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ORIGINAL ARTICLE

The Effect of UCP2 45bp Insersi/Delesi Genetic Variation on the Body Composition Improvement of Woman with Obesity in Continuous Training and High-Intensity Interval Training (Randomized Controlled Trial Study)

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ABSTRACT

Background. Continuous Training (CT) is often considered an effective way to reduce obesity. However, in recent year, a popular protocol called High-Intensity Interval Training (HIIT) shows up as alternatives to CT. There is other factor affecting obesity named UCP2 45-bp Insersion/Deletion genetic marker. **Objectives.** This research aims to determine the effect of the UCP2 45-bp I/D gene as a genetic marker in response to obese training (CT and HIIT). **Methods.** This study was a randomized controlled trial (RCT) in two cycling training groups (CT and HIIT). Purposive sampling method was used to collect 28 women with obesity (BMI≥25 kg/m^2). Random allocation into two groups using the block randomization method. Exercise training interventions conducted for 12 weeks, with a frequency of 3 times per week. **Results.** Body composition data (body weight, BMI and Body Fat Percentage) before and after the intervention were analyzed with the Dependent T-Test and found that both the CT and HIIT groups had significant improvements in body composition ($P<0.05$). ANCOVA Test analyzed the effect of training type and UCP2 45-bp I/D variance on body composition. There was no effect of training type and genetic variation on body weight improvement ($P=0.145$), body mass index improvement ($P=0.153$), and body fat improvement ($P=0.159$). **Conclusion.** The reasearch conclude that both Continuous Training and High-Intensity Interval Training can improve the body composition of obese patients equally well. There was no effect of UCP2 45-bp I/D variance on the response to training in woman with obesity.

KEYWORDS: ***Body Composition; Continuous Training (CT); High-Intensity Interval Training (HIIT); Obesity; UCP2 45-bp Insersion/Deletion***

INTRODUCTION

Obesity defines as Body Mass Index (BMI) ≥25 kg/m2 for the Asia Pacific population (World Health Organization, 2018). Obesity related with higher mortality rate due to complications such as Diabetes Mellitus (DM) (1). The increase in BMI of 1 kg/m^2 will increase the risk of DM by 20%. The life expectancy of obese patients is reduced by about 5-20 years (1). Data by WHO shows that 39% of the world's population is obese (2). Inactive lifestyle is one factor that cause obesity. Physical activity is an effective way to reduce the incidence of obesity. Training is a structured and sustained physical activity that involves repetitive movements and aims to improve fitness and physical health (3). One of the training is Moderate Intensity Continuous Training (MICT) or Continuous Training (CT). Continuous training is the traditional methods of increasing physical activity. However, the effectiveness of CT relies on long duration sessions (4).This exercise involves a long enough duration at a moderate intensity that performed continuously

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without rest (5). Therefore, other time-efficient training modalities for obese people should be explored.

There comes various physical training models as alternatives to CT. One of them is High-Intensity Interval Training (HIIT), which is a combination of high-intensity and low-intensity physical training (6, 7). Recent studies have shown that HIIT effectively improves body composition (8, 9) and cardiopulmonary fitness (10-12).

Exercise therapy does not always work for obese people. Genetic marker is other factor affecting obesity by 40-90% (13-15). The polygenic obesity approach that studies gene interactions has found the uncoupling protein (UCP) coding gene (16). UCP is associated with proton leakage during oxidative phosphorylation in mitochondria. There are three polymorphisms of the UCP2 gene, one is in the promoter region (-866G/A, rs659366), one missense variant in exon 4 (rs660339, Ala55Val, C/T), the other is 45-basepair insertion/deletion (45 -bp I / D) is in the 3' untranslated region (UTR) exon 8 (17). The UCP2 45-bp I/D variation can cause a decrease in energy expenditure, reduce fatty acid oxidation, affect mRNA transcription and stabilization, thereby increasing the risk of obesity (18). A study in Malaysia revealed the correlation of UCP2 45-basepairs (bp) I/D gene polymorphism and obesity in women. The UCP2 45-bp minor allele frequency (I/D) was 14%. Previous studies reported that the allele I of UCP2 45-bp was more at risk for developing obesity and BMI than the D allele (19). Previous research found that genetic variation can affect physical fitness and body response (20, 21). Therefore, researchers interested in knowing the effect of the UCP2 45-bp (I/D) gene as a genetic marker on the response to exercise training (CT and HIIT) in obesity. This research hypothesises that there is an effect of UCP2 45-bp I/D gene variation on the improvement of body composition in response to training.

MATERIALS AND METHODS

Research Methods. A Randomized Controlled Trials (RCT) study was conducted. There were two groups of cycling training interventions (HIIT and CT) using the block randomization method.

Research Participants. Purposive sampling method was used to collect this study population. The sample size required 28 women with obesity which divided into two groups of 14. The inclusion criteria for study subjects were 18-34 years old, had a history of inactive lifestyle for the last six months, were declared healthy and fit for training, and were willing to be participant. Participant who taking weight-loss drugs/supplements were excluded. The exclusion criteria were that they could not finished the intervention according to the predetermined program.

Measurement of Research Variables. Measurement of Body Composition Body Mass Index (BMI) uses data on body weight (BW) and height (H) and the formula for body weight (kg)/height (m^2) in units of kg/m^2. Measurement of Body Fat Percentage using the Bioelectric Impedance Analysis (BIA) method in Body Composition Monitors Tanita®, in percent (%). The examination of the genetic variation of UCP2-45 bp I/D used the Polymerase Chain Reaction-Restriction Fragment Length Polymorphism (PCR-RFLP) method.

Training Protocol. Exercise training interventions conducted for 12 weeks, with a frequency of 3 times per week and based on the COVID-19 prevention health protocol. The type of training is cycling. The control group receives continuous training (CT). Continuous training is an exercise that performed with the same intensity throughout the program, without any recovery period. The intervention performed with an exercise regimen at an intensity of 60 - 80% maximum Heart Rate.

The intervention group is a group that receives High-Intensity Interval Training. High-Intensity Interval training is an intermittent period of exercise separated by a recovery period. The intervention was performed with an exercise regimen of 4 x 4 minutes intervals (85 - 95% maximum HR) and 3 x 3 minutes of rest/recovery (60% maximum Heart Rate) (22, 23).

Data Analysis. Body composition data (body weight, BMI and Body Fat Percent) before and after the intervention were analyzed using the Dependent T-Test as bivariate analysis. To evaluate the effect of training type and UCP2 45-bp (I/D) genetic variation on body composition, we use ANCOVA Test as multivariate analysis. The test results considered significantly different if $P<0.05$.

Research Ethics. The study has received an Ethics Committee Approval from the Medical and Health Research Ethics Committee (MHREC)

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RESULTS

Table 1 showes the results of Body Weight, BMI and Body Fat results before and after intervention.

Table 1. Body Composition of Participants

Variabel	N	Mean ± SD	Median	(Min – Max)	95% CI
Continuous Training					
Body Weight pre-intervention	14	75.86±9,92	77.95	(55.70 – 91.60)	(70.13 – 81.59)
Body Weight post-intervention	14	74.31±9.52	75.60	(55.90 – 89.60)	(68.82 – 79.81)
BMI pre-intervention	14	30.18±3.19	29.65	(25.40 – 37.60)	(28.34 – 32.02)
BMI post-intervention	14	29.5±3.05	29.0	(25.50 – 36.80)	(27.82 – 3.35)
Body Fat pre-intervention	14	41.23±3.56	40.90	(35.60 – 49.40)	(39.17 – 42.29)
Body Fat post-intervention	14	40.45±3,65	40.05	(35.10 – 48.80)	(38.34 – 42.56)
High-Intensity Interval Training					
Body Weight pre-intervention	14	79.05±7.55	77.80	(69.60 – 93.80)	(74.68 – 83.40)
Body Weight post-intervention	14	76.82±7.25	76.45	(67.50 – 93.20)	(72.63 – 81.01)
BMI pre-intervention	14	31.61±3.24	30.95	(27.70 – 37.10)	(29.73 – 33.48)
BMI post-intervention	14	30.70±3.06	30.25	(27.20 – 36.20)	(28.94 – 32.48)
Body Fat pre-intervention	14	42.69±3.74	42.05	(38.10 – 48.40)	(40.53 – 44.85)
Body Fat post-intervention	14	41.54±3.44	41.0	(37.20 – 46.90)	(39.55 – 43.52)

Table 2. The Effect of Training Intervention on Body Composition

Variable	N	Mean ± SD	P
Continuous Training			
Body weight : pre – intervention	14	75.86±9.92	0.006
Post - intervention	14	74.31±9.52	
BMI: pre-intervention	14	30.18±3.19	0.005
Post-intervention	14	29.5±3.05	
Body Fat: pre-intervention	14	41.23±3.56	0.001
post-intervention	14	40.45±3.65	
High Intensity Interval Training			
Body weight : pre – intervention	14	79.05±7.55	0.004
Post - intervention	14	76.82±7.25	
BMI: pre-intervention	14	31.61±3.24	0.003
Post-intervention	14	30.70±3.06	
Body Fat: pre-intervention	14	42.69±3.74	0.0001
Post-intervention	14	41.54±3.44	

Table 3. The Effect of Training Intervention and UCP2 45-bp (I/D) Genetic Variation on Body Composition

Group	N	Body Weight	Body Mass Index	Body Fat
CT with DD allele gene	6	75.63±0.82	30.19±0.31	41.10±0.32
CT with DI allele gene	8	75.99±0.70	30.33±0.27	41.22±0.28
HIIT with DD allele gene	7	76.28±0.76	30.41±0.29	41.21±0.30
HIIT with DI allele gene	7	74.33±0.79	29.65±0.31	40.44±0.31
ANCOVA Test		P=0.145	P=0.153	P=0.159

Bivariate analysis to determine whether exercise training interventions affect Body Weight, BMI and Body Fat Percent using Dependent T-Test with the results as shown in Table 2. Table 2 showes that both CT and HIIT significantly improved Body Weight, BMI and Body Fat Percentage with $P<0.05$.

The genetic variation of UCP2 45-bp (I/D) expected to have influences on energy expenditure thus affects the development of obesity. A total of 28 study subjects conducted to UCP2 45-bp (I/D) genetic examination, the results were that 13 subjects (46%) had the DD genotype variant and 15 study subjects (54%) had the DI genotype variant.

Table 3 showes the results of multivariate analysis with ANCOVA Test. This test analysed the effect of training type (CT/HIIT) and UCP2 45-bp (I/D) genetic variation on body composition. This table showes no significant difference ($P\geq 0,05$) in the mean of Body Weight, BMI and Body Fat Percentage in four intervention groups. There was no effect of training type and genetic variation on body weight improvement (P=0.145). There was no effect of training type and genetic variation on body mass index improvement (P=0.153). There was no effect of training type and genetic variation on body fat improvement (P=0.159).

DISCUSSION

This study proved that 12 weeks of CT and HIIT interventions can improve body composition by reducing body weight, BMI and body fat percentage of obese women. Previous studies related to CT interventions also proved similar results as Wewege's study, which stated that CT intervention for approximately ten weeks could significantly reduce BMI in overweight and obese subjects (5). King's study showed different results, and there was no decrease in BMI after eight weeks of CT intervention (24). The results difference could be due to differences in the length of CT intervention. Meanwhile, studies related to HIIT interventions, such as the Ouerghi's study, also stated that the HIIT intervention for eight weeks resulted in a significant decrease in BMI value (25). Putra's study also reported similar results, that HIIT interventions could significantly reduce BMI in obese women (26). However, the study conducted by Hughes and Higgins reported different results, there was no significant decrease in BMI values after six weeks of HIIT intervention (27). Difference in HIIT intervention duration is one factor that determines the impact of HIIT on body composition (25).

Body composition improvement on CT happened through proliferation of capilary skeletal muscle, increase of mitochondrial volume, carnitine transferase enzyme that increases fatty acid transportation, and fatty acid binds protein that regulates myocite fatty acid transportation (28). All of these things can reduce the percentage of body fat, which then also reduces BMI. Meanwhile, the mechanism for improving body composition in HIIT occurs through the high intensity of training in HIIT, which stimulates metabolism after physical activity, named Excess Post-exercise Oxygen Consumption (EPOC) (29). EPOC defines as an increase in oxygen consumption during the recovery period after training (30). After a training session, the body's metabolic rate remains high to return the body to a resting state. The process of returning the body to homeostasis involves more elevated levels of fatty acids in the Krebs Cycle, increases influence of Growth Hormone (GH), insulin, ACTH, cortisol, and thyroid hormones, sympathetic activity, and mitochondrial respiration. Therefore, EPOC is associated with an increase in energy expenditure (31). HIIT also causes a decrease in lactate and hydrogen ions which triggers an increase in fat oxidation (29). In HIIT, increased fat oxidation also occurs in the mitochondria through increased mitochondrial volume, lipolysis of triacylglycerol adipose tissue into fatty acids, and the transport of fatty acids into cells, intramuscular lipolysis, and transportation of fatty acids to mitochondria (32).

A study comparing the effect of CT and HIIT interventions showed no significant difference in body composition improvement in the two groups. Both CT and HIIT have an equally good impact on improving Body Weight, BMI and Body Fat Percentage of obese patients. This result was consistent with Wewege's study, which found that MICT and HIIT had the same effect on body fat and waist circumference (5). Ram stated that the effect of HIIT and MICT on body composition did not differ significantly between the groups. Ram used short term (6 weeks) cycling training as the intervention of study (33). A systematic review and meta-analysis by Keating also stated that MICT and HIIT were able to reduce body fat percentage by up to 5% significantly. HIIT could be an alternative to MICT with the same good results, even more, efficient in implementation time (34).

The results on the effect of genetic variation of UCP2 45-bp (I/D) study proved that there was no effect of genetic variation on the response to training either as a whole or in each training group. There was no difference between the DD and DI genotype variants in response to body composition improvement after training intervention. The genetic variation of UCP2 45-bp I/D can cause a decrease in energy expenditure, reduce fatty acid oxidation, affect mRNA transcription and stabilization, thereby increasing the risk of obesity (18). However, the study could not prove the effect of genetic variation on body composition improvement after training intervention.

The limitation of this study was that it was unable to obtain a complete genetic variation of UCP2. From 28 research participants, only two genotype alleles were obtained, namely DD and DI. This study did not find genotype II as UCP2 45 bp I/D variation gene, so it is difficult to conclude the effect of UCP2 gene variation on improving body composition on CT and HIIT interventions. The small number of participants may decrease the power to detect differences between the intervention groups.

Based on the study's limitations, it can be suggested to be able to take more participants and meet the availability of three types of genotypes in the 45 bp I/D UCP2 genetic variation.

CONCLUSION

The research conclude that the 12-week CT and HIIT interventions improved body composition by reducing body weight, BMI and body fat of obese women. HIIT can be an alternative to MICT with the same results, even more efficient in implementation time. There was no effect of UCP2 45-bp I/D genetic variation and the type of training on the body composition improvement.

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APPLICABLE REMARKS

Time-saving High Intensity Interval Training (HIIT) can be used as an alternative to exercise therapy for people with obesity. To see the effect of the 45 bp I/D UCP2 gene requires further research with a larger number of participants and the availability of a more complete 45 bp I/D UCP2 genotype.

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