



Sub-Acute Toxicity of Banana (*Musa Paradisiaca* L.) Heart Extract on Haematological Parameters in Mice

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ARTICLE INFO

Article history:

Received: 01 April 2024

Revised: 16 July 2024

Accepted: 21 July 2024

Published online : 01 October 2024

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ABSTRACT

Banana heart is a rich source of antioxidant compounds with numerous health benefits. The aim of the present study is to determine the sub-acute toxicity of banana heart extract on haematological parameters *in vivo*. Kepok banana heart extract was obtained by maceration in ethanol (70%). The extract was made into a suspension using 0.5% sodium carboxymethyl cellulose (Na CMC). Thirty-six (36) male albino mice were divided into four groups (1 – 4) of nine animals each. Group 1 was used as the control mice and were administered 0.5% Na CMC, while groups 2 – 4 served as the treatment groups, and were administered 200 mg/kg, 400 mg/kg, and 800 mg/kg dose of kepok banana extract suspension, respectively. The extract suspension was administered orally once daily for 21 days. Blood samples were collected on weekly interval at 7, 14 and 21 days of extract administration, and were used for haematological analysis using a hematology analyzer. The parameter investigated were erythrocyte count, leukocyte count, platelet count, hemoglobin concentration, hematocrit, and erythrocyte indices which include mean cell volume (MCV), mean cell haemoglobin (MCH), and mean cell haemoglobin concentration (MCHC). The results showed that kepok banana extract administration did not cause significant variation in the haematological parameters. All the parameters were within normal values. This therefore indicates that kepok banana heart extract is relatively safe on sub-acute administration.

Keywords: Kepok banana heart, *Musa paradisiaca* L., Hematological parameters, Toxicity.

Introduction

The use of natural medicines is currently undergoing significant development in various aspects of human life. In addition to being more economical, natural medicines tend to be safer than synthetic drugs.¹ Medicinal plants have thousands of different species. Of the approximately 40,000 species of medicinal plants identified worldwide, about 30,000 are found in Indonesia. This figure covers about 90% of the total medicinal plants in the Asian region. Of these, 25%, or about 7,500 species have been shown to have medicinal properties. However, only about 1200 plant species have been used as raw materials for herbal medicines.²

Indonesia is a tropical country with fertile land that yields a diverse range of traditional medicinal plants, including the banana plant. The banana heart, known as 'ontongpisang,' in Indonesia is a flower produced by the banana tree from which the banana fruits are produced. The banana heart has multiple layers of reddish-brown-purple skin and a milky cream-white interior. Between the layers of skin lies a jejunal-shaped arrangement of flowers with a soft core.³

Banana heart contains flavonoids, phenols, alkaloids, tannins, saponins, and coumarins. Further research is needed to determine the safety of this material, including toxicity testing. The present study aimed to investigate the effect of sub-acute administration of banana heart extract on blood parameters in mice.

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Citation: Dhipa S, Aldi Y, Husni E. Sub-Acute Toxicity of Banana (*Musa Paradisiaca* L.) Heart Extract on Haematological Parameters in Mice. Trop J Nat Prod Res. 2024; 8(9): 8332-8335. <https://doi.org/10.26538/tjnpr/v8i9.13>

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria

Materials and Methods

Collection and identification of plant materials

Fresh kepok banana heart (*Musa paradisiaca* L.) were obtained from banana plantations in Nagari Sumanik, Salimpaungsub district, Tanah Datar Regency, West Sumatra, Indonesia. The kepok banana was identified at Herbarium Andalas (ANDA), Department of Biology, Faculty of Mathematics and Natural Sciences, Andalas University, Indonesia. The plant material was assigned voucher number 39/K-ID/ANDA/1/2024.

Preparation of extract

Kepok banana heart (10 kg) was extracted by maceration in 70% ethanol at room temperature. The extract was concentrated in a rotary evaporator at 45°C and a pressure of 150 bar to obtain a thick extract.

Preparation of 0.5% sodium carboxymethyl cellulose (Na CMC) suspension

Na CMC powder (500 mg) was placed in a mortar, 10 mL of hot water was added gradually until the powder dissolved. The suspension was grind until homogeneous and then made up to 100 mL with distilled water.

Preparation of extract suspension

The Na CMC suspension (0.5%) was mixed with kepok banana heart extract according to the planned dose calculation. The extract-Na CMC suspension was homogenized, and then made up to volume with distilled water.

Experimental animals and extract administration

Thirty-six (36) male albino mice age 2-3 months weighing between 20 – 30 g were used for the study. The animals were grouped into four (4) of nine animals each, and treated as follows:

Group I: This was used as the control group, and they were administered 0.5% Na CMC suspension without the banana heart extract.

Group II: The animals were administered 200 mg/kg dose of banana heart extract suspension.

Group III: The animals were administered 400 mg/kg dose of banana heart extract suspension.

Group IV: The animals were administered 800 mg/kg dose of banana heart extract suspension.

The extract was administered orally once a day for 21 days. The volume of the test extract administered to the animals was 0.32 mL/20 g body weight of the animal.

Haematological analysis

On weekly interval (on day 7, day 14, and day 21) of extract administration, blood samples were collected in EDTA tubes, and were analyzed using the Exigo™ H400 digital Hematology Analyzer. The analysis employs the flow cytometric method, which measures cells based on the electrical or optical impedance produced on passage of the cells. The following hematological parameters were analysed; erythrocyte count, leukocyte count, platelet count, hemoglobin concentration, hematocrit, mean cell volume (MCV), mean cell hemoglobin (MCH), mean cell hemoglobin concentration (MCHC).

Statistical analysis

The data were analyzed using a two-way analysis of variance (ANOVA). The data was presented as means \pm standard deviation (SD).

Results and Discussion

Effect of kepok banana heart extract on erythrocyte count

Administration of kepok banana heart extract did not cause significant alteration in erythrocyte count ($p > 0.05$). The highest mean erythrocyte count was observed in group 4 (800 mg/kg) with a mean value of 9.56 million cells/ μ L. Although high, the erythrocyte count is still within normal limit. The high erythrocyte count may be due to the antioxidant effect of kepok banana heart extract. Antioxidants can stimulate the formation of red blood cells (erythropoiesis) in the bone marrow.⁴ The average value of erythrocytes obtained from the study results is within the normal range of erythrocyte counts in mice, which ranges from 3.57-15.2 million/ μ L.⁵ This observation suggests that erythrocyte count is not significantly affected by variation in the dose and duration of administration of kepok banana heart extract (*Musa paradisiaca* L.) (Figure 1).

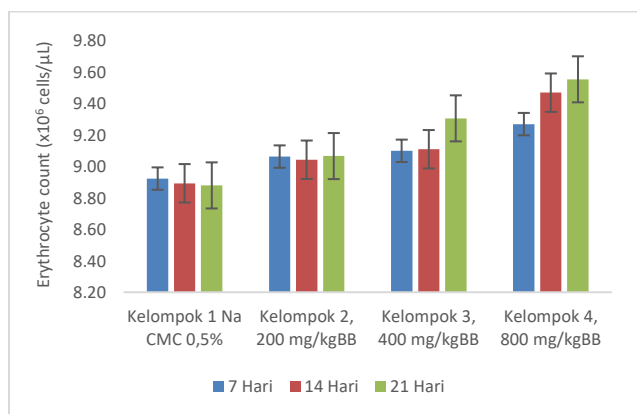


Figure 1: Effect of kepok banana heart extract on erythrocyte count in male mice

Effect of kepok banana heart extract on leukocyte count

Kepok banana heart extract administration results in an increase in leukocyte count across the treatment groups with animals in group 3 (400 mg/kg) having the highest leukocyte count of 7.58×10^3 cells/ μ L. Statistical analysis showed that the increase in the leukocyte count in the treatment groups was not significant compared to the control ($p > 0.05$). The increase in leukocyte count in mice may be attributed to the constituents of kepok banana heart extract which may have immunostimulatory effect. Phenolic compounds can enhance the body's immune system by increasing the effectiveness of lymphokine proliferation produced by T cells, thus stimulating phagocytic cells. In addition, phenolic compounds can also increase the activation of effector cells such as lymphocytes and macrophages, which produce and release cytokines such as interleukins (IL-1, IL-6, IL-2), and tumor necrosis

factor-alpha (TNF-alpha).⁶ An increase in the number of white blood cells may be an indication of disease or infection in the body.⁷ However, in this test, the elevated white blood cell count was still within the normal range. The average leukocyte count in mice ranges from $1.06-56.08 \times 10^3$ cells/ μ L.⁵ It can be interpreted that the leukocyte count of the mice is not affected by variations in the dose and duration of administration of kepok banana heart extract (*Musa paradisiaca* L.) (Figure 2).

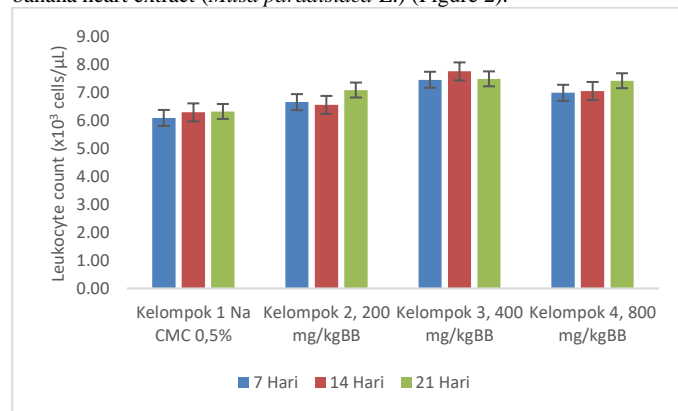


Figure 2: Effect of kepok banana heart extract on leukocyte count in male mice

Effect of kepok banana heart extract on platelet count

There was a decrease in the mean platelet count in the treatment groups compared to the control. The lowest platelet count of 672,000 cells/ μ L of blood was observed in group 4 (800 mg/kg) mice. However, the decrease in platelet count following the sub-acute administration of kepok banana heart extract was not statistically significant when compared to control ($p > 0.05$). The decrease in platelet count may be due to the antioxidant properties of kepok banana heart extract, which has been shown to have antithrombotic effect in mice. Flavonoids as a source of antioxidants can inhibit arachidonic acid synthesis and the enzyme cyclooxygenase (COX), which plays a role in the production of prostaglandins and thromboxane. Prostaglandins play an important role in inflammatory responses, while thromboxane is an important mediator of platelet aggregation. Inhibition of arachidonic acid synthesis by flavonoids can produce anti-inflammatory and antiplatelet effects.⁸ Despite the reduction in platelet levels as a result of kepok banana heart extract administration, the values were still within the normal range of $59 - 2633 \times 10^3$ cells/ μ L.⁵ This observation suggests that administration of kepok banana heart extract has no toxic effect on the platelets in mice (Figure 3).

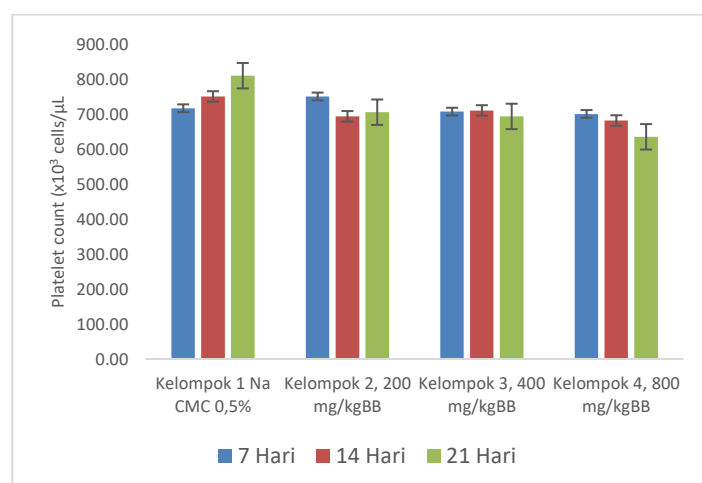


Figure 3: Effect of kepok banana heart extract on platelet count in male mice

Effect of kepok banana heart extract on hemoglobin concentration

Kepok banana heart extract administration results in no significant alteration in the hemoglobin levels in mice. This may be attributed to the flavonoid content of the extract. Flavonoids play an important role in

hemoglobin production by influencing iron absorption and release. This relationship is very important because the formation of methemoglobin is related to the form of iron in the heme molecule, which is maintained by the antioxidant properties of flavonoids.^{8,9} Although, the hemoglobin level tended to increase following the extract administration, but were within the normal range. The average normal value of hemoglobin in mice is between 6.1-21.7 g/dL.⁵ It could be inferred that the amount of hemoglobin in the blood is not influenced by the variation in dose and duration of administration of kepok banana heart extract (Figure 4).

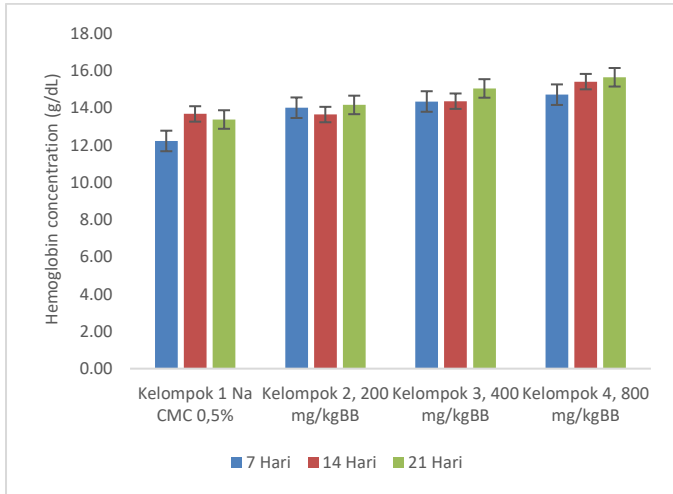


Figure 4: Effect of kepok banana heart extract on haemoglobin concentration in male mice

Effect of kepok banana heart extract on hematocrit

The hematocrit level varied among the treatment group compared to the control, but these values were still within the normal range. The average hematocrit level in mice ranges from 16.7 - 69.8%.⁵ Hematocrit values may increase due to an increase in red blood cell levels or a decrease in blood plasma volume. A decrease in hematocrit may be caused by red blood cell damage, decreased red blood cell production, or affected by the number and size of red blood cells.¹⁰ These factors can cause hematocrit values to vary from person to person. A decrease in hematocrit can also be caused by overuse of anticoagulants. This can cause the erythrocytes to shrink, resulting in a lower hematocrit value.¹¹ However, the result obtained from this study showed that the hematocrit levels in the mice were not altered significantly after sub-acute administration of kepok banana heart extract (Figure 5).

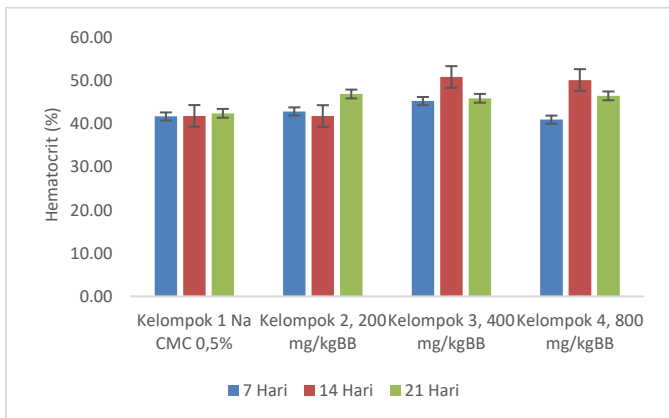


Figure 5: Effect of kepok banana heart extract on hematocrit in male mice

Effect of kepok banana heart extract on erythrocyte indices

Although, there were variations in the erythrocyte indices among the treatment groups, but the indices measured including mean cell volume (MCV), mean cell haemoglobin (MCH), and mean cell haemoglobin

concentration (MCHC), were all within the normal range. The average normal values for MCV, MCH, and MCHC in mice ranged from 39 - 90.8 fL, 12.6 - 31 pg, and 27 - 37.6 g/dL, respectively.⁵ Analysis showed that the variation in the erythrocyte indices were not statistically significant ($p > 0.05$). High MCV values indicate a larger average volume of erythrocytes (macrocytes), while low values indicate a smaller average volume of erythrocytes (microcytes). For MCH, high values indicate the presence of erythrocytes that contain more hemoglobin, and low values indicate erythrocytes that contain less hemoglobin. In addition, high MCHC values indicate red blood cells that are rich in hemoglobin, while low values indicate red blood cells that are deficient in hemoglobin.¹¹ Since the erythrocyte indices were within normal values, it therefore suggests that kepok banana heart extract has no toxic effect on the red blood cells (Figures 6, 7, and 8).

Conclusion

Findings from the present study has shown that kepok banana heart extract has no toxic effect on haematological parameters in mice. All haematological parameters were within normal values after sub-acute administration of the extract. These observations suggest that kepok banana heart extract is relatively safe following sub-acute administration. Further studies may be required to investigate its effect on other parameters as well as the histological effects on vital organs.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

Acknowledgments

This study was supported by Andalas University (Indonesia) through Dana Penelitian DIPA Fakultas Farmasi No.08/UN16.10.D/PJ.01./2023

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