

Electronic Thesis and Dissertation Repository

---

8-23-2016 12:00 AM

## Effect of Modified Corn Starch Ingestion on Soccer Skills and Physical/Cognitive Performance During a Simulated Soccer Game

Manuel D. Quinones  
*The University of Western Ontario*

Supervisor  
Dr. Peter Lemon  
*The University of Western Ontario*

Graduate Program in Kinesiology  
A thesis submitted in partial fulfillment of the requirements for the degree in Master of Science  
© Manuel D. Quinones 2016

Follow this and additional works at: <https://ir.lib.uwo.ca/etd>



Part of the [Sports Sciences Commons](#)

---

### Recommended Citation

Quinones, Manuel D., "Effect of Modified Corn Starch Ingestion on Soccer Skills and Physical/Cognitive Performance During a Simulated Soccer Game" (2016). *Electronic Thesis and Dissertation Repository*. 4056.

<https://ir.lib.uwo.ca/etd/4056>

This Dissertation/Thesis is brought to you for free and open access by Scholarship@Western. It has been accepted for inclusion in Electronic Thesis and Dissertation Repository by an authorized administrator of Scholarship@Western. For more information, please contact [wlsadmin@uwo.ca](mailto:wlsadmin@uwo.ca).

## Abstract

Generation UCAN® (UCAN), which contains heat treated non Genetically Modified Organism (GMO) corn starch, has the potential to enhance endurance exercise performance via altering substrate use; however, whether its effects are similar during high intensity, intermittent exercise, like that performed in soccer is unknown. Therefore, the purpose of this study was to assess the effects of UCAN on physical, skill and cognitive performance during and following a simulated soccer game. Eleven trained male soccer players completed a 60-min soccer simulated game on a treadmill, under two experimental conditions: UCAN and an isoenergetic carbohydrate (CHO) placebo. There were no significant treatment differences in physical performance; however, UCAN improved skills and some measures of cognitive performance ( $p < 0.05$ ) suggesting that UCAN supplementation may attenuate the decline in cognitive and skill performance often seen toward the end of games. More study of UCAN is warranted, especially on other aspects of cognitive and physical performance.

## Keywords

Low Glycemic Starch, Dietary Supplements, High Intensity Intermittent Exercise, Muscular Fatigue, Mental Fatigue, Motor Skills, Fat oxidation

## Acknowledgments

"No one can whistle a symphony. It takes an orchestra to play it." H.E. Luccock.

Thus, I would like to take this opportunity to thank very important and influential people in my life.

Foremost, I would like to thank my supervisor, Dr. Peter Lemon for giving me the opportunity to pursue my academic dreams. Thank you for giving me a chance to explore my own research interests. These past two years here at Western have been a great learning experience, and have also make me not only a better person but also more professional. Thanks for your guidance and support.

To all the soccer players that participated in this study. Thanks for your hard work and commitment to the study without expecting anything in return. This attitude only shows the true spirit of an athlete. To my lab mates Arash Bandegan and Victor Nguyen. I really appreciate your help during the data collection process.

I would also like to thank my family. To my mother, thanks for your understanding and constant encouragement to help me be the best I can be. To my brother Mauricio, words cannot express the admiration I have for your tenacity and positivism in life. You have always believed in me no matter the circumstances and that only motivates me to keep pursuing my dreams. Thank you brother!

To my beautiful wife, thanks for your endless love and support during this endeavor. Thank you for being so kind, loving and supportive and for all the sacrifices you have made to help me achieve my goals. I would like to dedicate this thesis to my children Sara and Camila. You girls are the reason I wake up every day with the motivation to keep pursuing my dreams.

Thank you all very much for your contributions, without your knowledge, guidance and support this would not have been possible.

# Table of Contents

Abstract.....	i
Acknowledgments.....	ii
Table of Contents.....	iii
List of Tables.....	vi
List of Figures.....	vii
List of Appendices.....	viii
List of Abbreviations.....	ix
1 Introduction.....	1
2 Literature Review.....	4
2.1 Carbohydrate (CHO) Metabolism.....	4
2.2 Fatigue and Exercise.....	6
2.2.1 Central Fatigue Hypothesis.....	7
2.2.2 CHO Supplementation and Central Fatigue.....	8
2.3 CHO Supplementation and Hypoglycemia.....	9
2.4 CHO Supplementation and Intermittent Exercise Performance.....	10
2.5 CHO Supplementation and Cognitive/Skill Performance.....	14
2.5.1 Sport-Specific Skills Performance.....	16
2.6 Generation UCAN <sup>®</sup> (UCAN).....	17
3 Methods.....	19
3.1 Participants.....	19
3.2 Preliminary Visits and baseline testing.....	19
3.3 Experimental overview.....	20
3.4 Supplementation Treatments.....	24
3.5 60-minute Simulated Soccer Game Protocol.....	24

3.6	Measurements .....	27
3.6.1	Body Composition .....	27
3.6.2	Aerobic Capacity .....	27
3.6.3	Sprint test .....	28
3.6.4	Heart Rate .....	28
3.6.5	Blood Glucose and Lactate .....	28
3.6.6	Fluid Ingestion .....	29
3.6.7	Cognitive Function.....	29
3.6.8	Rate of Perceived Exertion (RPE) .....	32
3.6.9	Repeated Sprint Ability test (RSA).....	32
3.6.10	Skill tests.....	33
3.7	Statistical Analysis.....	38
4	Results.....	39
4.1	Descriptive Analysis .....	39
4.2	Blood Metabolites.....	39
4.2.1	Blood Glucose.....	39
4.2.2	Blood Lactate .....	40
4.3	Respiratory Exchange Ratio (RER).....	41
4.4	Ratings of Perceived Exertion .....	42
4.5	Measures of Exercise performance.....	43
4.5.1	Slalom Dribble Test .....	43
4.5.2	Loughborough Soccer Passing Test.....	44
4.5.3	Repeated Sprint Ability Test (RSA).....	46
4.6	Measures of cognition.....	46
4.6.1	Eriksen-Flanker Task – Mean Reaction Time .....	46
4.6.2	Eriksen-Flanker Task – Response Accuracy .....	47

4.6.3	Eriksen-Flanker Task – Conflict Cost.....	47
4.6.4	Serial Seven Subtraction Test.....	48
5	Discussion .....	50
5.1	Effects on Cognition .....	53
5.2	Effect on Soccer Skills.....	56
5.3	Effects on repeated sprint performance (RSA).....	58
5.4	Limitations .....	59
5.5	Future Directions .....	60
5.6	Summary/Conclusion.....	61
	References.....	62
	Appendices.....	72
	Curriculum Vitae .....	85

## List of Tables

Table 1. Participant Characteristics .....	39
Table 2. Loughborough Soccer Passing Test.....	45
Table 3. Repeated Sprint Ability Test.....	46
Table 4. Measures of cognition.....	49

## List of Figures

Figure 1. Experimental protocol .....	23
Figure 2. Exercise Intensity Profile of each Running Block. ....	26
Figure 3. Set up for the Slalom Dribble Test .....	34
Figure 4. Diagram of the Slalom Dribble Test .....	35
Figure 5. Picture of the set up for the LSPT .....	37
Figure 6. Diagram of the LSPT.....	38
Figure 7. Blood Glucose Concentrations Before and During the Simulated Soccer Game. .	40
Figure 8. Blood Lactate Concentrations Before and During the simulated soccer game. ....	41
Figure 9. Respiratory Exchange Ratio (RER).....	42
Figure 10. Ratings of Perceived Exertion (RPE).....	43
Figure 11. Dribbling Time Before and After the Simulated Soccer Game. ....	44
Figure 12. Total Performance Time for the Loughborough Soccer Passing Test (LSPT) after the Simulated Soccer Game.....	45
Figure 13. Eriksen-Flanker Conflict Cost.....	48



## List of Appendices

Appendix A. Human Ethics Approval .....	72
Appendix B. Letter of Information and Informed Consent .....	73
Appendix C. Participant Health Information Form .....	81
Appendix D. Physical Activity Readiness Questionnaire.....	82
Appendix E. Ratings of Perceived Exertion (RPE) Scale.....	83
Appendix F. Loughborough Soccer Passing Test (LSPT) Trial Order Sheet.....	84

## List of Abbreviations

<b>ACSM</b>	American College of Sports Medicine
<b>BBB</b>	Blood Brain Barrier
<b>BCAA</b>	Branched Chain Amino Acids
<b>BPM</b>	Beats per minute
<b>CON</b>	Congruent
<b>CHO</b>	Carbohydrates
<b>ENRL</b>	Exercise Nutrition Research Laboratory
<b>FFA</b>	Free Fatty Acids
<b>f-TRP</b>	Free Tryptophan
<b>GABA</b>	<i>gamma</i> -Aminobutyric acid
<b>GLUT- 4</b>	Glucose Transporter 4
<b>HR</b>	Heart Rate
<b>INC</b>	Incongruent
<b>LSPT</b>	Loughborough Soccer Passing Test
<b>Non – GMO</b>	Non Genetically Modified Organism
<b>O<sub>2</sub></b>	Oxygen
<b>PAR – Q</b>	Physical Activity Readiness Questionnaire
<b>PEBL</b>	The Psychology Experiment Building Language

<b>RER</b>	Respiratory Exchange Ratio
<b>RPE</b>	Rating of Perceived Exertion
<b>RSA</b>	Repeated Sprint Ability
<b>UCAN</b>	Generation UCAN®
<b>VO<sub>2</sub>max</b>	Maximal rate of oxygen consumption

# 1 Introduction

In team sports of moderate to long duration such as soccer, fatigue is an inevitable aspect that often becomes apparent towards the later portion of the event. Typically, this is characterized by a drop in work rate (Bangsbo et al., 1991; Rampinini et al., 2007), i.e. players cover less total distance and spend less time running at high intensities in comparison to the first half. For example, Bangsbo et al. (1991) found that professional soccer players covered 5% more distance in the first half compared to the second half. Furthermore, Rampinini et al. (2007) reported that distance run at high exercise intensities during the first half is significantly greater than in the second half. Importantly, this fatigue may also lead to impaired cognitive function resulting in a deterioration of technical skills such as passing, shooting or dribbling (Rampinini et al., 2009a) and/or an increase in poor strategy decisions so critical in sports (Baker et al., 2014).

It is known that hypoglycemia and depleted carbohydrate (CHO) stores are conditions associated with the onset of fatigue (Stone & Oliver, 2009) and these are likely at least partially responsible for the performance decrements later in games or practices. Some investigators have observed that the majority of goals in soccer are scored in the last 30 minutes of a game (Ostojic & Mazic, 2002; Yiannakos & Armatas, 2006). Perhaps, this results from the greater reduction in the work rate seen during this time, from mental fatigue or perhaps a combination of the two for one team rather than an augmented performance for the other team. Certainly, athletes and coaches often attribute the final

outcome of a game to mental mistakes that happen often in the latter stages of a game. In light of this, nutrient supplementation strategies to avoid or delay the onset of fatigue has always been of considerable interest to many.

Generation UCAN® (UCAN) is a CHO drink (made from heat treated, non-GMO corn starch called Superstarch®) that due to its slow rate of absorption, causes a relatively small release of insulin into the blood, which not only helps sustain normal blood glucose, but also may help spare endogenous CHO stores by promoting the use of fats for fuel during the early stages of exercise. Theoretically, these mechanisms could delay the onset of fatigue (both physical and cognitive) and, therefore, enhance performance. For example, storage of glucose in the brain is limited so a constant supply of CHO delivered via the blood is best to preserve optimal functioning (Pardridge, 1983). Moreover, the release of free fatty acids (FFA) from adipose tissue is very sensitive to insulin and even small increases in insulin can reduce lipolysis considerably, which in turn reduces the mobilization of FFA from adipocytes leading to lower overall fat oxidation rates (Bonadonna et al., 1990) resulting in earlier CHO depletion. Therefore, not surprisingly, ingesting CHO regularly, as opposed to one time, during exercise of moderate to long duration has been shown to be beneficial for performance (Mitchell et al., 1989; Welsh et al., 2002; Winnick et al., 2005; Ali et al., 2007a; Cermak & Van Loon., 2013). However, many athletic competitions are designed in such way that supplementation during exercise becomes difficult. Additionally, most sports drinks are composed of sucrose, fructose, or maltodextrin, which are broken down and absorbed relatively rapidly. This creates an unhealthy cycle of a rapid post ingestion increase and subsequent decrease (spike and crash) in blood glucose that could accelerate the onset of fatigue symptoms by

inducing low blood glucose concentrations [sometimes called rebound hypoglycemia (Costill et al., 1977; Moseley et al., 2003).

Superstarch® (the key ingredient in UCAN) is made utilizing heat treatment of non-GMO corn starch, a novel processing technique intended originally to help patients with a rare genetic disorder called glycogen storage disease. This condition is characterized by a failure to be able to utilize glycogen stores, due to a genetic deficiency of phosphorylase, resulting in chronic hypoglycemia. Some studies have shown UCAN treatment to be better than conventional treatments for prevention of hypoglycemia (Bhattacharya et al, 2007; Correia et al, 2008). Furthermore, one study has shown UCAN has the potential to enhance endurance exercise performance (Roberts et al, 2011). However, whether the effects of UCAN treatment are similar during high intensity, intermittent exercise, like that performed in a soccer game (or other similar ball games), is still unknown.

Therefore, the purpose of the present study was to assess whether UCAN supplementation can enhance physical, technical (ball skills) or cognitive performance during a simulated soccer game.

We hypothesized that UCAN supplementation would enhance all three aspects of sports performance.

## 2 Literature Review

### 2.1 Carbohydrate (CHO) Metabolism

Skeletal muscle uses CHO and fats primarily as fuel for energy during any type of sustained or intermittent activity that requires significant muscle contraction (Van Loon et al., 2001). The relative contribution of these macronutrients for energy depends largely on factors such as duration and intensity of exercise (Van Loon et al., 2001), but can also depend on other factors such as training status, environmental conditions, or even gender (Coggan & Coyle, 1991; Van Loon et al., 1999). Carbohydrate stores in muscle (muscle glycogen) are the preferred substrate for energy during high intensity exercise. Many years ago, Gollnick et al. (1974) showed substantial increases in the rate of muscle glycogenolysis, (breakdown of glycogen), during very intense exercise compared to submaximal exercise on a cycle ergometer. At the molecular level, there appears to be several mechanisms responsible for this intensity-dependent glycogen use. The activity of the enzyme glycogen phosphorylase, the rate limiting enzyme for glycogenolysis, is regulated by intramuscular and systemic hormonal factors. More specifically, the increase in sarcoplasmic calcium and inorganic phosphate generated by muscle contractions during intense exercise stimulates the use of glycogen early in exercise and later epinephrine promotes muscle glycogen breakdown (Richter et al., 1982). This dual control allows muscles to utilize glycogen primarily during high intensity exercise in an attempt to match the energetic demands of the activity (Richter et al., 1982).

Blood glucose is also an important source of muscle fuel during exercise. Depending on exercise intensity and duration, glucose uptake into skeletal muscle can increase several fold (Katz et al., 1986). Specifically, with exercise onset, there is a substantial increased

extraction of glucose from blood into skeletal muscle (insulin-like effect of muscle contraction). This is achieved not only due to a very large increase in skeletal muscle blood flow (via capillary dilation) in the exercising muscles but also due to an increased movement (translocation) of the glucose transporter isoform 4 (GLUT 4) to the plasma membrane of muscle, which has been demonstrated to be extremely important for glucose uptake (Zisman et al., 2000). In the past, it was understood that activation of the insulin signaling pathways was required for cellular glucose uptake but insulin is not necessary with muscle contraction. Newer evidence has shown that muscle contraction-induced GLUT 4 translocation, can even have an additive effect when combined with insulin (Holloszy, 2003). These findings suggest that insulin-mediated GLUT 4 translocation is independent of contraction-induced GLUT 4 translocation. Once glucose enters the cell via facilitated diffusion, it is quickly phosphorylated and subsequently oxidized to generate energy for muscle contraction (Katz et al., 1986). Consequently, the ingestion of insulin stimulating CHO shortly before exercise can result in rebound hypoglycemia and can cause impaired exercise performance even when muscle glycogen stores are substantial (Costill et al., 1977).

Liver glucose output during dynamic exercise is also very important in the maintenance of blood glucose. As mentioned, glucose uptake in muscle increases during exercise so the liver becomes an important organ responsible for maintaining normal glycaemia. Consequently, liver glucose output is also dependent on exercise duration and intensity. Further, with intense exercise liver glucose output can be greater than muscle glucose uptake resulting in hyperglycemia (Hultman and Harris, 1988). The quantity of glucose released into the circulation by the liver to regulate glycaemia during exercise is obtained



not only from the breakdown of liver glycogen stores but also from gluconeogenesis, the process of creating new glucose from substrates such as lactate, pyruvate, alanine, and glycerol (Kristiansen et al., 1977). Although early in an exercise bout, the contribution of gluconeogenesis for liver glucose output is quite small, as the duration of exercise increases and liver glycogen stores become depleted, the liver starts to rely more on this metabolic pathway to obtain glucose. Consequently, an increased uptake of these gluconeogenic precursors as well as an increased activity of liver gluconeogenic enzymes have been shown during prolonged exercise (Ahlborg et al., 1974). Unfortunately, despite being a very important metabolic process to help regulate glycaemia as shown in rats by John-Alder et al., (1986), gluconeogenesis cannot fully compensate for the decrease in liver glycogen availability observed during prolonged exercise and thus liver glucose output starts to decrease resulting in low blood glucose concentrations and subsequently a decrease in both muscle glucose uptake (Coggan et al., 1995) and performance.

## 2.2 Fatigue and Exercise

Fatigue is often defined as the inability of muscles to generate (or maintain) force (Rahnama et al., 2003). Typically, a reduction in force with intense exercise is attributed to a reduced glycogen content in the active musculature (Saltin, 1973; Yaspelkis et al., 1993) or low blood glucose concentrations (Coggan & Coyle, 1987). However, clearly the reduction in force output seen with fatigue could also be due to a failure in the central nervous system to provide an optimal neural drive to the contracting skeletal muscles (Newsholme et al., 1987; Nybo and Nielsen, 2001). In team sports, fatigue is manifested through a progressive reduction in muscle force and power output which affects not only the athlete's ability to perform high intensity efforts (Bangsbo et al., 1991; Rampinini et

al., 2007), but also in their ability to perform complicated motor skills (Rampinini et al., 2009a; Stone and Oliver, 2009). Nevertheless, the causes of fatigue are very complex and could be derived from both peripheral and/or central mechanisms (Winnick et al., 2005).

### 2.2.1 Central Fatigue Hypothesis

This hypothesis was conceived based on the assumption that prolonged exercise can influence the synthesis and metabolism of some monoamines such as serotonin, dopamine, and noradrenaline (Meeusen et al., 2006). The first suggestion of a central fatigue hypothesis was by Newsholme et al. (1987). They proposed that variations in serotonergic neural activity during prolonged exercise contribute to the development of central fatigue by increasing lethargy and loss of drive which, in turn, may affect motor unit recruitment negatively resulting in a decreased mental and physical ability to perform exercise (Newsholme et al., 1987). Importantly, serotonin cannot cross the blood-brain barrier (BBB) and thus it needs to be synthesized by cerebral neurons (Meeusen et al., 2006). This process in the brain is dependent on the cerebral concentrations of the precursor amino acid tryptophan (TRP) (Dahlstroem & Fuxe, 1964; Ashcroft et al., 1965). Further, TRP binds to the protein transporter albumin in blood, which also serves as a transporter for free fatty acids (FFA). In blood, only a small amount is present as free TRP (f-TRP). When glycogen stores become depleted, there is an increase in the mobilization of FFA from adipose tissue. Consequently, TRP can be displaced from its protein transporter albumin making it readily available for transport across the BBB (Curzon et al., 1975). Subsequent increases in cerebral tryptophan concentration are correlated with increases in serotonin synthesis (Eccleston et al., 1965; Fernstrom & Wurtman, 1971; Carlsson and Lindqvist, 1972) and fatigue. Moreover, the

rate of cerebral uptake of f-TRP is also determined by the concentration of branched chain amino acids (BCAA) as these competing amino acids use the same transport system (Fernstrom, 1983). Thus, the synthesis and release of serotonin may not only be dependent on cerebral TRP concentrations but also on other factors such as the ratio of f-TRP to BCAA (Newsholme et al., 1987), availability of oxygen (Cooper et al., 2003), cerebral glucose availability (Bequet et al., 2002), and/or the activity of other neurotransmitters such as dopamine and *gamma*-Aminobutyric acid (GABA). Consequently, Davis and Bailey (1997) suggested a revised central fatigue hypothesis arguing, based on their work, that the ratio of serotonin to dopamine would determine the performance outcome. An increased ratio of serotonin to dopamine would affect performance negatively by augmenting feelings of tiredness and lethargy whereas a low ratio would enhance it by maintenance of motivation and arousal (Davis and Bailey, 1997).

### 2.2.2 CHO Supplementation and Central Fatigue

CHO supplementation during exercise has been proposed as an effective nutritional strategy to regulate the synthesis of certain neurotransmitters, and thus delay the onset of central fatigue (Messier, 2004; Meeusen et al., 2006). For example, glucose is needed for acetylcholine synthesis and consequently the increase seen with ingestion of CHO is believed to enhance cognitive function (Messier, 2004). In addition, CHO intake is known to suppress lipolysis by increasing the concentration of circulating serum insulin (Bonadonna et al., 1990). If the concentration of plasma FFA is reduced more TRP would be bound to the protein carrier albumin and, therefore, the increase in f-TRP seen with exercise would be mitigated (Davis et al., 1992). The end result should be a decreased

synthesis of serotonin and reduced central fatigue. A few studies have investigated the effects of CHO feeding on the development of central fatigue. For example, Davis et al. (1992) reported that the progressive increase in the plasma concentration ratio of f-TRP to BCAA seen during prolonged exercise could be attenuated by CHO feeding. In addition, their subjects receiving the CHO drinks were able to exercise longer compared to placebo. However, the direct effect of CHO supplementation on central fatigue is debated due to the ethical complications associated with measuring cerebral concentrations of neurotransmitters in humans.

Welsh et al. (1997) have explored this issue in rodents exercising for either 60, 90 or 120 minutes after ingesting CHO, BCAA or water-placebo. At the end of their corresponding exercise bout, the rodents were killed and brain concentrations of serotonin, dopamine and their primary metabolites, specifically in the striatum and brainstem regions, were measured. Neither BCAA nor CHO feedings affected brain serotonin or dopamine at 60 and 90 min of exercise. However, at 120 min, both BCAA and CHO feedings lowered the concentration of these two neurotransmitters in the brainstem. Interestingly, only CHO lowered serotonin in the striatum. These results suggest a possible positive influence of CHO and BCAA on the development of central fatigue, at least with prolonged exercise. Unfortunately, the animals were not exercised to exhaustion and, therefore, the relationship between the cerebral concentrations of neurotransmitters and their role in central fatigue and intense prolonged exercise in humans remains to be elucidated.

### **2.3 CHO Supplementation and Hypoglycemia**

Another possible benefit of CHO feeding during exercise is to avoid hypoglycemia and sustain adequate substrate delivery to the brain late in exercise. Exercise-induced

hypoglycemia has been shown not only to affect cerebral metabolic rate negatively by reducing glucose uptake in brain (Nybo et al., 2003) but also to attenuate the ability to sustain a high neural drive to the exercising muscles (Nybo, 2003). Perhaps, CHO ingestion can delay this situation by sustaining optimal blood glucose concentrations to allow normal glucose uptake in brain and provide an additional source of muscle CHO, thereby enhancing performance late in exercise. Recent evidence suggests that another potential exercise enhancing mechanism of CHO might involve CHO-sensitive receptors in the mouth increasing central activation. This evidence comes from mouth rinsing studies (no actual swallowing) with a 6% CHO solution (Jeukendrup, 2013; Turner et al., 2014). Surprisingly, this research has shown that CHO mouth rinsing activates certain areas of the brain responsible for motor control (Chambers et al., 2009; Turner et al., 2014), improving performance during short duration (~60 minutes) high intensity (<75%  $\text{VO}_2\text{max}$ ) exercise (Jeukendrup, 2010). However, these receptors have not been identified yet, so, specifically how they affect the central nervous system or the role that they might play on central fatigue remains unclear.

## 2.4 CHO Supplementation and Intermittent Exercise Performance

Many sports are characterized by high intensity bouts of exercise interspersed with short bouts of lower intensity exercise or relative rest. These include most team sports such as soccer, basketball, rugby, hockey, football as well many individual sports such as tennis, squash, badminton, etc (Cermak & Van Loon, 2013). The importance of CHO for performance in these types of sports is well known (Cermak & Van Loon, 2013). As an example, soccer is an activity of long duration played at an average intensity of 70 - 80%

VO<sub>2</sub>max, with an average respiratory exchange ratio (RER) of 0.88 (Bangsbo, 1994). This value indicates a heavy reliance on muscle and liver glycogen to supply the energy needed to meet the demands of the exercise (Bangsbo 1994; Jeukendrup, 2004). Perhaps the first to demonstrate how critical pre-game glycogen content can be for soccer performance was Saltin (1973). In his study, two groups of soccer players started a soccer game with differing amounts of glycogen in muscle by manipulating their diet and exercise on the days preceding the game. One group started with 96 mmol glucose units • kg<sup>-1</sup> of muscle which is deemed to be normal, and the other group had only half that concentration. At half time, the group with a greater glycogen content depleted about 2/3 of their stores whereas the other group had depleted almost all their stores. At the end of the game, glycogen in muscle was very low in both groups. Interestingly, both groups covered less distance in the second half, but the low group covered 25% less distance than the high group. Furthermore, the low group covered 50% of the total distance walking and only 15% running at maximal speed whereas the high group walked for only 27% of the total distance covered during the game and were able to run at maximal speed 24% of the time.

As mentioned earlier, blood glucose can also impact sports performance significantly. With the onset of exercise, glucose in blood rises and remains elevated during activity as long as liver glucose output is optimal. With the depletion of glycogen stores in liver, and in the absence of supplementation, serum glucose drops to a point where it can affect athlete's performance negatively (Ekblom, 1986; Leatt, 1986; Coggan & Coyle, 1987). Exercise CHO supplementation has been shown to reduce this drop in work rate during intermittent activity (Mitchell et al., 1989; Welsh et al., 2002; Winnick et al., 2005; Ali et

al., 2007a; Cermak & Van Loon, 2013;). Nevertheless, the mechanisms underlying these beneficial effects are not well established. One of the plausible benefits is ascribed to the ability of the body to spare muscle glycogen stores when CHO are consumed during exercise (Yaspelkis et al., 1993). For example, in a study by Yaspelkis et al. (1993) subjects cycled for 190 minutes, alternating between low to moderate intensities while consuming either a solid form of CHO, a liquid form of CHO or water-placebo. Muscles biopsies were taken before and after exercise and at the end of the exercise. Individuals who consumed CHO had a significantly greater amount of muscle glycogen indicating that a sparing of muscle glycogen during exercise occurs with CHO ingestion. Although there is other evidence to support this hypothesis (Tsintzas et al., 1995; Stellingwerff et al., 2007), some investigators have failed to demonstrate it (Flynn et al., 1987; Hargreaves & Briggs, 1988; Mitchell et al., 1989). In the Flynn et al. (1987) study, subjects performed four exercise sessions where they consumed a CHO drink (three sessions) or a water-placebo (one session) prior to and during 120 minutes of cycling exercise. When all trials were compared, there was no significant difference in muscle glycogen content at the end of exercise. Perhaps importantly, these subjects had performed a glycogen depleting exercise session two days before the experimental trials and then consumed the same diet (high CHO) in an attempt to match and maximize muscle glycogen stores before all sessions. These results may indicate that when muscle glycogen stores are great, the dependence on exogenous CHO is reduced. However, it could be argued that either the duration of exercise influenced the results as muscle glycogen changes have been shown to be time-dependent (Stellingwerff et al., 2007) or the specific muscle fibre type recruited (Tsintzas et al., 1995) because both depend upon

the type and intensity of exercise. For instance, Stellingwerff et al. (2007) reported that CHO supplementation only helped spare muscle glycogen during the early stages of exercise while Tsintzas et al. (1995) showed that CHO ingestion only helps in sparing of glycogen in type I (slow twitch) fibres. Interestingly, the intensity in both studies was similar while the duration of exercise in the Tsintzas et al. (1995) was 60 minutes, and in the Stellingwerff et al. (2007) was 180 minutes. Clearly, these methodological factors play a role when determining differences in glycogen use.

Another possible mechanism responsible for performance enhancement with CHO supplementation is the maintenance of blood glucose concentration within an adequate range, i.e., avoiding low blood glucose concentrations during exercise (Coyle et al., 1986; Coggan & Coyle, 1987; Ali et al., 2007a; Roberts et al., 2011). In the study by Coyle and colleagues (1986), seven subjects who cycled at ~70% of  $\text{VO}_2$  max took four hours to fatigue after ingesting a glucose polymer solution, which was one hour longer than ingesting water. Further, in the water trial, fatigue was preceded by a gradual decrease in blood glucose concentration, something that was not observed in the CHO trial.

Similarly, Febbraio et al. (2000) showed that ingestion of CHO during a 2-hour exercise session helped maintain plasma glucose concentrations and improved performance on a subsequent exercise time trial. In contrast, some have not been able to demonstrate this benefit (Jentjens et al., 2003; Claassen et al., 2005; Russell et al., 2014). For example, Russell et al. (2014) had soccer players ingest a 6% CHO-electrolyte solution before and every 15 minutes during a 90-minute soccer game and found no effect on blood glucose concentrations during the latter stages of the game vs those who ingested an electrolyte placebo solution. Specifically, there was a transient reduction in blood glucose



concentration during the CHO trial, similar to a water-placebo trial, at the start of the second half (following a 15-minute passive recovery time), a phenomenon sometimes known as rebound hypoglycemia. This response depends on the timing of CHO ingestion and exercise, and is thought to be due to a combination of the insulin effect on blood glucose and the insulin-like effect of muscle contraction. Here, it is important to note that in the Russell et al. study (2014), the first CHO ingestion was two hours before initiation of the soccer game. Others have reported that ingesting high glycemic index CHO (CHO that are absorbed rapidly and cause a rapid increase in blood glucose) within an hour or two of activity can induce hypoglycemia (Costill et al., 1977; Moseley et al., 2003). Similarly, Jentjens et al. (2003) found that ingestion of CHO prior to exercise did not maintain plasma glucose concentrations during the latter stages of the exercise. Further, it caused hypoglycemia in 6 out of 9 subjects and did not improve performance.

Other factors such as quantity and type of CHO have also received considerable attention in the literature. There seems to be a ceiling value for CHO intake (~30g/h – 60g/h) where no further benefit is seen if the dose increases perhaps due to delayed gastric emptying (Leiper et al., 2001; Rodriguez et al., 2009). So, although there is plenty of evidence supporting CHO as an ergogenic aid for performance, some studies failed to show any improvements suggesting that a number of factors such as timing, type of CHO and/or quantity should be considered in order to develop better supplementation strategies.

## 2.5 CHO Supplementation and Cognitive/Skill Performance

Low blood glucose concentrations can also impair cognitive and/or psychomotor functions (Amiel, 1998). As mentioned above, ingestion of CHO can avoid/delay

exercise-induced hypoglycemia and thus could attenuate the deterioration on cognitive and psychomotor performance often seen during the latter stages of exercise. However, the effect of CHO supplementation on cognitive performance is unclear. There is some evidence supporting enhanced cognitive performance (Collardeau et al., 2001; Bottoms et al., 2006) but there is also evidence suggesting no benefit (Welsh et al., 2002; Winnick et al., 2005; Bottoms et al., 2012). For example, cognitive function was measured using the Stroop color-word test during an intermittent exercise bout designed to simulate the physical demands of team sports but there was no effect on cognitive performance despite observed improvements in physical performance measures such as jumping and sprinting ability as well as in a whole body motor skill test (a pseudo hopscotch task) (Winnick et al., 2005). A recent review of the literature by Baker et al. (2014) suggested cognitive tests may not be sensitive enough to detect any positive effect given by exercise CHO intake. Although this may be true, there are several methodological factors that must be controlled before this suggestion can be substantiated. For instance, cognitive assessment timing is a very important factor to consider. Dietrich and Sparling (2004) suggested that the point at which cognitive tests are administered is very important because neural activation associated with exercise rapidly returns to baseline following the cessation of exercise. Further, a meta-analysis suggested that exercise led to an impairment in cognitive performance when the tests were performed during exercise while small improvements were seen when tests were performed following exercise (Lambourne and Tomporowski, 2010). One possible explanation for the discrepancy may be dual-task interference. Of course, athletes must complete complex motor skills, at intense rates, while making complex decisions simultaneously and this could result in

reduced attention perhaps due to the metabolic needs of the neural circuitry necessary to execute the complex motor skills required (the so called “hypofrontality” hypothesis proposed by Dietrich, 2003). So it would likely be best to measure cognitive performance during exercise rather than following it.

### 2.5.1 Sport-Specific Skills Performance.

Clearly, the technical aspects (sport skills) become extremely important for performance because the main objective of sports games is to score more goals/points than the opposition (Russell and Kingsley, 2014). Some authors have even identified these aspects as being more important than high intensity physical performance for success in soccer (Hughes and Franks 2005; Di Salvo et al., 2009). According to Rampinini et al. (2009a), passing and dribbling skills are the most frequent skills performed during soccer games. Despite some studies showing no effect of CHO consumption on sport-specific motor skills performance (Baker et al., 2007; Roberts et al., 2010), there is overwhelming evidence supporting the efficacy of CHO supplementation before and during exercise to enhance skill performance in sport disciplines such as basketball, tennis, soccer, etc (Vergauwen et al., 1998; Dougherty et al., 2006). Specifically, in soccer, CHO supplementation has been shown to attenuate the decrease in skill performance seen during the latter stages of a game (Northcott et al., 1999; Ostojic and Mazic, 2002; Ali et al., 2007a; Ali and Williams, 2009; Currel et al., 2009; Russell et al., 2012). Of course, this can contribute to more success.

## 2.6 Generation UCAN<sup>®</sup> (UCAN)

UCAN sports drink is a, relatively new, commercially available supplement intended to enhance performance by optimizing energy use. Superstarch<sup>®</sup> its key ingredient, is a CHO made from non-GMO corn starch that, due to its slow rate of absorption into the blood stream, causes a relatively small release of insulin in blood (Roberts et al., 2011), which may help spare body CHO stores by promoting the use of fats for fuel. This would be good for athletes as CHO stores are limited and thus sparing them may represent an advantage especially toward the end of the event. UCAN is cornstarch which has undergone a heat-moisture treatment which alters the way it is absorbed. Heat treatment of corn starch is a novel processing technique intended to help children with a rare genetic disorder called glycogen storage disease. This disease involves an inability to utilize glycogen due to absence of the enzyme phosphorylase resulting in life threatening hypoglycemia. To survive these patients must eat every 2 hours even through the night. The slow absorption of UCAN has been a near miracle type treatment allowing patients to sleep through the night and to participate in regular activities by maintaining normal blood glucose. Studies have shown Superstarch<sup>®</sup> treatment to be better than conventional treatments for prevention of hypoglycemia (Bhattacharya et al, 2007; Correia et al, 2008). As a result, it has been theorized, that UCAN may be a useful supplement for athletes because low blood glucose concentrations and depleted glycogen stores are often associated with the onset of fatigue in different sport disciplines; however, to our knowledge, only one study has been published and it investigated endurance performance of cyclists (Roberts et al., 2011). Therefore, the purpose of the present study was to assess the effect of Generation UCAN supplementation on soccer skills and physical/

cognitive performance during intense, intermittent, repeated exercise like that found in many ball games. It was hypothesized that UCAN supplementation would enhance all three aspects of sports performance.

## 3 Methods

### 3.1 Participants.

Fifteen experienced male soccer players were recruited but four participants did not complete the entire experimental protocol. One participant vomited during the first experimental trial, two other participants had work obligations during the study and could no longer complete all measures and a fourth participant had unexpected academic obligations. These four participants were excluded from data analysis (Table 1 in Results section). All participants were competitive athletes from a range of outfield playing positions and had been participating in soccer training at least ~3 d/wk for 6 months prior to the study. Each completed a health information form (Appendix C) and the PAR-Q (Canadian Society for Exercise Physiology, 2002, Appendix D) to screen out any potential contraindications to the exercise. All risks and discomforts were explained fully prior to any testing. All data collection was conducted in the Exercise Nutrition Research Laboratory (ENRL) and Alumni Hall Gym (AH Gym) at The University of Western Ontario. All participants provided written, informed consent of the protocol approved previously by the Office of Research Ethics at Western.

### 3.2 Preliminary visits and baseline testing

Prior to any experimental testing, participants visited the ENRL/AH Gym on three separate occasions for familiarization to laboratory and field testing procedures and screening/baseline measures. On the first visit, participants had their body composition measured and then completed an incremental running test to measure their maximal

oxygen consumption ( $\text{VO}_{2\text{max}}$ ).

On a subsequent day, at least 48 hours after the first visit, each was given an opportunity to familiarize himself with both the cognitive and skill tests in order to limit any potential learning effect during the study. The cognitive tests used included the Eriksen-Flanker test and the Serial seven subtraction test. The soccer skill tests used were the Slalom dribble test and the Loughborough soccer passing test (LSPT) (Ali et al., 2007b). All participants completed 8 familiarization repeats for each test. In addition, they completed a familiarization with the simulated soccer game protocol by performing a 15-minute (1 block) run on the treadmill at the intensities to be used during the experimental simulation. During this time, cognitive performance was assessed in the same way it was to be done during the experimental sessions. During this familiarization session, players were introduced to the 20-point Borg Scale of perceived exertion (RPE; Borg, 1973; see Appendix E) and were provided clear directions on how to use it to represent their perceptions of effort during the simulated game.

On the third day of preliminary testing, baseline scores for the two skill tests were collected. Specifically, after a standardized 10 min warm-up, all participants performed four trials on the LSPT. The first two were “re-familiarization” trials and the average score of the last two trials was used as the final score.

### 3.3 Experimental overview.

A double-blind, randomized, crossover research design was implemented involving two experimental treatments heat treated corn starch (UCAN) or Dextrose (CHO Placebo). Each treatment consisted of a 4 h test day in the ENRL and AH Gym. Conditions were

separated by at least one week. Briefly, the first participant was assigned randomly to one experimental treatment and thereafter treatments were systematically rotated to avoid order effects. Each participant completed both conditions (UCAN and Placebo) and neither the subject nor investigator knew which condition was which (supplement preparation and assignment was completed by a Graduate student not involved in the study). All drinks were given to the participants in opaque plastic 1L Green Gatorade® drink bottles.

Participants recorded their food/drink intake for the two days prior to the first test day in order to replicate nutrient consumption the two days preceding their next trial. Further, all were provided a standardized low carbohydrate meal consisting of two burgers with lettuce buns, boiled yellow mini-potatoes and apple juice ( $\sim 9 \text{ kcal}\cdot\text{kg}^{-1}$  and  $1 \text{ g}\cdot\text{kg}^{-1}$  of carbohydrate [CHO]) at 1930 h on the eve of testing in order to minimize the intra- and inter-variability of nutritional status (Jeacocke & Burke, 2010). This was the last food intake prior to the experimental day but water intake was allowed *ad libitum*.

On test days, participants reported to the ENRL at 0730 h after a 12 h overnight fast having abstained from strenuous exercise, caffeine or alcohol consumption for 24 h prior to testing. Upon arrival at the laboratory, measures of baseline blood glucose, blood lactate and resting expired gases were obtained. Subsequently, participants performed the Eriksen-Flanker test and the Serial Seven subtraction test 3 times each. The first two were “re-familiarization” attempts and the last test score was used as a baseline score. Once the cognitive tests were finished, participants were provided with the first bottle of the experimental or placebo drink and were allowed 30 minutes for gastric emptying/absorption. Blood glucose concentration was measured 15 and 30 minutes after



ingestion of the experimental drink. Then, participants executed a standardized 10-minute exercise warm-up, blood glucose and lactate were measured again and then the 60-minute simulated soccer game protocol began. This game was comprised of 4 x 15-minute intermittent running blocks interspersed with 3 minutes of passive recovery. The first 3 blocks corresponded to the first half of the game, then a 15-minute half time break was given. During this time, the second bottle of the corresponding experimental drink was provided. The Serial Seven subtraction test and the Eriksen-Flanker test were completed at consistent time points during each standardized 15-minute running block and expired gas samples were collected during the last 4 minutes of each exercise block (Figure 1). In addition, heart rate was monitored throughout. Measures of blood glucose and lactate as well as ratings of perceived exertion were collected during the passive recovery after each exercise block. Ten minutes after completion of the last block, participants performed a repeated sprint ability test (RSA), followed by two soccer skill tests, the Slalom Dribbling test and the Loughborough soccer passing test (LSPT). The final measures of blood glucose and lactate as well as ratings of perceived exertion were collected following the Slalom Dribbling test (Figure 1).

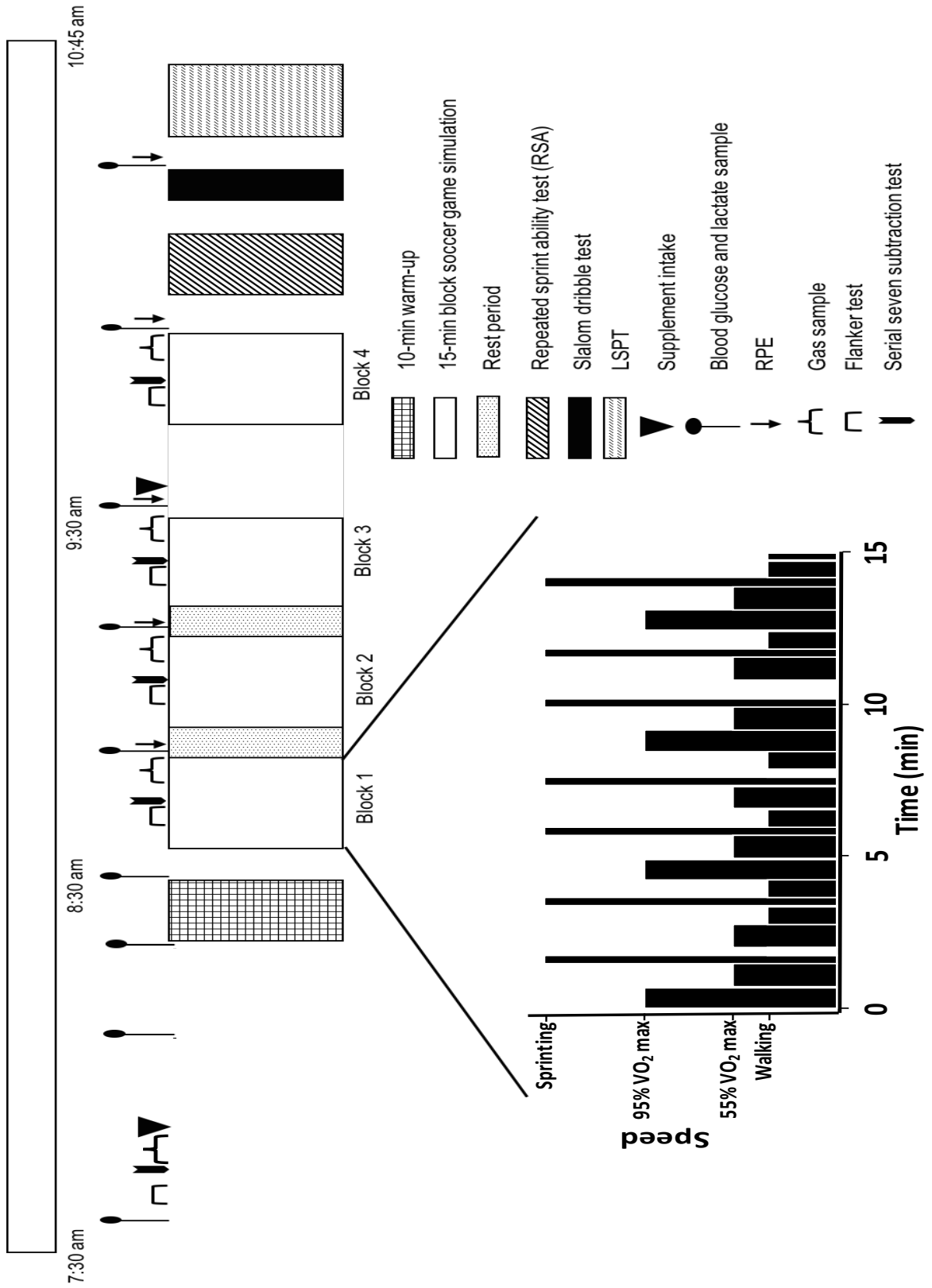


Figure 1. Experimental protocol

### 3.4 Supplementation Treatments

Both treatments were carbohydrate beverages because CHO or energy ingestion enhances exercise performance and all participants completed both trials. Experimental treatments included UCAN and Dextrose (placebo). Both drinks consisted of an 8% CHO solution containing a total of  $0.7\text{g}\cdot\text{kg}^{-1}\cdot\text{h}^{-1}$  ( $2.8\text{ kcal}\cdot\text{kg}^{-1}\cdot\text{h}^{-1}$ ) of the corresponding CHO. The total amount of CHO was provided at two time points, the first one 30 minutes prior to commencement of exercise ( $497\pm 48\text{ ml}$ ; containing  $0.5\text{g}\cdot\text{kg}^{-1}$ ) and the second one was provided at half time ( $199\pm 18\text{ ml}$  containing  $0.2\text{g}\cdot\text{kg}^{-1}$ ). The quantity of CHO ingested and the volume of water used to prepare the beverages were determined based on ACSM guidelines (2009). The electrolyte concentration in both beverages was controlled using a non-caloric flavoured electrolyte replacement powder (UCAN Hydrate®) that contained  $94\text{ mg}\cdot\text{L}^{-1}$  of magnesium,  $281.7\text{ mg}\cdot\text{L}^{-1}$  of chloride,  $563.4\text{ mg}\cdot\text{L}^{-1}$  of sodium,  $187.9\text{ mg}\cdot\text{L}^{-1}$  of potassium and  $28\text{ mg}\cdot\text{L}^{-1}$  of calcium. These quantities were determined based on manufacturer's guidelines. As mentioned, both experimental drinks were administered in identical, opaque drink bottles. Prior to ingestion, participants were told not to open the bottle nor discuss their supplement with anyone. The beverages were indistinguishable by taste and texture. Participants were asked after ingesting each drink to try to identify the solution as placebo or treatment. Their responses (only 3/11 guessed correctly) clearly indicating that they did not know the order of treatments.

### 3.5 60-minute Simulated Soccer Game Protocol

The simulated soccer game was adapted from a protocol developed previously by Drust et al. (2000) and later modified by Clarke et al. (2008). The latter investigators used the same absolute speeds for every participant. The participants in the present study ran at the

same relative intensities based on their recorded maximum ability and at exercise intensities known to occur in soccer games (Nicholas et al., 2000). The simulation was conducted on a treadmill (Desmo Pro, Woodway®, Wisconsin, USA) interfaced with computer based, interactive Software (PT Pro, Version 2.2). The Software was used to ensure that changes of treadmill speed were done at the same times during all trials. The protocol was comprised of four, 15-minute blocks running at varying intensities to replicate the activity pattern of soccer such as sprinting, jogging, cruising and walking. Backwards movements, sideways movements and actions with the ball were not included in the protocol because of the difficulty to execute these actions when using a motorized treadmill. The first half was comprised of three blocks, each separated by three minutes of passive recovery. The last intermittent running block was completed after a 15-minute half time break. The proportion of time spent at each exercise intensity was allocated based on a time-motion analysis of professional soccer players observed by Reilly and Thomas (1976). The times used for each of these exercise intensities in each block were as follows: full stopping (15 s), walking (35 s), jogging (46 s at 55 % VO<sub>2</sub>max), cruising (42 s at 95% VO<sub>2</sub>max) and sprinting (17 s at 90% of their speed during the 15 sec treadmill sprint). The duration of each bout included the time lag for changes in treadmill speed. The different speeds used during the running block were determined using the following equations:

For walking: (*comfortable gait speed in males 20-29-year-old*)

$$\text{Speed (mph)} = (0.788 \text{ cm/s} \times 0.0223694 \text{ mph}^{-1}) * \text{Height (cm)} \text{ (Bohannon, 1997).}$$

For jogging:

$$0.55 \text{ } VO_{2 \text{ max}} = 0.2 * \text{Speed}(m \cdot s^{-1}) + 0.9 * \text{Speed}(m \cdot s^{-1}) * \text{fractional grade} + 1$$

*MET (ACSM, 2000)*

For Cruising:

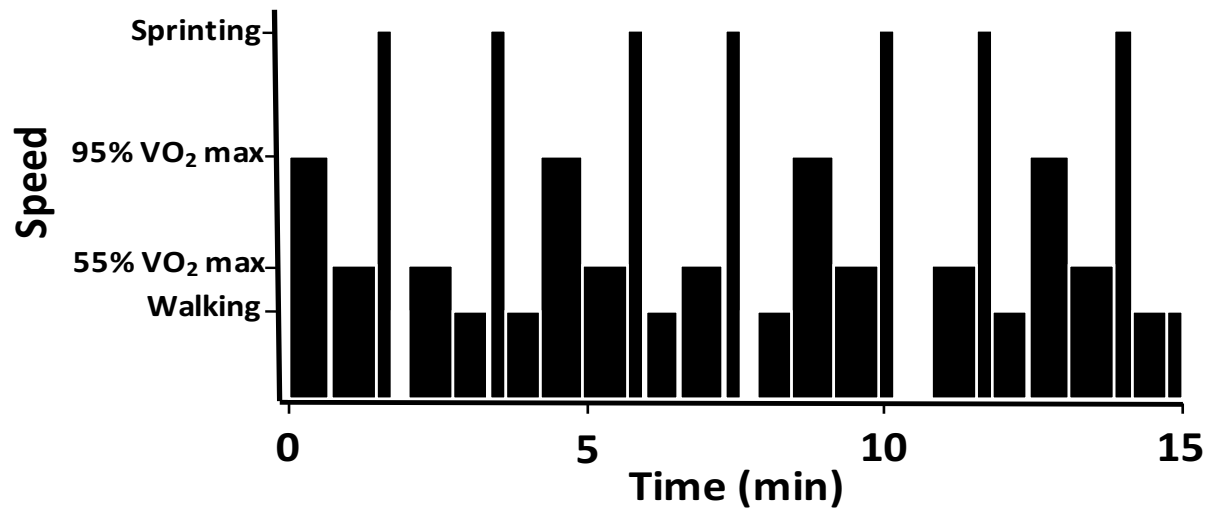
$$0.95 \text{ } VO_{2 \text{ max}} = 0.2 * \text{Speed}(m \cdot s^{-1}) + 0.9 * \text{Speed}(m \cdot s^{-1}) * \text{fractional grade} + 1$$

*MET (ACSM, 2000)*

For Sprinting:

*90% of their peak sprint speed in a 15-sec all out test on a non-motorized treadmill.*

Throughout the block, participants were informed 15 seconds and 5 seconds prior to all upcoming changes in speed. In addition, they were given feedback on time elapsed and time remaining to complete the block.



**Figure 2. Exercise Intensity profile of each running block.**

## 3.6 Measurements

### 3.6.1 Body Composition

Air displacement plethysmography body volume (BodPod®, Life Measurements, Concord, CA) and body mass were used to determine body density. Participants were required to fast three h prior to entering the BodPod®, and to wear approved clothing (compression shorts and lycra swim cap) to minimize errors due to air in hair or under clothing. Thoracic volume was estimated via a calculation integral to the BodPod® software. Body composition was estimated from the measured body density using the Siri (Siri, 1961) equation.

### 3.6.2 Aerobic Capacity

VO<sub>2</sub>max was determined via an incremental speed protocol on a treadmill (Desmo Pro, Woodway®, Wisconsin, USA). Briefly, participants started running at 9.7 km•h<sup>-1</sup> with the treadmill set at a constant gradient of 1%. Subsequently, increases in speed of 0.16 km •h<sup>-1</sup> every 12 sec were applied until volitional exhaustion. Heart rate (HR) was monitored throughout the test (Cardio 660™, Sportline, Elmsford, New York). Expired gases were collected via a breath-by-breath collection system (Sensormedics Vmax 29, Yorba Linda, CA). The system was calibrated according to manufacturer's guidelines using known gases volumes and composition of air samples. The greatest value achieved over a 20-second collection period was considered max whenever a plateau in VO<sub>2</sub> occurred (<50% of the expected increase in oxygen uptake for the increased workload) or when two of the following three criterion measures were attained (±10 bpm of age predicted maximum HR, RER >1.15 [RER = volume of CO<sub>2</sub> produced/volume of O<sub>2</sub> consumed] or volitional exhaustion).

### 3.6.3 Sprint test

Peak sprint speed was determined using a 15-second all-out effort on a non-motorized treadmill (Desmo Pro, Woodway®, Wisconsin, USA). For this test, the treadmill running belt was disengaged so that participants were able to propel it themselves. At a 2% incline, participants grabbed onto the treadmill handle bars and ran as fast as they could for 15-seconds. The highest speed shown on the treadmill's display board during the test was recorded as peak speed.

### 3.6.4 Heart Rate

HR was monitored continuously throughout the simulated soccer game protocol via a Cardio 660<sup>TM</sup> heart rate monitor (Sportline, Elmsford, USA). Highest HR recorded for two runs at each different exercise intensity throughout each block were averaged to provide an estimation of HR for each exercise intensity and throughout the whole block.

### 3.6.5 Blood Glucose and Lactate

Finger prick blood glucose and lactate measurements were taken at baseline (fasted; 0730 h), before exercise (0830 h), after completing each of the four exercise blocks (0850 h, 0910 h, 0930 and 1005 h) and after completing the Slalom dribble test (1025 h). In addition, blood glucose concentrations were also measured 15 and 30 minutes after ingestion of the experimental drink. While sitting down on a chair within the ENRL, participant's finger was first washed with soap and water and subsequently cleaned with an alcohol swab, air-dried then pierced with a FreeStyle Lancing Device® (Abbott Diabetes Care Limited, Saint-Laurent, Quebec). The first drop of blood was discarded and the second drop was analyzed via FreeStyle Freedom Lite® Glucometer (Abbott Diabetes Care Limited, Saint-Laurent, Quebec) and Lactate Scout+ analyzer (EKF

Diagnostics, Cardiff, United Kingdom). The FreeStyle Freedom Lite® Glucometer and the Lactate Scout+ analyzer were chosen as they required very little blood (0.3 µL of blood and 0.2 µL of blood, respectively) and were shown previously to be accurate within the range of expected blood glucose and lactate concentrations, respectively (Freckmann et al., 2012; SensLab GmbH., 2012).

### 3.6.6 Fluid Ingestion

Prior to each simulated soccer game, participants were instructed to engage in normal hydration practices and consume water *ad libitum* throughout the protocol. Participants were provided a 1L Green Gatorade® squeeze bottle, filled with a pre-weighed volume of water. At the end of the exercise session, the water bottle was reweighed to the nearest 1.0 g on a Mettler-Toledo® Scale PB3002 (Mettler-Toledo Canada, Mississauga, Ontario). The volume of fluid consumption was quantified as the difference in pre- and post-exercise water bottle mass.

### 3.6.7 Cognitive Function

Cognition was evaluated during exercise. Briefly, participants completed several basic cognitive tasks, which have been shown previously to measure sport-specific cognitive demands (Voss et al., 2009). A review conducted by Tenenbaum and Bar-Eli (1993) concluded that decision making in sport is contingent on short-term/working memory, visual search strategies, concentration and attentional allocation. Therefore, in order to assess some of these fundamental cognitive demands of sport effectively, two cognitive tests were conducted throughout the simulated soccer game. A modified one-minute serial-seven subtraction test, as previously described by Kennedy & Scholey (2000), and the Flanker test were chosen based on previous validation as reliable measures of at least



one cognitive outcome of interest. See below for details.

The PEBL (Michigan Tech University, Houghton, MI), a widely available Windows computer software program, provided the platform for the Flanker test. Previous research conducted by Piper and colleagues (2012) concluded that PEBL demonstrated similar plasticity as other previously validated cognitive batteries when evaluating executive function (umbrella term for the management of many of the aforementioned cognitive traits of interest). Furthermore, as open-source software, our research team was able to alter source codes and manipulate the number of trials to fit the current protocol during the periods of time where participants were not sprinting. During each experimental trial, the cognitive tests were conducted at baseline (prior to first supplement ingestion) and during each exercise block of the simulated soccer game.

### 3.6.7.1 Modified Eriksen-Flanker Task

The Flanker task was incorporated as an assessment of attentional control - the ability to direct attention dynamically while ignoring distractions (Eriksen and Eriksen, 1974). Attentional control is often included in sport cognition batteries, as the ability to direct attention and pick out relevant cues is believed to be associated with sport expertise (Williams and Davids, 1998; Mann et al., 2007). Specifically, the Eriksen-Flanker task presented five arrows centered linearly on the computer screen. Participants were instructed to focus their attention on the center arrow, and ignore the two flanking arrows on either side. Each test was comprised of 40 random trials (20 congruent and 20 incongruent). The congruent trials consisted of the target arrow being flanked by other arrows that faced the same direction (e.g., ←←←←← or →→→→→). The incongruent trials consisted of the target arrow being flanked by other arrows that faced the opposite

directions (e.g.,  $\leftarrow\leftarrow\rightarrow\leftarrow\leftarrow$  or  $\rightarrow\rightarrow\leftarrow\rightarrow\rightarrow$ ). To respond, participants pressed a push button on a hand held device interfaced with the computer, while running on the treadmill. The push button for each hand also corresponded to the direction of the center arrow on the computer screen, regardless of condition. This test was performed twice within each exercise block for a total of 8 times during the soccer game simulation. The task was scored as mean choice reaction time (ms), accuracy (%) and conflict cost (also known as the 'Flanker effect') (ms). Eriksen-Flanker conflict cost was quantified as the difference in mean choice-reaction time between congruent and incongruent trials.

### 3.6.7.2 Serial Seven Subtraction

Originally developed in 1942 by Hayman, serial subtraction is a mental math cognitive task, which is postulated to measure working memory (DeStefano and LeFevre, 2004; Kase et al., 2009). Participants completed a 1-minute verbal version of the serial-seven subtraction task once for each of the four exercise blocks throughout the protocol. Starting numbers for all trials were between 800-999 and chosen by means of a random number generator. Prior to testing, participants were instructed to subtract seven recursively from the starting number as quickly and as accurately as possible within the allotted one minute. Time commenced once participants repeated the original number. If a mistake was made, participants were asked to continue subtracting seven from the erroneous value and subsequent responses were scored in relation to the incorrect number. All verbal responses were recorded audibly to ensure answers were quantified accurately. In order to minimize any potential learning curve, participants completed practice trials during familiarization visits as well as before each simulated game. Each trial was quantified as the proportion of correct responses over one minute as follows:

$$\% \text{ correct} = \text{correct responses} / \text{correct} + \text{incorrect responses}$$

### 3.6.8 Rate of Perceived Exertion (RPE)

Subjective ratings of perceived exertion were collected after completing each of the four exercise blocks (0850 h, 0910 h, 0930 and 1005 h) and after completing the Slalom dribble test (1025 h) using the 20- point Borg Scale (Borg, 1973) (Appendix E). The scale was shown to the participants at the end of each exercise block as well as after the Slalom dribble test, and they were asked to rate how hard the exercise felt based on their current state.

### 3.6.9 Repeated Sprint Ability test (RSA)

Soccer players' ability to perform repeated high-intensity actions is considered a key factor in elite soccer (Rampinini et al., 2009b). Therefore, a repeated sprint ability test was implemented to assess physical performance after the 60-minute simulated soccer game. The test was comprised of 12 x 30 m sprints interspersed with 35 seconds of recovery (Glaister et al., 2008). All sprints were performed on a flat, non-slippery wooden surface in Alumni Hall gym at Western University. Timing gates (Western Engineering Electronic shop, London, CA) were placed at 0, 20m and 30m. Each sprint was initiated one meter behind the start line to avoid false triggering of the first timing gate, and finished one meter in front of the last timing gate to ensure participants did not slow down before crossing the 30 m point. Participants were required to give a maximal effort in each sprint. The following measures of performance were calculated from the RSA:

- 1) Mean sprint time = average time, expressed in seconds, for all the sprints performed.

- 2) Best sprint time = best time out of all the sprints performed.
- 3) Percent sprint decrement =  $100 \times (total\ sprint\ time / ideal\ sprint\ time) - 100$ ,  
where:

Total sprint time = sum of sprint times from all sprints.

Ideal sprint time = number of sprints x fastest sprint time (Glaister et al., 2008).

### 3.6.10 Skill tests

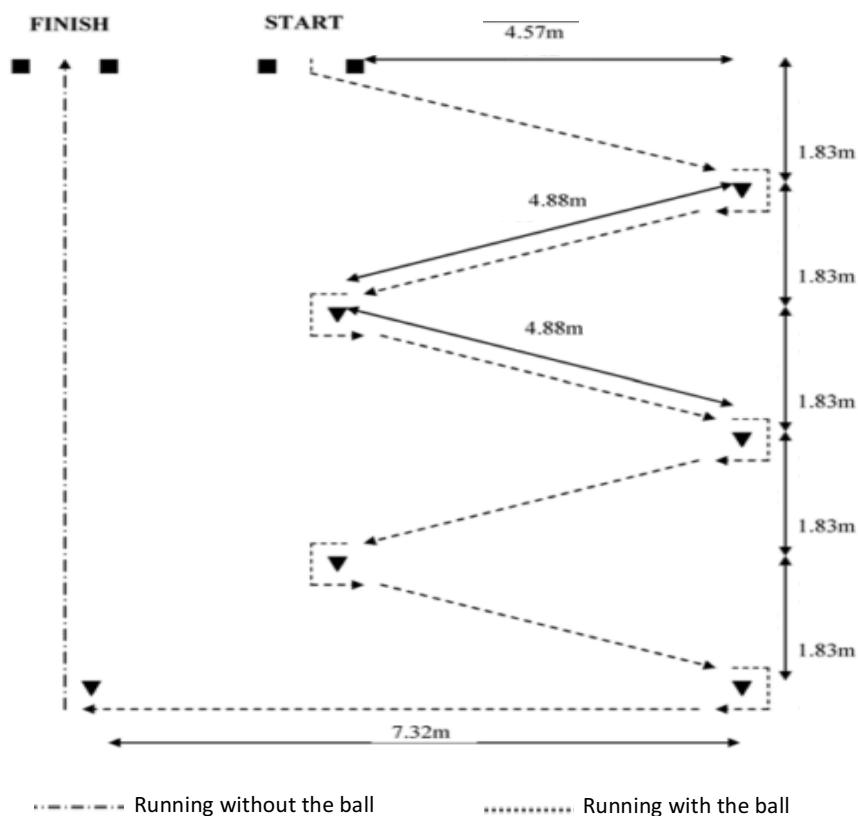
All testing protocols were carried out on a flat, non-slippery wooden surface in Alumni Hall gym at Western University. Grids and distances were measured using a 30m tape, and were marked out with cones and green painters tape.

#### 3.6.10.1 Slalom Dribble Test

Soccer dribbling skill was assessed using the slalom dribble test. This test has been shown to have good ecological and construct validity as well as high test-retest reliability ( $r = .95$ ; Reilly and Holmes, 1983). It evaluates total body movement, challenging participants to dribble a ball around a set obstacle course as quickly as possible (Figures 3 and 4). Timing gates (Western Engineering Electronic shop, London, CA) were placed at the start and end lines in order to measure accurately the time taken to complete the course as shown (Figure 3). Participants stood with the ball 1 meter behind the starting line. On the command go, they dribbled the ball towards the right of the first cone and then alternately around the outside of the remaining cones in a zig-zag fashion. Once they got to the sixth cone, the ball was stopped and subsequently they sprinted in a straight line to the end line. Participants were required to perform the slalom dribble test four times with a rest of 1 minute between trials. The first two were “re-familiarization” trials and the average score of the last two trials was used as the final score.



**Figure 3. Set up for the Slalom dribble test**



**Figure 4. Diagram of the Slalom dribble test**

### 3.6.10.2 Loughborough Soccer Passing Test (LSPT)

The LSPT is a psychomotor skill test that incorporates various psycho-technical aspects of soccer such as passing, control, dribbling and decision-making (Ali et al., 2007b).

Participants must perform 16 passes to four different coloured targets, while negotiating a coned area, as quickly as possible (Figures 5 and 6). The four coloured target areas measured 30 cm x 60 cm each with an inner aluminum target measuring 15 cm x 10 cm.

These targets were attached to wooden gym benches to allow rebounding of the ball.

Participants started with the ball in the middle of the inner grid. As soon as the first colour was called, participants dribbled into the passing area and passed the ball to the

target as quickly and accurately as possible. As the ball was travelling back to them, the examiner called the next colour target. Then, participants controlled the ball, moved in the inner grid and then out to the corresponding passing area to perform the next pass. The passing sequence was assigned randomly (Appendix F). There were 8 long and 8 short distance passes in total, and participants were allowed 43 seconds to complete the test before they were penalized. Penalty and bonus time were accumulated according a pre-set criterion (Ali et al., 2007b):

5 s for missing the bench completely or passing to the wrong bench

3 s for missing the target area (60 cm x 30 cm)

3 s for handling the ball

2 s for passing the ball from outside of the designated area

2 s for not crossing two inner lines

2 s if the ball touches any cone

1 s for every second over the allocated 43 s to complete the test

1 s was deducted from the total time if the ball hit the 10-cm strip in the middle of the target

Three parameters of performance were calculated from the LSPT including:

1) Test Time: Time to complete all 16 passes as measured with a stopwatch (Cardio 660<sup>TM</sup>, Sportline, Elmsford, USA).

2) Penalty Time: Total penalties accumulated from the mistakes made and the bonuses scored during the test execution.

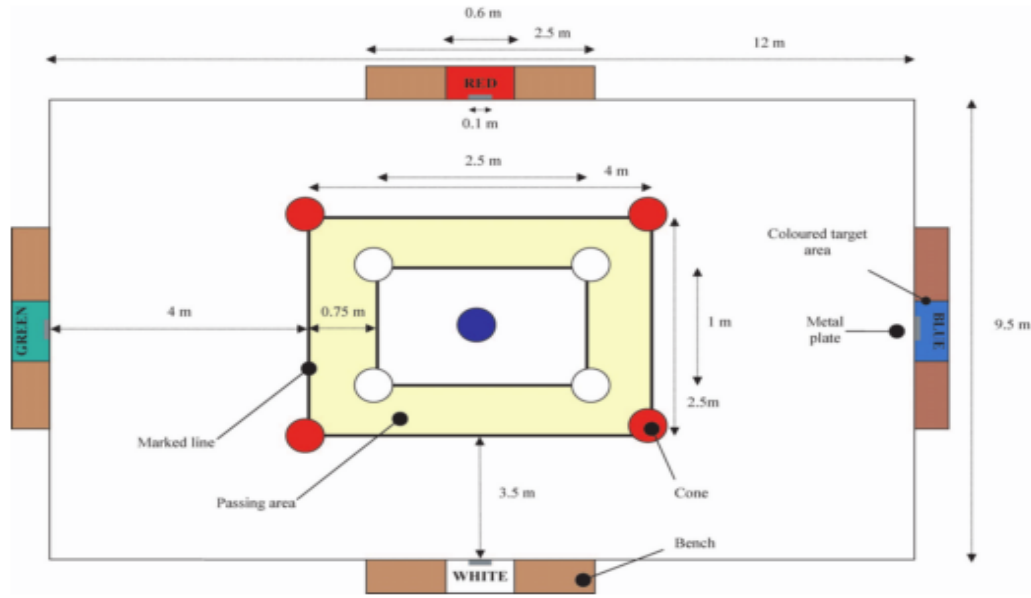
3) Total Performance Time: Time to complete the test after adjusting for penalties and

bonus time.



**Figure 5. Picture of the set up for the LSPT**





**Figure 6. Diagram of the LSPT**

### 3.7 Statistical Analysis

Statistical analyses were performed using SigmaPlot for Windows (Version 12.0). Blood metabolites concentrations, expired gases samples, ratings of perceived exertion, accuracy and reaction times during cognitive performance results were analyzed using two-way (condition by time) repeated measures ANOVA. The Slalom dribble test was analyzed using one-way ANOVA. Tukey's HSD was used for post-hoc analysis of any significant effect. Furthermore, The Eriksen-Flanker conflict cost, RSA and LSPT data were analyzed using a paired t test. Significance was set at  $p \leq 0.05$ . Data are presented as means  $\pm$  SD.

## 4 Results

### 4.1 Descriptive Analysis

With respect to physical characteristics the 11 experienced male soccer players studied appeared to be representative of young, elite soccer players (Table 1). The total distance run during the 60-minute simulated game was 8.7 km, comparable with the distance covered in a game by professional soccer players (Rampinini et al., 2009a).

**Table 1. Participant Characteristics (n=11)**

Height, cm	177.7 ± 6.8
Mass, kg	77.3 ± 7.9
Age, y	22 ± 3
Body fat, %	12.8 ± 4.9
VO <sub>2</sub> max, ml•kg <sup>-1</sup> •min <sup>-1</sup>	57.1 ± 3.9

Cm = centimeters; kg= kilograms; y = years, ml = milliliters, min = minutes.  
Data are reported as mean ± SD.

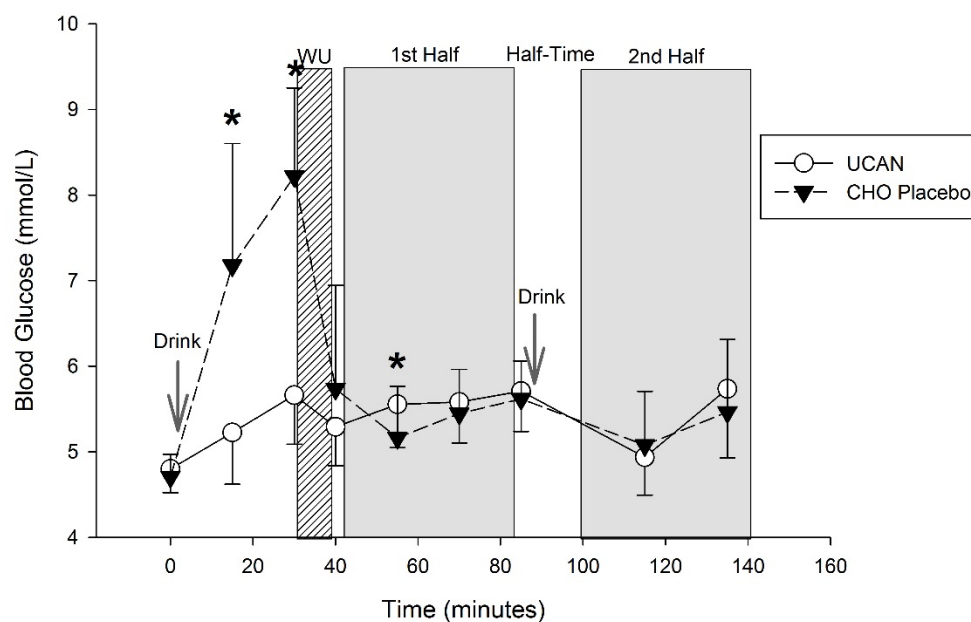
### 4.2 Blood Metabolites

#### 4.2.1 Blood Glucose

Due to technical difficulties, two participants were excluded from the analysis (n=9).

There was a main effect of time ( $p < 0.001$ ) for blood glucose concentration. Blood glucose concentration at 30 minutes post drink was greater compared to any other time during the experimental protocol. Additionally, there was a main ( $p= 0.006$ ) effect of treatment as well as a significant treatment x time interaction ( $p < 0.001$ ). Pairwise

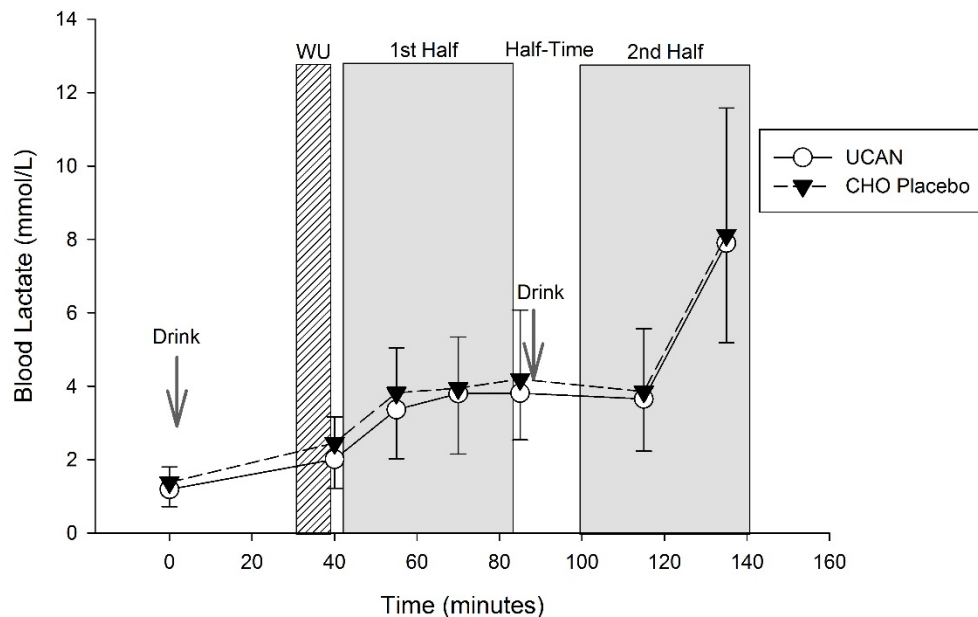
comparisons showed that blood glucose concentration was lower for UCAN at both 15 minutes ( $p < 0.001$ ) and 30 minutes ( $p < 0.001$ ) post treatment ingestion, and greater after the first exercise block ( $p = 0.004$ ) compared to CHO-Placebo (Figure 7).



**Figure 7. Blood glucose concentration before and during a simulated soccer game. Values are means  $\pm$  SD for CHO-Placebo (filled triangles,  $n = 9$ ) and UCAN (open circles,  $n = 9$ ). \*Significantly different than UCAN ( $p < 0.005$ ).**

#### 4.2.2 Blood Lactate

There was a main effect of time ( $p < 0.001$ ) for blood lactate. Blood lactate concentration was greater after performing the repeated sprint ability test, compared to baseline or any other time during exercise (Figure 8). Further, blood lactate concentration for block 3 was greater than baseline and post warm-up concentrations (Figure 8). However, no treatment ( $p = 0.13$ ) or interaction ( $p = 0.99$ ) effects were found.

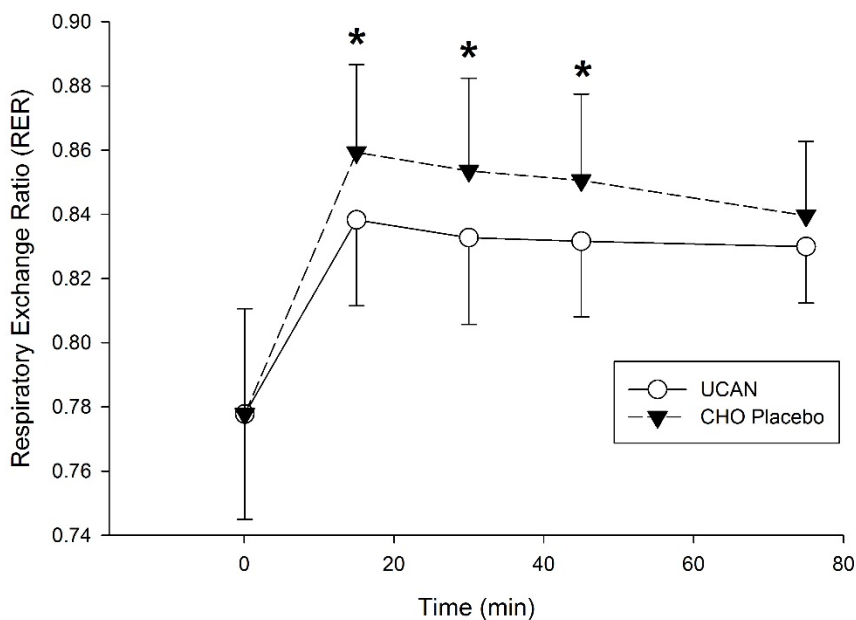


**Figure 8. Blood lactate concentrations at rest and during a simulated soccer game. Values are means  $\pm$  SD for CHO-Placebo (filled triangles,  $n = 11$ ) and UCAN (open circles,  $n = 11$ ).**

### 4.3 Respiratory Exchange Ratio (RER)

There was a main effect of time ( $p < 0.001$ ) and treatment ( $p = 0.02$ ) for RER.

Specifically, for both experimental drinks, RER was greater for all exercise blocks compared to baseline. Furthermore, a treatment x time interaction was detected ( $p = 0.002$ ). Pairwise comparisons revealed that RER values were lower for UCAN during block 1 ( $p = 0.003$ ), block 2 ( $p = 0.004$ ) and block 3 ( $p = 0.007$ ) when compared to CHO Placebo (Figure 9).

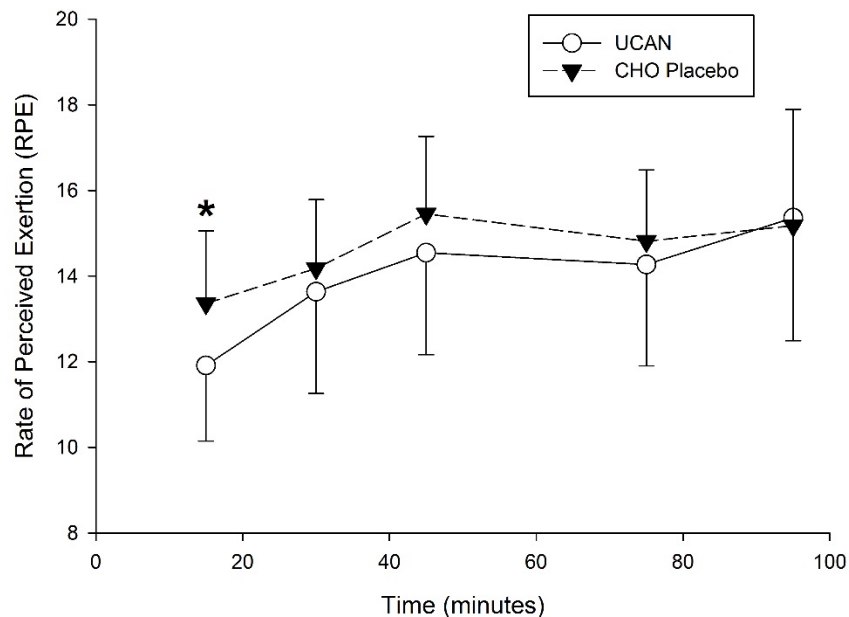


**Figure 9. Mean respiratory Exchange Ratio (RER) at baseline and during a 60-minute simulated soccer game. Values are means  $\pm$  SD for CHO-Placebo (filled triangles,  $n = 11$ ) and UCAN (open circles,  $n = 11$ ). \* Significantly greater than UCAN ( $p= 0.02$ ).**

#### 4.4 Ratings of Perceived Exertion

A main effect of time ( $p < 0.001$ ) was detected for RPE. Average RPE for block 1 was lower than block 3, block 4 and post- Repeated Sprint Ability test (RSA). No treatment effect ( $p= 0.24$ ) was observed but there was a treatment x time interaction ( $p= 0.013$ ).

Pairwise comparisons using a Tukey analysis showed that average RPE for block 1 was lower with UCAN ( $p= 0.025$ ) compared to CHO-Placebo (Figure 10).



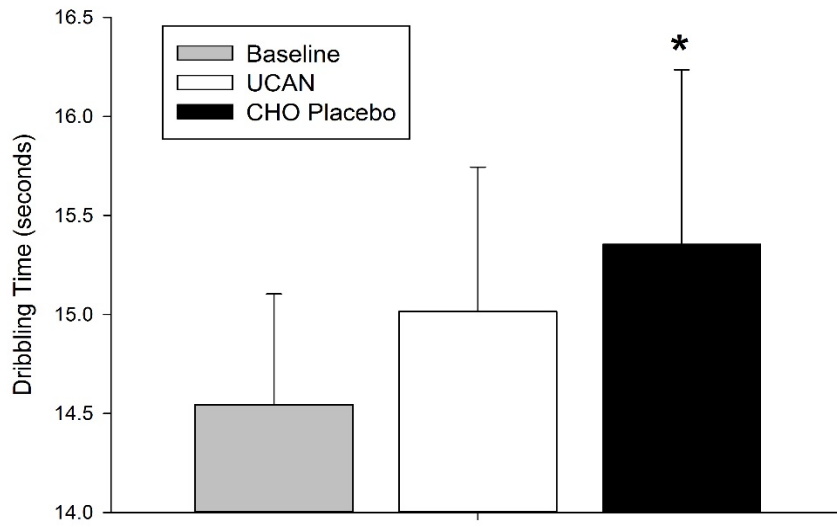
**Figure 10. Ratings of perceived exertion during a simulated soccer game. Values are presented as mean  $\pm$  SD for CHO-Placebo (filled triangles, n = 11) and UCAN (open circles, n = 11). \*Significantly greater than UCAN ( $p = 0.025$ ).**

## 4.5 Measures of Exercise performance

### 4.5.1 Slalom Dribble Test

A main effect of treatment was observed ( $p = 0.03$ ). Comparisons using a Tukey test revealed that dribbling time for CHO Placebo was ( $p < 0.05$ ) greater (slower) than baseline. No statistically significant differences were observed between UCAN and

baseline (Figure 11).



**Figure 11. Dribbling time at baseline and after 60 minutes of a simulated soccer game. Values are presented as mean  $\pm$  SD for Baseline (n = 11), CHO-Placebo (n = 11) and UCAN (n = 11). \*Significantly greater than Baseline ( $p < 0.05$ ).**

#### 4.5.2 Loughborough Soccer Passing Test

*Test Time:* No significant effects were observed ( $p = 0.85$ ) comparing UCAN to a CHO Placebo (Table 2).

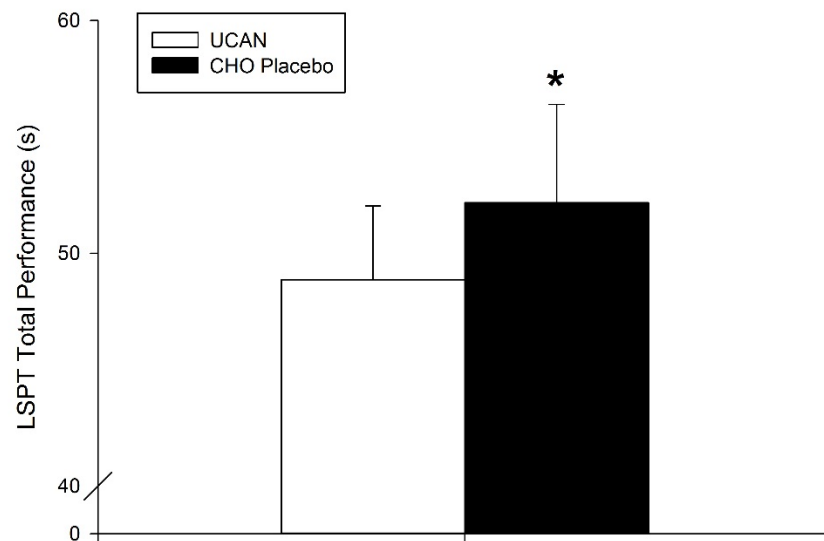
*Penalty Time:* A difference was found comparing UCAN to a CHO Placebo. Average penalty time was lower ( $p = 0.004$ ) with UCAN compared to CHO Placebo (Table 2).

**Table 2. Loughborough Soccer Passing Test (means  $\pm$  SD)**

<b>Test Time (s)</b>	UCAN	44.59 $\pm$ 2.19
	PLACEBO	44.46 $\pm$ 1.80
<b>Penalty time (s)</b>	UCAN	4.27 $\pm$ 2.23 *
	PLACEBO	7.73 $\pm$ 4.50

s= seconds; \* p= 0.004 vs CHO Placebo

*Total Performance Time:* A significant difference was observed ( $p= 0.006$ ) comparing UCAN to the CHO Placebo. Total performance time was reduced (faster) with UCAN compared to CHO Placebo (Figure 12).



**Figure 12. Total performance time during the Loughborough Soccer Passing Test (LSPT) after 60 minutes of a simulated soccer game. Values are presented as mean  $\pm$  SD for CHO-Placebo (filled bar,  $n = 11$ ) and UCAN (open bar,  $n = 11$ ).**

**\*Significantly less (faster) with UCAN ( $p= 0.006$ ).**



### 4.5.3 Repeated Sprint Ability Test (RSA)

There were no differences in mean sprint time between treatments for 20 meters ( $p=0.68$ ) and 30 meters ( $p=0.35$ ). Additionally, no differences were observed for best sprint time between treatments for 20 meters ( $p=0.82$ ) and 30 meters ( $p=0.26$ ). Furthermore, no significant differences were found in percent sprint decrement for 20 meters ( $p=0.73$ ) and 30 meters ( $p=0.54$ ) when comparing UCAN to a CHO Placebo (Table 3).

**Table 3. Repeated Sprint Ability Test (means  $\pm$  SD)**

		<b>20 m</b>	<b>30 m</b>
<b>Mean Sprint Time (s)</b>	UCAN	3.08 $\pm$ 0.11	4.43 $\pm$ 0.15
	PLACEBO	3.10 $\pm$ 0.13	4.48 $\pm$ 0.22
<b>Best Sprint Time (s)</b>	UCAN	2.94 $\pm$ 0.13	4.26 $\pm$ 0.15
	PLACEBO	2.95 $\pm$ 0.11	4.32 $\pm$ 0.23
<b>Percent Sprint Decrement (%)</b>	UCAN	4.8 $\pm$ 2.0	4.0 $\pm$ 2.0
	PLACEBO	5.0 $\pm$ 1.3	3.6 $\pm$ 1.3

s= seconds; \*  $p<0.05$

## 4.6 Measures of Cognition

### 4.6.1 Eriksen-Flanker Task – Mean Reaction Time

*Congruent:* There was a main effect of time ( $p=0.008$ ) for the congruent trials. Block 1 mean reaction times were greater than both baseline and block 4. However, no treatment ( $p=0.55$ ) or interaction ( $p=0.69$ ) effects were found (Table 4).

*Incongruent:* A main effect of time ( $p = <0.001$ ) was detected for incongruent trials.

Block 1 mean reaction times were greater than all other times. However, no treatment ( $p=0.22$ ) or interaction ( $p=0.30$ ) effects were found (Table 4).

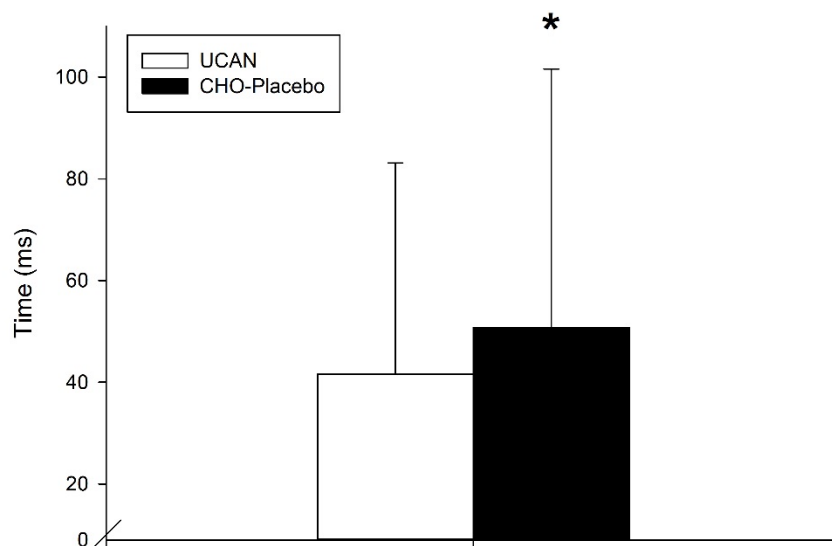
#### 4.6.2 Eriksen-Flanker Task – Response Accuracy

*Congruent:* There were no main (time,  $p=0.51$ ; treatment,  $p=0.71$ ) or interaction (time x treatment,  $p=0.35$ ) effects for congruent trials. (Table 4).

*Incongruent:* There were no main (time,  $p=0.09$ ; treatment,  $p=0.98$ ) or interaction (time x treatment,  $p=0.12$ ) effects for incongruent trials. (Table 4).

#### 4.6.3 Eriksen-Flanker Task – Conflict Cost

There was a main effect of time ( $p=0.04$ ) and treatment ( $p=0.03$ ). However, no interaction ( $p=0.89$ ) effects were detected (Table 4). When averaged for all exercise blocks, the Conflict Cost for each participant during exercise under both treatments, a paired t-test indicated that UCAN supplementation was faster ( $p=0.019$ ) in Flanker Conflict Cost between the two experimental treatments (Figure 13).



**Figure 13. Averaged Flanker Conflict Cost during a 60-minute simulated soccer game. Values are means  $\pm$  SD for CHO-Placebo (filled bar, n = 11) and UCAN (open bar, n = 11). \*Significantly greater (slower) than UCAN (p= 0.019).**

#### 4.6.4 Serial Seven Subtraction Test

*Number of responses:* There was a main effect of time ( $p < 0.001$ ) on the number of responses provided for one minute during exercise. The number of responses at baseline was lower compared to all blocks during exercise. However, no treatment ( $p=0.73$ ) or interaction ( $p=0.71$ ) effects were found (Table 4).

*Response Accuracy:* There was a main effect of time ( $p= 0.04$ ). Block 1 response accuracy was lower ( $p= 0.03$ ) compared to baseline. No treatment ( $p=0.32$ ) or interaction ( $p=0.30$ ) effects were observed (Table 4).

**Table 4. Measures of cognition (means  $\pm$  SD)**

		<b>Baseline</b>	<b>Block 1</b>	<b>Block 2</b>	<b>Block 3</b>	<b>Block 4</b>
<b>Flanker Con-MRT (ms)</b>	UCAN	448 $\pm$ 46	465 $\pm$ 55	455 $\pm$ 57	454 $\pm$ 54	451 $\pm$ 38
	PLACEBO	446 $\pm$ 47	477 $\pm$ 69	460 $\pm$ 48	462 $\pm$ 51	453 $\pm$ 40
<b>Flanker Inc-MRT (ms)</b>	UCAN	489 $\pm$ 55	512 $\pm$ 67	493 $\pm$ 56	491 $\pm$ 61	494 $\pm$ 42
	PLACEBO	487 $\pm$ 56	529 $\pm$ 74	504 $\pm$ 67	505 $\pm$ 61	501 $\pm$ 50
<b>Flanker Con-Accuracy (%)</b>	UCAN	98 $\pm$ 5	99 $\pm$ 2	99 $\pm$ 1	100 $\pm$ 1	100 $\pm$ 1
	PLACEBO	99 $\pm$ 2	99 $\pm$ 2	100 $\pm$ 1	99 $\pm$ 3	99 $\pm$ 2
<b>Flanker Inc-Accuracy (%)</b>	UCAN	95 $\pm$ 8	95 $\pm$ 7	94 $\pm$ 8	94 $\pm$ 8	95 $\pm$ 6
	PLACEBO	98 $\pm$ 3	93 $\pm$ 8	94 $\pm$ 6	93 $\pm$ 8	96 $\pm$ 5
<b>Flanker Conflict Cost (ms)</b>	UCAN	41 $\pm$ 20	46 $\pm$ 21	38 $\pm$ 15	38 $\pm$ 17	43 $\pm$ 17
	PLACEBO	41 $\pm$ 10	52 $\pm$ 21	44 $\pm$ 23	43 $\pm$ 16	48 $\pm$ 21
<b>Serial Subtraction (% Correct)</b>	UCAN	91 $\pm$ 9	82 $\pm$ 20	87 $\pm$ 16	89 $\pm$ 15	90 $\pm$ 11
	PLACEBO	88 $\pm$ 15	86 $\pm$ 14	84 $\pm$ 16	82 $\pm$ 18	85 $\pm$ 14
<b>Serial Subtraction (# Answers)</b>	UCAN	20 $\pm$ 7	23 $\pm$ 11	23 $\pm$ 10	24 $\pm$ 9	25 $\pm$ 10
	PLACEBO	21 $\pm$ 10	24 $\pm$ 10	24 $\pm$ 10	24 $\pm$ 11	24 $\pm$ 11

**MRT=mean reaction time; Con= Congruent; Inc= Incongruent; \*p<0.05**

## 5 Discussion

Compared to an isoenergetic CHO Placebo drink ingested during a simulated soccer game, the major findings of the present study include that UCAN supplementation: 1) resulted in better soccer skills (dribbling and passing) performance, 2) maintained lower respiratory exchange ratio values suggesting a possible CHO sparing effect, and 3) displayed more stable blood glucose concentrations and reduced perceived exertion, early in the simulated game. Considering the present study was the first to investigate UCAN supplementation efficacy on different parameters of sports performance during/following repeated, intense, intermittent exercise, it is not possible to directly compare the findings of the present study to others in the literature. In fact, to our knowledge, only one study has used UCAN supplementation previously and that study examined the effect on a number of physiologic and metabolic parameters during much more prolonged exercise at a lower intensity ( $\sim 70\%$   $VO_2\text{max}$ ) (Roberts et al., 2011). Those investigators reported that UCAN supplementation blunted the initial increase in serum insulin resulting in a more steady serum glucose response as well as an increase in both serum free fatty acid and glycerol concentrations compared to a maltodextrin placebo drink. Further, they assessed substrate oxidation (indirect calorimetry) and, although they found no significant differences between trials in respiratory exchange ratio (RER), the observed lower values with UCAN supplementation did approach significance ( $p=0.07$ ) at both 60 and 90 minutes of exercise. These observations suggested a greater use of fats for fuel during this type of exercise and the resulting sparing of CHO could have important implications on both physical and cognitive performance during the latter stages of

prolonged exercise events. However, the investigators did not observe any differences in physical performance during a time to exhaustion test done after 150 minutes of exercise at 70% of  $\text{VO}_2$  peak indicating any physical performance benefit of UCAN intake with this type of exercise is small at best. Further, they did not attempt to assess cognitive performance effects so whether UCAN affects exercise cognitive function is unknown. Nonetheless, this type of exercise test (time to exhaustion) may not be the best choice to assess any potential UCAN benefit on many types of athletic performance because it did not include the repeated, intense, intermittent exercise used by athletes in many sports such as soccer, hockey, basketball, etc. Therefore, those previous results are quite limited. In the current study with intense, repeated, intermittent exercise, UCAN ingestion also displayed a small rise in blood glucose concentrations initially, and presumably blunted the initial response of serum insulin resulting in more steady blood glucose concentrations similar to those seen in the Roberts et al. (2011) study. During exercise, blood glucose concentration were similar for both treatments, except after block 1. One could argue that these results indicate a similarity in substrate use during the majority of the exercise time in both experimental days. However, blood glucose concentration is simply reflecting the balance between liver glucose output and muscle glucose uptake. Therefore, until liver glycogen stores are depleted, it is possible for participants to show a similar glucose exercise response. Coyle and colleagues (1986) showed that blood glucose concentrations in a water-placebo group were similar during the first several hours of exercise, compared to a CHO group yet observed significant differences in performance after 3 hours of cycling. Perhaps our simulated soccer game was not long enough to show differences in blood glucose concentrations. Further, RER values with

UCAN supplementation were lower ( $p=0.002$ ) at several time points during the simulated soccer game indicating there were differences in substrate use and suggesting a possible carbohydrate (CHO) sparing effect. Although the role of fat utilization during high intensity exercise ( $>85\%$  of  $\text{VO}_2$  max) is often considered negligible (Achten et al., 2002), Romijn et al. (1993) demonstrated its use can be substantial, and a recent study by Hetlelid et al. (2015) showed that the difference in performance between a group of elite and recreational runners was due mainly to increased fat use. Specifically, the elite runners had nearly a threefold increase in fat oxidation despite reporting similar ratings of perceived exertion (RPE), blood lactate concentrations, and carbohydrate oxidation rates vs the recreational runners during six bouts of four minutes of running just above second ventilatory threshold. These data indicate that fat oxidation can be very important even during high intensity exercise performance.

Importantly, RPE during the present simulated soccer game was lower with UCAN, particularly during the first 15 minutes of exercise ( $p=0.025$ ) indicating that the participants found the exercise to be less difficult. This could mean that in an actual soccer game when intensity is not fixed, athletes ingesting UCAN might be able to exercise harder and, therefore, assuming soccer skills are similar, improve their performance. Interestingly, blood glucose concentrations 15 minutes into the simulated soccer game were lower ( $p=0.004$ ) with CHO placebo following a large increase pre-exercise. This rapid decrease following the large increase with CHO Placebo ingestion may be due to the combined effects of greater circulating insulin and the insulin-like effect of muscle contraction (Costill et al., 1977; Moseley et al., 2003). Regardless, in the present study, blood glucose concentrations were above clinical hypoglycemic thresholds

consistently and, therefore, participants were able to complete the simulated soccer game despite the increased RPE with CHO Placebo ingestion.

## 5.1 Effects on Cognition

As expected, reaction time was shorter and accuracy improved during the congruent trials of the Eriksen-Flanker test under both experimental drinks, compared to the incongruent trials. Further, these scores were better (faster) with UCAN supplementation compared to CHO placebo. Although the individual time point differences did not reach statistical significance, there appears to be a positive effect for UCAN supplementation, which would benefit athletes playing in activities requiring intense, intermittent exercise, especially requiring numerous split-second decisions (Anzender and Bosel, 1998; Williams et al., 1999). As a result, conflict cost or Flanker interference effect (difference in reaction time between congruent and incongruent trials) was less ( $p= 0.04$ ) with UCAN supplementation suggesting more cognitive control with UCAN. This should allow participants to overcome conflict faster and could represent an advantage for performance in many activities, especially team sports like soccer.

Based on the central fatigue hypothesis (Newsholme et al., 1987), one might expect that UCAN supplementation could be detrimental to cognitive performance because a greater mobilization of free fatty acids (FFA) in blood could displace the amino acid tryptophan (TRP), increasing the concentration of free TRP (fTRP) in blood. A resulting increased production of serotonin could cause lethargy and tiredness. However, in the present study, RPE and scores during the Flanker tests were improved (lower) with UCAN supplementation. Perhaps serotonin synthesis is not the only factor determining central



fatigue. In fact, the ratio of serotonin to other monoamines such as dopamine may also be important (Davis and Bailey, 1997).

Interestingly, reaction times for the Eriksen-Flanker test were greater (slower) during exercise vs baseline (Table 2), which seems counterintuitive; however, these observations are in agreement with a recent meta-analysis, which showed that exercise led to cognitive performance impairment when the tests and physical exercise were performed concurrently (Lambourne and Tomporowski, 2010). One possible explanation for this outcome may be dual-task interference. Specifically, it is believed that executing two tasks simultaneously can result in reduced attention (Dietrich, 2003). Thus, the present data appear to support the “transient hypofrontality hypothesis” proposed by Dietrich (2003). Basically, tasks, such as those in the Flanker test, which depend on the frontal lobe (frontal dependent cognition), result in transient impairments during exercise because exercise increases the cognitive demand responsible for gross body movement. This increase occurs without a concurrent increase in available metabolic resources, which in turn, causes a reallocation of resources that results in the temporary inhibition of neural networks (Pontifex and Hillman, 2007). Surprisingly, these slower reaction times during exercise did not affect accuracy. This may be because most scores at baseline were perfect (100% accuracy), consistent with previous studies (Kraemer et al., 1994; Zeef et al., 1996). Obviously, this makes it difficult to establish whether or not there is a treatment effect of UCAN because participants cannot perform any better than baseline and especially so if the test is insensitive to small decreases in accuracy. Use of a more challenging cognitive task where the baseline scores are much lower might have been better. Further, a major limitation when analyzing the cognitive results in the present

study was the observed variability. Despite repeated pre-study practice trials, there was substantial between-subject variability as well as day to day variability, obscuring any treatment effect, i.e. a potential statistical type II error. As expected, the responses during the incongruent trials generated greater (slower) reaction times that were also less accurate for both experimental drinks. These findings are also consistent with the literature (Pontifex and Hillman, 2007; Davranche et al., 2009). Future studies need to address these limitations by increasing the number of participants, by recruiting participants with very similar baseline performances, and perhaps developing more complicated cognitive tests.

In contrast, participants with UCAN did not perform better on the Serial Seven Subtraction test, compared to a CHO placebo. As with the Eriksen-Flanker test, there was considerable between-subject and within-subject variability, making it hard to find a treatment effect, if any, i.e. a possible statistical type II error. Specifically, the number of answers in one minute for participants during this test ranged from 8 to 42. Further, only one participant provided the same number of answers at baseline (no treatment) during both experimental days while the rest of the participants had a score that varied from 2 to 9 answers displaying substantial day to day variability. Generally, those who had a low number of answers during the test, made more mistakes, as well. Although this test has been used as a measure of working memory (Kase et al., 2009), the task itself requires a reasonable level of arithmetic skill and perhaps this is the critical determinant of performance with this type of test. Nonetheless, working memory is defined commonly as the ability to retain information while processing the same or other information concurrently (Salthouse and Babcock, 1991). Interestingly, there was a main effect of

time indicating that performance actually improved with exercise duration, as opposed to deteriorating as with the Eriksen-Flanker test. This suggests that this task may not be dependent fully on the frontal lobe; perhaps integrating other areas of the brain, as suggested by some authors (Smith and Jonides, 1999; Bledowski et al., 2009).

Furthermore, patients with frontal lobe injuries have been reported to perform normally on conventional intelligence tests (Hebb, 1939), suggesting that even if the frontal lobe is affected adversely, other areas of the brain can perform normally.

## 5.2 Effect on Soccer Skills

One of the most important findings in the present study was that UCAN supplementation improved soccer skill performance. Specifically, participants were able to perform the dribbling test faster and the LSPT better with UCAN supplementation. These results indicate an advantage of UCAN supplementation over a CHO placebo relative to soccer skill performance, at least in the latter stages of a soccer game. Some investigators have suggested that, although related, success in soccer is more a result of maintaining soccer skill performance throughout the game than one's ability to exercise intensely (Hughes and Franks, 2005; Di Salvo et al., 2009). Of course, the main objective of soccer is to score more goals than the opposition and, skill performance is critical to attain this objective (Russell and Kingsley, 2014). In the present study, as expected, mean slalom dribble time increased (got worse) vs baseline as a result of the simulated soccer game but interestingly with UCAN supplementation, the deterioration was only 3% vs 6% for CHO placebo trial ( $p < 0.05$ ). Additionally, UCAN supplementation allowed participants to perform better on several aspects of the LSPT. One of the strengths of this test is that it measures skills dynamically, i.e., it measures the ability to perform a pass (technique) in a

dynamic setting, by incorporating psycho-technical aspects of soccer such as passing, control, dribbling and decision-making (Ali et al., 2007b). As such, it is likely a good predictor of actual soccer performance.

The parameters measured during the LSPT are the time taken to complete each sequence of the passes, plus the additional penalty time accrued for inaccurate passes or poor ball control, which when combined make up the total performance time. Interestingly, there were no differences between trials for the time taken component of the test so the difference in the assessed performance was due to the additional penalty time (Table 4) with the CHO placebo trial. This result may indicate that there were no differences in gross motor performance but rather in fine motor performance, which could be extremely important when competing in soccer at an elite level. A plausible mechanism explaining the observed results could be that the players in the CHO placebo trial could have been more glycogen depleted vs UCAN. Jacobs (1981) suggested that anaerobic performance can be affected negatively when muscle glycogen concentrations fall below a critical threshold of  $175 \text{ mmol} \cdot \text{kg}^{-1}$  dry muscle mass. While we did not measure muscle glycogen concentrations, the exercise protocol used during the present study replicates the physical demands and activity patterns of a soccer game (Drust et al., 2000). Further, muscle glycogen content has been shown to decrease substantially during the second half of a soccer game, even when players have consumed a large amount of CHO in the days preceding the event (Saltin, 1973). Therefore, it is possible that muscle glycogen content was lower at the end of the CHO placebo trial resulting in a poorer passing performance. This suggestion is consistent with the observed greater RER values, i.e., greater reliance

on CHO for energy, seen during the CHO Placebo trial. More study with muscle glycogen measures is needed to confirm this.

A limitation of the present study was that calculation of the percent decrement of the passing test relative to baseline scores was not completed because the baseline measurements produced a very high variability perhaps due to warm-up time allowed. We used a 10-minute warm-up before executing these skill tests at baseline. Some studies have found that skills are more consistent with 30 minutes of exercise vs a five or ten-minute warm up (Northcott et al., 1999; Ali et al., 2007a). For instance, in the Northcott et al. (1999) study, participants performed a five minute warm up followed by two x 45 minutes of a simulated soccer game. Soccer skills were measured every 15 minutes during the simulated soccer game. Reported skill performance was better at 30 minutes compared to 0 and 15 minutes. Clearly, participants need to attain a desirable body core and muscle temperature for top skill performance (Ali et al., 2007a). Consequently, it may be that our warm-up time was not long enough prior to the baseline test causing the large variability observed. Of course, this was not a problem with the experimental treatments because the simulated soccer game provided more than adequate warm up. For these reasons, the trials during the baseline measurement of the LSPT were considered practice trials and excluded from the analysis.

### **5.3 Effects on repeated sprint performance (RSA)**

In the present study, there were no differences in any of the parameters measured during the RSA test following the simulated soccer game. This test was employed as a standardized assessment of physical capacity/performance as this ability has been considered key in elite performance (Rampinini et al., 2009). However, in hindsight the

relative energy systems used in these short efforts might have made the test insensitive to detect differences in performance dependent on CHO utilization. Specifically, it is known that anaerobic glycolysis uses CHO as a substrate and, during six-second sprints, supplies ~40% of the total energy needed (Gaitanos et al., 1993). Moreover, this relative contribution has also been reported to decrease by 8-fold from the first to the last sprint of 10, six-second maximal sprints interspersed with 30-second recovery periods (Gaitanos et al., 1993). In the present study, participants did 12x30m (~4.5-second) sprints with 35-second recovery periods, which reflects a similar effort. Although glycogen stores in muscle prior to the test were not measured, it is possible that glycogen was adequate to complete these sprints under both treatments due to the pre-experimental treatment control used, even if treatment differences did exist. Perhaps a test with similar intensity but of longer duration and/or with less recovery time could detect UCAN treatment differences, if they exist. Another factor that can contribute to the decrease in sprint performance is the accumulation of certain metabolites such as inorganic phosphate, hydrogen ions, lactate, etc. For instance, lactate accumulation has been shown to decrease power output in repeated sprint tests (Rampinini et al., 2009). In the present study, blood lactate concentrations were similar with both treatments, so perhaps similar decrements in performance should not be surprising, i.e., accumulation of these metabolites may have impaired performance similarly, regardless of substrate availability.

## 5.4 Limitations

There were a few additional limitations with the present study that need to be considered for future projects. As discussed, the Eriksen-Flanker test presented a considerable amount of between-subject and within-subject variability. Often, in this situation the

recommendation is to increase the sample size to minimize this problem, i.e., prevent a type II error. Studies that only measure cognitive performance with this type of test often use larger sample sizes (up to 500 participants), where variability is much less of a factor. However, it is very challenging to recruit so many to participate in research projects requiring the methods used in the present study. Therefore, attempting to better match participants with similar baseline reaction times prior to the experiment as well as devising cognitive tests that are more consistent are recommended. Finally, muscle glycogen content was not measured during the present study and, should be assessed in future studies.

## 5.5 Future Directions

Based on the current findings, UCAN supplementation appears to augment soccer skill performance and perhaps cognition via increased attentional allocation. Future studies should assess directly whether the present findings are related to differing glycogen use between treatments. Further, to clarify the potential role of UCAN supplementation on cognitive function during exercise other cognitive tests should be utilized in order to measure additional domains of executive function and to minimize measurement variability. Additionally, it would be interesting to test skills from different sports to determine the generalizability of the results of the present study. Finally, although the simulated soccer game protocol replicated the metabolic demands of the sport enabling a fair assessment of the treatments studied, field studies (actual games) are also needed to evaluate the extent of any benefits of UCAN supplementation on both sport skill and cognitive performance when the effort level is selected by the athletes.

## 5.6 Summary/Conclusion

Eleven trained male soccer players performed a simulated soccer game on a treadmill (Desmo Pro, Woodway®, Wisconsin, USA) under two experimental conditions: UCAN and an isoenergetic CHO placebo. Measures of cognitive, physical and soccer skill performance were collected before, throughout and after the simulated soccer game. Interestingly, UCAN supplementation improved both cognitive and soccer skill performance. Specifically, better reaction times, faster dribbling speed and better performance time on the LSPT were observed with UCAN supplementation.

Therefore, it is concluded that UCAN, a hydrothermally modified non-GMO corn starch sport drink, improves some important parameters of soccer performance such as soccer skills and may improve attention allocation during a simulated soccer game. Likely, similar results would be found for other sports requiring intense, repeated, intermittent exercise bouts, i.e., many individual and team sports. It is suggested that differences in exercise substrate use are responsible but future more direct measures are required to confirm this and/or to elucidate any other factors responsible for this ergogenic effect. Finally, to document further how UCAN supplementation may affect cognitive function with intense, repeated exercise other domains of executive function need to be investigated.



## References

- Achten, J., Gleeson, M., and Jeukendrup, A. E. (2002). Determination of the exercise intensity that elicits maximal fat oxidation. *Medicine and Science in Sports and Exercise*. 34(1), 92-97.
- Ahlborg, G., Felig, P., Hagenfeldt, L., Hendler, R., and Wahren, J. (1974). Substrate turnover during prolonged exercise in man. *The Journal of Clinical Investigation*. 53(4), 1080–1090.
- Ali, A., and Williams, C. (2009). Carbohydrate ingestion and soccer skill performance during prolonged intermittent exercise. *Journal of Sports Sciences*. 27, 1499-1508.
- Ali, A., Williams, C., Nicholas, C. W., and Foskett, A. (2007a). The influence of carbohydrate-electrolyte ingestion on soccer skill performance. *Medicine and Science in Sports and Exercise*. 39(2), 1969-1976.
- Ali, A., Williams, C., Hulse, M., Strudwick, A., Reddin, J., Howarth, L., Eldred, J., Hirst, M., and McGregor, S. (2007b). Reliability and validity of two tests of soccer skill. *Journal of sports sciences*. 25(13), 1461-1470.
- American College of Sports Medicine. Guidelines for Exercise Testing and Prescription, 6th Edition. Baltimore, MD: Lippincott Williams & Wilkins, 2000, pp.163–176.
- Amiel, S. A. (1998). Cognitive function testing in studies of acute hypoglycaemia: rights and wrongs? *Diabetologia*. 41, 713–9.
- Anzeneder, C. P., and Bosel, R. (1998) Modulation of the spatial extent of the attentional focus in high-level volleyball players. *European Journal of Cognitive Psychology*. 10(3), 247-267.
- Ashcroft, G. W., Eccleston, D., and Crawford, T. B. (1965). 5-hydroxyindole metabolism in rat brain. a study of intermediate metabolism using the technique of tryptophan loading- I Methods. *Journal of Neurochemistry*. 12, 483-492.
- Baker, L. B., Dougherty, K. A., Chow, M., and Kenney, W. L. (2007). Progressive dehydration causes a progressive decline in basketball skill performance. *Medicine and Science in Sports and Exercise*. 39(7), 1114-1123.
- Baker, L. B., Nuccio, R. P., and Jeukendrup, A. E. (2014). Acute effects of dietary constituents on motor skill and cognitive performance in athletes. *Nutrition Reviews*. 72(12).
- Bangsbo, J. (1994). Energy demands in competitive soccer. *Journal of Sports Sciences*. 12, S5-S12.
- Bangsbo, J., Norregaard, L. and Thorsoe, F. (1991). Activity profile of competition soccer. *Canadian Journal of Sport Sciences*. 16, 110–116.
- Béquet, F., Gomez-Merino, D., Berthelot, M., and Guezennec, C. Y. (2002). Evidence that brain glucose availability influences exercise-enhanced extracellular 5-HT

- level in hippocampus: A microdialysis study in exercising rats. *Acta Physiologica Scandinavica*. 176(1), 65-69.
- Bhattacharya, K., Orton, R. C., Qi, X., Mundy, H., Morley, D. W., Champion, M. P., Eaton, S., Tester, R. F., and Lee, P. J. (2007). A novel starch for the treatment of glycogen storage diseases. *Journal of Inherited Metabolic Disease*. 30(3), 350–357.
- Bledowski, C., Rahm, B., and Rowe, J. B. (2009). What "works" in working memory? Separate systems for selection and updating of critical information. *The Journal of Neuroscience: The Official Journal of the Society for Neuroscience*. 29(43), 13735-13741.
- Bohannon, R. W. (1997). Comfortable and maximum walking speed of adults aged 20-79 years: reference values and determinants. *Age and Ageing*. 26, 15-19.
- Bonadonna, R. C., Groop, L. C., Zych, K., Shank, M., and DeFronzo, R. (1990). Dose-dependent effect of insulin on plasma free fatty acid turnover and oxidation in humans. *The American journal of physiology*. 259(5), E736-50.
- Borg, G. (1973). Perceived exertion: A note on 'history' and methods. *Medicine and Science in Sports and Exercise*. 5, 90-93.
- Bottoms, L. M., Hunter, A. M., and Galloway, S. D. R. (2006). Effects of carbohydrate ingestion on skill maintenance in squash players. *European Journal of Sport Science*. 6(3), 187-195.
- Bottoms, L., Sinclair, J., Taylor, K., Polman, R., and Fewtrell, D. (2012). The effects of carbohydrate ingestion on the badminton serve after fatiguing exercise. *Journal of Sports Sciences*. 30(3), 285-293.
- Carlsson, A., and Lindqvist, M. (1972). The effect of L-tryptophan and some psychotropic drugs on the formation of 5-hydroxytryptophan in the mouse brain in vivo. *Journal of Neural Transmission*. 33(1), 23-43.
- Cermak, N. M., and Van Loon, L. J. C. (2013). The use of carbohydrates during exercise as an ergogenic aid. *Sports Medicine*. 43, 1139-1155.
- Chambers, E. S., Bridge, M. W., and Jones, D. (2009). Carbohydrate sensing in the human mouth: effects on exercise performance and brain activity. *The Journal of Physiology*. 587(Pt 8), 1779-94.
- Claassen, A., Lambert, E. V., Bosch, A. N., Rodger, I. M., St Clair Gibson, A., and Noakes, T. D. (2005). Variability in exercise capacity and metabolic response during endurance exercise after a low carbohydrate diet. *International journal of sport nutrition and exercise metabolism*. 15(2), 97-116.
- Clarke, N. D., Drust, B., Maclaren, D. P., and Reilly, T. (2008). Fluid provision and metabolic responses to soccer-specific exercise. *European Journal of Applied Physiology*. 104(6), 1069-1077.
- Coggan, R., and Coyle, E. F. (1991). Carbohydrate Ingestion During Prolonged Exercise: Effects on Metabolism and Performance. *Exercise & Sport Sciences Reviews*. 19(1), 1-40.

- Coggan, R., and Coyle, E. F. (1987). Reversal of fatigue during prolonged exercise by carbohydrate infusion or ingestion. *Journal of Applied Physiology*. 63(6), 2388-2395.
- Coggan, R., Swanson, S. C., Mendenhall, L., Habash, D. L., and Kien, C. L. (1995). Effect of endurance training on hepatic glycogenolysis and gluconeogenesis during prolonged exercise in men. *The American journal of physiology*. 268(3), E375-E383.
- Collardeau, M., Brisswalter, J., Verduyssen, F., Audiffren, M., and Goubault, C. (2001). Single and choice reaction time during prolonged exercise in trained subjects: Influence of carbohydrate availability. *European Journal of Applied Physiology*. 86, 150-156.
- Cooper, J. R., Bloom, F. E., and Roth, R. H. (2003). The biochemical basis of neuropharmacology. 8th ed. New York: *Oxford University Press*.
- Correia, C. E., Bhattacharya, K., Lee, P. J., Shuster, J. J., Theriaque, D. W., Shankar, M. N., Smit, G. P., and Weinstein, D. (2008). Use of modified cornstarch therapy to extend fasting in glycogen storage disease types Ia and Ib. *American Journal of Clinical Nutrition*. 88(5), 1272-1276.
- Costill, D. L., Coyle, E., Dalsky, G., Evans, W., Fink, W., and Hoopes, D. (1977). Effects of elevated plasma FFA and insulin on muscle glycogen usage during exercise. *Journal of Applied Physiology*. 43, 695-699.
- Coyle, E. F., Coggan, R., Hemmert, M. K., and Ivy, J. L. (1986). Muscle glycogen utilization during prolonged strenuous exercise when fed carbohydrate. *Journal of Applied Physiology*. 61(1), 165-172.
- Currell, K., Conway, S., and Jeukendrup, A. E. (2009). Carbohydrate ingestion improves performance of a new reliable test of soccer performance. *International Journal of Sport Nutrition and Exercise Metabolism*. 19(1), 34-46.
- Curzon, G., Kantamaneni, B. D., Fernando, J. C., Woods, M. S., and Cavanagh, J. B. (1975). Effects of chronic porto-caval anastomosis on brain tryptophan, tyrosine and 5-hydroxytryptamine. *Journal of Neurochemistry*. 24(5), 1065-1070.
- Dahlstroem, A., and Fuxe, K. (1964). Evidence for the existence of monoamine-containing neurons in the central nervous system. I. Demonstration of monoamines in the cell bodies of brain stem neurons. *Acta Physiologica Scandinavica, Supplementum*. 232, 1-55.
- Davis, J. M., and Bailey, S. P. (1997). Possible mechanisms of central nervous system fatigue during exercise. *Medicine and Science in Sports and Exercise*. 29(1), 45-57.
- Davis, J. M., Bailey, S. P., Woods, J. A., Galiano, F. J., Hamilton, M. T., and Bartoli, W. P. (1992). Effects of carbohydrate feedings on plasma free tryptophan and branched-chain amino acids during prolonged cycling. *European Journal of Applied Physiology and Occupational Physiology*. 65(6), 513-519.

- Davranche, K., Hall, B., and McMorris, T. (2009). Effect of acute exercise on cognitive control required during an Eriksen flanker task. *Journal of Sport and Exercise Psychology*. 31(5), 628-639.
- DeStefano, D., and LeFevre, J. (2010). The role of working memory in mental arithmetic. *European Journal of Cognitive Psychology*. 16(3), 353-386.
- Dietrich, A. (2003). Functional neuroanatomy of altered states of consciousness: The transient hypofrontality hypothesis. *Consciousness and Cognition*. 12(2), 231-256.
- Dietrich, A., and Sparling, P. B. (2004). Endurance exercise selectively impairs prefrontal-dependent cognition. *Brain and Cognition*. 55(3), 516-524.
- Di Salvo, V., Gregson, W., Atkinson, G., Tordoff, P., and Drust, B. (2009). Analysis of High Intensity Activity in Premier League Soccer. *International Journal of Sports Medicine*. 30(3), 205-212.
- Dougherty, K. A., Baker, L. B., Chow, M., and Kenney, W. L. (2006). Two percent dehydration impairs and six percent carbohydrate drink improves boys basketball skills. *Medicine and Science in Sports and Exercise*. 38(9), 1650-1658.
- Drust, B., Reilly, T., and Cable, N. T. (2000). Physiological responses to laboratory-based soccer-specific intermittent and continuous exercise. *Journal of sports sciences*. 18(11), 885-892.
- Eccleston, D., Ashcroft, G. W., and Crawford, T. B. (1965). 5-hydroxyindole metabolism in rat brain. a study of intermediate metabolism using the technique of tryptophan loading- II Applications and drug studies. *Journal of Neurochemistry*. 12, 493-503.
- Ekblom, B. (1986). Applied physiology of soccer. *Sports Medicine*. 3, 50-60.
- Eriksen, B.A., and Eriksen, C.W. (1974) Effects of noise letters upon the identification of a target letter in a non-search task. *Perception & Psychophysics*. 16(1), 143-149
- Febbraio, M., Chiu, A., Angus, D. J., Arkinstall, M. J., and Hawley, J. (2000). Effects of carbohydrate ingestion before and during exercise on glucose kinetics and performance. *Journal of Applied Physiology*. 89(6), 2220-2226.
- Fernstrom, J.D. (1983). Role of precursor availability in control of monoamine biosynthesis in brain. *Physiological Reviews*. 63(2), 484-546.
- Fernstrom, J.D., and Wurtman, R.J. (1971) Brain serotonin content: physiological dependence on plasma tryptophan levels. *Science*. 173(3992), 149-152.
- Flynn, M. G., Costill, D. L., Hawley, J. A., Fink, W. J., Neuffer, P. D., Fielding, R. A., and Sleeper, M. D. (1987). Influence of selected carbohydrate drinks on cycling performance and glycogen use. *Medicine and science in sports and exercise*. 19(1), 37-40.
- Freckmann, G., Schmid, C., Baumstark, A., Pleus, S., Link, M., and Haug, C. (2012). System accuracy evaluation of 43 blood glucose monitoring systems for self-

- monitoring of blood glucose according to DIN EN ISO 15197. *Journal of Diabetes Science and Technology*. 6(5), 1060-1075.
- Gaitanos, G., Willians, L., Boobis, L., and Brooks, S. (1993). Human muscle metabolism maximal exercise. *Journal of Applied Physiology*. 75(2), 712- 719.
- Glaister, M., Howatson, G., Pattison, J. R., and McInnes, G. (2008). The reliability and validity of fatigue measures during multiple-sprint work: an issue revisited. *Journal of strength and conditioning research / National Strength & Conditioning Association*. 22(5), 1597-1601.
- Gollnick, P. D., Piehl, K., and Saltin, B. (1974). Selective glycogen depletion pattern in human muscle fibres after exercise of varying intensity and at varying pedalling rates. *The Journal of Physiology*. 241(1), 45-57.
- Hargreaves, M., and Briggs, C. (1988). Effect of carbohydrate ingestion on exercise metabolism. *Journal of Applied Physiology*. 65(4), 1553-1555.
- Hayman, M. (1942). Two minute clinical test for measurement of intellectual impairment in psychiatric disorders. *Archives of Neurology Psychiatry*. 47(3),454-464.
- Hebb, D. O. (1939). Intelligence in man after large removal of cerebral tissue: Report of four left frontal lobe cases. *Journal of General Psychology*. 21, 73–87
- Hetlelid, K. J., Plews, D. J., Herold, E., Laursen, P. B., and Seiler, S. (2015). Rethinking the role of fat oxidation: substrate utilisation during high-intensity interval training in well-trained and recreationally trained runners. *British Medical Journal Open Sport & Exercise Medicine*. 0(1), e000047.
- Holloszy, J. O. (2003) A forty-year memoir of research on the regulation of glucose transport into muscle. *American Journal of Physiology - Endocrinology and Metabolism*. 284(3), E453-E467.
- Hughes, M., and Franks, I. (2005). Analysis of passing sequences, shots and goals in soccer. *Journal of Sports Sciences*. 23(5), 509-514.
- Hultman, E., and Harris, R. C. (1988). Carbohydrate metabolism. In: Poortmans JR, ed. *Principles of Exercise Biochemistry*.
- Jacobs, I. (1981). Lactate, muscle glycogen and exercise performance in man. *Acta Physiologica Scandinavica, Supplementum*. 495, 1-35.
- Jeacocke, N. A., and Burke, L. M. (2010). Methods to standardize dietary intake before performance testing. *International Journal of Sport Nutrition and Exercise Metabolism*. 20, 87-103.
- Jentjens, R., Cale, C., Gutch, C., and Jeukendrup, E. (2003). Effects of pre-exercise ingestion of trehalose, galactose and glucose on subsequent metabolism and cycling performance. *European Journal of Applied Physiology*. 88(4-5), 444-452.
- Jeukendrup, A. E. (2004). Carbohydrate Intake During Exercise and Performance. *Nutrition*. 20, 669–677.

- Jeukendrup, A. E. (2010). Carbohydrate and exercise performance: the role of multiple transportable carbohydrates. *Current Opinion in Clinical Nutrition and Metabolic Care*. 13(4), 452-457.
- Jeukendrup, A. E. (2013). Oral carbohydrate rinse: Placebo or beneficial?. *Current Sports Medicine Reports*. 12(4), 222-227.
- John-Alder, H. B., McAllister, R. M., and Terjung, R. L. (1986). Reduced running endurance in gluconeogenesis-inhibited rats. *American Journal of Physiology - Regulatory, Integrative and Comparative Physiology*. 251(1), R137-R142.
- Kase, S.E., Ritter, F.E., and Schoelles, M. (2009). Serial subtraction errors revealed. In proceedings of the 31st annual conference of the Cognitive Science Society, 1551-1556.
- Katz, A., Broberg, S., Sahlin, K., and Wahren J. (1986). Leg glucose uptake during maximal dynamic exercise in humans. *American Journal of Physiology - Endocrinology and Metabolism*. 251(1), E65-E70.
- Kennedy, D.O., Scholey, A.B. (2000) Glucose administration, heart rate and cognitive performance: effects of increasing mental effort. *Psychopharmacology*. 149, 63-71.
- Kramer, A. F., Humphrey, D. G., Larish, J. F., Logan, G. D., Strayer, D. L., (1994). Aging and inhibition: beyond a unitary view of inhibitory processing in attention. *Psychology and Aging*. 9(4), 491-512.
- Kristiansen, S., Hargreaves, M., and Richter, E. A. (1997). Progressive increase in glucose transport and GLUT-4 in human sarcolemmal vesicles during moderate exercise. *American Journal of Physiology - Endocrinology and Metabolism*. 272(3), E385-E389.
- Lambourne, K., and Tomporowski, P. (2010). The effect of exercise-induced arousal on cognitive task performance: A meta-regression analysis. *Brain Research*. 1341, 12-24.
- Leatt, P. B. (1986). The effect of glucose polymer ingestion on skeletal muscle glycogen depletion during soccer match play and its resynthesis following a match. *Master's thesis*, University of Toronto, Canada.
- Leiper, J. B., Prentice, A. S., Wrightson, C. and Maughan, R. J. (2001). Gastric emptying of a carbohydrate-electrolyte drink during a soccer match. *Medicine and Science in Sports and Exercise*. 33, 1932-1938.
- Mann, D.T.Y., Williams, A.M., Ward, P., and Janelle, C.M. (2007). Perceptual-cognitive expertise in sport: A meta-analysis. *Journal of Sport & Exercise Psychology*. 29, 457-478.
- Meeusen, R., Watson, P., Hasegawa, H., Roelands, B., and Piacentini, M. F. (2006). Central fatigue: The serotonin hypothesis and beyond. *Sports Medicine*. 36(10), 881-909.
- Messier, C. (2004). Glucose improvement of memory: A review. *European Journal of Pharmacology*. 490(1-3), 33-57.

- Mitchell, J. B., Costill, D. L., Houmard, J. A., Fink, W. J., Pascoe, D. D., and Pearson, D. R. (1989). Influence of carbohydrate dosage on exercise performance and glycogen metabolism. *Journal of Applied Physiology*. 67(5), 1843-1849.
- Moseley, L., Lancaster, G. I., and Jeukendrup, A. E. (2003). Effects of timing of pre-exercise ingestion of carbohydrate on subsequent metabolism and cycling performance. *European Journal of Applied Physiology*. 88(4-5), 453-458.
- Newsholme, E.A., Acworth, I.N., and Blomstrand, E. (1987). Amino acids, brain neurotransmitters and a functional link between muscle and brain that is important in sustained exercise. *Advances in Myochemistry*. London, UK. John Libbey Eurotext. pp. 127-138.
- Nicholas, C. W., Nuttall, F. E., and Williams, C. (2000). The Loughborough Intermittent Shuttle Test: a field test that simulates the activity pattern of soccer. *Journal of sports sciences*. 18, 97-104.
- Northcott, S., Kenward, M., Purnell, K. and McMorris, T. (1999). Effect of a carbohydrate solution on motor skill proficiency during simulated soccer performance. *Applied Research in Coaching and Athletics Annual*. 14, 105-118.
- Nybo, L. (2003). CNS fatigue and prolonged exercise: Effect of glucose supplementation. *Medicine and Science in Sports and Exercise*. 35(4), 589-594.
- Nybo, L., and Nielsen, B. (2001). Hyperthermia and central fatigue during prolonged exercise in humans. *Journal of Applied Physiology*. 91(3), 1055-1060.
- Nybo, L., Nielsen, B., Blomstrand, E., Moller, K., and Secher, N. (2003). Neurohumoral responses during prolonged exercise in humans. *Journal of Applied Physiology*. 95, 1125-1131
- Ostojic, S. M., and Mazic, S. (2002). Effects of a carbohydrate-electrolyte drink on specific soccer tests and performance. *Journal of Sports Science and Medicine*. 1, 47-53.
- Pardridge, W. M. (1983). Brain metabolism: a perspective from the blood-brain barrier. *Physiological reviews*. 63(4), 1481-1535.
- Piper, B.J., Li, V., Eiwaz, M.A., Kobel, Y.V., Benice, T.S., Chu, A.M., Olsen, R.H., Rice, D.Z., Gray, H.M., Mueller, S.T., and Raber, J. (2012) Executive function on the Psychology Experiment Building Language tests. *Behavior Research Methods*. 44(1), 110-123.
- Pontifex, M. B., and Hillman, C. H. (2007). Neuroelectric and behavioral indices of interference control during acute cycling. *Clinical Neurophysiology*. 118(3), 570-580.
- Rahnama, N., Reilly, T., Lees, A., and Graham-Smith, P. (2003). Muscle fatigue induced by exercise simulating the work rate of competitive soccer. *Journal of Sports Sciences*. 21(11), 933-942.
- Rampinini, E., Coutts, A. J., Castagna, C., Sassi, R., and Impellizzeri, F. M. (2007). Variation in top level soccer match performance. *International Journal of Sports Medicine*. 28(12), 1018-1024.

- Rampinini, E., Impellizzeri, F. M., Castagna, C., Coutts, A. J., and Wisløff, U. (2009a). Technical performance during soccer matches of the Italian Serie A league: Effect of fatigue and competitive level. *Journal of Science and Medicine in Sport*. 12(1), 227-233.
- Rampinini, E., Sassi, A., Morelli, A., Mazzoni, S., Fanchini, M., and Coutts, A.J. (2009b). Repeated- sprint ability in professional and amateur soccer players. *Applied Physiology Nutrition and Metabolism*. 34, 1048-1054.
- Reilly, T., and Holmes, M. (1983) A preliminary analysis of selected soccer skills. *Physical Education Review*. 61, 64-71.
- Reilly, T., and Thomas, V. (1976). A motion analysis of work-rate in different positional roles in professional football match-play. *Journal of Human Movement Studies*. 2, 87-97.
- Richter, E. A., Ruderman, N. B., Gavras, H., Belur, E. R., and Galbo, H. (1982). Muscle glycogenolysis during exercise: dual control by epinephrine and contractions. *American Journal of Physiology - Endocrinology and Metabolism*. 242(1), E25-E32.
- Roberts, M. D., Lockwood, C., Dalbo, V. J., Volek, J., and Kerksick, C. M. (2011). Ingestion of a high-molecular-weight hydrothermally modified waxy maize starch alters metabolic responses to prolonged exercise in trained cyclists. *Nutrition*. 27, 659-665.
- Roberts, S. P., Stokes, K. A., Trewartha, G., Doyle, J., Hogben, P., and Thompson D. (2010). Effects of carbohydrate and caffeine ingestion on performance during a rugby union simulation protocol. *Journal of Sports Sciences*. 28(8), 833-842.
- Rodriguez, N., Di Marco, N., and Langley, S. (2009). American College of Sports Medicine position stand: nutrition and athletic performance. *Medicine and Science in Sports and Exercise*. 41(3), 709-31.
- Romijn, J. A., Coyle, E. F., Sidossis, L. S., Gastaldelli, A., Horowitz, J. F., Endert, E., and Wolfe, R. R. (1993). Regulation of endogenous fat and carbohydrate metabolism in relation to exercise intensity and duration. *American Journal of Physiology*. 265 (Endocrinol. Metab. 28), E380-E391.
- Russell, M., and Kingsley, M. (2014). The efficacy of acute nutritional interventions on soccer skill performance. *Sports Medicine*. 44, 957-970.
- Russell, M., Benton, D., and Kingsley, M. (2014). Carbohydrate ingestion before and during soccer match play and blood glucose and lactate concentrations. *Journal of Athletic Training*. 49(4), 447-453.
- Russell, M., Benton, D., and Kingsley, M. (2012). Influence of carbohydrate supplementation on skill performance during a soccer match simulation. *Journal of Science and Medicine in Sport*. 15(4), 348-354.
- Salthouse, T. A., and Babcock, R. L. (1991). Decomposing adult age differences in working memory. *Developmental Psychology*. 27, 763-776.



- Saltin, B. (1973). Metabolic fundamentals in exercise. *Medicine and Science in Sports*. 5(3), 137-146.
- SensLab GmbH. (2012) EKF Lactate Scout+ product specification sheet, Leipzig, Germany.
- Siri, W.E. (1961) Body composition from fluid space and density. Techniques for measuring body composition. Washington, DC. *National Academy of Science*. pp. 223-244.
- Smith, E. E., and Jonides, J. (1999). Storage and executive processes in the frontal lobes. *Science*. 283(5408), 1657–61.
- Stellingwerff, T., Boon, H., Gijsen, A. P., Stegen, J. H., Kuipers, H., and Van Loon, L. J. C. (2007). Carbohydrate supplementation during prolonged cycling exercise spares muscle glycogen but does not affect intramyocellular lipid use. *European Journal of Physiology*. 454, 635–647.
- Stone, K.J., and Oliver, J. (2009). The Effect of 45 Min of Soccer-Specific Exercise on the Performance of Soccer Skills. *International journal of sports physiology and performance*. 4(2), 163-175.
- Tenenbaum, G., & Bar-Eli, M (1993). Decision making in sport: A cognitive perspective, pp. 171-192. *Handbook of research on sport psychology*. New York, NY. Macmillan
- Tsintzas, O., Williams, C., Boobis, L., and Greenhaff, P. (1995). Carbohydrate ingestion and glycogen utilization in different muscle fibre types in man. *The Journal of Physiology*. 489(1), 243-250.
- Turner, C. E., Byblow, W. D., Stinear, C. M., and Gant, N. (2014). Carbohydrate in the mouth enhances activation of brain circuitry involved in motor performance and sensory perception. *Appetite*. 80, 212-219.
- Van Gool, D., Van Gervan, D. and Boutmans, J. (1988). The physiological load imposed on soccer players during real match-play. *In Science and Football*. 51–59.
- Van Loon, L. J. C., Jeukendrup, A. E., Saris, W. H. M., and Wagenmakers, A. J. M. (1999). Effect of training status on fuel selection during submaximal exercise with glucose ingestion. *Journal of Applied Physiology*. 87(4), 1413–1420.
- Van Loon, L. J. C., Greenhaff, P. L., Constantin-Teodosiu, D., Saris, W. H. M., and Wagenmakers, A. J. M. (2001). The effects of increasing exercise intensity on muscle fuel utilisation in humans. *Journal of Physiology*. 536(1), 295–304.
- Vergauwen, L., Brouns, F., and Hespel, P. (1998). Carbohydrate supplementation improves stroke performance in tennis. *Medicine and Science in Sports and Exercise*. 30(8), 1289-1295.
- Voss, M.W., Kramer, A.F., Basak, C., Prakash, R.S., and Roberts, B. (2009). Are expert athletes ‘expert’ in the cognitive laboratory? A meta-analytic review of cognition and sport expertise. *Applied Cognitive Psychology*. 24(6), 812-826.

- Welsh, R. S., Waskovich, M., Alderson, N. L., and Davis, J. M. (1997). Carbohydrate and branched-chain amino acid feedings suppress brain 5-HT during prolonged exercise. *Medicine & Science in Sports & Exercise*. 29(5), S192.
- Welsh, R. S., Davis, J. M., Burke, J. R., and Williams, H. G. (2002). Carbohydrates and physical/mental performance during intermittent exercise to fatigue. *Medicine and Science in Sports and Exercise*. 34, 723-731.
- Williams, A.M., and Davids, K. (1998). Visual search strategy, selective attention, and expertise in soccer. *Research Quarterly for Exercise and Sport*. 69(2):111-128.
- Williams, A. M., Davids, K., and Williams J. G. (1999) Visual perception and action in sport. London, UK. E & FN Spon
- Winnick, J. J., Davis, J. M., Welsh, R. S., Carmichael, M. D., Murphy, E. A., and Blackmon, J. A. (2005). Carbohydrate feedings during team sport exercise preserve physical and CNS function. *Medicine and Science in Sports and Exercise*. 37(7), 306-315.
- Yaspelkis, B. B., Patterson, J. G., Anderla, P. A., Ding, Z., and Ivy, J. L. (1993). Carbohydrate supplementation spares muscle glycogen during variable-intensity exercise. *Journal of Applied Physiology*. 75(4), 1477-1485
- Yiannakos, A., and Armatas, V. (2006). Evaluation of the goal scoring patterns in European Championship in Portugal 2004. *International Journal of Performance Analysis in Sport*. 6(11), 178-188.
- Zeef, E. J., Sonke, C. J., Kok, A., Buiten, M. M., and Kenemans, J. L., (1996). Perceptual factors affecting age-related differences in focused attention: Performance and psychophysiological analyses. *Psychophysiology*. 33, 555-65.
- Zisman, A., Peroni, O. D., Abel, E. D., Michael, M. D., Mauvais-Jarvis, F., Lowell, B. B., Wojtaszewski, J. F., Hirshman, M. F., Virkamaki, A., Goodyear, L. J., Kahn, C. R., and Kahn, B. B. (2000). Targeted disruption of the glucose transporter 4 selectively in muscle causes insulin resistance and glucose intolerance. *Nature Medicine*. 6, 924 - 928

# Appendices

## Appendix A. Human Ethics Approval



Research Ethics

### Western University Health Science Research Ethics Board HSREB Full Board Initial Approval Notice

**Principal Investigator:** Dr. Peter Lemon  
**Department & Institution:** Health Sciences\Kinesiology, Western University

**Review Type:** Full Board  
**HSREB File Number:** 107705  
**Study Title:** The effects of starch produced from heat-treated corn (Superstarch) supplementation on skills, cognitive function and physical performance during/following a simulated soccer game

**HSREB Initial Approval Date:** March 21, 2016  
**HSREB Expiry Date:** March 21, 2017

**Documents Approved and/or Received for Information:**

Document Name	Comments	Version Date
Data Collection Form/Case Report Form	health information form	2015/08/25
Instruments	RPE scale	2015/08/20
Instruments	PAR-Q	2015/08/20
Advertisement	Ad Poster	2016/01/28
Western University Protocol	Western Protocol Superstarch & Soccer	2016/02/26
Letter of Information & Consent	LOI & Consent Form SS & Soccer Study	2016/02/26
Other	Email Recruitment Script	2016/02/26

The Western University Health Science Research Ethics Board (HSREB) has reviewed and approved the above named study, as of the HSREB Initial Approval Date noted above.

HSREB approval for this study remains valid until the HSREB Expiry Date noted above, conditional to timely submission and acceptance of HSREB Continuing Ethics Review.

The Western University HSREB operates in compliance with the Tri-Council Policy Statement Ethical Conduct for Research Involving Humans (TCPS2), the International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use Guideline for Good Clinical Practice Practices (ICH E6 R1), the Ontario Personal Health Information Protection Act (PHIPA, 2004), Part 4 of the Natural Health Product Regulations, Health Canada Medical Device Regulations and Part C, Division 5, of the Food and Drug Regulations of Health Canada.

Members of the HSREB who are named as Investigators in research studies do not participate in discussions related to, nor vote on such studies when they are presented to the REB.

The HSREB is registered with the U.S. Department of Health & Human Services under the IRB registration number IRB 00000940.

Ethics Officer, on behalf of Dr. Joseph Gilbert, HSREB Chair

Ethics Officer to Contact for Further Information: Erika Basile \_\_\_ Katelyn Harris \_\_\_ Nicole Kanaki \_\_\_ Grace Kelly \_\_\_ Vikki Tran \_\_\_

*This is an official document. Please retain a copy for your files.*

Western University, Research, Support Services Bldg., Rm. 5150  
London, ON, Canada

## Appendix B. Letter of Information and Informed Consent



Title of Study: The effects of starch produced from heat-treated corn (Superstarch) supplementation on skills, cognitive function and physical performance during/following a simulated soccer game

Principal Investigator: Peter W.R. Lemon (PhD)  
Graduate Student: Manuel Quinones (B.ASc.)

Exercise Nutrition Research Laboratory (Room 2235 – 3M Centre)  
School of Kinesiology, Western University.

### LETTER OF INFORMATION AND CONSENT

#### INVITATION TO PARTICIPATE

You are being invited to participate in a research study at the Exercise Nutrition Research Laboratory (Room 2235, 3M Centre) investigating the effects of a heat modified corn starch supplementation (Superstarch) on skills, cognitive function and physical performance during and following a simulated soccer game.

#### PURPOSE OF THE LETTER

The purpose of this letter is to provide you with information required for you to make an informed decision regarding participation in this research.

#### PURPOSE OF THIS STUDY

In team sports of moderate to long duration such as soccer, fatigue occurs toward the end of the event and is characterized by a drop in work rate. This drop in work rate may be accompanied by impairment in thinking (cognition) that leads to deterioration in technical skills such as passing, shooting or dribbling. Often, athletes and coaches in team sports attribute the final outcome of a game to mental mistakes that happen in the latter stages of a game. In light of this, nutrient supplementation strategies to avoid or delay the onset of fatigue have been of considerable interest to many. Superstarch is a carbohydrate (CHO) drink that due to its slow rate of absorption into your blood causes a relatively small release of insulin in blood, which helps spare body CHO stores by promoting the use of fats for fuel. This may delay the onset of fatigue. Heat treatment of corn starch is a novel processing technique intended to help children with a rare genetic disorder called glycogen storage disease. This condition is characterized by failure to be able to use stored CHO resulting in decreased blood glucose. Some

01/18/2016

\_\_\_\_\_ initials

1 of 8

studies have shown this treatment to be better than conventional treatments for prevention of low blood glucose. Moreover, it has also been shown to enhance endurance exercise performance. However, whether the effects of this treatment are similar during high intensity intermittent exercise, like performed in a soccer game, is still unknown. Therefore, the purpose of this study is to assess whether Superstarch can enhance physical or cognitive performance during and following a simulated soccer game.

#### INCLUSION CRITERIA

In order to be eligible to participate in this study you must be a healthy male or female elite soccer player, involved in regular training (3-5 x week), and 15 to 35 year old.

#### EXCLUSION CRITERIA

You will be excluded from this study if you:

- Are not involved on regular soccer training (at least 3x week)
- Have symptoms or take medication for respiratory, cardiovascular, metabolic, neuromuscular disease
- Been diagnosed with a cognitive impairment and/or learning disability
- Use any medications with side effects of dizziness, lack of motor control, or slowed reaction time
- For women, if you are pregnant or become pregnant during the study.
- Have a history of concussion/head injuries.
- Have an excessive alcohol intake (>2 drinks/day)

#### STUDY OUTLINE:

This is a double blind (meaning neither the participants nor the investigators will know which treatment is which until the study is completed), placebo (sugar) - controlled study, i.e., there will be two treatments studied (sugar and Superstarch) with a total of 12 subjects. All participants will complete both treatments on separate days and all study activities will be completed in the Exercise Nutrition Research Laboratory (Room 2235, 3M Centre) or in the Alumni Hall Gym. **\*\*\*For the test days, you will need to arrive at the laboratory at 7:30 am following an overnight fast (no food or drink except water after 7:30pm). Further, you will need to refrain from exercise and from consuming caffeine or alcohol for 24 hours prior to the two simulated soccer games. You will be given a standardized pre-fast meal of ~10 kcals/kg containing 1g/kg of carbohydrate (two hamburgers, potatoes, lettuce and apple juice [250 ml]) to ensure you will have the same energy for both testing days \*\*\***

#### STUDY PROCEDURES

If you volunteer to participate in this study, you will need to do the following things: Visit the lab for two practice sessions and one baseline session prior to the experimental treatment days. The practice sessions will be held within the same week and the baseline session will be held no less than one week before the initial treatment day. The first session will involve filling out forms to ensure your safe participation in the study, a measurement of body composition, a maximum oxygen

consumption test and a sprint test to determine the treadmill speeds to be used during study days (details are below). This will require about 60-70 minutes. The second practice session will be used to acclimate you to the running exercise intensities and to practice the cognitive and soccer skill tests. This will require less than 60 minutes. The third session before the experimental treatment days will be to collect baseline scores on skill tests. This session will be ~30 minutes.

2. For the experimental treatment days you visit the laboratory on two additional occasions. Both will involve the identical exercise bout (simulated soccer game) and the consumption of an experimental drink (a drink containing glucose or containing Superstarch – both will have equal amounts of energy) at two times (30 minutes before starting the simulated games and ~50 minutes later, i.e., half time). The order of these two experimental treatments will be randomized, i.e., like a coin flip, and you will complete both trials. The exercise session for both study days will consist of a 60 minute simulated soccer game divided into 4 x 15-minute blocks of treadmill running at varying intensities that replicate the activity pattern of soccer such as sprinting, jogging, cruising, walking etc. Subsequently, you will perform a repeated sprint ability test (RSA) followed by two skill tests (described below).
3. We will measure cognitive function and breath samples within each block during the 60 minute running exercise.
4. We will also measure blood glucose and blood lactate before ingesting the treatment drinks, before starting the game, every 15 minutes, i.e., at the end of each exercise block, and post-exercise. Therefore, we will collect a total of 7 samples during each study visit for a total of 14 samples over a 2-week period. Each sample is a drop of blood from a finger prick with a sterile lancet. These samples will be discarded right after the analysis is completed.

**Practice sessions: Before your inclusion in the study sessions,** you will be asked to fill out a physical activity readiness questionnaire and a participant information form for personal and familial health history. During the first practice session you will have your body composition determined via BodPod® and maximal oxygen consumption test on a treadmill. The BodPod® is a chamber which determines body volume by measuring the space your body takes up and together with your body weight allows us to calculate lean and fat content of the body. The maximal oxygen consumption test involves a continuous, incremental running test on a treadmill (0.1 miles per hour per 10 seconds, starting at 6mph) until you reach volitional fatigue, i.e., when you step off the treadmill. Ending the test is not difficult and you will be given time to practice prior to the test. The sprint test involves a 15 second treadmill sprint to determine your peak running speed. For safety reasons, this is completed with the treadmill in manual mode meaning you provide the treadmill belt movement so it slows down as you do. During the second practice session you will complete one 15-minute block of treadmill running to allow you to become familiar with the running intensities that will be used during the simulated soccer game and you will have an opportunity to practice the cognitive and soccer skill tests to minimize any learning effect on the treatment days.

**Baseline session:** After performing a standardized 10 minute warm-up (~6mph), you will be scored on both soccer skill tests. For each test, you will perform 4 attempts.

**Test day 1 (Total duration is approximately 4 hours – For overview see Experimental Protocol Figure (below):**

- The evening before the experiment you will visit the lab to pick up dinner (two hamburgers, potatoes, lettuce and apple juice [250 ml]) so you will be well fuelled for the simulated soccer game.
- Arrive at Rm 2235 3M Centre at 7:30 am with limited prior physical activity (drive/use of the elevator to get to the lab) and having consumed no food or drink, except water, since 7:30pm the night before. This strategy is used to ensure that you are in a rested physiological state.
- At 7:35 am, the first fingerpick for blood lactate and blood glucose samples will be collected.
- At 7:40 am, we will collect baseline data for the cognitive tests (serial seven subtraction and flanker tests). You will attempt each test 3 times.
- At 7:50 am, we will provide one of the treatment drinks and you will be allowed time to read/study while the drink is digested. The drink is an 8% CHO solution containing 0.5 g CHO per kg body weight and some electrolytes typical of sports drinks (94 mg per litre of magnesium, 282 mg per litre of chloride, 563 mg per litre of sodium, 188 mg per litre of potassium and 28 mg per litre of calcium).
- At 8:20 am, you will perform a 10 minute standardized treadmill warm-up.
- At 8:30 am, the second fingerpick blood lactate and blood glucose samples will be collected, followed by initiation of the first 15 minute exercise block. During this block, you will perform both the serial seven subtraction and flanker tests. Also, expired breath samples will be collected during the last 4 minutes to estimate your carbohydrate and fat use.
- At 8:50 am, the first exercise block is completed. You will be given 4 minutes of passive rest (sitting) and the third fingerpick blood lactate and blood glucose samples will be collected.
- At 8:54 am, the second 15-minute exercise block will start. During this block, you will perform the similar serial seven subtraction and flanker tests. Also, breath samples will be collected during the last 4 minutes.
- At 9:09 am, the second exercise block will end. You will be given 4 minutes of sitting rest and the fourth fingerpick blood lactate and blood glucose samples will be collected.
- At 9:13 am, the third 15-minute exercise block will start. During this block, you will again perform the serial seven subtraction and flanker tests. Further, breath samples will be collected during the last 4 minutes.
- At 9:28 am, the third exercise block will end. You will be given 15 minutes (half time) of sitting rest and the fifth fingerpick blood lactate and blood glucose samples will be collected. During this time a second drink will be provided. The drink is an 8% CHO solution this time containing 0.2 g CHO per kg body weight as well as the electrolyte mixture described above.
- At 9:43 am, the fourth 15-minute exercise block will commence. During this block, you will perform the serial seven subtraction and flanker tests. Also, breath samples will be collected during the last 4 minutes.

- At 9:58 am, the fourth exercise block will end and the sixth fingerpick blood lactate and blood glucose samples will be collected.
- At 10:03 am, Jog to the Alumni Hall Gym.
- At 10:07 am, the repeated sprint ability test (RSA) begins.
- At 10:15 am, end of the RSA. You will be given 5 minutes of sitting recovery before starting the first skill test.
- At 10:20 am, beginning of the Slalom Dribble test.
- At 10:25 am, the Slalom Dribble test ends and the final fingerpick blood lactate and blood glucose samples will be collected.
- At 10:30 am, the LSPT (passing test) starts.
- At 10:40 am, the first experimental treatment day ends.

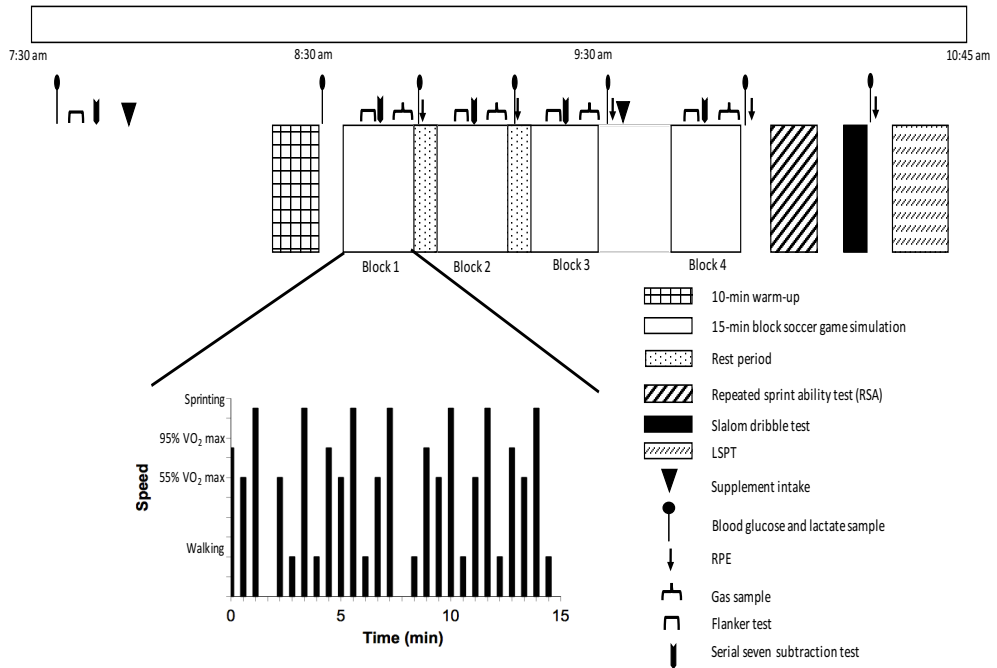


Figure – Experimental Protocol



**Test day 2 (Total duration is approximately 4 hours):** This visit will take place at least one week after the first study visit. You will perform the same study procedures you did during the first study visit except with the other experimental treatment drink.

#### POSSIBLE RISK AND HARMS

Both treatments to be studied are available commercially for several years and there are no known risks to their ingestion. This study does involve strenuous exercise that may pose a risk of minor muscle injury, discomfort or soreness. Further, all exercise involves some health risk (primarily cardiovascular or hydration-related) and you may experience symptoms of fatigue while participating in this study. Importantly, similar exercise to that used in this study is completed daily on campus by kinesiology students and Mustang athletes. Further, the risks of cardiovascular complications are much reduced in young, healthy, physically trained individuals. Importantly, research has determined that regular exercise is safer than no exercise with respect to a wide variety of health markers. Finally, to avoid dehydration concerns you will be encouraged to consume adequate fluids to replace sweat losses.

**Maximal oxygen consumption test:** you may experience muscle fatigue, discomfort, dizziness and/or nausea.

**Blood Collection:** The fingerprick method may result in some bleeding or bruising; however, this risk is low because we use pressure on the puncture site and a bandaid, where necessary. Infection is possible but extremely rare as sterile technique will be used.

#### POTENTIAL BENEFITS

There is no direct benefit to you from participating in this study but you will have access to some information about your body composition, exercise capacity, and perhaps some strategies to maximize performance through nutrition interventions. You will also receive the experimental results (mean±SD) once everything is completed, if you request them.

#### COMPENSATION

You will not be compensated for your participation in this study nor will you be reimbursed for any additional costs incurred such as parking or transportation.

#### VOLUNTARY PARTICIPATION

Participation in this study is voluntary. You may refuse to participate, refuse to answer any questions or withdraw from the study at any time without a penalty of any kind. The investigator may withdraw you from this research if circumstances arise which warrant doing so.

#### RIGHTS OF A PARTICIPANT (in the event of a study related injury)

If you suffer any study related injury during your participation in this study care will be provided to you at no cost.

**CONFIDENTIALITY**

If you agree to join this study, only members of the study team will look at your personal information (e.g., name, experimental results, etc) and only the information they need for the study will be collected.

Further, all information that is collected for the study will be coded so you cannot be identified and kept in a secure area (locked in a file cabinet or on a University server behind a fire wall) by the study doctor for 5 years. Only the study team or the people or groups listed below will be allowed to look at your records. Identifiable data will only be kept for 5 years, whereas de-identified data will be kept indefinitely on a kinesiology server. These data will be in numerical form only. No personal identifiers will be present. And these data may be compared to the results of future similar studies.

Representatives of the University of Western Ontario Health Sciences Research Ethics Board may look at the study records and at your personal health information to check that the information collected for the study is correct and to make sure the study followed proper laws and guidelines.

All information collected during this study, including your personal health information, will be kept confidential and will not be shared with anyone outside the study unless required by law. You will not be named in any reports, publications, or presentations that may come from this study.

If you decide to leave the study, the information about you that was collected before you leave the study will still be used in order to answer the research question. No new information will be collected without your permission.

**Consent Form**

The effects of starch produced from heat-treated corn (Superstarch) supplementation on skills, cognitive function, and physical performance during/following a simulated soccer game

Investigators: Dr. Peter W.R. Lemon and Manuel Quinones, B.ASc.

I have read the accompanying "Letter of Information", have had the nature of the study explained to me and I agree to participate. All questions have been answered to my satisfaction.

If you wish to participate in future studies in the Exercise Nutrition Research Lab, the research team will collect your contact information.

I wish to be contacted for future studies in the Exercise Nutrition Research Laboratory.

Yes \_\_\_\_\_ (check mark)      No \_\_\_\_\_ (check mark)      Date: \_\_\_\_\_

By signing below, I agree to participate in this study.

Name of Participant (please print): \_\_\_\_\_

Signature of Participant: \_\_\_\_\_ Date: \_\_\_\_\_

Signature of Parent if participant under 18 years \_\_\_\_\_ Date: \_\_\_\_\_

Name of Person Obtaining Informed Consent: \_\_\_\_\_

Signature of Person Obtaining Informed Consent: \_\_\_\_\_

Date: \_\_\_\_\_

You will receive a copy of the consent form after it has been signed. You do not waive any legal rights by signing the consent form.

This letter is for you to keep for future reference.

Sincerely,

Dr. Peter Lemon

Manuel Quinones, B.ASc.

## Appendix C. Participant Health Information Form

### Participant Health Information Form

Participant I.D.: \_\_\_\_\_ Date: \_\_\_\_\_

Age: \_\_\_\_\_ Height: \_\_\_\_\_ Weight: \_\_\_\_\_ Smoker: **Yes / No**

Ethnic Background: \_\_\_\_\_

#### Medical History (please check any and all that apply)

Family history of heart disease	<input type="checkbox"/>	Endocrine disorder	<input type="checkbox"/>
Heart murmur	<input type="checkbox"/>	Raynaud's syndrome	<input type="checkbox"/>
Phlebitis	<input type="checkbox"/>	Polycystic ovary syndrome	<input type="checkbox"/>
Other heart disorder (please specify)	<input type="checkbox"/>	Seizures	<input type="checkbox"/>
_____		Digestive/gastrointestinal problems	<input type="checkbox"/>
Family history of stroke	<input type="checkbox"/>	Asthma	<input type="checkbox"/>
Migraines	<input type="checkbox"/>	Bronchitis	<input type="checkbox"/>
Sinus problems	<input type="checkbox"/>	Other respiratory disorder (please specify)	<input type="checkbox"/>
Hypertension	<input type="checkbox"/>	_____	
Diabetes	<input type="checkbox"/>		

Have you ever fainted? **Yes / No**

If yes, under what circumstances:

\_\_\_\_\_

Are you taking any medications? **Yes / No**

If yes, please specify: \_\_\_\_\_

Have you had any major surgeries, illnesses or injuries? **Yes / No**

If yes, please specify (include dates): \_\_\_\_\_

Have you had a concussion or serious head injury? **Yes / No**

If yes, please specify (include dates): \_\_\_\_\_

Do you consume alcohol or any caffeinated beverages on a regular basis? **Yes / No**

If yes, please specify the quantity: \_\_\_\_\_

Have you ever been diagnosed with a learning impairment? **Yes / No**

If yes, please specify: \_\_\_\_\_

Do you have any food allergies? **Yes / No**

If yes, please specify \_\_\_\_\_

Do you consume any dietary supplements (including protein powder)? **Yes / No**

If yes, please specify the type, frequency, and typical daily dose:

\_\_\_\_\_

## Appendix D. Physical Activity Readiness Questionnaire

Physical Activity Readiness  
Questionnaire - PAR-Q  
(revised 2002)

# PAR-Q & YOU

(A Questionnaire for People Aged 15 to 69)

Regular physical activity is fun and healthy, and increasingly more people are starting to become more active every day. Being more active is very safe for most people. However, some people should check with their doctor before they start becoming much more physically active.

If you are planning to become much more physically active than you are now, start by answering the seven questions in the box below. If you are between the ages of 15 and 69, the PAR-Q will tell you if you should check with your doctor before you start. If you are over 69 years of age, and you are not used to being very active, check with your doctor.

Common sense is your best guide when you answer these questions. Please read the questions carefully and answer each one honestly: check YES or NO.

YES	NO	
<input type="checkbox"/>	<input type="checkbox"/>	1. Has your doctor ever said that you have a heart condition <u>and</u> that you should only do physical activity recommended by a doctor?
<input type="checkbox"/>	<input type="checkbox"/>	2. Do you feel pain in your chest when you do physical activity?
<input type="checkbox"/>	<input type="checkbox"/>	3. In the past month, have you had chest pain when you were not doing physical activity?
<input type="checkbox"/>	<input type="checkbox"/>	4. Do you lose your balance because of dizziness or do you ever lose consciousness?
<input type="checkbox"/>	<input type="checkbox"/>	5. Do you have a bone or joint problem (for example, back, knee or hip) that could be made worse by a change in your physical activity?
<input type="checkbox"/>	<input type="checkbox"/>	6. Is your doctor currently prescribing drugs (for example, water pills) for your blood pressure or heart condition?
<input type="checkbox"/>	<input type="checkbox"/>	7. Do you know of <u>any other reason</u> why you should not do physical activity?

**If  
you  
answered**

### YES to one or more questions

Talk with your doctor by phone or in person BEFORE you start becoming much more physically active or BEFORE you have a fitness appraisal. Tell your doctor about the PAR-Q and which questions you answered YES.

- You may be able to do any activity you want — as long as you start slowly and build up gradually. Or, you may need to restrict your activities to those which are safe for you. Talk with your doctor about the kinds of activities you wish to participate in and follow his/her advice.
- Find out which community programs are safe and helpful for you.

### NO to all questions

If you answered NO honestly to all PAR-Q questions, you can be reasonably sure that you can:

- start becoming much more physically active — begin slowly and build up gradually. This is the safest and easiest way to go.
- take part in a fitness appraisal — this is an excellent way to determine your basic fitness so that you can plan the best way for you to live actively. It is also highly recommended that you have your blood pressure evaluated. If your reading is over 144/94, talk with your doctor before you start becoming much more physically active.

### DELAY BECOMING MUCH MORE ACTIVE:

- if you are not feeling well because of a temporary illness such as a cold or a fever — wait until you feel better; or
- if you are or may be pregnant — talk to your doctor before you start becoming more active.

**PLEASE NOTE:** If your health changes so that you then answer YES to any of the above questions, tell your fitness or health professional. Ask whether you should change your physical activity plan.

**Informed Use of the PAR-Q:** The Canadian Society for Exercise Physiology, Health Canada, and their agents assume no liability for persons who undertake physical activity, and if in doubt after completing this questionnaire, consult your doctor prior to physical activity.

**No changes permitted. You are encouraged to photocopy the PAR-Q but only if you use the entire form.**

NOTE: If the PAR-Q is being given to a person before he or she participates in a physical activity program or a fitness appraisal, this section may be used for legal or administrative purposes.

"I have read, understood and completed this questionnaire. Any questions I had were answered to my full satisfaction."

NAME \_\_\_\_\_

SIGNATURE \_\_\_\_\_

DATE \_\_\_\_\_

SIGNATURE OF PARENT \_\_\_\_\_

WITNESS \_\_\_\_\_

or GUARDIAN (for participants under the age of majority)

**Note: This physical activity clearance is valid for a maximum of 12 months from the date it is completed and becomes invalid if your condition changes so that you would answer YES to any of the seven questions.**

**Appendix E. Ratings of Perceived Exertion (RPE) Scale**

Rating	Perception of effort
6	
7	Very, very light
8	
9	Very light
10	
11	Fairly light
12	
13	Somewhat hard
14	
15	Hard
16	
17	Very hard
18	
19	Very, very hard
20	

From Borg (1973, p. 92). © by Lippincott, Williams & Wilkins. Adapted by permission.

## Appendix F. Loughborough Soccer Passing Test (LSPT) Trial Order Sheet

### LSPT Trials

1	2	3	4	5	6	7
Green	Red	Green	Blue	Green	Yellow	Red
Red	Blue	Blue	Red	Yellow	Blue	Green
Yellow	Yellow	Red	Yellow	Blue	Green	Yellow
Blue	Blue	Yellow	Blue	Green	Blue	Blue
Yellow	Green	Green	Green	Red	Red	Yellow
Green	Yellow	Blue	Red	Blue	Green	Green
Blue	Green	Yellow	Yellow	Red	Yellow	Blue
Yellow	Red	Green	Red	Green	Red	Red
Red	Blue	Red	Blue	Yellow	Blue	Blue
Green	Yellow	Green	Green	Green	Green	Red
Yellow	Red	Yellow	Yellow	Blue	Yellow	Green
Blue	Blue	Blue	Red	Yellow	Green	Yellow
Red	Green	Red	Green	Red	Yellow	Red
Green	Red	Yellow	Yellow	Yellow	Red	Yellow
Blue	Yellow	Red	Blue	Blue	Blue	Green
Red	Green	Blue	Green	Red	Red	Blue
8	9	10	11	12	13	14
Red	Green	Yellow	Yellow	Blue	Red	Green
Blue	Yellow	Red	Red	Green	Yellow	Yellow
Red	Green	Blue	Blue	Red	Red	Blue
Green	Yellow	Yellow	Red	Blue	Green	Yellow
Yellow	Red	Blue	Green	Red	Yellow	Green
Blue	Yellow	Red	Yellow	Green	Blue	Blue
Yellow	Green	Yellow	Green	Yellow	Green	Red
Green	Blue	Green	Blue	Red	Blue	Green
Yellow	Green	Red	Red	Green	Yellow	Red
Red	Red	Green	Green	Yellow	Green	Yellow
Blue	Blue	Blue	Blue	Blue	Red	Blue
Yellow	Red	Red	Yellow	Red	Blue	Green
Red	Blue	Yellow	Blue	Green	Red	Red
Green	Yellow	Green	Yellow	Yellow	Blue	Yellow
Blue	Red	Blue	Green	Blue	Yellow	Red
Green	Blue	Green	Red	Yellow	Green	Blue

## Curriculum Vitae

**Name:** Manuel Quinones

**Post-secondary Education and Degrees:** Fanshawe College  
London, Ontario, Canada  
2010-2012

University of Guelph-Humber  
Toronto, Ontario, Canada  
2012-2014 B.A.Sc

The University of Western Ontario  
London, Ontario, Canada  
2014-2016 M.Sc.

**Honours and Awards:** Lippincott Williams & Wilkins/Wolters Kluwer Health Book Prize  
for outstanding undergraduate abstract  
OEP (Ontario Exercise Physiology) 2014

Western Research Graduate Scholarship  
2014-2015, 2015-2016

**Related Work Experience** Teaching Assistant  
The University of Western Ontario  
2014-2016