inform individualised carbohydrate strategies by maximising exogenous oxidation. Eleven trained endurance athletes performed three 150-minute cycling trials at 95% of LT1, ingesting either 90g/h or a personalised carbohydrate dose, derived from the first experimental trial; exogenous oxidation rates for each participant were divided by an oxidation efficiency of 80% to identify the rate to provide carbohydrates in the personalised trial. Results indicated that exogenous carbohydrate oxidation rates were comparable between the high-dose and personalised carbohydrate trials. Mean peak exogenous oxidation rates in the personalised trial did not significantly differ from the 90g trial; 0.91 ±0.19 *vs* 0.91 ±0.15 g/min, respectively; ($p = 0.847$). Heart rate, ratings of perceived exertion, and gastrointestinal fullness were significantly lower in the personalised than the 90g trial, (142 ±12 *vs* 148 ±13 bpm; 13 ±1 *vs* 12 ±2; 2.5 ±1.1 *vs* 1.7 ±1.2 respectively). There were no significant relationships between peak exogenous carbohydrate oxidation and body mass, height, or workload ($p \geq 0.05$) in either the 90g or personalised trial. Plasma glucose and lactate were not significantly different ($p = 0.812$; $p = 0.360$ respectively) between 90g and personalised trials. The low coefficient of variation (19% ±0.3) and limits of agreements between peak and average exogenous CHO oxidation rates across all timepoints in the 90g and personalised trials indicate a consistent response among participants, strengthening the reliability of these findings. Furthermore, total carbohydrate and fat oxidation rates were unaffected by any trial. The second objective examined the reliability of these exogenous carbohydrate oxidation rates across repeated trials; by repeating the initial 90g trial; providing 1.5 g/min of carbohydrates. Mean peak exogenous carbohydrate oxidation rates were not significantly different between the 90g and 90gR trials (0.89 ±0.19 *vs* 0.96 ±0.21 g/min, respectively; $p = 0.25$). Ratings of perceived exertion, measures of gastrointestinal comfort and heart rate were all not significantly different between the two repeat 90g trials ($p = 0.393$; $p > 0.05$; $p = 0.900$, respectively). In conclusion, it is possible to personalise carbohydrate ingestion rates to optimise exogenous oxidation and improve gut comfort during endurance exercise, supporting the potential for individualised dosing strategies. Additionally, whilst tighter controls are required, the determination of exogenous carbohydrate oxidation is somewhat reliable and research should extend to investigate the effect of multiple carbohydrates to determine whether the observed trends are evident, and therefore more applicable with current CHO recommendations during exercise.

Abbreviations

BM	Body Mass
BMI	Body Mass Index
CHO	Carbohydrate
CHO-OX	Carbohydrate Oxidation
EE	Energy Expenditure
EI	Energy Intake
FAT-OX	Fat Oxidation
FFM	Fat-Free Mass
FM	Fat Mass
GI	Gastrointestinal
GLUT4	Glucose Transporter 4
HR	Heart Rate
LT1	First Lactate Threshold
LT2	Second Lactate Threshold
RER	Respiratory Exchange Ratio
RMR	Resting Metabolic Rate
RPE	Rate of Perceived Exertion
VO_2max	Maximal Aerobic Capacity
VO_2peak	Highest value of VO_2max
WMAX	Maximal power

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CHAPTER 1

Introduction

1.1. Thesis Outline

This thesis focuses on the potential to develop a personalised carbohydrate dose based on individual exogenous carbohydrate (CHO) oxidation rates during exercise. It also identifies the reliability of measuring exogenous CHO oxidation rates during exercise. The thesis begins with an introduction to the topic area followed by a discussion of the current literature and the requirement for future investigation. The methods describe the approach employed in the study and where appropriate, additional justification for them. Following this, in the results, the experimental data are presented and analysed to investigate the aims of the study. The latter part of this thesis contains a discussion to explore and interpret the results, followed by a conclusion and future directions.

1.2. Exercise, fuel utilisation and fatigue

Exercise refers to physical activities requiring effort that are carried out to sustain and improve health and fitness. Exercise is commonly classified into two broad categories: aerobic (endurance) and resistance exercise (Cartee *et al.,* 2016). Endurance exercise is defined as exercise performed for an extended period at intensities >50% maximal aerobic capacity (VO_2max), (Joyner & Coyle, 2008), and usually involves sustained work efforts lasting longer than 1 minute (Chamari & Padulo, 2015).

To sustain endurance exercise, two major substrates must be utilised for energy: CHO and fat. The contribution of these substrates to overall energy production changes in an equal-and-opposite manner as a function of intensity and duration; higher intensity endurance exercise will require greater CHO utilisation and reduced fat utilisation and vice versa. Even at moderate intensity, and particularly during strenuous endurance exercise, CHO predominates as the fuel source. For example, it has been observed that at higher intensities (70-80% of maximal aerobic power), exhaustion during cycling exercise has coincided with muscle glycogen depletion of the quadriceps femoris. However, at 60-70% of maximal aerobic power, the onset of fatigue and glycogen depletion occurs later (Hermansen et al, 1967).

Humans have limited ability to store CHO. Fatigue during exercise is commonly associated with reduced muscle glycogen content or low blood glucose; the latter being reflective of low liver glycogen content. The combination of these factors has the ability to impair performance. The time to which muscle fatigue occurs may be influenced by glycogen stores

preceding exercise and the rate of CHO ingestion *during* exercise (Enoka *et al.,* 1992). At rest, muscle glycogen levels are usually around 400-600 mmol/kg, depending on training status (Vigh-Larsen *et al.,* 2021). High intensity exercise lasting 60-80 minutes can reduce muscle glycogen substantially (Bosch *et al.,* 1994), causing muscle fatigue and forced reduction in intensity, or the cessation of exercise. The quantity of CHO ingested *following* a bout of exercise will determine the restoration rate of muscle glycogen.

As a result, many nutritional strategies attempt to elevate or maintain body CHO availability to help prevent performance decline, by maximising glycogen stores and providing a consistent supply of glucose. It has been illustrated that when participants are fed glucose during exercise, they can extend exercise duration (Coyle, 1992). The mechanisms behind fatigue underpin the development of several guidelines and approaches to use CHO as a strategy for optimising exercise performance and recovery (Jeukendrup, 2007).

1.3. Carbohydrate feeding during exercise

CHO ingestion during exercise bouts >45 minutes appears to enhance performance by maintaining plasma glucose concentration, sustaining total CHO oxidation, preserving liver glycogen and, in some cases, sparing muscle glycogen, delaying fatigue. These clear performance and metabolic benefits have prompted the development of specific guidelines tailored to exercise duration (Jeukendrup, 2014).

As the primary fuel for endurance exercise, CHO forms the basis of guidelines for feeding to sustain performance during these exercise bouts. Current guidelines recommend individuals to consume CHO in doses of 30-90 grams per hour depending on the intensity and duration of exercise (Burke *et al.,* 2013). Carbohydrates that are commonly ingested during exercise include glucose or glucose polymers such as maltodextrin, which are often combined with fructose or sucrose. Some research suggests a positive relationship between CHO intake and performance (Pfeiffer *et al.,* 2012) and as a result, when ingesting single transportable CHOs (e.g. glucose), athletes should aim for doses of 30-60 g/h, with some athletes benefiting from doses of up to 90 g/h (Burke *et al.,* 2013). However, numerous studies have demonstrated that using multiple transportable CHO sources during cycling can enhance exogenous oxidation rates, thereby supporting training and racing performance (Wallis *et al.,* 2008). The consumption of high rates of just glucose has several limitations, the most important of which is the saturation of the transport protein, SGLT1, which aids in the absorption of glucose from the intestine. Further ingestion of glucose above 60-70 g/h will not yield greater intestinal absorption and increased oxidation rates (Jentjens *et al.,* 2004), but instead risk gastrointestinal distress as CHO remains in the gut.

The idea, therefore, of consuming multiple transportable CHOs which utilise different absorption pathways to glucose (e.g. GLUT5 for fructose), may increase CHO oxidation rates beyond the capacity of glucose alone. In fact, increases in oxidation rates have been seen as high as 50% with fructose coingestion (Jentjens *et al.,* 2004). Galactose ingestion has previously been observed to have a slower oxidation rate compared to combined glucose and fructose ingestion (Stannard, Hawke, & Schnell, 2009). Furthermore, the co-ingestion of glucose and galactose does not increase the oxidation of ingested galactose during exercise (Odell *et al.*, 2022). Finally, it has been observed that the ingestion of drinks containing glucose and fructose (in a 2:1 ratio at a rate of 1.8 g/min) results in nearly a 10% improvement in performance, via higher cycling power output, compared with glucose ingestion at the same rate (Currell & Jeukendrup, 2008).

1.4. Carbohydrate and fuel metabolism

Exogenous CHO are absorbed in the small intestine, transported to the liver via the portal vein, and subsequently distributed from the liver through several hepatic veins. Glycogen, a storage form of CHO (≤3000 kcal), is depleted during exercise (Koivisto *et al.,* 1985). Endogenous CHO refers to glycogen stored within skeletal muscle and the liver. The metabolism of muscle glycogen accelerates with increasing exercise intensity, with CHO becoming the primary substrate at intensities above ~65% VO2max due to the higher ATP generation efficiency (Hearris *et al*., 2018).

Muscle *glucose* uptake, the transport of glucose from the bloodstream into muscle cells, is facilitated by both insulin (insulin-mediated) and exercise (exercise-induced), (Mul *et al.,* 2015). To support the elevated energy demands during exercise, the rate of muscle glucose uptake increases significantly (Jensen *et al.,* 2011). Glucose transporters, known as GLUT isoforms, facilitate the absorption of monosaccharides, including GLUT4 for glucose and GLUT5 for fructose (Fuchs *et al.,* 2019). GLUT4 is the primary transporter responsible for muscle glucose uptake during exercise. Once taken in from the bloodstream, glucose can be directly oxidised by muscle cells to support energy production during exercise.

As previously noted, CHO provision during exercise enhances endurance capacity and performance (Stellingwerff & Cox, 2014). Consuming CHO before, during and after exercise effectively supports both muscle and central nervous systems. Karelis *et al.* (2010) provided a comprehensive review explaining the potential mechanisms through which CHO supports performance during prolonged exercise. CHO intake during exercise helps maintain plasma glucose levels and exogenous CHO oxidation by directly using CHO as a fuel. Additional

benefits of CHO consumption include reduced central fatigue, potential sparing of muscle and liver glycogen, changes in muscle metabolite levels, reduced exercise-induced strain, and improved maintenance of excitation-contraction coupling. These factors are associated with fatigue, and CHO ingestion during exercise can mitigate them, thereby avoiding fatigue and maintaining endurance performance.

1.5. Investigating exogenous carbohydrate oxidation rates

Aside from mouth-rinsing, the primary advantage of CHO feeding during exercise is the provision of an exogenous fuel source. Consequently, extensive research has been conducted to investigate exogenous CHO oxidation under a variety of conditions and has helped to inform and develop nutritional guidelines.

The oxidation of ingested CHO is usually assessed using stable carbon isotopes, known as tracers. Within research studies, participants are provided with drinks containing the CHO fuel source to be investigated and a small amount of the stable isotope tracer, for example, U-$^{13}C_6$-glucose. CHO is broken down into CO_2 and H_2O by complete oxidation. By measuring the enrichment of ^{13}C in the breath, it is possible to determine the oxidation of ingested CHO during exercise, using several equations as outlined by Pirnay *et al.* (1997) and outlined further in the methods section of this thesis.

1.6. Oxidation of ingested carbohydrates during exercise

The ingestion of CHO during exercise to enhance performance, as discussed by Thomas *et al.* (2016) and the subsequent development of guidelines, are in part based on the rationale of maximising exogenous CHO oxidation. On average, the maximal oxidation rate of ingested CHO is up to approximately 60 grams per hour when glucose-based CHO is consumed alone, further increasing with glucose-fructose mixtures. A review demonstrated no difference in exogenous CHO oxidation rates when ingesting glucose at 1.2 g/min *vs* 1.8 g/min. However, a difference was apparent in *endogenous* CHO oxidation, with lower rates observed in the more moderate CHO condition. This example highlights the theory that overfeeding is not beneficial, and personalised strategies to optimise performance may be advantageous (Jeukendrup, 2010).

1.7. Factors affecting exogenous carbohydrate oxidation rates during exercise.

1.7.1. Exercise intensity and duration

At rest, the body derives energy from the oxidation of CHO and fat. However, during exercise, skeletal muscle contraction increases the demand for energy and results in a heightened fuel consumption by muscles. During low-intensity exercise, such as light

walking, the energy required by working muscles is predominantly provided by the oxidation of free fatty acids available in blood plasma (Mul *et al.,* 2015). As exercise intensity increases, there is a shift towards a greater reliance on CHO as a fuel source, making CHO crucial for supporting active muscle contractions during exercise (Burke *et al.,* 2013).

Despite this shift, the CHO oxidation rates of ingested CHO remain constant at exercise intensities above 50-60%, and during high-intensity exercise, the oxidation of plasma fatty acids decreases, with CHO oxidation accounting for over ~70% of the energy required (Pirnay *et al.,* 1982). Specifically, within this paper it was observed that at lower exercise intensities, between 22-51% $\dot{V}O_2$max, exogenous glucose oxidation was linearly correlated with the relative workload, but when this increased to 51-62% $\dot{V}O_2$max, exogenous glucose oxidation levelled off. This is likely a result of the decrease in oxidation within the muscles and/ or a lesser availability of exogenous glucose for use.

Additionally, fuel usage varies with exercise duration, with a higher demand for CHO as the duration increases. It is evident that, irrespective of the duration, CHO consumption during exercise can prevent hypoglycaemia, maintain CHO oxidation rates, and enhance endurance capacity (Jeukendrup, 2014). Jeukendrup *et al.* (2006) observed an increase in exogenous CHO oxidation rates with increasing duration of exercise to a certain extent. When participants were fed 1.5 g/min of glucose, exogenous oxidation rates increased linearly for ~120 minutes, reaching a peak of 1.24 g/min. Beyond this, no further increase was observed and exogenous oxidation rates levelled off. It is suggested that this could be a result of increased hepatic glucose output during the later stages of exercise.

1.7.2. CHO dose and feeding schedule

Pre-exercise CHO recommendations suggest consuming approximately 1-4 grams of CHO per kg of body mass in the 1-4 hours before exercise (i.e. 1g/kg, 1 hour before, and 4g/kg, 4 hours before; Burke *et al.,* 2013). During exercise, the optimal CHO dose should attempt to match the maximal rate of exogenous CHO oxidation, without causing GI discomfort. Inadequate CHO consumption can limit endurance capacity and performance (Jeukendrup, 2007). For exercise sessions lasting <45 mins, specific CHO ingestion is generally unnecessary, as glycogen stores and daily CHO intake typically meet the body's needs. However, it has been observed that CHO mouth rinsing, without actual ingestion, can positively affect performance (Carter, Jeukendrup & Jones, 2004). The proposed mechanism suggests that CHO is detected by receptors in the oral cavity, which send afferent neural signals to the brain, resulting in improved performance (Jeukendrup, Rollo & Carter, 2013).

Current recommendations suggest that athletes should consume CHO during exercise bouts longer than one hour, with quantities from 60 g/h up to 90 g/h (Burke *et al.,* 2013). When consuming doses of CHO beyond 60 g/h, the aim should be to use multiple transportable sources (for example, glucose-fructose). For exercise sessions ranging from 1-2 hours, 30-60 g/h of CHO is considered sufficient, while individuals completing endurance exercise sessions lasting 2.5-3 hours or longer, may benefit from higher CHO intakes. Although limited literature exists to suggest variability in athletes CHO metabolism and personal oxidation rates, further research and personalised nutritional intake are necessary. Smith *et al.* (2010) assessed glucose ingestion across various doses and found that consuming 15-60 g/h of glucose during prolonged exercise enhances endurance performance, with a clear dose-performance relationship observed for exercise lasting 150 minutes. Individuals responded positively to CHO ingestion rates of both 39 and 64 g/h, supporting the ergogenic effect of CHO for endurance performance (Newell *et al.,* 2015). Recent investigations into CHO feeding and the co-ingestion of multiple CHO sources have resulted in updated guidelines recommending CHO doses beyond 90 g/h. These CHO oxidation-focused studies demonstrate a significant performance effect, both reinforcing and guiding fueling strategies for athletes.

Wagenmaker *et al.* (1993), assessed the effect of changing CHO dose on exogenous oxidation rates during cycling exercise in men. It was confirmed that despite changes in CHO doses with solutions increasing from 4-16%, in both maltodextrin and sucrose, increases in exogenous CHO oxidation occurred but a plateau was observed after 90-120 minutes of exercise. Wallis *et al.* (2007), further confirmed this and that the CHO dose provided is influential up to 60-70 g/h where, with glucose alone, further feeding shows no further increase in exogenous CHO oxidation rates.

With regard to feeding schedule, Mears *et al.* (2010), investigated the pattern of CHO ingestion during running and identified that ingesting a larger volume of CHO at less frequent intervals (every 20 minutes) increased exogenous CHO oxidation rates, compared with smaller, more frequent ingestions (every 5 minutes) but with the same total volume of fluid. Within this, exogenous CHO oxidation rates were on average, 23% higher with the less frequent ingestion of CHO.

1.7.3. Exercise mode

A 2011 study by Pfeiffer and colleagues investigated the potential differences in CHO oxidation rates between cycling and running. Participants engaged in either running or cycling at approximately 60% VO2max whilst ingesting CHO at a rate of 1.5 g/min. The

results indicated no significant differences in peak exogenous CHO oxidation rates, with both exercises displaying similar time courses. The study findings suggest both running and cycling exercise can be used interchangeably to investigate CHO oxidation rates. Therefore, the current study, which focuses on CHO personalisation using cycling as the chosen mode, can provide valuable insights applicable to other endurance sports including running.

1.7.4. Habitual diet

An individual's habitual diet has been investigated sparsely when considering the influence on exogenous CHO oxidation rates. One paper of note identified that by altering total daily intake of CHO, it is possible to modify metabolic adaptations to exercise, including changes in CHO oxidation rates in male endurance athletes (Cox et al., 2010). Conversely, Burke et al. (2021) found the opposite in elite male race walkers, and that changes in diet didn't modulate exogenous CHO oxidation rates. There is some indication that athletes adhering to lower CHO diets may exhibit reduced reliance on CHO and enhanced muscle adaptation compared to those with higher CHO diets (Burke, 2019). However, these diets may also lead to lower CHO availability during exercise, potentially limiting an individual's capacity to train at the necessary volume and intensity to enhance performance. This is particularly important to consider for elite endurance athletes, who can require >25 hours of endurance exercise training per week to improve performance. Further investigation into the impact of diet on substrate utilisation and exogenous CHO oxidation rates is clearly required to help further inform the 'prescription' of CHO doses.

1.7.5. Sex

Sex has been demonstrated to impact substrate oxidation due to hormonal and sex-specific differences in lipid storage within muscles and the liver (Beaudry & Devries, 2019). In previous literature, studies investigating the effect of CHO ingestion on exercise performance have predominantly used male participants, limiting our understanding of the metabolic responses to CHO ingestion in females. In males, the oxidation of ingested CHO and the suppression of hepatic glucose production during exercise depends on the rate of CHO intake (Jeukendrup et al., 1999). There is an increasing volume of literature available to address CHO metabolism and fuel utilisation in females.

More recently, Wallis et al. (2006) identified that the metabolic responses to CHO ingestion between males and females during cycling exercise were similar, with no significant difference in the oxidation rates of ingested CHO. Two other studies have also investigated the sex-related differences in substrate metabolism when CHO is ingested during exercise; one of which identified strong trends to suggest that females may oxidise a greater relative

proportion of exogenous CHO during endurance training compared to males (Riddell et al., 2003).

On the other hand, M'Kaouar et al. (2004) found that despite lower rates of absolute exogenous CHO oxidation in females relative to men, the contributions of fat and CHO to the total energy yield, at the same relative exercise intensity, were similar. Further to this, Wallis et al. (2007) demonstrated that increasing CHO ingestion rates from 0.5 to 1.0 g/min increased exogenous CHO oxidation by 42%. Beyond 1.5 g/min, further increases in CHO ingestion showed no further increases in exogenous CHO oxidation rates in trained females. This supports the comparable literature analysing trained male athletes when measuring glucose ingestion during exercise. Despite peak exogenous CHO oxidation rates in females occurring at ~0.5 g/min, these author's findings underpin the theory that CHO oxidation rates do not increase when CHO ingestion exceeds 1.0-1.2 g/min during exercise.
More recently, Pettersson et al. (2019) measured both CHO and fat oxidation rates across 120 minutes of skiing exercise, following the supplementation of CHO at a rate of 2.2g/min. They found no differences between male and female substrate utilisation or, interestingly, performance, following this supplementation of CHO.

Expanding research into CHO utilisation and oxidation in females would help tailor nutritional strategies to individual needs, supporting the case for personalisation. If a clearer relationship between sex and CHO intake were established, it could potentially influence current guidelines on exercise fueling practices.

1.7.6. Body size and body mass

It has been suggested that the primary limitation on exogenous CHO oxidation rates is the capacity for intestinal CHO absorption (Fuchs, Gonzalez, Van Loon, 2019). Therefore, as taller, larger individuals typically possess a greater intestinal surface area for CHO absorption, it stands to reason that exogenous CHO oxidation rates might scale with body size. These individuals also tend to have greater liver and muscle mass, which both facilitate higher rates of CHO metabolism. Evidence supporting this has been conducted on resting individuals, and demonstrated that individuals who are taller and/or have higher fat-free mass, can absorb glucose more rapidly (Anderwald *et al.,* 2011; Færch *et al.,* 2013).

However, until recently, evidence from studies conducted during exercise did not consistently show a clear relationship between total body mass and exogenous CHO oxidation rates (Jeukendrup, 2010). Accordingly, current sports nutrition guidelines recommend CHO intake based on *grams per hour* rather than *grams per kilogram* of body mass (or kilogram of

fat-free mass) per hour, reflecting evidence that shows no consistent relationship between body mass and exogenous CHO oxidation rates.

Despite the theoretical expectation that body size influences intestinal CHO absorption capacity and therefore, exogenous CHO oxidation rates during exercise, empirical evidence supporting this hypothesis remains limited. Therefore, there is a critical need to investigate whether body mass is indeed related to exogenous CHO oxidation rates during exercise. Establishing such a relationship would provide a basis for potentially revising current sports nutrition guidelines for CHO intake. More recently, Ijaz *et al.* (2024) has established a positive relationship between exogenous glucose oxidation during exercise and body size, with smaller athletes (<70 kg) oxidising on average, 13 g/h less than larger athletes (>70 kg) during the same relative exercise intensity. Further research is needed to explore these relationships, but mitigating GI discomfort and optimising CHO utilisation may be achieved through the personalisation of CHO intake. This would involve tailoring intake to match the individual gut absorption capacities, thereby maximising CHO oxidation rates.

1.8. Factors affecting the variability of oxidation rates

A number of factors occurring within day-to-day variations, as outlined above, contribute to variability in exogenous CHO oxidation rates. It is therefore crucial to explore this variability and evaluate the reliability of the measurement methods used to understand the necessity for personalised CHO intake.

Exogenous CHO oxidation rates with glucose feeding have generally been illustrated to increase in a dose-dependent manner. It has been noted that despite this, when ingesting >1.0 to 1.2 g/min of CHO, plateaus in peak oxidation can range from 0.5 to 1.1 g/min (Odell et al., 2022). Hearris et al. (2022) examined exogenous CHO oxidation rates while participants consumed 2 g/min CHO in various forms. The study highlighted significant interindividual variability in peak CHO oxidation rates, further emphasising the importance of personalised fuelling strategies to optimise CHO intake and avoid GI discomfort from a one-size-fits-all recommendation. Some of the previous research available has shown no significant differences in exogenous CHO oxidation rates between trained and untrained men (Jeukendrup et al., 1997). This investigation by Jeukendrup and colleagues compared the substrate utilisation between both groups at an exercise intensity of 60% VO2max. Despite differences in absolute exercise intensity, both groups demonstrated the same CHO oxidation rates (with 0.96 g/min for trained men and 0.96 g/min for untrained men) when provided with 1.1 g/min of CHO. Subsequent studies (e.g. Van Loon et al., 2001) have supported these initial findings.

1.9. Gastrointestinal discomfort

Gastrointestinal (GI) discomfort can significantly impact an individual's ability to consume the necessary volume of CHO to optimise performance. Oxidation rates aside, issues related to GI comfort are equally important to consider in terms of CHO personalisation. In endurance events, inadequate nutritional planning can contribute to GI discomfort from over-fuelling, preventing athletes from consuming the doses necessary and could result in under-fueling and reduced CHO availability, ultimately impairing performance. These GI issues typically arise from an accumulation of CHO in the gut, which occurs when individuals consume CHO in doses exceeding their absorption capacity. Additionally, GI discomfort is frequently reported by athletes during both training sessions and competitive events at higher intensities, despite adherence to recommended CHO fueling guidelines (Peters *et al.,* 1999; Pfeiffer *et al.,* 2012). Enhancing an individual's CHO tolerance and gut absorptive capacity could mitigate these issues. Regular CHO consumption, and 'gut training' with CHO-based products during exercise are strategies suggested to improve CHO absorption (Ferraris, 2001). Studies have established the possibility of maintaining high oxidation rates while minimising Gi discomfort (Jeukendrup, 2014).

1.10. Personalisation

The personalisation of CHO intake is increasingly looked at within the field of sports nutrition (Jeukendrup, 2014). The variability in oxidation rates observed between individuals suggests that personalisation could be a method to optimise the effectiveness of CHO supplementation during exercise, and prevent GI distress. The investigation and understanding of the extent of this inter-individual variability may encourage the personalisation of CHO and support further research. Currently, both practitioners and athletes can help personalise energy and CHO intake based on preference and tolerance (Podlogar & Wallis, 2022). The use of power meters and heart rate monitors have enabled individuals to determine energy turnover during exercise and, subsequently, calculate and adjust energy and CHO intakes. Aforementioned tracer-based methodologies have been used to investigate exogenous CHO oxidation rates, and Podlogar & Wallis (2022) have suggested that it might be possible to personalise CHO intakes based exogenous CHO oxidation rates, but this is yet to be tested. Identifying suitable CHO doses for individuals will likely face issues surrounding the reliability and repeatability of these tests and methods. As a result, a suitable aim within the present investigation and for future research is to identify the reliability and validity of these methods, to understand whether it would be possible to provide one test to identify CHO oxidation rates in athletes, and confidence that it is accurate for subsequent use in both training and racing.

This introduction has highlighted gaps in the current literature that underpin the necessity for personalised CHO strategies to enhance athletes' performance during prolonged exercise. Furthermore, personalised approaches are crucial to mitigate potential GI discomfort and issues associated with excessive and insufficient CHO intake. Therefore, the main aims of this thesis are outlined as follows.

1.11. Aims

The aims of this study are:

1. To establish if maximal exogenous CHO oxidation rates obtained after large amounts of glucose are provided during prolonged exercise, could be used to determine a personalised CHO dose.

   It is expected that similar exogenous CHO oxidation rates will be observed within participants with both the larger (90g) dose and the personalised carbohydrate dose.

2. To investigate the repeatability of determining exogenous carbohydrate oxidation rates during exercise with the larger (90g) CHO dose; this will be performed using two identical trials.

   It is expected that despite daily variations, exogenous CHO oxidation rates will be similar within participants in the two repeated trials.

CHAPTER 2

Materials and Methods

2.1. Participants

Between January 2023 and September 2023, fourteen healthy individuals were assessed for their eligibility to participate in the study. Eleven of the individuals completed the protocol having successfully met the minimum inclusion criteria; generally healthy, partaking in endurance-based exercise ≥3 times per week, $\dot{V}O_2$ max in females ≥50 mL/kg/min and in men ≥55 mL/kg/min.

Data collection was completed by September 2023 (see Figure 2.1 for a flow chart of participant recruitment and involvement in the study). Participants were recruited by email, social media, and contacting local running, cycling and triathlon clubs. Participants were excluded from taking part if they were: <18 or >50 years old, with BMI <18.0 or >27 kg/m^2, or taking any medication or supplements with the potential to interfere with normal metabolism (e.g., beta-blockers, insulin, anti-inflammatory agents, bronchodilators, thyroxine), history of smoking, engaging in prolonged periods of fasting or dietary restraint or existence of food intolerances, pregnant or breastfeeding, bone, or joint problems. Participants were also excluded if they suffered from claustrophobia. Participants provided written informed consent in accordance with the Helsinki Declaration of 1975 as revised in 1983 to take part in the study. The study was approved by the University of Birmingham Science Technology Engineering and Mathematics Ethics Committee (Ref: ERN_ 0733).

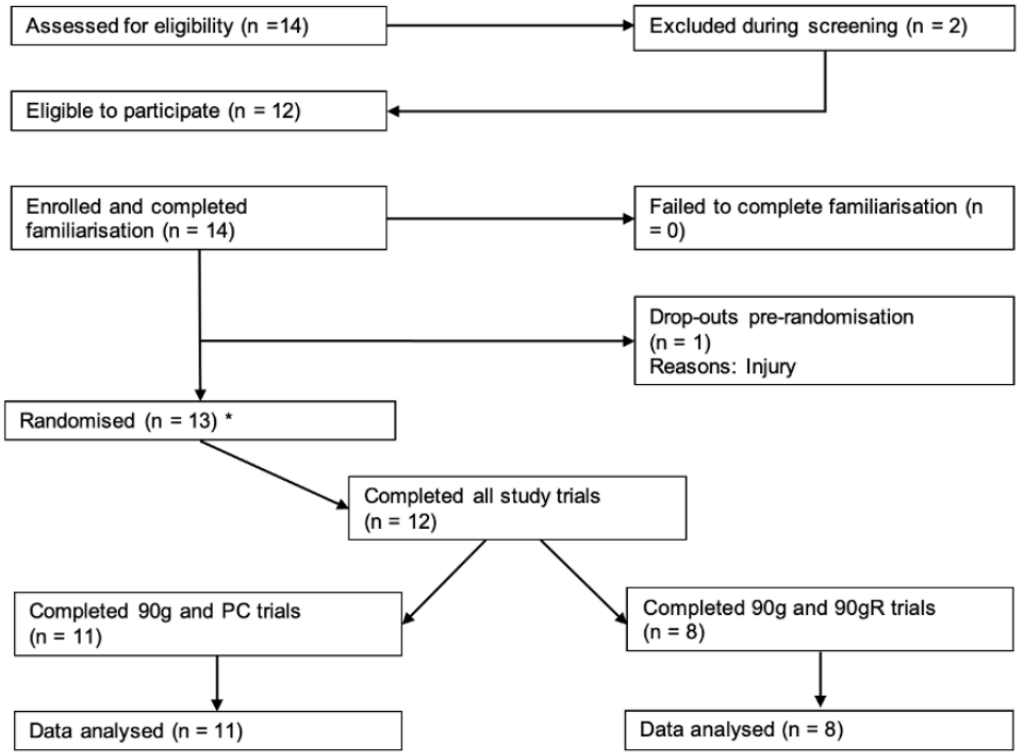

*Figure 1. A schematic to illustrate the number of participants within the present study. *The participants main trial 2 and main trial 3 were randomised where appropriate.*

All volunteers were deemed to be healthy as assessed by a general health questionnaire. The mean ±SD characteristics at baseline of the 11 participants who completed the study are shown in Table 1 below.

2.2. Menstrual cycle

Female participants were asked to self-report details on their menstrual cycle and possible contraceptive use before testing. Of the six who participated, two females reported to have a regular menstrual cycle with no contraceptive use. One female participant reported not having a regular menstrual cycle. Three female participants reported the use of contraception; this included, the Mirena IUD, and the oral contraceptives Rigevidon and Microgynon. No control for the menstrual cycle phase or contraceptive use was utilised throughout the study.

Table 1. Participant characteristics. All values are presented as Mean ±SD. Female participants, n=6. Male participants, n=5. Overall, n=11.

	All	Male	Female
Age, yrs	23±2	24±3	22±0
Height, cm	171±11	179±10	164±5
Body Mass, kg	66.4±9.3	73.7±8.4	60.3±4.1
Fat Mass, kg	11.0±5.4	6.6±3.5	14.7±3.5
Fat Free Mass, kg	55.5±12.6	67.1±8.0	45.9±4.3
Body Fat, %	17.2±9.3	8.9±4.6	24.2±5.2
$\dot{V}O_2$ max, mL/kg/min	59.2±7.3	65.4±6.0	54.1
LT1, W	194±48	229±51	166±18
LT1, W/kg	2.9±0.6	3.2±0.8	2.7±0.2
LT2, W	236±55	285±41	195±17
LT2, W/kg	3.5±0.6	3.9±0.8	3.2±0.1
Workload for exercise trials, W	184±46	217±49	157±17

2.3. Study Design

This study followed a partially randomised design. After preliminary testing, each participant completed a familiarisation trial and three experimental trials. Each experimental trial consisted of one 150-minute exercise session. The experimental trials differed in the carbohydrate dose provided to be ingested during the exercise bout. In the first experimental trial, all participants received 1.5 g/min CHO. In the second and third experimental trials, participants received a specific dose (dependent on their exogenous oxidation rate, determined during Trial 1) or 1.5 g/min CHO again, in a randomised order. Experimental trials were separated by approximately 7-14 days. All testing took place at the University of Birmingham.

2.4. Screening, baseline testing and familiarisation

After gathering informed consent from participants and noting their completion of a general health questionnaire, characteristic data was recorded including height (Model 220; Seca,

Germany), body mass (Champ II, OHAUS, Switzerland) and body composition (BOD POD GS-X, COSMED Italy, Inc). The BOD POD is a computerised device which measures body volume by monitoring changes in pressure using Air Displacement Plethysmography within a closed chamber. Participants then undertook two incremental tests (one to task failure) on a cycle ergometer (WattBike Atom, WattBike Ltd, Nottingham, UK). The first was a submaximal aerobic exercise test to determine both lactate thresholds followed by a 20-minute rest and then a second test which was a maximal exercise test to assess $\dot{V}O_2$ max and Wmax. This was used to confirm eligibility.

2.5. Sub-maximal test:

The first test protocol started between 50-100 W depending on the individual (determined by estimated thresholds, training status and body mass). It involved 4-minute-long stages of easy-to-moderate intensity cycling, each stage increasing by 25 W and finishing before volitional exhaustion. During the submaximal test, at the last minute of each stage (3 minutes), a lancet was used to take a small sample of blood from the participant's finger to identify blood lactate concentration. The lancet was used to provide 20 µL of blood which was then collected into a capillary tube. This capillary was then placed into a cup before mixing. The cup was placed directly into a Biosen slot (Biosen C-Line Glucose and Lactate analyser, EKF-diagnostic GmbH, Germany) to be analysed, reporting lactate values in mmol/L. Heart rate was continuously monitored and recorded using telemetry, with participants wearing a chest strap heart rate monitor (Polar 10, Polar Electro Oy) and RPE (Borg, 1982) was recorded at the end of each stage. The first(LT1) and second (LT2) lactate thresholds were determined using the “ExPhysLab App”, (ExPhysLab, August 2022). Using this App, the baseline plus method (Bsln+) is implemented. This considers LT1 as the exercise intensity at which lactate increases to 0.5 mmol/L above baseline (resting) values (Berg *et al.,* 1990; Zoladz *et al.,* 1995). LT2 is typically the maximum workload that precedes a rapid increase in blood lactate values resulting from an imbalance between lactate production and clearance, which is accompanied by a rise in blood H^+ (Binder *et al.,* 2008). As above, the ExPhysLab App was used to calculate this by identifying a lactate curve, fitted using a 3rd-order polynomial regression curve. LT2 was therefore identified as the exercise intensity yielding the maximum perpendicular distance to the straight line between the data point preceding the first rise in lactate greater than 0.4 mmol/L and the last data point (Bishop, Jenkins & Mackinnon, 1998). The researcher stopped the sub-max test when participants were reaching near maximal effort based on perceived exertion and predicted maximal effort. On completion of the sub-max test, participants rested for 10-20 minutes before commencing the maximal exercise test.

2.6. Maximal ($\dot{V}O_2$max) test:

The maximal test began at the same intensity as the sub-max test. The intensity increased by 25 W every minute until cadence could not be sustained. During the maximal test, a facemask was securely fitted to the participant to enable breath-by-breath respiratory measurements, including minute ventilation, $\dot{V}E$; oxygen consumption, $\dot{V}O_2$; carbon dioxide production, and $\dot{V}CO_2$. Gas exchange measurements were made using an automated online gas analysis system (Vyntus, Vyaire Medical, IL, US). The gas analysers used were calibrated with a known gas mixture (15.04 % O_2, 5.06 % CO_2; BOC Gases, Surrey, UK) as per the manufacturer's recommendations. Before each test, a 3-litre directional syringe (Jaeger, Wurzburg, Germany) was used to calibrate the volume transducer.

The data output from the Vyntus was then used to determine $\dot{V}O_2$max by identifying the highest 30-second average of O_2 uptake across the maximal testing duration. This was reported in ml/kg/min. The test ceased when participants reached task failure. Like that of the sub-max test, participants wore a chest strap to record their heart rate throughout the test, (Polar 10, Polar Electro Oy). Following the maximal test, participants were given a 10-minute break before being familiarised with the testing protocol to be used in the main experimental trials to remove the procedure's novelty and ensure they could tolerate the CHO ingestion protocol. This involved completing a 60-minute familiarisation exercise session at the intensity correlating to 95% LT1 and the carbohydrate ingestion rate prescribed during the first of the main exercise trials (i.e., 1.5 g/min, equivalent to 90 g/h).

After establishing eligibility for the study, participants were scheduled for the three experimental trials. Experimental trials 1 and 2 were scheduled to be separated by 2-3 weeks, due to analysis of the breath required before estimating the amount of glucose oxidised in visit 1. Following this, visits 2 and 3 were separated by approximately 7 days.

2.7. Main Experimental Trials

An illustration of the experimental trial design can be seen in Figure 2.

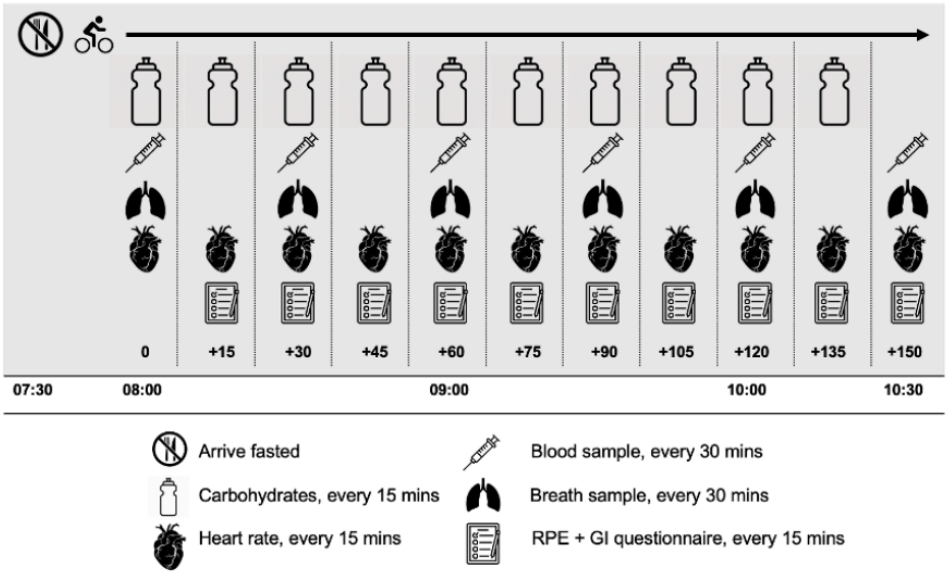

Figure 2. A visual illustration of the experimental trials. Participants arrived at the laboratory fasted, before completing 150-minutes at the intensity corresponding to 95% LT1. At 15-minute intervals, they received a carbohydrate-containing drink. Heart rate and questionnaires assessing GI and RPE were administered every 15 minutes. Blood and breath samples were taken every 30 minutes.

Participants arrived at the laboratory in the morning (between 7 and 9 a.m.) following a 10-hour overnight fast except for water. Participants were asked to refrain from exhaustive exercise and alcohol for 24 hours prior to the experimental trials (visits 2, 3, 4). To ensure standardisation of the diet preceding each experimental trial, participants were asked to record all food and fluid consumed on the day before the first experimental trial. This eating pattern was repeated before each subsequent experimental trial. Participants were asked to maintain exercise patterns and physical activity levels across the trial period. On arrival, body mass was measured (Champ II, OHAUS, Switzerland). The environmental conditions of the laboratory were monitored throughout each visit using the Vyntus (Vyaire Medical, IL, US). On average (± SD), ambient air pressure (Pa), temperature (ºC) and humidity (%) were 99725 (1466), 20 (2) and 60 (7) respectively. Participants were asked to put on a chest strap heart rate monitor. An antecubital venous cannula (BD Venflon IV Cannula, 20g, MidMeds Ltd, UK) was inserted and connected to a three-way stopcock to obtain 6 ml samples before exercise and every 30 minutes during exercise until completion. This enabled the collection of blood throughout the trial without the need to change or stop participants' cycling cadence. All blood samples were immediately stored on ice. Resting expired breath samples were collected into 10mL evacuated tubes (Exetainer Breath Vial, Labco Ltd.: Buckinghamshire, UK) and were filled from a mixing chamber to subsequently determine the ratio of $^{13}C/^{12}C$

ratio at rest and every 30 minutes during the 150-minute exercise bout. The experimental trial consisted of 150 minutes of exercise at the intensity corresponding to 95% of LT1, determined previously described. As mentioned, the first lactate threshold (LT1) is defined as the lowest intensity at which there is a sustained increase in blood lactate concentration above resting values, categorised at 0.5 mmol/L. Expressing workloads relative to this threshold has been shown to minimise between-subject variation when compared with using $\dot{V}O_2$max or W_{max}. In addition, discrepancies in $\dot{V}O_2$max values have been observed between participants who possess similar athletic capabilities (Jones *et al.,* 2021). The intensity of 95% LT1 was favourable because it provided sufficient energy demands to favour CHO oxidation, and participants were able to maintain the required effort for the set duration without failure. Questionnaires to assess self-rated perceived exertion (RPE) using a 6-20 scale (Borg, 1982) and gastrointestinal (GI) issues were administered every 15 mins during the exercise period of the experimental visits. GI was assessed using a 10-point Likert scale (Thorburn *et al.,* 2006) to include the assessment of nausea, abdominal cramping, and stomach fullness. It should also be noted that 8 out of the total 11 participants completed the reliability trial due to limited resources for testing and participant's time commitments.

2.8. Nutritional manipulation – test beverages

Participants ingested a total of 2.5 L of fluid during each exercise trial. During the first 2-3 minutes of exercise, participants were asked to consume the first CHO-containing drink (250mL). Following this, participants were asked to ingest 9 equal doses of carbohydrate-containing drinks at 15-minute intervals during the 150-minute exercise bout. The drinks contained carbohydrates in the form of glucose (Dextrose, Bulk Powders, Essex, UK). The first experimental trial provided carbohydrates at a rate of 1.5 g/min (90 g/h CHO). Previous literature suggests that when ingesting glucose only during exercise, exogenous CHO oxidation rates appear to peak at ~ 1 g/min (Jeukendrup, 2014). This is likely to be limited by intestinal glucose absorption. As a result, in this study, a saturating dose of CHO was provided at a rate of 1.5 g/min of glucose to ensure peak exogenous glucose oxidation was achieved. This also accounted for any possible individual variation where individuals may exhibit exogenous CHO oxidation rates greater than 1 g/min. The second and third experimental trials were random in order. One of these repeated the dose of 1.5 g/min (90 g/h) of CHO to assess reliability. The other experimental trial (known as the personalised or 'PC' trial) provided the participants with the same amount of fluid but a different dose of CHO. The dose was determined during the first of the three experimental trials and informed based on each participant's exogenous CHO oxidation rate when provided with 1.5 g/min CHO across the 150-minute exercise trial. See the section below (Breath Analysis) for how the dose was determined.

The drinks contained a small amount of the stable isotope tracer U-$^{13}C_6$-glucose with 99% purity (Cambridge Isotope Laboratories, Inc; Massachusetts, USA), to determine exogenous carbohydrate oxidation during exercise from expired breath measurements. Drinks were enriched to 0.5 mg/g relative to each dose of glucose provided (calculated before each trial, based on individual oxidation rates, and required carbohydrate doses), with the stable isotope U-$^{13}C_6$-glucose. Drink enrichments as averages (±SD) for 90g, PC, and 90g repeat trials were: 34.74 (2.23), 33.70 (2.46), and 34.22 (2.41) [$d^{13}C_{V\text{-}PBD}$ (‰)], respectively. The ^{13}C enrichment of the ingested CHO was determined by isotope ratio mass spectrometry (see section below, Breath Analysis).

2.9. Breath Analysis

2.9.1. Indirect Calorimetry

The principles of indirect calorimetry were utilised within this project to assess carbohydrate and fat oxidation during exercise. The use of indirect calorimetry to measure oxygen uptake ($\dot{V}O_2$) from and carbon dioxide production ($\dot{V}CO_2$) and calculate substrate oxidation is based on several assumptions; a) the complete oxidation of carbohydrates and fats results in the production of carbon dioxide and water, and b) the chemical structure and amount of oxygen needed for the complete oxidation of fats and carbohydrates differ. Additionally, c) proteins present a negligible part within substrate oxidation, and consequently, all the energy for ATP synthesis came from the complete oxidation of CHO and fats. In the present study, this assumption should not influence the data as it is likely protein oxidation is minimal, particularly when individuals are fed CHO, (Jeukendrup and Wallis, 2005).

Calculations of fat and total carbohydrate oxidation rates were calculated using the following stoichiometric equations, with protein oxidation assumed to be negligible (Jeukendrup and Wallis, 2005).

$$\text{Total CHO oxidation} = 4.210\ \dot{V}CO_2 - 2.962\ \dot{V}O_2$$

$$\text{Fax oxidation} = 1.695\ \dot{V}O_2 - 1.701\dot{V}CO_2$$

Where $\dot{V}CO_2$ and $\dot{V}O_2$ are measured in litres per minute (L/min) and oxidation rates in grams per minute (g/min).

2.9.2. Stable Isotopes

The isotopic enrichment ($\delta^{13}C$) of expired breath samples was determined using gas chromatography isotope ratio mass spectrometry (analysed at an external laboratory, Iso-Analytical Ltd, Crew, UK). In this technique, the contents of the samples were flushed

from the sample vials via a double-holed needle through a packed column gas chromatographer held at 75° C. The resultant chromatographic peak passed into a Europa Scientifica Hydra 20-20 where the isotopomers at a mass-to-charge ratio (m/z) of 44, 45, and 46 for CO_2 were measured. This was expressed as **δ** per millilitre difference between the $^{13}C/^{12}C$ of the sample and a known laboratory reference standard. $\boldsymbol{\delta}^{13}C$ was then related to an international standard; the reference gas used to determine the $\boldsymbol{\delta}^{13}C$ value of the sample gases was IA-CO2-9 ($\boldsymbol{\delta}^{13}C$ = -39.57 ‰ vs. V-PBD) which was prepared to 3.3 % CO_2 by volumetric dilution. This is traceable to NBS-18 Calcite ($\boldsymbol{\delta}^{13}C$ value of –5.01 ‰ vs. V-PBD) and IAEA-CO-1 Marble ($\boldsymbol{\delta}^{13}C$ value of +2.49 ‰ vs. V-PBD), which are distributed as isotope reference standards by the International Atomic Energy Agency, Vienna.

The ^{13}C enrichment of ingested carbohydrates was determined by elemental analyser isotope ratio mass spectrometry (Iso-Analytical Ltd, Crewe, UK). The sample aliquots were pipetted into capsules and dried before analysis. These capsules were loaded into an auto-sampler on a Europa Scientific elemental analyser. CO_2 is separated from the other gases in a column gas chromatograph held at an isothermal temperature. The resultant CO_2 chromatographic peak enters the ion source of the Europa Scientific 20-20 where it is ionised and accelerated. Gas species of different masses are separated in a magnetic field and then simultaneously measured using a Faraday cup collector array to measure the isotopomers of CO_2 at m/z 44, 45, and 46. The reference material used during $\delta^{13}C$ analysis of the samples was IA-R080 (beet sugar, $\boldsymbol{\delta}^{13}C_{V\text{-}PBD}$ = -25.53 ‰). For quality control purposes check samples of IA-R006 (cane sugar, $\boldsymbol{\delta}^{13}C_{V\text{-}PBD}$ = -11.64 ‰) and IA-R071 (middle sugar [beet/cane 50:50 mix], $\boldsymbol{\delta}^{13}C_{V\text{-}PDB}$ = -19.26 ‰) were analysed with the samples. IA-R005, IA-R006, and IA-R071 are in-house standards and are calibrated against and traceable to IAEA-CH-6. IAEA-CH-6 is an inter-laboratory comparison standard distributed by the International Atomic Energy Agency (IAEA), Vienna.

2.9.3. Calculation of Oxidation Rates and the Personalised Dose

Exogenous CHO oxidation rates were calculated using the following equation (Pirnay *et al.,* 1997):

$$\text{Exogenous carbohydrate oxidation} = \dot{V}CO_2 \left[\frac{\delta Exp - \delta Exp_{bkg}}{\delta Ing - \delta Exp_{bkg}}\right]\left(\frac{1}{k}\right)$$

Where CO_2 is in L/min, is the ^{13}C enrichment of expired air during exercise at different timepoints, is the ^{13}C enrichment of the ingested CHO solution, is the ^{13}C enrichment of

expired air at time point 0, and (0.74326 L/g) is the volume of CO_2 produced when 1 g of glucose is completely oxidised.

Endogenous carbohydrate oxidation rates were then calculated as the difference between total CHO oxidation and exogenous CHO oxidation rate. Exogenous CHO oxidation rates were analysed for the whole duration of the study as 30 minutes is sufficient for the bicarbonate pool to turn over, which means that the $^{13}CO_2$ observed in the breath reflects that of the $^{13}CO_2$ produced from the muscle during exercise (Podlogar and Wallis, 2020). The peak exogenous oxidation rate was determined as the highest observed value for each participant during the exercise trial and used to calculate the dose for the “personalised” trial. The method to calculate this involved using oxidation efficiency. Oxidation efficiency describes the % of ingested CHO that are oxidised. A greater oxidation efficiency would suggest that less CHO is remaining in the gut and consequently reduces the risk of causing GI discomfort. It has been observed that the oxidation efficiency of exogenous CHO (glucose) during exercise of a similar intensity falls ~70-90% (Jentjens *et al.,* 2004), this was consequently estimated at 80% efficiency. A calculation could then be implemented to identify CHO doses for the remaining exercise trial; the exogenous oxidation rate from the first experimental trial for each participant was divided by 0.8 (80%) to identify the rate at which to provide CHO in the personalised trial. For example, if a participant's exogenous CHO oxidation rate was 1.0 g CHO/min, carbohydrates would be provided based on this equation: 1.0 x 60 / 0.8 = 75 g CHO/hour.

2.10. Blood Sampling and Plasma Analysis

*Capillary blood samples; analysis using the Biosen C-line Glucose and Lactate analyser (*EKF-diagnostic GmbH, Germany).

Finger-prick capillary blood samples were used for analysis. The Biosen was calibrated before each exercise threshold test. A new micro-tube of multi-standard was inserted at the start of each testing day to calibrate the system according to the manufacturer’s manual. To measure the blood sample, a capillary tube was filled with free-flowing blood from the tip of the finger (pricked using a lancet). This was placed into a microtube containing glucose haemolysing solution, sealed, and inverted ten times, before being placed into the sample tray. The concentration was then measured (mmol/L). This was completed for each blood sample across the sub-maximal test. The Biosen works based on an electrochemical principle. The glucose contained in the sample is enzymatically converted with immobilised enzyme glucose oxidase found on the chip sensor. The H_2O_2 generated by the reaction is detected at the electrode. The sensor current is proportional to the glucose concentration in

the sample and is used to provide the measurement. Following each measurement, the system is flushed.

2.10.1. Venous blood samples; analysis of plasma using an automated clinical analyser

The venous blood samples were stored on ice immediately following collection. The blood samples were centrifuged at 3000 g for 15 minutes at 4 ºC. Aliquots containing cell-free plasma were immediately stored and frozen at -70 ºC until later analysed for the determination of metabolite concentrations. Plasma samples were analysed using commercially available kits using an automated photometric clinical chemistry analyser (RX Daytona+ Randox, London, UK) to determine glucose (Glucose, Randox, UK) and lactate (Lactate, Randox, UK) concentrations. The RX Daytona+ operates based on the principles of measuring the absorbance of light. Before the analysis of all blood glucose and lactate samples, quality checks were carried out for each metabolite as per the manufacturer's instruction and where necessary, calibrations were made. In the instance of measuring glucose concentration, using the glucose kit (Randox, UK), the method is as follows. The plasma sample is initially incubated with hexokinase to phosphorylate the glucose to glucose-6-phosphate. This is then oxidised by glucose-6-phosphate dehydrogenase to form NADH. NADH presents a strong absorbance at 450 nm. The Daytona+ measures absorbance at this wavelength and using the calibration curve, can provide the concentration of the metabolite (glucose) in the solution (plasma). In the instance of measuring lactate concentration, the lactate kit (Randox, UK), is used based on the same principle where the subsequent oxidation of the sample to form NADH can be measured (based on the absorbance of NADH and the calibration curve). The coefficient of variations (CV) for this can be seen below in Table 2.

Table 2. Analytes and measurement variation.
All analyte intra-assay CVs were calculated from technical repeats from the same analysis or plate. Inter-assay CVs for plasma glucose and lactate were calculated from repeats of control solutions.

	Coefficient of Variation (%)	
Analyte	Intra-assay	Inter-assay
Plasma Glucose	1.0	3.5
Plasma Lactate	1.5	3.4

2.11. Statistics

All data was processed in either Excel (Microsoft Excel, version 16.78.3) or Jamovi (Jamovi, version 1.2.27.0) as appropriate and all values are presented as mean ±SD unless otherwise stated. The 90 vs PC trials and the 90 vs 90R trials were treated as separate parts throughout the thesis and thus are statistically analysed as such. Participant average (over the whole trial) physiological and perceptual responses, substrate oxidation AUC (area under the curve; the trapezoid rule) and/ or peak exogenous CHO oxidation responses to the conditions were compared by paired sample T-tests. Substrate oxidation rates and physiological and/ or perceptual responses across time were analysed using repeated measures ANOVA. Bland-Altman plots were used to produce quantitative estimates of agreements between the trials. Pearson's Correlation and the correlation coefficient, R^2 were used to explore relationships between body mass, height, and absolute workload with exogenous CHO oxidation rates of participants across all trials. All values are presented as mean ±SD. Statistical significance was set at $p < 0.05$.

CHAPTER 3

Results

Participants completed three exercise trials at the exercise intensity corresponding with 95% LT1. Trial 1 (90g) informed the CHO dose for the personalised (PC) trial. The other trial was a repeat of Trial 1, referred to as 90gR. During trial 1, all participants were given 90 g of carbohydrates per hour of exercise (225 g total for the exercise duration). Expired breath analysis was used to quantify the peak exogenous CHO oxidation rate for each participant (reported as g/min). This was used to calculate the dose per hour for the PC trial for participants (ie., x*60/0.8), assuming 80% oxidation efficiency.

This section is presented in two parts. The first part will compare the 90g and PC trials to reflect the first aim of the thesis in identifying whether the same rates of exogenous CHO oxidation are observed with a set (90g) dose and a personalised dose of carbohydrates.
The second part will compare the 90g and 90gR trials to reflect the reliability aim of this thesis; assessing whether across different days, exogenous CHO oxidation is similar, and the trial is repeatable.

3.1. Personalisation

3.1.1. Carbohydrate dose

The average peak exogenous CHO oxidation rate derived from the 90g trial was 0.91±0.15 g/min. This resulted in a personalised dose of 68g±12 g/h CHO, on average (*n = 11*). The individual CHO dose for each participant in the personalised (PC) trial is illustrated in Table 3. This was established based on their oxidation rates, determined in the first exercise trial.

Table 3. The carbohydrate doses that were provided to participants in the PC trial.

Participant ID	P1	P2	P3	P4	P6	P7	P8	P9	P12	P13	P14
CHO (g/h)	83	60	61	52	83	75	78	52	70	76	63

3.1.2. Physiological responses

Table 4 illustrates a direct comparison between the physiological responses of the participants between the 90g and PC trials, averaged over the entire exercise duration. A paired samples t-test was used to identify any significant differences between the PC and 90g trials. $\dot{V}CO_2$ and $\dot{V}E$ were marginally but significantly greater in the 90g and PC trials ($p < 0.05$). $\dot{V}O_2$, $\dot{V}E$ and CHO oxidation showed no clear significant difference between the 90g trial compared with the PC trial. RER did not differ between trials. Fat oxidation showed no difference between 90g and PC trials.

Table 4. The physiological responses in both the 90g and PC exercise trials; including $\dot{V}O_2$, $\dot{V}CO_2$, RER, $\dot{V}E$, carbohydrate, and fat oxidation rates. Data are represented as mean ±SD, averages across the trial durations. n = 11.

	Trial 90g	Trial PC
$\dot{V}O_2$ (L/min)	2.69±0.62	2.62±0.62
$\dot{V}CO_2$ (L/min)	2.47±0.55	2.39±0.51
RER	0.92±0.03	0.92±0.03
$\dot{V}E$ (L/min)	67±14	64±14
CHO oxidation (g/min)	2.44±0.51	2.28±0.38
Fat oxidation (g/min)	0.36±0.15	0.38±0.20

Changes in $\dot{V}O_2$, $\dot{V}CO_2$, $\dot{V}E$, and RER between trials and over time were also statistically analysed using repeated measures ANOVA. There was no change in $\dot{V}O_2$ over time ($p = 0.107$) and there was no difference between trials ($p = 0.064$) nor was there a time x trial interaction ($p = 0.866$). There was a significant increase in $\dot{V}CO_2$ over time ($p < 0.05$) but there was no difference observed between trials nor a time x trial interaction; ($p = 0.062$) and ($p = 0.982$), respectively. There was a significant increase in $\dot{V}E$ over time ($p < 0.001$) but there was no clear difference between trials ($p = 0.050$) nor was there a time x trial interaction ($p = 0.903$). There was no change observed in RER over time ($p = 0.975$) and there was no difference observed between trials nor was there a time x trial interaction; ($p = 0.726$) and ($p = 0.665$), respectively.

3.1.3. Substrate oxidation rates

As shown in Figure 3, the exogenous, endogenous and total carbohydrate oxidation rates followed similar trends in both trials. Total, endogenous, and exogenous CHO oxidation rates were statistically analysed using repeated measures ANOVA. Total carbohydrate oxidation rates (Figure 3.a) did not change over time ($p = 0.445$) and there was no difference between trials ($p = 0.243$) nor was there a time x trial interaction ($p = 0.975$). Resting breath ^{13}C enrichment was similar at the start of both the PC and 90g exercise trials, averaging -26.34±1.13 and -26.11±0.91 δ‰ vs PDB respectively. Expired breath ^{13}C is shown below in (Figure 3.d). Exogenous CHO oxidation rates (Figure 3.b) increased over time ($p < 0.001$) but there was no difference between trials observed ($p = 0.315$) nor a time x trial interaction ($p = 0.529$). Endogenous CHO oxidation rates (Figure 3.c) decreased over time ($p < 0.001$) but there was no difference between trials observed ($p = 0.229$) nor a time x trial interaction ($p = 0.439$). The Area Under the Curve (AUC) was calculated using the trapezoidal rule to quantify overall total, endogenous and exogenous carbohydrate oxidation across the three exercise trials. This was calculated from the period of 30-150 minutes. In the 90g trial, the total, exogenous and endogenous carbohydrate oxidation (in grams, g) was 291.6±0.5, 80.6±6.5 and 208.9±6.8 respectively. In the PC trial total, exogenous and endogenous oxidation was 274.9 ±0.4, 83.6±6.0 and 191.3±5.7 respectively. There was no significant difference in the AUC of total ($p = 0.09$), exogenous ($p = 0.06$) and endogenous ($p = 0.77$) CHO between the 90g and PC trials. Fat oxidation was statistically analysed using repeated measures ANOVA. There was no difference observed between trials ($p = 0.879$), nor was there a difference between trials or a time x trial interaction; ($p = 0.738$) and ($p = 0.696$), respectively.

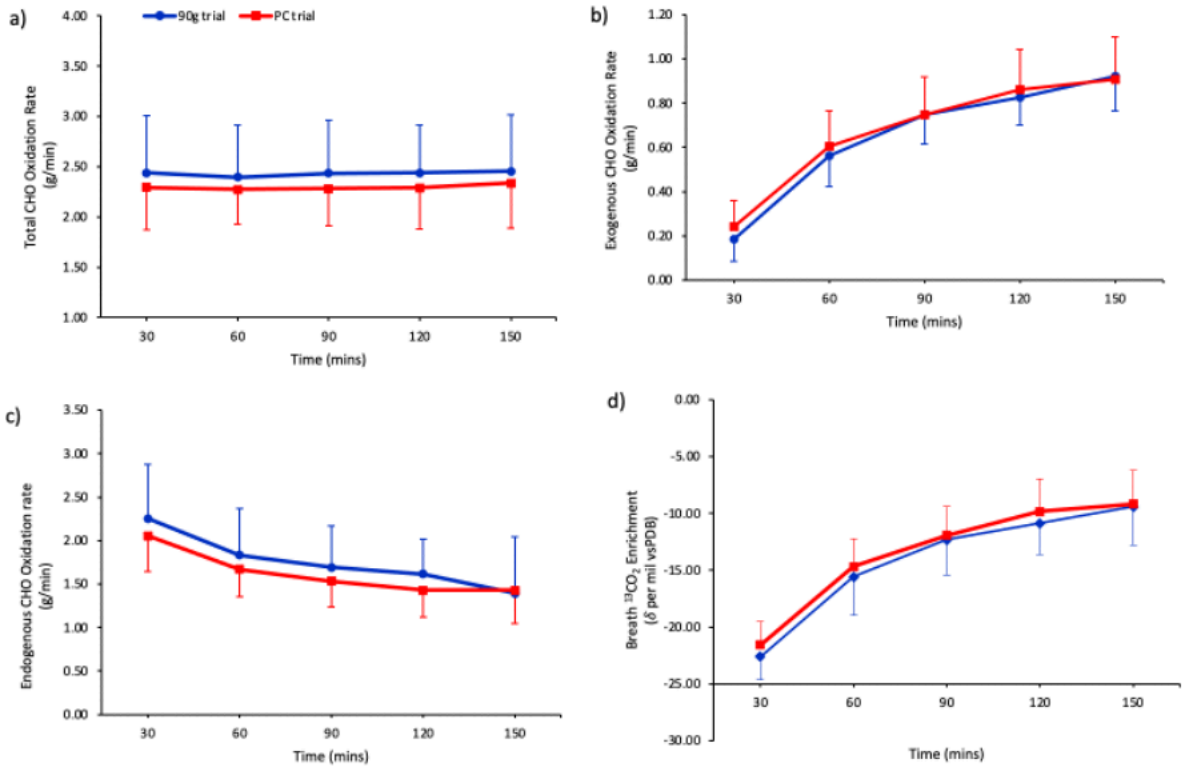

*Figure 3. Total CHO **a,** exogenous CHO **b,** endogenous CHO **c,** oxidation rates and breath enrichment **d,** were measured every 30 min in 90g and PC conditions.*

3.1.4. Peak and average exogenous CHO oxidation rates

There was no difference in the average peak exogenous CHO oxidation rates between 90g and PC trial conditions (Figure 4) (i.e., highest measured value at one time point) 0.91±0.15 vs 0.91±0.19 g/min for the 90g and PC trials, respectively; (p =0.847). The CV of peak exogenous CHO oxidation rates in 90g and PC trials was 19±0.3% as determined in Excel (CV = SD / Mean). Figure 4 illustrates the individual peak exogenous CHO oxidation rates for reference. Figures 5 and 6 are both Bland Altman plots to present the peak exogenous CHO oxidation rates and average exogenous CHO oxidation rates (respectively) across all time points and present the data as a cluster surrounding the average difference.

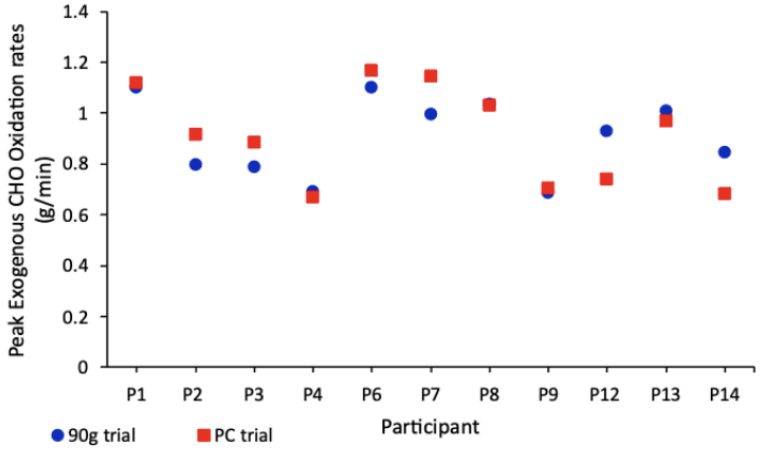

Figure 4. A graph to present the peak exogenous CHO oxidation rates (g/min) per participant in 90g and PC trials; n=11.

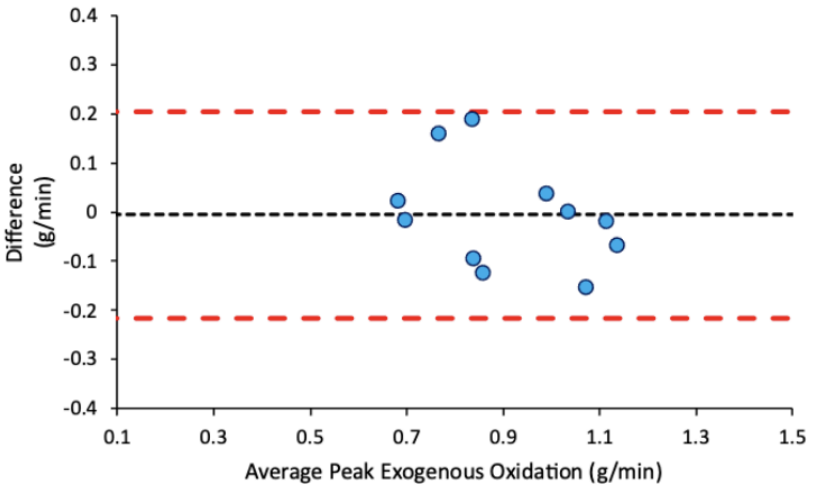

Figure 5. Bland-Altman Plot: Peak exogenous CHO oxidation rates (g/min) in 90g vs PC trial. Legend: average difference = 0.004, upper limit of agreement = 0.227, lower limit of agreement = -0.220 (g/min). The average difference is illustrated by a solid line and 95% limits of agreements by dashed lines.

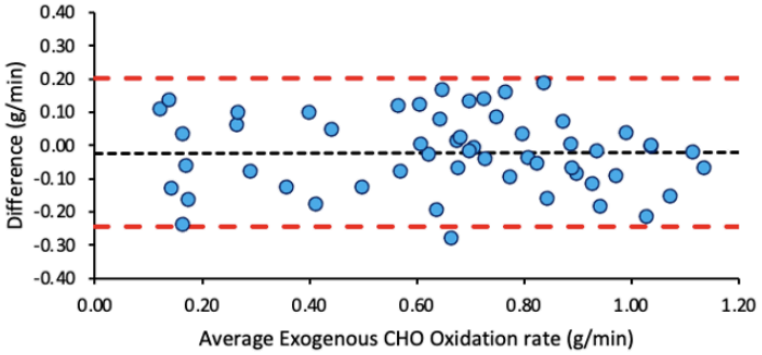

Figure 6. Bland-Altman Plot: Average exogenous CHO oxidation rates (g/min) in 90g vs PC trial in all participants across all time points. Legend: average difference = -0.02, upper limit of agreement = 0.20, lower limit of agreement = -0.25 (g/min). The average difference is illustrated by a black solid line and 95% limits of agreements by red dashed lines.

3.1.5. RPE, Heart Rate and GI Comfort

Average scores for RPE and ratings of GI comfort including nausea, stomach fullness and abdominal cramping are presented in Table 5 compared with t-test values. No significant differences were observed in nausea and abdominal cramping between 90g and PC trials. A t-test illustrated significant differences in stomach fullness, heart rate and RPE; ($p < 0.05$), all of which were marginally higher in the 90g trial than in the PC trial.

*Table 5. Participant GI, HR, RPE. Data are represented as mean ±SD, averages across the trial durations, using a 1-10 scale. n= 11. * Indicates a significant difference between trials of p <0.05.*

	Trial 90g	Trial PC
GI- Nausea	2.4±1.5	2.0±1.3
GI- Stomach Fullness *	2.5±1.1	1.7±1.2
GI-Abdominal Cramping	1.6±1.3	1.6±1.3
HR (bpm) *	148±13	142±12
RPE (Borg scale) *	13±1	12±2

RPE, HR and GI (nausea, stomach fullness, and abdominal cramping) were also analysed between trials and across time using repeated measures ANOVA. There was a significant increase in RPE over time ($p < 0.001$); RPE was significantly greater in the 90g trial than in the PC trial ($p < 0.05$), but there was no time x trial interaction ($p = 0.912$). There was a significant increase in HR over time ($p < 0.001$); HR was significantly higher in the 90g trial than in the PC trial ($p < 0.001$), but there was no time x trial interaction ($p = 0.262$). There was a significant increase in GI nausea over time ($p < 0.001$), but no significant difference between trials, nor was there a time x trial interaction; ($p = 0.080$) and ($p = 0.156$), respectively. There was a significant increase in GI stomach fullness over time ($p < 0.001$); this was significantly higher in the 90g trial than the PC trial ($p < 0.05$). There was also a time x trial interaction (<0.05). Post-hoc comparisons illustrated these differences between 30-120 minutes, and 30-150 minutes in both the 90g and PC trials, ($p < 0.001$). There was no increase in GI abdominal cramping observed over time ($p = 0.130$), nor a difference between trials ($p = 0.557$). there was no time x trial interaction ($p = 0.753$).

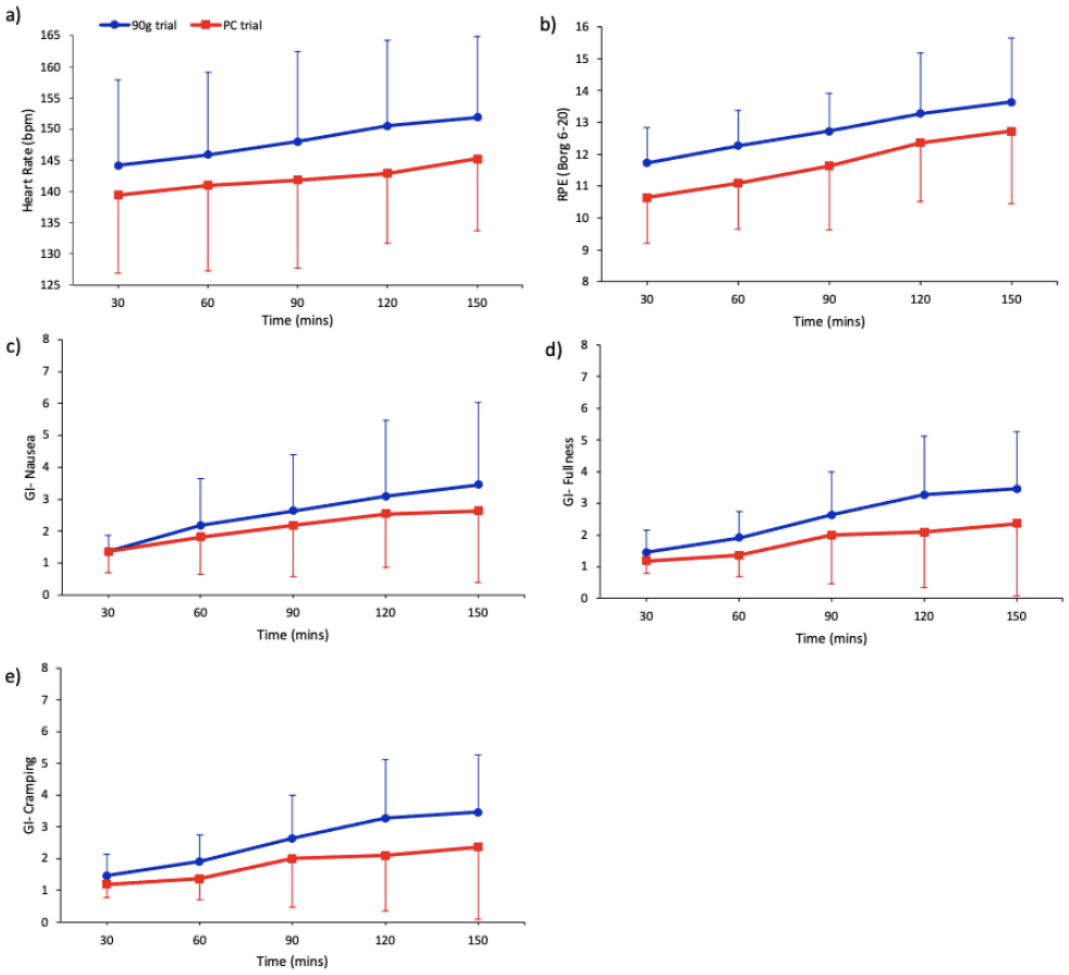

Figure 7. Heart rate (HR) ***a,*** *RPE b, GI- nausea* ***c****, GI- stomach fullness* ***d,*** *GI- abdominal cramping* ***e****, were measured every 30 min in 90g and PC conditions.*

*** Indicates a significant difference of p <0.001*

3.1.6. Plasma glucose and lactate

Plasma glucose

The average plasma glucose concentration for the 90g exercise trial across the 150-minute exercise duration was 5.74±0.50 (mmol/L). The average plasma glucose concentration for the PC exercise trial across the 2.5-hour exercise duration was 5.69±0.55 (mmol/L). A repeated measures ANOVA was performed to compare possible differences in plasma glucose between the PC and 90g trials and across time. There was a significant increase in plasma glucose over time ($p < 0.05$), but no difference was observed between trials ($p = 0.812$) and there was no time x trial interaction ($p = 0.546$).

Plasma Lactate

The average plasma lactate concentration for the 90g exercise trial across the 150-minute exercise duration was 2.04±0.73 (mmol/L). The average plasma lactate concentration for the PC exercise trial across the 150-minute exercise duration was 1.73±0.43 (mmol/L). A repeated measures ANOVA was performed to compare possible differences in plasma lactate between PC and 90g trials and across time. There was a significant increase in plasma lactate over time ($p < 0.001$), but no difference was observed between trials ($p = 0.360$) and there was no time x trial interaction ($p = 0.822$).

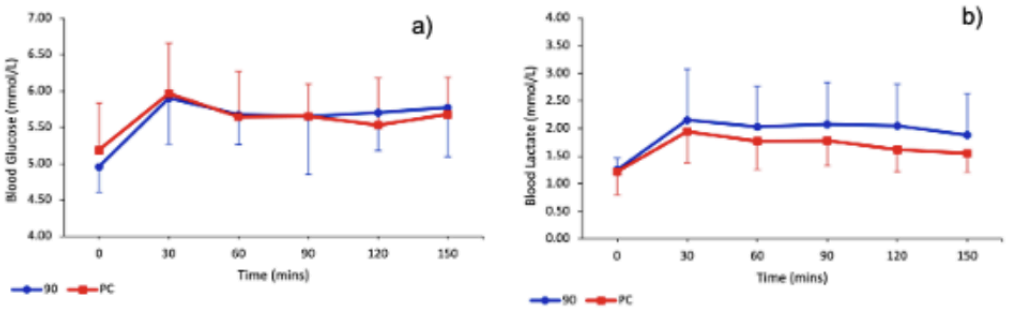

Figure 8. Changes in plasma glucose (a) and plasma lactate (b) concentrations (mmol/L) across the exercise duration in both trials.

3.2. Reliability

3.2.1. Physiological responses

Table 6 illustrates a direct comparison between the physiological characteristics of the participants (on average) between the 90g and 90gR trials. A t-test was used to identify significant differences between the 90g and 90gR trials. No significant differences were observed when comparing the 90g and 90gR trials. $\dot{V}O_2$, $\dot{V}CO_2$, RER, CHO and fat oxidation

were the same in both trials. $\dot{V}E$ was slightly but significantly higher in the 90gR trial than the 90g trial. CHO oxidation was also marginally greater in the 90gR trial.

Table 6. Participant physiological responses. The participant's physiological responses in the 90g and 90gR exercise trials; including $\dot{V}O2$, $\dot{V}CO2$, RER, $\dot{V}E$, carbohydrate, and fat oxidation rates. Data are presented as mean ±SD, averages across the trial durations. n = 8.

	Trial 90g	Trial 90gR
$\dot{V}O_2$ (L/min)	2.67±0.71	2.68±0.73
$\dot{V}CO_2$ (L/min)	2.46±0.62	2.49±0.66
RER	0.92±0.03	0.93±0.05
$\dot{V}E$ (L/min)	66±16	67±18
CHO oxidation (g/min)	2.44±0.52	2.53±0.77
Fat oxidation (g/min)	0.34±0.18	0.33±0.21

$\dot{V}O_2$, $\dot{V}CO_2$, $\dot{V}E$, and RER were all statistically analysed using repeated measures ANOVA. There was no change in $\dot{V}O_2$ over time ($p = 0.061$) nor was there a difference between exercise trials ($p = 0.790$) and there was no time x trial interaction observed ($p = 0.187$). There was no increase in $\dot{V}CO_2$ over time ($p = 0.207$), no difference between trials ($p = 0.566$) and no time x trial interaction observed ($p = 0.105$). There was a significant increase in $\dot{V}E$ over time ($p < 0.05$), but no difference between trials and there was no time x trial interaction observed; ($p = 0.342$) and ($p = 0.081$), respectively. There was no change in RER over time ($p = 0.679$), no difference observed between trials ($p = 0.272$) and no time x trial interaction ($p = 0.492$).

3.2.2. Stable isotope measurements

Resting breath ^{13}C enrichment was similar at the start of both the 90g and 90gR exercise trials, averaging – 26.27±1.23 and – 25.97±0.54 δ‰ vs PDB respectively. Expired breath ^{13}C is shown below in (Figure 9.d).

3.2.3. Substrate oxidation rates

The total, exogenous, and endogenous CHO oxidation rates were statistically analysed using repeated measures ANOVA. Total carbohydrate oxidation rates (Figure 9.a) did not change over time ($p = 0.813$) and there was no difference observed between trials ($p = 0.433$). There was no time x trial interaction ($p = 0.104$). Exogenous CHO oxidation rates (Figure 9.b) increased over time ($p < 0.001$) but there was no difference between trials observed ($p = 0.619$) nor a time x trial interaction ($p = 0.117$). Endogenous CHO oxidation rates (Figure 9.c) decreased over time ($p < 0.001$) but there was no difference between trials observed ($p = 0.479$) nor a time x trial interaction ($p = 1.799$). The Area Under the Curve (AUC) was calculated using the trapezoidal rule to quantify overall (mean average) total, exogenous and endogenous carbohydrate oxidation across the exercise trials. This was calculated from the period of 30-150 minutes. In the 90g trial, the total, exogenous and endogenous carbohydrate oxidation (in grams, g) was 291.6±0.5, 80.6±6.5 and 208.9±6.8 respectively. In the 90gR trial, the total, exogenous and endogenous carbohydrate oxidation was 305.5±2.5, 85±7.4 and 220.5±9.8 respectively. There was no significant difference in the AUC of total ($p = 0.09$), exogenous ($p = 0.09$) and endogenous ($p = 0.16$) CHO between the 90g and 90gR trials. Fat oxidation was statistically analysed using repeated measures ANOVA. There was no change in fat oxidation over time ($p = 0.525$), no difference observed between trials ($p = 0.494$) and no time x trial interaction ($p = 0.577$). The CV for the average total CHO oxidation rates across participants in the 90gR trial was 30±0.8%.

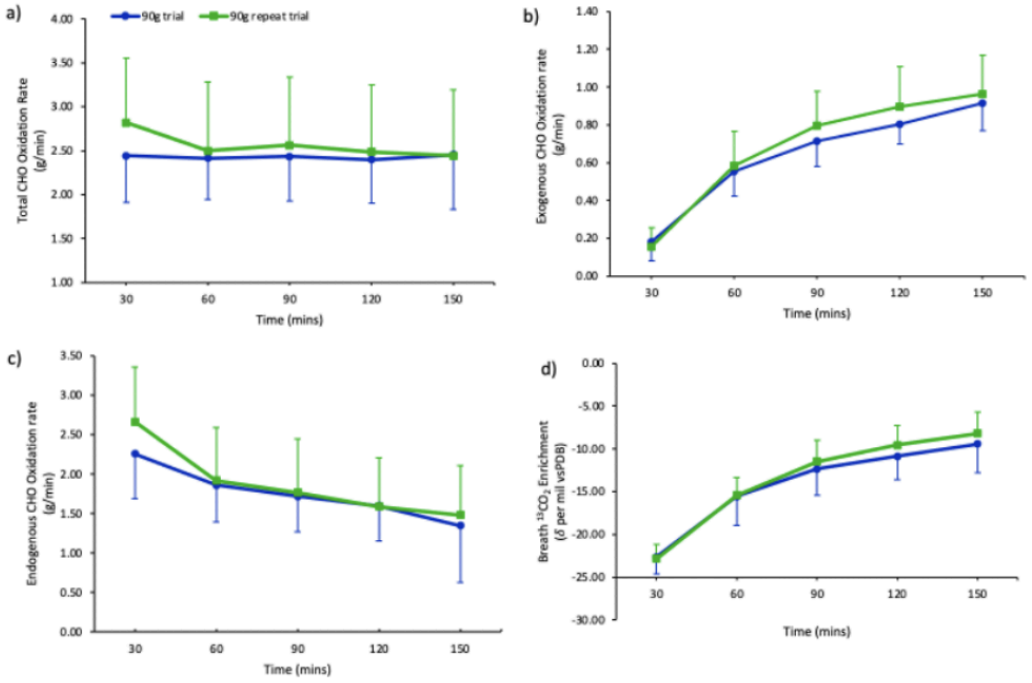

*Figure 9. Total CHO **a,** exogenous CHO **b,** endogenous CHO **c,** oxidation rates and breath enrichment **d,** were measured every 30 min in 90g and 90gR conditions.*

3.2.4 Peak exogenous CHO oxidation rates

There was no difference in peak exogenous CHO oxidation rates (Figure 10) (i.e., highest measured value at one-time point) in the 90g trial as compared to 90gR trial (0.89±0.19 vs 0.96±0.21 g/min for the 90g and 90gR trials, respectively; p =0.25). The CV of peak exogenous CHO oxidation rates in both 90g and 90gR trials was 22±0.5%. Figure 3.8 illustrates the individual peak exogenous CHO oxidation rates for reference. Figures 11 and 12 are both Bland Altman plots to present the peak exogenous CHO oxidation rates and average exogenous CHO oxidation rates (respectively) across all time points and present the data as a cluster surrounding the average difference.

There was no difference in peak exogenous CHO oxidation rates in the 90gR trial as compared to the PC trial (0.96±0.22 vs 0.90±0.21 g/min for the 90gR and PC trials, respectively; p =0.36). The CV of peak exogenous CHO oxidation rates in 90gR and PC trials was 21±0.5%.

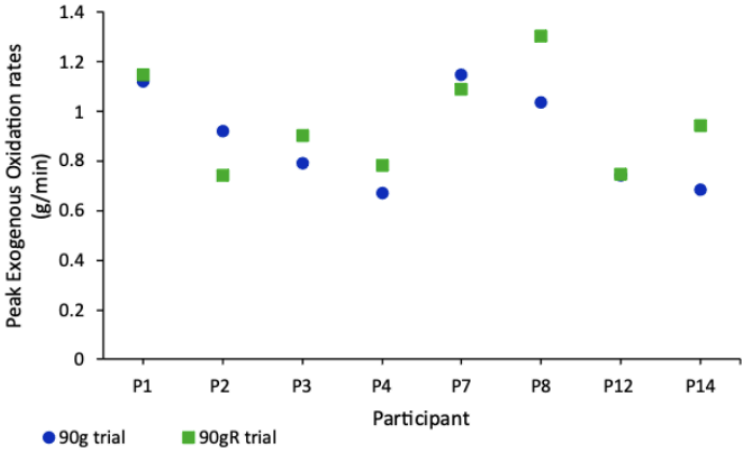

Figure 10. A graph to present the peak exogenous CHO oxidation rates (g/min) per participant in 90g and 90gR trials. N = 8.

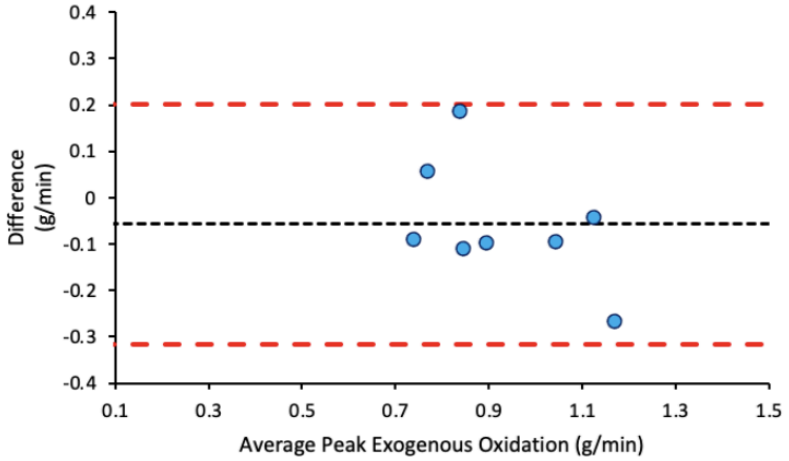

Figure 11. Bland-Altman Plot: Peak exogenous CHO oxidation rates (g/min) in 90g vs 90R trial. Legend: average difference = -0.057, upper limit of agreement = 0.222, lower limit of agreement = -0.336 (g/min). The average difference is illustrated by a solid line and 95% limits of agreements by dashed lines.

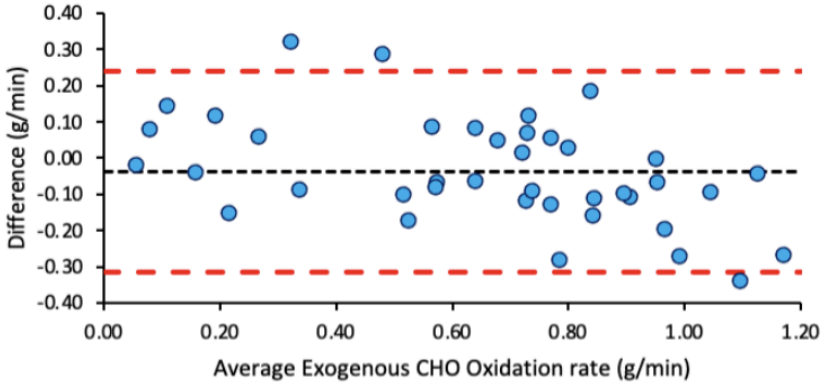

Figure 12. Bland-Altman Plot: Average exogenous CHO oxidation rates (g/min) in 90g vs 90gR trial in all participants across all time points. Legend: average difference = -0.04, upper limit of agreement = 0.24, lower limit of agreement = -0.31 (g/min). The average difference is illustrated by a solid line and 95% limits of agreements by dashed lines.

3.2.5. RPE, Heart Rate and GI Comfort

The average scores for RPE and ratings of GI comfort including nausea, stomach fullness and abdominal cramping are presented in Table 7 and compared with t-test values. No significant differences were observed between trials in any of the variables, but significances over time were present in RPE, GI nausea and GI stomach fullness.

Table 7. Participant GI, HR, RPE scores. Data are represented as mean ±SD, averages across the trial durations, using a 1-10 scale. n = 8.

	Trial 90g	Trial 90g Repeat
GI- Nausea	2.2±1.5	1.8±1.2
GI- Stomach Fullness	2.2±0.7	1.6±1.0
GI- Abdominal Cramping	1.1±0.1	1.2±0.3
Heart Rate (bpm)	150±10	150±5
RPE (Borg scale)	13±1	12±1

RPE, HR and GI (nausea, stomach fullness, and abdominal cramping) were analysed using repeated measures ANOVA. There was a significant increase in RPE over time ($p < 0.001$), no difference between trials observed ($p = 0.393$) and no time x trial interaction ($p = 0.491$). There was a significant increase in HR over time ($p < 0.001$), but no difference between trials ($p = 0.900$), and no time x trial interaction ($p = 0.628$). There was a significant increase in GI nausea over time ($p < 0.05$), but no difference was observed between trials ($p = 0.099$) and no time x trial interaction ($p = 0.922$). There was a significant increase in GI stomach fullness over time ($p < 0.001$), but no difference was observed between trials ($p = 0.189$) and no time x trial interaction ($p = 0.158$). There was no change in GI abdominal cramping over time ($p = 0.073$), no difference was observed between trials ($p = 0.265$) and no time x trial interaction ($p = 0.821$).

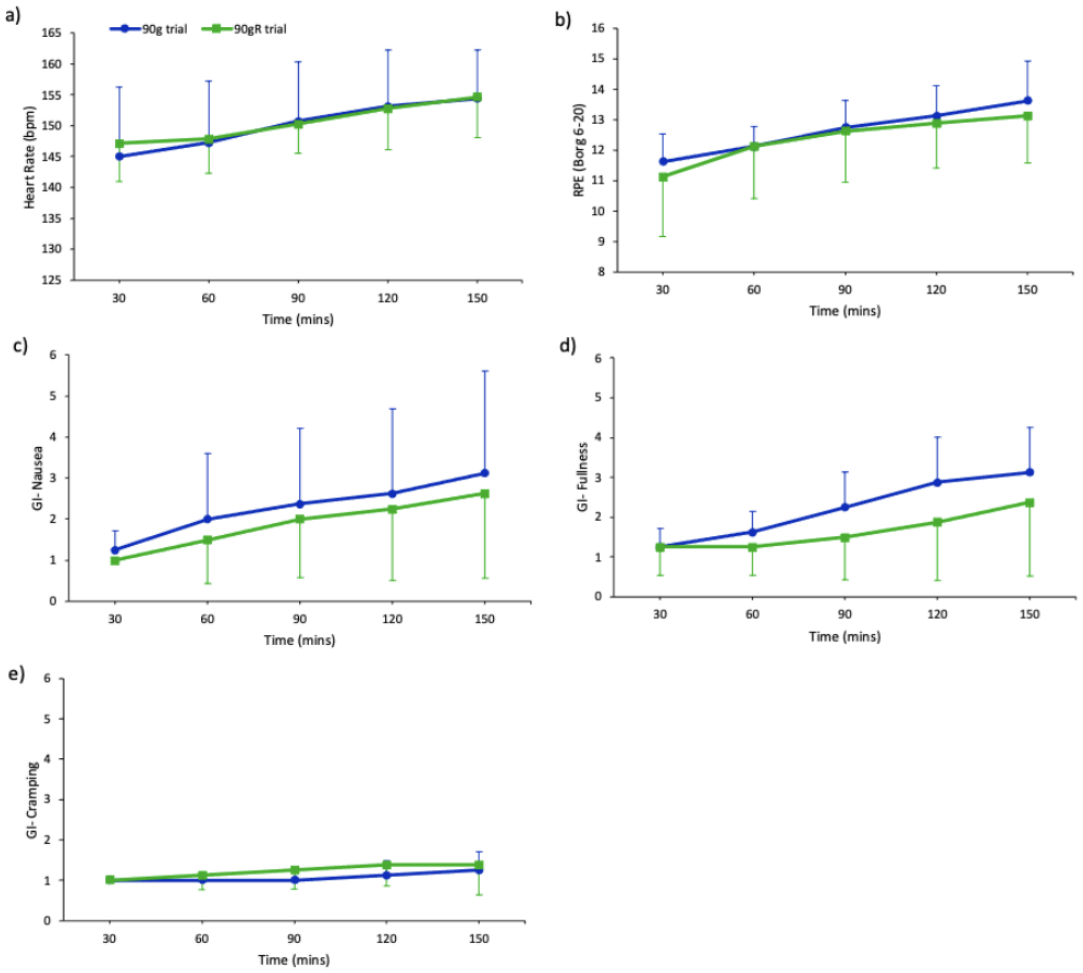

Figure 13. Heart rate (HR) ***a,*** *RPE* ***b,*** *GI- nausea* ***c,*** *GI- stomach fullness* ***d,*** *GI- abdominal cramping* ***e,*** *were measured every 30 min in 90g and PC conditions.*

3.3. Observed differences involving exogenous CHO oxidation rates and other variables between 90g, PC and 90gR

The Figures 14.a, 14.b, and 14.c below illustrate the relationships between body mass, height and workload with peak exogenous CHO oxidation rates. Pearson's correlation (Jamovi) and the correlation coefficient (Excel) were used to identify the R-value, R^2, the p-value and quantify the relationships between peak exogenous CHO oxidation rates and body mass, height, and workload of participants in 90g *vs* PC and in 90g *vs* 90gR. A correlational analysis was run on each trial separately.

In the group comparing 90g *vs* PC, there was no significant relationships in the 90g trial between body mass (R =0.465, R^2 =0.216, p =0.150), height (R =0.590, R^2 =0.348, p =0.056) or workload (R =0.602, R^2 =0.362, p =0.050) with peak exogenous CHO oxidation

rate. Similarly, no significant relationships were identified in the PC trial (body mass: R =0.570, R^2 =0.324, p =0.067; height: R =0.563, R^2 =0.317, p =0.071; workload: R =0.539, R^2 =0.291, p =0.087).

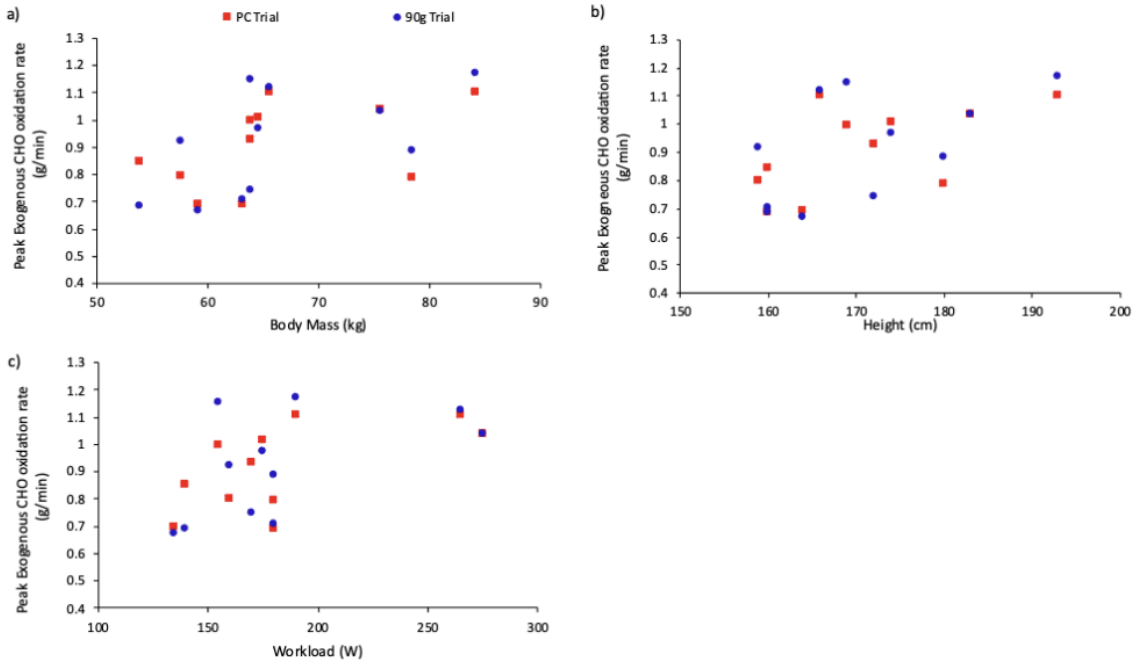

Figure 14. Relationship between peak exogenous CHO oxidation rate and body mass ***a,*** *height* ***b,*** *and workload* ***c,*** *in 90g and PC trials.*

In the group comparing 90g *vs* 90gR, displayed in Figures 15.a, 15.b, and 15.c, there was no significant relationships in the 90g trial between body mass (R =0.303, R^2 =0.0916, p =0.466), height (R =0.202, R^2 =0.0409, p =0.631) or workload (R =0.620, R^2 =0. 384, p =0.101) with peak exogenous CHO oxidation rate. No significant relationships were identified in the 90gR trial between body mass (R =0.477, R^2 =0.227, p =0.232) or height (R =0.486, R^2 =0.236, p =0.222) with peak exogenous CHO rate, but a significant relationship was observed between workload and peak exogenous CHO rate (R =0.767, R^2 =0.588, p =0.026).

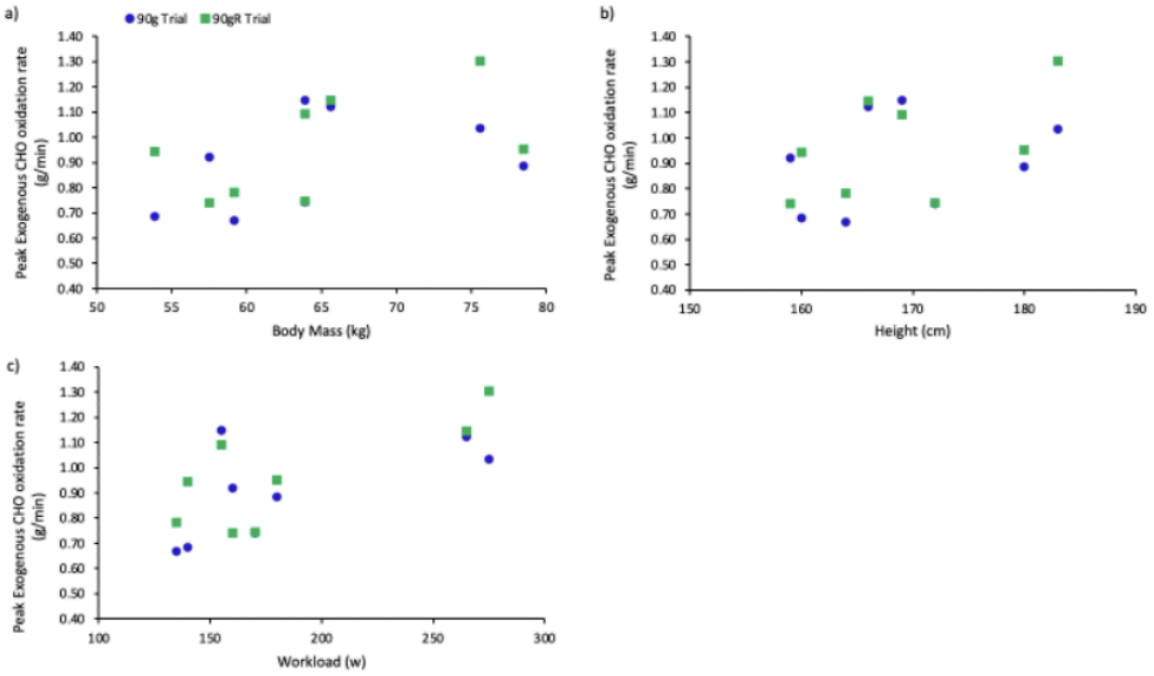

Figure 15. Relationship between peak exogenous CHO oxidation rate and body mass ***a,*** *height* ***b,*** *and workload* ***c,*** *in 90g and 90gR trials.*

Independent T-tests were used to establish significant differences between the three trials. In the 90g trial and the PC trial, there were significant differences between average peak exogenous CHO oxidation of the male (90g; 1.04±0.11, PC; 1.01±0.13) and female (90g; 0.81±0.19, PC; 0.83±0.12) participants ($p = 0.040$ and $p = 0.045$ respectively). There was no significant difference observed between the averages of the peak exogenous CHO oxidation rate of the male (90gR; 1.13±0.18) and female (90gR; 0.86±0.15) participants in the 90gR trial ($p = 0.060$).

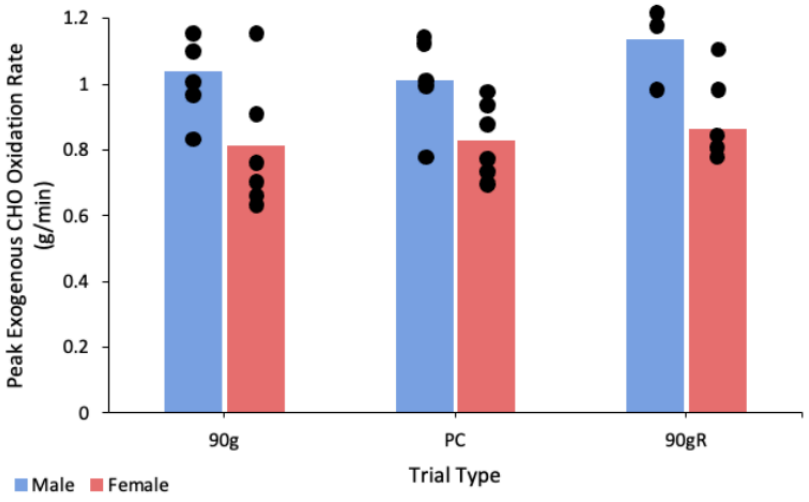

Figure 16. A sex comparison to illustrate differences in peak exogenous CHO oxidation rates (g/min) across all three exercise trials; 90g, PC and 90gR. Individual peak oxidation rates for all participants are illustrated by the black dots.

CHAPTER 4

4. Discussion

The primary aim of this study was to determine whether maximal exogenous CHO oxidation rates achieved through large glucose intakes during prolonged exercise could inform a personalised CHO dosing strategy. As hypothesised, the study results indicate that similar exogenous CHO oxidation rates were observed in the personalised trial as initially found in the high-dose (90g) trial. Moreover, personalised dosing appeared to reduce participants' GI discomfort, specifically stomach fullness. The secondary aim of this study was to assess the repeatability of exogenous CHO oxidation rate measurements during exercise, using a large CHO dose to evaluate the reliability of the technique. Importantly, it was found that exogenous CHO oxidation rates were remarkably similar between the two high-dose trials, suggesting that personalising CHO feeding during exercise based on estimated exogenous CHO oxidation rates could be implemented, but further consideration is necessary to assess the reliability of this. This discussion will explore these findings in more detail in the context of the existing literature, and identify strengths, limitations and areas for future research.

4.1. Personalised glucose feeding elicits comparable exogenous CHO oxidation rates to high-dose glucose feeding.

The findings of this study indicated no significant differences in peak exogenous CHO oxidation rates between high glucose feeding (90 g/h) and personalised carbohydrate (PC) feeding during exercise. Jeukendrup *et al.* (1997) reported similar peak exogenous CHO oxidation rates when comparing between trained and untrained participants (0.95 ±0.07 *vs* 0.96 ±0.03 g/min, respectively) using a glucose dose of 63 g/h. Studies (e.g. Smith *et al.,* 2010) suggest a dose response relationship between glucose intake and exogenous CHO oxidation rates when provided with glucose in doses of 15, 30, and 60 g/h, whereby exogenous glucose oxidation increased with ingestion rates; 0.17 ±0.04, 0.33 ±0.04, and 0.52 ±0.09 g/min, respectively. This is further supported by the findings of a 2004 review by Jeukendrup, which demonstrated that exogenous glucose oxidation rates do not significantly increase with increasing CHO intake beyond 60-90 g/h during prolonged exercise.

Following the initial 90g/h trial, peak exogenous glucose oxidation rates were divided by 0.8 to account for an 80% average oxidative efficiency (Jentjens *et al.,* 2003); subsequently, 8 out of 11 participants from the present study were fed with >60 g/h of glucose in the PC trial.

Oxidation efficiency is known to reduce as CHO ingestion increases. Wagenmakers *et al.* (1993) concluded after providing 0.6, 1.2, 1.8 and 2.4 g/min glucose solutions over 120 minutes of cycling, that despite ingestion rates increasing four-fold, average CHO oxidation rates plateaued at 1.07 g/min. The present study aimed to account for this by providing greater CHO than the observed peak exogenous oxidation in the 90 g/h trial (e.g. 1.5 g/min provided in the personalised trial, when peak exogenous CHO oxidation was 1.3 g/min during the 90g trial). Clearly ingestion isn't a measure of oxidation rate, but up until ~1 g/min, increases in oxidation are seen with increases in ingestion. Individuals in the present study oxidised up to 1.3 g/min, and so the previous evidence suggesting an exogenous glucose oxidation peak rate of ~1-1.1 g/min (Jeukendrup, 2004), may not be appropriate for every individual to optimise CHO oxidation and energy availability. Furthermore, the higher exercise intensity used in the present study relative to previous literature was also designed to maximise the use of exogenous CHO and may explain some elevated individual CHO oxidation rates. These findings, combined with the current research that illustrates increases in oxidation rates with the co-ingestion of multiple transportable CHOs, suggests that even higher rates of oxidation could be achieved by using multiple transportable CHOs, such as glucose *and* fructose (Jeukendrup & Jentjens, 2000).

However, the results of the present investigation also revealed when ingesting 1.5 g/min of glucose that 54% of participants oxidised <1-1.1 g/min, with the lowest oxidising 0.69 g/min. Individuals with lower CHO oxidation rates may, therefore, when ingesting peak rates observed in the literature, encounter increased undigested CHO in the gut and subsequently increase the prevalence of GI discomfort. Personalisation may therefore be necessary to avoid this undigested CHO in the gut and associated GI discomfort.

A dose of 90 g/h of glucose was used to determine peak exogenous CHO oxidation rates to subsequently personalise CHO intake in the PC trial. The lack of significant difference between the 90g and PC trials in this study suggests that for trained athletes (male and female $VO2_{max}$ ≥55 ml/kg/min and ≥50 ml/kg/min, respectively), consuming 90g of glucose per hour is unlikely to provide additional benefits in terms of exogenous CHO oxidation rates. This is where the recommendation of 90 g/h of CHO is focused on the co-ingestion of CHO. This finding has practical implications for endurance athletes in that when glucose feeding is based on oxidation rates, GI discomfort can be minimised compared to a 'more-is-better' approach. The relatively low coefficient of variation (CV = 19%±0.3) and limits of agreements between peak and average exogenous CHO oxidation rates across all timepoints illustrated in Figures 4, 5 and 6 indicate a consistent response among participants, strengthening the reliability of these findings.

4.2. Similar metabolic responses between conditions across participants.

When comparing the 90g and PC conditions, similar metabolic responses were observed. CHO provided most of the energy during the exercise trials, as determined through expired breath analysis. CHO oxidation accounted for most energy requirements during the exercise trials at ~85-87% of total substrate oxidation rates, while fat oxidation ranged from ~13-15%. This finding aligns with extensive literature indicating that CHO is the preferred fuel source during moderate-high-intensity exercise (Coyle, 1991; Jeukendrup, 2004). Maximising exercise intensity was essential in this study to create a high demand for CHO and to promote maximal exogenous CHO oxidation rates. No significant differences were observed in the mean CHO and fat oxidation rates between the 90g and PC trials, or between the 90g and 90gR trials.

Despite different provisions of CHO during the two contrasting dosed trials (90g and PC), they yielded a similar proportion of energy from both endogenous and exogenous CHO sources, as well as fats. There were no significant differences in total, endogenous or exogenous oxidation rates between the 90g and PC trials, despite the provision of different CHO doses. However, a significant interaction effect was observed over exercise time, supporting the current literature indicating that exogenous CHO oxidation rates increase with prolonged exercise to support the energy demands and facilitate muscle contractile activity, and this is consistent with an increased reliance in blood borne CHO sources in the face of declining muscle glycogen (Hawley & Leckey, 2015; Jeukendrup, 2010).

4.3. Variation in physiological and psychological responses between trials.

In the PC trial, the average CHO intake was 68 ±12 g/h, totaling 170 ±30 grams of CHO over the 2.5 hours of exercise; with the highest individual dose provided at 83 g/h and the lowest at 52 g/h. This represents a 24% decrease on average in CHO consumption compared to the 90g trial, translating to an average reduction per participant of ~22 g/h. Notably, in the PC trial, two participants consumed ~48 g/h less than in the initial 90g trial. Conversely, two other participants had CHO doses close to the 90g trial with only a ~7 g/h reduction; all of which was calculated based on their individual exogenous CHO oxidation rates. Despite these variations in CHO intake, the metabolic responses between the 90g and PC trials were on average, largely similar, even for participants with significantly reduced CHO doses, and no effect on total nor endogenous CHO oxidation was observed. It should be noted that, although in some cases CHO intake was significantly lower in the PC trial, current recommendations for CHO intake are based on the co-ingestion of glucose and fructose at 90 g/h, not glucose alone. Based on this, when comparing the PC dose to the current

glucose only recommendations of 60 g/h reported in literature (Jeukendrup, 2014), eight out of eleven participants were provided with CHO doses beyond this recommendation, with the highest ~38% greater than the suggested 60 g/h. Only two out of the eleven participants were provided with intakes less than this, and one was equal to the 60 g/h recommendation. Using the present studies results, this suggests that only 9% of the participants fell in line with the current recommendations for CHO intake; a considerable difference.

Other dose-response studies have illustrated that overfeeding with glucose has been shown to increase the reliance on CHO and contribute to an increased use of CHO in doses of 60 g/h *vs* 30 g/h (Wallis et al., 2007) but further increases (90 g/h) showed no significant difference. This was like the present study, where the higher CHO doses did not further increase exogenous CHO oxidation rates beyond those seen when ingesting ~90 g/h.

In the present study, it was observed that HR, RPE and GI (fullness) were all significantly elevated in the 90g trial compared with the PC trial. However, due to the lack of time-matched familiarisation compared to the experimental trial, it was deemed more appropriate to compare differences between PC and 90gR, with the initial 90g trial acting as a true familiarisation. Whilst RPE was similar between PC and 90gR, HR and GI (fullness) were significantly reduced in the PC trial, compared to both 90g and 90gR. This suggests the personalised CHO dose may cause reduced cardiovascular strain and increased GI comfort. Previous studies have shown that high CHO intakes can lead to GI distress, which negatively impacts performance (Jeukendrup, 2010; Costa *et al*., 2017). In the present study, all participants received lower CHO doses in the PC trial than in the 90gR trial, potentially minimising the volume of undigested CHO in the gut and associated discomfort. Fluid intake was consistent across all trials, totaling 2.5 litres in each trial. Despite the uniform fluid volume, the increase in GI comfort in the PC trial indicates that prescribing CHO doses based on individual oxidation rates may help alleviate GI distress and sustain fuelling strategies, and thus performance. This is supported by previous research and reviews (Stellingwerff & Cox, 2014) suggesting that individualising nutrition strategies can optimise performance and limit adverse symptoms.

With regard to overall comfort and stress, Wallis *et al.* (2007) has previously illustrated the link with an increased dose of CHO and subsequently higher HR and prevalence of GI discomfort, than with a lower dose. In the 2007 study, it was reported that participants experienced less GI discomfort and associated complaints during a two hour exercise period where ingestion of CHO was lower. Speculation was made to imply that the excess CHO in the higher dose trials remained undigested, accumulating in the gut. Additionally,

Jeukendrup (2007) reported that higher CHO intakes during prolonged exercise led to an elevated perception of effort, potentially due to the metabolic demands of digesting and oxidising large amounts of CHO. Similarly, the finding in this study of significantly higher HR in the 90g trial aligns with the work of Achten *et al*. (2003), who observed increased HR in athletes consuming high (1.25 g/min) CHO doses prior to cycling-based endurance exercise. This could be attributed to the increased metabolic rate and cardiovascular demands associated with higher CHO oxidation.

As discussed, this study provides further insights into GI symptoms (specifically: nausea, cramping, and fullness) during exercise with CHO supplementation that both align and deviate from previous research. The significant increase in GI, specifically nausea over time, with no significant differences between trials, supports findings by Jeukendrup and Jentjens (2000), who noted that prolonged exercise, regardless of CHO intake, can exacerbate GI discomfort due to reduced blood flow to the gut. The lack of a significant difference in nausea between trials further suggests that factors other than CHO intake might play a more critical role in nausea development during exercise. The significant increase in stomach fullness over time, and greater prevalence in the 90g trial compared to the PC trial, vallidates findings by Rehrer *et al.* (1992), who observed increased feelings of fullness and bloating with higher CHO ingestion. This is likely due to slower gastric emptying and increased osmolality from higher CHO concentrations in the stomach. The absence of significant changes in abdominal cramping over time and between trials, however, contrasts with some reports in the literature. For example, during a triathlon race, Pfeiffer *et al*. (2012) documented a higher incidence of abdominal cramping with increased CHO intake, particularly when the intake exceeded the gut's absorptive capacity. However, the environmental conditions, and higher relative intensity of both running and swimming when compared with cycling may have contributed to these results observed. The lack of significant differences in our study might be due to individual variability in GI tolerance, the type of CHO used, or the exercise protocol.

4.4. Plasma glucose and lactate responses.

Individuals participating in endurance exercise will require frequent CHO ingestion to maintain plasma glucose concentrations, CHO oxidation rates and subsequently sustain performance. The results of the present study illustrate the provision of glucose at either 90 g/h or a personalised dose (68 ±12 g/h) maintained participants' plasma glucose levels across both the 90g and PC exercise trials (5.75 mmol/L and 5.69 mmol/L, respectively). It is possible that the absorption of glucose from the gut is a limiting factor for oxidation rates, with lowering feeding rates not consequently lowering plasma glucose levels, but without

supporting data assessing insulin responses and tracer kinetics, this is only speculative. Although plasma glucose increased slightly over time, all participants maintained similar concentrations in both trials despite changes in absolute CHO dose.

In both the 90g and PC trials, a similar trend in concentrations was observed with glucose across the exercise duration. The significant increase in plasma glucose over time and lack of significant difference between trials algins with existing literature that suggests CHO ingestion during exercise helps maintain stable plasma glucose levels, which is crucial for sustaining performance and delaying fatigue (Coyle et al., 1986; Coggan & Coyle 1991).

The exercise intensity of the trial was set to be in the moderate exercise intensity domain, where generally no rise in plasma lactate is expected. However, it was observed that plasma lactate increased over time, with no differences between the 90g and PC trials, likely due to CHO provision. This is consistent with the understanding that while CHO can influence lactate production, the differences may not always be pronounced between dosing strategies if overall CHO availability is sufficient (Hawley & Leckey, 2015). Consequently, as CHO absorption rates appear to be the limiting factor, with the same quantity of CHO being absorbed and delivered to the muscle, lactate production is also similar between 90g and PC trials. The similarity in lactate responses and absorption rates between the trials suggests that despite differences in CHO doses, both trials supported the metabolic demands of the exercise sufficiently.

4.5. The oxidation rates achieved with a personalised dose of CHO appear consistent when compared to a high CHO dose and have no significant effect on total CHO or fat oxidation.

Peak and average exogenous CHO oxidation rates exhibited considerable inter-individual variation (Figures 5 and 6), underscoring the necessity for personalised CHO dosing strategies during endurance exercise. Figures 5, 6, 11 and 12 reveal broader limits of agreement for the 90g *vs* 90gR trials compared to the 90g *vs* PC trials, suggesting a greater variability when replicating the same CHO dose. A conservative CHO oxidation efficiency factor of 80% was chosen, based on previous studies reporting oxidation efficiency rates ranging from ~75-95% (Hawley & Leckey, 2015; Jeukendrup & Jentjens, 2000). This allows potential estimation errors to be accounted for, while still allowing for consistent exogenous CHO oxidation rates across the three trials. Whilst the present study shows some variation in exogenous CHO oxidation rates, the individual rates observed across the three trials were similar (±0.2 g/min 95% of the time), so the *personalised* prescription of CHO dosing for a fixed duration of cycling at the same relative intensity is reliable.

4.6. Reliability

When assessing the reliability of personalised CHO recommendations, all physiological responses and substrate oxidation rates were unchanged between 90g and 90gR trials. This supports the use of one-time measurements of oxidation rates for the purpose of personalisation. HR, RPE and GI discomfort were also unchanged between the 90g and 90gR trials, further supporting this.

When assessing the reliability of individualised glucose feeding, no significant differences were observed in mean peak exogenous glucose oxidation rates between 90g and 90gR trials, suggesting that, on average, exogenous glucose oxidation rates also do not change over time. Despite this, individual data found variances of 14 g/h and -20 g/h between the two trials. A possible explanation for these variates was the lack of strict dietary controls in the present study. Margolis *et al.* (2019) observed no differences in exercising exogenous CHO oxidation between groups fed with low (<1.5 g/kg) or adequate (6 g/kg) doses of CHO in the 24h preceding the trials, suggesting no impact of diet on exogenous CHO oxidation. However, a more recent review by the same authors (Small & Margolis, 2022), found that differences in dietary CHO intake *did* significantly impact rates of exogenous CHO oxidation on average. Additionally, Jeukendrup *et al.* (1996) investigated the effect of low pre-exercise endogenous CHO availability on CHO oxidation rates and observed that exogenous CHO oxidation rates were 28% lower in the low-glycogen trial compared with higher pre-exercise CHO availability or ‘CHO loading’. This highlights the requirement for stricter dietary controls to enhance the reliability of metabolic measurements in exercise-related studies (Jeukendrup & Wallis, 2005). As a result, the ‘personalised’ dose used within the present study should be considered as an estimation and range to account for possible daily variations in dietary intake.

Factors that affect exogenous CHO oxidation rates.

4.6.1. Sex and CHO oxidation rates.

There was a significant difference in peak exogenous CHO oxidation rates observed between the 90g and PC trials in both male and female participants, suggesting that males can oxidise a greater absolute dose of CHO. This was not uniform in the study, as differences between the 90g and 90gR were directionally consistent but not significant, and so the trend remains unclear.

Previously literature has thoroughly investigated differences in substrate oxidation rates in males *vs* females; Horton *et al*. (1998) found that females had a lower CHO oxidation rate

compared to males, attributed to an increased reliance on fat as a fuel source during moderate-intensity exercise. This was further supported a few years later with findings that males have higher total CHO oxidation rates during exercise because of a greater volume of muscle mass and storage capacity (Mittendorfer *et al.*, 2002). In most studies that have investigated substrate utilisation in males and females, exercise intensity is often matched relative to VO_2 (max or peak). It's been observed that at the same percentage of VO_2 max, females oxidise less CHO and more fat than males (Horton *et al.,* 1998), likely a result of the lower absolute workload despite the same relative intensity.

Fewer studies however, have illustrated differences in exogenous CHO oxidation rates in male and females, with clear comparisons. Both M'Kaouar *et al.* (2004) and Riddell *et al.* (2003) showed that the contribution of CHO oxidation to the energy yield from exogenous glucose oxidation during cycling exercise was consistently higher in females than in males. Despite this finding, none of the studies had sufficient statistical power to conclude this difference, and so, at present, there is little evidence to suggest significant differences between males and females regarding exogenous glucose oxidation rates. Wallis *et al.* (2006) established that when expressed in absolute terms, similar rates of exogenous CHO oxidation in males and females were observed with CHO ingestion during exercise. It is possible that the *slightly* higher exogenous glucose oxidation rates observed in *some* studies could be a result of the higher dose of glucose ingested when expressed in milligrams or grams per kilograms of FFM per minute; showing relevance to the below section on body mass and differences in peak exogenous CHO oxidation rates.

As a result of the previous literature mentioned above, and the limited statistical power due to a small sample size in the present study, it would be appropriate to suggest further research is required to confirm the differences in exogenous CHO oxidation rates between sexes. When a dose based on individual oxidation rates cannot be derived, it seems appropriate to recommend both males and females to ingest similar glucose doses, or in line with a dose relative to their body mass.

4.6.2. Body Mass, Workload, Height and Exogenous CHO Oxidation rates.

It has previously been suggested that the capacity for intestinal CHO absorption affects exogenous CHO oxidation rates and that, larger body size or height and therefore a larger intestinal surface area may affect this (Fuchs, Gonzalenze, Van Loon, 2019). Jeukendrup & Jentjens (2000) and Jeukendrup *et al*. (2010) both previously found no relationship between body mass and CHO oxidation rates, and further stated that body mass should not be used as a significant predictor of exogenous CHO oxidation rates. The present study also

established no significant relationship between body mass and peak *exogenous* CHO oxidation rates across the participants. It is apparent from Jeukendrup & Wallis's (2005) investigations that absolute CHO oxidation rates may be elevated in larger individuals, but that *relative* CHO oxidation rates may not be significantly different from that of smaller individuals, and the present study's results support this.

It is well-documented that with increased exercise intensity and thus workload, CHO oxidation rates increase because of greater energy demands and shifts towards CHO utilisation. Despite a higher workload requiring more muscle mass recruitment, and therefore a higher metabolic rate, in the present study there were no such significant correlations between workload and peak exogenous CHO oxidation rates in either the 90g or PC trials. In the 90g trial, trends were identified that suggest a possible relationship, but statistical significance was borderline (p = 0.05). This, combined with the significant relationship between workload and peak exogenous CHO oxidation rate identified in the 90gR trial, indicates a potential overall trend, which needs further investigation to establish the importance of workload in exogenous CHO oxidation rates. Two studies have previously shown *significant* effects of workload and CHO oxidation rates, whereby greater rates are observed when energy expenditure is increased at higher exercise intensities (Achten & Jeukendrup, 2004), (Ijaz *et al.,* 2024). Ijaz and colleagues (2024) investigated differences in exogenous glucose oxidation rates during cycling exercise in two groups; large (>70kg) and small (<70kg) individuals. When larger individuals exercised at the same *relative* exercise intensity to small individuals (e.g. 95% lactate threshold), it was apparent that body size was positively correlated with peak exogenous glucose oxidation rates, with larger individuals oxidising great amounts of exogenous glucose. This is likely explained by the relationship with total workload, whereby increases in workload also increase metabolic demand and consequently an increased requirement to oxidise exogenous glucose. The present study only investigated 4 individuals >70kg, therefore it's possible with a greater proportion of larger individuals, our results may have observed similar findings.

The relationship between participant height and CHO oxidation rates during exercise has not been extensively studied. There is strong anecdotal evidence to suggest that with increasing height, both body and muscle mass are greater and subsequently, absolute CHO oxidation rates would increase due to elevated metabolic rate, energy demands and substrate utilisation during exercise. With elevated metabolic rates, it's likely that GI comfort from increased CHO tolerance would also be elevated in taller individuals. There was, however, no significant relationship observed in the present study between height and peak

exogenous CHO oxidation, but it is likely that it is more directionally constant and consistent and needs further investigation.

4.7. Limitations

4.7.1. Poor dietary control

In the present study, participants were requested to record their food intake on the day prior to their first experimental trial and replicate this intake for the subsequent two trials. However, because the food consumed was neither supplied nor monitored by the researchers, precise dietary control was challenging. Research indicates that the macronutrient composition of an athlete's diet can significantly affect substrate utilisation, with high-CHO diets being shown to enhance CHO oxidation during exercise (Jeukendrup, 2010; Burke *et al*., 2011), however this investigation needs more work. The timing of CHO intake before exercise sessions may also influence exogenous CHO oxidation rates by increasing and sparing muscle glucose stores (Hawley *et al.,* 1997) and maintaining blood glucose concentration. This can, in turn, support sustained high-intensity performance and improve endurance whilst enhancing oxidation rates (Cermak & van Loon, 2013; Jeukendrup, 2014). Studies have demonstrated that even small differences in pre-exercise nutrition can result in significant variations in metabolic responses during exercise (Tarnopolsky *et al.,* 2001). Unfortunately, despite attempting to standardise dietary intake, the results may be limited due to poor dietary control within this study, which could have introduced variability in exogenous CHO oxidation rates across participants. This approach is supported by previous literature further advocating for stricter dietary controls to enhance the reliability of metabolic measurements in exercise-related studies (Jeukendrup & Wallis, 2005). As a result, the ‘personalised’ dose used within this study should be considered as an estimation and range to account for possible daily variations in dietary intake.

4.7.2. Lack of ^{13}C control prior to trials

In the present study, participants were not asked to avoid foods with a high natural abundance of ^{13}C. Despite seeing little differences between resting ^{13}C breath enrichment at the start of exercise trials, we were unable to control or minimise the likely background shift from glycogen stores in the days preceding the experimental trials. However, it is likely this is more of an issue when using naturally high abundance ^{13}C carbohydrates to determine exogenous CHO oxidation. When the ingested CHO is enriched with $U^{13}C$-glucose as in the present study, the change in breath ^{13}C is large and can minimise these background issues.

4.7.3. Menstrual Cycle Control

The menstrual cycle of the six female participants was recorded but not controlled when scheduling metabolic trials. Given the well-documented effects of the menstrual cycle on substrate metabolism (e.g. Oosthuyse and Bosch, 2010), there is a possibility this could have influenced the results.

4.7.4. Order effects

All participants were required to begin the testing protocol with the same 90g trial to calculate CHO doses for subsequent exercise trials, hence the part-randomisation of the study. To limit the risk of an order effect, it would have been beneficial to familiarise participants with the testing protocol more than once. Although the second aim of the study was to assess the repeatability and/ or reliability of the methods used to investigate exogenous CHO oxidation rates, the limited number of participants (n =8) as a result of difficulties with scheduling and availability also hinders this, and findings should be further investigated to solidify the evidence that this protocol is sufficient.

4.7.5. Future work

The present study focused solely on glucose to assess the feasibility of personalising CHO intake. Given the positive results, future research should expand on this to explore the potential benefits of personalising CHO intake using multiple transportable CHO to investigate any additional performance enhancements (Currell & Jeukendrup, 2008; O'Brien & Rowlands, 2011). In the present study, the personalised dose was compared to an impractical dose of glucose, as this was required to encourage maximal oxidation rates. In future it might be useful to also compare the personalised dose to the 'recommendations' but in particular, with respect to gut comfort and performance to identify if personalising has a *functional* benefit. Addressing the limitations would enhance the reliability of these findings and provide a stronger foundation for future research. Improvements to the methods used by increasing the research confidence in prior dietary intake and control of the menstrual cycle, whilst increasing participant numbers, both the personalisation and reliability aspects of the study would show stronger cases. Additionally, for practical relevance, it would be beneficial to investigate this with the addition of fructose to see if the observed trends are evident and therefore more applicable with current CHO recommendations during exercise. Although strategies involving the co-ingestion of multiple CHO have been associated with performance benefits, glucose remains a critical CHO for evaluating oxidation rates during exercise. This study focused solely on glucose, given its fundamental role in evaluating oxidation rates during exercise. Personalising CHO intake is essential given the variability in

factors affecting exogenous CHO oxidation rates. With limited studies addressing the issue, further research into CHO personalisation is warranted.

5. Conclusion

In summary, the present study has provided critical insights into establishing the possibility of providing nutritional personalisation for endurance athletes and has established several key findings. Firstly, as hypothesised, it may be possible to personalise ingestion rates to ensure peak oxidation is achieved by using a high-glucose feeding trial. This apparently induced largely similar metabolic effects but some evidence of improved gut comfort, likely a result of lower CHO intake. Secondly, whilst further work with tighter controls is warranted, the determination of exogenous CHO oxidation rates is somewhat reliable. Further work could now build on these observations in pursuit of individualisation of CHO feeding strategies to maximise absorption, minimise GI distress and optimise performance.

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