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The Effect of Black Garlic Extract as a Curative Therapy on Increased Total Cholesterol in Obesity Model Rats

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Abstract**:** *Obesity is a condition of excess body fat accumulation that often triggers an increase in total cholesterol levels in the blood, thereby increasing the risk of cardiovascular disease. Black garlic, a fermented garlic product, is known to contain bioactive compounds such as S-allylcysteine (SAC) which is effective in lowering cholesterol levels. Objective: To determine the effect of black garlic extract as a preventive therapy in obesity model rats induced by a high-fat and fructose diet. Method: The research method used an experimental design with a pre- and post-test group design approach. A total of 28 male Sprague Dawley rats were divided into four groups: negative control (standard diet AIN 93), positive control (high-fat and fructose diet), and two treatment groups each given black garlic extract at a dose of 1.6 ml/day and 0.8 ml/day. Total cholesterol levels were measured before and after the intervention using the spectrophotometric method. Data analysis used SPSS with the Independent t-test. Results: The results showed that administration of black garlic extract significantly reduced total cholesterol levels in the treatment group compared to the positive control group. A dose of 1.6 ml/day produced a greater cholesterol-lowering effect than a dose of 0.8 ml/day. Conclusion: Black garlic extract was effective in lowering total cholesterol levels in an obese rat model.*
Keywords: black garlic, total cholesterol, obesity

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INTRODUCTION

Obesity is a condition in which there is an accumulation of excess fat in adipose tissue, and is one of the most common health problems (Hales et al., 2020). A general assessment of obesity is done through body weight measurements, but a more accurate method is to use the Body Mass Index (BMI), which indirectly measures the level of body fat or adipose tissue.

Based on the results of research from the 2018 Basic Health Research (Riskesdas), the percentage of obesity in Indonesia in the adult population aged over 18 years with a Body Mass Index (BMI) of more than 27 kg/m2 has increased. In 2013, the percentage reached 14.8%, while in 2018 it increased to 21.8%.(Indonesian Health Research and Development Agency, 2019). This

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condition of obesity can result in increased lipid levels in the body, which has the potential to cause atherosclerosis.(Kawai et al., 2021).

One of the causes of obesity is excessive food consumption. Decreased physical activity, especially accompanied by excessive eating habits, can producean imbalance between calorie intake and expenditure, which ultimately leads to obesity.Being overweight can increase the risk of hypertension, diabetes mellitus, heart disease, dyslipidemia, kidney failure, and inflammatory responses. Dyslipidemia, characterized by elevated levels of total cholesterol, triglycerides, and low-density lipoprotein (LDL), as well as low levels of high-density lipoprotein (HDL), can lead to atherosclerosis and cardiovascular disease. Total cholesterol is one indicator for assessing the risk of cardiovascular disease. Hypercholesterolemia, or elevated total cholesterol levels, often has no symptoms, making it important to conduct preventive checkups and regularly monitor cholesterol levels, especially for individuals at high risk.(Henning, 2021; Jebari-Benslaiman et al., 2022).

Increased cholesterol levels in the body can be triggered by lifestyle changes and unhealthy diets that tend to be high in fat and carbohydrates. Fructose is a type of simple carbohydrate that is a natural sweetener in fruits, honey, and vegetables. A high-fructose diet, especially from processed foods, can cause increased cholesterol levels through the activation of the enzyme hydroxy-methylglutaryl-coenzyme A (HMG-CoA) reductase. High-fat consumption can also trigger an increase in cholesterol of 1.9 mg/dl for every 1% increase in saturated fatty acids. Consuming a high-fat and high-fructose diet simultaneously causes a higher increase in plasma cholesterol levels compared to a high-fat diet alone.(Samir Softic et al., 2017; Susanti et al., 2019)

Elevated blood cholesterol levels can be controlled by using the active ingredient in garlic known as alliin. Alliin is a sulfur-containing chemical compound that gives garlic its distinctive aroma (Ha et al., 2015) The purpose of this research isForexploring the possibility of using Black garlic as an intervention in managing cholesterol levels in obese individuals, so that it is hoped that it can make a significant contribution in efforts to prevent and treat obesity and related complications.

METHODOLOGY

This research is an experimental study with a pre- and post-control group design conducted to evaluate the effect of treatment on cholesterol levels in male Sprague Dawley rats. The study population consisted of male white rats obtained from the Biomedical Laboratory of the Faculty of Medicine, Baiturrahmah University, Padang, Indonesia. The samples used were rats that met the inclusion and exclusion criteria, with the sampling technique using total sampling. The study involved four groups: a negative control group, a positive control group, and two treatment groups that received interventions in the form of Black Garlic Extract at doses of 0.8 and 1.6 mg, respectively. Data analysis was performed using univariate and bivariate statistical tests to determine significant differences between the treatment and control groups, with a p-value <0.05 as the significant limit.

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RESULTS AND DISCUSSION

The Effect of Black Garlic Administration on the Body Weight of Obese Mice Models

The results showed that all mice in the positive control group (K+), treatment 1 (PL1), and treatment 2 (PL2) successfully experienced obesity after induction of a high-fat and fructose diet for 12 weeks as shown in Figure 1 below. After the administration of black garlic intervention, there was a change in body weight in each group. In the positive control group (K+) which did not receive black garlic intervention, body weight increased from 299.14 g to 311.42 g (Δ=12.28 g; $p<0.05$). In contrast, the PL1 group which received a dose of 1.6 ml/day of black garlic experienced a decrease in body weight from 284.85 g to 262.14 g (Δ=-22.71 g; $p<0.05$). The PL2 group that received black garlic at a dose of 0.8 ml/day also experienced a decrease in body weight from 285.85 g to 269.57 g (Δ=-16.28 g; $p<0.05$).

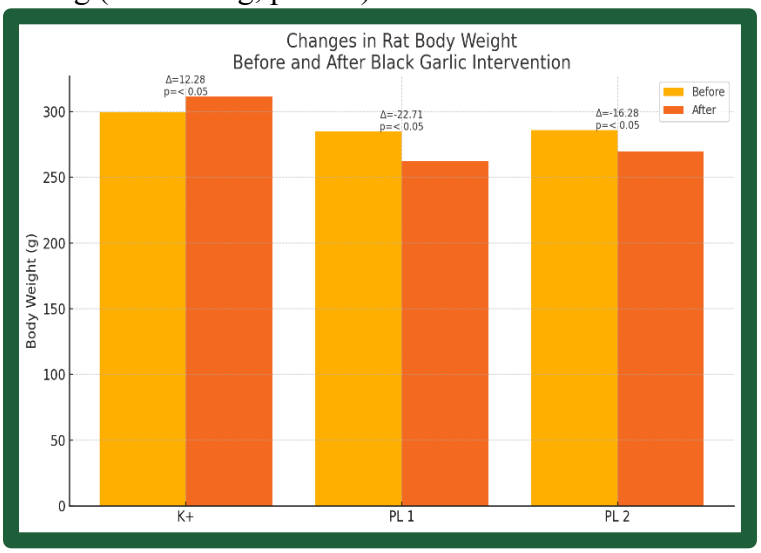

Figure 1. Changes in Rat Body Weight

These results indicate that black garlic administration can inhibit weight gain in obese mice and even tends to reduce weight compared to the positive control group. The greater weight loss in the PL1 group indicates a dose-dependent effect.

The weight loss mechanism is thought to be related to the bioactive compounds in black garlic, such as S-allyl cysteine (SAC), flavonoids, and polyphenols, which increase during the fermentation process. These compounds possess potent antioxidant activity, reducing oxidative stress and chronic inflammation, which play a role in the pathogenesis of obesity. Black garlic is also known to improve lipid metabolism and reduce adipose tissue accumulation in obese animals.(Kimura et al., 2017).

The results of this study are in line with research(Chae et al., 2023a)which reported that aged black garlic can reduce obesity and improve muscle atrophy in obese mice induced by a high-fat diet. Other studies have also shown that black garlic consumption is associated with improved metabolic profiles and reduced body fat accumulation.(Ha et al., 2015).

However, a Bonferroni post hoc analysis found no significant differences between the

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treatment groups. This suggests that the weight loss effect was not large enough to produce a statistical difference between the groups. This is likely due to the relatively small sample size and the limited duration of the intervention.

Effect of High Fat and Fructose Diet on Total Cholesterol Levels

The results showed that all mice fed a high-fat and fructose diet developed hypercholesterolemia. In contrast, all mice in the negative control group fed the standard AIN-93 diet had total cholesterol levels within normal limits. These findings indicate that the high-fat and fructose diet model successfully produced lipid metabolism disorders in the form of increased total cholesterol levels. A high-fat diet increases saturated fatty acid intake, which contributes to increased atherogenic lipoprotein synthesis. Meanwhile, high fructose consumption can increase the activity of the HMG-CoA reductase enzyme, which plays a role in cholesterol synthesis in the liver.

In this study, all mice fed a high-fat and high-fructose diet developed hypercholesterolemia. These results are consistent with research conducted by(Susanti et al., 2019)which showed that a high-fructose diet significantly increased total cholesterol, LDL, and triglyceride levels in Wistar rats. These results are also supported by research by Ndung'u et al. (2024) which showed that long-term exposure to HFHFr causes lipid metabolism disorders and vascular damage, which are early stages of atherosclerosis. Increased total cholesterol levels due to a high-fat and fructose diet are thought to contribute to lipid accumulation in blood vessel walls, endothelial dysfunction, and increased oxidative stress, which accelerates the process of atherogenesis.(Jebari-Benslaiman et al., 2022; Kawai et al., 2021)Therefore, intervention efforts using natural ingredients with antioxidant and hypolipidemic activity, such as black garlic, are important to prevent the progression of vascular damage in obesity.

3.3 The Effect of Black Garlic on Total Cholesterol Levels

After administration of black garlic, there was a tendency for total cholesterol levels to decrease in the treatment group compared to before the intervention, as shown in Figure 2 below. In the positive control group (K+), total cholesterol levels decreased from 108.37 mg/dL to 99.00 mg/dL (Δ=-9.37 mg/dL). The PL1 group experienced a decrease from 116.45 mg/dL to 109.01 mg/dL (Δ=-7.44 mg/dL), while the PL2 group experienced a decrease from 103.58 mg/dL to 100.92 mg/dL (Δ=-2.66 mg/dL).

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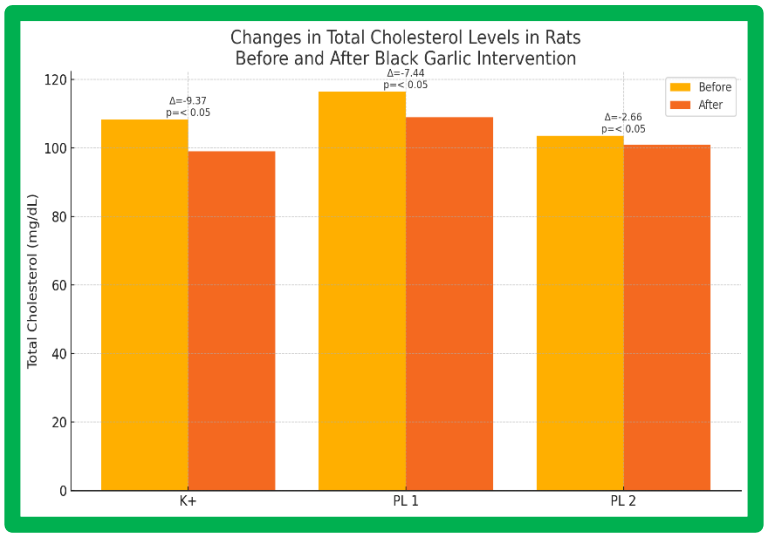

Figure 2. Total Cholesterol Levels Before and After Black Garlic Administration

General Linear Model analysis showed a change in total cholesterol levels before and after the intervention (p=0.001). However, further Bonferroni tests showed no significant difference between treatment groups (p>0.05).

Although it has not yet reached statistical significance, the trend of decreasing total cholesterol levels in the group receiving black garlic indicates the potential hypocholesterolemic effect of this ingredient.

The SAC compound found in black garlic is known to inhibit the activity of the HMG-CoA reductase enzyme, which plays a role in cholesterol biosynthesis. Furthermore, the polyphenol and flavonoid content can increase the activity of the Lecithin Cholesterol Acyltransferase (LCAT) enzyme, which plays a role in reverse cholesterol transport, thereby helping reduce cholesterol accumulation in the circulation.(Kimura et al., 2017)Research by Ha et al. (2015) showed that aged black garlic can improve lipid profiles and antioxidant status in animals fed a high-fat diet. Furthermore, Chae et al. (2023) reported that black garlic is effective in improving metabolic disorders associated with obesity.(Chae et al., 2023b; Ha et al., 2015)

The insignificant difference in cholesterol levels between groups in this study is likely due to several factors. First, the duration of black garlic administration was relatively short, resulting in a suboptimal hypolipidemic effect. Second, the biological response to the active compounds in black garlic can vary among individuals. Third, the stability of the bioactive compounds during fermentation and storage can also influence the effectiveness of the therapy. Corzo-Martínez et al. (2007) stated that the active compound content of garlic products can change depending on processing and storage conditions.

CONCLUSIONS

A high-fat and fructose diet successfully induced obesity and hypercholesterolemia in male Sprague Dawley rats. Administration of black garlic extract at doses of 1.6 mL/day and 0.8 mL/day

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showed a tendency to reduce body weight and total cholesterol levels, but the reduction in total cholesterol levels did not reach statistical significance. Thus, black garlic has potential as an adjuvant therapy in the management of obesity-related metabolic disorders, but further research is needed to determine the optimal dose and duration of administration.

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