Simpo PDF Merge and Split Unregistered Version - http://www.simpopdf.com

Original Article

Age Related Changes in Cardiovascular Response to Oxidative Stress Induced by Exercise

Affan E. Hassen Al-Taai(M.B.Ch.B, M.Sc)*
Hedef D. El-Yassin (Ph.D. Post Doctoral) **

Summary:

Background:
There are many circulatory changes that occur during exercise in order to supply the tremendous blood flow required by the muscles during the stimulatory effects on circulation by the mass sympathetic discharge, the increased arterial pressure and cardiac output. The metabolic effects and the oxidative stress as a result of the work load on cardiac and skeletal muscles could also show changes.

Objective:
This study was designed to investigate the effects of aging process on the vascular response during exercise and also in the oxidative stress according to age.

Subjects and Methods:
Eight healthy Iraqi subjects were enrolled in this study. Divided into three groups according to age, group I (age range 20-29), group II (age range 30-39), and group III (age range 40-49). They were asked to exercise according to modified Bruce protocol. Blood samples were taken from each subject pre and post exercise for biochemical tests. The test included were creatine kinase, uric acid, malonedialdehyde (MDA) , lipid profile (triglycerides, cholesterol, high density lipoprotein HDL). Low density lipoprotein LDL was calculated.

Results:
There is a statistical significance increase in heart rate and systolic blood pressure after exercise in all groups however diastolic blood pressure showed a decrease. A positive linear correlation is present. Mean serum levels of uric acid was shown to be elevated after exercise, meanwhile triglycerides levels decreased significantly in all age groups after exercise and also cholesterol. LDL on the other hand showed a decrease in the younger age group.

Conclusion:
The results obtained set a normative data for the studied parameters for the age group included in the study to be used in the future for the detection and differentiation of any cardiovascular abnormality from age related changes. These data have important clinical implications if we are to prevent the frailty and morbidity associated with old age.

Keywords: oxidative stress, exercise, lipid profile

Fac Med Baghdad 2007; Vol.49, No.4
Received July 2006
Accepted Jun.2007

Introduction:

The ability to impact the environment is mainly dependent on our capacity for physical activity and movement represents more than just a convenience. It is fundamental for our evolutionary development not less important than the complexities and emotion [(1)]. Moreover there are no other normal stresses to which the body can be exposed than even nearly approach the extreme stress of heavy exercises. In fact if some of the exercises were continued for prolonged periods they might easily be lethal[(2)].

Meanwhile reports show increasing mortality rate from cardiovascular disorders with advancing age and sedentary life style over the world. The impact of physical activity as well as habitual exercise practicing in reducing cardiovascular events in older adults has been noticed. There are many circulatory changes that occur during exercise to supply the tremendous blood flow required by the muscles during the stimulatory effects on the

**Department of Physiology, College of Medicine, University of Baghdad.*
***Department of Physiological Chemistry, College of Medicine, University of Baghdad.*

Simpo PDF Merge and Split Unregistered Version - http://www.simpopdf.com

circulation by the mass sympathetic discharge, the increased arterial pressure and cardiac output. These cardiovascular responses could vary between children , adults and elderly normal people due to age related effects on the heart as well as the circulatory system[(2)]. Moreover the metabolic effects and the antioxidants stresses as a result of the work load on cardiac and skeletal muscles could show changes between different groups[(3)]. In the meantime increased metabolism during exercise is a potential source of oxidative stress with a situation increased between oxidants and antioxidant levels. Free radicals which are highly reactive chemical species may increase during exercise [(4)].

This study was designed to investigate the effects of aging process on the cardiovascular responses during exercise and to explore the changes in the oxidative stress according to age in an attempt to set a normative data for groups included to be used in the future for the detection and differentiation of any cardiovascular abnormalities from age related changes.

Subjects and methods

Subjects

Eighty healthy Iraqi subjects were enrolled in this study. They were categorized according to their age, into three groups. Group I included subjects of age range 20-29 years, while group II included subjects of age range between 30-39years, and the last group (group III) was consisted of subjects with age range 40-49 years.

All subjects were volunteers selected from medical students and other volunteers. They were thoroughly examined by a physician to exclude any unhealthy subjects. All were asked to have a good night sleep and not to do any physical activity prior to the experiment in order to avoid any possibility of affecting the secretion of growth hormone.

Height was measured to the nearest centimeter using a measuring tape while weight was measured using (Berkel weight measuring device) BMI was calculated from height and weight using the formula wt (kg/ht(m^2)[(5)].

The Bruce (modified) protocol for treadmill testing was applied.

Venous blood samples were collected from all subjects 5ml. aspirated prior to the exercise test and similar amount after the exercise. Blood samples were withdrawn by vein puncture from each subject using disposable plastic syringe. The blood was allowed to coagulate at room temperature and then centrifuged for 10 minutes at 2500rpm at 4˚C. The serum was stored at -20˚C until analysis. The biochemical parameters studied included, serum albumin, uric acid, creatine kinase, malonedialdehyde (MDA) and lipid profile (cholesterol, triglycerides, high density lipoprotein HDL, low density lipoprotein LDL and very low density lipoprotein VLDL).

Uric acid was estimated enzymaticly.Kit was supplied by Biomaghraeb. While the activity of CK in serum was determined by the increase in absorbance at 340 nm pH 6.6 at 30˚C. Triglycerides, total cholesterol and HDL in the serum were also determined by the enzymatic method. Kits were supplied by BioMérieux Company -France. Serum level of LDL-C was calculated by Friedwald formula [(6)]. Friedwald, Levy and Friedrickson in 1972 had postulated a formula to calculate LDL-C value, which was based on the assumption that VLDL-C is present in serum at a concentration equal to one fifth of the triglyceride concentration.

Therefore:

$$\text{LDL-C} = \text{TC} - [\text{HDL-C} + \text{VLDL}]$$

$$\text{LDL-C} = \text{TC} - [\text{HDL-C} + \text{TG}/5]$$

The formula is only valid at serum triglyceride concentration of less than 400mg/100ml.

All statistical analysis have been made by applying the excel programm 2002 (10.2614.2625), t-test was applied to estmate the significancy. P value was taken to the closest (P<0.05).

Results:

The general characteristics of the (80) subjects studied who were grouped according to age are shown in table 1.

Simpo PDF Merge and Split Unregistered Version - http://www.simpopdf.com

Table 1: Characteristics of the subjects studied according to age

	GroupI (20-30) n=25	GroupII (30-40) n=26	GroupIII (40-<50) n=29
Mean age (yrs)±SD	24.845 ± 2.625	34.19 ±3.093	45.63 ± 2.557
Mean height (m)±SD	1.714 ± 0.099	1.667 ±0.071	1.688 ± 0.075
Mean weight (kg)±SD	70.08* ± 13.167	78.195* ± 13.455	80.46* ± 14.443
Mean BMI (kg/m^2)±SD	23.886* ± 3.689	28.317* ± 4.308	28.332* ± 4.876

*significance ($p<0.05$) between groups I&II and between groups I&III

The data show positive correlation between age and BMI.

Table 2 presents the mean heart rate results showing significant statistical differences in the mean heart rate after exercise between group I and II and between group I and III ($p<0.05$). Also there is a significant difference in the mean heart rate difference (ΔHR) between group I and II and between group I and III ($p<0.05$). No significant differences in mean heart rate before the test among the three groups.

Table 2: Mean heart rate measurements

	GroupI (20-30) n=25	GroupII (30-40) n=26	GroupIII (40-<50) n=29
Mean HR (at rest) (beat/min)±SD	82.81 ± 11.37	83.32 ± 11.197	80.695 ± 10.4
Mean HR (after exercise) (beat/min)±SD	132.19* ± 20.793	121.215* ± 22.379	118.575* ± 28.88
Mean ΔHR (beat/min)±SD	49.38* ± 20.435	35.055* ± 22.629	37.615* ± 24.85

*significance ($p<0.05$) between groups I&II and between groups I&III

The correlation between age and heart rate after exercise showed a negative linear correlation.

Mean systolic blood pressure results are shown in table 3. It shows after exercise a significant statistical difference between group II and group III; however no significant difference in the mean systolic blood pressure could be noticed for the three groups

Simpo PDF Merge and Split Unregistered Version - http://www.simpopdf.com

Table 3: Mean systolic blood pressure measurements.

	GroupI (20-30) n=25	GroupII (30-40) n=26	GroupIII (40-<50) n=29
Mean SBP (at rest) (MmHg)±SD	123.98 ± 17.533	127.29 ± 13.767	132.25 ± 12.603
Mean SBP (after exercise) (MmHg)±SD	139.81 ± 22.998	148.25* ± 17.007	158.275* ± 15.924
Mean Δ SBP (MmHg)±SD	15.965 ± 16.836	21.355 ± 15.209	24.23 ± 19.736

*significance ($p<0.05$) between groups II&III

A positive linear correlation between age and systolic blood pressure is present.

The mean diastolic blood pressure results are shown in table 4, which shows that there is a decrease in the mean diastolic blood pressure after exercise from the mean diastolic pressure before exercise in all the three groups, but without significant statistical differences among them. Also there is no significant difference in the mean diastolic blood pressure among the three groups.

Table 4: Mean diastolic blood pressure measurements.

	GroupI (20-30) n=25	GroupII (30-40) n=26	GroupIII (40-<50) n=29
Mean DBP (at rest) (MmHg)±SD	78.785 ± 7.15	76.535 ± 7.988	80.835 ± 5.934
Mean DBP (after exercise) (MmHg)±SD	73.635 ± 11.05	72.23 ± 11.323	76.81 ± 8.149
Mean Δ DBP (MmHg)±SD	-4.855 ± 7.234	-4.305 ± 9.513	-3.61 ± 8.338

Membrane integrity is thought to be compromised by oxidative stress and is evaluated through the measurement of creatine kinase. Results are shown in figure (1)

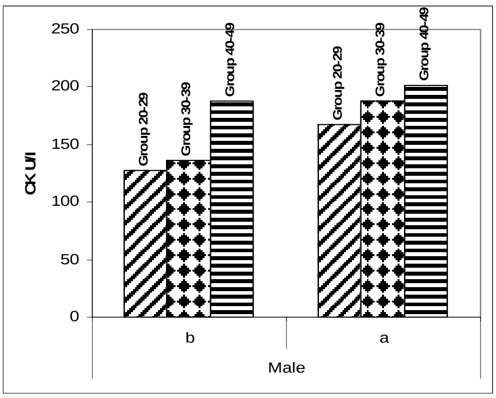

Figure (1): Serum CK level in all three groups' pre (b) and post (a) exercise.

Ongoing oxidative stress induced by maximal exercise was analyzed by measurements of antioxidants which are the defense mechanisms against such stress.

Simpo PDF Merge and Split Unregistered Version - http://www.simpopdf.com

Mean serum levels of uric acid are shown in figure (2). As shown in the figure there is an increase in mean uric acid level after the exercise from its level before exercise. A significant difference was found between group I and group II.

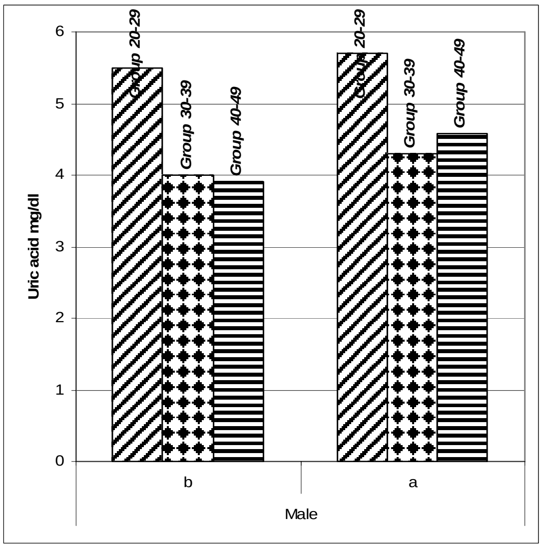

Figure (2): Serum uric acid level in all three groups' pre (b) and post (a) exercise.

Mean value of triglyceride levels (TG) decreased significantly in all age groups after exercise as shown in figure (3). The pre and post exercise values showed a linear positive correlation with increasing age.

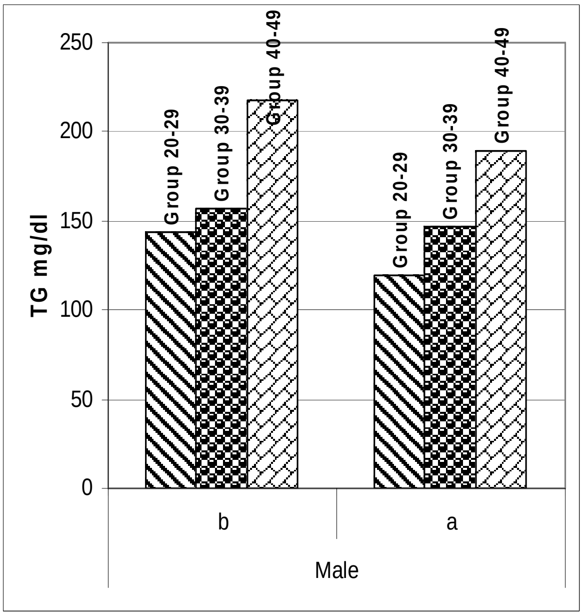

Figure (3): Serum TG level in all three groups male and females' pre (b) and post (a) exercise.

It was found that mean total cholesterol concentration values decreased after exercise in all age groups as shown in figure (4).

Simpo PDF Merge and Split Unregistered Version - http://www.simpopdf.com

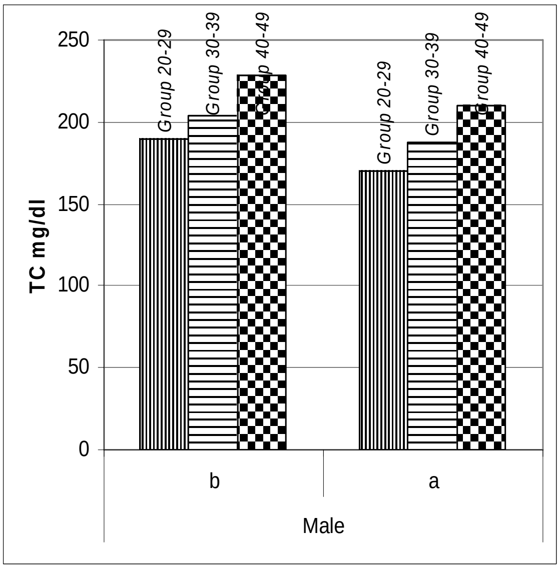

Figure (4): Serum TC level in all three groups male and females' pre (b) and post (a) exercise.
The studied parameters could all be compared as shown in figure (5).

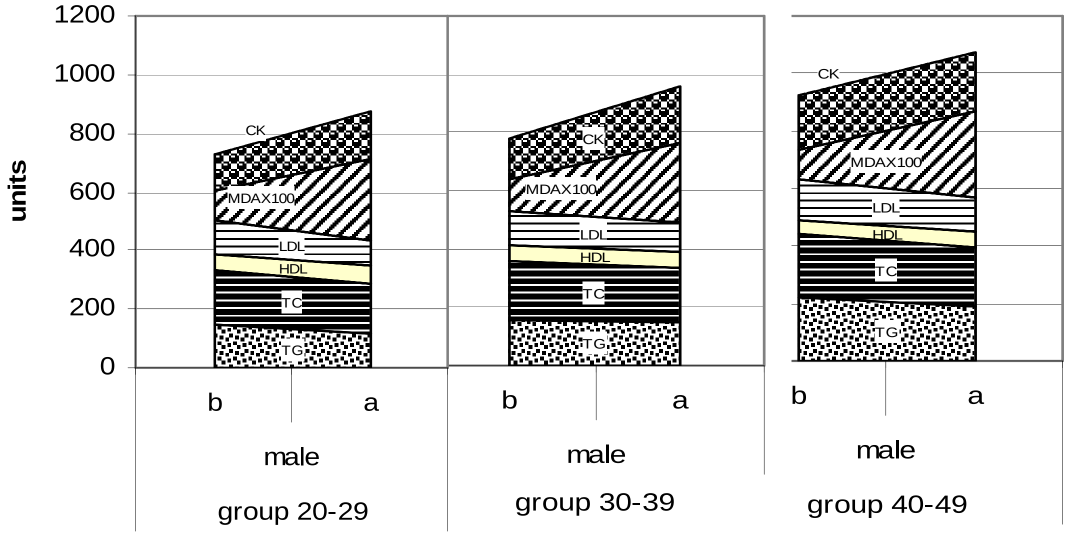

Figure (5): Comparison of all the studied parameters in the three groups, pre (b) and post (a) exercise.

This figure compares the values of all the biochemical parameters studied in this work (in form of area under the curve) in all three groups before and after exercise. The figure shows the same trend of having high triglyceride level in all groups before and after exercise and decrease after that. However the level is lower in the age group 20-29 (group I) and increases with increasing age. Same can be said about the cholesterol. But looking at the HDL area in the figure one can postulate that it has a plateau pattern in all three groups but it decreases with increasing age. LDL on the other hand shows a significant decrease after exercise in the small age group. Such a behavior can also be observed in the two age groups but with less significance. Although the area under the curve is small in the small age group and increases with increasing age. The behavior of MDA and CK is the same in all age groups with a significant increase after exercise. The area under the curve increases with increasing age.

Simpo PDF Merge and Split Unregistered Version - http://www.simpopdf.com

Discussion:

As aging seems to be an increasing phenomena of the world population, there is a renewed interest on the effect of age on the cardiovascular function and on physical fitness [7]. Maximal exercise of short duration is not only associated with sport performance, but it is a factor in many daily activities like climbing stairs hurrying across a busy street or carrying a heavy grocery bag. The ability to perform these types of activities can have a major influence on the independence of older adults, and may serve as an index of an individual ability to live independently [8]. This study shows a significant difference (table 1) in the mean weight and BMI of the subjects studied according to age where the mean weight and BMI increased with age as was reported [9, 10]. These findings could be explained by the fact that the proportion of body fat increases with advancing age and with progressive decline in energy intake and total daily energy expenditure, which is mainly due to decrease in physical activity and to a lesser extent a decrease in basal metabolic rate[11]. In this series the mean heart rate prior to the exercise testing did not differ significantly among the subjects of the three groups. While the mean heart rate increase significantly post exercise in each age group indicating an increase sympathetic stimulation rates during exercise or stress. This finding is similar to other findings reported [12,13]. Table 3 shows a significant increase in the mean systolic pressure after exercise in each group. Similar results were also reported by other authors[14]. Dynamic exercise causes an increase in arterial blood pressure due to the combination of an increase in cardiac output and in the total systemic vascular resistance as a concrescence of active muscles and the arterial baro receptors are reset so they operate normally around the higher blood pressure[15].

Maximal exercise induced oxidative stress means the formation of ROS (reactive oxygen species) and affection of the antioxidant levels. Direct detection of ROS and other free radicals are difficult because these molecules are short-lived and highly reactive in a non-specific manner. On going oxidative damage is thus generally analyzed by measurements of secondary products including derivatives of amino acids, nucleic acids, uric acid, and albumin and lipid peroxidation[16].

Membrane integrity is thought to be compromised by oxidative stress and through the measure of CK in the serum. It was found than the mean CK level prior to exercise testing differs significantly among the three age groups (figure 1). Its level increased significantly post exercise in each age group indicating a damage of the skeletal muscle caused by exercise. An increase in total CK activity, and the appearance of considerable amounts of muscle protein in plasma precipitated by the muscle strain during exercise explained by damage of skeletal muscle detected by histological studies[17].

Figure 1 shows a positive leaner correlation between the CK level and the mean age of the subjects before and after exercise, so CK level increased with increasing age. However it was pointed that older adults experience greater muscle damage following exercise than young subjects which is due in part to the smaller muscle mass and lower VO_2max seen in older men.During exercise albumin was increased in young and decreased in old but there were no age related differences in total protein changes[18].

Also it was found that albumin concentration was not associated with decline in functional performance in men and women but combination of low albumin and low cholesterol levels may increase the risk of future functional decline [19].

The other secondary antioxidant measured was uric acid Uric acid is one of the most important secondary antioxidant chosen to be studied in this work. As shown in figures 2 and 5 there was a significant increase in mean uric acid concentration post exercise in all age groups. Stress caused by exercise enhances the catabolism of ATP to hypoxanthine by virtue of the enzyme xanthine oxidase, this is further converted to uric acid in the process. Free radicals including the toxic superoxide and hydroxyl radicals are produced and cause muscle damage. At the same time uric acid that accumulates in cells leaks or secrets out of the cells causing a significant increase in plasma level [20]. Mean value of triglyceride levels (TG) decreased significantly after exercise in all age groups as shown in figures 3 and 5, due to intramyocellular triglyceride hydrolysis during exercise. The pre and post exercise values showed a linear positive correlation with increasing age being in female

Simpo PDF Merge and Split Unregistered Version - http://www.simpopdf.com

less than in male. Similar findings have been reported, this may be attributed to age related hyperlipidemia due to decreased rate of removal from the plasma.

Mean value of total cholesterol levels (TC) decreased in all age groups after exercise as shown in figures 4 and 5. In figure 5 there is a decrease in lipid profile after exercise when compared to that before exercise in all age groups. This trend is typical for short-term lipid modifications that occur after a single session of aerobic exercise. This might be due to increase in lecithin cholesterol acyltransferase[(21)]. It also be due to the fact that exercise induces modification in hormonal factors promoting lipid mobilization during exercise, plasma catecholamines, growth hormone, cortisol levels increased while insulin decrease[(22)] .

Figure (5) also shows an increase in mean MDA concentration values significantly after exercise in all age groups. After short time resistance exercise there becomes an increase in mobilization of fat-soluble antioxidants despite mobilization of antioxidants, oxidative stress occurs during submaximal exercise which was indicated by increased MDA concentration. While successive increase in MDA formation with increasing age (figure 5) provides evidence for accumulation of an oxidatively damaged lipid components with age [(23)].

Although the behavior of lipid profile and lipid peroxiadtion is the same in all age groups. But it is clear that the extent of variation in these parameters is different between these groups. As shown in figures the levels of triglycerides and cholesterol are higher in group III than in group I and II. Age related hyperlipidemia could be explained by either increased lipid production and /or decreased removal from the plasma. It was proposed that a change in cholesterol fraction carried by different lipoprotein fractions could explain such age related hypercholesterimia. Alterations in metabolism in these lipoprotein macromolecules are accompanied by a decrease in rate of lipoprotein cholesterol estrification [(24)].

References:

1. McArdle WD, Katch FI and Katch VL. "Essentials of exercise physiology" 2nd ed. Philadelphia; Lippincott; Williams and Wilkins, 2000.

2. Guyton AC and Hall JE. "Textbook of Medical Physiology" 11th ed. WB Saunders Co. 2000

3. Turley KR and Wilmore JH. "Cardiovascular responses to treadmill and cycle ergometer exercise in children and adults" J. Appl. Physiol 1997; 83: 948-957.

4. Clarkson PM and Tompson HS. "Antioxidants: what role they play in physical activity and health" Am.J.Clin Nutr. 2000; 72: 637-646.

5. Calle EE. "Body mass index and mortality rate in a prospective cohort of U.S. adults. N Engl. J. Med. 1999; 341: 1097-1000.

6. Friedwald W. T., Levy R.I., and Fredrickson D.S.; Estimation of the concentration of LDL-Chol. In plasma without use of the preparative ultra centrifugation. Clin. Chem. 1972, 18: 499-509.

7. Lakatta EG" Cardiovascular regularity mechanisms in advanced age " Physol Rev 1992; 73: 413-417)

8. Martin J" The effects of age and exercise on short-term maximal performance " J Sport Sciences 1993; 13: 450-458.

9. Medghini F., Dally LFazzini PF, Menotti A" Normal behavior of circulatory parameters during exercise. Reference values for heart rate and systemic blood pressure " G Iral Cardiol 1995; 25(8): 967-75,

10. Hsieh SD, Yoshinaga H and Muto T." Waist-to height ratio, a simple and practical index for assessing central fat distribution and metabolic risk in Japanese men and women" Int.J.Obes. Relat.Metabol. Disod. 2003; 27(5): 610-6.

11. Elia M." Obesity in the elderly" Obes Res 2001; 9(4): 2445-2485.

12. Alonso Dde O, Forjaz CL, Rezende LO and Braga AM." Heart rate response and its variability during different phases of maximal graded exercise" Arq Bras Cardiol 1998; 71(6): 787-92.

13. Lenk MK, Alehan D, Celiker A, Alpay F. "Bruce treadmill test in healthy Turkish children: endurance time, heart rate, blood pressure and

Simpo PDF Merge and Split Unregistered Version - http://www.simpopdf.com

electrocardiographic changes" Turk. J. Pediatric. 1998; 40(2): 167-75.

14. Finnoff JT, Smith J, Low PA, Dahm DL, Harrington SP. "Acute hemodynamic effects of abnormal exercise with and without breath holding" Arch. Phys. Med. Rehabil. 2003; 84(7): 1017-22.

15. Shepherd JT." Circulatory response to exercise inn health" Circulation 1987; 76(1): 3-10.

16. Kohen R and Nyska A. " Oxidation of biological systems: oxidative stress phenomena, antioxidants, redox reactions and methods for their quantification" Toxocol Pathol. 2002; 30(6): 620-50.

17. Ziegler S, Niessner A., Slany J. Muller MM and Geyer UG. "Muscle cell proteins are selectively released into the blood stream by marathon running" Acta Med. Australia 2003; 30(2): 55-8.

18. Morgan AL, Sinning WE, Weldy DL. "Age effects on body fluid distribution during exercise in the heart" Aviat Space Environ Med. 2002; 73(8): 750-7.

19. Ima H, Hayashi T. Negawa T. and Tomida M. "Strenuous exercise induced change in redox state of human serum albumin during intensive Kendo training" Jpn J. Physiol 2002; 52(2): 135-40.

20. Chevion S, Moran DS, and Heled Y. "Plasma antioxidant status and cell injury after severe physical exercise". PNAS, 2003, 100(9): 5119.

21. Aguilo A, Tauler P, Pilar-Guia M. Villa G. et al. "Effects of exercise intensity and training on antioxidants and cholesterol profile in cyclists" J Nutr Biochem, 2003; 14(6): 319-325.

22. Garcia –Araujo M, Semprun-Fereira M, Sulbaran TA et al." National and metabolic factors as risk factors for cardiovascular disease in an adults population in the city of Maracibo , Etadio Zulla Venezuela" Invest. Clin. 2001; 42(1): 23-42.

23. Ramel A, Wagner KH and Elmadfa I." Plasma antioxidants and lipid oxidation after submaximal resistance exercise in men. Eur J Nutr 2004, 43(1): 2-6

24. DeSchrijver R. "Cholesterol metabolism in mature and immature rats fed animal and plant protein. J Nutr. 1990; 120(12): 1624-1632.