Biotecnia
Revista de Ciencias Biológicas y de la Salud
http://biotecnia.unison.mx
Universidad de Sonora
"El saber de mis hijos hará mi grandeza"

THE INFLUENCE OF AN AMARANTH-BASED BEVERAGE ON CYCLING PERFORMANCE: A PILOT STUDY

INFLUENCIA DE UNA BEBIDA A BASE DE AMARANTO SOBRE EL RENDIMIENTO EN CICLISMO: ESTUDIO PILOTO

Ever Espino-González[1], María J. Muñoz-Daw[1]*, Juan M. Rivera-Sosa[2], María L. De la Torre-Díaz[1], Gabriel E. Cano-Olivas[3], Juan C. De Lara-Gallegos[1], María C. Enríquez-Leal[4]

[1] Sport and Exercise Nutrition Laboratory, Faculty of Physical Culture Sciences, Autonomous University of Chihuahua, Chihuahua, México.
[2] Physical Activity and Health Laboratory, Faculty of Physical Culture Sciences, Autonomous University of Chihuahua, Chihuahua, México.
[3] Cano's Bicycle Laboratory, Chihuahua, México.
[4] Faculty of Chemical Sciences, Autonomous University of Chihuahua, Chihuahua, México.

ABSTRACT

The present study aimed to evaluate the effectiveness of an amaranth-based beverage (CHO-P) on cycling performance and hydration status, despite containing a total caloric content higher than that of a commercial sports beverage (CHO-P: 52.48 kcal per 100 mL vs CHO: 24 kcal per 100 mL). In a randomized, crossover design, six cyclists performed two exercise tests separated by seven days. Each test comprised two time-trials (32.20 km and 5 km) separated by 10 min of rest. Participants consumed either an amaranth-based beverage (CHO-P; 10% and 1.5% concentrations) or a commercial sports beverage (CHO; 6%). Changes in hematocrit and body mass, ratings of perceived exertion, and average power were assessed throughout both tests. 32.2-km time-trial performance was enhanced with CHO-P compared to CHO (54.3 ± 4.1 min vs 55.6 ± 4.8 min; $p<0.05$). However, no other variable measured in this study was significantly different between beverage types. Further laboratory based research should be performed to further explore the ergogenic potential of amaranth supplementation during endurance exercise.

Keywords: Amaranthus, Physical Endurance, Sports Nutritional Sciences, Carbohydrates, Plant Proteins.

RESUMEN

El objetivo del presente estudio fue evaluar los efectos de una bebida a base de amaranto en el rendimiento físico y el estado de hidratación, a pesar de tener un contenido calórico mayor al de las bebidas deportivas comerciales (CHO-P: 52.48 kcal por 100 mL vs CHO: 24 kcal por 100 mL). En un diseño aleatorizado y cruzado, seis ciclistas realizaron dos pruebas de esfuerzo separadas por siete días. Cada prueba consistió en dos carreras contra reloj separadas por 10 min de descanso, una de 32.20 km y otra de 5 km. Los participantes consumieron una bebida a base de amaranto (CHO-P; 10% y 1,5%) o una bebida deportiva comercial (CHO; 6%). Durante ambas pruebas se evaluaron los cambios en el hematocrito y la masa corporal, la percepción del esfuerzo y la potencia muscular. El rendimiento mejoró en la carrera contra reloj de 32.2 km cuando los sujetos se hidrataron con CHO-P en comparación con CHO (54,3±4,1 min frente a 55,6±4,8 min; $p<0,05$). Sin embargo, ninguna otra variable fue significativamente diferente entre los dos tratamientos. Futuras investigaciones permitirán explorar el potencial ergogénico de la ingesta de amaranto durante el ejercicio de resistencia.

Palabras Clave: Amaranto, Resistencia Física, Ciencias Nutricionales Deportivas, Hidratos de Carbono, Proteínas Vegetales.

INTRODUCTION

The development and evaluation of new nutritional ergogenic sports beverages require a combination between food engineering and sports nutrition science. Studies lead by physiologist David Costill in the 1980s formed the basis for the use of sports beverages (CHO) with different nutrients (Fielding *et al*., 1985; Neufer *et al*., 1987). Subsequent research concluded that sports beverages help significantly improve endurance performance due to their contribution of carbohydrate, electrolytes and water (Aragón, 2009).

Currently, there is an interest in adding protein to carbohydrate beverages in order to improve endurance performance. It has been found that carbohydrate-protein (CHO-P) ingestion during exercise improves time-trial (Roberts *et al*., 2012; Seifert *et al*., 2012), time-to-exhaustion (Mccleave *et al*., 2011), multiple-sprint sport (Highton *et al*., 2013), and subsequent cycling time-trial performance (Hall *et al*., 2013). In addition, some studies have reported that CHO-P ingestion improves blood levels of insulin and glucose during exercise compared to placebo (Ivy *et al*., 2003), reduces body weight loss induced by dehydration through exercise (Cathcart *et al*., 2011), and helps to diminish post-exercise muscle damage (Seifert *et al*., 2005; Hall *et al*., 2013). However, these performance benefits of ingesting CHO-P beverages during exercise remain unclear. In 2013, Coletta *et al*. devised a study in order to assess the influence of three different beverages on endurance performance during an outdoor running field trial

*Autor para correspondencia: María de Jesús Muñoz Daw
Correo electrónico: mmunoz@uach.mx
Recibido: 22 de diciembre de 2017
Aceptado: 04 de marzo de 2018

(>60 minutes): a carbohydrate (CHO), a double-carbohydrate (CHO-CHO), and a carbohydrate-protein (CHO-P) beverage compared to a placebo (PLA). The results showed no significant difference between beverages in time to complete a 19.2 km run (PLA = 88.6±11.6 min, CHO = 89.1±11.3 min, CHO-P = 89.1±11.8 min, and CHO-CHO = 89.6±11.9 min) (Coletta *et al*., 2013). Other authors have obtained similar results that demonstrate that consumption of CHO-P beverages during exercise does not enhance endurance (Gasier and Olson, 2010; Martínez-Lagunas *et al*., 2010) or resistance performance (Smith *et al*., 2017).

In the present study amaranth was used to elaborate a homemade CHO-P beverage due to the quality of its protein and its proportion of carbohydrate. The nutritional quality of amaranth is better than that of many cereals, and its balance of essential amino acids is higher than that of many vegetable proteins (Juan *et al*., 2007). The WHO (World Health Organization) and the FAO (Food and Agricultural Organization) state that on an ideal protein value scale of 100, amaranth is 75, soybean is 68, wheat is 60, and corn is 44 (Herrera and Montenegro, 2012). Species of the genus *Amaranthus* have been described as non-traditional sources of protein, with a protein percentage between 12 and 16%. These values are higher than wheat (10%), corn (10%) and rice (7%) (Arcila and Mendoza, 2006).

Based on its higher protein value, the purpose of the present study was to determine if an amaranth-based beverage (CHO-P) given during cycling exercise would improve physical performance and hydration status, despite containing a total caloric content higher than that of a commercial sports beverage (CHO-P: 52.48 kcal per 100 mL vs CHO: 24 kcal per 100 mL).

MATERIAL AND METHODS

Experimental Approach to the Problem

This study used a randomized, counterbalanced, and crossover design. After preliminary measurements, participants performed two exercise tests separated by seven days; each test comprised two time-trials, one of 32.20 km and another of 5 km, separated by 10 min of rest. Participants consumed either an amaranth-based beverage (CHO-P; 10% and 1.5% concentrations) or a commercial sports beverage (CHO; 6%).

Subjects

Six trained cyclists (five men and one woman) between 20 and 32 years of age were recruited from both the Autonomous University of Chihuahua and a local running group. The subjects had been cycling for at least two years and all of them had raced in national competitions. These entrance criteria were considered so that the findings of the study could be properly used in competitive athletic populations. Men weighed 77.8±9.3 kg and had a relative power of 3.72±0.75 W/kg, while the woman weighed 54.7 kg and had a relative power of 2.73 W/kg. Following approval from The Ethics Committee of the Angels Hospital, Chihuahua, México, all participants provided written informed consent before their participation. All procedures were performed in accordance with the Declaration of Helsinki.

PROCEDURES

Design of the amaranth-based beverage

First, the amaranth was examined to ensure it was fit for human consumption. The microbiological analysis was performed according to the Mexican Official Standards for the preparation and dilution of food samples (NOM-110-SSA1-1994), counting aerobic bacteria (NOM-092-SSA1-1994), molds and yeasts (NOM-111-SSA1-1994) and coliform bacteria (NOM-112-SSA1-1994). The results showed absence of fungus, yeast, aerobic mesophilic bacteria and coliforms.

Then, for the proximate analysis of the beverage, the nitrogen concentration of the amaranth was calculated according to the Kjeldahl method (Bradstreet, 1965) (6500000; Labconco 050740015 H Micro-Kjeldahl Equipment, Missouri, U.S.A.). Based on the nitrogen content, protein concentration was determined using the conversion factor of 6.25 (equivalent to 0.16 g nitrogen per gram of protein). Lipid content was determined with a Goldfisch apparatus (EG-600; Craft C05158 Goldfisch Equipment DE 6 U, D.F., México). Moisture content was obtained using a moisture balance (VE-50-5; Velab 07335, D.F., México). Minerals content was determined using a muffle (FE-292; Felisa 1311052 Muffle, Jalisco, México) and an oven (FE-292; Felisa 1312016 Oven, Jalisco, México). Ultimately, carbohydrate content was obtained by difference.

Once the proximate analysis of the amaranth was completed, 88.8 g of amaranth grains, 41.3 g of sugar, 0.35 g of sodium chloride, and grape or orange flavor were added to one liter of water to make a beverage with a protein concentration of 1.5%, 10% carbohydrates, and 0.35 g/L of electrolytes (sodium chloride).

This beverage is not yet approved by the FDA; it is still an investigational supplement.

Preliminary measurements

Before starting experimental tests, all participants completed a preliminary test on their own bicycles to determine relative cycling power. Tests were performed on a Computrainer (RacerMate Inc., Seattle, USA), using the Computrainer coaching software (V 4.0.2.; RacerMate One). The test protocol consisted of 20 min of cycling at high resistance. Body mass and height were also measured for demographic purposes. Participants were weighed on a digital scale to the nearest tenth of a kg, and were measured in their cycling clothing only, without shoes or socks, as previously implemented in a similar study (Breen *et al*., 2010).

Experimental Protocol

All tests were conducted in the laboratory of cycling (Cano's bicycle laboratory, Chihuahua, Mexico). No less than a week after preliminary measurements, participants reported to the laboratory for the experimental tests. All tests were performed on a Computrainer (RacerMate Inc., Seattle, USA) using each subject's own bicycle. The Computrainer was connected to a computer loaded with the Computrainer coaching software (V 4.0.2.; RacerMate One). After a 10-min warm-up, participants performed the two experimental tests separated by seven days. They were instructed to complete the trials as a competitive event but without verbal or physiological feedback during the tests. They could only see the distance covered, which was displayed in a monitor.

Data and Sample Collection and Analyses

Body mass was measured before the 32.20 km trials and immediately after the 5 km trials to determine fluid losses during exercise. Participants were weighed on a digital scale to the nearest tenth of a kilogram, and were measured in their cycling clothing without shoes and socks, as previously described. To determinate hematocrit changes, capillary blood samples (2 capillary tubes) from participant's fingers were taken before and immediately after the 32.20 km trials. Samples were analyzed immediately after the tests using a centrifuge (M-250; SOLBAT, Puebla, México) and a hematocrit reader (SOLBAT, Puebla, México).

During the 32.20 km time-trials, average muscular power was obtained by using the Computrainer coaching software (V 4.0.2.; RacerMate One), whilst subjective ratings of perceived exertion on a Borg scale (ranging from 6 to 20) (Guidelines for Exercise Testing and Prescription, ACSM, 2006) were recorded every 5 min throughout the trials (see Figure 1).

Supplementation

Participants consumed either an amaranth-based beverage (CHO-P; 10% and 1.5% concentrations) or a commercial sports beverage (CHO; 6%). The amaranth-based beverage was formulated by using the methods described above. The CHO beverage provided carbohydrates in the form of dextrose and sucrose. The caloric content of the amaranth-based beverage was higher than that of the commercial sports beverage (CHO-P: 52.48 kcal per 100 mL vs CHO: 24 kcal per 100 mL). Beverages were supplied by the Sport and Exercise Nutrition Laboratory (Autonomous University of Chihuahua, Chihuahua, Mexico), and were prepared by investigators not directly involved in the study. The protocol was designed to ensure that participants ingested exactly 150 mL of fluid every 10 min during the 32.20 km time-trials (see Figure 1).

Statistical analyses

SPSS v 18.0 statistical software (SPSS Inc., Chicago, IL, USA) was used for all statistical analyses. Variables that included multiple measures per test as changes in hematocrit and body mass were analyzed using Student's t-test for paired samples. All the other variables were also analyzed using paired-samples t-tests to determine whether measurements were different between the CHO and CHO-P beverages. Ratings of perceived exertion data were analyzed by using a two-way analysis of variance with independent t-test used to differentiate means upon an interaction. The level of significance was set at $p<0.05$ for all analyses. All data from these analyses are reported as mean±standard deviation.

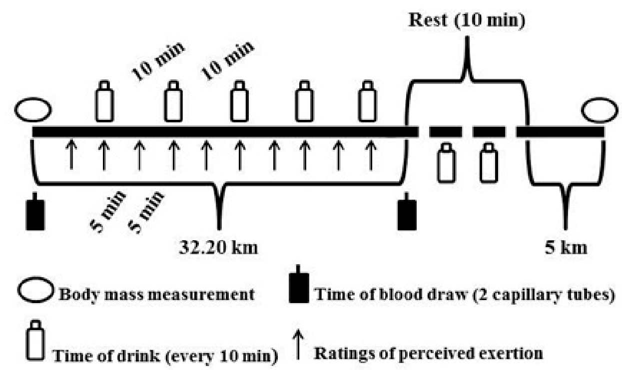

Figure 1. Diagram of the experimental protocol indicating work rates and times for blood draws, body mass measurements, ratings of perceived exertion, and time of supplementation.
Figura 1. Diagrama del protocolo experimental indicando las zonas de trabajo y los tiempos para la extracción de sangre, las mediciones de la masa corporal, los registros del esfuerzo percibido y la suplementación.

RESULTS

There were no statistically significant differences in ambient temperature (dry-bulb temperature: CHO: 28.3±1.6 °C, CHO-P: 29.2±4.2 °C; p = 0.78) (wet-bulb temperature: CHO: 21.5±3.1°C, CHO-P: 20.2±2.4 °C; p = 0.384) and relative humidity (CHO: 57.0±11.6 %, CHO-P: 46.3±5.7 %; p = 0.338) measurements of the cycling laboratory between test days.

Table 1. Muscular power, performance times, and ratings of perceived exertion.
Tabla 1. Potencia muscular, tiempos de ejecución y percepción del esfuerzo.

Test days	MP (Watts)	T 32.20 km (min)	T 5 km (min)	RPE
CHO	230.3±43.3	55.6±4.8	8.4±0.6	15.1±1.3
CHO-P	233.5±41.1	54.3±4.1	8.0±0.4	14.8±1.1
p	0.52	0.05*	0.06	0.33

CHO = commercial sports beverage, CHO-P = amaranth-based beverage, MP = muscular power, T = time, RPE = rating of perceived exertion. Values are presented as means ± SD. * Significant difference (p<0.05) between groups.

Differences in muscular power, performance times, and ratings of perceived exertion are displayed in Table 1. Average time to complete the 32.2-km time-trial was lower with CHO-P (54.3±4.1 min) compared with CHO (55.6±4.8 min), ($p<0.05$) and there was a tendency to significance in time to complete the 5-km time-trial (p = 0.06). There was no significant difference in ratings of perceived exertion (RPE) between treatments (p = 0.33) (Figure 2 shows the RPE values during the exercise trials). No other differences between tests were observed for the variables displayed in Table 1.

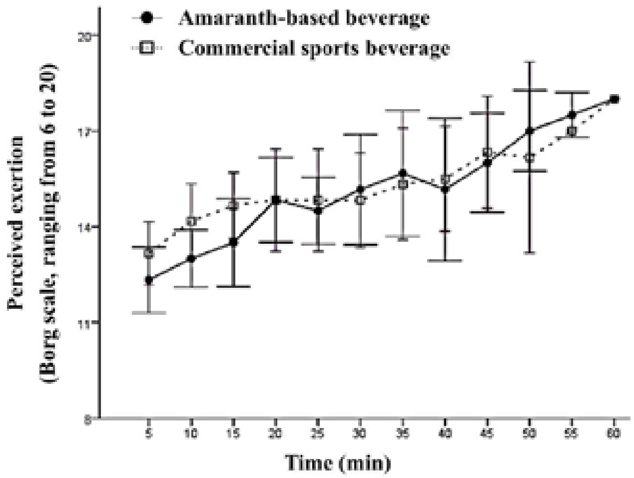

Figure 2. Perceived exertion during prolonged continuous exercise.
Figura 2. Esfuerzo percibido durante el ejercicio continuo.

Table 2. Changes in hematocrit and body mass during prolonged exercise.
Tabla 2. Cambios en el hematocrito y la masa corporal durante el ejercicio prolongado.

	Hematocrit BE (%)	Hematocrit PE (%)	p
CHO	44.78±1.2	45.79±1.7	0.324
CHO-P	45.68±2.2	46.28±1.6	0.269
	Body mass BE	**Body mass PE**	**p**
CHO	73.9±12.1	73.2±11.9	0.003*
CHO-P	73.9±12.3	73.3±12.1	0.030*

CHO = commercial sports beverage, CHO-P = amaranth-based beverage, BE = before exercise, PE = post-exercise. Values are presented as means ± SD. * Significant difference (p<0.05) between before and after exercise.

Changes in hematocrit and body mass during prolonged exercise were similar with both treatments. These data are illustrated in Table 2. There was a significant difference in body mass between before and after exercise with both treatments (CHO-P: p = 0.030, CHO: p = 0.003).

DISCUSSION

The primary objective of this study was to evaluate the effects of an amaranth-based beverage on endurance performance, physiological markers of hydration status and subjective ratings of perceived exertion. These variables can be influenced by climatic factors, the mood of the participants, the type of performance test, and primarily the composition of the supplements. In this case, no significant differences were observed in climatic factors between test days, and all participants completed the trials without any verbal or physiological feedback that can affect their mood. Although physical performance does not depend on the exercise tests used to evaluate it, it has been reported that there is less variability in time-trial tests than in similar time-to-exhaustion tests (Laursen *et al*., 2007). However, several time-to-fatigue studies have found significant improvements on endurance performance in CHO-P groups compared to CHO alone or placebo (Ivy *et al*., 2003; Saunders *et al*., 2004; Valentine *et al*., 2008; Ferguson-Stegall *et al*., 2010; Alghannam, 2011; Mccleave *et al*., 2011). In this case, we used time-trial tests and found significant improvements on cycling performance, which may be explained in part by the higher total caloric content in the CHO-P beverages.

Before the experiment, 30 volunteers were asked to choose between an amaranth-based beverage and an amaranth-based gel (considering their taste and texture). The result was the liquid version, a beverage with a total caloric content higher than that in commercial CHO beverages (CHO-P: 52.48 kcal per 100 mL vs CHO: 24 kcal per 100 mL). Mechanistically, this energy difference between beverages might be responsible for the time-trial performance differences. However, some studies have found significant improvements on endurance performance when using the same or a similar beverage caloric content (isocaloric studies) (Alghannam, 2001; Highton, 2013), which highlights the possible ergogenic potential of adding small amounts of protein or amino acids to CHO beverages.

Amino acids have several transport pathways from the intestines and are known to boost the absorption of water and electrolytes (Na+, H+, K+ and Cl-) (Poncet and Taylor, 2013; Hellier, 1973). Based on these facts, it is possible that little amounts of protein in CHO beverages helps to improve endurance performance by increasing the fluid transport across the lining of the intestine. However, transportation of energy substrates and electrolytes across the intestinal wall was not measured in the current study. Amino acids have also been proposed to exert its ergogenic effect by retarding central fatigue. Highton and colleagues have suggested that amino acids might decrease free fatty acid levels causing higher albumin availability which in turns bind to free tryptophan and avoid its conversion to serotonin, a monoamine neurotransmitter known to regulate anxiety and mood which may alter effort perception and psychological skills important for athletes (Highton *et al*., 2013). In this case the amino acid profile of the CHO-P beverage was not determined; thus, it cannot be elucidated whether the performance improvements were the result of the higher total energy content or due to specific protein (amino acids) mechanisms of the amaranth. Future research should consider the amino acid composition of beverages and measure plasma amino acids levels to continue exploring the possible ergogenic potential of different sources of protein in CHO-P beverages.

In terms of how to evaluate hydration status, changes in body mass have shown to be a stable physiological indicator to monitor fluid balance and acute changes in body fluids (Harvey *et al*.,2008; Meir *et al*., 2011). This method was used in the study by Cathcart *et al*. (2011), where the results showed that body mass changed significantly in the PLA group (-0.75±0.22 kg, P = 0.01), but did not change in the CHO–P group (0.42±0.42 kg, P = 0.35). This may have been possible due to the higher total caloric content of the CHO-P beverages (CHO: 76 g L^{-1} vs CHO-P: 72 g L^{-1} CHO, 18 g L^{-1} P).

In a study by Highton *et al*. (2013), the caloric content of the beverages also could have influenced changes in body mass. In this case, there was no difference between treatments (P>0.05), perhaps because the total caloric content of the beverages was similar (CHO: 8% vs CHO-P: 6% CHO, 2% P). Although the caloric content of the beverages appears to be an important factor influencing changes in body mass, the results have not always been as expected. In the present study, changes in body mass were similar with both treatments (CHO-P: p = 0.030, CHO: p = 0.003), even though the caloric content of the amaranth-based beverage was higher than that of the commercial sports beverage. Similarly, changes in hematocrit during prolonged exercise were similar with both treatments. Perhaps, it needs to be measured for a longer period than in the present study to observe differences in hydration status between treatments.

Limitations of the study

One of the limitations of the present study was that a double blind design was not possible due to the organoleptic properties of the amaranth. Moreover, there was possibly a lack of statistical power due to the lower number of subjects. Nevertheless, there are some studies that have used the same or a similar number of subjects (Alghannam, 2011; Tarpey *et al*., 2013), one of which obtained results similar to ours (Alghannam, 2011).

CONCLUSIONS

To our knowledge, this is the first study to explore the ergogenic potential of amaranth. Our results suggest that an amaranth-based beverage may be equally effective as a commercial sports beverage for supporting optimal performance and hydration. However, further laboratory based studies with a standardized protocol and adequate number of subjects should be performed to determine whether these effects were the result of the energy difference between beverages or due to the amaranth properties and its nutritional value. Future research should include different amaranth concentrations and use food engineering techniques that allow researchers to perform double-blind trials. The results of this pilot study also showed that the effective caloric concentration of sports beverages remains debatable.

ACKNOWLEDGMENTS

The authors would like to thank the subjects in this study for their time, effort, and dedication. This study was funded by Chihuahua Youth Institute (ICHIJUV, for its acronym in Spanish). The results of the present study do not constitute endorsement of the product by the authors.

REFERENCES

Alghannam, A.F. 2011. Carbohydrate–Protein Ingestion Improves Subsequent Running Capacity Towards the End of a Football-Specific Intermittent Exercise. Applied Physiology, Nutrition, and Metabolism. 36: 148-157.American College of Sports Medicine. 2006. Guidelines for Exercise Testing and Prescription. Baltimore. 7th ed. Lippincott Williams & Wilkins. p. 123.

Aragón, L.F. 2009. Milk ... Sports Drink?. Kérwá. (consulted 11/08/2015). Available from: http://hdl.handle.net/10669/444.

Arcila, N., Mendoza, Y. 2006. Elaboration of an instant beverage based on grains amaranth (*Amaranthus cruentus*) and its potential use in human consumption. Revista de la Facultad de Agronomía LUZ. 23: 110-119.Bradstreet, R.B. 1965. The Kjeldahl Method for Organic Nitrogen. 1st Edition. Hilside, N.J. Academic Press Inc. p. 169-224.Breen, L., Tipton K.D., Jeukendrup A.E. 2010. No effect of carbohydrate-protein on cycling performance and indices of recovery. Medicine and Science in Sports and Exercise. 42: 1140-1148.

Cathcart, A.J., Murgatroyd, S.R., McNab, A., Whyte, L.J., Easton, C. 2011. Combined carbohydrate-protein supplementation improves competitive endurance exercise performance in the heat. European Journal of Applied Physiology. 111: 2051-2061.Coletta, A., Thompson, D.L., Raynor, H.A. 2013. The influence of commercially-available carbohydrate and carbohydrate-protein supplements on endurance running performance in recreational athletes during a field trial. Journal of the International Society of Sports Nutrition. 10: 17-24.

Ferguson-Stegall, L., Mccleave, E.L., Ding, Z., Kammer, L.M., Wang, B., Doerner, P.G., Liu, Y., Ivy, J.L. 2010. The Effect of a Low Carbohydrate Beverage With Added Protein on Cycling Endurance Performance in Trained Athletes. Journal of Strength and Conditioning Research. 24: 2577-2586.

Fielding, R.A., Costill, D.L., Fink, W.J., King, D.S., Hargreaves, M., Kovaleski, J.E. 1985. Effect of carbohydrate feeding frequencies and dosage on muscle glycogen use during exercise. Medicine and Science in Sports and Exercise. 17: 472-476.

Gasier, H., Olson, C. 2010. The Effects of a Carbohydrate-Protein Drink on Performance and Mood in U.S. Pararescue Trainees. Journal of Exercise Physiologyonline. 13: 22-31.

Hall, A.H., Leveritt, M.D., Ahuja, K.D., Shing, C.M. 2013. Coingestion of carbohydrate and protein during training reduces training stress and enhances subsequent exercise performance. Applied Physiology, Nutrition, and Metabolism. 38: 597-604.

Harvey, G., Meir, R., Brooks, L., Holloway, K. 2008. The use of body mass changes as a practical measure of dehydration in team sports. Journal of Science and Medicine in Sport. 11 :600-603.

Hellier, M.D., Thirumalai, C., Holdsworth, C.D. 1973. The effects of amino acids and dipeptides on sodium and water absorption in man. journal of the British Society of Gastroenterology. 14: 41-45.

Herrera, S., Montenegro, A. 2012. Amaranth: the prodigious food for longevity and life. Kalpana. 8: 50-66.

Highton, J., Twist, C., Lamb, K., Nicholas, C. 2013. Carbohydrate-protein coingestion improves multiple-sprint running performance. Journal of Sports Sciences. 31: 361-369.

Ivy, J.L., Res, P.T., Sprague, R.C., Widzer, M.O. 2003. Effect of a Carbohydrate-Protein Supplement on Endurance Performance During Exercise of Varying Intensity. International Journal of Sport Nutrition and Exercise Metabolism. 13: 382-395.

Juan, R., Pastor, J., Alaiz, M., Megías, C., Vioque, J. 2007. Protein characterization of eleven species of grains amaranth. Grasas y Aceites. 58: 49-55.

Laursen, P.B., Francis, G.T., Abbiss, C.R., Newton, M.J., Nosaka, K. 2007. Reliability of time-to-exhaustion versus time-trial running tests in runners. Medicine and Science in Sports and Exercise. 39: 1374-1379.

Martínez-Lagunas, V., Ding, Z., Bernard, J.R., Wang, B., Ivy, J.L. 2010. Added Protein Maintains Efficacy of a Low-Carbohydrate Sports Drink. Journal of Strength and Conditioning Research. 24: 48-59.

Meir, R.A., Brooks, L.O., Rogerson, S. 2011. What do changes in prematch vs. postmatch, 1, 2, and 3 days postmatch body weight tell us about fluid status in English Premiership rugby union players?. Journal of Strength and Conditioning Research. 25: 2337-2343.

Mccleave, E.L., Ferguson-Stegall, L., Ding, Z., Doerner III, P.G., Wang, B., Kammer, L.M., Ivy, J.L. 2011. A low carbohydrate-protein supplement improves endurance performance in female athletes. Journal of Strength and Conditioning Research. 25: 879-888.

Neufer, P.D., Costill, D.L., Flynn, M.G., Kirwan, J.P., Mitchell, J.B., Houmard, J. 1987. Improvements in exercise performance: effects of carbohydrate feedings and diet. Journal of Applied Physiology. 62: 983-988.

Poncet, N., Taylor, P.M. The role of amino acid transporters in nutrition. 2013. Current Opinion in Clinical Nutrition & Metabolic Care. 16: 57-65.

Roberts, J.D., Tarpey, M.D., Kass, L.S., Roberts, M.G. 2012. An investigative study into the influence of a commercially available carbohydrate-protein electrolyte beverage on short term repeated exercise performance. Journal of the International Society of Sports Nutrition. 9: 5-16.

Saunders, M.J., Kane, M.D., Todd, M.K. 2004. Effects of a Carbohydrate-Protein Beverage on Cycling Endurance and Muscle Damage. Medicine and Science in Sports and Exercise. 36: 1233-1238.

Seifert, J.G., Kipp, R.W., Amann, M., Gazal, O. 2005. Muscle damage, fluid ingestion, and energy supplementation during recreational alpine skiing. International Journal of Sport Nutrition and Exercise Metabolism. 15: 528-536.

Seifert, J.G., Kipp, R.W., Bacharach, D.W. 2012. The effects of a carbohydrate-protein gel supplement on alpine slalom ski performance. Journal of Sports Science and Medicine. 11: 537-541.

Smith, J.W., Krings, B.M., Shepherd, B.D., Waldman, H.S., Basham, S.A., McAllister, M.J. 2017. Effects of Carbohydrate and Branched Chain Amino Acid Beverage Ingestion during Acute Upper-Body Resistance Exercise on Performance and Post-Exercise Hormone Response. Applied Physiology, Nutrition, and Metabolism. 2-28.

Stearns, R.L., Emmanuel, H., Volek, J.S., Casa, D.J. 2010. Eeffects of Ingesting Protein in Combination with Carbohydrate During Exercise on Endurance Performance: A Systematic Review with Meta-Analysis. Journal of Strength and Conditioning Research. 24: 2192-2202.

Tarpey, M.D., Roberts, J.D., Kass, L.S., Tarpey, R.J., Roberts, M.G. 2013. The ingestion of protein with a maltodextrin and fructose beverage on substrate utilisation and exercise performance. Applied Physiology, Nutrition, and Metabolism. 38: 1245-1253.

Valentine, J.R., Saunders, M.J., Todd, M.K., Laurent, T.G. 2008. Influence of Carbohydrate-Protein Beverage on Cycling Endurance and Indices of Muscle Disruption. International Journal of Sport Nutrition and Exercise Metabolism. 18: 363-378.