

The increase of uncoupling protein-1 expression after moderate intensity continuous exercises In obese females

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Abstract

This study aims to prove an increase in the expression of uncoupling protein-1 (UCP-1) after moderate-intensity continuous exercise in obese females. The present study employed a true-experimental with the randomized control group pretest-posttest design. A total of 12 obese females were enrolled in this study and given moderate-intensity continuous exercise. Continuous exercise was done with an intensity of 60-70% HR_{max} for 40 minutes using a treadmill. The Enzyme-Linked Immunosorbent Assay (ELISA) method was used to examine the expression of UCP-1. Data were analyzed using independent samples t-test with statistics packet for social science (SPSS) software version 17. The results obtained mean UCP-1 expression on K₁ between pre-exercise (3.68±0.46) ng/mL, post-exercise (3.73±0.69) ng/mL and (p=0.875). Mean UCP-1 expression on K₂ between pre-exercise (3.64±1.52) ng/mL, post-exercise (6.83±0.64) ng/mL and (p=0.001). Based on the results of the study, it can be concluded that there is an increase in UCP-1 expression between before and after moderate-intensity continuous exercise in obese females. Future studies are suggested to compare the effect of acute exercise with moderate intensity chronic exercise on increasing UCP-1 expression in obese females.

Keywords: UCP-1 expression, Continuous exercise, Moderate-intensity, Obese females

INTRODUCTION

Obesity is an extremely massive health problem in almost all countries (Bhurosy & Jeewon, 2014). Basic Health Research (Riskasdas) reports that there has been an increase in obesity at the age of 18 years since 2007 (10.5%), 2013 (14.8%), and 2018 (21.8%) (Riskasdas, 2018). The increase in overweight and obesity is closely related to dietary behavior, passive lifestyle, lack of movement, lack of physical activity, and physical exercise (Oh et al., 2017). These behavioral factors can lead to an energy imbalance, which has an impact on the activation of white fat tissue (Kim et al., 2018). Thus it causes non-optimal metabolic performance of fatty acid oxidation (Flouris et al., 2017). Therefore, lifestyle modification through regular, measurable, and continuous physical exercise can be used to increase fat oxidation metabolism, and increase brown fat tissue activity (Oh et al., 2017).

Physical exercise is considered a very strategic approach in preventing and reducing the prevalence of overweight and obesity (Rodrigues et al., 2018). This is because physical exercise can increase fat

oxidation and activate brown fat tissue (Purdom et al., 2018) to activate increased expression of uncoupling protein-1 (UCP-1) (Kim & Plutzky, 2016). However, physical exercise also has the potential to disrupt the body's metabolism and increase oxidative stress (Brondani et al., 2012). This can cause impaired fat oxidation and decrease protein expression in the mitochondria membrane, as well as increase white fat deposits in adipose tissue (Huh, 2018). On the other hand, physical exercise with the right intensity can increase fat metabolism, so that it can activate brown fat tissue, which is promoted by the irisin hormone (Jabbour & Iancu, 2017).

Several studies have reported that low-moderate-intensity physical exercise increases fat metabolism, but high-intensity physical exercise tends to decrease fat metabolism (Purdom et al., 2018; Oh et al., 2017). In addition to the intensity factor, the duration of physical exercise is also associated with fat metabolism which can play a role in changing protein composition in mitochondria and causing the efficiency of the oxidation process through the multiplication of H protons across the membrane to electron transport to regulate the synthesis of Adenosine Triphosphate (ATP) and energy transduction (Befroy et al., 2008). Increased fat metabolism is also associated with muscle contraction during physical exercise, thus causing an increase in irisin secretion (Brondani et al., 2012). Increased irisin secretion can stimulate the browning process in white adipose tissue (WAT) by stimulating UCP-1 expression through signaling p38 mitogen-activated protein kinase (p38-MAPK), thereby increasing energy expenditure through heat (thermogenesis) and reducing body fat accumulation (Boström et al., 2012; Perakakis et al., 2017; Fatouros, 2018). Previous research by Zhang et al. (2017) reported that endurance exercise and combined physical exercise using weights for 12 weeks in healthy and pre-diabetic adults had no significant effect on UCP-1 mRNA in subcutaneous White Adipose Tissue (WAT), although UCP1 mRNA increased (1.82 times from baseline). However, a study done by Oh et al. (2017) reported different results that physical exercise with an intensity of 60%-64% VO_{2max} in trained people and intensity of 47%-52% VO_{2max} for

untrained people significantly increased fat oxidation. Based on a systematic review conducted by [Dinas et al. \(2017\)](#), it is reported that moderate-intensity physical exercise performed for 6 weeks did not affect changes in UCP-1 expression. Therefore, how appropriate and effective dose of physical exercise to increase UCP-1 expression in preventing the increasing prevalence of overweight and obesity is still not exploited.

Based on the explanation above, this study was conducted to prove the response of moderate-intensity continuous exercise to increase UCP-1 expression in obese women, with the hypothesis that moderate-intensity continuous exercise increased UCP-1 expression.

METHOD

The method used in this experimental research is the randomized pretest-posttest control group design. A total of 12 obese women were enrolled in the study. Subject of this study were recruited using a consecutive sampling technique. Some criteria were set on the subjects such as aged 19-24 years, body mass index (BMI) > 27.5 kg/m², normal resting heart rate, normal blood pressure, and normal Hb. The subjects were randomly divided into two groups, namely the control group (n=6, K₁) and the moderate-intensity continuous exercise group (n=6, K₂). This research has complied with the provisions of the Health Research Ethics Commission (KEPK), Faculty of Medicine, Brawijaya University, East Java, Indonesia.

Continuous exercise is carried out with an intensity of 60-70% of max heart rate (HR_{max}) for 40 minutes with details of 5 minutes of warming up (50-60% HR_{max}), 30 minutes of continuous core (60-70% HR_{max}) and 5 minutes of cooling down (50-60% HR_{max}) ([Rejeki et al., 2021](#)). Moderate-intensity continuous exercise using a treadmill was performed in the morning. Monitoring heart rate during moderate-intensity continuous exercise using Polar Heart Rate Monitor H10.

Blood samples were taken before and 10 minutes post-exercise in the cubital vein as much as three milliliters ([Daskalopoulou et al., 2014](#)). Blood was centrifuged for fifteen minutes at 3000 rpm ([Tsuchiya et al., 2015](#);

Tsuchiya et al., 2014). Then the serum was separated, stored at -80°C for analysis of UCP-1 expression the next day. UCP-1 expression examination was carried out in the Lab. UB Medical Faculty Physiology uses the ELISA kit method (Catalog number: E-EL-H1661; Elabscience., China) with a detection range of 0.16-10 ng/mL and a sensitivity level of UCP-1 in the kit 0.10 ng/mL.

Measurement of body height (BH) was carried out using a Stadiometer. Body weight (BW) was measured using the Electronic Scale. Body mass index (BMI) was measured by calculating weight (kg) divided by height (m²) (Aktar et al., 2017; Nimptsch et al., 2019). Blood pressure was checked using an OMRON HEM-7130 L digital blood pressure meter. Hb was measured using Mission Hemoglobin (Mission® Hb Test Strips, San Diego, USA).

Statistical analysis using SPSS software version 17. Shapiro-Wilk was used to test for normality, while the Independent Samples T-Test was used to test differences between groups. All data are shown with mean ± Standard Deviation (SD). All statistical analyzes used a significant level (p<0.05).

RESULTS

The results of descriptive analysis and independent samples t-test on anthropometric research subjects: age, BH, BW, BMI, Hb, SBP, and DBP are presented in Table 1.

Table 1. Results of the different bioanthropometric tests of subjects

Variable	Group				P-Value
	K ₁ (n=6)		K ₂ (n=6)		
	mean	SD	mean	SD	
Age (years)	20.67	1.03	20.50	1.05	0.787
BH (m)	1.59	0.05	1.58	0.06	0.835
BW (kg)	75.23	6.74	74.10	7.47	0.788
BMI (kg/m ²)	29.85	1.60	29.55	0.99	0.705
Hb (g/dL)	15.32	1.99	15.07	0.90	0.785

SBP (mmHg)	120.00	8.94	120.00	8.94	1.000
DBP (mmHg)	78.33	7.53	80.00	8.94	0.734

Descript⁷n: K₁: control group; K₂: moderate intensity continuous exercise group; BH: body height; BW: body weight; BMI: body mass index; Hb: hemoglobin; SB¹⁹: systolic blood pressure; DBP: diastolic blood pressure. p-Value was obtained by using the Independent Samples T-Test to compare bioanthropometric subjects between K₁ and K₂.

Based on ²²Table 1, there is no significant difference in bioanthropometric research subjects between K₁ and K₂ (³ $p > 0.05$). The results of the different UCP-1 expression tests between pre-exercise and post-exercise K₁ and K₂ are presented in Figure 1.

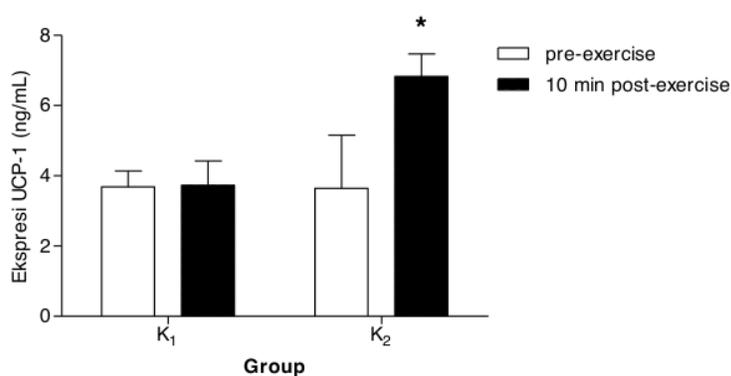


Figure 1. Average expression of UCP-1 in each group
*Significantly different from pre-exercise in K₂ ($p < 0.05$).

Based on Figure 1, it can be seen that the average expression of UCP-1 10 minutes post-exercise was higher than that of pre-exercise ²in each group. Based on the results of the analysis, there was an insignificant difference in the mean UCP-1 expression ¹between pre-exercise and 10 minutes post-exercise at K₁ ($p > 0.05$), while UCP-1 expression was between ¹pre-exercise and 10 minutes post-exercise ³at K₂ showed a significant difference ($p < 0.05$). The results of the analysis of the mean expression of UCP-1 pre-exercise (K₁*K₂) and 10 minutes post-exercise (K₁*K₂) can be seen in Figure 2.

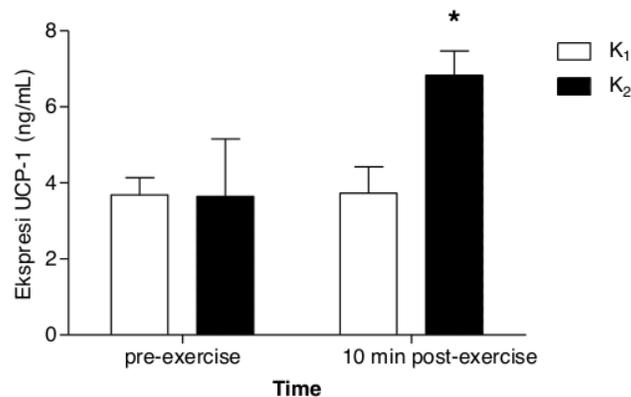


Figure 2. Expression of UCP-1 pre-exercise (K₁*K₂) and 10 minutes post-exercise (K₁*K₂)
*Significantly different from K₁ (p<0.05).

Figure 2 shows that the mean expression of UCP-1 pre-exercise tends to be the same, while the mean expression of UCP-1 10 minutes post-exercise in K₂ is higher than in K₁. The results of the analysis showed that there was no significant difference in pre-exercise UCP-1 expression between K₁ and K₂ (p>0.05), while the mean post-exercise UCP-1 expression between K₂ and K₁ showed a significant difference (p<0.05).

DISCUSSION

The analysis indicates that bioanthropometric subjects: age, BH, BW, BMI, Hb, and blood pressure between K₁ (control group) and K₂ (moderate intensity continuous exercise group) did not show significant differences (p>0.05). It is reported that there were insignificant differences in the characteristics of study subjects, such as BH, BW, BMI, hip circumference, waist circumference, and the ratio of hip and waist circumference between groups (p>0.05). Research by Brandao et al. (2019) also reported that there were no differences in the characteristics of research subjects, such as body weight, body mass index, fat mass and free fat mass between groups (p>0.05). Thus, both groups were at the same starting point before the moderate intensity continuous exercise intervention.

This study uses obese female subjects because women have a higher risk of obesity than men (Mitchell & Shaw, 2015). Based on the

prevalence rate, women have a higher obesity prevalence rate of 15% compared to men at 11% (WHO, 2016). The prevalence rate of obesity in Indonesia in women aged > 18 years is estimated to be 29.3%, an increase of 15.4% from 2013 (13.9%) and a decrease of 3.6% from 2013 (32.9%) (Risikesdas, 2018). In addition, women are also more prone to obesity. This is due to hormonal factors where women's body composition has more fat mass than men. In general, ¹³women have a higher percentage of body fat ²³than men, which is 26.9%, while the percentage of body fat in men is 14.7%. In women, excess fat is located in the chest, abdomen, and upper body (Rahmat & Raharjo, 2018).

The method applied in classifying obesity in this study is the anthropometric method (Sudargo et al., 2018). Anthropometric methods are commonly applied in various studies to determine body composition and identify obesity (Nuttall, 2015). BMI is one of the anthropometric methods that is often used in classifying obesity. In general, BMI is recommended by The Centers for Disease Control and Prevention to determine weight status and classify it simply. However, BMI has a weakness in the level of accuracy because BMI cannot describe the composition of free fat mass and fat mass (Akpınar et al., 2007; Chooi et al., 2019). In addition, BMI ¹⁰also cannot distinguish between fat mass and muscle mass (Nimptsch et al., 2019).

The findings of this study showed that the mean expression of UCP-1 pre-exercise in each group had no difference ($p > 0.05$), while the expression of UCP-1 ¹⁶10 minutes post-exercise in K₂ (moderate intensity continuous exercise group) was higher than in K₁ (control group). Based on the results of the different test, there was a significant difference in the average increase in UCP-1 expression ($p < 0.05$). Reisi et al. (2016) concluded that the response to moderate-intensity continuous exercise significantly increased serum UCP-1 expression. Likewise, previous research by Rui et al. (2017) concluded that continuous exercise significantly increased UCP-1 expression. Dewal and Stanford (2019) also reported that exercise increased UCP-1 expression and increased ¹⁷sympathetic nerve excitability. The results also showed an increase in UCP-

1 expression in the moderate intensity continuous exercise group. This proves that exercise is a thermogenic activity, increasing muscle temperature and core temperature, which will stimulate an increase in sympathetic nervous system response, to increase browning so that UCP-1 expression increases (Aldiss et al., 2018).

The increase in UCP-1 expression in moderate-intensity continuous exercise may also be influenced by increased sympathetic nerve activity. This is because the intensity of exercise increases sympathetic nerve activity and increases catecholamine secretion (Sanchez-Delgado et al., 2015). Increased catecholamine secretion will be captured by adrenergic receptors, so there is an increase in cyclic adenosine monophosphate (cAMP) then activates PKA and causes increased lipolysis, thereby increasing UCP-1 (Rodrigues et al., 2018). Furthermore, UCP-1 induces non-shivering thermogenesis for increased energy consumption, with increased Brown Adipose Tissue (BAT) activity (Kim et al., 2018). Sympathetic nerve stimulation is not only responsible for the cardiorespiratory hemodynamic performance, but is also responsible for body metabolism by activating BAT stimulation.⁹ This is because the sympathetic nervous system stimulates BAT via noradrenergic nerve endings, promoting nonshivering thermogenesis. Noradrenaline binds to beta-3 (β_3), alpha-1 (α_1) and 2-adrenergic receptors, thus this causes an increase in signal transduction that can interfere with lipolysis, thermogenesis, apoptosis, and gene expression. In this way, activation of 3 receptors in BAT increases metabolism, leading to increased activation of UCP-1 (de Queiroz et al., 2012; Brondani et al., 2012). Limitations in this study include 1) the size of the subject used is small, 2) only one parameter measurement (UCP-1), so evidence of fat browning markers is still lacking, and 3) the intervention carried out is acute exercise.

CONCLUSION

Overall, it can be concluded that giving a moderate-intensity continuous exercise intervention for 40 minutes/exercise session increased UCP-1 expression in obese female. Future studies are suggested to

² compare the effect of acute exercise with moderate intensity chronic exercise on increasing UCP-1 expression in obese women. In addition, further research is also needed by measuring other variables, such as PGC-1 α , FNDC-5, irisin, PRDM16, and IL-6. These variables have a strong correlation with the increased expression of UCP-1.

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