

Adaptation to the levels of MDA and SOD Enzyme Activity of MICT and HIIT Exercise on Wistar

Samsul Mu'arif

Post-graduate Program
Universitas Negeri Yogyakarta
Yogyakarta, Indonesia
samsul.muarif2016@student.uny.ac.id

Mr. Widiyanto

Post Graduated Program
Universitas Negeri Yogyakarta
Yogyakarta, Indonesia
Widi@uny.ac.id

Abstract—The purpose of this study is to reveal the influence of moderate intensity continuous training (MICT) and high-intensity interval training (HIIT) against the levels of malondialdehyde (MDA) and superoxide dismutase enzyme activity (SOD) in blood circulation. The sample in this study used 21 male Wistar rat tail based on criteria for inclusion and was divided into three treatment groups (control, MICT, HIIT). MICT method applied with exercise intensity 60-80% baseline maximum ability, whereas the method of exercise HIIT applied with 100% intensity baseline maximum ability. Exercise interventions in this study provided for 6 weeks with a frequency of four times per week. MDA levels have known data collection based on the reading of the spectrophotometry of blood serum. SOD enzyme activity is known by observation rate of inhibition of the reduction of ferisitokrom c by superoxide anion from xanthine/xanthine oxidase. The results showed that exercise HIIT and influence the MICT method changes the levels of MDA (Sig. < 0.05), but no effect on enzyme activity of SOD (Sig. > 0.05). It shows, MICT method considered more safe and effective in improving metabolic status based on the trend of changes in average levels of MDA and SOD enzyme activity.

key words—MDA, SOD, MICT, HIIT.

I. INTRODUCTION

Passive lifestyle and nutritional intake against the inefficient provision of output energy impact on overweight and obesity [4, 8, 17]. The Lancet journal published in national geographic (2014) mentioned that based on the survey results number of individuals known to obesity in the world reached 875 million in 1980 have elevated into a 2.1 billion in 2013 [25]. Health research database (RISKESDAS) also mentioned that the prevalence of obesity in Indonesia is still experiencing an increase each year and tends to occur in the population with age > 18 years. Percentage of population males with 7.8% in 2007 increased to 19.7% in 2013, the percentage of women with 15.5% in 2010 increased to 32.9% in 2013, while the overall increase in numbers of all ages and genders during the year 2010-2013 shows the percentage of 18.8% increased to 26.6% [16].

Obesity can trigger an increase in the degree of oxidative stress due to adipose tissue mainly on visceral compartment capable of unleashing biological active molecules in the form of various adipocytokines (adipokine) which is on a mechanism for performance generates reactive oxygen species (ROS) [10,

18]. Other opinions also delivered by Manna, et al., that the obese are able to induce oxidative stress due to high formation of superoxide (O₂^{•-}) through various biochemical mechanism in the form of adenine dinucleotide nicotinamide phosphate (NADPH oxidases), oxidative phosphorylation, activation of protein kinase C (PK-C), activation of the polyol pathway and hexosamine [12]. While according to Birben, et al., oxidative stress on obesity next able to flourish and triggers the onset of various pathological problems such as cancer, neurological disorders, hypertension, atherosclerosis, ischemia/perfusion, diabetes mellitus, idiopathic pulmonary fibrosis, and asthma [2].

The sport with the right dosage, measurable, and regular are overweight and obesity handling strategy through a nonpharmacological approach [7, 19, 20, 21]. An exercise program to decrease the levels of body fat according to Mikami, et al., can be done by the method of moderate intensity continuous training (MICT) at 60-70% VO₂ peak intensity for 45 minutes in continuous [13]. Yet another opinion delivered by Maillard, et al., that high-intensity interval training (HIIT) [60 x (8 seconds at 77-85% HR_{max}, with 12 minutes active recovery)] has been shown to significantly lower levels of total fat capable, abdominal subcutan, and visceral [11]. These conditions because of the exercise by the method of HIIT responded bodily through the increased activity of the enzyme cytochrome c oxidase (COX) as well as the components of cardiometabolic marked by declining fat content and hormone insulin in blood plasma [15, 5].

Although the exercise with the method more effective than HIIT rated MICT in lowering the levels of body fat, but the biochemical adaptation on the complex free-radical and endogenous antioxidant enzymes activity of the body based on a dose of exercise should still be Note: for the methods of exercise is known to involve high intensity even reached the verge of supramaximal. These problems need attention in particular due to exercises conducted with high intensity can increase the levels of malondialdehyde (MDA), which is the product of the concentration of lipids is at once an indicator of oxidative stress [3]. While the high production of ROS and the degree of oxidative stress subsequent capable aggravate, master, even triggering the failure of endogenous antioxidant enzymes in particular performance superoxide dismutase

(SOD) in the reduction of exposure to free radicals [9, 14, 23]. This underlying condition causes damage to components of the biomolecules of cells and normal tissue mainly on deoxyribonucleic acid (DNA), polyunsaturated fatty acids (PUFA), amino acids, and a variety of proteins active in the body [14, 26]. Based on the background of problems, needed research on biochemical adaptation of body of MICT and HIIT on levels of MDA and SOD enzyme activity.

II. METHOD

A. Animal Research

This research is a true experimental laboratories conducted in in vivo involving animals try to form rat *rattus norvegicus* wistar strain types. The number of samples in the study involves 21 rats that consists of a control group (n = 7), MICT (n = 7), and HIIT (n = 7). Counting the number of sample refers to the "guideline for the care and use mammals in neuroscience and behavioral research".

Incubation place animals try (animal house) in this research have the temperature maintained at 22 ° C, with humidity 50-55%, and do control of light-dark cycle of 12 hours. Inclusion criteria for animal statutes try in this research include the rat *Rattus norvegicus* type strain wistar males, age 2-3 months, untrained, overweight, healthy and are not defects. The use of animals try to the criteria of inclusion in this research have been awarded the legality of the Commission of ethics of the research University of Brawijaya Malang with numbers: 900-KEP-UB that have passed and declared to be "ELIGIBLE to CONDUCT".

B. Exercise Protocol

Implementation of the exercise protocol against animals was conducted using a special treadmill for mice under the brand CIS3 IDEAS industries of electronic & software. The MICT method exercise was applied with a maximum intensity of 50-60 % baseline capability with an initial speed of 40 m / min, whereas the HIIT method exercise was applied with an intensity of 100% maximum baseline with an initial speed of 40 m / min.

Progress increased load exercise performed with the increased speed of the treadmill of 1 m/min for HIIT, and 0.6 m/min for MICT is applied in each of the first exercises start the 2nd week. The total duration of the exercise HIIT done for 15 minutes (9 minutes "on" and "off" 6 minutes) intervals. While the MICT performed for 30 minutes in continuous in each practice session. The exercise intervention in this study was given for 6 weeks with the frequency of 4 times a week.

C. Blood Sampling and Laboratory Analysis

Blood sampling is done by taking blood from the heart of mice by as much as 5 ccs. To avoid coagulation and damage, blood samples included on anticoagulant tubes ethylenediamine tetraacetate (EDTA) purple. Separation of serum and blood plasma is carried out by centrifuge at 2000 rpm rotation speed for 10 minutes.

Serum MDA levels are known to be based on the results of serum reaction with TCA 100%, Na Thio 1%, and HCl 1N, which is then incubated and observed results using spectrophotometry.

The serum of SOD enzyme activity to known based on the rate of inhibition of the reduction of ferisitokrom c by superoxide anion generated by the xanthine/xanthine oxidase activity of the reaction is read using spectrophotometry with wavelength 500-600 nm.

III. RESULT AND DISCUSSION

TABLE I. MEAN ± SD OF MDA LEVEL AND SOD ACTIVITY

Group	MDA level	SOD activity
Control	146.45 ± 41.45	6.90 ± 0.81
MICT	88.83 ± 3.04	9.33 ± 3.28
HIIT	184.07 ± 6.19	6.98 ± 1.53

^a. MDA level (ng/ml)
^b. SOD activity (Unit/ml)

TABLE II. ANOVA OF MDA LEVEL

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	26.679	2	13.339	2.913	.080
Within Groups	82.435	18	4.580		
Total	109.113	20			

TABLE III. ANOVA OF SOD ACTIVITY

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	32213.968	2	16106.984	27.360	.000
Within Groups	10596.887	18	588.716		
Total	42810.856	20			

TABLE IV. TUKEY-HSD OF MDA LEVEL

Dependent Variable: MDA Level					
Test	(I) Treatment group	(J) Treatment group	Mean Difference (I-J)	Std. Error	Sig.
Tukey HSD	Control	MICT	57.62000*	12.96937	.001
		HIIT	-37.62000*	12.96937	.025
	MICT	Control	-57.62000*	12.96937	.001
		HIIT	-95.24000*	12.96937	.000
	HIIT	Control	37.62000*	12.96937	.025
		MICT	95.24000*	12.96937	.000

*. The mean difference is significant at the 0.05 level.

A. The Effect of MICT on MDA Level and SOD Activity

Based on the results of the anova test (TABLE 4) show that the MICT effect significantly changes the levels of MDA (Sig. < α, with α = 0.05). The results of this study support previous research conducted by Narasimhan & Rajasekaran, the findings of such research that high ROS on exercise that includes the concept of endurance training method as on MICT, turns Note that the increased production of ROS low to moderate levels of not always cause loss and damage to the body, for the improvement of the conditions of the ROS also

functioned as up-regulator of activation mechanism of antioxidants in detoxification system [22]. It is also proven in this study showing that the average value of the activity of the enzymes SOD on higher MICT method when compared with the control group and HIIT (TABLE 1). Earlier findings about mechanisms and increased MDA levels decrease enzyme activity of SOD on exercise moderate has been described by Gounder, et al., that the long-term impact on the molecular level of moderate exercise in continuous causes activation of the signaling oxidizing nuclear erythroid p-45 related factor-2 (Nrf-2) [24], as a regulator of transcription factor genes contain various antioxidant response element (ARE) to establish homeostasis with radical formation against reactive [22]. In this research, a variety of conditions such as one of the causes of the decline in the levels of MDA significantly followed by an increase in the average activity of SOD enzymes.

B. The Effect of HIIT on MDA Level and SOD Activity

The results of the ANOVA test (TABLE 4) show that HIIT effect significantly to increased levels of MDA (Sig. < α , with $\alpha = 0.05$), which was not followed by an increase in average enough SOD enzyme activity means (TABLE 1). The results of this study resemble the previous research conducted by the Goto, with high-intensity exercise intervention for 12 weeks proved to increase the oxidative stress marker in the form of MDA [6]. Findings from such research that high-intensity exercise causes the body is on oxygen deficit condition further enlarge the oxygen consumption of the network of post-exercise, other sources refer to it as excess post-exercise oxygen consumption (EPOC) that could potentially increase the formation of ROS quite rapidly, even Nita & Grzybowski, explaining that in this the high concentration of lipids through the ROS work against PUFA produce derivatives of the pillars in the form of MDA [14]. Other detrimental impacts from this condition also advanced by Aldred, high levels of ROS that will suppress endogenous antioxidant activity [1]. The occurrence of the antioxidant deficiency which was alleged to be one of the factors the causes of the high levels of MDA and SOD enzyme activity rate low in the study.

IV. CONCLUSION

This study found the presence of a significant change in the method of MICT against MDA levels (TABLE 2), but there was no increase in the activity of the SOD enzymes (TABLE 3) and the control group comparison. It shows that the practice is still recommended as one of the metabolic statuses of the repair strategy and controlling deposits of body fat that is safe and effective, because of the low levels of MDA at the MICT than HIIT and control is MICT has indicated that risk against oxidative stress and potential tissue damage due to the work of ROS.

REFERENCES

- [1] Aldred, S. *Oxidative and nitrative changes seen in lipoproteins following exercise*. University of Birmingham. Atherosclerosis, pp. 1-8, 2007.
- [2] Birben, E., Sahiner, U. M., Sackesen, C., et al. Oxidative stress and antioxidant defense. *Journal World Allergy Organization*, pp. 9-19, 2012.
- [3] Cunningham, P., Geary, M., Harper, R., et al. High intensity sprint training reduces lipid peroxidation in fast-twitch skeletal muscle. *Journal of Exercise Physiology*, 8, 6, pp. 18-25, 2005.
- [4] Faghri, P., Stratton, K., & Momeni, K. Nutritional disorders & therapy sedentary lifestyle, obesity, and aging: implication for prevention. *Journal Nutritional Disorders & Therapy*, 5, 1, pp. 1-2, 2015.
- [5] Gibala, M. J., Little, J. P., Essen, M. V., Wilkin, G. P., Burgomaster, K. A., Safdar, A., Raha, S., and Tarnopolsky, M. A. Short-term sprint interval versus traditional endurance training: similar initial adaptations in human skeletal muscle and exercise performance. *The Physiological society*, 575.3, pp. 901-911, 2006.
- [6] Goto, C., Higashi Y, Kimura M, et al. Effect of different intensities of exercise on endothelium-dependent vasodilation in humans: role of endothelium-dependent nitric oxide and oxidative stress. *Circulation*, vol. 108, pp. 530-5 (2003).
- [7] Iguichi, M., Littmann, A.E., Chang, S., et al. Heat stress and cardiovascular, hormonal, and heat shock protein in human. *Journal of Athletic Training*, vol. 47, pp. 184-190, 2012.
- [8] Jung, U. J., & Choi, M. Obesity and its metabolic complications: the role of adipokines and the relationship between obesity, inflammation, insulin resistance, dyslipidemia and nonalcoholic fatty liver disease. *International Journal of Molecular Sciences*, vol. 15, pp. 6184-6223, 2014.
- [9] Lubrano, V. & Balzan, S. Enzymatic antioxidant system in vascular inflammation and coronary artery disease. *World Journal of Experimental Medicine*, vol. 5, pp. 218-224, 2015.
- [10] Marseglia, L., Manti, s., D'Angelo, G., et al. Oxidative stress in obesity: a critical component in human diseases. *International Journal of Molecular Sciences*, vol. 16, pp. 378-400, 2015.
- [11] Maillard, F., Rousset, S., Pereira, B., et al. High intensity interval training reduces abdominal fat mass in postmenopausal women with type 2 diabetes. *Journal of Diabetes & Metabolism*, pp. 1-9, 2016.
- [12] Manna, P., Obesity, oxidative stress, adipose tissue dysfunction, and the associated health risk: causes and therapeutic strategies. *Metabolic Syndrome and Related Disorders*, vol. 13, pp. 423-444, 2015.
- [13] Mikami, M. E., Sato, K., Kurihara, T., & Hasegawa, N. Endurance training-induced increase in circulating irisin levels is associated with reduction of abdominal visceral fat in middle-aged and older adults. *Plos One*, vol. 20, pp. 2-13, 2015.
- [14] Nita, M., & Grzybowski, A. The role of the reactive oxygen species and oxidative stress in the pathologism of the age-related ocular diseases and other pathologies of the anterior and posterior eye segments in adults. *Oxidative Medicine and Cellular Longevity*, pp. 1-23, 2016.
- [15] Racil, G., Coquart, J. B., Elmoutassar, W., et al. Greater effects of high-compared with moderate-intensity interval training on cardio-metabolic variables, blood leptin concentration and ratings of perceived exertion in obese adolescent females. *Biology of Sport*, vol. 33, pp. 145-152, 2016.
- [16] Riset Kesehatan Dasar (Riskesdas). Badan Penelitian dan Pengembangan Kesehatan. *Laporan Nasional*. Jakarta: Badan Litbangkes Depkes, 2013.
- [17] Sahoo, K., Sahoo, B., & Choudhury, A. K. Childhood obesity: causes and Consequences. *Journal of Family Medicine and Primary*, 4, 2, pp. 187-192, 2015.
- [18] Sanchez, A. F., Santillan, E. M., Bautista, M., et al. Inflammation, oxidative stress, and obesity. *International Journal of Molecular Science*, vol. 12, pp. 3117-3132, 2011.
- [19] Sugiharto. *Fisiologi Olahraga: Teori dan Aplikasi Pembinaan Olahraga*. Malang: UM Press, 2014.
- [20] Sword, D. O., & Carolina, S. Exercise as a management strategy for the overweight and obese: where does resistance exercise fit in?. *Strength and Conditioning Journal*, vol. 34, pp. 47-55, 2012.
- [21] Wadden, T. A., Neiberg, R. H., Wing, R. R., et al. Four-year weight losses in the look AHEAD study: factors associated with long-term success. *National Institute of Health*, vol. 19, pp. 1-25, 2011.
- [22] Narasimhan, M., & Rajasekaran, N. S. Exercise , nrf2 and antioxidant signaling in cardiac aging. *Frontiers in Physiology*, vol. 7, pp. 1-8, 2016.
- [23] Winarsi, H., Wijayanti, S. P. M., & Purwanto, A. Aktivitas enzim superoksida dismutase, katalase dan glutation peroksidase wanita penderita sindrom metabolik. *MKB*, vol. 44, 7-12, 2012.

- [24] Gounder, S. S., Kannan, S., Devadoss, D., *et al.* Impaired transcriptional activity of nrf2 in age-related myocardial oxidative stress is reversible by moderate exercise training. *Plos one*, vol. 7, pp. 1–12, 2012.
- [25] [Http://www.nationalgeographic.co.id/berita/2014/06/jumlah-orang-obesitas-di-indonesia-terus-meningkat](http://www.nationalgeographic.co.id/berita/2014/06/jumlah-orang-obesitas-di-indonesia-terus-meningkat) (acces 8 jan 2018).
- [26] Yavari, A., Javadi, M., Mirmiran, P., & Bahadoran, Z. Exercise-induced oxidative stress and dietary antioxidants. *Asian Journal Sports Medicine*, vol. 6, pp. 1-7, 2015.