Chapter 8

Oxidative Stress and Antioxidant Defenses Induced by Physical Exercise

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Additional information is available at the end of the chapter

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Abstract

This chapter intends to present the physiological and biochemical mechanisms by which exercise induces the appearance of oxidative stress, as well as the characteristics of the physical exercise that involve the appearance of oxidative stress in the human organism.

On the other hand, in this chapter are also presented antioxidant defenses that are magnified with physical exercise and the mechanisms of action, as well as other types of antioxidant defenses that can be incorporated to the body to increase the total antioxidant capacity.

The understanding of possible mechanisms associated with physiological responses that explain how exercise increases oxygen toxicity, and the design of appropriate measures to minimize this toxicity, are critical to:

1) Increase the effectiveness of the exercise as a preventive and therapeutic tool in clinical practice.

2) Control the exercise-induced tissue damage.

Oxidative stress is a situation in which the cells are exposed to pro-oxidant agent and antioxidant defense mechanisms are exceeded, affecting the cellular redox state. This fact occurs during and/or at a high intensity exercise phase.

The knowledge of how the antioxidants interact provides rational bases to develop nutritional strategies to confront the progress in exercise activities and the health maintaining in amateur and professional subjects.

Keywords: oxidative stress, physical exercise, antioxidant defense, nutritional strategies

1. Introduction

The beneficial effects of regular non-exhaustive physical exercise have been known for a long time. Exercise is part of the treatment of common diseases such as diabetes mellitus or coronary heart disease. It improves plasma lipid profile, increases bone density, and helps to lose weight. However, the benecial effects of exercise are lost with exhaustive exercise and with a lack of training. Some of this damage is due to the production of free radicals. Exhaustive exercise causes muscle damage and inflammation, producing free radicals (Fig 1). Phagocytes, such as neuthrophyls, located in the inflamed area may all contribute to free radical production.

As a consecuence, oxidative stress occurs. Oxidative stress is a disturbance in the prooxidant-antioxidant balance in favor of the former, giving an overproduction of free radicals, causing insuffucient antioxidant defenses.

Exercise increases energy demands, increasing the oxygen consumption 10–20 times (200 times in active muscles). It is well known that one of the forms of radical production during exercise is due to a leak in the mitochondrial electron transport chain (ETC). Molecular oxygen typically reacts with species generated by single electron transfer. In such reactions, molecular oxygen is reduced by one electron to form a superoxide radical (O_2^-).

Free radicals are chemical compounds produced in cells in a wide range of processes by reactive species oxygen (ROS) (superoxide, hidroxile, alcoxile, peroxile, hydrogen peroxide) and nitrogen (nitric oxide, nitrogen dioxide, peroxinitrile). Free radicals are widely thought to be of cardinal importance in effecting both the damage and the adaptation that accompany significant physical activity or exercise. They are reactive prooxidant agents to carbohydrates, proteins, and lipids

Submaximal long-duration exercise training may augment the physiological antioxidant defenses in several tissues. Antioxidant defenses can be enzimatic (SOD, GPx, and catalase) and non-enzimatic (glutation, vitamin C and E, carotenoids, lipoic acid, transferrine, and polyphenols).

The effect of antioxidant suplementation (vitamin C, vitamin E, carotenoids, polyphenols, etc.) in the oxidative stress during the exercise has been demonstrated. Within antioxidants, flavonoids are the most important polyphenolic compounds found in rich abundance in all land plants. Flavonoids often exhibit strong antioxidant properties due to its ability to trap free radicals. Furthermore, it is currently unclear whether regular vigorous exercise increases the need for dietary intake of antioxidants. Clearly, additional research that analyzes the antioxidant requirements of individual athletes is needed. This controversy may be due to different reasons: age and sex, training methods, differences in biomaker analysis methods, etc.

Chronic exercise also leads to the upregulation of the body's antioxidant defence mechanism, which helps minimize the oxidative stress that may occur after an acute bout of exercise. Recent studies show a beneficial role of the reactive species, produced during a bout of exercise, that lead to important training adaptations: angiogenesis, mitochondria biogenesis, and muscle hypertrophy. The adaptations occur depending on the mechanic, and consequently biochem-

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ical, stimulus within the muscle. This is a new area of study that promises important findings in the sphere of molecular and cellular mechanisms involved in the relationship between oxidative stress and exercise.

Healthy exercise is being done on a regular basis (several days a week) at a moderate intensity so that the human body in its capacity for homeostatic adaptation (with this type of exercise) increases the physiological antioxidant defenses (enzyme systems such as glutathione peroxidase, catalase, superoxide dismutase), and will offset the appearance of oxidizing species upon exercise (radical and non-radical) with this increased enzyme activity; however, the exercise leads to an increase in the oxidative body state. When more oxidizing species are generated, the body can counteract the so-called oxidative stress, which appears to be unhealthy. If the body does not sufficiently increase the physiological antioxidant defense, it is necessary to provide these through dietary antioxidants such as those included in fruits and vegetables.

Brites et al. (1999) observed an increase in plasma levels of low molecular weight antioxidants (ascorbic acid, uric acid, and α-tocopherol) in a group of trained players to sedentary controls. This increase can be attributed to a mobilization of these antioxidants from tissues into plasma, which would justify the improvement of the total plasma antioxidant status with training.

It would actually be very convenient for researchers, clinicians, coaches, etc. if optimal levels for relevant parameters would be decided and are available, but to date, and for a number of reasons, there are no clear data, such as definite reference intervals. The best practice seems to be thefrequent monitoring, comparison of the individualized values, and relative assessment of the training settings.

The optimal response to training would be the achievement of the relevant adaptations, such as the enhancement of the antioxidant capacity and the subsequent health improvement. Well-designed exercise training is regarded as a preferred way to attain these benefits.

2. Free radicals and exercise

Free radicals during exercise can be formed by different sources [1]:

1. It may be due to an electron leak in the mitochondrial ETC on the ubiquinone-citocrome b, giving superoxide radical anion ($O_2^{\bullet -}$). In this reaction, hydrogen ions and electrons are transported by the electron transporter to the oxygen giving water as final product.

$$4H^+ + 4e^- + O_2 \rightarrow 2H_2O$$

The ETC releases energy and resynthesized ATP in associated reactions. The transport of one electron pair produces enough energy to resynthesize an average of three ATP moles. A total of 12 electron pairs are produced by 180 g of glycogen degradation, producing 36 ATP moles.

Therefore, during aerobic metabolism, the major part of the 36 ATP moles are resynthesized in the ETC.

Due to the increasing O_2 consumed during the exercise (10–20 times) and considering the flow of O_2 in the muscle is 10 times higher [2], the production of superoxide radical anions ($O_2^{\bullet -}$) is equally increased. The superoxide ions' consequent reactions produce other ROS, hydrogen peroxide (H_2O_2), and hidroxile radical (OH^-) [1].

The muscle contraction is connected with the generation of ROS [3] and the consequent muscle fatigue [4].

The prolonged reduction of the strength muscle is probable due to the muscle damage produced by the superoxide radical after the exercise. After exercising, neuthrophils produce ROS, causing the inflammatory response [5]. Neutrophils are the predominant phagocytes of circulating blood, and they are the first cells to arrive at sites of infection. ROS produced during the exercise favor the neutrophils' muscle infiltration, promoting the increase of vascular permeability.

The interaction with the vascular endothelium is produced through membrane receptors: adhesion molecule interleukocyte and leukocyte endothelial adhesion molecule. It has been demonstrated that body temperature increase the leukocytes adhesion to the endothelial cells causing cellular damage. Moderate exercise increases cellular respiration and high intensity exercise tends to suppress cellular respiration [6].

It has been recently demonstrated that the neuthrophils activation factor is induced by the gram-negative lipopolysaccharide. The activation of the leukocytes have toxic effects such as proteinases release, ROS, and ecosanoids.

The stimulation of xantine-oxidase located in endothelial cells during the ischemia reperfusion also produces superoxide radicals. In this reaction, oxygen penetrates the cells producing urate and superoxide radicals with high toxicity [7].

Superoxide radicals stimulate the neuthrophils activation, increasing leukocyte activity in different organs, causing damaged tissues.

Another endogenic form to obtain superoxide radicals is the peroxidation of araquidonic acid, which activates lipoxigenase and ciclooxigenase rutes [8]. It is important to consider that ROS also have beneficial biological effects [9].

The production of septic shock produces nitric oxide derived from L-arginine. This compound is generate in nervous cells and hepatocytes stimulated by citoquines and leukocytes giving a vasodilating effect.

Nitric oxide has several functions: inmunosupression, neurotoxicity, and alteration of the sensorial transmission. Human studies reveal high levels of this compound during sepsis [10].

2. Ischemia-reperfusion. Periods of intensive exercise can cause temporary ischemia or hypoxia in certain regions of the body (kidney, splanic region). Hipoxia is higher as the intensity of the activity increases. After the intensive exercise, the damaged regions are reoxigenated, and then ischemia-reperfusion producing free radicals occurs [11].

Ischemia-reperfusion can occur with intensive exercise, such as rowing, using more oxygen when the resistance of the shoulder, arms, back, and legs is tested [12].

3. Catecholamines autoxidation. The level of catecholamines increases when exercise intensity increases.

4. Xanthine oxidase. Free radical production during exhaustive exercise may also be caused by the enzyme xanthine oxidase [7]. Periods of intensive exercise can cause temporary ischemia or hypoxia, causing ATP to be converted to ADP, AMP, inosine, and finally hypoxanthine. Under such ischemic conditions, intracellular xanthine dehydrogenase (XD) can be converted to xanthine oxidase (XO) by cysteine residue.

Under normal physiological conditions, XD is the dominant form of the enzyme and oxidizes both hypo-xanthine and xanthine (to uric acid) in a process that, concomitantly, reduces NAD^+ to NADH. Xanthine oxidase, on the other hand, can no longer utilize NAD^+ as the electron acceptor, and instead, preferentially reduces oxygen directly to superoxide and hydrogen peroxide. During ischemia, oxygen concentrations are low and intracellular concentrations of XO and hypoxanthine can rise. When oxygen is finally reintroduced (reperfusion) a burst of O_2^- and H_2O_2 can occur. It should be noted that XO generates both O_2^- and H_2O_2 directly.

5. Lipid peroxidation of arachidonic acid produces superoxide radicals [8].

6. The alteration of calcium homeostasis produced when the muscle is stressed during exhaustive exercise, particulary anaerobic/excentric exercise leads to ischemia (hipoxantine formation), creating an excess of contracted activity and muscle damage (with proteases activation) [13].

7. As a result of the respiratory reaction due to the activation of leukocytes after muscle damaged induced during exercise.

3. Oxidative stress induced by exercise

3.1. Oxidative stress induced by extenuant exercise

The increase in energy consumed during exercise increases the oxygen demands of the active tissues, increasing up to 20 times in comparison with basal state [14]. The oxygen flow in the peripheral skeletal muscle tissue can increase up to 200 times, increasing 30 times the blood flow, and the oxygen difference in the arteriovenous blow increases 3 times. As a result, the oxidative metabolism is increased, maximizing the energy produced by unit of substrate and avoiding lactate accumulation [15].

Dillard et al. (1978) [16] first described that extenuant exercise induced lipid damage in tissues. After that, many other investigations focused on the effects of exercise and training in oxygen toxicity and the body defense response. It is accepted that oxygen toxicity can be implicated in some pathologic situations.

The understanding of the mechanisms associated with physiological responses that explain how exercise increases the oxygen toxicity and the design of appropiate measures to minimize toxicity are indispensable to:

1. Increase exercise efficacy as a preventive and therapeutic instrument in clinical practise
2. Control the damaged tissue induced by exercise

Oxidative stress induced by extenuant exercise is a situation by which cells are exposed to a prooxidant environment and defense mechanisms are not enough, affecting the redox estate of the cells. Due to this, nutritional supplements of antioxidants such as vitamin C, vitamin E, carotenids, and polyphenols in the diet are important [13].

In humans, antioxidant defenses in the skeletal muscle and heart are limited. Basal metabolism in the heart is 100% higher than in the liver. This involves a higher risk of oxidative damage in the heart [17]. In basal state, the oxygen consumed of a kilogram of heart and liver is 94 and 44 mL/min, respectively [18]. In adults, superoxide dismutase (SOD) and catalase (CAT) activities are 40 and 16 times smaller in muscles compared with the liver activity of these enzimes [19].

Davies et al. (1982) [20] showed using spin electronic resonance (SER) that exhaustive exercise increases free radical concentration in the liver and muscles and induce oxidative damage in these tissues. Futhermore, they verify the decrease of antioxidant levels and free radical damage could be implicated in the mitochondrial biosynthesis.

Sakellariou et al. (2014) [3] observed the effect of intense muscular contraction activity during 30 min of exercisse and showed 70% increment of free radicals in the active muscles compared with the muscles in basal state.

The theory explains that muscle damage, particularry after eccentric muscle exercise, is responsible for the inflammatory stress after the exercise.

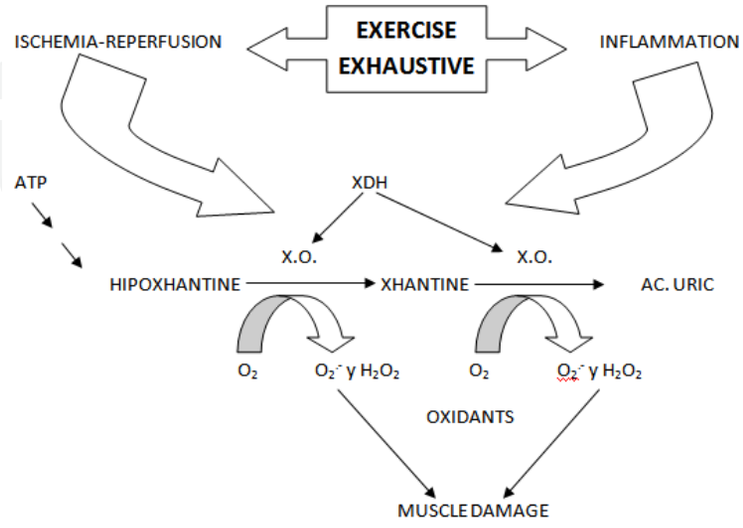

Figure 1. Scheme of the relationship between exhaustive exercise and muscle damage.

After exercise, neuthrophils, monocytes and macrofagues go to the damaged area and provoke the elimination of degraded proteins and cellular remainders. These cells are able to produce ROS and proinflammatory cytokines such as IL-1 TNF-α or IL-8, producing oxidative stress and eventually inflammation. Concentric exercise is associated with an increase in inflammation markers (IL-6) but not in muscle damage parameters (CK). However, excentric muscular exercise shows a typical increase in CK after 72 h. In this case, there is no increase of IL-6 [13].

Barclay et al. (1991) [21] suggest that oxidative stress is implicated in muscle fatigue. There is no evidence of the effect of superoxide radicals in the presence of the free radical hidroxil trapper, blocking the xanthine oxidase activity. Powers et al. (2008) [22] observed a relation between oxidative stress and the mscle strengh.

The factor that triggers the muscle contraction is an electric signal based on the Na^+ entry and followed by a K^+ release of the cell. Animal and human studies have demonstrated a rise in the K^+ plasma concentrations after muscle concentration. As a result of this increase of K^+ that escape to the extracellular medium and water introduction to the cell, intensive muscle contraction decreases by 6–20% of the intracellular K^+ contraction. This can promote the fatigue [23].

Glutathione (GSH) oxidation in different tissues is a valid parameter to appreciate oxidative stress. In this situation, intracellular GSH rapidily oxidizes to GSSG. Intracellular GSSG can be reduced to GSH in the presence of a reductase glutathione and NADPH as cofactor. When the oxidative stress is high, the relation between GSSG/GSH can be higher than the reduction ability of the cells. In this situation, the heart and skeletal muscle cells pour GSSG out of the cells [24].

In extenuant exercise, an increase of GSSG and a decrease of total glutation (GSSG + GSH) in the skeletal muscle tissues such as the liver and heart has been observed [25, 26].

This increasing production of GSSG exceeds the reductase glutathione's ability to reduce disulfide group, thus explaining that the GSSG spill from the tissue to the plasma [27]. The increasing oxidized glutathione plasma concentration as a result of the exercise has been demonstrated in many studies [28, 26, 29].

Gohil et al., (1988) [30] showed that submaximal exercises at 65% VO_{2max} increased the oxidation of blood glutathione during the first 15 min of the exercise. In another study, the level of GSSG in blood increased significantly after 14 min during a maximal test in the cycle ergometer or after pedalling for 30 min in an aerobic threshold or after pedalling 30 min in an anaerobic threshold [26]. In contrast, they did not find significant changes in GSSG in the blood after 60 min and 120 min of the exercise [25]. Sen et al. (1995) [26] demonstrated that 24 hours of recuperation is enough to establish GSSG values in the blood before the exercise.

The glutathione synthesis ability in the liver is high and exercise induces a decrease of glutathione, promoting a protective response of the liver [26].

Studies in hepatectomized (HX) rats reveal that the GSH level in the heart muscle depends on its supply in the liver; however, this fact does not apply to skeletal muscles [31]. These cells are very active in glutathione production. It has been estimated that muscle cells are able to produce 3 mM concentrations of glutathione [27].

The use of gluthatione oxidation as a parameter to detect free radical damage in exercise has demontrated that the damage only appears in exercise exhaustion, meaning that the effect of free radicals only occurs when the subject do exercise above the anaerobic threshold [32].

ROS synthesis induced by neutrophils in exercise has been demostrated by many authors [33, 34].

In mammals, oxidative DNA damage is related to the metabolism rate [35]. After racing for 10 hours, the relationship between oxidase nucleosides/creatine is 1.3 higher with respect to basal state [36]. However, Viguie et al., (1993) [37] did not observe significant changes in 8-hidroxiguanosine after 90 min of racing.

Oxidants as hydroxyle radicals and peroxide radicals can react with proteins. Oxidase proteins rapidily break down into amino acids. Some of these, such as methionine, tryptophan, histidine, and sulfhydryl residue are very sensitive to oxidative damage. The protein oxidation include receptor modification, alteration in translated signals, and other processes (Aoi et al., 2014) [38].

Reznick et al. (1992) [39] observed that exercise increases the protein oxidation of skeletal muscles in rats. Rajguru et al. (1994) [40] showed that after exercise, there is a decrease of sulfhydryl groups in the skeletal muscle. This fact is important in protein crosslinking.

3.2. Exercise as an oxidative stress protector

Up to now, the work has been focused in the damaging effect of exhaustive exercise. However, moderate exercise results in a healthy and beneficial practise that prevents diseases, due to its ability to prevent oxidative stress [41].

Oxidative stress induced by exercise depends on the type, intensity, and the length of the exercise. However, interindividual variability is attributed to the level of training, sex, nutrition, and genetic factors [13].

Undesirable effects of exhaustive exercise can be avoided with progress in training. Salminen et al. (1983) [42] showed that training reduces free radical susceptibility to free radicals. On the other hand, Gómez-Cabrera et al. (2008) [43] observed that training increases antioxidant enzymes. These authors previously showed that training protects against glutathione oxidation associated with exhaustive exercise. Regular exercise creates an adaptation against oxidative stress due to a decrease in DNA damage and maintained levels of protein oxidation [44]. There are many studies that confirm that antioxidant supplements can interfere with the free radical metabolism damaged training adaptations. This fact suggests the recommendation of a diet rich in antioxidant compounds (fresh fruits and vegetables)

Antioxidant defenses in the skeletal muscles, heart, and liver are regulated due to the effect of exercise in the body [45, 46] and showed that exhaustive exercise increased the rate of catalase activity in the liver, muscle, and heart. Since then, a great number of works have confirmed the effect of different resistance training in antioxidant defenses [47–50].

Moderate daily exercise and long duration exercise (resistance training) produce an increase in mitochondrial content in the muscle. However, high intensity exercises have demonstrated

muscle damage derived from the sensibility increment of oxidant agents, the liberation of proteolytic enzymes in the muscle and liver, and loses in the integrity of membranes.

Ginsburg et al. (1996) [52] described a decrease of 47% ($p<0.001$) in lipid peroxidation in the plasma compared with the result obtained before the test. The same work demonstrated that the lipidic peroxidation values were smaller in basal state in trained subjects than in sedentary subjects. These results indicate that accmmulative effects of training tend to decrease lipidic peroxidation in the plasma.

Criswell et al., (1993) [53] studied the effect of training for 12 weeks and observed favorable changes in the skeletal muscles in rats. The authors demosntrated that 5 min of high intensity exercise, was better for antioxidant defense regulation than continuous exercise with moderate intensity.

Daily exercise is important to mantain and promote the ability to defend the organism against the toxicity of reactive oxygen. In prokaryotes, some of the dependent mechanisms of ROS in the induction of defense antioxidant proteins are known [26]. In mammals, cells have been identifying transcription factors responsible for the activation of protein-1 and NF-kB sensitive to redox balance [27]. The redox-tiol state in the different compartments of these cells seems to be implicated in the regulation of these transcription factors. For example, a high cystosolic concentration of GSSG promotes the deactivation of NF-kB, but low cystosolic concentration of GSSG inhibits the fixation of the activate dimmer to the diana oligonucleotids.

Exercise that promotes changes in the redox-tiol state of the tissues can influence the intracellular signal of the translate process, causing the expression of defense antioxidant proteins [43].

Large amount of works support that chronic exercise increases the antioxiant defenses [47–50]. Erythrocyte catalase activity and glutathione reductase show a significant increase after 10 weeks of training [54].

In other studies, the antioxidant state of highly trained runners (128–230 km/week), moderate and low trained runner (26–70 km/week), and sedentary subjects were studied. The results demosntrated a direct relation between the weekly distance and the erythrocyte activity of the antioxidant enzymes. It was found that trained marathon runners have higher levels of MDA and conjugated dienes (CD) in basal state than sedentary subjects. At the end of the half marathon, trained subjects showed a significant increase in the MDA and CD values, however test values decreased in the recuperation period (24–48 hours) to lower values, even lower than when they were determined in basal state.

These results suggest that aerobic training improves the enzymatic antioxidant activity in erythrocytes in basal state and in the recovery period after exercise. This improvement, along with the increase of muscle blood flow and the activity of mitochondrial deshydrogenase-aldehydo activity in the muscle, could be reponsible for the significant decrease of lipidic peroxidation index after exercise in trained subjects [55].

Lipidic peroxidation in blow decrease in response to the increment of training time in 60-year-old women, indicating an adaptation effect [56].

Another study in rats demonstrated after control their training for 5 days that muscle damage induced because of a race could be eliminated. The experiments conclude significant reductions in the pain sensation and proteolysis after training. The authors suggest that training can induce a protective effect against muscle damage when the intensity and the duration of the exercise was moderate [57].

Child et al. [2, 58] studied trained runners subjected to exhaustive exercise. The study suggested a considerable increase of ROS and observed that variations in oxygen consumed can underestimate the real increase in free radical formation during intensive exercise as a consecuence of the reduction of mitochondrial control repiratory and the increase of the formation of free radicals derived from non-mitochondrial sources [59].

Brites et al. (1999) [60] observed an increase in plasmatic levels of low molecular weight antioxidants (ascorbic acid, uric acid, and α-tocopherol) in a footballer trained group with respect to sedentary subjects. This increase can be atributted to a mobilization of the antioxidants from the tissues to the plasma, explaining the improvement of the total plasma antioxidant state with the training [61].

Various authors suggest that physical training promotes parallel adaptation of the mitochondrial antioxidant enzymes and the antioxidant capacity of mitochondrial enzymes. However, Laughlin et al., (1990) [62] studied the relation between the oxidative ability and the antioxidant muscle enzymes and a relation between antioxidant ability and the activity of SOD and catalase was found. Altough training promotes an increase in the muscle's antioxidant ability, there was no effect in the SOD activity, promoting a significant decrease in catalase activity. This coincides with the result found by Ji et al. (1992) [25].

4. Antioxidant defenses

The demonstrated contribution of ROS to muscle damage and muscle fatigue as a consequence of intensive or prolongued exercise induces the defense mechanisms in skeletal muscle cells to reduce the risk of oxidative damage [63, 21]. There are two protective mechanisms: enzymatic and nonenzymatic. They act as a unique antioxidant system to reduce the ROS damage in the cells. Antioxidants (enzymatic and nonenzymatic) exist in extracellular and intracellular space [64]. Antioxidants can be both synthesized in vivo and absorbed through diet.

4.1. Enzymatic antioxidants

The main antioxidant cellular enzymes are superoxide-dismutase (SOD), catalase (CAT), and glutation-peroxidase (GPx). Each of these enzymes is responsible for the reduction of a different ROS, and they are located in different cellular compartments. (1) SOD has three isoforms, two of them are present within cells, whereas the other one is located in the extracellular space. Specifically in skeletal muscle cells, the highest percentage of SOD (65–85%) is found in the cytosol, and the remaining (15–35%) is present in the mitochondria of the muscles. SOD catalyses the reaction of superoxide radicals into oxygen and hydrogen peroxides (H_2O_2).

(2) GPX is located in both the cytosol and the mitochondria of cells. It is responsible for the removal of a wide range of hydroperoxides—from complex organic hydroperoxides to H_2O_2—thus, it may protect membrane lipids, proteins, and nucleic acids from oxidation. GPX is also present in muscle cells, but its activity varies depending on the muscle fiber type, with the greatest activity present in slow twitch muscle fibers (type I) that have higher oxidative capacity. (3) CAT is extensively distributed within the cells and its main function is to degrade H_2O_2 into H_2O and O_2. Nevertheless, it has a lower affinity for H_2O_2 compared with GPX. Similar to the latter, CAT can be found in higher concentrations in type I muscle fibers [22].

The SOD activity shows a significant increase with training, and there is evidence that SOD-Mn is mainly responsible for this increase. The increase in SOD-Mn with training is relatively small compared with the increase in the activity of other mitochondrial enzymes. Furthermore, this rise is not related to a significative improvement in antioxidant protection [65].

Exercise increases SOD activity only in type I muscle fibers, and the SOD activity increase is higher in length than in intensity. An intensive exercise test causes an increase in SOD activity in tissues such as the heart, liver, lungs, and skeletal muscles [25].

GPx activity increases with training only in type II fibers, and this adaptation to training depends on the duration more than the intesity of the exercise. After intensive exercise, GR activity appears to increase in skeletal muscles. GR activity also increase in humans after prolonged exercise [66]. A study on sedentary subjects, marathon athletes, and sprinter trained subjects resulted in a significant increase in GPx compared with sedentary subjects [67].

SOD activity was higher (52%, $p<0.01$) in the trained footballer with respect to sedentary subjects [68].

The effect of training in the catalase activity is controversial. Several studies showed an increase, decrease, and absence of the variation in the catalase activity with chronic exercise. Calderera et al. (1973) [69] observed an important increased CAT activity in the heart, liver, and skeletal muscles after an intensive test exercise.

The activation of the antioxidant enzymatic defenses after intensive exercise can reflect an increase in ROS production. However, due to the differences in oxygen consumption and intrinsic differences in the enzymatic activities, skeletal muscles are subjected to a higher oxidative stress than the liver and heart during exercise [25].

Although evidence has revealed that training controls and regulates antioxidant enzymes in active tissues used in exercise, there is still controversy. In general, antioxidant enzymes of skeletal muscles show the best adaptation response to the training.

In humans, there exists a correlation between the high activity of antioxidant enzymes and the maximum oxygen consumed. Training athletes have a higher SOD and CAT activity in skeletal muscles. Professional and amateur cyclists have higher SOD activity in erythrocytes than sedentary subjects [25]. Due to this, resistance training reduces oxidative damage due to the increase of mitochondrial antioxidant enzymes and a reduction of the oxygen flow in the respiratory chain.

4.2. Nonenzymatic antioxidants

The nonenzymatic antioxidant group includes glutathione, vitamin C, vitamin E, carotenoids, uric acid, polyphenols, and others. Similar to enzymatic antioxidants, these are present in different cellular compartments and elicit distinct antioxidant properties that maximize their effectiveness [70].

GSH exerts various essential functions in the body. Amongst these functions is its major antioxidant role. It efficiently scavenges ROS and free radicals, preventing an increase in the oxidative stress process. In these reactions, the reduced GSH is oxidized, via the enzyme glutathione peroxidase, to form glutathione disulfide (GSSG). Note that GSSG is formed by two GSH molecules linked via a disulfide bond due to the oxidation of the thiol (SH) groups. Once oxidized, GSSG can be reduced back to its original GSH form by the enzyme GSSG reductase and nicotinamide adenine dinucleotide phosphate (NADPH). Nevertheless, when there is a high level of oxidative stress, NADPH becomes depleted and there is an intracellular accumulation of GSSG. This excess GSSG can either be exported out of the cell or it can form a mixed disulfide. Measuring the plasma level of GSH or its oxidized form (GSSG) is a widely accepted method of detecting oxidative stress and can be reported as redox potential, GSH or GSSG concentration, or GSH/GSSG ratio. It is not only a good indicator of systemic oxidative status but also a useful indicator to indicate the free radical production during exercise [71, 72–74].

GSH is the major source of tiol groups in the cells. GSH has several defense antioxidant functions. The practise of 90 min of exercise decreases GSH and increases blood levels of GSSG [30].

Sen et al., (1994) [26] made a maximal test for 14 min. After the test, the subject showed no variation in the blood level of (GSH+GSSG) or GSH, but there was an increase of GSSG and also in the relation GSSG/(GSH+GSSG). When the subjects were subjected to a 30 min test (77% VO_2 max) of anaerobic exercise, the level of GSH was mantained. However, there was an increase of GSSG, the total glutathione, and also the relation GSSG/(GSH+GSSG). After 24 h, all the results recovered to the levels found before the test.

The human levels of GHS are almost undetectable (<0.01 μM), the oxidation of GSH could be in the erythrocytes, in a specific system dependent of the energy to export the excess intracellular GSSG. Due to fact that the test does not reflect the increase of GSH in the blood, the increasing total glutathione could be due to the GSSG exportation of the tissues and the blood GSSG [26].

Plasma levels of GSH is approximately three times lower than blood levels. Moreover, the changes in GSH plasma due to the accelerated flow of liver GSH, produced during exercise, are not detected in the blood in GSH form or total glutathione. The oxidative stress due to intense physical activity produce a rapid oxidation in intracellular GSH in muscle cells and a GSSG production liberated in blood circulation. Thus, a decrease in intracellular glutathione level is observed. This suggests that the GSSG flow of muscle cells to the blood is due to a mechanism dependent of energy [26].

The effect of the training in GSH content seem to vary in different types of muscle fibers and different tissues.The content of GSG in erythrocytes increased at the same time with glutathione reductase after 20 weeks of training in humans who were previously sedentary [25]. In trained subjects, after 2.5 race, a diminution of plasma GSH and the GSH/GSSG. However, the level of GSSG showed a rise at the end of the test compared with the basal state [75].

Short-term training does not improve the adaptation of antioxidant system. A study made on both sex subjects subjected to 8 weeks of aerobic training (3 times/week) showed no variations in the SOD, CAT, and GPx enzymes activity. Even though were changes in the vitamin E concentration in the muscles and in glutathione levels (GSH, GSSG, total glutation, and GSH/GSSG) [76].

However, it has been demonstrated that training protects against glutathione oxidation associated to exhaustive exercise [32].

Similar to vitamin C, vitamin E has important antioxidant properties. Due to its capacity for scavenging ROS and free radicals, particularly peroxyl radical (R O O ·), it exerts the important function of protecting cellular membranes and plasma lipoproteins against lipid peroxidation. This is possible because vitamin E has a great affinity for reducing peroxyl radicals, preventing their interaction with the membrane phospholipids or lipoproteins [77].

Vitamin E has been found to protect cellular membranes from lipid peroxidation. Hence, it is logical to assume that this vitamin could protect muscle cells against exercise-induced damage. Early studies analyzing the effects of vitamin E supplementation and exercise investigated its effect on performance. Most of the studies, however, report no benefit of vitamin E neither for muscle strength nor for endurance performance [78]. Furthermore, it has been hypothesized that vitamin E supplementation could have a protective effect against the contraction-induced muscle damage oxidative stress that may occur after an intense exercise bout. This rationale is based on the knowledge that this vitamin can stabilize muscle membranes by interacting with its phospholipids that would, this way, provide some protection against the increase in oxidative stress or muscle damage observed after certain types of exercises [78]. Altough vitamin E is an effective capture of free radical, the reaction of vitamin E with radicals produces a functional decrease of vitamin E and the formation of free radicals-vitamin E.

The oxidative stress produces a significative decrease of vitamin E levels in tissues. However, the radical vitamin E can be synthetized with the cooperation of other antioxidants. As a result, the investigations conclude that vitamin E's ability to act as an antioxidant is related with the ability of other antioxidants to recycle vitamin E during stress oxidative periods [79].

The exercise could induce an alteration of plasma levels of vitamin E. During human exercise, an increase in vitamin E concentration in plasma and erytrhocytes was observed, suggesting that exercise could promote vitamin E mobilization from tissues to plasma, and the skeletal muscle could use the circulating vitamin E to protect against oxidative damage [25].

Other authors did not find variations in vitamin E levels in humans after a half marathon race [79]. In trained footballer, the levels of vitamin E were higher (10%) than in sedentary subjects [80].

Vitamin E changes are better appreciated when the results are expressed by unit of mitochondrial ubiquinone. The reduction of vitamin E in the inner mithochondrial membrane can justify the susceptibility of the mitochondria to free radicals damage. The content of vitamin E in the heart can decrease. Vitamin E heart content suffered a light decrease after a training program in treadmill, compared with the disminution in skeletal muscles. Different responses to the training of vitamin E and skeletal muscle can be explained in part due to the high vitamin E content in the heart (≈70nmol/g) [25]. In an ultraresistance race (thriatlon), no variation of vitamin E concentration were found before or after the race [81].

Ascorbic acid is the main form of the vitamin found in vivo. This vitamin, also referred to as ascorbate, is found in relatively high levels in different tissues throughout the body. Ascorbate has clearly been shown to play an essential role in connective tissue biosynthesis. During oxidation reactions, only small amounts of ascorbate are lost because, once it is oxidized, it can be reduced back to ascorbic acid by reductants such as glutathione, nicotinamide adenine dinucleotide (NADH), and NADPH. Similarly, vitamin C is also known to regenerate other antioxidants, such as vitamin E and glutathione, back to their reducing state; thus, maintaining a balanced network of antioxidants. The increase of vitamin C levels can protect against oxidative damage of free radicals. However, high concetration of vitamin C (1 mM) acts as a prooxidant in the presence of metals such as Fe^{2+} or Cu^{2+}. Duthie et al. (1990) [79] studied the response of vitamin C after a half marathon race and they observed an increase in the levels of the vitamin. However, Ginsburg et al. (1996) [81] realized a study with triathletes (ultraresistance exercise) and did not observe a variation in the plasma concentration of vitamin C before and after the race.With respect to the trained effect, the vitamin C level of the trained footballer level was higher than in sedentary subjects [81].

As vitamin C, β-carotene can acts as an antioxidant and as a pro-oxidant. A physiological oxygen partial pressure (<100 mm Hg) β-carotene shows a capture activity of radicals. However, the partial pressure >150 mm Hg promotes prooxidant activity of β-carotene [82].

Polyphenolic antioxidants have demonstrated their efficacy against oxidative stress induced by exercise. It has demonstrated the decrease of oxidized proteins in a study subjected to intensive exercise [83, 84].

5. Conclusions

During exercise, an important free radical production is predictable and as a consequence a major requirement of defense mechanisms. Some of the antioxidant defenses can be adequated with training and in the presence of an appropriate diet. Defenses can be insufficient when the exercise exceeds the level by which they were adapted.

The knowledge of how antioxidants interact provides rational bases to develop nutritional strategies to put forward the progress in exercise activities and in mantaining the health of amateur and profesional subjects.

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References

[1] Niess AM, Simon P. Response and adaptation of skeletal muscle to exercise—the role of reactive oxygen species. Front Biosci. 2007 Sep 1;12:4826-38.

[2] Child RB, Wilkinson DM, Fallowfield JL, Donnelly AE. Elevated serum antioxidant capacity and plasma malondialdehyde concentration in response to a simulated half-marathon run. Med Sci Sports Exerc. 1998;30(11):1603-1607.

[3] Sakellariou GK, Jackson MJ, Vasilaki A. Redefining the major contributors to superoxide production in contracting skeletal muscle. The role of NAD(P)H oxidases.Free Radic Res. 2014 Jan;48(1):12-29.

[4] Kuwahara H, Horie T, Ishikawa S, Tsuda C, Kawakami S, Noda Y, Kaneko T, Tahara S, Tachibana T, Okabe M, Melki J, Takano R, Toda T, Morikawa D, Nojiri H, Kurosawa H, Shirasawa T, Shimizu T. Oxidative stress in skeletal muscle causes severe disturbance of exercise activity without muscle atrophy. Free Radic Biol Med. 2010 May 1;48(9):1252-62.

[5] Yamada M, Suzuki K, Kudo S, Totsuka M, Simoyama T, Nakaji S, Sugawara K. Effect of exhaustive exercise on human neutrophils in athletes. Luminescence. 2000 Jan-Feb; 15(1):15-20.

[6] Neiman D. Immune response to heavy exertion. J Appl Physiol. 1997; 82:1385-1394.

[7] Viña J, Gimeno A, Sastre J, Desco C, Asensi M, Pallardó FV, Cuesta A, Ferrero JA, Terada LS, Repine JE. Mechanism of free radical production in exhaustive exercise in humans and rats; role of xanthine oxidase and protection by allopurinol. IUBMB Life. 2000 Jun;49(6):539-44.

[8] Jiang J, Borisenko GG, Osipov A, Martin I, Chen R, Shvedova AA, Sorokin A, Tyurina YY, Potapovich A, Tyurin VA, Graham SH, Kagan VE. Arachidonic acid-induced carbon-centered radicals and phospholipid peroxidation in cyclo-oxygenase-2-transfected PC12 cells.J Neurochem. 2004 Sep;90(5):1036-49.

[9] Smith ChV. Free radical mechanisms of tissue injury. In: Molen HT, Smith ChV, editors. Free radical mechanisms of tissue injury. Boca Raton; 1992. p. 1-22.

[10] Evans T, Carpenter A, Silue A, Cohen J. Inhibitions of nitric oxide syntethase inesperimetal gram negative sepsis. J Infect Dis. 1994;169:343-349.

[11] Sjódin B, Hellsten Westing Y, Apple FS. Biochemical mechanism for oxygen free radical formation during exercise. J Sports Med. 1990;10:236-254.

[12] Kanter MM, Nolte NA, Holloszy JO. Effect of an antioxidant vitamin micture on lipid peroxidation et rest and post-exercise. J Appl Physiol. 1993;74:965-969.

[13] König D, Berg A. Exercise and oxidative stress: Is there a need for additional antioxidants. Öster J Sport Med. 2002;3(1):6-15.

[14] Wilmore JH, Costill DL. (2005) Physiology of Sport and Exercise: 3rd Edition. Champaign, IL: Human Kinetics.

[15] Keul J, Doll E. Oxydative energy supplementation. In: E.Jokl, editor. Energy Metabolism of Human Muscle. Basel:Karger: 1972;214-237.

[16] Dillard CJ, Litov RE, Savin WM, Dumelin EE, Ttappel AL. Effects of exercise, vitamin E and ozone on pulmonary function and lipid peroxidation. J Appl Physiol. 1978; 45:927-932.

[17] Kumar CT, Reddy VK, Prasad M, Thyagaraju K, Reddanna P. Dietary suppplementation of vitamin E protects heart tissue from exercice-induced oxidant stress. Mol Cell Biochem. 1992;111:109-115.

[18] Diem K, Lentner C. Scientific Tables. Documenta Geigy. 7ª ed. Switzerland: Basel; 1970.

[19] Jenkins RR, Friedland R, Howald H. The relation-ship of oxygen uptake to superoxide dismutase and catalase activity in human skeletal muscle. Int J Sports Med. 1984;5:11-14.

[20] Davies KJA, Quintanilha AT, Brooks GA, Packer L. Free radicals and tissue damage produced by exercice. Biochem Biophys Res Commun. 1982;107(4):1198-1205.

[21] Barclay JK, Hansel M. Free radicals may contribute to oxidative skeletal muscle fatigue. Can J Physiol Pharmacol. 1991;69:279-284.

[22] Powers SK, Jackson MJ.Exercise-induced oxidative stress: Cellular mechanisms and impact on muscle force production. Physiol Rev. 2008 Oct;88(4):1243-76.

[23] Sjogaard G. Exercice-induced muscle fatigue: The significance of potassium. Acta Physiol Scand. 1990;140(593S):1-63.

[24] Strobel NA, Matsumoto A, Peake JM, Marsh SA, Peternelj TT, Briskey D, Fassett RG, Coombes JS, Wadley GD.Altering the redox state of skeletal muscle by glutathione depletion increases the exercise-activation of PGC-1α.Physiol Rep. 2014 Dec 23;2(12). pii: e12224.

[25] Ji LL. Exercice-induced oxidative stress in the heart. In: Sen CK, Packer L, Hanninem O, editors. Exercice and Oxygen Toxicity. Amsterdam: Elsevier Science; 1994. p. 249-267.

[26] Sen ChK, Hänninen O. Physiological antioxidants. In: C.K.Sen LPaOH, editor. Exercice and Oxygen Toxicity. Amsterdam: Elsevier Science: 1994c. p. 89-126.

[27] Sen ChK, Marin E, Kretzschmar M, Hanninen O. Skeletal muscle and liver glutathione homeostasis in response to training, exercice and inmovilization. J Appl Physiol. 1992;73:1265-1272.

[28] Viguie CA, Frei B, Shigenaga MK, Ames BN, Packer L, Brooks GA. Antioxidant status and indexes of oxidative stress during consecutive days of exercice. J Appl Physiol. 1993;75:566-572.

[29] Hillman AR, Vince RV, Taylor L, McNaughton L, Mitchell N, Siegler J. Exercise-induced dehydration with and without environmental heat stress results in increased oxidative stress. Appl Physiol Nutr Metab. 2011 Oct;36(5):698-706.

[30] Gohil K, Viguie CA, Stanley WC, Brooks GA, Packer L. Blood glutathione oxidation during human exercice. J Appl Physiol. 1998;64:115-119.

[31] Kretzschmar M, Pfeifer U, Machnik G, Klinger W. Glutathione homeostasis and turnover in the totally hepatectomized rat: Evidence for a high glutathione export capacity of extrahepatic tissues. Exp Toxicol Pathol 1992;44:273-281.

[32] Gómez-Cabrera MC, Gimeno A, Lloret A, Miñana JB, Marquez R, Viña J. Deporte de alta competición y daño oxidativo: Papel de los nutrientes antioxidantes. Antioxidantes y calidad de vida [en línea] [fecha de acceso 21 de mayo de 2001]. URL disponible en: www.antioxidantes.com.ar /12/Art118.htm.

[33] Smith JA, Teldorfd IB, Mason IB, Weidemann MJ. Exercice. Int J Sports Med. 1990;11:179-187.

[34] Nunes-Silva A, Bernardes PT, Rezende BM, Lopes F, Gomes EC, Marques PE, Lima PM, Coimbra CC, Menezes GB, Teixeira MM, Pinho V. Treadmill exercise induces neutrophil recruitment into muscle tissue in a reactive oxygen species-dependent manner. An intravital microscopy study. PLoS One. 2014 May 5;9(5):e96464.

[35] Ames BN, Shigenaga MK, Hagen TM. Oxidants, antioxidants and the degenerative diseases of aging. Proc Natl Acad Sci U S A. 1993;90:7915-7922.

[36] Alessio HM, Cutler RG. Evidence that DNA damage and repair cycle activity increases following a marathon race. Medicine and Science in Sports and Exercise. 1990;22(S126):751-756.

[37] Viguie CA, Frei B, Shigenaga MK, Ames BN, Packer L, Brooks GA. Antioxidant status and indexes of oxidative stress during consecutive days of exercice. J Appl Physiol. 1993;75:566-572.

[38] Aoi W, Naito Y, Yoshikawa T. Potential role of oxidative protein modification in energy metabolism in exercise. Subcell Biochem. 2014;77:175-87.

[39] Reznick AZ, Cross CE, Hu ML, Suzuki YJ, Khwaja S, Safadi A, et al. Modification of plasma proteins by cigarette smoke as measured by protein carbonyl formation. Biochem J. 1992c;286:607-611.

[40] Rajguru SU, Yeargans GS, Seidler NW. Exercice causes oxidative damage to rat skeletal muscle microsomes while increasing cellular sulfhidryls. Life Sci. 1994;54:149-157.

[41] Farney TM, McCarthy CG, Canale RE, Schilling BK, Whitehead PN, Bloomer RJ. Absence of blood oxidative stress in trained men after strenuous exercise. Med Sci Sports Exerc. 2012 Oct;44(10):1855-63.

[42] Salminen A, Vihko V. Lipid peroxidation in exercise myopathy. Exp Mol Pathol. 1983;38:380-388.

[43] Gomez-Cabrera MC1, Domenech E, Viña J. Moderate exercise is an antioxidant: Upregulation of antioxidant genes by training. Free Radic Biol Med. 2008 Jan 15;44(2): 126-31.

[44] Radak Z, Pucsuk J, Boros S. Exercise predonditioning against hydrogen peroxide-induced oxidative damage in proteins of rat myocardium. Arch Biochem Biophys. 2000;376:248-251.

[45] Leeuwenburg C, Fiebig R, Chandwaney R, Ji LL. Aging and exercise training in skeletal muscle: Responses of glutathione and antioxidant enzyme systems. Am J Physiol. 1994;267(R):439-445.

[46] Caldarera CM, Guarnieri C, Lazzari F. Catalase and peroxidase activity of cardiac muscle. Boll Soc Ital Biol Sper. 1973;49:72-77.

[47] Cardoso AM, Bagatini MD, Roth MA, Martins CC, Rezer JF, Mello FF, Lopes LF, Morsch VM, Schetinger MR. Acute effects of resistance exercise and intermittent intense aerobic exercise on blood cell count and oxidative stress in trained middle-aged women.Braz J Med Biol Res. 2012 Dec;45(12):1172-82.

[48] Cunha TF, Bacurau AV, Moreira JB, Paixão NA, Campos JC, Ferreira JC, Leal ML, Negrão CE, Moriscot AS, Wisløff U, Brum PC. Exercise training prevents oxidative stress and ubiquitin-proteasome system overactivity and reverse skeletal muscle atrophy in heart failure. PLoS One. 2012;7(8):e41701.

[49] Hübner-Woźniak E, Morgulec-Adamowicz N, Malara M, Lewandowski P, Okęcka-Szymańska J. Effect of rugby training on blood antioxidant defenses in able-bodied and spinal cord injured players. Spinal Cord. 2012 Mar;50(3):253-6.

[50] Vincent HK, Powers SK, Stewart DJ, Demirel HA, Shanely RA, Naito H. Short-term exercise training improves diaphragm antioxidant capacity and endurance. Eur J Appl Physiol. 2000 Jan;81(1-2):67-74.

[51] Heavens KR, Szivak TK, Hooper DR, Dunn-Lewis C, Comstock BA, Flanagan SD, Looney DP, Kupchak BR, Maresh CM, Volek JS, Kraemer WJ. The effects of high intensity short rest resistance exercise on muscle damage markers in men and women. J Strength Cond Res. 2014 Apr;28(4):1041-9.

[52] Gingsburg GS, Agil A, O'Toole M, Rimm E, Douglas PS. Effects of a single bout of ultraendurance exercise on lipid levels and susceptibility of lipids to peroxidation in Triathletes. J Am Med Assoc. 1996;276(3):221-225.

[53] Criswell D, Powers S, Dodd S, Lawler J, Edwards W, Renshler K, et al. High intensity training-induced changes in skeletal muscle antioxidant enzyme activity. Med Sci Sports Exerc. 1993;25:1135-1140.

[54] Ohno H, Yahata T, Sato Y, Yamamura K, Taniguchi N. Physical training and fasting erythrocyte activities of free radical scavenging enzyme sistems in sedentary men. Eur J Appl Physiol. 1988;57:173-176.

[55] Robertson JD, Maughan RJ, Duthie GG, Morrice PC. Increased blood antioxidant systems of runners in response to training load. Clin Sci. 1991;80:611-618.

[56] Yagi K. Lipid peroxides and exercise. Med Sci Sports Exerc. 1992;30:40-42.

[57] Komulainen J, Vihko V. Training-induced protection and effect of teminated training on exercise-induced damage and water content in mouse skeletal muscles. Int J Sports Med. 1995;16:293-297.

[58] Child RB, Browm S, Day S, Donelly AE, Ropper H, Saxton JM. Changes in indices of antioxidants status, lipid peroxidation and inflamation in human skeletal muscle after eccentric muscle actions. Clin Sci. 1999;96(1):105-115.

[59] Sjódin B, Hellsten Westing Y, Apple FS. Biochemical mechanism for oxygen free radical formation during exercise. J Sports Med. 1990;10:236-254.

[60] Brites FD, Evelson PA, Christiansen MG, Nicol MF, Basílico MJ, Wikinski RW, et al. Soccer Players under regular training Show Oxidative Stress but an Improved Plasma Antioxidant Status. Clin Sci. 1999;96:381-385.

[61] Rosety-Rodríguez M, Díaz-Ordoñez A, Rosety I, Fornieles G, Camacho-Molina A, García N, Rosety MA, Ordoñez FJ. Aerobic training improves antioxidant defense system in women with metabolic syndrome. Medicina (B Aires). 2012;72(1):15-8.

[62] Laughlin MH, Simpson T, Sexton WL, brown OR, Smith JK, Korthuis RJ. Skeletal muscle oxidative capacity, antioxidant enzymes and exercise training. J Appl Physiol. 1990;68:2337-2343.

[63] Spriet LL, Whitfield J. Taurine and skeletal muscle function. Curr Opin Clin Nutr Metab Care. 2015 Jan;18(1):96-101.

[64] Powers SK, Hamilton K. Antioxidants and Exercice. Clin Sports Med. 1999; 18(3): 525-536.

[65] Marsh SA, Laursen PB, Coombes JS. Effects of antioxidant supplementation and exercise training on erythrocyte antioxidant enzymes. Int J Vitam Nutr Res. 2006 Sep; 76(5):324-31.

[66] Marzatico F, Pansarasa O, Bertorelli L, Somenzini L, Della Valle G. Blod free radical antioxidant enzymes and lipid peroxides following long-distance and lactacidemic performances in highly trained aerobic and sprint athletes. J Sports Med Phys Fitness. 1997;37(4):235-239.

[67] Brites FD, Evelson PA, Christiansen MG, Nicol MF, Basílico MJ, Wikinski RW, et al. Soccer Players under regular training Show Oxidative Stress but an Improved Plasma Antioxidant Status. Clin Sci. 1999;96:381-385.

[68] Caldarera CM, Guarnieri C, Lazzari F. Catalase and peroxidase activity of cardiac muscle. Boll Soc Ital Biol Sper. 1973;49:72-77.

[69] Dufaux B, Heine O, Kothe A, Prinz U, Rost R. Blood Glutathione Status Following Distance Running. Int J Sports Med. 1997;18(2):89-93.

[70] Powers SK, DeRuisseau KC, Quindry J, Hamilton KL. Dietary antioxidants and exercise. Journal of Sports Sciences. 2004; 22(1): 81-94.

[71] Gomes EC, Stone V, Florida-James G. Impact of heat and pollution on oxidative stress and CC16 secretion after 8km run. European Journal of Applied Physiology. 2011;111(9):2089-2097.

[72] Sastre J, Asensi M, Gasco E, et al. Exhaustive physical exercise causes oxidation of glutathione status in blood: prevention by antioxidant administration. American Journal of Physiology. 1992;263(5): R992-R995.

[73] Sen CV, Rankinen T, Vaisanen S, Rauramaa R.Oxidative stress after human exercise: Effect of N-acetylcysteine supplementation. Journal of Applied Physiology. 1994;76(6): 2570-2577.

[74] Medved I, Brown MJ, Bjorksten AR, Leppik JA, Sostaric S, McKenna MJ. N-acetylcysteine infusion alters blood redox status but not time to fatigue during intense exercise in humans. Journal of Applied Physiology. 2003;94(4):1572-1582.

[75] Tiidus PM, Pushkarenko J, Houston ME. Lack of antioxidant adaptation to short-term aerobic training in human muscle. Am J Physiol. 1996;271(R):832-836.

[76] Packer L. Protective role of vitamin E in biological systems. Am J Clin Nutr. 1991;53(4 S):1050-1055.

[77] Institute of Medicine, Dietary Intakes for Vitamin C, Vitamin E, Selenium, Carotenoids, National Academy Press, Washigton, DC, USA, 2000.

[78] Urso ML and Clarkson PM. Oxidative stress, exercise, and antioxidant supplementation. Toxicology. 2003;189(1-2):41-54.

[79] Duthie GG, Robertson JD, Maughan RJ, Morrice PC. Blood antioxidant status and erythrocyte lipid peroxidation following distance running. Arch Biochem Biophys. 1990;282(1):78-83.

[80] Brites FD, Evelson PA, Christiansen MG, Nicol MF, Basílico MJ, Wikinski RW, et al. Soccer Players under regular training Show Oxidative Stress but an Improved Plasma Antioxidant Status. Clin Sci. 1998;96:381-385.

[81] Gingsburg GS, Agil A, O'Toole M, Rimm E, Douglas PS. Effects of a single bout of ultraendurance exercise on lipid levels and susceptibility of lipids to peroxidation in Triathletes. J Am Med Assoc. 1996;276(3):221-225.

[82] Palozza P, Luberto C, Calviello G. Antioxidant and prooxidant role of β-carotene in murine normal and tumor thymocytes: Effects of oxygen partial pressure. Free Radic Biol Med. 1997;22:1065-1071.

[83] Morillas-Ruiz JM, Villegas García JA, López FJ, Vidal-Guevara ML, Zafrilla P. Effects of polyphenolic antioxidants on exercise-induced oxidative stress. Clin Nutr. 2006 Jun;25(3):444-53.

[84] Morillas-Ruiz J, Zafrilla P, Almar M, Cuevas MJ, López FJ, Abellán P, Villegas JA, González-Gallego J. The effects of an antioxidant-supplemented beverage on exercise-induced oxidative stress: results from a placebo-controlled double-blind study in cyclists. Eur J Appl Physiol. 2005 Dec;95(5-6):543-9.

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