

Cepero González, M.; Padial Ruz, R.; Rojas Ruiz, F.J.; Romero Sánchez, D. y De la Cruz Márquez, J.C. (2016). Efectos de bebidas carbohidratadas y proteicas sobre la recuperación del esfuerzo / Effects of Carbohydrate-Protein Beverages on Recovery of the Exercise. Revista Internacional de Medicina y Ciencias de la Actividad Física y el Deporte vol. 16 (62) pp.373-401 [Http://cdeporte.rediris.es/revista/revista62/artefectos701.htm](http://cdeporte.rediris.es/revista/revista62/artefectos701.htm)

REVISIÓN / REVIEW

EFFECTS OF CARBOHYDRATE AND PROTEIN BEVERAGES ON RECOVERY FROM EXERCISE

EFECTOS DE BEBIDAS CARBOHIDRATADAS Y PROTEICAS SOBRE LA RECUPERACIÓN DEL ESFUERZO

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Código UNESCO / UNESCO Code: 2411.06 Fisiología del Ejercicio / Exercise Physiology, 3206.02 Metabolismo Energético / Energy Metabolism.

Clasificación Consejo de Europa / Council of Europe Classification: 2: Bioquímica del deporte / Biochemistry Sport, 6: Fisiología del ejercicio / Exercise Physiology, y 11: Medicina del Deporte / Sports Medicine.

Recibido 14 de abril de 2013 **Received** April 14, 2013

Aceptado 5 de enero de 2014 **Accepted** January 5, 2014

ABSTRACT

This manuscript undertakes a review of the effects resulting from the intake of proteins and/or amino acids administered in carbohydrate drinks as well as whey protein and casein protein on recovery and the parameters of muscle damage during prolonged resistance exercise. The research was conducted in April 2013 in the databases of ISI Web of Knowledge, SCOPUS, PubMed, Medline, SportDiscus, and databases on Spanish networks CINDOC CTI-CSIC, RESH, DICE, and DIALNET crossing the descriptors "Exercise", "Resistance training" and "Recovery" with the terms "Ergogenic Beverage", "Casein Protein" and "Whey Protein". The most popular nutritional strategies are based on a carbohydrate beverage which combines different protein sources on prolonged exercise tests similar to those found in both team and individual sports with contrastive results.

KEY WORDS: Ergogenic Beverage, Recovery, Casein Protein, Whey Protein.

RESUMEN

Este artículo aporta una revisión del efecto de la coingesta de proteínas y/o aminoácidos administrados en bebidas carbohidratadas, la proteína de suero de leche y proteína caseína, sobre la recuperación y los parámetros del daño muscular en ejercicios de larga duración. La búsqueda se ha realizado en abril de 2013 en las bases de datos del ISI Web of Knowledge, SCOPUS, Sport Discuss, PubMed, Medline, Sportdiscus, y en las bases de datos CINDOC en las redes CTI-CSIC, RESH, DICE y DIALNET cruzando los descriptores “Exercise”, “Resistance training” y “Recovery” con los términos “Ergogenic beverage”, “Casein Protein” y “Whey Protein”. La estrategia nutricional más respaldada es la ingesta de un preparado líquido carbohidratado en donde se combinan proteínas de diferentes fuentes sobre pruebas de esfuerzos prolongados similares a la competición tanto en deportes individuales como en colectivos, con resultados discrepantes.

PALABRAS CLAVES: Bebida ergogénica, recuperación, proteína caseína, proteína de suero de leche.

1. INTRODUCTION

There is an increase of the plasma free fatty acid levels during prolonged exercise and all those situations where the deposition of glycogen is very low because fats are in such circumstances responsible for the supply of most of the energy. There is also an increased use of the branched-chain amino acids by muscles as sources of energy to the extent that their concentration in the bloodstream decreases.

The intake of nutrients following a strenuous exercise session affects the anabolic processes irrespective of the type of exercise. Proteins and carbohydrates are of paramount importance since these two macronutrients represent different functions as anabolic agents. There is no doubt that proteins and the capture of amino acids resulting from the ingestion are necessary in order to achieve positive protein/nitrogen balance whereas the intake of carbohydrates during recovery is considered as the most important means for the restoration of glycogen after strenuous exercise (Jeukendrup and Jentjens, 2000; Saunders, Kane and Todd, 2004; Van Loon, et al. 2000ab).

There are various factors that play an important role in the efficiency of protein and carbohydrates on the glycogen synthesis after exercise, so the inappropriate ingestion of these factors may undermine the capacity to reach an anabolic state. The evidence provided about the ingestion before and after the exercise clearly denote the importance of these two macronutrients as regards nutrition and anabolism after the exercise (Betts, and cols. 2007; Poole, et al. 2010).

Although most athletes may satisfy their nutritional needs before and/or after the exercise, prolonged activities require a nutritional supply during the exercise. Resistance exercises demand the use of a higher amount of energy, which leads to significant increases in the consumption of carbohydrates and fat oxidation. Additionally, there may be considerable losses of liquids and electrolytes due to sweating especially during prolonged exercise in a hot environment. Subsequently, the inappropriate intake of liquids and nutrients during the performance of the resistance exercise may result in dehydration, hyponatremic hyperhydration, glycogen depletion, hypoglycemia and central fatigue. Moreover, the nutritional deficiencies during a prolonged activity may undermine the capacity of a quick recovery after the exercise, which may affect performance later (Betts et al. 2007; Jentjens and Jeukendrup, 2003; Saunders, 2007).

Several studies have been conducted into the issue of nutritional needs in an attempt to clarify the points raised above, which have led to two nutritional strategies that provide positive effects for resistance athletes:

- 1) Carbohydrate consumption. There is a clear general consensus in the bibliography on the benefit of carbohydrate intake during prolonged exercise (2 hours or over) that almost always delays the start of fatigue and improves performance such as in activities of

short duration but higher intensity. For instance, continuous exercise that lasts for about 1 hour and high intensity intermittent exercise (Jeukendrup, 2007). The effects of post-exercise consumption of high-molecular-weight versus low-molecular-weight carbohydrate solutions on recovery following high-intensity interval training seem to be insignificant (McGlory and Morton, 2010).

- 2) Ingestion of carbohydrate and protein beverages (CHO + P): This strategy is the real concern of this revision as it is being used more regularly by athletes to improve performance in resistance exercises and since it reduces the signs of muscle damage and also improves recovery after the exercise, but there are significant methodological differences between the different references.

Some studies reveal the effect of the addition of proteins and/or amino acids in carbohydrate sports drinks on physical performance (table 1). Most of them show an improvement related to that addition (Betts et al., 2007; Burke, 1999; Fogt and Ivy, 2000; Ivy, et al. 2003; Moore et al., 2007; Niles et al., 2001; Ready, Seifert and Burke, 1999; Saunders et al., 2009, 2006; Saunders, Luden and Herrick, 2007; Saunders, Kane, and Todd, 2004; Schedl, Muaghan, and Gisolfi 1994; Williams, Ivy and Raven, 1999; Williams, et al., 2003; Zawadzki, Yaspelkis and Ivy, 1992) whereas others do not show any difference between the protein and/or amino acid plus carbohydrate supplementation in contrast with those who only use carbohydrates (Anderson, 2001; Breen, et al. 2010; Cepero et al., 2009, 2010; Davis, Welsh and Alerson, 2000; Gasier and Olson, 2010; Osterberg, Zachwieja and Smith, 2008; Romano-Ely et al. 2006; Skillen et al., 2008; Tonne and Betts, 2010; Van Essen and Gibala, 2006; Valentine et al., 2008; Van Hall, et al. 1995).

Neither have we noticed improvements in muscle injuries induced by exercise through the ingestion of carbohydrate drinks with added proteins (Green, et al. 2008) even though they do restrict the perception of pain after exhaustive aerobic exercise (McBrier et al. 2010) and stimulate the synthesis of skeletal muscle protein if they are ingested during the recovery from resistance exercise (Howarth, et al. 2009).

Table 1. Comparison of the characteristics of the drinks in representative research on the performance in resistance.

| Study | Liquid (ml/h) | Drink | CHO (g/h) | Proteins(gr/l) | Type of protein | Meaning |
|-------------------------|---------------------------------------|-------------------------|----------------|-----------------|---------------------------------|---------|
| Cepero et al. (2010) | 1000 | CHO CHO+CP CHO+WP | 9% 7% 7% | 0 2% 2% | Casein protein Whey protein. | No |
| Gasier and Olson (2010) | 600 ml in 5 doses 3000 ml in total | CHO CHO+WP | 8,9% 1,81% | 0 7,22% | Whey protein. | No |
| Tonne and Betts (2010) | 1053 ml + 75 ml | CHO CHO+P | 95 72+ 5 | 0 22+-2 | Whey protein | No |
| Cepero et al. | 1000 | CHO | 9% | 0 | Casein protein | No |

| | | | | | | | |
|---|---|--|----------|-----------|----------|--|----------------------|
| (2009) | | | CHO+P | 7% | 2% | | |
| Saunders et al. (2009) | 200 ml every 5 Km+150 ml after exercise | | CHO | 60 | 0 | Hydrolyzed casein protein. | Yes in the last 5km. |
| | | | CHO+P | 60 | 14,4 | | |
| Osterberg et al. (2008) | 250 ml each 15 minutes. | | CHO | | | | No |
| | | | CHO+P | | | | |
| Skillen et al. (2008) | 1500 (500 ml before, during and after) | | CHO | 23 (4,6%) | 0 | Leucine, valine, isoleucine and arginine | No |
| | | | CHO+AA | 18 (3,6%) | 5 (1%) | | |
| Valentine et al. (2008) | 250 ml each 15 minutes. | | CHO | 7,75% | 0 | Concentrated whey | No |
| | | | CHO+CH O | 9,69% | 0 | | |
| | | | CHO+P | 7,75% | 1,94% | | |
| Betts, Williams, Duffy and Gunner, (2007) | 0.8-1.1 grams of CHO/Kg of mass | | CHO | 8% | | Whey protein | No. |
| | | | CHO+CH O | 8% | | | |
| | | | CHO+P | 8% | 0,3 g/kg | | |
| Moore et al. (2007) | 577 | | CHO | 35 | 0 | Hydrolyzed casein | Yes |
| | | | CHO+P | 35 | 7 | | |
| | | | CHO+P | 35 | 14 | | |
| Saunders et al. (2007) | 560 | | CHO | 41 | 0 | Concentrated whey | Yes |
| | | | CHO+P | 41 | 10 | | |
| Saunders et al. (2007) | 1000 | | CHO | 60 | 0 | Hydrolyzed casein | Yes |
| | | | CHO+P | 60 | 18 | | |
| Romano-Ely et al. (2006) | 600 | | CHO | 56 | 0 | Concentrated whey | No |
| | | | CHO+P | 45 | 11 | | |
| Van Essen and Gibala. (2006) | 1000 | | CHO | 60 | 0 | Isolated whey | No |
| | | | CHO+P | 60 | 20 | | |
| Saunders et al. (2004) | 508 | | CHO | 37 | 0 | Concentrated whey | Yes |
| | | | CHO+P | 37 | 9 | | |
| Ivy et al. (2003) | 600 | | CHO | 47 | 0 | Concentrated whey | Yes |
| | | | CHO+P | 47 | 12 | | |

Note: CHO= Carbohydrate drink; CHO+P; Carbohydrate + protein drink; CHO+AA; Carbohydrate + Amino Acid drink.

Many female athletes reduce their consumption of energy, more specifically fat consumption, in order to modify their body composition, but such a nutritional practice is usually not advisable. As opposed to men, women appear to be less dependent on glycogen during exercise and less sensitive to carbohydrates, because of glycogen synthesis, during recovery. Resistance female athletes may require more proteins than their male counterparts in order to achieve a positive nitrogen balance and boost the protein synthesis. Therefore, resistance female athletes need to focus less on carbohydrate consumption and focus more on the quality of the proteins and fat consumption within the context of energy balance in order to improve the adaptations to the training as well as general health and wellbeing (Volek, Forsythe and Kraemer, 2006). Attention at the time of the ingestion of nutrients, the quality of macronutrients and dietary supplements such as creatine are still being discussed in the case of women.

As a result of those disagreements highlighted in various studies, this research aims to assess the effect of the ingestion of different carbohydrate beverages with protein supplementations on the recovery from physical exercise. As a matter of fact, this study is a revision of the most recent

investigations which dealt with the effects of whey protein (WP) and casein protein (CP) on performance and recovery from prolonged exercises. Furthermore, we wish to determine whether there is a consensus in the bibliography used as to whether the administration of carbohydrates with proteins improves its absorption and sport performance.

2. METHOD

2.1. *Bibliographical search strategy*

The bibliographical search approach was based on the analysis of secondary sources from encyclopedia, books and research reviews generally conducted on the athlete's diet related to sport performance in order to, from there, analyze the primary sources mentioned and expand the research to specialized databases using common descriptors and keywords in line with the purpose of this study (Thomas, Nelson, and Silverman, 2011). From a sequential viewpoint, the phases of this revision are as follow:

1. To analyze the generic bibliography on the athlete's diet using secondary sources (Antonio et al., 2008; González-Gallego, 2006; Kern, 2005; Lowery, 2012; MacLaren, 2007; Maughan and Murray, 2009; McDonalds, 2009; Westerterp, 2013).
2. To determine the keywords, topics and research descriptors: "Exercise", "Resistance training" and "Recovery" and to cross them with the terms "Ergogenic beverage", "Casein Protein" and "Whey Protein" in English as well as in Spanish.
3. To search for primary sources online through the electronic library of the University of Granada which allows to directly download the articles contained in the magazines to which the university has subscribed. We used the ISI Web of Knowledge, SCOPUS, PubMed, Medline and Sportdiscus. Among the national databases, we used the databases of the Scientific Information and Documentation Centre (CINDOC) affiliated to the institute of Documentary Studies on Science and Technology (IEDCYT) which belongs to the Centre of Social and Human Sciences (CCHS) of the Spanish National Research Council (CSIC) on the CTI-CSIC, RESH, DICE networks and the DIALNET platform for documentary resources and services of the University of La Rioja. With the same crossed terms, the identification of bibliographical references was performed until the month of April 2013 in the databases mentioned. All the articles were classified as revisions or experiments.
4. To read, analyze and assess the references found in the bibliographical searches.
5. To edit the bibliographical revision according to the classification performed earlier.

2.2. *Inclusion and exclusion criteria*

The following inclusion criteria were used for the final selection of the articles with which this revision is concerned:

- a) The article had at least to be found in a database in line with the crossed descriptors that were pointed out, preferably in the last ten years. However, articles were also selected as long as they had largely been used as a reference on previous occasions.
- b) The results had to be related to physical activity or sport and the participants had to be athletes irrespective of their age, gender or competitive level. Studies on animals were ruled out.
- c) The published article had to comply with the inherent characteristics of a scientific article. The contents published in magazines which were indexed in the Web of Knowledge and Scopus is a guarantee that the articles comply with the minimum quality requirements.
- d) The administration route for the supplements had to be oral, with it being the ideal way of processing the proteins, discarding those who had exclusively used the parenteral route (Manninen, 2004).

2.3. *Methodology of analysis*

A total of 2485 scientific articles were found, from which the main lines of investigation were identified, which were related to recovery after sport performance through the ingestion of drinks with casein protein or whey protein, with a limited number of 107 scientific references since they had at least already been quoted by other authors once according to the Web of Knowledge database.

The classification of the articles was carried out following the assessment of administration strategies of the drinks in order to lead the amino acids towards the bloodstream:

- 1) By ingesting food with total proteins;
- 2) Supplements with intact proteins;
- 3) By ingesting free amino acids; and
- 4) By ingesting hydrolyzed proteins, in which small chains of amino acids called peptides are produced.

3. RESULTS OF THE REVISION OF THE RELEVANT STUDIES AND INVESTIGATIONS

The following revision criteria have been established according to the classification of the procedures used in the ingestion of beverages with and without proteins, which was mentioned earlier:

3.1. Benefits of the intake of proteins and/or amino acids in carbohydrate sports supplements on recovery after sport performances

The addition of carbohydrates to a protein supplement is based on the stimulus of insulin secretion, which is crucial for the regulation of the absorption of glucose by tissues. Exercise allows to improve the response of the skeletal muscles to glucose causing their higher sensitivity to the effects of insulin (Richter, et al., 1989). Therefore, the combination of carbohydrates and proteins or amino acids in a supplement may contribute to a more effective absorption of the proteins and to an improved synthesis rate of muscle proteins (Koopman, et al., 2005).

On an empty stomach, a sedentary person weighing between 70 and 90 kg may lose approximately 40/60 g of proteins every day. Consequently, athletes, in accordance with the latest recommendations, ought to follow the following guidelines in order to ensure the repair, reshaping, adaptation and gain of lean mass, regardless of whether they are performing aerobic or resistance exercises (Philips, 2013):

- The daily intake of proteins have to be higher than the RDA (from 1,2 until 1,6 g of protein/kg of the athlete's weight/day).
- Dairy proteins need to be enriched with leucine.
- To consume proteins in doses of 20-25 g/ration in order to maximize the adaptation responses.
- To deliver the intake of soy flour throughout the day in a balanced way.
- To immediately consume the proteins after the exercise.

The capture of amino acids from ingested sources of proteins is variable (Tipton et al., 2007), and depends on the type of proteins (Tipton et al., 1999ab, Tipton et al., 2004; Wilkinson et al., 2007) or amino acids (Borsheim, Aarsland and Wolfe, 2004), of the ingested nutrients (Borsheim et al., 2004; Elliot et al., 2006; Miller et al., 2003), and the time of ingestion in relation to the exercise (Tipton et al., 2001; 2007).

Several studies have compared the effect of added proteins and/or amino acids to carbohydrate sports drinks on physical performance (table 1) as well as on recovery (Millard-Stafford et al., 2005). Many athletes have improved their performance with that supplement (Burke, 1999; Fogt an Ivy, 2000; Ivy et al., 2003; Niles et al., 2001; Ready, Seifert and Burke, 1999; Saunders, Kane and Todd, 2004; Saunders,

2007; Saunders et al., 2009; Schedl, Muaghan and Gisolfi, 1994; Williams, Ivy and Raven 1999, Williams, et al. 2003; Zawadzki, Yaspelkis and Ivy, 1992) even at the end of intermittent exercises, especially during football practice (Alghannam, 2011).

In most of the situations analyzed until now, the voluntary restriction of protein intake during the Ramadan for instance produced side effects, albeit limited, for muscle development as well as competitive performance (Shephard, 2012ab).

Other studies show no difference between protein and/or amino acid plus carbohydrate supplementations regarding drinks with only carbohydrates (Cepero et al., 2009, 2010; Davis, Welsh and Alerson, 2000; Gasier and Olson, 2010; Goh et al., 2012; Madsen et al., 1996; Osterberg, Zachwieja and Smith 2008; Romano-Ely et al., 2006; Skillen et al., 2008; Tonne and Betts, 2010; Van Essen and Gibala, 2006; Van Hall et al., 1995).

And some studies reveal that a mixture of carbohydrates and a moderate amount of proteins can improve aerobic resistance in the exercise carried out near the ventilatory threshold (Ferguson-Stegall, et al., 2010) and that mixture has proved beneficial for performance in extreme situations, especially when it is hot (Cathcart, et al., 2011).

There are studies which claim that the consumption of CHO and proteins during the first stages of recovery positively affects the performance of the following exercise and that it could be particularly beneficial for the athletes who take part in championships held on the same or consecutive days, which we therefore consider more consistent. This is a fact that we have to seriously take into account. In fact, despite the general belief that the oral intake of amino acids is not as effective for the stimulation of protein synthesis as it is with the intravenous infusion, there are studies that prove that both options do stimulate protein synthesis in a similar way (Biolo et al., 1997; Tipton et al., 1999ab). The practice according to which the intake of small amounts of proteins in the diet 5 or 6 times every day improves the synthesis of protein muscle is the most acclaimed.

Nevertheless, there very few studies that use exercise protocols with a length of more than an hour to analyze the ergogenic effects of protein on performance and the parameters of muscle damage since the vast majority of studies are either conducted using a cyclo-ergometer with exercises lasting a few seconds and recoveries of 24 seconds on average or using a race with a few sprints of less than 100 m with a 25-second recovery on average.

3.1.1. Essential amino acids v non-essential amino acids

One of the commonly mentioned benefits of the amino acid supplementation is that certain amino acids (arginine, histidine, lysine, methionine, ornithine and phenylalanine) may stimulate the release of growth hormone, insulin and/or glucocorticoids and may so stimulate the anabolic processes (Kreider, Miriel and Bertun, 1993).

Tipton et al., (1999ab) pointed that the essential amino acids are more effective than the non-essential ones and that the consumption of essential amino acids after training is as effective as the mixture of non-essential amino acids with carbohydrates for the stimulation of protein synthesis (Naclerio, 2007 and Rasmussen et al., 2000) (table.2).

Tipton et al., (2001) have proved that the simultaneous intake of essential amino acids and carbohydrates in solution, one or three hours after the training session, is able to cause an increase of up to 400% in the protein synthesis when compared to normal values at ease.

More specifically Beelen et al., (2010) conclude that the consumption of approximately 20 g intact protein, or the equivalence of around 9 g of essential amino acids during the first hours of recovery after the exercise, improves the synthesis of muscle proteins.

However, the oral intake of non-essential amino acids such as arginine combined with CHO is unsuccessful at achieving an increase in the plasma insulin levels (Van Loon, et al., 2000c) and the speeds of muscle glycogen synthesis (Yaspelkis and Ivy, 1999) in contrast with the intake of only CHO. Therefore, amino acids are also successful at increasing protein synthesis rate but they appear to be more effective if consumed shortly before the training session than after it (Hoffman, 2007) (Table 3).

3.1.2. Hydrolyzed proteins

They are proteins that have been broken down into their most basic components, amino acids, in an aqueous solution. Hydrolyzed proteins contain di- and tri-peptides which are absorbed more quickly than free amino acids and even more quickly than intact proteins (Di Pasquale, 1997). It was noticed that the intake of hydrolyzed proteins has a strong insulinotropic effect. Therefore, the sports recovery drinks containing hydrolyzed proteins may have great ergogenic value (Manninen, 2004).

The most interesting findings on this matter are revealed by Manninen (2006) who demonstrates that nutritional mixtures containing hydrolyzed proteins with leucine and high glycemic index carbohydrates increase the secretion of insulin in comparison with preparations containing only high glycemic index carbohydrates. With them, the post-exercise hyperinsulinemia is sustained with an hyperaminoacidemia induced by the intake of the hydrolyzed protein and by leucine, which produces a better protein absorption. Thus, the consumption of post-exercise recovery drinks containing these nutrients may lead to the skeletal muscle hypertrophy and increased strength when combined with appropriate resistance training (Reitelseder, et al., 2010). However, the long-term effects on body composition and performance during exercise have not been determined.

The higher absorption rate of amino acids in the form of di-peptides and in contrast with a mixture of free amino acids, appear to be related to a higher amino acid transport capacity (Di Pasquale, 1997). That is to say that it is a benefit for those athletes who wish to maximize the transport of amino acids towards the muscles.

Van Loon et al., (2000b) proved that the intake of that mixture of highly insulinotropic hydrolyzed protein amino acids combined with a moderate CHO intake (0,8 g/kg/h), produced increased speeds in the muscle glycogen synthesis in comparison with the intake of only CHO.

However, there are no studies that can confirm if such an advantage has an impact in terms of a quicker increase in the muscle mass or improvement in the recovery. Nevertheless, the advantages mentioned earlier (greater amino acid absorption, greater biological value) are still attractive for consumers (Manninen, 2004).

In multiple studies conducted by Van Loon et al., (2000ac) in order to analyze the insulinotropic capacity of various free amino acids, hydrolyzed and intact proteins, the findings showed that the oral intake of hydrolyzed proteins and amino acids combined with carbohydrates produce an insulinotropic effect as big as 100% higher than that noticed with the intake of only carbohydrates. Van Loon et al., (2000ac) based their studies on the clarification of the type, combination and amount of free amino acids or protein sources required in order to maximize the response of insulin when a drink containing CHO is added. They proved that the intake of a drink containing a mixture of hydrolyzed wheat protein, free leucine and free phenylalanine (0,4 g/kg/h) combined with CHO (0,8 g/kg/h) considerably increased the insulin levels without causing any gastrointestinal disorders. Mixtures containing big amounts of free amino acids (arginine, leucine, phenylalanine and glutamine) produced similar or even higher insulin levels, but these mixtures were not pleasant and caused gastrointestinal disorders (Van Loon et cols., 2000c).

3.1.3. Branched- chain amino acids (BCAA; leucine, isoleucine and valine)

These cannot be synthesized by humans and must be ingested in the diet. They are found in meat, eggs, milk, cheese, fish, etc. at a rate of some 15-20 g/100 g of protein (Burke et al., 2012).

The anabolic effect of BCAA in the human skeletal muscle was first demonstrated with the subject at ease, then studies that show similar effects in the recovery period after some resistance exercises (Tipton and Wolfe, 2004). Under certain circumstances, the BCAA supplements can improve physical performance even though most studies have not found any effect on performance when they are ingested with carbohydrates.

The recommended amount of BCAA is 0,03-0,05 g/kg of body mass per hour or 4,2 g/hour ingested several times during the exercise and recovery, preferably as a drink.

The ergogenic effects and those on the physiological responses to exercise of the BCAA supplementation are as follow:

- Reduction of protein degradation induced by exercise and/or the release of muscle enzymes (a sign of muscle damage) which possibly enhances an anti-catabolic hormonal profile (Carli et al., 1992; Wagenmakers, 1998).
- The effect of the BCAA, especially leucine, is mediated through the activation of the enzymes responsible for the regulation of the protein synthesis mechanism (Karlson et al., 2004).
- The increase of the BCAA plasma level during exercise can reduce the transport of tryptophan in the brain and the synthesis of 5-hydroxytryptamine (5-HT). The 5-HT is generally believed to be at the center of general fatigue, that is to say, fatigue from the brain related to muscle exercise (Newsholme and Blomstrand, 2006). The BCAA supplementation sustained during physical activity has produced positive effects on cognitive performance and the perception of exercise (Newsholme et al., 1991; Portier et al., 2008). An improvement in central fatigue was mentioned by Newsholme et al., (2006) and Blomstrand, Hassmen and Newsholme (1991) who claimed that resistance activities and mental performance could increase with the intake of BCAA, but later studies did not find any improvements in this performance with the administration of BCAA (Madsen et al., 1996; Van Hall et al., 1995).
- Oxidative improvement. During resistance exercises, the BCAA are more absorbed by the muscles than the liver in order to contribute to the oxidative metabolism. Without exogenous supplementation, the BCAA source for the muscle oxidative metabolism during exercise is the BCAA plasma stock, which is completed through the full protein catabolism during the resistance exercise (Davis, 1995; Kreider, 1998; Newsholme et al., 1991).
- Undesirable effects. McLean, Graham and Saltin, (1996), Madsen et al., (1996) and Van Hall et al., (1995) have proved that the intake of BCAA causes some negative metabolic effects such as the increase of ammonium in the plasma levels leading to effects of mental confusion during high levels of exercise, especially when very high doses are ingested (30 g per day) although they are tolerated at the gastric level.

Table 2. Classification of the essential and non-essential amino acids. * Amino acids considered essential in certain special circumstances where the organic demands are increased (training, competition, etc.). Naclerio (2007)

| Essential amino acids | Non-essential amino acids |
|-----------------------|---------------------------|
| Phenylalanine | Aspartic Acid |
| Isoleucine | Glutamic Acid |
| Leucine | Alanine* |
| Lysine | Arginine* |

| | |
|------------|------------|
| Methionine | Asparagine |
| Threonine | Cysteine* |
| Tryptophan | Glycine |
| Valine | Glutamine* |
| | Proline |
| | Histidine* |
| | Ornithine |
| | Prolina |
| | Serine |
| | Taurine* |
| | Tyrosine* |

3.2. The intake of Whey Protein (WP) and Casein Protein (CP) combined with Carbohydrates

The proteins found in milk are distributed in two components:

- a) Whey protein (WP).
- b) Casein protein (CP).

Both proteins derive from milk, but each protein is different in the absorption rate and the bioavailability. Therefore, each type of protein may contribute in a different way to the adaptations generated through resistance training in particular.

3.2.1. Whey proteins (WP)

Cow milk protein contains high levels of essential and branched-chain amino acids and is absorbed more quickly than casein. The whey proteins provide cysteine (2,5%), sulphur-containing amino acid and initiator of the glutathione synthesis (essential antioxidant that protects the organism against the damage caused by the production of free radicals) and other micro fractions that reinforce the release of growth factors such as somatomedin (IGF-1) which stimulates recovery and muscle growth. It lacks phenylalanine (essential amino acid with crucial functions for the synthesis of brain neurotransmitters), glutamine, arginine, and taurine, which are amino acids considered essential in highly physically demanding situations (Di Pasquale, 1997, Naclerio, 2007).

Therefore, in accordance with this analysis, the manufactured preparations based on whey proteins should be strengthened with phenylalanine, glutamine peptides, arginine and taurine (Di Pasquale, 1997, Hoffman and Falvo, 2004).

3.2.2. Casein protein (CP)

Casein is a protein which is commonly found in milk making up to 80% of its content. It exists in the form of micelles, which are big colloidal particles. Casein micelle produces gel in the stomach, which slows down its digestion. As a result,

casein provides a sustained but slow release of amino acids in the bloodstream, which sometimes lasts for several hours (Boirie et al., 1997). That means a better retention and use of nitrogen.

Like the proteins found in whey, casein is a complete protein containing calcium, phosphorus and other minerals (Hoffman and Falvo, 2004). In some studies, casein produced a more sustained and effective response as opposed to whey proteins which were absorbed more quickly (Dangin et al., 2002, Kerksick et al., 2006, Tipton et al., 2004).

3.2.3. Differences between Whey Protein (WP) and Casein Protein (CP)

Whey as well as casein proteins are essential for an appropriate nutrition. However, whey protein can be absorbed quickly whereas casein releases amino acids in the bloodstream in a more sustained way. Moreover, calcium, vitamins D and A can be supplied in a balanced way through the mixture of whey and casein proteins (Wein and Miraglia, 2011).

Both casein and whey are complete proteins, but their amino acid composition is different. More specifically, leucine content, which plays an important role in the metabolism of muscle proteins, is higher in whey than in casein. Thus, the digestion rate of proteins can be more important than the amino acid structure of proteins. These results were supported by Tipton et al., (2001), who also pointed that the differences in the digestive characteristics between casein and whey result in a minor synthesis of muscle proteins with casein. However, the net synthesis of muscle proteins in a 5-hour period was not different between both proteins when the intake (20 g of each protein) was performed one hour after the overload training session.

Several studies have compared the benefits from the intake of both types of proteins using hydrolyzed whey proteins in order to determine the differences between the whey and casein proteins (Cepero et al., 2009; 2010 and Hoffman, 2007). Table 3 shows the types of proteins used and the results of the most significant studies.

The most relevant study was conducted by Zawadzki, Yaspelkis and Ivy (1992) who, with the intake of a combined CHO+whey supplement, noticed an increase in the speed of the glycogen synthesis during a 4-hour period after the exercise in contrast to a CHO supplement only. Highton et al., (2013) insist on the advantages of a CHO (6%) + WP (2%) solution in comparison with a CHO (8%) solution as a way of improving the average speed of resistance exercise.

Boirie et al., (1997), after comparing the casein supplementation with that of whey, showed that the intake of 30 g of casein against 30 g of whey produced significantly different effects on the gain of post-prandial proteins. He showed that the production of amino acids in plasma is more rapid after the whey intake; It is of higher magnitude but more transitory. In contrast, casein is absorbed far more slowly, producing a far less increase in the plasma concentration of amino acids. The intake of whey proteins stimulated the synthesis of proteins by 68%

while the casein intake stimulated the protein synthesis by 31%. When the researchers compared the leucine post-prandial balance 7 hours after the intake, the casein consumption resulted in a significantly higher leucine balance whereas while no changes were noticed in relation to the basal value after the consumption of whey. These findings suggest that whey stimulates a rapid protein synthesis, but most of these proteins are oxidized (used as fuel) whereas casein produces a greater protein accumulation during a more prolonged period of time.

Tipton et al., (2001, 2004), Boire et al., (1997) Dangin et al., (2002), Hoffman and Falvo (2004) and Hoffman (2007) agree that the ingestion of whey protein is more efficient than casein protein. Whey protein can provide a greater instant increase in the rate of protein synthesis. Nevertheless, the mixture of whey and casein proteins can prove effective for the production of instant and prolonged increases in the protein synthesis rate.

Colombani et al., (1999) compared the metabolic consequences of the CHO and CHO+P consumption using hydrolyzed whey proteins during a marathon. He noticed an increase in the levels of plasma amino acids during the marathon with the CHO+P supplement, without any alterations in the ammonium levels, which is a reminder of fatigue, whereas Van Hall, Shirreffs and Calbet (2000) found no differences in the release of insulin between the intake of a carbohydrate drink (sucrose) and the same drink containing whey protein.

Hoffman (2007) has analyzed the differences between the whey and casein intake in protein accumulation pointing that both can have different digestive characteristics. But, apparently both whey and casein are effective proteins for the stimulation of muscle protein synthesis. At the same time, the differences in the digestive characteristics of proteins result in a different pattern of protein synthesis with the whey intake and in a greater and faster response in comparison with a more gradual protein synthesis after the casein intake. Although the net total synthesis of muscle proteins appears to be similar between both proteins, it is unclear whether the fast increase noticed after the whey intake represents a greater advantage for the improvement of the recovery and reshaping of the skeletal muscles (Hoffman, 2007).

Hoffman and Falvo (2004), Tipton and Wolfe (2004), Tipton et al., (2004) have assessed the differences in the organic responses that are determined with the intake of whey or casein proteins. They have found significant differences in the post-prandial absorption speed caused by a slower and more sustained assimilation of casein proteins in contrast to whey even though the results of the whey or casein intake shortly after a resistance training session produce a similar response in the muscle protein synthesis in spite of the temporary differences in insulin and amino acid concentrations.

Wilborn et al., (2013) have conducted a study into women who practice basketball. They ingested 24 g of WP before and after the exercise as opposed to others who ingested 24 g of CP with the same exercise protocol during 24 weeks. He noticed some changes in both groups in the fat mass, limbs strength lower than 1RM, high jump and long jump, but there seems not to be a difference between WP and CP (table 3).

Thus, a few hours shortly after the intake of whey proteins, they showed a net higher balance of muscle proteins. Most of the amino acids captured by the muscle may be oxidized instead of being used as raw material for the production of an increased protein synthesis (Hoffman and Falvo, 2004). Accordingly, it was suggested that in order to assess the long-term metabolic effects, the analysis of the plasma absorption rate of amino acids caused by the intake of different proteins should be a more important factor than the structure (Tipton and Wolfe, 2004).

Dangin et al., (2002) showed that the continuous intake of whey proteins (a similar amount of proteins but ingested in a prolonged period of time of 4 hours in comparison with a single intake) produced a better net leucine oxidation than a single whey or casein intake. The fractioned intake provides a more sustained flow of amino acids and improves muscle anabolic response even in contrast to when the same amount of proteins is ingested using casein as the source.

According to this, the best way of supplying whey proteins in order to enhance the anabolic effects is to ingest small doses of whey proteins (2,3 g) every 20 minutes during 2 hours, (Bilsborough and Mann, 2006) since the maximum rate of protein synthesis stimulated by the increasing flow of amino acids was established between 6-7 g per hour.

This flow level is achieved with a single intake of casein proteins (although it takes more time to achieve it) or with sustained supply of whey proteins, which when ingested in small and frequent doses that do not cause a sudden increase or decrease of its concentrations like the ones observed when a single dose of 20-30 g is ingested (Bilsborough and Mann, 2006).

Pérez-Guisado (2009), sustains that the best protein mixture is the that containing whey and casein proteins (in an approximate proportion of 4 and 1 respectively), which is even higher than the mixture of whey-amino acid branched-chain glutamine proteins (Kerksick et al., 2006). As regards whey protein, although it can provide an instant greater increase than casein in the protein synthesis rate, the mixture of both has the advantage of producing instant and prolonged increases in that protein synthesis rate (Hoffman, 2007). If we intend to maximize the recovery of the consumed muscle glycogen, we should continue with the intake of carbohydrates at an approximate pace of 1,2 g/kg/hr (Ivy, 2004).

Table 3. Most representative studies comparing the whey protein (WP) and casein protein (CP) intake.

| STUDIES | CHO+P AND CHO DRINKS COMPARISON | RESULTS |
|------------------------|--------------------------------------|---|
| Highton et al., (2013) | CHO 8% v CHO 6% + CP 2% | Improved race average speed with CHO+WP |
| Wilborn et al., (2013) | WP v CP Pre/post-resistance exercise | Post-exercise improvements but without difference between WP an CP. |

| | | |
|---|--|--|
| Pérez-Guisado (2009) Hoffman (2007) | Combination of WP+CP | The best proportion to combine WP and CP is 4 and 1 respectively. More effective production of instant and prolonged increases in protein synthesis. |
| Bilsborough and Mann (2006) | WP | Best whey administration method is 2,3 g/20 minutes during 2 hours. |
| Tipton et al., (2004) Hoffman and Falvo (2004) | WP v CP | WP is more absorbed more quickly than CP whose absorption can last for hours. |
| Tipton and Wolfe (2004) | Combination of WP+CP | More effective production of instant and prolonged increases in protein synthesis. |
| Calbet & McLean (2002) | CHO+WP v total milk proteins | Greater absorption of hydrolyzed peptides through the intestine and higher availability of AA during the post-prandial period with WP. |
| Dangin et al., (2002) | WP v CP WP administered during 4hrs v 1 single intake of WP and CP. | Higher net leucine oxidation with the prolonged intake of WP than with a single intake of WP or CP. |
| Tipton et al., (2001) | WP v CP | Greater leucine content with WP. |
| Boire et al., (1997) | WP v CP | A more rapid and higher-magnitude appearance of amino acids in plasma and transitory WP. However, CP obtained a higher increase of leucine 7 hours later. |
| Zawadzki, Yaspelkis & Ivy, (1992) | CHO CHO+WP | Higher speed in the synthesis of glycogen during 4 hours with CHO+WP. |

Calbet and McLean (2002) noticed that the combined administration of glucose and hydrolyzed proteins stimulates the synergetic release of insulin without considering the source of proteins. These authors concluded that hydrolyzed peptides are absorbed at a higher rate in the small intestine than the total of whey proteins administered as a milk solution, which is reflected as a rapid increase in the plasma concentration of branched-chain amino acids in peripheral blood (table 3). Therefore, we can claim that the metabolic benefits obtained with a protein supplement or balanced amino acid profiles are higher than when BCAA supplements are ingested.

Furthermore, hydrolyzed whey proteins produced a greater availability of amino acids during the 3-hour- post-prandial period. According to Calbet and McLean (2002), the combination of high levels of plasma amino acids and insulin could explain the superiority of the hydrolyzed peptides on all the other proteins in order to stimulate a better use of nitrogen, especially when they are administered in combination with glucose.

Most authors agree that the best strategy to properly enhance muscle anabolism would be the intake of a preparation in which different proteins from different sources are mixed up (Tipton and Wolfe, 2004). Consequently, muscle protein regenerates more efficiently after the intake of hydrolyzed whey or soy casein when at ease as well as after resistance exercise in young males (Tang et al., 2009). Although both proteins are absorbed quickly, the ingestion of hydrolyzed whey is quicker than that of soy after resistance exercise. These differences may be related to the speed with which proteins are ingested (that is to say, fast v slow) or possibly to minor differences in the leucine content of each protein.

4. CONCLUSIONS

- 1- The studies we have revised consider that the availability of carbohydrates is the main undermining factor during prolonged exercise, but have not found any studies that may determine the maximum absorption capacity of carbohydrates compatible with the administration of a greater amount of proteins in order to enhance performance during exercise.
- 2- The most effective drinks are those combining casein and whey proteins together with CHO proteins since they produce instant and prolonged increases in the protein synthesis rate. Amino acids are equally effective to increasing the protein synthesis rate even though they are more effective when they are ingested shortly before the training session, but in order to appropriately enhance muscle anabolism, the most suitable strategy is to ingest a preparation in which proteins from various sources are mixed up.
- 3- The most relevant studies suggest testing the drinks during resistance training sessions similar to competition in team sports with variable efforts as well as individual sports. The analysis contained in the revision reveals statistically significant differences in sport performance with resistance training protocols prolonged over an hour where the participants end up exhausted.

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