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EFFECT OF PHYSICAL AND BIOCHEMICAL STRESS ON ANTIOXIDANT ACTIVITY IN THE SPORTS WOMEN

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Abstract:

Physical activity is recognized as an important component of healthy life style and recommended throughout life by scientists and clinicians. Exercise training is recommended for improving physiological and functional capacity in the elderly. Intracellular levels of reactive oxygen species may be crucial for determining whether the cells undergo differentiation, proliferation or apoptosis because it has been shown that at low concentration. H_2O_2 induces necrosis in fibroblasts, while at high concentrations it causes a transient cell growth arrest by inducing the expression of gadd 45,153 and adapts 15 genes. The human body is constantly under attack from free radicals. Free radicals are highly reactive molecules generated by the biochemical redox reaction that occur as part of normal cell metabolism and by exposure to environment factors. The main free radical oxygen species which occur in human body include; super oxide radical ($O_2^{\bullet -}$), hydroxyl radial ($OH^{\bullet}$), nitric oxide radical ($NO^{\bullet}$), peroxyl radical ($ROO^{\bullet}$). Once formed free radicals attack cell structures within the body. Decomposition of peroxidised lipids yields a wide variety of end products including malondialdehyde. The imbalance between process forming oxygen free radicals and endogeneous defence system results in oxidative stress, it may play a major role in numerous diseases artherosclerosis, cancer, diabetes, respiratory diseases, liver damage, AIDS, central nervous system disorders, Parkinson disease, etc.

Keywords: Catalase, glutathione peroxidase, lipid peroxidation, reduced glutathione, superoxide dismutase, vitamin-A, C and E.

Introduction

Oxygen derived free radicals include superoxide, hydroxyl, hydroperoxyl and alkoxyl radicals. Other common reactive oxygen species (ROS) produced in the body includes nitric oxide and peroxy nitrite anion [1]. Several investigations have shown an increased in the production of reactive oxygen species; which occur as consequence of electron transport chain deficiency [2, 3]. There is no uniformity in the results on how the antioxidant defense system is affected in old age. However, exercise like ageing, is one of the physiological conditions characterized by increased production of free radicals [4]. ROS are also produced by exogenous agents like radiation, xenobiotic and various environmental factors [5]. The other endogenous sources for ROS production include the reactions of lipoxygenases, cyclooxygenases, xanthine oxidase and NADPH oxidase [6]. The production of free radicals increase is parallel with increase in oxygen consumption during exercise and increase is directly related to the intensity and the duration of exercise. Recent studies suggest that both, ROS and lipid peroxidation products are also involved in the intracellular signaling mechanisms which determine the cells final fate.

Lipid peroxidation (LH) can be defined as the oxidative deterioration of lipid containing a number of carbon-carbon double bonds [7]. A large number of toxic by products are found during lipid peroxidation. These have effects at a site away from the area of their generation. Hence, they behave as toxic second messenger. Membrane lipids are particularly susceptible to lipid peroxidation. In the general process of lipid peroxidation is the target is PUFA and the initiating, oxidizing radical. The major fatty acids that undergo lipid peroxidation in the cell membranes are linoleic acid (18:2), arachidonic acid (20:4) decosahexaenoic acid (22:6), and other poly unsaturated acids. When the fatty acids undergo peroxidation 2, 6 and 10 different hydroperoxide species result from 18:2, 20:4 and 22:6 respectively [8].

The hydroxyl radical is considered potentially the most potent oxidant encounter in biological systems. Highly reactive hydroxyl radicals readily react with a variety of molecules such as those found in organic lipids by removal or addition of hydrogen to unsaturated bonds [9]. The hydroxyl radicals are derived from the decomposition of hydrogen peroxide via the Fenton reaction; the interaction of superoxide with hydrogen peroxide through the Harber-Weiss reaction [10]. Hydroxyl radicals can also be generated when the reduced

forms of transition metal ions such as copper come into contact with H_2O_2. Superoxide is the best known free radical of all oxygen derived species. It is the first intermediate in the sequential univalent reductions of that leads to the formation of H_2O [11]. Superoxide radical is unique it can lead to the formation of many other reactive species, including hydroxyl free radical, hydrogen peroxide and perhydroxyl radicals [12]. At a neutral or higher pH, the dismutation of O_2 is catalysed by superoxide dismutase [13].

The interest in H_2O_2 as a biologically active oxygen derived intermediate in evident from the enormous amount of data available in the literature [14]. Although H_2O_2 is not by definition considered an oxygen free radical. It remains the most extensively studied oxygen metabolite. H_2O_2 is a secondary product of one election oxidation. The dismutation of O_2 by superoxide dismutase is a major source of H_2O_2. As shown earlier, H_2O_2 has the ability to generate reactive hydroxyl free radicals through its interaction with redox active transitional metals [15, 16]. Radical-mediated damage to cellular biomembranes results in lipid peroxidation, a process that generates a variety of products including reactive electrophiles such as epoxides and aldehydes and are exemplified as MDA a by product of lipid degradation is a tautomer that is both highly electrophilic and nucleophilic. This characteristic allows not only reaction with cellular nucleophiles but also the formation of MDA oligomers [17]. Lipid peroxidation is a major source of cytotoxoid products [18] such as aldehydes. Malondialdehyde can destabilize lipids asymmetry of their membrane bilayer. This can result in cellular dehydration and reduction in whole cell deformability and cell survival.

Antioxidants are substances which react chemically with free radicals and render them, harmless and at the same time break the vicious circle, which involves the decomposition of fatty acid and proteins the creation of new free radicals and eventually cell death. Thus, they act as weapons for combating free radicals. The various antioxidants exert their effect by scavenging superoxide or by activating a battery of detoxifying or defense proteins [19]. Antioxidants are more necessary for women than men. Women may have more reason to reach for their vitamins than men, according to a recent study in the American epidemiology that shows women undergo more oxidation than men. Oxidation is the process that forms free radicals and is thought to play a part in cardiovascular disease, stroke and other conditions [20]. This study indicates women would be wise to consume more antioxidants. The antioxidants in the body can be derived from exogenous or endogenous

biosynthesized sources. Radial scavenging antioxidants have been exposed to have a novel functions by which they regulate gene expression of the cell [21].

Catalase is a major primary antioxidant defense component and also a tetrameric hence containing protein and it catalyses the decomposition of H_2O_2 to H_2O and oxygen. Sharing this function with GPx, both enzymes detoxify oxygen reactive radicals by catalyzing the fermentation of H_2O_2 derived from superoxide. Glutathione peroxidase (GPx) a well known selenoenzyme present in the cytosol and mitochondrial matrix remove the majority of H_2O_2 [22]. It catalyses the reduction of harmful peroxide by glutathione and protect cells against oxidative damage. Most species exhibit GPx, intracellularly located in the cytosol and mitochondrial matrix. GPx catalyzed the reduction of H_2O_2 and organic hyphoperoxides. Superoxide dismutases (SOD) are localized to the cytosol and mitochondria and it functions to reduce superoxide anion to H_2O_2 and molecular oxygen [23].

Vitamin-C is the most effective water soluble antioxidant in human plasma against lipid peroxidation induced by aqueous, peroxyl radicals, activated neutrophils or the gas-phase of cigarette smoke [24]. Where $HO^{\bullet}$ hydroxyl radical, LOOH is lipid hydroperoxide, $LO^{\bullet}$ is alkoxyl radical, AH^{-} is radical and helps to preserve α-tocopherol (vitamin-E) in lipoproteins. Vitamin-C prevents lipid hydroperxoide formation in plasma lipoprotein. Ascorbate is a strong antioxidant capable of scavenging a wide variety of reactive oxygen and nitrogen species. Ascorbate can both consume oxygen free radicals before they can oxidize α-tocopherol and can reduce α-tocopherol in LDL in the face of an oxidant stress [25]. Dehydroascorbate by itself is a peroxide scavenger by virtue of its action as an α-keto acid after its hydrolysis to the carboxylate form. Dehydroascorbate can prevent oxidative damage which could be attributed to the acceleration of the HMP shunt as well as through its direct antioxidant action after its reduction by GSH. Ascorbic acid serves as both an antioxidant and a prooxidant [26].

Reduced glutathione readily interacts with free radicals, notably hydroxyl and carbon radicals. By donating a hydrogen atom such reactions scan provide protection by neutralizing reactive $OH^{\bullet}$, which is considered a major source of free radical damage. The existence of a GSH-dependent factor that inhibits lipid peroxidation in membrane has been suggested [27]. Such inhibition may be related to a catalytic amount of a

vitamin that undergoes a series of reaction steps involving the regenerations of GSH. As with most antioxidant defenses the levels of GH fluctuate under various physiological conditions, including again [28] and some neoplastic disease, which are usually accompanied by increased lipid peroxide [29]. The structural arrangement of β-carotene with their long chain of conjugated double bonds suggests that they would make excellent scavenger for reactive free radicals. Carotenoids have long been considered antioxidants because of their capacity to scavenge free radicals [30]. Carotenoids protect lipids against peroxidation by quenching free radicals and other biactive oxygen; species notably singlet oxygen [31]. β-carotene displays an efficient biological radical trapping antioxidant activity through its inhibition of lipid peroxidation induced by the xanthine oxidase system [32].

The mechanism responsible for the sparing action of the ability of ascorbic acid is to reduce tocophoroxyl radicals formed by the scavenging of other free radicals [33]. Chromanol ring of tocopherols donates its phenolic hydrogen to reduce the free radical and is itself oxidized to the quinone form. It is the most widely distributed antioxidant in nature, being found in both the plant and animal kingdoms. The generic term vitamin-E refers to atleast eight structural isomers of tocopherol. Among these, α-tocopherol is the best known isomer and possesses the most potent antioxidant activity [34]. Because of the lipophilic property of the tocopherol molecule, vitamin-E is the major free radical chain terminator in the liphophilic environment. High levels of tocopherol are found in selected mammalian tissues this preferential distribution may result from its high lipid solubility.

The present study is carried out to assess the change in the level of lipid peroxidation, vitamin-A, E, C, GSH, SOD, catalase and glutathione peroxidase in the sports women.

Materials and Methods

Blood samples were collected from the sports students who are studying in VELTECH MULTITECH Dr.Rangarajan Dr.Sakunthala Engineering College, Avadi, Chennai. Venous blood was collected into tubes containing EDTA as anticoagulant. The plasma was separated and used for further analyzes. In all samples, general parameters studied include, lipid peroxidation, antioxidants in a RBC's, the antioxidant enzymes SOD and glutathione peroxidase were assayed in haemolysate while catalase was assayed in red cell membrane. Antioxidants vitamin-A and E were assayed in plasma and vitamin-C in whole blood. Malondialdehyde was

also assayed in plasma. The particulars like name, age, height, food habits were noted. Non-parametric studies included blood pressure, sugar, hypertension and cholesterol. The subjects were compared with normal subjects.

Separation of Red Blood Cells and Isolation of Erythrocyte Membrane

Erythrocyte membrane was isolated with a change in buffer. Packed cells remaining after the removal of plasma was washed three times with isotonic saline to remove the buffy coat. 4ml of packed cells were then washed 3 times with isotonic tris-HCl buffer, pH 7.4. Hemolysis was performed by precipitating out the washed red blood cell suspension into polypropylene centrifuge tubes which contain hypotonic buffer, pH 7.2. Erythrocyte ghosts were sedimented in a high speed refrigerated centrifuge at 20,000 rpm for 40 minutes. The supernatant was decanted carefully and ghost button was resuspended by swirling. Sufficient buffer of the same strength was added to reconstitute the sedimented membrane to the original volume. The ratio of the cells to washing solution is approximately 4:3 by volume. The ghosts or the membrane were washed three times subsequent to haemolysis. The supernatant after the last wash was either pale pink or colorless. The pellet of the erythrocyte membrane was resuspended in 10ml of tris-HCl buffer, pH 7.4 and aliquots of this reconstituted membrane preparation was taken for various analyses.

Lipid Peroxidation

Lipid peroxidation was assayed as the thiobarbituric acid reactive substance (TBARS) and malondialdehyde was used as a standard. Lipid peroxidation was measured in plasma and erythrocyte membrane. Lipid peroxide levels in plasma were determined using thiobarbituric acid (TBA) reagent reaction. The water soluble substances that react with TBA were eliminated by precipitating lipid peroxides along with plasma proteins in a phosphotungstic acid system. The interference from sialic acid and bilirubin, both of which react with TBA, was avoided by performing TBA reaction in an acetic acid solution. To 0.5ml of plasma, 1ml of phosphotungstic acid was added, mixed well and centrifuged. The supernatant was discarded and the sediment was mixed with 2ml of $^{N}/_{12}$ sulphuric acid and 0.3ml PTA. The mixture was centrifuged and the sediment was dissolved in 4ml distilled water and 1ml of TBA reagent. The contents were heated in a boiling water bath for 60 minutes. After cooling, 5ml of butanol was added and the contents shaken and centrifuged for 20 minutes. The upper butanol layer was read at 515nm. A stock solution of malondialdehyde containing 2–10 nmoles was taken

mixed with TBA reagent and processed in the same manner as above. Lipid peroxidation values are expressed as nmoles of MDA/dl plasma.

Superoxide Dismutase

Superoxide dismutase catalyses the superoxide anion (O_2^-) into hydrogen perxoide and molecular oxygen. Superoxide dismutase in the haemolysate was assayed based on the oxidation of epinephrine–adenochrome transition by the enzyme. 0.5ml haemolysate was diluted with 0.5ml of water. To this, 0.25ml of ethanol and 0.15ml of chloroform were added and centrifuged. The enzyme acclivity in the supernatant was determined. To 0.5ml of the supernatant, 1.5ml of buffer was added. The reaction was initiated by the addition of 0.4ml of epinephrine and change in optical density per minute was determined at 480nm. The change in optical density per minute measured at 50% inhibition of epinephrine to adrenochrome by the enzyme is taken as one enzyme unit. The enzyme activity is expressed as units/mg hemoglobin.

Catalase

This enzyme catalyses the reaction and is assayed in the erythrocyte membrane suspension. 1ml of buffer, 0.4ml of water and 0.1ml of membrane suspension, were taken and brought to 37°C. The reaction was initiated by the addition of 0.5ml hydrogen peroxide and the reaction mixture was incubated at 37°C for 1 minute. Addition of 2ml of dichromate acetic acid reagent terminated the reaction. Standard hydrogen peroxide solution in the range of 4–20 μmoles were taken and treated in the same manner. The tubes were heated in a boiling water bath for 15 minutes, cooled and read at 510nm. Enzyme activity is expressed as μmoles of hydrogen peroxide utilized/mg protein/minute.

Glutathione Peroxidase

Glutathione peroxidase was assayed in the haemolysate. 0.2ml each of EDTA, sodium azide, glutathione and hydrogen peroxide incubation mixture containing 0.4ml of buffer and 0.1ml of lysate was incubated at 37°C for 10 minutes. The reaction was arrested by the addition of 0.5ml of TCA. Protein was sedimented by centrifugation and 0.5 ml of the supernatant was taken. 3ml of phosphate solution and 1ml of DTNB were added and the color developed was read immediately at 412nm. Suitable aliquots of standard solution were taken and

treated in the same manner to obtain a standard curve for comparison. Glutathione peroxidase activity is expressed as μgGPx utilized/mg hemoglobin/minute.

Vitamin-A

Vitamin-A was assayed in plasma. 3ml of light petroleum ether, 1.5ml of ethanol and 1.5ml of plasma was added, and shaken well for about 10 minutes and allowed to stand to obtain a clear supernatant. 2.5ml of the supernatant as well as standard in the range 150-350 μg was used for the assay. 2ml of TCA-chloroform reagent was added and the optical density was read at 620nm within 15 seconds. Values are expressed as μmoles/L plasma.

Vitamin-E

Vitamin-E was assayed in plasma after extraction. After saponification and solvent extraction of lipids to remove the interfering substances, α-tocopherol was estimated spectrophotmetrically using bathopenanthroline reagent. 1ml of plasma, 2ml of 2% solution of pyrogallol in purified ethanol was mixed thoroughly. The mixture was heated at 70°C for 2 minutes. Added 0.3ml of saturated potassium hydroxide and mixed again and incubated at 70°C for 30 minutes. The tubes were shaken and centrifuged for 10 minutes to separate the two phases. The hexane extract was estimated for vitamin-E. 3ml of hexane extract was evaporated to dryness under vacuum. 3ml of absolute ethanol was added to the residue. 0.2ml of 2% bathophenanthroline reagent was added and mixed thoroughly. Care was taken not to expose the tubes to direct light. 0.2ml of orthophosphoric acid reagent and ferric chloride reagent was added and mixed well. Standard vitamin-E in the range of 50–100μg was also treated in a similar way. The absorbance of the pink color was read at 536nm against a reagent blank with ethanol.The values are expressed as μmoles/L of plasma.

Vitamin-C

Vitamin-C (ascorbic acid) was estimated in an aliquot of whole blood after protein precipitation. 1ml of blood was mixed with 2ml 10% TCA and allowed to stand for 5 minutes. 1ml of the supernatant was taken after centrifugation and made upto 3ml with 5% TCA then treated with 0.1ml of the DTC reagent. The tubes were shaken well and incubated for 3 hours at 37°C. 0.75ml of 65% sulphuric acid reagent was added and the color developed was read after 30 minutes at 520nm. Blank contained 3ml of 5% TCA. Standard ascorbic acid

solution in the range of 10–50μg was treated in the same way. Vitamin-C values are expressed as μmoles of ascorbic acid/L of blood.

Reduced Glutathione

Reduced glutathione in blood was measured. 0.2ml of blood was collected with EDTA as anticoagulant and then hemolyzed with 1.8ml of EDTA solution and then 3ml of precipitating reagent was added and mixed thoroughly. It was allowed to stand for 5 min before filtering. Then 2ml of filtrate was added to 4ml of 0.3M disodium hydrogen phosphate solution and 1ml of DTNB reagent. Blank was prepared in a similar manner using distilled water instead of blood filtrate. A set of standards containing 20–70μg of reduced glutathione were taken and made upto 2ml with water. To each tube 4ml of DSHP and 1ml of DTNB reagent was added and the optical density wad read at 412nm. Values were expressed as μmoles/L of blood.

Statistical Analysis

All quantitative measurements are expressed as mean ± SD for the different groups. Statistically significant differences between the different groups studied using student 't' test.

Results and Discussion

The investigation aims to study the effect of physical and biochemical stress on antioxidant activity in the sports women.

Table – 1: Lipid Peroxidation Product Expressed as Malondialdehyde in Plasma.

Subject	Plasma nmoles/dl
Healthy control (n = 26)	0.21 ± 0.062
Group – I (n = 28)	0.22 ± 0.087*

Statistically significant difference are expressed as *P < 0.001, when compared with normal subjects

Lipid peroxidation levels for sports women are significantly elevated compared with the normal subject. Peroxidation and the formation of thiobarbituric acid reactive substance in the tissues is induced by free radicals

formed due to external stressors, leading to a chain reaction, generation of free radical acting on polyunsaturated fatty acid (PUFA) and lipid hydroperoxide. The higher susceptibility of erythrocytes to lipid peroxidation arises out of its high levels of molecular oxygen and iron content. A prominent increase in lipid peroxidation was observed in all tissues. Free radical production and oxidant stress, which are known to increase during exercise, may contribute to the oxidant damage proposed to play a role in the ageing process [35]. Results indicating increase or decrease is even no change of antioxidant enzyme activities in various tissues in old age have been reported. The result of the study showed that amenorrheic female athletes demonstrate an increased potential for lipid peroxidation after exercise.

Table – 2: Antioxidant Enzyme Activities in the Erythrocyte (Mean ± SD).

Subject	GPx (lysate) µgGSH utilized/min/mg Hb	SOD (lysate) IU/mg Hb	Catalase (membrane) µmole H_2O_2 consumed/min/mg protein
Healthy control (n = 26)	0.03 ± 0.023	0.85 ± 0.129	0.01 ± 0.008
Group (n = 27)	0.02 ± 0.012	0.60 ± 0.378	0.03 ± 0.09

Statistically significant differences are expressed as *P < 0.001, when compared with normal subject

Table-2 shows the antioxidant enzymes GPx and SOD activity for sports persons are decreased compared with the normal subjects. Catalase activity is slightly increased with the level of the normal subjects [36]. GPx activities were reduced to certain extent in the exercise group. GPx reveals the most prominent alteration among the antioxidant enzymes. The enzymatic antioxidants defense of striated muscle differs from that of other tissue in old age. Mitochondrial oxidative capacity was reduced in the muscles of old subject while no changes were

observed in antioxidant enzyme activities when compared to the young ones. There are contradictory results on how the antioxidant enzymes are affected by ageing [37]. The changes in the antioxidant enzymes with ageing usually show a different pattern in various tissues.

Table – 3: Non–enzymatic Antioxidant Levels in Blood (Mean ± SD).

Subject	Vitamin-A μmol/L plasma	Vitamin-E μmole/L plasma	Vitamin-C μmole/L whole blood	GSH μmole/L whole blood
Healthy control (n = 26)	0.04 ± 0.014	0.50 ± 0.106	0.12 ± 0.095	0.79 ± 0.322
Group (n = 27)	0.05 ± 0.055*	0.47 ± 0.117*	0.11 ± 0.009*	0.71 ± 0.100*

Statistically significant difference are expressed as * P < 0.001, when compared with normal subjects

In table-3 shows that GSH, vitamin-E and C levels are decreased with the normal subjects and vitamin-A levels are slight increased with the normal subjects. Vitamin-E and C are nutrient antioxidants that can scavenge or reduce free radical; β-carotene functions as a radical trapping antioxidant [32]. It was not only found to quench singlet oxygen but also inhibit photochemical reactions leading to lipid peroxidation. GSH is the principal cellular non protein sulfhydryl compound; it has a variety of functions in bioreduction and detoxification process. Vitamin-A deficient guinea pigs using exhaled pentane and ethane as markers for *in vivo* lipid peroxidation, found that β-carotene treated animals exhibited diminished lipid peroxidation when exposed to free radical forming carbon tetrachloride [18]. Exercise-induced increased in oxidative stress can result in lipid peroxidation which has been implicated in a number of destructive biological process including DNA modification, aging, membrane ion transport damage, cancer and atherosclerosis. A definite increase in lipid peroxidation parameters in athletes were observed in our study in agreement with earlier reports.

Oxidative stress cause by free radicals has become an area of interest in understanding the process of human disease. The term oxidative stress has rarely been defined in a universally accepted way. One accepted definition is a disturbance in the prooxidant-antioxidant balance in favor of the former, leading to potential damage for a disturbance in this balance to occur, it follows that one or both of the following scenarios must be present: (i) A reduction in antioxidant (ii) An increase in reactive species. Tissue damage can be a cause of oxidative stress, but oxidative stress can also be a cause of tissue damage. Oxidative stress occurs when the production of damaging free radicals and other oxidative molecules exceeds the capacity of the body's antioxidant defenses to detoxify them. Oxidative stress and pulmonary inflammations have developed pulmonary fibrosis. Oxidative stress contributes to the pathogenesis of vascular disease one of the prominent feature of arteriosclerosis is the oxidation of low density lipoproteins [38]. Oxidative stress is though to play a crucial role in the development of age related cataract. Vitamin-C prevents the oxidative damage in lens. Oxidative stress has been implicated in both apoptosis and the pathogenesis of cancer. Free radicals may be involved in the cancers of the lungs, cervix skin, stomach, prostate, colon and oesophagus.

Free radicals are generally very reactive molecular moieties possessing unpaired electrons. ROS include hydroxy radicals, superoxide radicals, peroxy radicals and H_2O_2. ROS are produced by both exogenous and endogenous sources. Certain cell type endogenously synthesizes H_2O_2 as a response to activation by specific cytokines and growth factors and show stimulation of protein kinas cascades.

Reactive oxygen species damage cellular macromolecules such as DNA, lipid, carbohydrate and protein [39]. Some essential growth regulatory proteins lose their functions where it is damaged by free radicals and these free radicals converts amino acids to carbonyl derivatives [40]. The inactivation of enzyme by free radical and the accumulation of oxidized proteins may play a critical role in the alterations of cellular function and cell death. Reactive oxygen species leads to oxidative damage of the nucleobase and sugar components of nucleotides in double standard DNA and has been implicated in DNA mutagenesis and covalent cross linking to DNA binding proteins [41]. The reduction of oxygen generates reactive intermediates called free radicals. With the exception of unusual circumstances such as the influence of ionizing radiation free radicals are generally produced in cells by electron transfer reactions. Free radical production in cells can be greatly increased by certain foreign

compounds. Recent studies suggest that arsenic also exerts its toxicity through the generation of reactive oxygen species, which include hydrogen peroxide and other chemical forms known as free radicals. Organic free radicals species can be numerous but the number of oxygen-derived from free radicals that occur in biological system is limited [42].

Antioxidant enzymes like glutathione peroxidase, superoxide dismutase levels are studied in sports person were found to be reduced, while catalase activity is slightly increased. Increased level of lipid peroxidation and antioxidant scavengers like vitamin-E, C and reduced glutathione levels are decreased, but vitamin-A levels were slightly increased. Prolonged exercise leads to increase the lipid peroxidation level, thus the sports women to take the balanced diet especially vitamins and healthy diet is recommended.

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References

1. R.L. Prior, G. Cao, 2000, Vol 35, pp588–592.

2. R.J. Feuers, R. Weindruch, R.W. Hart, 1993, Vol 295, pp191–200.

3. S.W. Fannin, E.J. Lesnefsky, T.J. Slabe, M.O. Hassan, C.L. Hoppel, 1999, Vol 372, pp399–407.

4. P.M. Clarkson, 1995, Vol 35, pp141-145.

5. P.A. Riley, 1994, Vol 65, pp27–33.

6. R. Cross, O.T.A. Jones, 1991, Vol 1057, pp281–288.

7. C. Rice–Evans, R. Burden, 1993, Vol 32, pp71–100.

8. R.H. Esterbaue, H. Zollner, R.J. Schaur, 1990, Vol 45, pp240–268.

9. K.L. Fong, P.B. Mccay, J.L. Poyer, 1976, Vol 15, pp77–89.

10. R.F. Harbe, J.J. Weiss, 1934, Vol 147, pp332–351.

11. T.M. Florence, 1990, Vol 15, pp88–93.

12. H.E. Esterbauer, R.G. Koller, J.F. Koster, 1986, Vol 239, pp405–409.

13. I. Fridovich, 1975, Vol 44, pp147–159.

14. E.D. Harris, 1992, Vol 6, pp2675-2683.

15. O.I. Aruoma, B. Halliwell, 1987, Vol 241, pp273–278.

16. O.I. Aruoma, B. Halliwell, E. Gajewski, M. Dizdaroglu, 1991, Vol 273, pp2601–2604.

17. E.N. Frankel, 1985, Vol 754, pp264–270.

18. H. Esterbauer, 1982, Vol 76, pp101–128.

19. S. Dhakshinamoorthy, D.J. Long, A.K. Jaigwal, 2000, Vol 36, pp201–216.

20. G. Block, M. Dietrick, E.P. Norkus, J.D. Morrow, M. Hudes, B. Caan, L.F. Packer, 2002, Vol 156(3), pp274–85.

21. N. Noguchi, A. Watanable, H. Shi, 2000, Vol 33, pp809–817.

22. P.M. Abuja, R. Albertini, 2001, Vol 306, pp1–17.

23. C.L. Faltman, L.M. Schaefer, T.D. Qury, 2003, Vol 35, pp236–256.

24. B.L. Frei, B.N. Ames, 1989, Vol 86, pp6377–81.

25. I.S. Jialal, S.M. Grnady, 1991, Vol 87, pp597–601.

26. A. Bendich, P. D'Spolito, E. Gabriel, L.J. Machlin, 1984, Vol 114, pp1588–1593.

27. R.F. Burk, 1990, Vol 45, pp383–385.

28. W.A. Al-Turk, S.J. Stohs, F.H. Eu–Rashidy, S. Othman, O. Shaheen, 1987, Vol 34, pp1-8.

29. M.Y. Farooqui, W.W. Day, D.M. Zamorano, 1987, Vol 88, pp177–180.

30. N.I. Krinsky, S.M. Denace, 1982, Vol 69, pp205–209.

31. C.S. Foote, 1979, Vol 34, pp139–171.

32. N.I. Krinsky, 1979, Vol 151, pp649–660.

33. A.L. Tappel, 1997, Vol 47 pp111–131.

34. G.W. Burton, A. Joyce, K.U. Ingold, 1982, Vol 2, pp327-333.

35. L. Deak, R. Darad, 1991, Vol 59, pp123–128.

36. F. Gundug, 2003, Vol 15, pp203–207.

37. L. Abbasoglu-Dogrus, C. Toptani-Tamers, B. Ugurnal, N. Kocak-Yoker, G. Aykac-Toker, M. Uysal, 1997, Vol 98, pp177–180.

38. C.J. Cheng, Y.M. Huseh, M.S. Lai, M.P. Shyu, Y. Chens, K. Wumm, J. Kuotl, 1995, Vol 25, pp53–60.

39. B.A. Freeman, 1982, Vol 47, pp412–426.

40. W.A. Pyror, 1976, Vol 1, pp1–49.

41. P. Floke, 1982, Vol 5, pp223-253.

42. H.A.O. Hill, 1979, Vol 23, pp5-18.

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