



THE EFFECT OF TOFU EXTRACT CONSUMPTION ON WEIGHT LOSS IN MALE WHITE MICE (*Mus Musculus*) GIVEN A HIGH-FAT DIET

Zakia Himmiliyani^{1*}, & Shahrul Rahman²

^{*1}Faculty of Medicine, Muhammadiyah University of North Sumatra, Indonesia

²Department of Internal Medicine, Muhammadiyah University of North Sumatra, Indonesia

e-mail: zakiahimmiliyani8@gmail.com*, shahrulrahman@umsu.ac.id

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Abstrak: Pendahuluan: Obesitas merupakan gangguan metabolisme kompleks yang disebabkan oleh ketidakseimbangan antara asupan energi dan pengeluaran energi sehingga terjadi akumulasi lemak berlebih pada berbagai jaringan adiposa dan organ. Konsumsi makanan dengan kandungan protein tinggi diketahui dapat menekan nafsu makan, salah satunya melalui konsumsi tahu. Tahu mengandung protein dan isoflavon yang diketahui memiliki efek dalam membantu penurunan berat badan. **Tujuan:** Mengetahui pengaruh pemberian ekstrak tahu terhadap penurunan berat badan pada mencit putih jantan (*Mus musculus*) yang diberikan diet tinggi lemak. **Metode:** Penelitian ini menggunakan metode eksperimen dengan desain *Randomized Pretest and Posttest with Control Group Design*. Hewan uji yang digunakan adalah mencit putih jantan (*Mus musculus*) dengan jumlah sampel sebanyak 32 ekor. Peningkatan berat badan dilakukan melalui pemberian kuning telur, kemudian diberikan ekstrak tahu dengan dosis 0,8 dan 1,2 mg/hari selama 14 hari. **Hasil:** Hasil analisis statistik menggunakan *Paired T-Test* menunjukkan adanya penurunan berat badan yang signifikan setelah pemberian ekstrak tahu pada dosis 0,8 dan 1,2 mg/hari ($p < 0,05$). **Kesimpulan:** Terdapat pengaruh pemberian ekstrak tahu terhadap penurunan berat badan pada mencit putih jantan (*Mus musculus*).

Kata kunci: Tahu, Berat Badan, Obesitas, Protein, Isoflavon.

Abstract: Introduction: Obesity is a complex metabolic disorder caused by an imbalance in energy intake and energy expenditure that causes excess fat accumulation in various adipose tissues and organs. Consuming foods with high protein content is known to suppress appetite, one of which is tofu. Tofu contains isoflavon protein which has a weight loss effect. **Objective:** To determine the effect of administering tofu extract on weight loss in male white mice (*Mus musculus*) given a high-fat diet **Method:** This study used an Experimental method with a research design in the form of *Randomized Pre and Posttest with Control Group Design*, using male white mice (*Mus musculus*), with a sample size of 32 mice. Increased body weight by administering egg yolk and administering Tofu Extract at doses of 0.8 and 1.2 mg/day for 14 days. **Results:** The results of statistical analysis with *Paired T-Test* showed a significant decrease in Tofu Extract levels at doses of 0.8 and 1.2 mg/day ($p < 0.05$). **Conclusion:** There is an effect of administering tofu extract on weight loss in male white mice (*Mus musculus*).

Keywords: Tofu, Weight, Obesity, Protein, Isoflavones.

How to Cite

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INTRODUCTION

The rise in obesity in recent decades is due to changes in diet and increasingly unhealthy lifestyles. Consumption of fast food, high-energy foods, and lack of physical activity are the main factors causing an imbalance in energy intake and expenditure, leading to excess fat accumulation in various adipose tissues and organs (Anyanwu, 2023; Hall et al., 2019).

The development of obesity is associated with hyperinsulinemia, insulin resistance, and abnormalities in lipid metabolism. Insulin resistance is considered the most common underlying abnormality in human obesity and is influenced by genetic and environmental factors, particularly changes in diet and physical activity. Lipid abnormalities associated with obesity include increased overall lipid production with increased concentrations of fatty acids, triacylglycerols, low-density lipoprotein (LDL), and very low-density lipoprotein (VLDL) (Willey et al., 2021).

Preventive or therapeutic strategies to control obesity must target changes in these abnormalities. Various dietary modifications designed to control overweight and dyslipidemia focus on manipulating the amount and nature of dietary energy and fat intake. In recent years, increasing attention has shifted to the role of protein intake in obesity management (Hall et al., 2019).

Consuming foods high in protein is known to suppress appetite and food intake in humans. Of the three macronutrients (carbohydrates, fats, and proteins), protein has the strongest effect on suppressing food intake. Furthermore, dietary protein has been shown to induce greater satiety and thermogenic effects and greater weight loss than carbohydrates. One type of protein is plant-based protein, derived from soybeans, which are widely consumed in foods such as tofu (Rahman, 2022; Skov et al., 2020).

Tofu is a source of plant protein rich in isoflavones, substances that have a cholesterol-lowering effect by increasing bile acid excretion and inducing cholesterol utilization in the body. Furthermore, the protein in tofu contains arginine, which is known to suppress cholesterol levels and increase satiety by increasing the gut hormone peptide YY (PYY), which plays a role in appetite suppression (Utari et al., 2020; Bhathena & Velasquez, 2007).

Nagasawa et al.'s (2002) study examined the effects of dietary calories containing soy protein isolate (SPI) in tofu on body fat composition, plasma glucose, lipid and adiponectin levels, and the expression of genes involved in sugar and fatty acid metabolism in obese male rats. Body weight and the weight of adipose tissue, mesenteric fat, epididymis, and brown fat were lower in rats fed the SPI diet. Plasma cholesterol, triglyceride, FFA, and glucose levels also decreased with the SPI diet. Body fat content and plasma glucose levels in rats fed the SPI diet were still lower than those in rats fed an isocaloric casein protein diet.

In a randomized trial in overweight and obese subjects, high protein consumption (25% of total energy) in a low-fat ad libitum diet for 6 months resulted in greater weight loss and body fat loss compared with high carbohydrate consumption (12% of total energy) (Skov et al., 2020).

Previous research aimed to study soybean tempeh extract on weight loss in white mice fed a high-fat diet, but there has been no research examining tofu extract, so the author was interested in conducting research to see the effect of consuming tofu extract on weight loss in male white mice (*Mus musculus*) fed a high-fat diet.

RESEARCH METHODS

This study used an experimental method with a Randomized Pre and Posttest with Control Group Design to assess the effect of tofu extract on weight loss in male white mice (*Mus musculus*) fed a high-fat diet. This study was conducted at the Laboratory Animal Management Unit, Pharmacology and Biochemistry Laboratory, Faculty of Medicine, University of Muhammadiyah North Sumatra, from June 2024 to January 2025. The samples used were 2–3-month-old male white mice in healthy condition. The number of samples was calculated using the Federer formula, with a minimum of 6 mice per group. To anticipate a 20% dropout rate and to accommodate research needs, the number of samples used was 8 mice per group, resulting in a total of 32 mice required in this study.

Sample selection was conducted using a simple random sampling method. Thirty-two mice that met the criteria were numbered and randomly divided into four groups of equal size. Each group received a different treatment according to the research design, which aimed to evaluate the effectiveness of tofu extract in reducing weight in mice fed a high-fat diet.

RESULTS AND DISCUSSION

The following are the average results of weighing the research animals before and after being given egg yolk and after being given tofu extract, namely:

Table 1. Mean and Standard Deviation of Body Weight Measurements Before Egg Yolk Administration

K (-)	K (+)	P1	P2
21.72	22.46	25.07	24.29
21.61	22.44	24.87	24.25
21.57	22.41	24.76	24.21
21.50	22.39	24.50	24.16
21.43	22.35	24.48	24.00
21.37	22.33	24.45	23.77
21.31	22.29	24.39	23.68
21.24	22.27	24.33	23.54
21.46 ± 0.16	22.36 ± 0.06	24.60 ± 0.26	23.98 ± 0.28

The table results obtained the average value of the BB of the research animals in the negative control group was 21.46 ± 0.161 gr, the positive control group was 22.36 ± 0.069 gr, treatment group 1 was 24.60 ± 0.262 gr and treatment group 2 was 23.98 ± 0.288 gr.

Table 2. Mean and Standard Deviation of Body Weight Measurements After Egg Yolk Administration

K (-)	K (+)	P1	P2
22.46	31.08	49.52	33.33
22.25	30.46	35.43	32.42
21.75	30.51	32.30	30.46
22.00	31.00	34.12	30.82
21.76	30.25	34.25	31.22
21.52	30.32	31.23	30.38
21.43	30.20	31.07	31.36
22.02	30.18	33.47	31.40
21.89 ± 0.35	30.50 ± 0.35	35.17 ± 5.99	31.42 ± 1.00

Based on the results of table 2, the average value of the body weight of research animals in the negative control group was 21.89 ± 0.352 gr, the positive control group was 30.50 ± 0.353 gr, treatment group 1 was 35.17 ± 5.992 gr and treatment group 2 was 31.42 ± 1.009 gr.

Table 3. Average and Standard Deviation of Body Weight Measurements After Administration of Tofu Extract

K (-)	K (+)	P1	P2
23.63	32.61	46.11	28.80
23.10	31.55	32.50	27.23
22.13	31.47	29.44	25.72
22.26	32.34	31.07	25.29
22.17	30.87	31.15	26.25
22.22	30.75	29.10	25.07
23.01	31.24	29.00	26.33
22.36	31.40	30.02	26.52
22.61 ± 0.56	31.52 ± 0.65	32.29 ± 5.70	26.40 ± 1.00

Based on the results of table 3, the average value of the body weight of research animals in the negative control group was 22.61 ± 0.561 gr, the positive control group was 31.52 ± 0.651 gr, treatment group 1 was 32.29 ± 5.708 gr and treatment group 2 was 26.40 ± 1.003 gr.

Table 4. Difference in Body Weight Pre and Post Treatment

Group	Pretest	Post Test	BB Difference	%
Control (-)	21.89	22.61	0.72	3.2%
Control (+)	30.5	31.52	1.02	2.2%
Treatment 1	35.17	32.29	-2.88	-8.5%
Treatment 2	31.42	26.4	- 5.02	-17.3%

Based on the results of the table above, the difference in body weight in the negative control group was 0.72 gr (3.2%), the positive control group which was only given 1.02 gr egg yolk (2.2%), treatment group 1 which was given 0.8 mg/day tofu extract, the difference in body weight was -2.88 gr (-8.5%) and treatment group 2 which was given 1.2 mg/day tofu extract was -5.02 gr (-17.3%).

Table 5. Paired T-Test Pre and Post Administration of Tofu Extract

Group	Pre Test	PostnTest	Weight Difference Body	P
Control (-)	21.89	22.61	0.72	0.314
Control (+)	30.5	31.52	1.02	0.241
Treatment 1	35.17	32.29	-2.88	0,000
Treatment 2	31.42	26.4	- 5.02	0,000

With the results of the paired T-Test on the negative control $p = 0.314$ positive control 0.241, treatment 1 $p = 0.000$ and treatment 2 $p = 0.000$, where in treatment groups one and two the p value < 0.05 so that the administration of tofu extract can reduce body weight in this research animal.

Table 6. Independent Sample T-Test Between Groups

	<i>P Value Levene's Test for Equality of Variances</i>	<i>Sig. (2-tailed) T Test Equality of Means</i>
BB Pre Test – Post Test	0.154	0.045

Based on the results of the homogeneity test using Levene's Test for Equality of Variances, a significance value (Sig.) of 0.154 was obtained, which is greater than 0.05 ($0.154 > 0.05$). These results indicate that the data variance between the pre-test and post-test groups is homogeneous or has the same variance. Therefore, the interpretation of the Independent Samples t-Test results is carried out by referring to the Equal Variances Assumed row.

Based on the results of the Independent Samples t-Test in the Equal Variances Assumed row, the Sig. (2-tailed) value was 0.045, which is smaller than 0.05 ($0.045 < 0.05$). Based on the decision-making criteria, H_0 is rejected and H_a is accepted. Thus, it can be concluded that there is an effect of tofu extract on weight loss in research animals.

DISCUSSION

According to the results of this study, there was a decrease after being given tofu extract, with the difference in body weight in the negative control group which was only given feed being 0.72 grams, in the positive control group which was only given egg yolk was 1.02 grams, in treatment group 1 which was given tofu extract 0.8 mg/day the difference in body weight was -2.88 grams and treatment group 2 which was given tofu extract 1.2 mg/day was -5.02 grams (Rahman et al., 2024).

The results of this study are consistent with those of Iritani et al. (2020) who observed weight loss in obese male Sprague-Dawley rats fed soy protein isolate and hydrolysate compared to casein protein for 1 week, at doses of 0.63 and 1.39 g/100 g diet, respectively. The results showed a decrease in body fat and plasma glucose in the rats. The weight loss was 317 ± 24.7 and 217 ± 12.2 g, respectively. The obese rats exhibited glucose intolerance and insulin hypersecretion. These results are also consistent with those of Nagasawa et al. (2002), who stated that a calorie-restricted diet containing soy protein isolate (SPI) reduced body weight and the weight of mesenteric adipose tissue, epididymis, and brown fat in rats. Furthermore, plasma cholesterol, triglyceride, free fatty acid (FFA), and glucose levels were also decreased by the SPI diet.

Park et al.'s (2021) study compared steamed soybeans and soybean koji. Soybean koji is soybeans supplemented with *Bacillus amyloliquefaciens* CJ 14-6. The study assessed weight loss in the steamed soybean group of 6.9 ± 0.44 g and the soybean koji group of 12.4 ± 0.61 g, with a p-value of 0.001. The study also measured the amino acid content of soybeans, namely threonine, glutamic acid, glycine, valine, isoleucine, leucine, tyrosine, phenylalanine, histidine, and tryptophan, with an average total (mg%) of amino acids in steamed soybeans of 15.42 ± 0.41 and soybean koji of 65.8 ± 2.21 .

Obesity occurs due to a complex metabolic disorder caused by an imbalance between energy intake and energy expenditure, leading to fat accumulation in various adipose tissues. Various dietary modifications are designed to control excess weight. Consuming foods high in protein is known to promote weight loss. These high-protein foods can be obtained from soybeans. The proteins contained are 7S globulin (conglycinin) and 11S globulin (glycinin), which comprise approximately 80% of the total protein. Soy protein is considered a complete

protein because it contains most of the essential amino acids found in animal proteins (Bhathena & Velasquez, 2007; Rahman & Santika, 2022).

Soybeans have biological effects in preventing metabolic disorders, such as hyperlipidemia, cardiovascular disease, and type 2 diabetes. These beneficial effects stem from their high content of unsaturated fats, fiber, protein, and isoflavones. The weight loss effects of soy products can be explained by three aspects: soy protein, soy isoflavones, and soy fiber. Protein can reduce body weight by increasing satiety and regulating the mTOR signaling pathway. Natural soy protein can strongly bind soy isoflavones and protect them from degradation after heat treatment. Soy isoflavones can reduce fat accumulation by inhibiting fat production and increasing fatty acid (FA) oxidation. Several studies have reported that isoflavones play an important role in lipid metabolism by regulating genes regulated by peroxisome proliferator-activated receptor (PPAR) and sterol regulatory element binding protein (SREBP) factors that are closely related to glucose and fatty acid metabolism. In addition, isoflavones can affect stearoyl-coenzyme A desaturase 1, which is a key enzyme in obesity (Rietjens et al., 2019; Mullen et al., 2023).

In previous studies, soy protein has been reported to affect obesity by reducing body weight, fasting glucose levels, and liver fat accumulation in animal models. Soy polysaccharides and genistein prevent high-fat diet-induced weight gain, dyslipidemia, oxidative stress, and inflammation in mice. Inhibition of hepatic lipogenic enzymes and fatty acid (FA) synthesis, muscle FA oxidation, increased plasma adiponectin concentrations, and increased fecal triglyceride excretion are some of the mechanisms proposed to explain the antiobesity effects of soy protein. Isoflavones in soybeans have broad metabolic effects. Isoflavones inhibit lipogenesis and increase FA β -oxidation, leading to a reduction in body fat stores, thus being identified as a mechanism of action of isoflavones against obesity (Lu et al., 2019; Akhlaghi et al., 2017).

Isoflavones can inhibit adipogenesis and lipogenesis by interacting with transcription factors and signaling molecules to inhibit fat cell formation. Furthermore, obesity is closely linked to visceral fatty acid metabolism. Soy isoflavones increase visceral fatty acid metabolism in high-fat diet-induced obese male mice, resulting in reduced body weight and visceral fat cell area, as well as suppression of visceral fat synthesis and accelerated fat hydrolysis (Shen & Liang, 2010).

This theory is consistent with research by Tan et al. (2019), which showed that soy isoflavones reduced body weight and serum concentrations of triglycerides (TG), total cholesterol (TC), and low-density lipoprotein (LDL). In the study, mice were fed a basal diet and a high-fat diet for nine weeks, then observed through adipose tissue using hematoxylin and eosin staining. As the main area of energy storage, visceral adipose tissue always accumulates excessively in obese animals. The average area of one visceral lipocyte in male mice with diet-induced obesity (DIO) was significantly larger than in the control group after being fed a high-fat diet. After administration of soy isoflavones, the average area of one visceral lipocyte was significantly reduced in all observed fat depots compared to mice with DIO. The obese mice group also showed increased lipid accumulation in visceral fat, and this lipid accumulation was significantly reduced after administration of soy isoflavones.

This study is in accordance with the research of Iritani et al. (2020), Park et al. (2021), and Lu et al. (2019), which showed that tofu extract can reduce body weight in research animals. The results showed that the weight loss before and after treatment in the group given

1.2 mg/day of tofu extract was -5.02 grams, with the results of the Paired t-Test of 0.003 ($p < 0.05$), so there is an effect of tofu extract on weight loss in this research animal.

CONCLUSION

This study shows that the administration of tofu extract has an effect on weight loss in male white mice (*Mus musculus*) fed a high-fat diet. The treatment groups receiving tofu extract at doses of 0.8 mg/day and 1.2 mg/day experienced significant weight loss compared to the control group, with the largest reduction occurring at a dose of 1.2 mg/day of 5.02 grams or 17.3%. The results of the Paired t-Test showed a significance value of 0.000 in both treatment groups, while the results of the Independent Samples t-Test showed a Sig. (2-tailed) value of 0.045, so it can be concluded that the administration of tofu extract has a significant effect on weight loss in research animals. These findings strengthen the evidence that the protein and isoflavone content in tofu has the potential to play a role in controlling obesity through the mechanisms of increasing satiety, inhibiting lipogenesis, and increasing fat metabolism.

This study has several limitations, including a relatively limited sample size, a short intervention duration, the use of experimental animals, which makes the results not directly generalizable to humans. Furthermore, biochemical parameters such as lipid profiles, glucose, insulin, and adiponectin levels, and histopathological examination of adipose tissue were not measured, allowing for a more in-depth understanding of the mechanism of action of tofu extract. Therefore, further research is recommended using a larger sample size, a longer intervention period, a wider range of tofu extract dosages, and the addition of metabolic biomarker analysis and histological examination. Human clinical studies are also needed to confirm the effectiveness and safety of tofu extract as a supportive therapy for obesity. Practically, the results of this study indicate that tofu extract has the potential to be developed as an alternative functional food or natural supplement that can support obesity prevention and control efforts through a nutritional approach.

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