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Ameliorative effect of sixteen weeks endurance training on biochemical and oxidative damage in high fat diet induced obese rats

Seyed Reza Emami[1], Mahvash Jafari[2]*, Rouhollah Haghshenas[3] & Aliasghar Ravasi[4]

[1]Department of Biochemistry, Faculty of Medicine, Baqiyatallah University of Medical Sciences, Tehran, Iran
[2] Exercise Physiology Research Center, Baqiyatallah University of Medical Sciences, Tehran, Iran.
[3]Departments of Human Science, Semnan University, Semnan, Iran
[4]Department of Exercise physiology, Faculty of Physical Education and Sports Science, University of Tehran, Tehran, Iran.

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Obesity is mainly caused by consumption of high fat diet (HFD) and a lack of physical activity. Physical activity is an efficient strategy to delay development of obesity. In this studyv we tried to evaluate attenuating properties of 16 weeks endurance training on plasma oxidative stress and some biochemical parameters in HFD induced obese rats. Twenty-four male Wistar rats were randomly divided into four groups: standard diet group (SD), standard diet with endurance training group (ESD), HFD group and HFD with endurance training group (EHFD). After sixteen weeks, plasma was prepared and evaluated for measurement of different parameters. The results showed that HFD significantly decreased the activities of superoxide dismutase (33.58%), catalase (26.51%) and glutathione S-transferase (22.77%), while endurance training increased these enzymes activities. However, exercise ameliorated the increased malondialdehyde level and depletion of glutathione. In addition, it significantly reduced the increased levels of liver enzymes activities and lipid profiles. These findings suggest that endurance training has found to have beneficial effects against HFD-induced oxidative damage through increasing reduced antioxidants levels and inhibition of lipid peroxidation due to its antioxidant property. Thus, it can be considered an interesting strategy for the management of obesity related diseases.

Keywords: Antioxidants, Lipid peroxidation, Lipid profiles, Obesity, Physical exercise

Obesity is becoming a major challenge for public health that increases the risk for a number of diseases including diabetes, cardiovascular diseases, cancer, cognitive dysfunction and anxiety disorders. The prevalence of obesity is progressively increasing worldwide and one-third of the world population is currently classified as obese/overweight[1-3]. It has been estimated that worldwide, 650 million adults (overall prevalence of 13.0%) were obese in 2016. The worldwide prevalence of obesity nearly tripled between 1975 and 2016[4]. In 2015, globally, a burden of about 4.0 million deaths (7.1% of all deaths) and 120 million disability-adjusted life years (4.9% of all DALYs) were attributed to overweight and obesity. Overall, about 22.7% of the Iranian's adult population (15.3% of men and 29.8% of women) was obese in 2016[5].

Obesity is mainly caused by a sedentary lifestyle excess energy intake, the consumption of high fat diet (HFD), lack of physical activity and exposure to stress[6,7]. The consumption of a HFD is considered a major cause of the development of obesity-related health complications[1]. Oxidative stress caused by the consumption of a HFD is evident in most experimental models and patients with clinical conditions[1,2]. HFD-induced obesity stimulates the production of inflammatory cytokines, which is associated with synthesis of reactive oxygen species (ROS). Overproduction of ROS can cause extensive damage to macromolecules, including proteins, DNA and membrane phospholipids, which leads to lipid peroxidation and oxidative stress and finally cell death[2,6].

Exercise is useful way to prevent the development of obesity and its associated complications[6,7]. It is associated with numerous health benefits. It reduces body weight and adiposity, improves physiological and functional capacity in the humans, increases bone density, decreases tissue lipid content and enhances fatty acid oxidation and insulin activity[2,8]. Exhaustive exercise increases the production of ROS via an electron leak of the mitochondrial electron transport

*Correspondence:
Phone: +98 21 22289942; Fax: +98 21 22830262
E-Mail: m.jafari145@gmail.com

Table 1 — Effect of 16 weeks endurance training and high-fat diet on plasma biochemical parameters in control and experimental groups of rats

Parameters	SD	ESD	HFD	EHFD
Glucose (mg/dL)	137.13±4.98	129.54±4.75‡‡‡	184.49±3.94***$^{,\#}$	158.38±3.93*
Urea (mg/dL)	27.13±1.15	26.38±3.18	33.5±1.75	29.13±1.19
Creatinine (mg/dL)	0.51±0.025	0.50±0.044	0.59±0.024	0.57±0.025
AST (U/L)	31.28±2.83	30.41±2.04	45.04±1.77***$^{,\#}$	32.09±1.88
ALT (U/L)	22.06±1.90	21.88±0.61	32.16±1.57***$^{,\#}$	23.81±1.99
LDH (U/L)	155.53±8.49	149±4.96‡	225.54±13.63***$^{,\#}$	186.87±9.16
GGT (U/L)	30.80±2.20	27.29±3.15††	41.70±1.75*	32.44±3.46
ALP (U/L)	160.13±3.41	154.49±3.44	178.85±4.75*$^{,\#}$	156.5±6.32
CK (U/L)	37.97±1.79	39.55±2.22†,‡	53.61±3.20**	52.13±3.86**
TG (mg/dL)	70.5±6.58	62±4.31	94.38±3.49**$^{,\#}$	70.25±3.03
TC (mg/dL)	82.5±3.59	77.5±2.29	120.88±3.97***$^{,\#}$	85.5±2.78
VLDL (mg/dL)	14.11±1.32	12.4±0.86	18.88±0.70**$^{,\#}$	14.05±0.61
LDL-C (mg/dL)	36.53±5.33	29.85±2.76	78.95±3.37***$^{,\#}$	38.83±3.54
HDL-C (mg/dL)	31.88±2.05	35.25±2.25	23.05±1.89$^{*,\#}$	32.63±1.73
L-DL-C/HDL-C (mg/dL)	1.15±0.9	0.85±0.39	3.42±0.42**$^{,\#}$	1.19±0.22
TC/HDL-C (mg/dL)	2.59±0.38	2.20±0.39	5.24±0.64**$^{,\#}$	2.62±0.61

[Groups: SD, standard diet; ESD, Endurance training plus standard diet; HFD, high-fat diet; and EHFD, Endurance training plus high-fat diet. Parameters: AST, aspartate transaminase; ALT, alanine transaminase; LDH, lactate dehydrogenase; GGT, γ-glutamyl transferase; ALP, alkaline phosphatase; CK, creatin kinase;TC, total cholesterol; TG, triglyceride; HDL-C, high-density-lipoprotein-cholesterol; VLDL, very low-density-lipoprotein and LDL-C, low-density-lipoprotein-cholesterol. Values are expressed as mean±SEM (n=7). *P<0.05, **P<0.01 and ***P<0.001 *vs*. SD group; $^{\#}P$<0.05 *vs*. ESD and EHFD groups; $^{†}P$<0.05 and $^{††}P$<0.01 *vs*. HFD and $^{‡}P$<0.05 and $^{‡‡‡}P$<0.001 *vs*. EHFD group]

Fig. 2 — Effect of 16 weeks endurance training and high-fat diet on plasma oxidative stress biomarkers in control and experimental groups of rats. SD, standard diet group; ESD, endurance training plus standard diet; HFD, high-fat diet group; and EHFD, endurance training plus high-fat diet; SOD, superoxide dismutase; CAT, catalase; GST, glutathione S-transferase; GSH, glutathione; and MDA, malondialdehyde. [Values are expressed as mean±SEM (n=7). *P <0.05, **P <0.01 and ***P <0.001 *vs*. SD group; $^{\#\#}P$<0.01 and $^{\#\#\#}P$<0.001 *vs*. HFD group; $^{‡}P$<0.05 and $^{‡‡}P$<0.01 *vs*. EHFD group]

gain and food intake, which was in agreement with the various previously published studies[11,21]. This significant lower body weight in trained rats may be due to the reduction of the amount of adipose tissue resulting in decreased generation of sex hormones, glucose, leptin, the increase appetite-suppressing neuropeptide hormones levels, and negative energy and fat balance linked with increased energy expenditure and fat oxidation during the exercise[12,19]. Although a study showed that no significant difference was observed on average body weight and/or food intake of rats fed the HFD and under exercise[22]. Chaolu *et al* showed that the food intake of the mice in the exercise groups significantly increased as compared to the non-exercise groups[23].

HFD increases the generation of ROS by stimulating proinflammatory cytokines such as tumor-necrosis factor-alpha (TNF-α), interleukin-6 (IL-6) and interleukin-8 (IL-8), the well-known stimulators of NADPH oxidase as the main enzyme responsible for ROS generation[19,24]. Many cells are capable of responding to HFD-induced increases in ROS via endogenous protective mechanisms, including changes in enzymatic endogenous antioxidants such as SOD and CAT. SOD scavenges superoxide radicals to less reactive H_2O_2 and O_2, while CAT

scavenges H_2O_2 to water and molecular oxygen[25]. A significant decreased plasma SOD and CAT activities were noted in the present study in HFD group, which was the result of high oxidative stress. Inhibition of antioxidant enzyme activities may be a consequence of inactivation of enzyme proteins from increased ROS production[26]. In addition, the depletion of SOD activity increases the endogenous superoxide anion, which would have further consequences. It reacts with NO to form peroxynitrite radical and generates hydroxyl radical in conjunction with H_2O_2 through the Haber–Weiss and Fenton reactions, all exacerbating lipid peroxidation, DNA damage and cell death[14]. These results are consistent with previous studies showing that tissue antioxidant defenses may be compromised in high-fat diet-fed animals[2,27]. Cavaliere and coworkers showed that SOD activity was elevated after 1 week of HFD, while markedly decreased after 12 weeks. They suggested that the elevated SOD activity was able to balance ROS levels only for the first week and ROS levels increased progressively during the treatment reaching high values at 12 and 18 weeks when SOD activity was low[28]. In addition, both exercise groups in the present study increased SOD and CAT activities due to the capability to develop a compensation oxidative stress in tissue by means of an adaptation of the antioxidant and repair systems[29]. Exercise training reduces the white adipose tissue mass and the expression of NADPH oxidase and stimulate the expression of nuclear factor kappa B (NF-κB), which leads to increase the expressions of antioxidant enzymes like SOD and CAT[2,24]. Several studies have demonstrated that antioxidant enzymes activities were increased in blood or in tissues after aerobic exercise[2,30]. However, several other studies have reported a decrease in antioxidant enzymes activities in the muscle, liver, kidney and blood of animals by exhaustive exercise[8,9], and other study have found no change in antioxidant enzymes activities after exercise[31]. The response of training on oxidative stress would appear different according to the type, duration and intensity of exercise, previous exercise exposure, subject age and species, nutritional status, type of tissue, animal species and assay technique employed[8].

GST is a group of enzymes capable of conjugating GSH with ROS for protecting tissue from lipid peroxidation and oxidative stress and its levels can reflect the antioxidant capacity of the body[14,32]. In present study, the GST activity was decreased in rats fed HFD, while it was increased in both exercise groups. There were significant differences in the GST activity between HFD and exercise groups. The decreased GST activity in HFD-treated rats may be due to inactivation of enzyme by increased free radicals and depletion of GSH (Figure 2), which is essential for GST activity. These findings are in agreement with the results of the previous reports that HFD feeding significantly decreased the activity of GST in various tissues[20]. A study has showed that GST activity was significantly increased in the muscle and no change in liver after exercise[8]. Vukovic et al. showed that liver GST activity was significantly decreased in HFD and physical activity[33].

GSH as powerful antioxidant in the cells acts as an ROS scavenger and as a substrate for several enzymes in different metabolic pathways, including GST and glutathione peroxidase. It is synthesized in the liver cells and then distributed to different organs through blood[34]. Our data showed that a GSH depletion of 35.79% in HFD feeding rat may be due to increased utilization of GSH for participation of GSH in neutralizing free radicals produced by HFD, diminished GSH synthesis in liver and limited intracellular reduction of oxidized GSH level (GSSG). Diminished GSH indicates impair the cells defense against the toxic actions of ROS and may lead to oxidative stress and cytotoxicity. Depletion of GSH leads to the production of GSSG and finally decreased the ratio of GSH/GSSG in tissues of rats, which is indicative of oxidative stress[8]. The increased GSH content in training groups in comparison to the HFD group could be due to the antioxidant potential of the exercise[35]. Yol and coworkers reported that regular aerobic exercise for 4–6 weeks could improve total antioxidant capacity in male soccer athletes[36].

One of the molecular mechanisms involved in obesity-induced toxicity is lipid peroxidation leading to the damage of the cellular membrane structure and function. MDA is most frequent marker of lipid peroxidation and oxidative stress[8,37]. Elevation of MDA level in the present study leads to inactivation of antioxidant enzymes by cross-linking with MDA, which further cause increased accumulation of ROS, which further enhances more lipids peroxidation[38]. A significant decrease in lipid peroxidation in ESD and EHFD groups was observed. Several studies have reported an increase in lipid peroxidation products in animals by HFD feeding and endurance training in animals[27,33]. However, other study in rats has reported

no change in MDA level in response to physical activity[31]. The discrepancy between these results may be due to the exercise intensity, participant's age and body status[31].

Oxidative damage also leads to hepatocytes damage in HFD-fed rats, as evidenced by the increased plasma activities of the AST, ALT, ALP, GGT and LDH enzymes[27]. HFD-induced membrane lipid peroxidation leads to an increased permeability of liver and the leakage of these enzymes into the plasma[14]. The high levels of these enzymes in our study are attributed to fatty liver induced by HFD and return of their activities to its normal levels in both exercise groups, suggesting the amelioration of fatty liver[8]. This decrease may be due to the consequence of prevention of liver damage from the lipoperoxidation by the antioxidant potential of the exercise[35]. Adiponectin secretion is inhibited by several factors including high level of TNF-α and oxidative stress. Decreased adiponectin level in the HFD was demonstrated to be associated with decreased insulin sensitivity[2,28]. Exercise training significantly reduced the increased levels of blood glucose due to the increased adiponectin level and insulin sensitivity[6,22].

Hyperlipidemia, oxidative stress and obesity are the important causative factors for the development of cardiovascular diseases. The LDL to HDL and TC to HDL ratios are considered to be good prognostic indicators for atherosclerosis extent[21,39]. In present study, the plasma levels of TC, TG, VLDL and LDL-C in the HFD group were increased, while HDL-L level was decreased, all of which eventually trigger the development of lipotoxicity and lipid accumulation in the liver[27]. The data in our study showed that an increase in LDL-C was accompanied with a decrease in HDL-C, which may suggest a change from HDL-C into LDL-C. HFD induces the production of ROS, which react with lipoproteins to produce oxidation states, thus diminishing the cellular uptake of lipids from the blood[27,40]. This results are similar to results other studies[22,27]. However, the decreased these parameters in training groups in comparison to the HFD group could lead to a lowering of the atherogenicity and therefore a significant reduction in the potential incidence of coronary heart disease[2]. This decreased of lipids may be caused by the inhibition of impaired lipid digestion and absorption, improvement in glucose and lipid metabolism, increased antioxidant defense, and down-regulation of lipogenic enzymes[22]. Several studies have demonstrated that exercise decreases blood lipid and lipoprotein levels lipids in humans and rats[6,41,42]. Huang and coworkers showed that the increases in lipid indices induced by the administration of HFD were not affected by the endurance exercise intervention[21].

Our results suggest that the inhibition of antioxidant enzyme activities by HFD increases the endogenous free radicals and lipid peroxides, leading to intracellular Ca^{2+} influx and the release of cytochrome c from mitochondria to cytosol, which is a crucial step in apoptotic signaling through the activation of caspases[43,44]. In addition, overproduction of ROS by HFD activate NF-κB, which in turn increases the expression of cytokines genes such as TNF-α, leadings to inflammation and depletion of GSH and increased MDA levels[8,14]. Depletion of GSH induces apoptosis in cells by interacting with proapoptotic and antiapoptotic signaling pathways and activating several transcription factors, such as AP-1 and NF-κB[43]. Exercise training has been shown to reduce cell size of adipocytes, and decrease the level of inflammation in adipose tissue in mice[2]. Long-term exercise reduces the apoptosis in muscles by increasing the concentration of Bcl-2 as the antiapoptotic marker[43,44]. However, further studies are required to investigate the effects of various high fat diets formulas and exercise models on induction of cell death in variety of cell types using an *in vivo* system.

Conclusion

According to our results, oxidative stress is an essential mechanism involved in HFD-induced adversity effect, as evidenced by decreased GSH level and antioxidant enzymes activities as well as enhanced lipid peroxidation. Endurance training has ameliorative effect against HFD-induced oxidative damage through increasing the level of antioxidants and attenuating lipid peroxidation due to its antioxidant property. Thus, it can be considered an interesting strategy for the management of on development of obesity and obesity-related metabolic diseases. Further *in vivo* studies are required to investigate the effects of various high fat diets formulas and exercise models on induction of cell death.

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Conflict of Interest

Authors have no competing interests.

References

1 Gerardi G, Cavia-Saiz M, Rivero-Perez MD, Gonzalez-SanJose ML & Muniz P, Wine pomace product modulates oxidative stress and microbiota in obesity high-fat diet-fed rats. *J Funct Foods*, 68 (2020) 103903.

2 Al-thepyani M, Algarni S, Gashlan H, Elzubier M & Baz L, Evaluation of the Anti-Obesity Effect of Zeaxanthin and Exercise in HFD-Induced Obese Rats. *Nutrients*, 14 (2022) 4944.

3 Moholdt T, Parr EB, Devlin BL, Debik J, Giskeodegard G & Hawley JA, The effect of morning vs evening exercise training on glycaemic control and serum metabolites in overweight/obese men: a randomised trial. *Diabetologia*, 64 (2021) 2061.

4 Pourfarzi F, Sadjadi A, Poustchi H & Amani F, Prevalence of overweight and obesity in Iranian population: A population-based study in northwestern of Iran. *J Public Health Res*, 11 (2022) 2475.

5 Djalalinia S, Saeedi Moghaddam S, Sheidaei A, Rezaei N, Naghibi Iravani SS, Modirian M, Zokaei H, Yoosefi M, Gohari K, Kousha A, Abdi Z, Naderimagham S, Soroush AR, Larijani B & Farzadfar F, Patterns of obesity and overweight in the iranian population: findings of steps 2016. *Front Endocrinol*, 11 (2020) 42.

6 Juszczak F, Vlassembrouck M, Botton O, Zwakhals T, Decarnoncle M, Tassin A, Caron N & Decleves AE, Delayed exercise training improves obesity-induced chronic kidney disease by activating AMPK pathway in high-fat diet-fed mice. *Int J Mol Sci*, 22 (2020) 350.

7 Wang J, Zhang Q, Xia J & Sun H, Moderate Treadmill Exercise Modulates Gut Microbiota and Improves Intestinal Barrier in High-Fat-Diet-Induced Obese Mice via the AMPK/CDX2 Signaling Pathway. *Diabetes Metab Syndr Obes*, 15 (2022) 209.

8 Mousavi SR, Jafari M, Rezaei S, Agha-alinejad H & Sobhani V, Evaluation of the effects of different intensities of forced running wheel exercise on oxidative stress biomarkers in muscle, liver and serum of untrained rats. *Lab Animal*, 49 (2020) 119.

9 Baek KW, Gim JA & Park JJ, Regular moderate aerobic exercise improves high-fat diet-induced nonalcoholic fatty liver disease via monoacylglycerol O-acyltransferase 1 pathway suppression. *J Sport Health Sci*, 9 (2020) 472.

10 Henriquez-Olguin C, Renani LB, Arab-Ceschia L, Raun SH, Bhatia A, Li Z, Knudsen JR, Holmdahl R & Jensen TE, Adaptations to high-intensity interval training in skeletal muscle require NADPH oxidase 2. *Redox Biol*, 24 (2019) 101188.

11 Maharjan BR, Martinez-Huenchullan SF, Mclennan SV, Twigg SM & Williams PF, Exercise induces favorable metabolic changes in white adipose tissue preventing high-fat diet obesity. *Physiol Rep*, 9 (2021) e14929.

12 Haghshenas R, Jafari M, Ravasi A, Kordi M, Gilani N, Shariatzadeh M, Hedayati M & Rahimi M, The effect of eight weeks endurance training and high-fat diet on appetite-regulating hormones in rat plasma. *Iranian J Basic Med Sci*, 17 (2014) 237.

13 Hariri N & Thibault L, High-fat diet-induced obesity in animal models. *Nutr Res*, 23 (2010) 270.

14 Eshrati R, Jafari M, Gudarzi S, Nazari A, Samizadeh E & Ghafourian Hesami M, Comparison of ameliorative effects of Taraxacum syriacum and N-acetylcysteine against acetaminophen-induced oxidative stress in rat liver and kidney. *J Biochem*, 169 (2021) 337.

15 Tietze F, Enzymic method for quantitative determination of nanogram amounts of total and oxidized glutathione: applications to mammalian blood and other tissues. *Anal Biochem*, 27 (1969) 502.

16 Bradford MM, A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Anal Biochem*, 72 (1976) 248.

17 Friedewald WT, Levy RI & Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem*, 18 (1972) 499.

18 Kruk J & Duchnik E, Oxidative stress and skin diseases: possible role of physical activity. *Asian Pac J Cancer Prev*, 15 (2014) 561.

19 Fang J & Tang M. Exercise improves high fat diet-impaired vascular function. *Biomed Rep*, 7 (2017) 337.

20 Ansari JA, Bhandari U, Pillai K & Haque S. Effect of rosuvastatin on obesity-induced cardiac oxidative stress in Wistar rats—A preliminary study. *Indian J Exp Biol*, 50 (2012) 216.

21 Huang WC, Xu JW, Li S, Ng XE &Tung YT, Effects of exercise on high-fat diet–induced non-alcoholic fatty liver disease and lipid metabolism in ApoE knockout mice. *Nutr Metab*, 19 (2022) 1.

22 Kumar P, Bhandari U & Jamadagni S, Fenugreek seed extract inhibit fat accumulation and ameliorates dyslipidemia in high fat diet-induced obese rats. *Biomed Res Int*, 2014 (2014) 606021.

23 Chaolu H, Asakawa A, Ushikai M, Li YX, Cheng KC, Li JB, Zoshiki T, Terashi M, Tanaka C & Atsuchi K, Effect of exercise and high-fat diet on plasma adiponectin and nesfatin levels in mice. *Exp Ther Med*, 2 (2011) 369.

24 Celik H, Kilic T, Kaplan DS, Eren MA, Erel O, Karakilcik AZ & Bagci C, The effect of newly initiated exercise training on dynamic thiol/disulphide homeostasis in sedentary obese adults. *An Acad Bras Cienc*, 91 (2019) e20180930.

25 Chakraborty R & Sen S, Nephroprotective activity of Pisonia aculeata L. leaf extract against cisplatin induced nephrotoxicity and renal dysfunction in experimental rodents. *Indian J Exp Biol*, 58 (2020) 770.

26 Madhavan S, Rajkumar J & Dorai S, Evaluation of polyherbal formulation against isoniazid and rifampicin induced oxidative stress in rat kidney. *Indian J Tradit Know*, 20 (2021) 671.

27 Lasker S, Rahman MM, Parvez F, Zamila M, Miah P, Nahar K, Kabir F, Sharmin SB, Subhan N & Ahsan GU, High-fat diet-induced metabolic syndrome and oxidative stress in obese rats are ameliorated by yogurt supplementation. *Sci Rep*, 9 (2019) 1.

28 Cavaliere G, Viggiano E, Trinchese G, De Filippo C, Messina A, Monda V, Valenzano A, Cincione RI, Zammit C & Cimmino F, Long feeding high-fat diet induces hypothalamic oxidative stress and inflammation, and prolonged hypothalamic AMPK activation in rat animal model. *Front Physiol*, 9 (2018) 818.

29 Vezzoli A, Pugliese L, Marzorati M, Serpiello FR, La Torre A & Porcelli S, Time-course changes of oxidative stress response to high-intensity discontinuous training versus moderate-intensity continuous training in masters runners. *PLoS ONE*, 9 (2014) e87506.

30 Berzosa C, Cebrian I, Fuentes-Broto L, Gomez-Trullen E, Piedrafita E, Martinez-Ballarin E, Lopez-Pingarron L, Reiter R & Garcla J, Acute exercise increases plasma total antioxidant status and antioxidant enzyme activities in untrained men. *J Biomed Biotechnol*, 2011 (2011) 540458.

31 Saritas N, Uyanik F, Hamurcu Z & Coksevim B, Effects of acute twelve minute run test on oxidative stress and antioxidant enzyme activities. *Afr J Pharm Pharmacol*, 5 (2011) 1218.

32 Kazmi S, Abbas K, Shah S, Nisar U, Arif M, Ahmad R, Pavase T & Soomro M, Partial characterization of glutathione S-transferase (GST) isolated from hepatocytes of *Cyprinus carpio* from river Ravi Punjab, Pakistan. *Indian J Geo-Mar Sci*, 49 (2020) 1017.

33 Vukovic R, Blazetic S, Orsolic I, Heffer M, G Vari S, Gajdos M, Krivosikova Z, Kramarova P, Kebis A & Has-Schon E, Impact of ovariectomy, high fat diet, and lifestyle modifications on oxidative/antioxidative status in the rat liver. *Croat Med J*, 55 (2014) 218.

34 Tahmasebi K, Jafari M, Heydari J, Asgari A, Salehi M, Khazaie S & Abedini MS, Tissues toxicity attenuation by vitamin E on oxidative damage induced by diazinon. *J Environ Anal Health Toxicol*, 37 (2022) e2022036.

35 Ye Y, Lin H, Wan M, Qiu P, Xia R, He J, Tao J, Chen L & Zheng G, The effects of aerobic exercise on oxidative stress in older adults: A systematic review and meta-analysis. *Front Physiol*, 2021 (2021) 701151.

36 Yol Y, Turgay F, Yigittürk O, Asıkovalı S & Durmaz B, The effects of regular aerobic exercise training on blood nitric oxide levels and oxidized LDL and the role of eNOS intron 4a/b polymorphism. *Biochim Biophys Acta Mol Basis Dis*, 1866 (2020) 165913.

37 Upadhyay SD, Ahmad Y, Kohli S & Sharma R, Synergistic effect of folic acid and galantamine against experimentally induced oxidative stress in IMR 32 cells. *Indian J Exp Biol*, 60 (2022) 286.

38 Rana A, Kumar NR & Kaur J, Therapeutic effect of propolis on Staphylococcus aureus induced oxidative stress in spleen of Balb/c mice: A biochemical and histopathological study. *Indian J Nat Prod Resour*, 13 (2022) 346.

39 Chuffa LgdA & Seiva FRF, Combined effects of age and diet-induced obesity on biochemical parameters and cardiac energy metabolism in rats. *Indian J Biochem Biophys*, 50 (2013) 40.

40 Motshakeri M, Goh YM & Ebrahimi M, Metabolic effects of high sucrose and saturated oil feeding on insulin resistance in Sprague-Dawley rats. *Indian J Exp Biol*, 53 (2015) 264.

41 Hsu YJ, Chiu CC, Lee MC & Huang WC, Combination of treadmill aerobic exercise with bifidobacterium longum OLP-01 supplementation for treatment of high-fat diet-induced obese Murine model. *Obes Facts*, 14 (2021) 306.

42 Zhao S, Zhong J, Sun C & Zhang J, Effects of aerobic exercise on TC, HDL-C, LDL-C and TG in patients with hyperlipidemia: A protocol of systematic review and meta-analysis. *Medicine*, 100 (2021) e25103.

43 Salomao R, Neto IVdS, Ramos GV, Tibana RA, Durigan JQ, Pereira GB, Franco OL, Royer C, Neves FdAR & Carvalho ACAd, Nobrega OT, Haddad R, Prestes J & Marqueti RdC, Paternal resistance exercise modulates skeletal muscle remodeling pathways in fathers and male offspring submitted to a high-fat diet. *Front Physiol*, 12 (2021) 706128.

44 Ruan L, Li F, Li S, Zhang M, Wang F, Lv X & Liu Q, Effect of different exercise intensities on hepatocyte apoptosis in HFD-induced NAFLD in rats: the possible role of endoplasmic reticulum stress through the regulation of the IRE1/JNK and eIF2α/CHOP signal pathways. *Oxid Med Cell Longev*, 2021 (2021) 6378568