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Leptin Level Expression in Obese Women During Chronic Physical Exercise

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ABSTRACT
This study aims to analyze the effect of short-term combination training for 4 weeks in reducing leptin levels in obese women. The method used is a true experimental with a randomized pretest-posttest control group design, involving 14 obese female subjects who were divided into two groups: control (K1) and combination training (K2). The combination training consisted of endurance training (treadmill) and resistance with moderate intensity (60-70% 1-RM) which was done 5 times per week for 4 weeks. The results showed a significant decrease in leptin levels in the K2 group compared to K1, which actually showed a slight increase in leptin. The decrease in leptin levels in K2 was associated with a decrease in fat mass and body mass index (BMI), accompanied by an increase in muscle mass. Combination training has a significant effect on reducing leptin through increased energy expenditure and reduced adiposity. Although this study showed positive results, there are still limitations in controlling external factors such as stress levels that may affect leptin levels. Further research is recommended to consider these variables so that the results are more comprehensive.

Keywords: leptin, obesity, combination training, fat mass, women
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Introduction

High leptin levels in obese people are a risk factor for several diseases that contribute to increased premature death (Katsiki et al., 2018). Energy imbalance between unhealthy and excessive diets, lack of physical activity, and sedentary lifestyles cause high fat mass deposits which ultimately become the underlying factor causing high leptin in obese people (Suputra, 2023). Lifestyle changes through regular physical exercise have been shown to be an effective strategy to control the rate of increase in leptin in obesity.

In obese people, although leptin levels increase due to increased fat mass, the hypothalamus does not respond normally to leptin (novel-insights-into journal). This condition is known as leptin resistance which is characterized by hyperleptinemia and decreased sensitivity and failure of the brain to respond to leptin, thereby inducing increased appetite and weight (Rejeki et al., 2023). These high circulating leptin levels in obese individuals may contribute to a low-grade inflammatory background, which makes obese individuals more susceptible to increased risk of developing metabolic diseases such as cardiovascular disease, T2D, as well as degenerative diseases including autoimmune diseases (multiple sclerosis, thyroiditis, rheumatoid arthritis, inflammatory bowel disease and knee arthrosis) and cancer (Pérez-Pérez et al., 2020).

Circulating leptin levels are positively correlated with body fat (Li & Li, 2016). Obese individuals show high levels of leptin expression in adipose tissue and have high circulating leptin levels. Therefore, exercise is needed to reduce fat mass through adipose tissue reduction. Exercise can reduce serum leptin by increasing energy expenditure and decreasing adiposity (Yu et al., 2017).

However, the type (e.g., aerobic and/or resistance training), intensity and duration of exercise, as well as obesity and health status, are factors that may influence how exercise affects chronic inflammation associated with obesity (Lopes et al., 2016). Moderate-intensity aerobic exercise for 6 weeks has been shown to reduce leptin levels in obese women, whereas 70% 1RM resistance exercise for 6 weeks did not significantly reduce leptin levels in obese women (Abedi et al., 2019). Sajad 2013 also stated that short-term resistance training for 8 weeks did not significantly change leptin levels.

A recent study by Oh & Lee, (2023) showed that combined exercise for 8 weeks in a moderate-intensity group (50% VO2max) and a high-intensity group (80% VO2max) resulted in significant decreases in leptin levels. Research conducted by Li et al., (2020) showed that 10 weeks of combination training can reduce leptin levels, but these results vary depending on the duration of training; groups with a duration of 270 minutes and 450 minutes per week showed a significant decrease in leptin levels, while there was no significant change in the group with a duration of 150 minutes per week. In contrast, short-term studies such as those conducted by Rejeki (2024), which used moderate-intensity combination training for 4 weeks with a frequency of three times a week, showed that changes in body composition and adipokine levels could be achieved with a shorter duration. Therefore, the aim of this study was to analyze the effect of short-term combination training for 4 weeks in reducing leptin levels in obese women.

Methodology

The method used in this study was true experimental with the research design used was the randomized pretest-posttest control group design. The study was conducted for 4 weeks using 3 places, namely: 1) Sport Science Laboratory, Faculty of Sport Science, State University of Malang; 2) Atlas Sport Club Malang; and 3) Physiology Laboratory, Faculty of Medicine, Brawijaya University. The subjects in this study used 14 obese women as research subjects selected based on the following criteria: age 20-30 years, BMI 25-36 kg/m^2, blood pressure (systolic 110/120 mmHg and diastolic 70/80 mmHg), resting heart rate 60-80 bpm, oxygen saturation 96-100%, not actively exercising, non-smokers, and non-drinkers. The subjects were divided into two groups, namely the control group (K1; n = 7) and the endurance training group (K2; n = 7).

The combination exercise in this study used endurance and resistance training. Endurance training using a treadmill for 40 minutes. Resistance training using 6 gym equipment with the Cybex and Life Fitness brands which are divided into 2 muscle parts, namely the upper body and lower body. The upper body uses a chest press machine, shoulder press machine, and lat pull down machine, while the lower body uses a leg press machine, leg extension machine and hip abduction machine. The intensity of the exercise in this study used moderate intensity (60-70% 1-RM), which was done as many as 4-6 sets, 10-15 repetitions, with a frequency of 5x / week, for 4 weeks. During the combination exercise, heart rate was monitored using POLAR H10. Furthermore, blood samples were taken from the cubital vein as much as 4 ml during the pretest (before the exercise intervention) and posttest (after 4 weeks of the combination exercise intervention). Blood samples were centrifuged for 15 minutes at 3000 rpm, then the serum was separated and stored at -800C for leptin level analysis the next day using the ELISA method (REF.CAN-L-4260; Diagnostics Biochem Canada (DBC), Inc. London, ON, Canada) with a sensitivity of 0.5 ng/mL and a detection range of 1–100 ng/mL.

Data analysis in this study was carried out after the pretest and posttest were collected. Furthermore, the data that had been obtained were analyzed using the Statistical Package for the Social Science (SPSS) application with version 21.0. The normality test was carried out using the Saphiro-Wilk test with a significant level ($p>0.05$). The difference in leptin levels between the pretest and posttest in each group was analyzed using the paired sample t-test, while the difference in leptin levels between the pretest and posttest groups used the independent sample t-test with a significant level ($p<0.05$).

Result

The result of descriptive analysis, normality, and independent sample t-test of the average characteristics of the ressearch subjects can be seen in the table 1. Based on Table 1, the results of the descriptive analysis show that the differences in characteristics in each group are not significant. The results of the Independent Samples T-Test show that there is no significant difference in the average data on the characteristics of the research subjects in both groups (p>0.05). The results of the analysis of the average leptin levels between the pretest and posttest in both groups can be seen in Tables 2 and 3.

Table 1. Characteristics of Research Subjects in Both Groups

Variable	n	K1		K2		
		Mean	SD	Mean	SD	*P-Value*
Age (year)	7	23.14	1.86	25.57	2.57	0.066
TB (m)	7	1.57	0.068	1.54	0.039	0.336
BB (kg)	7	78.04	6.04	77.10	7.44	0.799
BMI (kg/m^2)	7	31.61	2.91	32.28	2.29	0,641
TDS (mmHg)	7	125.57	4.07	123.57	5.71	0.465
TDD (mmHg)	7	68.71	8.09	73.85	5.36	0.187
DJI (bpm)	7	89.14	13.98	90.28	9.06	0.859
SpO^2 (%)	7	98.00	0.577	98.42	0.534	0.175
ST (°C)	7	35.27	1.17	35.90	0.602	0.233
GDP (mg/dL)	7	98.71	13.17	99.00	3.91	0.957
HB (g/dL)	7	13.51	1.88	14.24	1.37	0.425

Description: K1: Control group; K2: Combined Exercise Group; SBP: Systolic blood pressure; SBP: Diastolic blood pressure; DJI: Resting heart rate; SpO^2: Oxygen saturation; ST: Body temperature; GDP: Fasting blood sugar; HB: Hemoglobin; TB: Height; BB: Weight; BMI: Body mass index.

Table 2. Results of Analysis of Leptin Levels Pretest and Posttest in Each Group

Groups	Observation	n	Mean (ng/mL)	SD	Normality	*p-value*
K1	Pretest	7	678.83	39.22	0.643	0.444
	Posttest	7	690.56	57.16	0.514	
K2	Pretest	7	688.92	101.26	0.649	0.000*
	Posttest	7	572.37	108.81	0.689	

Description: K1: Control group; K2: Combination exercise group. (*) Indicates significant value with pretest (p<0.05).

Based on Table 2, the results of the descriptive analysis show that there is a decrease in the average leptin levels between the pretest and posttest of the K2 group. The results of the Paired Sample T-Test show that there is a significant decrease in the average leptin levels between the pretest and posttest in K2 (p = 0.000; p <0.05), while in the K1 group there is no significant decrease in the average leptin levels between the pretest and posttest (p = 0.444; p> 0.05).

Table 3. Results of Analysis of Leptin Levels Pretest and Posttest in Both Groups

Groups	n	*Pretest*		p-value	*Posttest*		p-*value*
		Mean	SD		Mean	SD	
K1	7	678.83	39.22	0.810	690.56	57.16	0.026*
K2	7	688.92	101.26		572.37	108.81	

Description: K1: Control group; K2: Combination exercise group. (*) Indicates significant value with pretest (p<0.05).

Dicussion

Based on the results of the study, there was a significant change in inflammation between the combination exercise group (K2) compared to the control group (K1) (Table 2). In this study, the decrease in leptin levels only occurred in the combination exercise group, while the control group experienced an increase in leptin levels. The

difference in leptin levels in the two groups was due to the exercise intervention in the K2 group. In K1, there was no decrease in leptin levels because no intervention was given. The results of this study are in line with a study conducted by Lopes et al., (2016) which stated that combined exercise consisting of resistance training (three sets of 6-10 repetitions at 60-70% of 1 RM) followed by 30 minutes of aerobic exercise (walking/running at 50-80% VO2peak), performed three times a week for 12 weeks can reduce leptin levels in obese female adolescents. This decrease in leptin levels is related to reduced body fat and decreased insulin resistance. A long-term study conducted for 1 year by Dâmaso et al., (2014) stated that combined aerobic and resistance training was more effective in reducing leptin levels than aerobic training alone. The study showed the beneficial effects of aerobic training accompanied by resistance training in reducing visceral and subcutaneous fat associated with decreased hyperleptinemia and increased adiponekine concentrations. Rejeki et al., (2023) also stated that combined moderate-intensity training performed three times a week for 4 weeks was more effective in improving body composition and adipokine levels than aerobic and resistance training alone.

The decrease in leptin levels in the combined training group (K2) of this study was also followed by an increase in muscle mass and a decrease in fat mass, so the decrease in leptin levels is thought to be due to a decrease in fat mass which is indicated by a decrease in BMI. This is because leptin is a protein that is mainly secreted by adipose tissue and which circulates in the blood in proportion to adiposity. Leptin concentrations will change along with changes in fat stores, when fat mass decreases (Hayashino et al., 2014). Therefore, leptin is closely related to changes in BMI (Kazmi et al., 2016).

Aerobic exercise is often associated with greater energy expenditure, while endurance training is more associated with maintaining muscle mass that shifts the composition of fat mass. Aerobic exercise induces FA metabolism through oxidation in muscle mitochondria to produce large amounts of ATP. Conditions in aerobic exercise that require high ATP will trigger the activation of the AMPK pathway which functions to conserve ATP by inhibiting the biosynthesis and anabolic pathways (glycogen and protein synthesis), and stimulating the catabolic pathway by increasing glucose transport and fat metabolism. Meanwhile, endurance training that focuses on muscle hypertrophy will induce the release of anti-inflammatory myokines and activate the AMPK and phosphatidylinositol 3-kinase (PI3-kinase) pathways that produce muscle-adipose relationships that underlie fat burning in the body (Rejeki et al., 2023). Combination training provides more beneficial effects, due to the combination of aerobic training (increased energy systems through oxidative metabolism, changes in skeletal muscle fiber types, metabolic capacity, and cardiorespiratory fitness) with resistance training (changes in skeletal muscle mass or fiber diameter and increased muscle strength) (Sigal et al., 2014).

The limitation of this study is that it only focuses on leptin levels, while several other variables that also have the potential to affect leptin levels such as stress levels, are not controlled. For further research, it is recommended that factors such as stress be taken into account or controlled, considering that stress can increase appetite which has the potential to increase leptin levels. By considering these factors, further research will be more comprehensive and able to provide a deeper view of the relationship between combination training, leptin levels, and other factors that influence it.

Conclusions

The study showed that short-term combination training for 4 weeks had a significant effect on reducing leptin levels in obese women compared to the control group that did not receive training intervention. The decrease in leptin levels in the combination training group (K2) was likely related to a decrease in fat mass, which was also followed by an increase in muscle mass and a decrease in fat mass. The effectiveness of this combination training is supported by the findings of previous studies showing the benefits of aerobic and resistance training in reducing leptin levels through increased energy expenditure and decreased adipose tissue. Combination training

provides additional benefits by supporting fat metabolism and maintaining optimal body composition. However, this study has limitations, namely the lack of control for external factors such as stress levels, which also have the potential to affect leptin levels. For further research, it is recommended that these factors be controlled to obtain more accurate and in-depth results regarding the effects of physical exercise on leptin levels in the obese population.

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