



# Response of White Rat (*Rattus Norvegicus*) Leukocyte Numbers with Temulawak (*Curcuma Xanthorrhiza* Roxb) Extract in Severe Exercise

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**Abstract.** Strenuous exercise can increase oxygen uptake in active muscle cells, which in turn can affect the number of leukocytes. Temulawak can prevent an increase in the number of leukocytes. The aim of this study was to analyze the effect of giving ginger extract on the number of leukocytes in male Wistar white rats (*Rattus norvegicus*) with heavy exercise. This type of research was a pure experiment with a pretest-posttest with control group design on 28 rats that were given maximum physical activity and treated with ginger extract for 28 days. The test animals were divided into 4 groups, namely, the control group was not given ginger extract and 2 treatment groups were given ginger extract at doses of 6.75mg/day and 13.5mg/day. Statistical analysis uses the One way Anova test. There was no difference between groups before treatment ( $p=0.495$ ) with the highest leukocyte count of 13.5 mg/day. There were differences between groups after treatment ( $p=0.039$ ) with the highest leukocyte count in the control treatment. There was a difference between groups in delta ( $p=0.002$ ) with the highest decrease in leukocytes in the 13.5 mg/day treatment. Temulawak extract has an effect on reducing the number of leukocytes. Giving ginger extract in the amount of 13.5 mg/day is the most effective dose in reducing the number of leukocytes.

**Keywords:** Ginger Extract, Leukocyte Levels, Heavy Sports Activities

## 1 INTRODUCTION

Strenuous exercise impacts the number of leukocytes. The acute response to vigorous exercise is characterized by an increase in circulating leukocytes, primarily due to the mobilization of these cells from the spleen and bone marrow into the bloodstream. This phenomenon is driven by the release of stress hormones such as catecholamines (epinephrine and norepinephrine), which increase the movement of leukocytes and increase their number in circulation [1-2]. Research shows that during and immediately after vigorous exercise, there is an increase in the number of neutrophils. Research

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found neutrophil levels increased after vigorous exercise [3]. This increase is associated with an inflammatory response, in which cytokines such as IL-6 and TNF-alpha are released, which stimulate leukocyte activity [4]. The increase in leukocyte counts is not uniform, while neutrophils usually increase, lymphocyte counts may initially decrease before recovering within a few hours after exercise [5]. During strenuous exercise, the body experiences physiological stress triggering the release of cortisol and other hormones that modulate immune function [6]. This hormonal response is very important because it not only facilitates leukocyte mobilization but influences their functional capacity, potentially increasing their ability to respond to pathogens [7]. In addition, higher intensity exercise tends to produce more pronounced leukocytosis compared to moderate exercise [8-9]. However, excessive exercise can lead to increased risk of infection due to subsequent reduction in leukocyte function [8, 10].

Temulawak (*Curcuma xanthorrhiza* Roxb) has attracted attention due to its various pharmacological properties, including its potential effects on leukocytes. Research has found that bioactive compounds in ginger, namely curcumin and xanthorrhizol, show anti-inflammatory and immunomodulatory effects and influence leukocyte dynamics. Research has proven that ginger has an anti-inflammatory effect that has the potential to change the number of leukocytes in various conditions, including infections and inflammatory diseases [11-13]. Temulawak extract was found to influence immune cell function and leukocyte activity. Antioxidants are known to play a role in reducing oxidative stress, which is associated with inflammation and immune response. By reducing oxidative stress, ginger helps regulate leukocyte proliferation and activity [14]. The anti-inflammatory effect of ginger has been shown to reduce levels of pro-inflammatory cytokines. This decrease causes a decrease in leukocyte activation and recruitment, thereby potentially reducing the number of leukocytes [15-16]. This study aims to determine the effect of giving ginger on leukocyte levels in mice.

## 2 METHODS

The research used the True Experiment method with a Randomized Pre test post test control group design. This research used 7 white Wistar rats (*Rattus norvegicus* L.) as experimental animals in each group for 28 days. Maximum physical activity ability was measured by swimming using the forced swim test method. Rats were randomly divided into three groups, treated with 6.75 mg of ginger extract/day in group 1 (P1), 13.5 mg of ginger extract/day in group 2 (P2) and group 3 without treatment (K). Haemocytometer for checking mouse leukocyte levels. All data analysis using SPSS 24 software was tested using the One Way Anova test and continued with the Post Hoc LSD test. This research has received recommendations from the Health Research Ethics Commission, Faculty of Nursing and Health Sciences, Muhammadiyah University Semarang No. 001/KE/04/2024.

### 3 RESULTS AND DISCUSSION

#### 3.1 Rat Body Weight

The average daily weight development results of each group of mice can be seen in Figure 1. Based on Figure 1, it was found that the body weight of mice in the three groups tended to increase compared to their initial body weight with almost the same pattern of increase. Based on the results of the ANOVA repeated measure statistical test, there was no difference in the body weight of mice in the three groups ( $p=0.487$ ).

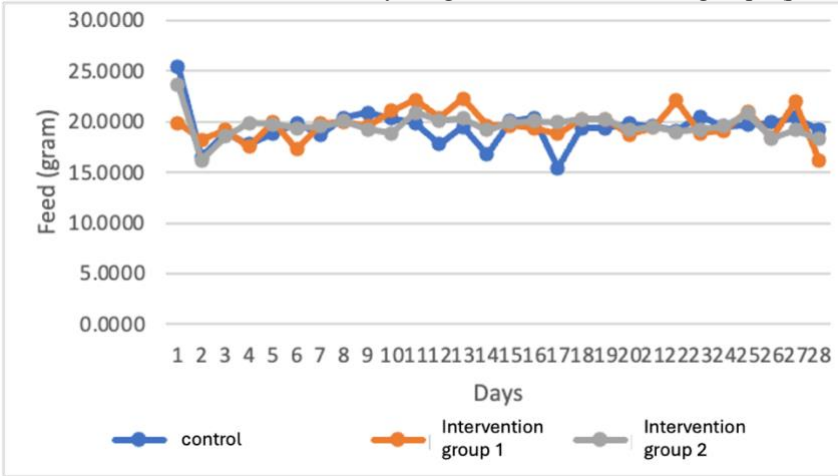
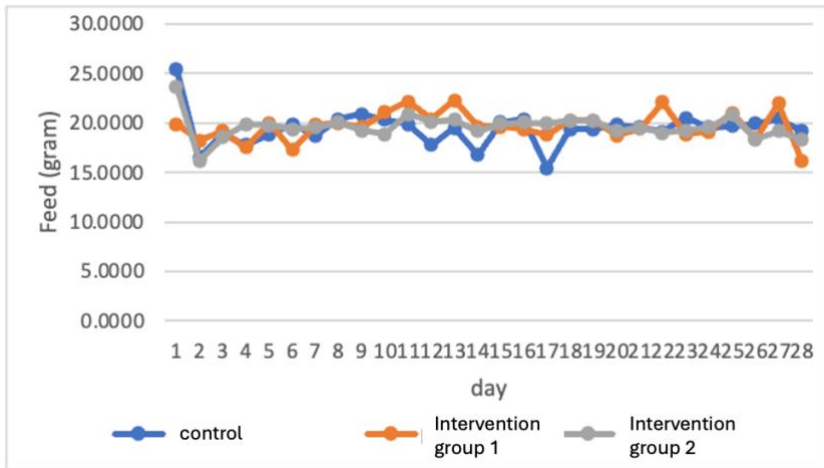


Fig. 1. Average weight development of mice

#### 3.2 Rat Feed Intake

Based on Figure 2, the food intake of rats in each final group tended to decrease compared to the initial intake. The results of statistical tests using the ANOVA repeated measure test showed no difference in the diet of the mice from the three treatment groups ( $p = 0.786$ ). The decrease in mouse food intake was thought to be due to stress resulting from heavy exercise. The higher the physical activity and the longer the duration, the greater the possibility of excessive physical activity causing loss of appetite and changes in appetite hormones [17]. Excessive physical activity can reduce ghrelin levels in the brain and also increase leptin in mice. Ghrelin itself is a hormone that stimulates appetite and leptin is a hormone that suppresses appetite [18].



**Fig. 2.** Average feed development of mice

### 3.3 The Effect of Giving Temulawak Extract on the Number of White Rat Leukocytes

The mean difference in the number of leukocytes can be seen in Table 1, there was a significant decrease in the number of leukocytes in the P2 group (dose 13.5mg/day) which had a mean decrease of  $-3.78 \times 10^3/\mu\text{L}$ , meaning that P2 was the best group in reducing the number leukocytes compared with other groups. The control group experienced an increase in the number of leukocytes with an average increase of  $2.35 \times 10^3/\mu\text{L}$ , this was because the control group was not given ginger extract but was only given drink and control feed. An increase in leukocytes will occur at maximum activity, leukocytes will increase if there is cell damage caused by free radicals which increases due to high oxygen consumption [19].

**Table 1.** Average Difference in Leukocyte Counts Before and After Treatment

	K	P1	P2	<i>p value</i>
	Mean±SD	Mean ±SD	Mean ±SD	
Pre	10.86±3.27	13.71±2.54	12.66±4.28	0.495
Post	13.21±2.68	11.9±3.57	8.88±1.41	0.039
Delta	2.35±2.17	-1.76±1.99	-3.78±3.23	0.002

K : Control group (group without treatment)

P1 : Temulawak extract treatment group with a dose of 6.75 mg

P2 : Temulawak extract treatment group with a dose of 13.5mg

The results of the One Way ANOVA test before treatment concluded that there was no difference in the mean number of leukocytes in each treatment group ( $p$ -value  $>0.05$ ). After treatment, there was an effect of giving ginger extract on reducing the

number of leukocytes (p-value <0.05) and likewise there was a difference in the effect of giving ginger extract on reducing the number of leukocytes (p-value <0.05). The results of the Post Hoc LSD test found that there were differences between groups P1 and P2.

**Table 2.** LSD Post Hoc Test Results

Groups	Groups		
	K	P1	P2
K	-	0.383	0.006*
P1	-	-	0.043*
P2	-	-	-

- K : Control group (group without treatment)
- P1 : Temulawak extract treatment group with a dose of 6.75 mg
- P2 : Temulawak extract treatment group with a dose of 13.5mg

The results of this research are supported by research by Supriyadi (2022) and Rosidi (2014) that giving ginger in drinking water has an effect on reducing the number of leukocytes [20-21]. Standardized ginger extract decreased inflammatory markers such as IL-1 $\beta$  and NF- $\kappa$ B in periodontal tissue, indicating a mechanism that can reduce the activity and number of leukocytes during the inflammatory response [22]. These findings suggest that ginger intake may cause a decrease in leukocyte counts due to its ability to modulate inflammatory pathways. Additionally, ginger's antioxidant properties are attributed to its high phenolic content, playing an important role in its health benefits. showed that ginger extract showed high total phenolic content, which was related to its antioxidant capacity [23]. Antioxidants can reduce oxidative stress, which is often associated with increased leukocyte activation and proliferation. By reducing oxidative stress, ginger can help reduce the overall number of leukocytes, because excessive leukocyte activation is a characteristic of chronic inflammation [16]. In addition, the immunomodulatory effects of ginger have been documented in various studies. Curcuma extract can influence breast milk production in postpartum mothers, which is closely related to immune function and leukocyte activity. Modulation of the immune response through food components such as Curcuma xanthorrhiza can produce a balanced number of leukocytes, especially in individuals with inflammatory conditions. [24].

#### 4 CONCLUSION

There is an effect of giving ginger extract on reducing the number of leukocytes. The greatest reduction was at a dose of 13.5 mg/day.

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