

Effect of moringa flower extract on liver function and histopathology appearance of obesity model rat's liver

Nuri Rahma Wahyuniasri¹, Leo Nardi^{2*}, Erwin Sopacua³

Abstract

This study aims to test and analyze the effectiveness of moringa flower extract on the liver function of male wistar strain white rats obesity model and how the histopathological picture. This research is a type of experimental quantitative research using post-test only group design. This study used test animals in the form of male white wistar rats with a body weight of 200-300 gr aged 2-3 months. The test animals were divided into 4 groups, the control group was only given regular food and distilled water, the treatment group was given high-fat food and Moringa flower extract at different doses, namely 200 mg / kgBB, 400 mg / kgBB, and 600 mg / kgBB. The results of the phytochemical test showed that moringa flower extract contains secondary metabolites in the form of saponins, tannins, flavonoids, alkaloids, and steroids. The administration of moringa flower extract at a dose of 600 mg/KgBB is effective in improving liver function in obese Wistar white rats. This improvement can be seen through the levels of ALT, AST, and histological structure of the liver that has improved. The results of histopathological observations of liver tissue in treatment group 3, namely moringa flower extract at a dose of 600 mg/KgBB, experienced the most significant improvement and approached the control group compared to other groups.

Keywords: moringa flower extract, liver, histopathology, obese rats

Introduction

Modern lifestyles are characterized by low levels of energy expenditure due to unhealthy dietary habits, increased screen time, low physical activity levels, and insufficient sleep.¹ The transition from traditional diets to processed foods, driven by factors such as urbanization and modernization, has led to a preference for foods that are palatable, shelf-stable, readily available, convenient to store and prepare, and often inexpensive. This shift has resulted in a higher consumption of energy-dense foods rich in oils and carbohydrates, while the consumption of nutrient-rich foods, including vitamins, macronutrients, and micronutrients, has declined.² The increased intake of sugar-sweetened beverages has further contributed to the rising prevalence of obesity and metabolic syndrome.³ These factors collectively lead to fat accumulation, resulting in overweight and obesity.²

Overweight and obesity are two major risk factors for a range of chronic diseases. Overweight is defined as having a body mass index (BMI) between 25 and 29.9, while obesity is defined as having a BMI of 30 or higher.⁴ Obesity is a complex, multifactorial chronic disease that develops when an individual's energy intake consistently exceeds energy expenditure, leading to an accumulation of excess calories.^{5,6} Traditionally, obesity has been defined as an abnormal or excessive accumulation of fat that may impair health, typically measured in clinical practice using the body mass index (BMI).⁷ Obesity has emerged as a significant global public health concern. Several countries worldwide have witnessed a two- or threefold increase in obesity prevalence over the past three decades, attributed to factors such as urbanization,

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sedentary lifestyles, and increased consumption of high-calorie processed foods.⁸ When the body's energy intake exceeds energy expenditure, excess calories are stored as fat, leading to overweight and obesity.⁹

Obesity is a prevalent non-communicable disease (NCD) and is recognized as the fifth leading cause of global mortality. Major risk factors contributing to NCDs include unhealthy dietary habits, physical inactivity, tobacco use, and alcohol abuse. Poor dietary patterns have been linked to an increased risk of chronic diseases and nutritional problems in the public health sector.¹⁰ NCDs have been expanded to encompass a wide range of health issues, endocrine, hematological, and neurological disorders, dermatological conditions, genetic abnormalities, trauma, mental disorders, kidney diseases, gastroenterology, and liver diseases.¹¹ The liver is the largest solid organ, the largest gland, and one of the vital organs that serves as a center for nutrient metabolism and excretion of metabolic waste products. Its primary function is to control the flow and safety of substances absorbed from the digestive system before distributing these substances to the systemic circulation. Total loss of liver function can lead to death within minutes, highlighting the importance of liver function.¹² In individuals with obesity, fat accumulation is distributed throughout many organs, one of which is the liver. Fatty liver disease is the most common liver disease found in obese individuals. This condition is classified as non-alcoholic fatty liver disease (NAFLD) if it occurs regardless of alcohol consumption. NAFLD is strongly associated with metabolic syndrome features, including obesity, insulin resistance, type 2 diabetes, and dyslipidemia. Risk factors associated with NAFLD development include poor dietary habits related to a sedentary lifestyle.¹³ Research on the effect of diet on NAFLD risk and management has found that long-term consumption of sugary drinks is positively correlated with NAFLD, and many individuals with NAFLD tend to consume high levels of sugary drinks compared to others.¹⁴

NAFLD can be divided into two types. The first type is associated with metabolic syndrome and insulin resistance. The second type of NAFLD is associated with infectious pathologies that can lead to liver steatosis. In this case, infections such as hepatitis C and HIV can be the cause, but it is also associated with medications (total parenteral nutrition, glucocorticoids, tamoxifen, tetracycline, amiodarone, methotrexate, valproic acid, vinyl chloride) and specific toxins or congenital/acquired metabolic diseases (e.g., lipodystrophy or cachexia or intestinal bypass surgery).¹⁵ There is no specific drug therapy for NAFLD; however, it is believed that a combination of lifestyle adjustments, increased physical activity, and smoking/alcohol cessation) can be beneficial.¹⁶ Individuals with NAFLD, whether living with obesity, should be encouraged to take part in a healthy lifestyle approach, regardless of weight loss. A healthy diet of reduced intake of calories and high glycemic index foods, increased consumption of monounsaturated fatty acids, omega-3 fatty acids, fiber, and specific protein sources such as fish and poultry is recommended to manage this condition.¹⁵

The consumption of plant extracts has gained significant attention for their health benefits, particularly in addressing Non-Alcoholic Fatty Liver Disease (NAFLD). Since ancient times, plants and their extracts derived from various parts have been utilized for their medicinal properties to treat specific ailments, enhance immunity, and bolster overall well-being.¹⁷ The use of plant extracts has become increasingly pertinent in the current era to harness nature's vital power to combat proliferating diseases such as cancer, heart attacks, diabetes, and accelerated skin aging.¹⁸ *Moringa oleifera*, commonly known as the Moringa tree, stands out as a remarkable plant with all its parts possessing medicinal value. Moringa is recognized as one of the most nutritionally rich food plants. It is exceptionally high in essential amino acids, protein, minerals, vitamins, and polyphenols. This remarkable plant serves as a rich source of phytochemicals, including flavonoids, anthocyanins, isothiocyanates, anthraquinones, alkaloids, essential oils, tannic acid, saponins, steroids, terpenoids, and cardiac glycosides.¹⁹ Moringa is also notable for its content of specific plant pigments, alpha-carotene, and beta-carotene. The therapeutic effects of Moringa are attributed to the combined action of various bioactive components found within the plant, including metal ions, vitamins, alkaloids, polyphenols, and other elements. These components collectively exert their influence on a wide range of physiological processes, including metabolism.²⁰ Moringa exhibits immense nutritional and pharmacological potential, demonstrating antimicrobial, anticancer, antihyperlipidemic, antidiabetic, antiulcer, analgesic, antifertility, and anticonvulsant properties. Notably, almost all parts of

the Moringa plant, including the leaves, roots, bark, flowers, pods, and seeds, have been extensively investigated for their efficacy in treating diabetes.²¹

Moringa is mostly located in rainforest regions and forest ecologies but has now adapted well to organized cultivation systems. When consumed as food, it has positive and preventive effects as well as a variety of powerful therapeutic properties and qualities with significant dietary advantages. Various parts of the moringa plant, including the leaves, flowers, fruits, seeds, and roots, are rich sources of protein, β -carotene, essential amino acids and minerals, and other phenolic compounds. Owing to its various health advantages, moringa is considered a necessary plant for its therapeutic potential. It has been found that the plant has several medicinal properties, including antitumor, anti-inflammatory, antiulcer, antipyretic, antiepileptic, antispasmodic, diuretic, antihypertensive, and antidiabetic. It lowers cholesterol levels, strengthens cells, and exerts hepatoprotective effects. In addition, it has been used traditionally in regional curative systems to cure heart problems, and its antifungal qualities are effectively used to treat various diseases.²² This study aimed to test and analyze the effectiveness of moringa flower extract administration on the liver function of white rat (*Rattus norvegicus*) male Wistar strain obesity model and the histopathological picture.

Method

Design and sampling

This research is a type of experimental quantitative research using a post-test only group design. Male Wistar rats weighing 160-200 gr and 2-3 months old were used in this study. Researchers chose Wistar male rats as research test subjects because these animals have characteristics and physiology that are almost the same as humans and are also one of the most widely used animals in biomedical research. The sample calculation was carried out using the Ferderer formula so that the minimum number of test animals obtained was as many as six heads per group. In this study, researchers used 6 Wistar rats for each experimental group with a total of 24 rats. Grouping of test animals was done randomly into four test groups.

Tools and materials

The tools used in this study include: rat cage, Ohaus scale, glass jar, rotary evaporator, blender, stirrer, rotary evaporator, porcelain cup, test tube, stopwatch, 3 ml syringe, gloves, mask, blunt-tipped sonde syringe, blood capillary pipette, spectrophotometer, recipitation buffer, colorimetric enzymatic kit, hematocrit capillary pipette, ependorf tubes. Meanwhile, the materials used were food and drinks for experimental animals, moringa flowers, alcohol, distilled water, 96% ethanol, etc. NaCl (10% formalin, 70%, 80%, 90% alcohol, xylol, paraffin), HE staining materials (xylol, 100%, 96%, 80%, 70% alcohol, hematoxylin, cosin), tissue, adhesive.

Preparation of Moringa flower extract

Fresh moringa flowers were selected, washed, and then dried. The dried Simplisia material was stored in a tightly closed plastic bag for further use. The simplisia material can be macerated using a suitable solvent. The extraction process of moringa flowers was carried out by the maceration method with 96% ethanol solvent. Moringa flowers were taken from the same area and garden, the same age of the tree (3 years old), in the same order of twigs, and at the same time. Moringa flower extract was prepared by maceration of fresh moringa flowers for 2 days at 30-35°C then crushed using a blender to obtain moringa flower powder. 100 g of moringa flower powder was added to 100 ml of 96% ethanol solvent, placed into a jar, and 1 L of 96% ethanol was added, then closed and left for 48 h protected from sunlight. The mixture was filtered to obtain macerate. Dregs were macerated with 96% ethanol using the same procedure. Maceration was carried out with a digital shaker at 50 rpm until a clear mask was obtained. The resulting liquid extract was evaporated with a rotary evaporator for 2 h.

Animal Preparation

The rats were fed a high-fat, high-cholesterol diet daily. The feed provided was quail egg yolk. This diet increases cholesterol levels exogenously, resulting in obesity. A high-fat, high-cholesterol diet was

given for 14 days before starting treatment with moringa flower extract. The parameters used to confirm obesity were body weight and abdominal circumference. Mice were considered obese if the Lee index was > 0.300 .¹⁴ The Lee index is one of the parameters used to assess whether mice are obese or not, which in humans is expressed in Body Mass Index (BMI).²³

$$\text{Lee index} = \frac{\sqrt{\text{Body weight (gram)} \times 10}}{\text{Nasoanal length (mm)}}$$

Treatment procedure

Test animals that passed the acclimation period and consumed high-fat and cholesterol-fed diets were then randomly divided into four groups, with each group consisting of six rats. Each rat was labeled on its tail using a waterproof marker. In the control group, the rats were only given distilled water. In the treatment group, rats were administered moringa flower extract liquid at different doses. According to research conducted by Wulandari *et al.*²⁴, the dose of moringa extract that effectively reduces cholesterol levels in white rats (*Rattus norvegicus*) male Wistar strain 500 mg/kg BB. Researchers adapted the study and modified the number of doses as follows: 1) Control Group (P0) (rat pellet feed + distilled water/day/head for 14 days); 2) Treatment Group I (P1) (high-fat diet + moringa flower extract at a dose of 200 mg/BB 1 ml and given distilled water/day/head for 14 days); 3) Treatment Group II (P2) (high-fat diet + moringa flower extract at a dose of 400 mg/BB 1 ml and given distilled water/day/head for 14 days); and 4) Treatment Group III (P3) (high-fat diet + moringa flower extract at a dose of 600 mg/BB 1 ml and given distilled water/day/head for 14 days). After treatment for 14 days, the rats were then euthanized under anesthesia and a laparotomy was performed to collect the liver. After the liver was excised, the rats were sacrificed.

Liver function assessment

Liver function can be measured by examining serum enzyme activity, including serum aminotransferase or transaminase. Aminotransferases are good indicators of liver damage. If both are elevated, then there is damage to the liver. The two aminotransferases are Aspartate Aminotransferase (AST), formerly called Serum Glutamic Oxaloacetic Transaminase (SGOT), and Alanine Aminotransferase (ALT), formerly called Serum Glutamic Pyruvic Transaminase (SGPT). ALT and AST are markers of liver functional status, as they can indicate damage to the liver parenchyma. The ALT parameter is used because this enzyme is one of the enzymes produced in the liver and released into the blood where its levels are directly proportional to the state of the liver itself; the higher the level in the blood, the more damaged the liver.²⁵ On day 15, rats were anesthetized for further blood sampling through the orbital vein with a capillary pipette as much as 3 cc collected into an EDTA tube (EthylenediamineTetraacetic Acid) and put into a cool box. Blood samples were then examined at the University of North Sumatra Laboratory to determine Alanine Aminotransferase (ALT) and Aspartate Aminotransferase (AST) levels by photometric examination using a production reagent kit (Analyticon®).

Histopathological preparation

Liver tissues were fixed in 10% Neutral Buffer Formalin solution, sliced, and placed in plastic specimen containers. The tissues were then dehydrated in a series of graded alcohols: 70%, 80%, 90%, absolute alcohol I, and absolute alcohol II, for 2 hours each. The tissues were cleared with xylene and embedded in paraffin, resulting in paraffin blocks stored in a refrigerator. The paraffin blocks were cut into thin sections (5-6 μm) using a microtome. The sections were floated in warm water at 60°C to stretch the tissue and prevent folding. The sections were then mounted on glass slides and stained with Hematoxylin and Eosin (HE) for microscopic examination.

Histopathological observation

Histological preparations were prepared by fixing the liver tissues and transferring them to plastic specimen containers. The sections were stained with hematoxylin-eosin (HE) and examined under a

microscope at 400x magnification. Histopathological changes in the rats were assessed using the Manja Roenigk scoring model. A score of 1 indicated no histological changes (normal), a score of 2 indicated parenchymal degeneration or hemorrhage, a score of 3 indicated hydropic degeneration or fatty infiltration, and a score of 4 indicated necrosis of liver cells.

Data analysis

Histopathological observations were scored and tabulated. The data were analyzed descriptively and statistically using SPSS version 25.0. Data normality was assessed using the Kolmogorov-Smirnov test ($p > 0.05$). One-way ANOVA was used to compare data between groups at a 95% confidence level ($p < 0.05$). Post-hoc analysis was performed using the LSD test.

Results

This study was conducted at the Laboratory of the Department of Pharmacology Pharmacy, Faculty of Medicine, University of North Sumatra, and the Anatomical Pathology Laboratory, University of North Sumatra. This study was conducted between October and December of 2023. This study used test animals in the form of Wistar male white rats with a body weight of 200-300 gr aged 2-3 months. The test animals were divided into four groups: the control group was given regular food and distilled water, and the treatment group was given high-fat food and moringa flower extract at different doses, namely 200 mg/kg BB, 400 mg/kg BB, and 600 mg/kg BB.

In general, the rats were healthy during this study, before and after treatment. A total of 24 test animals were included in this study until the end of the experiment. The body weight of 24 experimental animals was measured. The test animals in the treatment group were fed a high-fat diet for 14 days. A high-fat diet consists of ingredients that have a high fat content in the form of quail egg yolk. This food exogenously increases cholesterol levels in mice to induce the desired condition, namely obesity. The body weights of the test animals were measured on a digital scale. Body weight was measured before the high-fat diet administration and on day 14 after the induction of the high-fat diet to confirm whether the high-fat diet could induce obesity.

Based on the data obtained before the high-fat administration, the Lee index value in the treatment group was 0.28. This value is smaller than <0.3 or not yet included in the obesity category. After consuming a high-fat diet in the form of quail egg yolk for 14 days, body weight and nasoanal length of the rats were calculated to determine the Lee index value. In treatment group 1, the Lee index value changed to 0.31; treatment group 2, to 0.31; and treatment group 3, to 0.32. It can be concluded that the test animals in the treatment group were obese before testing the administration of moringa flower extract to improve liver function.

The results of observations made in all groups showed changes in ALT levels in the treatment group. Based on the average value of ALT levels, it can be seen that the control group had an average value of 119.93 U/L before treatment and after 14 days it became 120.56 U/L. The ALT levels of rats in the control group became normal or reference for the high and low ALT levels in the treatment group. Treatment group 1 had ALT levels of 188.45 U/L. After the administration of moringa flower extract at a dose of 200 mg/kg decreased to 144.2 U/L. Treatment group 2 after a high-fat diet was 187.26 U/L and after being given moringa flower extract at a dose of 400 mg/KgBB to 133.8 U/L. Finally, treatment group 3 after a high-fat diet 187.41 U/L and after being given moringa flower extract at a dose of 600 mg/KgBB to 120.73 U/L.

Table 1. Characteristics of test animals

| Component | Group | | | |
|-------------------------|--------------------------------------|--------|--------|--------|
| | Control | P1 | P2 | P3 |
| Rat type | Wistar strain white rat | | | |
| Sex | Male | | | |
| General condition | White fur color, healthy, and active | | | |
| Starting weight average | 239 gr | 237 gr | 245 gr | 241 gr |
| Average end body weight | 242 gr | 326 gr | 334 gr | 342 gr |

Table 2. Body weight of rats

| Parameter | Group | Average | |
|----------------------|---------|----------------------|---------------------|
| | | Before high fat diet | After high fat diet |
| Body weight (gr) | Control | 239 gr | 242 gr |
| | P1 | 237 gr | 326 gr |
| | P2 | 245 gr | 334 gr |
| | P3 | 241 gr | 342 gr |
| Nasoanal length (mm) | Control | 214 mm | 215 mm |
| | P1 | 216 mm | 221 mm |
| | P2 | 217 mm | 220 mm |
| | P3 | 216 mm | 217 mm |
| Lee index | Control | 0.28 | 0.28 |
| | P1 | 0.28 | 0.31 |
| | P2 | 0.28 | 0.31 |
| | P3 | 0.28 | 0.32 |

Table 3. ALT and AST levels

| Group | Repetition | After high fat diet (U/L) | | After treatment (U/L) | |
|----------------------------|------------|---------------------------|--------|-----------------------|--------|
| | | ALT | AST | ALT | AST |
| Control (Aquades) | 1 | 120.1 | 99.1 | 120.8 | 100.2 |
| | 2 | 118.5 | 97.3 | 119.5 | 98.9 |
| | 3 | 119.8 | 98.9 | 120.3 | 99.5 |
| | 4 | 121.4 | 99.4 | 121.8 | 100.4 |
| | 5 | 120.3 | 98.1 | 121.1 | 99.7 |
| | 6 | 119.5 | 100.2 | 119.9 | 101.1 |
| | Average | 119.93 | 98.83 | 120.56 | 99.96 |
| Treatment I (200mg/KgBB) | 1 | 189.3 | 160.3 | 142.2 | 131.2 |
| | 2 | 188.7 | 159.7 | 144.2 | 129.9 |
| | 3 | 189.9 | 158.2 | 146.3 | 123.2 |
| | 4 | 190.1 | 159.6 | 141.9 | 125.6 |
| | 5 | 185.2 | 160.2 | 145.5 | 128.7 |
| | 6 | 187.5 | 162.6 | 145.1 | 127.4 |
| | Average | 188.45 | 160.1 | 144.2 | 127.66 |
| Treatment II (400mg/KgBB) | 1 | 187.3 | 160.5 | 131.3 | 112.3 |
| | 2 | 189.9 | 161.3 | 135.2 | 115.6 |
| | 3 | 188.3 | 159.9 | 133.2 | 117.3 |
| | 4 | 189.2 | 161.2 | 132.6 | 120.3 |
| | 5 | 185.4 | 161.8 | 136.3 | 119.1 |
| | 6 | 183.5 | 161.4 | 134.2 | 118.3 |
| | Average | 187.26 | 161.01 | 133.8 | 117.15 |
| Treatment III (600mg/KgBB) | 1 | 191.8 | 158.9 | 122.2 | 100.9 |
| | 2 | 187.2 | 162.5 | 121.9 | 101.2 |
| | 3 | 188.4 | 160.3 | 120.2 | 104.6 |
| | 4 | 189.2 | 162.4 | 121.5 | 102.2 |
| | 5 | 184.7 | 161.2 | 119.7 | 105.4 |
| | 6 | 183.2 | 160.4 | 118.9 | 102.3 |
| | Average | 187.41 | 160.95 | 120.73 | 102.76 |

Based on the difference in the average value of ALT levels, the researchers concluded that treatment group 3, namely rats that were obese and given moringa flower extract at a dose of 600 mg/KgBB, had the greatest decrease in ALT levels and was close to the control group. Treatment group 1, which consisted of obese rats administered moringa flower extract at a dose of 200 mg/KgBB, experienced a decrease or improvement in ALT levels that were the lowest compared to treatment groups 2 and 3.

The results of the observations made in all groups showed that there were changes in AST levels in the treatment groups. Based on the average value of AST levels, it can be seen that the control group has an average value of 98.83 U/L before treatment and 99.96 U/L after 14 days. The values obtained in the control group were normal, as well as a reference for the high and low AST levels in the treatment group. Treatment group 1, after being fed a high-fat diet, had AST levels of 160.1 U/L and after being given moringa flower extract at a

se of 200 mg/KgBB to 127.66 U/L. Treatment group 2 after a high-fat diet 161.01 U/L and after being given moringa flower extract at a dose of 400 mg/KgBB to 117.15 U/L. Finally, treatment group 3 after a high-fat diet 160.95 U/L and after being given moringa flower extract at a dose of 600 mg/KgBB to 102.76 U/L. Based on the difference in the mean value of AST levels, the researcher concluded that treatment group 3, namely rats that were obese and given moringa flower extract at a dose of 600 mg/KgBB, had the greatest decrease in serum AST levels and was close to the control group. Treatment group 1, which was an obese rat given moringa flower extract at a dose of 200 mg/KgBB, experienced the least decrease or improvement in AST levels.

Based on the results of the phytochemical tests, it can be concluded that moringa flower extract contains secondary metabolites in the form of flavonoids, saponins, tannins, alkaloids, and steroids. Histopathological observations were made using a light microscope at a magnification of 400x. The purpose of this study was to observe the structure and morphology of the cells in each liver tissue specimen in the control and treatment groups. The administration of Moringa flower extract was administered daily in the morning.

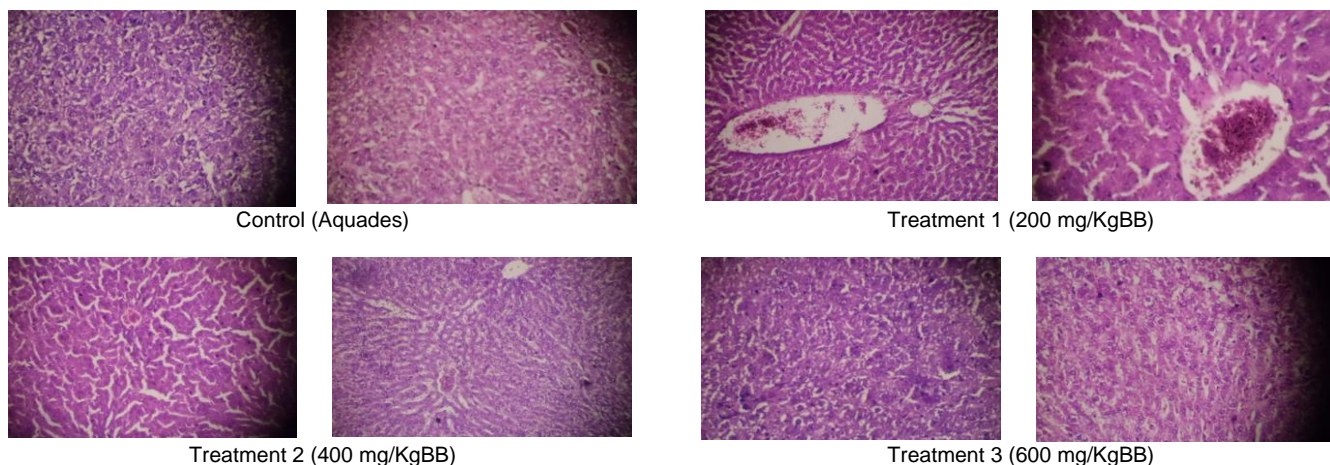


Figure 1. Histopathological image of liver tissue

Histopathological observations revealed different cellular appearances. The control group that was fed regular pellets and distilled water had a normal liver histology picture and was included in the score 1 category, namely no changes in liver histology structure. The histopathology of the liver in the control group was in normal form because it was not fed a high-fat diet; therefore, it was used as a reference to describe the other groups as well as a comparison with the treatment group that was fed a high-fat diet and moringa flower extract. In treatment group 1, there were differences in the shape of the liver structure because the organ was exposed to high-fat diet and obesity. In the histological picture of treatment group 1, necrosis was observed in the liver cells; therefore, it was included in the score 4 category (visible necrosis). In treatment group 2, there was an improvement in the histological structure of the liver, but there was still hydrophilic degeneration or fatty acids, so it was included in the score 3 category. In treatment group 3, there was a histological structure of the liver that was close to that of the control group, so it was included in the score 1 category.

These results indicate that the administration of Moringa flower extract at doses of 400 mg/KgBB and 600 mg/KgBB can improve the histological structure of the liver in obese male Wistar rats. However, the 600 mg/KgBB dose was more effective because it was closer to that of the normal group. This can be seen from the observation of liver histopathology in the control group and treatment group 3, which has a shape that is not significantly different. Improvement of the histological structure of the liver of obese Wistar male white rats is inseparable from the compound content of the moringa flower extract.

The results of the normality test showed that all data were normally distributed ($p > 0.05$); therefore, the homogeneity test was continued using the Levene test to determine whether each variant of the research population group was the same or homogeneous.

The homogeneity test results showed that the control group, treatment group 1, treatment group 2, and treatment group 3 came from populations with the same variance or homogeneous ($p > 0.05$). A one-way Anova test was then conducted to test the significance of the differences between the test groups.

Table 4. Normality test results

| Group | df | Sig |
|---------|----|------|
| Control | 6 | .200 |
| P1 | 6 | .200 |
| P2 | 6 | .200 |
| P3 | 6 | .200 |

Table 5. Homogeneity test results

| Levene static | df1 | df2 | Sig |
|---------------|-----|-----|------|
| 2.322 | 3 | 20 | .106 |

Table 6. One Way Anova test results on ALT levels

| | Number | df | Mean square | F | Sig |
|----------------|----------|----|-------------|---------|------|
| Between Groups | 2344.989 | 3 | 781.633 | 347.805 | .000 |
| Within Group | 44.947 | 20 | 2.247 | | |
| Total | 2389.845 | 23 | | | |

Table 7. Post-hoc LSD test results on ALT levels

| Group | | Mean difference | Sig |
|---------|-------------|------------------------|------|
| Control | Treatment 1 | -23.63333 | .000 |
| | Treatment 2 | -13.23333 [*] | .000 |
| | Treatment 3 | -.16667 | .849 |
| P1 | Control | 23.63333 [*] | .000 |
| | Treatment 2 | 10.40000 [*] | .000 |
| | Treatment 3 | 23.46667 [*] | .000 |
| P2 | Control | 13.23333 [*] | .000 |
| | Treatment 1 | -10.40000 [*] | .000 |
| | Treatment 3 | 13.06667 [*] | .000 |
| P3 | Control | .16667 | .849 |
| | Treatment 1 | -23.46667 [*] | .000 |
| | Treatment 2 | -13.06667 [*] | .000 |

AST level observation results

The One-Way Anova test results showed that there was a significant difference between the control and treatment groups ($p < 0.05$). Post-hoc LSD tests were conducted to analyze the average difference in AST levels between the groups.

ALT level observation results

The test results showed a significant difference between the control and treatment groups ($p < 0.05$). Post-hoc LSD tests were conducted to analyze the average difference in ALT levels between the groups. A significance value smaller than 0.05 means that the group has a significant difference from other groups and vice versa.

The results of the analysis showed that there was a significant difference between the control group and treatment groups 1 ($p = 0.000$) and 2 ($p = 0.000$). The control group and treatment group 3 showed no significant differences ($p = 0.849$).

Table 8. One Way Anova test results on AST levels

| | Number | df | Mean square | F | Sig |
|----------------|----------|----|-------------|---------|------|
| Between Groups | 3011.831 | 3 | 1003.944 | 193.896 | .000 |
| Within Group | 103.555 | 20 | 5.178 | | |
| Total | 3115.386 | 23 | | | |

Table 9. Post-hoc LSD test results on ALT levels

| Group | | Mean difference | Sig |
|---------|-------------|------------------------|------|
| Control | Treatment 1 | -27.70000 [*] | .000 |
| | Treatment 2 | -17.18333 [*] | .000 |
| | Treatment 3 | -2.80000 [*] | .340 |
| P1 | Control | 27.70000 [*] | .000 |
| | Treatment 2 | 10.51667 [*] | .000 |
| | Treatment 3 | 24.90000 [*] | .000 |
| P2 | Control | 17.18333 [*] | .000 |
| | Treatment 1 | -10.51667 [*] | .000 |
| | Treatment 3 | 14.38333 [*] | .000 |
| P3 | Control | 2.80000 [*] | .340 |
| | Treatment 1 | -24.90000 [*] | .000 |
| | Treatment 2 | -14.38333 [*] | .000 |

The results of the analysis showed that there was a significant difference between the control group and treatment groups 1 ($p = 0.000$) and 2 ($p = 0.000$), but there was no significant difference with treatment group 3 ($p = 0.340$).

Discussion

This study was conducted to test and analyze the effectiveness of moringa flower extract administration on the liver function of obese male Wistar strain white rats based on AST and ALT levels, as well as histopathology. The histological conditions of the rat liver that had un-

dergone the trial process were then analyzed. The control group had a normal liver histology. The results of the liver histopathology observations in the control group were used as a reference to describe the other groups as well as for comparison. In treatment group 1, which was fed a high-fat diet and moringa flower extract at a dose of 200 mg/KgBB, there were changes in the shape of the histological structure of the liver from the normal group, because the organ had been exposed to consumption of a high-fat diet. In the histological picture of treatment group 1, which was administered moringa flower extract at a dose of 200 mg/KgBB, necrosis was observed in the liver cells; therefore, it was included in the score 4 category (visible necrosis). Treatment group 2, which was administered moringa flower extract at a dose of 400 mg/KgBB, showed improvement in the histological structure of the liver, but there was still hydrophilic degeneration or fatty acids, so it was included in the score 3 category. Treatment group 3, which was given a high-fat diet and moringa flower extract at a dose of 600 mg/KgBB, showed a histological structure of the liver that was close to that of the control group, so it was included in the score 1 category (no change in shape or normal).

The results of this study indicate that the administration of 600 mg/kg BB moringa flower extract can improve the histological structure of the liver of obese Wistar male white rats. This can be seen from the results of liver histopathology observations in the control group and treatment group 3, which were not significantly different. Improvement of the histological structure of the liver of obese wistar male white rats is inseparable from the compound content of moringa flower extract. The content of secondary metabolites in moringa flower extract can repair cell tissue damaged by a high-fat diet and obese conditions experienced by white Wistar rats.

The phytochemical test results showed that moringa flower extract contains secondary metabolites, such as flavonoids, saponins, tannins, and triterpenoids. Previous studies have found that flavonoids, as natural substances with broad pharmacological activities and good therapeutic effects, have excellent antioxidant, anti-inflammatory, metabolic disease repair, anti-tumor, and other properties and can significantly reduce fatty liver.²⁶

Conclusion

Moringa flower extract contains secondary metabolites in the form of saponins, tannins, flavonoids, alkaloids, and steroids that help repair liver cells that experience fatty acids and necrosis due to obesity. Administration of 600 mg/KgBB was effective in improving liver function in obese Wistar white rats. This improvement can be seen in the levels of ALT and AST and the histological structure of the liver. The results of histopathological observations of liver tissue in treatment group 3, namely moringa flower extract at a dose of 600 mg/KgBB, showed the most significant improvement and approached the control group compared to other groups.

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