1.1 Classification of Nutritional Supplements

1.1.1 Useful Terms (pH, Absorption Mechanisms, Km from Receptors, Half-Life)

Supplement

The Oxford English Dictionary definition for supplement is: "Something that is added to make up for a deficiency" (Burke, Castell & Stear, 2009, p. 728).

In the FCB, supplements are commonly used among athletes and coaches to improve performance, for faster recovery and to improve overall health. It is important to note that supplements are rarely needed if the athlete's diet is healthy, varied and balanced.

There are exceptions where supplements can help performance or recovery, but in any case they would have to be consumed to supplement a healthy and balanced diet, never as a substitute. Thus, by definition, nutritional supplements should be used to supplement the diet, not to replace it (Jeukendrup & Gleeson, 2010).

The most important factor, if choosing to supplement, is that the chosen approach must comply with the code of conduct of the World Anti-Doping Agency (WADA). Specifically, it should ensure that all the supplements are free of banned substances.

Sports Foods

Sports foods are made up of the same nutrients as food, essentially carbohydrates, proteins and lipids. However, their portability, presentation and composition make them very practical for use at specific times in different sports, in order to improve performance and promote recovery.

Ergogenic Aids

According to the **International Society of Sports Nutrition (ISSN)**, an ergogenic aid is "any training technique, mechanical device, nutritional practice, pharmacological method, or psychological technique that can improve exercise performance capacity and/or enhance training adaptations" (Kreider et al., 2004, p. 2).

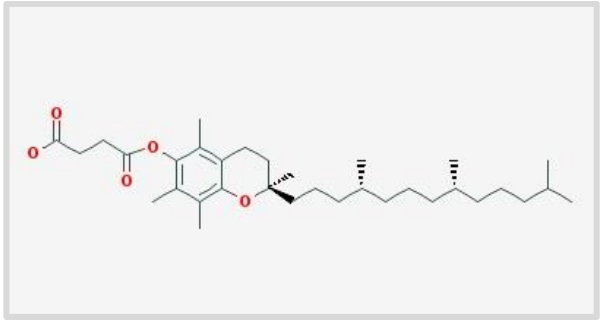
One can see that the definition is quite broad and covers many aspects. In the following course we will focus specifically on nutritional practices having to do with supplementation, which will not only allow for an increase in acute and chronic performance, but also increase tolerance to training, which in the long term may result in chronic performance improvement. Probably the most important aspect is that this improvement is achieved respecting the integrity of the immune system and supporting an improved level of recovery, which translates into better overall health for athletes of different levels, and thus a significant decrease in injuries and fatigue.

Chemical Structure

The branch of chemistry that studies carbon compounds (carbon chains with different functional groups) is called **organic chemistry** (Wade, 1993) and it is through this field that most ergogenic aids, chemical compounds and nutrients to be reviewed in these modules, may be analyzed.

The chemical structure of a molecule is composed of the atoms that form it, as well as their distribution in space. In organic chemistry, there are always carbon chains that consist of a different number of carbon atoms linked together and connected in turn to different atoms or functional groups such as acids, aldehydes and amino, among others. An example of a complex chemical structure of a molecule (or macromolecule in this case) is a protein, in which its primary structure indicates the amino acid sequence that shapes it. The secondary structure of a protein involves the folding of the chain of amino acids and its tertiary structure involves the arrangement in space of the whole molecule. Figure 1 shows the chemical structure of vitamin E (micronutrient).

Figure 1: Chemical Structure of Vitamin E



Source: Adapted from National Institutes of Health, 2005. Retrieved from https://goo.gl/yMxE6d

Functional groups

The reactive part of a molecule is called the **functional group,** because that is usually where the reactions take place (Wade, 1993). Among the functional groups of organic molecules that are of interest to the topics to be reviewed, one may consider, among others, the following:

- **Alcohols:** they are organic compounds that have the functional group hydroxyl (—OH).
- **Ethers:** they are formed by two radicals (R) linked to an oxygen atom (R—O—R').
- **Aldehydes and ketones:** the functional group is a carbonyl group (C=O).
- **Carboxylic acids:** contain the carboxyl group (—COOH).
- **Esters**: are formed from the reaction of a carboxylic acid with an alcohol (R—CO—O—R').
- **Amides:** are formed from the reaction of a carboxylic acid with an amino group (—NH2). Significantly, this type of chemical bond is what unites the different amino acids in a peptide or protein.

Physicochemical Properties

Among the inherent physicochemical properties of each molecule are: melting point, freezing point, solubility, molecular weight, specific heat capacity, among others. We shall describe molecular weight and some associated concepts.

Molecular Weight: numerically it equals the relative molecular mass of a molecule, expressed in daltons (Alberts et al., 1994).

The **dalton** is the unit of molecular mass. It is approximately equal to the mass of a hydrogen atom (1.66×10^{-24} g). One dalton equals one **atomic mass unit (amu).** The molecular mass of a water molecule is 18 daltons (16 from the oxygen and 2 from the hydrogen atoms composing the molecule).

The **relative molecular mass** is the mass of a molecule as a multiple of the mass of a hydrogen atom, which is equal to a dalton or an amu. Thus, the molecular mass of molecular nitrogen (N2) is 28.

Following is an introduction to some concepts related to the acid-base balance, in order to understand the characteristics of the medium where all biological reactions occur:

- **pKa and pKb:** Before giving the definition of pKa and pKb it is necessary to introduce some basic concepts.

- **pH:** by definition, is the negative logarithm of the concentration of protons in a given system (-log (H+)). The proton concentration ([H+]) must be expressed in units of molarity (M), that is, number of moles of each species (in this case protons) per liter of solution.

- **pOH:** by definition, is the negative logarithm of the hydroxyl concentration in a given system (-log [OH-]). The hydroxyl concentration ([OH-]) must be expressed in units of molarity (M), that is, number of moles of each species (in this case hydroxyls) per liter of solution.

- **Kw:** is the dissociation constant of water and is obtained from the product of [H+] and [OH-]. It has a constant value of 10^{-14} Thus, for any solution whose solvent is water, you have the following:

    - $Kw = [H+].[OH-] = 10^{-14}$.

- **pKw:** by definition, is the negative logarithm of the dissociation constant of water (-log (Kw)). It is important to note that $Kw = 10^{-14,}$ which means that pKw = 14. On the other hand, it is also noteworthy that pKw = pKa + pKb, for any weak acid and its conjugate base.
- **Ka and Kb:** the dissociation constant of an acid (organic acids of biological systems, such as lactic acid, pyruvic, acetoacetic, etc.), is defined as follows:

    - $Ka = [Conjugate\ Acid\ Base].[H_3\ O+] / [undissociated\ acid]$

    Where []: concentration; H_3O+: water molecule that has captured a proton.

    In the case of lactic acid, we have:

    - $HLac\ (ac) + H_2O <---> Lac-\ (ac) + H_3O+\ (ac)$

    Where, (ac): aqueous state, HLac: undissociated lactic acid Lac-: dissociated lactic acid (lactate), <--->: The reaction can proceed in either direction (this indicates that a dynamic chemical equilibrium can be reached).

    Therefore, the acid dissociation constant of an organic species indicates the tendency that it will have an acid to dissociate given an aqueous medium.

    Thus, by definition, the pKa is the negative logarithm of the acid dissociation constant (-log (Ka)). Notably, when the pH of a given

system is equal to the pKa of a given species, the concentration of the acid and its conjugate base are equal.

Furthermore, by definition, the pKb is the negative logarithm of the basic dissociation constant of a given chemical species (-log (Kb)). Notably, when the pOH of a given system is equal to the pKb of a given species, the concentration of the base and its acid conjugate are equal.

Henderson-Hasselbalch Equation

The Henderson-Hasselbalch equation can be derived from the first equation proposed in the section regarding Ka and Kb.

First, the negative logarithm is taken in base 10 (-log) to both terms of the equation:

- - log (Ka) = - log ([conjugate acid base].[H_3O+] / [undissociated acid]).
  If properties of logarithms are applied and considering that pKa = -log (Ka) and pH = - log ([H_3O+], we have:

- pKa = -log ([conjugate acid base] / [undissociated acid]) + pH.

Passing a term to the other side of the equation gives us:

- pH = pKa + log ([conjugate acid base] / [undissociated acid]).

This is effectively the equation that Henderson and Hassellbach derived, and it shows that it can be obtained from positing the equation of the equilibrium constant of a weak organic acid.

Biosynthesis

It constitutes the mechanism from which a given molecule is synthesized in a body tissue.

In the case of creatine, for example, its biosynthesis, according to what is generally accepted, occurs mainly in the liver in humans and involves two successive reactions where two enzymes are implicated (Greenhaff, 2000). The first reaction is catalyzed by the transamidinase glycine and involves the transfer of an amidine group from arginine to glycine to form the guanidinoacetic acid.

The second reaction involves the transfer of a methyl group from S-adenosylmethionine, a reaction that is catalyzed by the

guanidinoacetate methyltransferase, resulting in methylation of guanidoacetate to form methyl-guanidinoacetic acid or creatine. It is important to indicate that the two mentioned reactions are reversible.

Intestinal absorption

There are two kinds of membrane transport proteins, namely:

- **Transport proteins:** They bind to the specific solute to be transported and undergo a series of conformational changes that allow the transfer of solute through the membrane.
- **Channel proteins:** they do not bind to the solute, but rather form hydrophilic pores that cross the lipid bilayer formed by the cell membranes. When these pores are open they allow certain solutes to pass through them, and thus, may pass through the membrane (Alberts et al., 1994).

All channel proteins and many transport proteins only allow solutes to pass through the membrane passively in a process called **passive transport or facilitated diffusion.** If the transported molecule lacks electric charge (e.g., cholesterol or acetone), the passive transport direction is determined by the concentration difference on both sides of the membrane (concentration gradient). On the other hand, if the solute has a net charge (e.g., sodium ion or potassium), its transport is influenced by both its concentration gradient and by the electrical gradient through the membrane (membrane potential). The electrical gradient and concentration can be combined, thus allowing one to calculate the net force direction of flow or electrochemical gradient for each solute loaded (Alberts et al., 1994).

Cells also need transport proteins that actively pump certain solutes across the membrane against their electrochemical gradient. This process, known as active transport, is always mediated by transport proteins and is also coupled to a source of metabolic energy, such as provided by ATP hydrolysis or an ion gradient (Alberts et al., 1994).

Carbohydrates

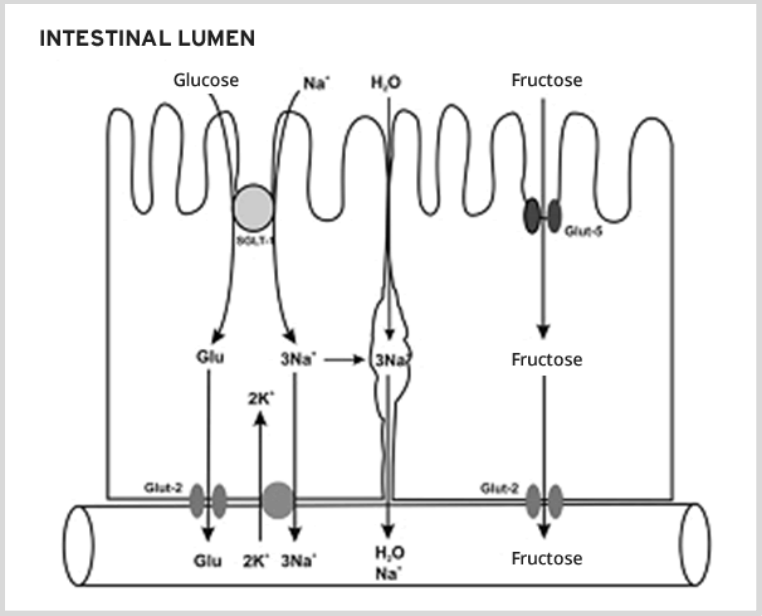
Following we will describe the transport of carbohydrates (CHO) in the intestine.

- **SGLT1 Transporter**

It is a cotransporter of monosaccharides and sodium (Na+). For each molecule of glucose transported, two sodium ions (Na +) are introduced in the epithelial cells. Glucose is pumped into the intestinal epithelial cells through the apical domain of the membrane by a glucose

unidirectional cotransporter driven by Na+; then it is sent into the extracellular fluid by facilitated diffusion through a glucose transport protein existing in the basal and lateral domains (Figure 2) (Alberts et al., 1994). The Na+ gradient that drives the coupled unidirectional glucose transport (symport) is maintained by the ATPase activity of the plasma membrane, which maintains the low intracellular Na+ concentration.

Figure 2: Glucose and Na+ transport through the intestinal epithelial cells



Source: Bermudez et al., 2007, p. 78.

Note the skewed distribution of the transport proteins in the plasma membrane of the intestinal epithelial cell, giving rise to transcellular transport of glucose through the intestinal epithelium. Note also that the adjacent cells that form the intestinal epithelium are connected through impermeable connections, called narrow or tight junctions.

- **GLUT5 transporter**

The independent and facilitated diffusion transporter of sodium (GLUT5) allows transport of fructose from the intestinal lumen to the epithelial cells (Bermudez et al., 2007).

In the basal face of epithelial cells, as can be seen in Figure 2, there is a third transporter for monosaccharides: GLUT2, which transports glucose, fructose and galactose (Bermudez et al., 2007).

Fats

The absorption of long chain fatty acids (> 11 carbons) through the membranes of the intestinal epithelium, occurs by simple diffusion, because they are highly soluble in lipid membranes (Jeukendrup & Gleeson, 2010). In epithelial cells, the fatty acids are reesterified to triacylglycerols in the endoplasmic reticulum, and then combine with cholesterol and phospholipids to form molecules called chylomicrons. In these, the nonpolar side (with affinity for lipids and not for water) of phospholipids is oriented toward the center and the polar side (with affinity for water and not lipids), towards the surface. This enables their transport through the lymph and blood plasma.

The medium chain fatty acids (8 to 10 carbons) and short (<7 carbons) are more soluble in water than the long chain, and therefore its absorption route is different. Such fatty acids enter the epithelial cells and, without being re-esterified, they diffuse directly through the basal membrane toward the portal vein, where they bind to the albumin plasma protein.

Probably, this particularity in its absorption, coupled with its transport across the outer and inner membranes of mitochondria without requiring a union with the L-carnitine, has stimulated the development of scientific investigation to study its potential application as an ergogenic during endurance training.

Amino acids

Amino acids, dipeptides and tripeptides, are absorbed by active transport, coupled to sodium transport in the small intestine (Jeukendrup & Gleeson, 2010). Then they are transported to the liver through the hepatic portal vein. Given that most amino acids are transported to the intestinal epithelium against a concentration gradient, a carrier protein is required for uptake. The luminal membrane transport systems are sodium dependent, while the contraluminal membrane are not (Jeukendrup & Gleeson, 2010).

Vitamins

Most of the vitamin absorption occurs in the jejunum and ileum and is generally a passive process (by simple diffusion, without energy expenditure) (Jeukendrup & Gleeson, 2010). Fat-soluble vitamins (A, D, E, and K) are absorbed with fatty acids and then incorporated with chylomicrons and transported to the liver and other tissues through the lymphatic system (Jeukendrup & Gleeson, 2010).

Water-soluble vitamins are absorbed mainly in the small intestine by simple diffusion. For instance, most of the vitamin C is absorbed in the distal small intestine; thiamine and vitamin B6 are mainly absorbed in the jejunum; biotin and riboflavin are absorbed primarily in the proximal small intestine; Niacin is partially absorbed in the stomach and mainly in the small intestine; while vitamin B12 is mainly absorbed in the ileum, and its absorption being more complex as it involves binding to a specific protein called intrinsic factor (Jeukendrup & Gleeson, 2010).

Minerals

Importantly minerals are not well absorbed in the human intestine, which is why their intake far exceeds the actual requirements (Jeukendrup & Gleeson, 2010). In the case of iron, approximately 15% of the heme iron, which is found within the hemoglobin protoporphyrin ring is absorbed in the small intestine, whereas non-heme iron, which is free iron in their oxidation states Fe^{2+} and $Fe^{3+,}$ is absorbed only from 2% to 10% (Jeukendrup & Gleeson, 2004).

In case of calcium, only 35% is absorbed, magnesium, between 20% and 30%, zinc between 14% and 41%, and chromium, less than 2%. (Jeukendrup & Gleeson, 2010).

Sodium, as was previously mentioned, is absorbed by facilitated diffusion along with glucose, and it is the concentration difference between the intestinal lumen and intestinal epithelial cells which determines its absorption.

Water

Most water absorption occurs in the small intestine, mainly in the duodenum (72%), by simple diffusion. Water moves from the intestinal lumen to the epithelial cells by an osmotic gradient. The osmolarity of most body fluids is 290 mOsm/L. Thus, when the osmolarity of the intestinal lumen is low (<280 mOsm/L), water enters intestinal epithelium cells, whereas when the chyme osmolarity is high (> 300 mOsm/L), water runs from epithelial cells into the lumen. With the absorption of solutes such as glucose and sodium, the osmotic gradient changes and draws water into the intestinal epithelium.

Apparently, for each glucose molecule absorbed, 260 water molecules are absorbed concomitantly (Lambert, Lanspa, Welch & Shi, 2008), or put another way, for every gram of glucose, 26 g of water are absorbed.

Several studies suggest that the maximum rate of fluid absorption in the intestine, at rest, is approximately 1.3 L/h (Noakes, 2001). During exercise, rates of fluid absorption have been reported in the first 50 cm

of the small intestine of 20 ml/cm/hour, which would mean absorptions higher than 1 L/h, assuming that the length of the small intestine is over 50 cm (Noakes, 2001). In reality, this is unlikely, since the tolerance to fluid intake is different for each athlete.

A relatively recent discovery is the existence of a specific transporter called aquaporin 1 (AQP1) or water channel protein (Noakes, 2001). However, apparently, there are humans that do not have this transporter. It is possible that people without AQP1, or with low amounts, present a reduced capacity to absorb fluids during exercise, and thus are more likely to suffer from nausea when they ingest large amounts of fluid (Noakes, 2001), although this hypothesis should be tested in future studies.

The Km of Receptors

The value of **Km**, or **Michaelis-Menten constant,** indicates the substrate concentration necessary to reach half the maximum speed catalyzed by an enzyme or, in the case of a transporter, the maximum rate of solute transport through a membrane. The Km value indicates the affinity between an enzyme or its transporter for its substrate. High values indicate low affinity and low values, high affinity. Thus:

- $V = (Vmax.[S])/(Km + [S])$.

Where V: velocity of the reaction or transport of a solute through a membrane; Vmax.: maximum speed; Km: Michaelis-Menten constant, and [S]: substrate concentration. Therefore, if you have the following:

- $V = 0.5/Vmax.$

This implies what is detailed below:

- $[S] = Km.$

Therefore, when the substrate concentration is equal to Km, the reaction reaches a maximum average speed.

Thus, the Km of the SGLT1 glucose transporter is 28 mM (Lambert et al., 2008). If one considers that the glucose concentration of a sports drink can reach 300 mM, it is clear that the transporter will be practically saturated. This justifies the inclusion of other carbohydrates such as fructose, which are captured by a different transporter (GLUT 5).

Classification of supplements

As you no doubt have noticed, there are a huge number of supplements available on the market in different doses and presentations. The money that turns over annually in this industry is definitely impressive. For instance, sales of supplements with ginseng in the United States reached sales of $10 million in 1997 (Bahrke, 2000), which represents more than 2% of the supplements market.
The remarkable thing is that there seems to be no compelling evidence that ginseng has an ergogenic effect (Bahrke, 2000). This is just one example, and one could continue with L-carnitine, chromium, coenzyme Q, hormone releasing factors, tribulus terrestris and dehydroepiandrosterone, among others.

Note that some supplements available on the market, such as L-carnitine, can be beneficial or ergogenic, but with an application different from that generally recommended in the market. It is very interesting also to note that there is data from studies that show a dissociation between the effect sought by athletes who ingest a given supplement, and the effect that has been tested and documented for it (Petróczi, Naughton, Mazanov, Holloway, & Bingham, 2009).

Thus, both the professional who does not have specific expertise in nutrition, and particularly in sports supplementation, and especially the athlete, are exposed to a very large number of products available on the market, information from scientific journals, textbooks and other sources, that can ultimately generate confusion.

Importantly, the majority of the products available on the market are heavily advertised by their manufacturers, usually with images of successful athletes, but without sufficient scientific evidence to support its use. It is also worth noting that companies often hire professionals to collaborate with independent research groups that conduct studies that are also supported by companies.

This calls on us to reflect and be very careful when interpreting the results of the studies; that is to say, the fact that these are scientific does not imply that they are completely reliable, as they are also subject to biases, competing interests and errors.

1.1.2 Classification from the Australian Institute of Sport Nutrition

Given the scenario posed above, the classifications of supplements proposed by the **Australian Institute of Sport (AIS)** (Table 1) is very useful to distinguish those few supplements that do have evidence of effectiveness and that, properly used, can significantly benefit the performance and health of athletes at all levels, from those that do not.

Table 1: Classification of nutritional supplements by Australian Institute of Sport

GROUP A

Characteristics	Subcategories	Substances
Level of Evidence: Its use is supported in specific sports situations Use in Supplementation programs: Provided for or permitted for use by some athletes in accordance with best practice protocols.	**Sports Foods:** Supplements intended to provide a practical source of nutrients when consuming traditional foods becomes impracticable	**Sport Drinks**
		Sports Gels
		Sports Candy
		Liquid Food
		Whey Protein
		Sports Bars
		Electrolyte Replacements
	Medical Supplements: Used in particular clinical situations such as deficits in certain nutrients. It is by individualized dose and under appropriate medical supervision.	**Iron (Fe)**
		Calcium (Ca)
		Multivitamins
		Vitamin D
		Probiotics
	Performance Supplements: Used to directly increase the performance of the athlete. Its provision should be individualized and overseen by qualified medical personnel. Despite having general evidence about these products, additional research may be needed about its specific use.	**Caffeine**
		B-alanine
		Baking Soda
		Beet Juice
		Creatine

GROUP B

Categories	Subcategories	Substances
Level of Evidence: Need for further research, they can be considered for use by athletes under research protocols or monitoring. **Use in Supplementation Programs:** Supplied to athletes in research situations or clinical monitoring.	**Food Polyphenols:** Food chemicals containing a particular bioactivity, such as antioxidants and anti-inflammatories. They can be ingested as food or as isolated elements.	**Quercetin**
		Tart Cherry Juice
		Exotic Berries (acai, goji, etc.)
		Curcumin
	Others	**Vitamins C and E**
		Carnitine
		HMB
		Glutamine
		Fish Oils
		Glucosamine

GROUP C

Categories	Subcategories	Substances
Level of Evidence: Little significant evidence of beneficial effects. **Use in Supplementation Programs:** They are not provided to athletes in supplementation protocols. They may be approved for use in athletes only through an official panel of supplementation.	Products from category A or B supplied outside the protocols approved for use.	**Equivalent to the lists of examples for groups A and B.**
	If a product does not qualify for membership in groups A, B or D, it is probably in this category.	**Some supplements of interest belonging to group C can be found on the page "Supplements A-Z" in the AIS sports nutrition section on the website of the ASC.**

GROUP D

Category	Subcategory	Substances
Level of Evidence: Prohibited or at high risk of contamination with substances that may lead to positive results in anti-doping tests **Uses in supplementation protocols:** They should not be used by athletes.	**Stimulants** http://list.wada-ama.org/	**Ephedrine**
		Strychnine
		Sibutramine
		Methylhexaneamine (DMAA)
		Herbal stimulants
	Prohormones and hormone stimulants http://list.wada-ama.org/	**DHEA**
		Androstenedione
		19-norandrostenedione
		Other Prohormones
		Tribulus Terrestris and other stimulants of testosterone.
		Maca root powder
	GH releasing peptides. http://list.wada-ama.org/ Technically, despite being sold as supplements, they tend to be unapproved pharmaceuticals.	
	Others http://list.wada-ama.org/	**Glycerol for hyperhydration strategies prohibited as a plasma expander.**
		Bovine Colostrum (not recommended by WADA due to the inclusion of growth factors in its composition).

1.1.3. Classification from the International Society of Sport Nutrition

The other classification we will study is that of the **International Society of Sports Nutrition (ISSN).** This classification constitutes a key tool for professionals, coaches, athletes and active persons to obtain information that is clear, evidence-based, unbiased and uncontaminated by interests of any kind, about the effectiveness of different supplements that are available or not on the market. This could be the first step before making decisions on whether a given supplement may be effective for a specific subject, with a given condition and a particular objective.

Thus we present below (Table 2), the latest classification of the ISSN (Kreider et al., 2010) on the effectiveness of nutritional supplements currently available on the market or being studied in scientific works.

Table 2: Classification of Nutritional Supplements of the International Society of Sports Nutrition

Category	Supplements for the development of muscle mass	Supplements for weight loss	Supplements for improving performance
Apparently Effective and Generally Safe	Weight gain powders, creatine, proteins, essential amino acids (EAA).	Low-calorie meals, powders for meal replacement and ready-to-drink supplements. Ephedra, caffeine and thermogenic supplements containing salicin, ingested in recommended doses in appropriate populations (ephedrine is now banned by the FDA).	Water and sports drinks, carbohydrates, creatine, sodium phosphate, sodium bicarbonate, caffeine, β-alanine.
Possibly Effective	HMB, in untrained subjects initiating training. Branched chain amino acids (BCAA).	Diets high in fiber, calcium, green tea extract, conjugated linoleic acid.	CHO/PRO post-exercise, EAA, BCAA, HMB, Glycerol.
Too Early to Conclude	Alpha-ketoglutarate, alpha ketoisocaproate (KIC), ecdysterones, growth hormone releasing peptides and secretors. Ornithine α-ketoglutarate, zinc aspartate / magnesium (ZMA).	Gymnema sylvestre, chitosan, phosphatidyl choline, betaine, coleus forskolin, DHEA, psychotropic herbs or nutrients.	Medium chain triacylglycerols.
Apparently Ineffective or Dangerous	Glutamine, Smilax, Isoflavones, sulfo polysaccharides (myostatin inhibitors), boron, chromium, conjugated linoleic acid (CLA), gamma oryzanol prohormone	Calcium pyruvate Chitosan, chromium (nondiabetic), garcinia cambogia (HCA), L-carnitine, phosphate, diuretic herbs.	Glutamine Ribose Inosine

	(ferulic acid), tribulus terrestris, vanadyl sulfate (vanadium).		

Source: Kreider et al., 2010, p. 16.

1.1.4 Contamination and Inadvertent Doping

Some ergogenic aids, such as creatine, are obtained from natural sources. In the particular case of this supplement, it is obtained from the muscles of animals (Mesa, Ruiz González-Gross, Sáinz & Castillo Garzón, 2002). Sarcocine and cyanamide are used during the industrial production of creatine. Thus, varying amounts of contaminants are generated such as dicyandiamide, dihydrotriazines, ions, etc., and that means that their tolerable concentrations must be defined and made available to the consumer.

In recent years, a greater number of dietary supplements have been identified with undeclared doping substances (Geyer, Braun, Burke, Stear & Castell, 2011). Importantly, the use of these supplements can lead to accidental positive doping cases, what is known as inadvertent doping.

The leading reasons for a case of doping are products containing ephedrine and analogues, such as sibutramine and methylhexanamine (Fajans Floyd, Knopf & Conn, 1967). In the case of supplements containing ephedrine, natural sources of ephedrine such as Ma Huang or Sinica ephedra are frequently mentioned on the label instead of the names of the active ingredients (ephedrine, pseudoephedrine, methylephedrine, etc.). Regarding supplements enriched with sibutramine, the ingredient is not stated on the label and it only says that the product contains "pure herbal ingredients" that supposedly have the ability to induce weight loss. Sibutramine is an anorectic synthetic drug, approved for pharmaceutical preparations and only available with a prescription. Because of its enormous side effects (risk of stroke and heart attack in patients with a history of cardiovascular disease), in January 2010 the European Medicines Agency recommended for the drug to be removed from the market.

Since 2008-2009, there has been a high risk of inadvertent doping with methylhexanamine, a stimulant which has been included in the prohibited list of WADA (World Anti-Doping Agency, WADA) since 2009 substances. It has been documented (Geyer et al., 2011) that approximately 15% of non-hormonal supplements, such as vitamins, minerals, protein and creatine containing anabolic androgenic steroids (mainly prohormones) not declared on the label. The reason for the contamination may be that prohormone manufacturers (which were legally marketed as dietary supplements in the United States until 2004) also made other supplements in the production line without sufficient cleaning.

Since 2002, dietary supplements have appeared on the market to which have probably been intentionally added high amounts (more than 1

mg/g) of anabolic steroids, undeclared or declared under non-approved label names. These supplements are advertised as leading to huge increases in strength and lean body mass (Geyer et al., 2011). Therefore, the risk of inadvertent doping is predominantly related to dietary supplements that are aggressively advertised by their physiological effects, for example, muscle mass gain or loss of fat, which cannot be attributed only to such products. Thus, athletes should buy supplements only from low-risk sources, although these sources still cannot guarantee that they are free of contaminants, but they do offer a minimization of risk. Such sources are established in countries like Germany (http://www.colognelist.com), Netherlands (http://antidoping.nl/nzvt), United Kingdom (http://www.hfl.co.uk) and United States (http://www.nsf.org/certified/dietary). Nutritional supplements produced by pharmaceutical companies may also represent another alternative, since these products have not been found to be contaminated with doping substances.