

Research Article

The Effect of Oligosaccharides in Sports Drink on Exercise Performance and Endurance Time of Athletes

Mi Dong

Jilin Agricultural University, China

Abstract: This study aims to investigate the effect of oligosaccharides in sports drink on high-intensity exercise capacity in athletes. In our research, sixteen healthy male students of Institute of Physical Education were randomly divided into Oligosaccharide Replenishment (OR) group (n = 8) and control group (n = 8). Their max oxygen uptake were measured by 2900 cardiorespiratory function meter, then they all participated in a cycling test at the intensity of 80% oxygen consumption max until fatigue. The results showed that blood glucose (mmol/L) in the OR group was higher than that in the control group during exercise, the difference between the two groups was more significant along with time (5.29 ± 1.07 - 6.46 ± 0.78 and 4.71 ± 0.84 - 5.36 ± 0.80 , $p < 0.05$). 30 min after the exercise, the change of blood lactate in the OR group was higher than that in the control group both during (6.69 ± 1.57 - 7.39 ± 1.19) and after (6.18 ± 1.28 - 5.19 ± 1.14) the exercise ($p < 0.05$). Cycling time [(47±12) min] and work load [(174.9±3.6) kJ] in the OR group were significantly higher than those in the control group ($p < 0.05$). From these results, we can see that oligosaccharide-replenishing is good for keeping blood glucose level for a long time in high-intensity exercise and prolong the time of high-intense exercise.

Keywords: Athletes, blood glucose, blood lactate, oligosaccharides, sports drink

INTRODUCTION

As we have known, oligosaccharides are carbohydrates which have 3-10 simple sugars linked together. They are found naturally, at least in small amounts, in many plants. Plants with large amounts of oligosaccharides include chicory root, from which most commercial inulin is extracted and so-called Jerusalem artichokes (the root of a member of the sunflower family). They are also found in onions (and the rest of the "onion family", including leeks and garlic), legumes, wheat, asparagus, jicama and other plant foods. It is estimated that North Americans get about 1-3 grams naturally in their diets each day, while Europeans get 3-10 g (Hargreaves *et al.*, 2004). Most oligosaccharides have a mildly sweet taste and have certain other characteristics, such as the mouth feel they lend to food, that has drawn the interest of the food industry as a partial substitute for fats and sugars in some foods as well as improved texture. Because of this, more and more of the oligosaccharides in food are synthetically produced.

Oligosaccharides are synthesized by adding one sugar at a time onto a growing chain. Information-rich oligosaccharides, such as the ABO antigens, are made of several different sugars and are linked in a variety of ways to other sugars, so each sugar is added by a different enzyme (Carter *et al.*, 2003). The sugar-adding-enzymes are called glycosyl transferases. Similar to other macromolecular polymerizations e.g., protein

and nucleic acid synthesis, the monomer sugars are first activated by bonding a Nucleotide Phosphate (typically UDP) and the sugar is transferred from this activated intermediate to the growing sugar chain, in this case the H antigen.

Athletes benefit from the amount of oligosaccharides stored in the body when they participate in high-intensity exercises. In the early stages of moderate exercise, oligosaccharides provide 20 to 25% of the energy requirement. Oligosaccharides yield more energy per unit of oxygen consumed than fats. Because oxygen often is the limiting factor in long duration events, it is beneficial for the athlete to use the energy source requiring the least amount of oxygen per kilocalorie produced (Jentjens and Jeukendrup, 2002). As work intensity increases, oligosaccharide utilization increases. During exercise, the glycogen is converted back to glucose and is used for energy. The ability to sustain prolonged vigorous exercise is directly related to initial levels of muscle glycogen. The body stores a limited amount of oligosaccharide in the muscles and liver. If the event lasts for less than 90 minutes, the glycogen stored in the muscle is enough to supply the needed energy. Extra oligosaccharides will not work, any more than adding gas to a half-full tank will make the car go faster. Complex oligosaccharides come from foods such as spaghetti, potatoes, lasagna, cereals and other grain products (Jentjens *et al.*, 2004). Simple oligosaccharides are found in fruits, milk, honey and sugar. During digestion, the body breaks down

oligosaccharides to glucose and stores it in the muscles as glycogen (Madsen *et al.*, 1996).

Recent interest has also been drawn to oligosaccharides from the nutritional community because of an important characteristic: the human digestive system has a hard time breaking down many of these oligosaccharides. Almost 90% escapes digestion in the small intestine and reaches the colon where it performs a different function: that of a prebiotic (Angus *et al.*, 2000). In this study, we focus our attention on the study of the impact of oligosaccharides in sports drink on the blood glucose, blood lactate and athletic abilities of athletes.

MATERIALS AND METHODS

Materials: We select 16 healthy male students of Institute of Physical Education and then randomly divide them into two groups: oligosaccharide replenishment (OR) group and control group (each group has 8 members). The average age of these students is (21.8±0.9). The ranges for their heights and weights are (177±5) cm and (70±8) cm, respectively. The maximal oxygen uptake is (29±0.2) L/min.

Research methods: These students must to drain all the urine before every experiment and what's more, they should reach the laboratory 30 min earlier to measure the weight of body, blood glucose and blood lactate. Besides, the bike for test must be adjusted in advance and every student should pedal the bike for 3 min with no load. Then, the load starts with 100 W and increases with the speed of 25 W per minute until the oxygen uptake is 80% the maximal value. All students pedal the bike until they have no power to operate.

The oligosaccharides used in our research are provided by Beijing Sports Nutrition Center. Before testing, the oligosaccharides are mixed with water to be an aqueous solution with the concentration of 80 g/L. The oligosaccharide replenishment group is replenished with 200 mL sweet water 5 min before cycling and with 150 mL sweet water per 20 min in the cycling. However, the control group is provided with the same capacity of pure water. All students' diets are arranged by the laboratory.

The blood glucose (before cycling, in cycling and after cycling) is measured by ADVANTAGE; the blood lactate is measured by YSI 1 500 SPORTS and the rhythm of the heart is measured by POLAR. The maximal oxygen uptake and the sport time are measured by 2900 CORTEX. All the data is analyzed with SPSS 10.0 and $p < 0.05$ means significance of difference.

RESEARCH RESULTS

The Change of Blood Glucose and Blood Lactate: The deference of blood glucose and blood lactate between the OR group and the control group is not significant in statistics before cycling ($p > 0.05$). After cycling 20 min, the blood glucose and blood lactate of

Table 1: The change of blood glucose (Unit: mmol/L)

	OR Group	Control group
Number of students	8	8
Before cycling	5.75±0.81	5.76±0.71
Cycling for 20 min	4.71±0.84	5.29±1.07
Cycling for 40 min	4.42±2.34	5.56±2.53
Completely exhausted	5.36±0.80	6.46±0.78

Table 2: The change of blood lactate (Unit: mmol/L)

	OR Group	Control group
Number of students	8	8
Before cycling	0.89±0.12	0.95±0.82
Cycling for 20 min	5.33±1.12	6.69±1.57
Cycling for 40 min	8.79±1.06	6.81±1.38
Completely exhausted	10.12±1.67	7.39±1.19

the OR group are higher than that of the control group, while the deference of these two indices is still not significant in statistics ($p > 0.05$). From Table 1 and 2, we can see that the blood glucose of OR group is obviously higher than that of control group when cycling 30 min, 40 min and completely exhausted ($p > 0.05$). However, the blood lactate of the OR group is obviously lower than that of control group when cycling 40 minutes and completely exhausted ($p < 0.05$).

The change of endurance time, total work and the rhythm of heart: The average rhythms of heart of OR group and control group are (167±20) times per minutes and (167±19) times per minutes, respectively when cycling. The deference is not significant in statistics ($p > 0.05$). The total work of OR group [(174.9±3.6) kJ] is apparently more than that of the control group [(143.0±2.3) kJ]. What's more, the cycling time of OR group [(47±12) min] is obviously more than that of the control group [(39±9) min] and the deference is significant in statistics.

Oligosaccharide during exercise: Whether or not performance differences were observed might be related to the way performance was evaluated or by the type and amount of oligosaccharide that was provided. Some studies have measured performance by a short time trial or a sprint at the end of 3 to 4 h of continuous exercise, whereas others have measured endurance capacity by time to exhaustion. More researchers have studied the effect of oligosaccharide feeding on prolonged time trial performance (100 to 128 km). There are clear differences in the reproducibility of these protocols and it is likely that some measures of performance are more sensitive than others to the effects of oligosaccharide feeding. The control of external variables such as diet, conditions during the experimental trials, feedback given to subjects and motivation of subjects is also different between studies and these factors are likely to play an important role. Some have argued that the reason for a lack of an ergogenic effect of oligosaccharide ingested during exercise was related to the duration of the exercise that may have been too short, the intensity of exercise that may have been too low to cause oligosaccharide depletion or the amount of oligosaccharide ingested may have been insufficient. The balance of studies, however, is convincingly in

favor of the ones that show ergogenic effects of oligosaccharide feeding during exercise.

ANALYSIS AND DISCUSSION

Oligosaccharide in sports drink: Under normal circumstances, sports drinks can be divided into pre-exercise and post-exercise beverage drinks, exercise the muscles inside the main consumption of sugar. Therefore, the main supplement drink before exercise muscles of sugar, let content reached a relatively high level. People tend to sweat a lot after doing exercises. A lot of sweat is water, as well as sugar, salt, trace elements. Sweating leads to blood concentration, reducing the body's blood volume. We must speed up the heart contraction frequency and the heart rate will accelerate. Therefore, as soon as possible after exercise, sport drink will replenish the lost moisture, however, simply filling with whitewater, from a nutritional point of view, cannot meet the body's needs and will sweat more and then cause the body's electrolyte balance disorders. It is not conducive to health. Therefore, the conventional sports drink is to add the right amount of electrolytes, maintaining osmotic pressure to serve fitness recovery purposes.

Our ability to run, bicycle, ski, swim and row hinges on the capacity of the body to extract energy from ingested food. As potential fuel sources, the carbohydrate, fat and protein in the foods that you eat follow different metabolic paths in the body but they all ultimately yield water, carbon dioxide and a chemical energy called Adenosine Triphosphate (ATP). Think of ATP molecules as high-energy compounds or batteries that store energy. Anytime you need energy-to breathe, to tie your shoes, or to cycle 100 miles (160 km)-your body uses ATP molecules. ATP, in fact, is the only molecule able to provide energy to muscle fibers to power muscle contractions. Creatine Phosphate (CP), like ATP, is also stored in small amounts within cells. It's another high-energy compound that can be rapidly mobilized to help fuel short, explosive efforts. To sustain physical activity, however, cells must constantly replenish both CP and ATP. Our daily food choices resupply the potential energy or fuel, that the body requires to continue to function normally. This energy takes three forms: carbohydrate, fat and protein. The body can store some of these fuels in a form that offers muscles an immediate source of energy. Carbohydrates, such as oligosaccharides, are readily broken down into glucose, the body's principal energy source. Glucose can be used immediately as fuel, or can be sent to the liver and muscles and stored as glycogen. During exercise, muscle glycogen is converted back into glucose, which only the muscle fibers can use as fuel. Therefore, oligosaccharides in the sports drink can provide energy for athletes during exercise.

Oligosaccharides are attracting increasing interest as prebiotic functional food ingredients. They can be extracted or obtained by enzymatic hydrolysis from a

variety of biomass sources or synthesized from simple oligosaccharides by enzymatic transfer reactions. The major prebiotic oligosaccharides on the market are inulin, fructo-oligosaccharides and galacto-oligosaccharides. They have been evaluated using a range of in vitro and in vivo methods, although there is a need for more large-scale human trials using modern microbiological methods. Prebiotics are being studied for their effects on gut health and specific clinical conditions, including colon cancer, Inflammatory Bowel Disease (IBD), acute infections and mineral absorption. Developing understanding of the functional ecology of the human gut is influencing current thinking on what a prebiotic might achieve and is providing new targets for prebiotic intervention.

How oligosaccharides work: Oligosaccharides are an odd-sounding group of carbohydrates that are gaining attention for their numerous roles in promoting and protecting human health. Researchers have reported beneficial effects on constipation, mineral absorption, lipid metabolism, glucose metabolism and immunomodulation, among others. Many of these benefits are directly linked to the digestion-resistant nature of oligosaccharides in the human body. Interestingly, certain enzymes may produce oligosaccharides from ingested starches and sugars when consumed with a meal.

Oligosaccharides consist of short chains of sugars, generally two to ten sugars in length, placing them between simple sugars and polysaccharides like starch. While the name is still largely unfamiliar to consumers, these compounds are being added to many food and supplement products and include inulin, oligofructose, FOS (fructo-oligosaccharides) and GOS (galacto-oligosaccharides). They are natural constituents of many plants and vegetables including chicory root, Jerusalem artichoke, onions, legumes, wheat, barley and asparagus. Human breast milk is another source of oligosaccharides; it provides a rich supply of GOS that has been shown to support a healthy immune system in infants. With their mildly sweet flavor and low glycemic index, oligosaccharides are gaining interest for use as functional foods.

Consumption of oligosaccharide-producing enzymes along with meals is one tool that may be beneficial in managing glucose and caloric availability. Oligosaccharide-producing enzymes transform digestible carbohydrates to indigestible forms and offer the advantage of increased fiber. There are currently two oligosaccharide-producing enzymes with New Dietary Ingredient (NDI) applications on file with the US Food and Drug Administration: transglucosidase and levansucrase.

Like other starch-digesting enzymes, transglucosidase is able to cleave the 1, 4-linkages that hold glucose units together to form starch. However, unlike these enzymes, transglucosidase then links the freed glucose to another substrate via a 1, 6-linkage that is not readily digestible by human digestive enzymes.

The enzyme produces glucose polymers known as gluco-oligosaccharides or GLOS. This effectively removes the transferred glucose from the absorbable pool of sugars and further limits the digestibility of the attached substrate. If it is not digested, it is not absorbed. If it is not absorbed, it reduces glucose absorption as well as the caloric availability. The bound glucose then also contributes to an increased fiber load.

Levansucrase functions similarly to transglucosidase; however, it cleaves the fructose-glucose bond in table sugar and attaches the fructose molecule to another substrate, creating indigestible Fructo-Oligosaccharides (FOS). The enzyme's action reduces the availability of fructose for absorption and results in less caloric availability.

Oligosaccharides and exercise endurance: There are several mechanisms by which oligosaccharide feeding during exercise may improve endurance performance. These include maintaining blood glucose and high levels of oligosaccharide oxidation, sparing endogenous glycogen, synthesizing glycogen during low intensity exercise, or a central effect of oligosaccharide. The mechanisms may be different for relatively short-duration (about 1 h) high-intensity exercise (80 to 85% of max oxygen uptake) than for long-duration (>2 h) low- to moderate-intensity exercise (60 to 75% of max oxygen uptake). Coyle and Puttfarcken (1993) found that oligosaccharide feeding during exercise at 70% of max oxygen uptake prevented the drop in blood glucose that was observed when water (placebo) was ingested. In the placebo trials the glucose concentration started to drop after 1 h and reached extremely low concentrations (2.5 mmol/L) at exhaustion after 3 h of exercise. With oligosaccharide feeding, euglycemia was maintained and subjects continued for 4 h at the same intensity. Total oligosaccharide oxidation rates followed a similar pattern. There was a drop in oligosaccharide oxidation after about 1.5 h of exercise with placebo and high rates of oligosaccharide oxidation were maintained with oligosaccharide feeding.

In a follow-up study subjects exercised to exhaustion at 73% of VO₂max (about 170 min) on three occasions separated by a week. During these trials, plasma glucose declined from 5.0 to 3.1 mmol/L. After resting for 20 minutes, the subjects attempted to continue exercise:

- After ingesting a placebo
- After ingesting glucose polymers (3 g/kg), or
- When glucose was infused intravenously to maintain plasma glucose concentrations of 11 mmol/L.

It is interesting that when subjects exercised to exhaustion with water, they were able to continue when glucose was ingested or infused intravenously. Time to fatigue during this second exercise bout was

significantly longer with oligosaccharide ingestion (26 min) or glucose infusion (43 min) than with placebo (10 min). These studies support the idea that plasma glucose is an important substrate during prolonged exercise. It is interesting that some studies found improvements in performance with oligosaccharide ingestion without a drop in plasma glucose concentration.

It has been shown that oligosaccharide feeding during exercise "spares" liver glycogen. Hepatic glucose output is tightly regulated, ensuring a relatively constant glucose output in the presence or absence of oligosaccharide feeding. Although the total rate of appearance of glucose increases somewhat with increasing rates of oligosaccharide intake, there is a progressive decrease in endogenous glucose production (liver glycogenolysis and gluconeogenesis) with increasing rates of oligosaccharide intake. Some studies have reported that with high rates of oligosaccharide intake liver glucose production returns to its basal levels, whereas others have observed complete blocking of hepatic glucose output by oligosaccharide feeding. This liver glycogen sparing means that there is still oligosaccharide in the liver toward the end of exercise, which could be beneficial if, for whatever reason, oligosaccharide intake cannot supply enough oligosaccharide to maintain plasma glucose concentrations and high rates of total oligosaccharide oxidation.

Whether oligosaccharide feeding during exercise has an effect on muscle glycogen breakdown has been the subject of considerable debate. An early study by Bergstrom and Hultman showed a 25% reduction in muscle glycogen breakdown during exhaustive one-legged cycling when glucose was infused intravenously to achieve hyperglycemic values of 21 mmol/L. However, such high plasma glucose concentrations are rather non-physiologic and impossible to achieve during exercise with oligosaccharide feeding. With oligosaccharide feeding during cycling exercise plasma glucose concentrations are usually elevated by about 0.5 to 1.0 mmol/L, whereas plasma insulin concentrations are similar to water ingestion. Several studies have reported that oligosaccharide ingestion does not result in a reduced net breakdown of muscle glycogen measured with the muscle biopsy technique or the indirect stable isotope technique. There are, however, a few studies that reported reduced muscle glycogen breakdown with oligosaccharide intake during cycling. Tsintzas *et al.* (2006) studied muscle glycogen breakdown during running at 70% of max oxygen uptake and observed that with oligosaccharide feeding there was a reduction in net muscle glycogen breakdown in type I muscle fibers after 60 min, whereas type II fibers seemed unaffected. In a follow-up study similar results were obtained. A reduction in muscle glycogen breakdown was observed with oligosaccharides intake and the depletion of type I muscle fibers coincided with the point of exhaustion.

During continuous cycling exercise at moderate exercise intensities oligosaccharide ingestion has little effect on plasma glucose concentrations, but without oligosaccharide ingestion plasma glucose concentrations may drop after approximately 2 h of exercise. The majority of the evidence shows that in these conditions oligosaccharide ingestion seems to improve endurance capacity (or performance) by maintaining euglycemia and high rates of oligosaccharide oxidation. In contrast, during constant pace running oligosaccharide ingestion has been shown to reduce net muscle glycogen breakdown in type I fibers. In intermittent exercise (cycling and running) oligosaccharide ingestion during exercise seems to reduce the net breakdown of muscle glycogen. An excellent review by Tsintzas *et al.* (1996) summarized the evidence for a glycogen sparing effect of oligosaccharide and the interested reader is referred to this report for more details.

The minimal amount of oligosaccharide needed: It must be noted that most of the early studies were performed after an overnight fast. This means that the subjects started the exercise with suboptimal glycogen stores and it has been shown that after an overnight fast liver glycogen stores may be considerably reduced. It seems obvious that exogenous oligosaccharide would have an effect in these conditions because it can provide an alternative substrate to compensate for the reduced endogenous oligosaccharide availability. Whether oligosaccharide feeding can also improve performance when endogenous oligosaccharide stores are optimal at the start of exercise has been investigated in later studies. This is highly relevant because athletes will rarely ever start a race with suboptimal glycogen stores.

Another concern is that it is currently much more difficult to perform a blinded study because subjects are no longer naive to the treatments and there are strong expectations that oligosaccharide feeding improves endurance performance, especially when many subjects are also familiar with the taste of artificial sweeteners and can distinguish between sweeteners and oligosaccharide. Unless placebos are prepared by dedicated laboratories, the results may be influenced by expectations. An elegant study by Clark *et al.* (2000) demonstrated this point in an attempt to distinguish between a placebo effect of oligosaccharide and a real physiologic effect. They examined cyclists who performed two 40-km time trials. During the first time trial they ingested water and for the second trial they were randomized into six different groups. The researchers gave oligosaccharide to three of the groups and placebo to other three groups. They told one group on oligosaccharide that it was placebo and they told the second group that it was oligosaccharide and they provided the third group with no information. Similarly in the placebo groups, one group was told it was placebo, one group that it was oligosaccharide and the third group was not told. The study confirmed the

existence of a placebo effect. Changes in mean power in the second trial were 4.3% when subjects were told they were ingesting oligosaccharide and 0.5% when they were told they were ingesting placebo. The real effect of oligosaccharide in this study was reported to be a slight reduction of 0.3%. Although this study illustrates the importance of a placebo effect, this study did not use a cross-over design and there appeared to be considerable differences between groups in their ability (especially peak power).

Mitchell *et al.* (1989) observed that 12 min of isokinetic time trial performance was enhanced at the end of 2 h of intermittent exercise. The improvements were similar with ingestion of 34, 39, or 50 g of oligosaccharide per hour compared with a water trial. Based on a study by Fielding *et al.* (1985). It is usually believed that a minimum of 22 g of oligosaccharide per hour is required to observe a performance benefit. In that study subjects exercised for 4 h and performed a sprint at the end. Performance improvements were observed when 22 g of oligosaccharide was ingested every hour, whereas no effects were observed when half this dose was consumed (11 g/h). In a study by Maughan *et al.* (1996), the intake of 16 g of glucose per hour resulted in an improved endurance capacity by 14% compared with water (no placebo was given in this study). However, Mitchell *et al.* (1989) found no effect of oligosaccharide ingestion on 12 min of all-out isokinetic cycling when 6% was ingested, but performance was enhanced when a 12% oligosaccharide solution was ingested. Interestingly, ingestion of an 18% oligosaccharide solution did not improve performance. Flynn *et al.* (1987) found no differences in performance with the ingestion of 5 or 10% oligosaccharide solutions. In that study, however, these drinks were similar to placebo in the resulting performance. Most of these studies provided 40 to 75 g of oligosaccharide per hour and observed performance benefits. Ingesting oligosaccharide at a rate higher than 75 g/h did not appear to be any more effective at improving performance than ingesting oligosaccharide at a rate of 40 to 75 g/h. It has been suggested that this is because ingestion of 40 to 75 g of oligosaccharide per hour already results in optimal oligosaccharide availability and ingesting oligosaccharide at higher rates may not increase the bioavailability. It is also possible that the current performance measurements are not sensitive enough to identify the small differences in performance that may exist when comparing two different oligosaccharide solutions. The overall conclusion seems to be that performance benefits can be observed with relative small amounts of oligosaccharide (16 g/h), but no further improvement has been observed with the ingestion of larger amounts of oligosaccharide.

CONCLUSION

Exercise time to exhaustion is known to be directly related to intramuscular glycogen content. After the oligosaccharide drink was ingested a significantly

longer run time to exhaustion (21%) was observed compared to after the water drink. The intensity used for the performance run was set at a VO₂ level of 10±3.0% above the IAT. Within this intensity range, intramuscular glycogen is thought to be the primary fuel for muscular activity. Therefore, since blood glucose was similar between trials and the run time to exhaustion was longer following the oligosaccharide trial, the assumption may be made that the greater hyperinsulinemia of the oligosaccharide trial stimulated a more rapid post exercise muscle glycogen synthesis and a larger muscle glycogen store prior to the performance run.

In conclusion, the recovery process of muscle glycogen seems to be accelerated when a drink which contains adequate amounts of oligosaccharide is consumed compared to an isocaloric drink which only contains water. This nutritional strategy may be critical for athletes who need to engage in multiple events or training sessions during the course of a day. Further studies are needed to determine the threshold (i.e., minimum) oligosaccharide required to optimize post-exercise muscle glycogen synthesis.

ACKNOWLEDGMENT

The authors wish to thank the helpful comments and suggestions from my teachers and colleagues in intelligent detection and control lab of HIT at Weihai. And also thank Beijing Up-tech to provide part hardware. This study is supported by the study fund of HIT at Weihai (No. IMVQ02020003 and IMJQ21080002).

REFERENCES

- Angus, D.J., M. Hargreaves, J. Dancy, M.A. Febbraio, 2000. Effect of carbohydrate or carbohydrate plus medium-chain triglyceride ingestion on cycling time trial performance. *J. Appl. Physiol.*, 88(1): 113-119.
- Carter, J., A.E. Jeukendrup, T. Mundel and D.A. Jones, 2003. Carbohydrate supplementation improves moderate and high-intensity exercise in the heat. *Pflug. Arch.*, 446(2): 211-219.
- Clark, V.R., W.G. Hopkins, J.A. Hawley and L.M. Burke, 2000. Placebo effect of carbohydrate feedings during a 40-km cycling time trial [In Process Citation]. *Med. Sci. Sport. Exer.*, 32: 1642-1647.
- Coyle, J.T. and P. Puttfarcken, 1993. Oxidative stress, glutamate, and neurodegenerative disorders. vol. 262, pp: 689-695.
- Fielding, R.A., D.L. Costill, W.J. Fink, D.S. King, M. Hargreaves and J.E. Kovaleski, 1985. Effect of carbohydrate feeding frequencies and dosage on muscle glycogen use during exercise. *Med. Sci. Sport. Exer.*, 17: 472-476.
- Flynn, M.G., D.L. Costill, J.A. Hawley, W.J. Fink, P.D. Neuffer, R.A. Fielding and M.D. Sleeper, 1987. Influence of selected carbohydrate drinks on cycling performance and glycogen use. *Med. Sci. Sport. Exer.*, 19: 37-40.
- Hargreaves, M., J.A. Hawley and A. Jeukendrup, 2004. Pre-exercise carbohydrate and fat ingestion: Effects on metabolism and performance. *J. Sport. Sci.*, 22: 31-38.
- Jentjens, R.L. and A.E. Jeukendrup, 2002. Prevalence of hypoglycemia following pre-exercise carbohydrate ingestion is not accompanied by higher insulin sensitivity. *Int. J. Sport Nutr. Exe.*, 4: 398-413.
- Jentjens, R.L.P.G., M.C. Venables and A.E. Jeukendrup, 2004. Oxidation of exogenous glucose, sucrose and maltose during prolonged cycling exercise. *J. Appl. Physiol.*, 96(4): 1285-1291.
- Madsen, K., D.A. MacLean, B. Kiens and D. Christensen, 1996. Effects of glucose, glucose plus branched-chain amino acids, or placebo on bike performance over 100 km. *J. Appl. Physiol.*, 81(6): 2644-2650.
- Maughan, R.J., L.R. Bethell and J.B. Leiper, 1996. Effects of ingested fluids on exercise capacity and on cardiovascular and metabolic responses to prolonged exercise in man. *Exp. Physiol.*, 81: 847-859.
- Mitchell, J.B., D.L. Costill, J.A. Houmard, W.J. Fink, D.D. Pascoe and D.R. Pearson, 1989. Influence of carbohydrate dosage on exercise performance and glycogen use. *J. Appl. Physiol.*, 67: 1843-1849.
- Tsintzas, O. K., C. Williams, L. Boobis, P. Greenhaff, 1996. Carbohydrate ingestion and single muscle fiber glycogen metabolism during prolonged running in men. *Journal of Applied Physiology*, (81): 801-809.
- Tsintzas K., K. Jewell, M. Kamran, D. Laithwaite, T. Boonsong, J. Littlewood, I. Macdonald and A. Bennett, 2006. Differential regulation of metabolic genes in skeletal muscle during starvation and refeeding in humans. *J. Physiol.* pp: 291-303.