The Effect of Regular Exercise Training on Serum Level of Malondialdehyde

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ABSTRACT

Background and objectives : Poly unsaturated fatty acids(PUFAs) that contain two or more double bonds are particularly susceptible to oxidation by free radicals and other highly reactive species. Malondialdehyde(MDA) is one of many low molecular weight end-products of lipid hydroperoxide decomposition and is the most often measured as an index of lipid peroxidation. The aim of the present study was to measure the serum level of MDA in healthy non athletics and athletics.

Methods : This study was carried out during the period from April 2007 to September 2007 on 53 healthy non athletics (27 males and 26 females), and 31 healthy athletics (16 males and 15 females). Serum MDA level was measured colorimetrically using thiobarbituric acid method .

Results: The mean value of serum MDA was significantly higher in healthy athletics than that of healthy non athletics ($p<0.05$). The mean value of serum MDA in females was non significantly higher than that of males ($p>0.05$) in both groups . The mean value of serum MDA was significantly higher in healthy non athletic smokers than that of non smokers ($p<0.001$) .

Conclusions : Based on the findings of the present study , it can be concluded that regular exercise training causes excess lipid peroxidation and generation of significant amounts of MDA, one of the most important harmful free radicals .Therefore athletics should take a diet rich in antioxidants or appropriate amount of antioxidant vitamins (A, C,and E) .

Keywords: Serum malondialdehyde (MDA), Exercise training, Reactive oxygen species (ROS) , Free radicals .

INTRODUCTION:

Lipid peroxidation is the most studied biologically relevant free radical chain reaction [1]. A Free radical ($R\cdot$) is simply defined as any species capable of independent existence that contain ≥ 1 unpaired electrons in its outer orbit and formed when oxygen interacts with certain molecules [2].

Free radicals include reactive oxygen species (ROS) and reactive nitrogen specie (RNS), such as oxygen radicals, superoxide ($O_2^{\cdot-}$), hydroxyl radical ($OH^{\cdot}$), peroxyl radical ($ROO^{\cdot}$), alkoxyl radical ($RO^{\bullet}$), HOCl, ozone (O_3), singlet oxygen ($^1O_2$), H_2O_2, peroxynitrite ($ONOO^-$), nitric oxide radical ($NO\cdot$) and nitrogen dioxide radical ($NO_2^{\cdot}$) [3,4].

ROS can cause peroxidation of lipids leading to damage of membrane permeability, inflammatory diseases, atherosclerosis, loss of enzyme activity involved in the decomposition of ROS, DNA damage leading to mutagenesis, aging and apoptosis of cells [5].

Lipid peroxidation consists of four steps (initiation, propagation, degradation and termination), as it is shown in (Figure 1).

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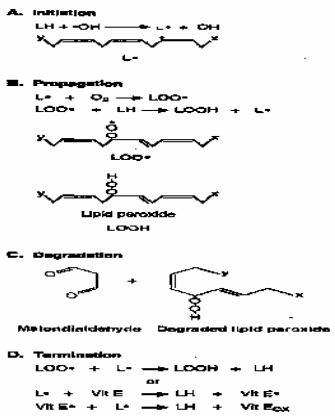


Figure(1): Lipid peroxidation: a free radical chain reaction. Peroxidation is initiated by a hydroxyl or other radical that extracts a hydrogen atom from a polyunsaturated lipid (LH), thereby forming a lipid radical (I^{-}). The free radical chain reaction is propagated by reaction with O2 forming the lipid peroxy radical ($LOO^{\cdot}$) and lipid peroxide ($LOOH\cdot$). Rearrangements of the single electron result in degradation of the lipid. Malondialdehyde one of the compounds formed, is soluble and appears in the blood. The chain reaction can he terminated by vitamin E and other lipid-soluble antioxidants that donate single electrons. Two subsequent reduction steps from a stable, oxidized antioxidant [6].

During oxidative metabolism, much of the oxygen consumed is bound to hydrogen, thus forming water. However, it has been estimated that 4-5% of the oxygen consumed during respiration is not completely reduced to water, instead forming free radicals. Thus as oxygen consumption increase during exercise, a concomitant increase occurs in free radical production and lipid peroxidation that makes a major contribution to (ROS)-induced injury. Moreover, exercise is postulated to generate free radicals by other means, including an increase in adrenaline and other catecholamines that can produce oxygen radical, when they are metabolically inactivated, production of lactic acid that can convert a weakly damaging free radicals (superoxide) into a strongly damaging one (hydroxyl), and inflammatory responses to secondary muscle damage incurred with overexertion [7,8]. Eventually lipid (fatty acids with two or more double bonds) degradation occurs, forming products such as malondialdehyde (MDA) [7]. MDA is one of many low molecular weight end- products of lipid peroxide decomposition and is the most often measured as a biomarker and an index of the overall lipid peroxidation [9-12]. Heavy physical exercise increases oxygen consumption and potentially initiates enhanced formation of (ROS). This is in turn leads to oxidative stress and cellular damage if not properly counteracted [10,13]. Heavy exercise training increases also excessive use of body muscles, skeletal muscle blood flow, oxygen extraction by three folds ,mobilization and transport of fatty acids, oxidative damage of LDL-C, systolic arterial and pulse pressures , inversely the total calculated peripheral resistance and free fatty acid (FFA) re-cycling by (45%) are decreased[14,15]. Data obtained by many investigators indicate that exercise training uses more fat for the same sub- maximal task due to the increased mitochondrial mass and activity, so it is one of the most important internal generated source of free radicals that cause impairment of oxidant/ antioxidant equilibrium[10]. Moreover, a vast volume of data has accumulated that has been interpreted to mean that regular exercise training increases the subject's capacity to oxidize lipids [16,17]. In addition Oostenbrug et al [18] reported that incorporation of the highly unsaturated fatty acids in membranes may increase the membrane's susceptibility to lipid peroxidation during prolonged regular exercise training. Cigarette contains high concentrations of various free radicals such as super oxide, hydroxyl and nitric oxide, so smoking increases the production of free radicals [9]. Shakir et al [19], conducted a study on the effect of cigarette on the lipid and lipoprotein metabolism and the activity of the enzymes involved such as lecithin cholestero acyl transferase (LCAT) and lipoproteinlipase (LPL) and

reported that cigarette smoking, most likely affects lipid and lipoprotein metabolism and these include the reduction of LCAT and LPL activities and increase nicotine will cause more lipolysis and generation of more unsaturated fatty acids, which are susceptible for peroxidation. In light of these data and to the best of our knowledge, no attempt has been made to study this important and unexplored field in Iraqi-Kurdistan region. Therefore the present study was designed to investigate the influence of long-term regular exercise training on the serum level of MDA as a biomarker and an index of lipid peroxidation in a group of physical education college students and comparing the results obtained with those of sedentary lifestyle students in Erbil city .

MATERIALS AND METHODS:

1-SUBJECTS

This study was conducted over a period of six months, from April 2007 to September 2007 at the dept. of medical biochemistry/ college of medicine/ Hawler medical university/ Erbil/ Iraq. The present investigation was conducted on 84 volunteer students, which were divided into two groups:

Group 1 (control group = non athletic group = non exercise training group): Included 53 healthy non athletic volunteer students (27 males and 26 females), of fourth year / medical colleges (Medicine, Dentistry, Pharmacy, and Nursing)/ Hawler medical university. The control subjects who participated in this study, none of them had engaged in any regular exercise program prior to this study. Their mean age was (22.5 ± 0.25) years, and the rang of age was (22-26) years.

Group 11 (exercise training group = athletic group): Included 31 healthy athletics (16 males and 15 females), in the fourth stage/ physical education college/ Salahaddin university. The exercise group underwent the aerobic training programs five days / week. Every day the training session lasted for 2-3 hours approximately, and including warming up and cooling down periods . Their mean age was (22.75) years and the range of age was (22-26) years. Details concerning both groups are shown in (Table 1). Non of these subjects included, in this study had clinical or biochemical evidences of any type of diseases, none were taking medications (anti-oxidants) and informed consent was obtained from each individual.

2-SAMPLES

Five to ten mls of venous blood was withdrawn from each individual using disposable syringes. Theblood samples were allowed to clot and serum was separated by centrifugation at 3000 rpm for 10 minutes . All serum samples were analyzed the same day.

3-METHODS

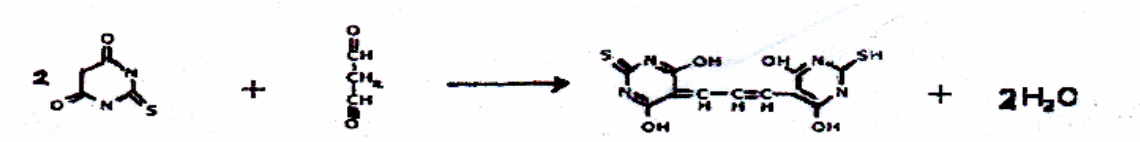
MDA, a secondary product of lipid peroxidation and a thiobarbituric acid-reactive substance (TBARS) was estimated according to the method described by Geuge and Aust [20], the principle of which was explained by Ge′rard Monnier et al [21] and Gutteridge and Halliwell [22] and depends on the colorimetric reaction with thiobarbituric acid. The intensity of the red-florescent MDA-TBA2 adduct is proportional to the amount of MDA in the sample, which is measured at 532 nm, (Figure 2). with thiobabituric acid (TBA). The intensity of the red-florescent MDA-TBA2 adduct is proportional to the amount of MDA in the sample which is measured at 532 nm, (Figure 2).

4-STATISTICAL ANALYSIS

The statistical evaluation of the results [mean, standard deviation (S, D) and standard error of mean (S.E.M)] were calculated using the scientific calculator [prob, 4h, 105]. The different variables were compared to each others, simple correlations were tested with the unpaired student ‘t’ test. $P < 0.05$ is regarded as significant [23].

Table (1): Details of the number, age and sex of the studied groups.

Groups	Sex	Number	Age (years) (Mean ±S.E.M)	Range of age (years)
Group 1	Males	27	22.8 ± 0.33	22—26
	Females	26	22.3± 0.21	22—26
	Total	53	22.5± 0.25	22—26
Group 11	Males	16	22.9 ± 0.51	22—26
	Females	15	22.6 ± 0.42	22—26
	Total	31	22.75 ± 0.45	22—26



Thiobarbituric acid Malondialdehyde MDA-TBA adduct

Figure(2): Formation of the MDA-TBA adduct. [21]

RESULTS:

Table (2) shows the mean values of serum MDA in group I (1.38±0.026 mmol/L), and group II (1.67 ±0.150 mmol/L). The mean level of serum MDA in group II was significantly higher than that of group 1 ($p< 0.05$).

Table (2): Details of serum MDA mmol/L (Mean ± S.E.M.) in both groups

Groups	MDA mmol/ L (Mean ± S.E.M.)	P value
Group 1	1.38 ± 0.026	
Group 11	1.67 ± 0.150	< 0.05

Table (3) shows the mean values for serum MDA in males and females in both groups . The mean level of serum MDA in females was non significantly higher than that of males ($P > 0.05$) Table (4)shows the mean levels of serum MDA in group I according to smoking (smokers and non smokers). The data obtained indicate also that the level of serum MDA in smokers was significantly higher than that of non smokers ($p <0.001$).

Table (3): Details of serum NDA mmol / L (Mean ± S.E.M) in males and females in both groups.

Groups	Sex	Mean± S.E.M.	P values
Group 1	Males	1.350 ± 0.034	N.S.
	Females	1.395 ± 0.020	
Group11	Males	1.61 ± 0.140	N.S.
	Females	1.72 ± 0.160	

Table (4): Details of serum MDA mmol/L (Mean ± S.E.M.) ,in group 1 (smokers and non smokers)

Subgroups of group 1	NO.	Serum MDA Mean±S.E.M	P value
Smokers	10	1.644 ±0.20	< 0.001
Non Smokers	43	1.088 ±0.16	
Total	53	1.38 ±0.15	

DISCUSSION:

The mean level of serum MDA in group 11 was significantly higher than that of group I ($p< 0.05$). This result is in agreement with those obtained by Child et al [24], who conducted a study to evaluate the effects of elevated serum antioxidant capacity and plasma MDA concentration in response to a stimulated half- marathon and found that the rise in total antioxidant did not prevent exercise-induced lipid peroxidation as MDA was elevated after exercise. The same authors concluded that, this may indicate inadequacies in the antioxidant defense system during the half-marathon run. Inversely, the data obtained in this study are in disagreement with those of Dixon [25], who conducted a study on the effects of moderate-intensity whole-body resistance exercise and found that it had no effect on serum MDA concentration. Exercise is one of the internally generated source of free radicals. Endurance exercise can increase oxygen utilization from 10 to 20 times over the resting state. This greatly increases the generation of free radicals. prompting concern about enhanced damage to muscles and other tissues, for that athletes need to take extra antioxidants to defend against the increased free radicals resulting from exercise [26]. Prolonged exercise increases the production and hence the concentration of free radicals in skeletal muscles and the heart, prolonged endurance exercise increases body temperature. The normal body temperature especially in response to frequent exposure to elevated temperatures cause denaturation of enzymes which contain metal ions, all enzymes that play as a catalytic in anti oxidant process are metalloenzymes [27].In both groups the mean level of serum MDA in females was non significantly higher than that of males ($p >0.05$). The data obtained in the present study are also in agreement with those obtained by EL-Yassin et al[28], and McMurrary et al. [29], who conducted studies on the effects of exercise training on the serum levels of MDA and found that physical exercise increases oxygen consumption above resting levels, and potentially initiates enhanced formation of MDA. This in turn leads to oxidative stress and cellular damage. The non significant higher level of serum MDA in females than that of males may indicate a state of wider deprivation of antioxidants in females and may be related to the hormonal imbalance in females [27]. The data obtained in group 1 indicate also that the level of serum MDA in smokers was significantly higher than that of non smokers ($p < 0.001$). This result is similar to the results of Altunas et al [30], who conducted a study on the effects of smoking on the serum level of MDA, and they reported that the concentration of serum MDA was higher in cigarette smokers than in non smoker control subjects. However no relation was found between lipid peroxidation, the serum level of MDA and the number of cigarette smoked by an individual daily. This finding supports the hypothesis that oxidative damage in smokers is due to the number of hours of active exposure to cigarette smoke [31]. In another study done by Khan, and Basser [32], it was found that smokers with coronary heart disease showed significantly increased ($p <0.025$) in the serum level of MDA, as compared to nonsmokers with coronary heart disease. The same authors concluded that elevated serum levels of MDA indicate an increase in the production level of oxygen free radicals, suggesting their possible role in atherogenesis, leading to coronary heart disease. Fraga et al [33], reported also that lungs and urine obtained from cigarette smokers contain elevated levels of free radicals. Finally Block et al. [34], conducted a study to examine the influence of factors associated with oxidative stress in human population and showed a positive relation between serum level of MDA and smoking.

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