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Omega-3 enriched chocolate milk: A functional drink to improve health during exhaustive exercise

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ABSTRACT

Omega-3 enriched chocolate milk (O3ECM) has a suitable combination of proteins, electrolytes and carbohydrates for post-workout recovery. Herein, the effects of an O3ECM on biochemical parameters and muscle damage in sedentary rats submitted to a session of intense and acute exhaustive exercise was evaluated. Forty-eight male Wistar rats were divided into the following test groups: (a) sedentary rats fed a chocolate milk; (b) sedentary rats fed an O3ECM; (c) exercised rats fed a chocolate milk; and (d) exercised rats fed an O3ECM. After 15 days, the analysis of muscle damage and health parameters was performed. The results showed that the O3ECM consumption decreased the muscle damage (creatine kinase –20% and lactate dehydrogenase –18.7%), as well as increased the activity of endogenous enzymes (catalase +41%, superoxide dismutase +26.7% and glutathione peroxidase +35.7%) and decreased the total cholesterol (–7.8%) and triacylglycerols (–16.2%). Overall, O3ECM can minimize the deleterious effects of exhaustive exercise.

1. Introduction

Functional foods have gained prominence in the market, with a large number of products being developed (Augustin et al., 2015; Kralovec et al., 2009; Menrad, 2003). Foods with wide acceptance and consumption have been added with ingredients that potentially improve the consumer's health. Such foods are also developed by the food industry to meet demands for specific audiences, such as the athletes. In this context, the formulation of a functional chocolate milk added with omega-3-rich chia oil is interesting due to the broad consumer acceptance of chocolate milk allied to the widely known functionality of omega-3.

The major vegetable oils traded and used by industry are palm, soybean, sunflower and canola oils. However, in recent

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years, interest in other plant sources of unsaturated fatty acids has increased since these products have nutritional, industrial and medicinal benefits (Bruneel et al., 2013). Among the unsaturated fatty acids, omega-3 fatty acids stand out by having a key role in foetal growth and prevention of cardiovascular diseases, being a potent antithrombotic and anti-inflammatory agent (Galli & Marangoni, 2006). Because of its functionality, the food industry has sought sources of vegetable oils that contain a high amount of omega-3, as well as a good ratio between omega-3 and omega-6 (Dubois, Breton, Linder, Fanni, & Parmentier, 2007).

Chia seeds contain about 30% oil and the oil fraction is primarily composed of unsaturated fatty acids. These seeds contain the highest percentage (60–68%) of α-linolenic acid when compared to other seeds, e.g. flaxseed (57%) (Ayerza & Coates, 2011). In recent years, the use of the chia seeds has been widespread and their consumption has increased considerably due to the health benefits associated with the high content of α-linolenic acid (Ayerza & Coates, 2011). Foods rich in omega-3, such as chia oil, can be considered potential functional products once omega-3 fatty acids are essential in the human diet, even though there is a mechanism in humans for producing these fats from other substances. Omega-3 fatty acids act as precursors of hormones that are important to the maintenance of metabolic functions (Calder, 2006) and bone homeostasis (Al-Nouri, Al-Khalifa, & Shahidi, 2012).

Chocolate milk has a good combination of proteins, electrolytes and carbohydrates for post-workout recovery. The ingestion of protein and carbohydrates can promote more pronouncedly the glycogen synthesis and improve the performance when compared to other beverages containing only carbohydrates (Valentine, Laurent, Saunders, Todd, & Flohr, 2006), and thus chocolate milk can be a valuable source for replenishing energy to athletes who perform vigorous exercise (Karp et al., 2006). It should also be noted that omega-3 fatty acids may have anti-inflammatory and antioxidant properties in athletes who perform intense and exhaustive exercise, which is recognized to be a potent generator of inflammation, free radicals and cell damage. The environmental damage induced by exercise can cause delayed-onset muscle soreness, which is characterized by a painful condition experienced after frequent exhaustive and intense exercises (Lembke, Capodice, Hebert, & Swenson, 2014). The objective of this study was to assess the effects of an omega-3 enriched chocolate milk on some biochemical parameters and muscle damage in sedentary rats submitted to intense and acute exhaustive exercise. Chia oil was used as the source of omega-3 due to its suitable balance between omega-3 and -6 fatty acids.

2. Materials and methods

2.1. Animals

Forty-eight male Wistar rats (specific-pathogen free) from the Multidisciplinary Center for Biological Research (University of Campinas, SP, Brazil) were maintained under controlled conditions (temperature: 22 °C, humidity 55%, reverse 12-hour light/dark cycle) in individual cages with free access to a commercial chow (Labina, Purina, Brazil) and water, until they reached 150 g of body mass. The Ethics Committee on Animal Experimentation of the University of Campinas approved all (n. 0654013) experimental procedures. The animals were randomly assigned to one of 4 experimental groups and received the following treatments for 15 days: (a) sedentary rats fed a chocolate milk; (b) sedentary rats fed an omega-3 enriched chocolate milk; (c) exercised rats fed a conventional chocolate milk; (d) exercised rats fed an omega-3 enriched chocolate milk. Wistar rats received the beverages (v = 4 ml/day) by gavage. This study adhered to the animal care standards of the American College of Sports Medicine.

2.2. Exhaustion protocol

The rats were familiarized with the treadmill by running for 10 min at 10 m/min on the day before being brought to a state of exhaustion. The exhaustion test was applied following a time–speed procedure: the exhaustion session was: 1–90 min at 15 m/min; 91–100 min at 20 m/min; 150 min to exhaustion at 22 m/min (Lollo et al., 2012). All groups performed the same protocol.

2.3. Chocolate milk processing

For the production of the conventional chocolate milk, the following ingredients were used: pasteurized milk, partially demineralized sweet whey powder (Cargill, São José do Rio Pardo, Brazil), alkalized cocoa powder (Cargill, Porto Ferreira, Brazil), stabilizer Avicell (FMC, Campinas, Brasil), vanilla and molasses flavor (Firminich, Genebra, Suíça), saccharose (União, Sertãozinho, Brazil) aspartame (Chicago, IL, USA), neotame (SweetMix, Sorocaba, Brazil), sucralose (SweetMix, Sorocaba, Brasil), stevia 95% rebaudioside (SweetMix, Sorocaba, Brazil), Blend made of sucralose/acesulfame K/neotame (5:3:0.1, w/w/w) (SweetMix, Sorocaba, Brazil) and chia oil (Benexia, Santiago, Chile).

Two chocolate milks were formulated. Table 1 shows the ingredients used in the formulation of the beverages. The chocolate powder was mixed with water and milk under intense agitation. Then the stabilizers were dispersed under intense agitation for 10 min and whey powder, cocoa and vanilla and molasses flavors were added. The formulation containing chia oil was added at approximately 50 °C and homogenized (6000 rpm/3 min in a L5M-A homogenizer, Silverson, East Longmeadow, MA, USA). These ingredients were homogenized and subjected to UHT sterilization for 5 min. The chocolate milks were packaged in aseptic plastic bottles of 0.5 L.

2.4. Antioxidant capacity

Following the recommendation of Powers, Smuder, Kavazis, and Hudson (2010), ferric reducing antioxidant power (FRAP) and oxygen radical absorbance capacity (ORAC) assays were performed in aqueous and ethanolic extracts. Firstly chocolate milk samples were freeze-dried and ground to a fine powder. To obtain the aqueous and ethanolic extracts, powdered samples were suspended in deionized water or 80% ethanol solution (1% w/v), respectively. The extracts were shaken for 30 min, centrifuged at 35,735 × *g* for 30 min at 10 °C, filtered through