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The Effect of Red Ginger (*Zingiber officinale* var. *rubrum* rhizoma) on Neutrophil Levels After Eccentric Exercise in Support of SDG 3

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ABSTRACT

Objective: Exercise-induced muscle damage (EIMD) following eccentric exercise triggers an acute inflammatory response characterized by increased neutrophil infiltration. Although non-steroidal anti-inflammatory drugs (NSAIDs) are commonly used to manage inflammation, their prolonged use may cause adverse effects. This study aimed to analyze the effect of red ginger (*Zingiber officinale* var. *rubrum*) extract on plasma neutrophil levels after eccentric exercise in untrained individuals as a natural anti-inflammatory strategy supporting SDG 3 (Good Health and Well-being). **Method:** The experimental study employed a randomized pretest-posttest control-group design involving 30 untrained medical students from Universitas Negeri Surabaya. Participants were randomly assigned to either a control group (placebo) or an experimental group receiving red ginger extract ($n = 15$ each). Blood samples were collected before exercise and after intervention to determine plasma neutrophil levels. Data were analyzed using normality tests followed by paired-sample t -tests with SPSS version 26. **Results:** The normality test confirmed that all variables were normally distributed ($p > 0.05$). The control group showed no significant change in plasma neutrophil levels after eccentric exercise ($p = 0.820$). In contrast, the experimental group demonstrated a significant reduction in neutrophil levels following red ginger supplementation ($p < 0.001$), indicating a decreased inflammatory response after exercise-induced muscle damage. **Novelty:** The study demonstrates the anti-inflammatory potential of red ginger extract using plasma neutrophil count as an early biomarker of EIMD after eccentric exercise. The findings provide evidence for a safe, affordable, and locally available natural alternative to conventional anti-inflammatory medication, contributing to healthier exercise recovery and supporting Sustainable Development Goal 3.

INTRODUCTION

An individual who does sports, whether an athlete, a trained or untrained layperson, will experience exercise-induced muscle damage (exercised-receptor damage/EIMD). EIMD occurs mainly in eccentric movements because fewer motor units are involved so that the mechanical stress on the muscle is greater (Douglas et al., 2017). EIMD is characterized by, among others, decreased muscle strength and pain (Hyldahl & Hubal, 2014), damage to muscle fibers (Heckel et al., 2019), sarcolemma and muscle extracellular matrix (Qian et al., 2023), and inflammation which, although it contributes to healing, also produces reactive oxygen species (ROS) (Bessa et al., 2016), so that it can reduce athlete performance and achievement and increase the number of sports contributions to society.

To reduce inflammation that occurs after exercise, both in the context of performance and recreation, anti-inflammatories are generally used, especially non-steroidal anti-inflammatory drugs (NSAIDs). However, long-term use of this drug causes 30% of hospitalizations due to drug side effects (Bindu et al., 2020), liver dysfunction

(Lundberg & Howatson, 2018), gastrointestinal bleeding and acute or chronic kidney disorders (Lucas et al., 2019).

Several studies have been conducted to replace NSAIDs in reducing inflammation, one of which is the use of ginger. Gingerol, shogaol and several other substances in ginger can reduce pro-inflammatory cytokines (Mashhadi et al., 2013). Another study stated that subjects given ginger supplementation had 20-45% lower TNF- α , IL-6, and IL-1 β than those given placebo (Wilson, 2015). Another study also showed that there was a decrease in plasma pro-inflammatory cytokines in trained men after giving ginger for 6 weeks, potentially reducing the effect of these cytokines in inducing fatigue (Zehsaz et al., 2014). Ginger can also reduce IL-1 α and TNF α which are some of the inflammatory markers in 32 untrained women (Barari, 2016).

The above studies prove the anti-inflammatory effects of ginger. However, the above studies use pro-inflammatory cytokines as markers of inflammation which generally increase in high-intensity exercise but are inconsistent because many other factors influence (Cerqueira et al., 2020). In addition, the above studies use endurance training, while for observation of muscle damage it is easier to use eccentric movements because there is greater mechanical stress on the muscles.

Beyond its scientific relevance, identifying a safe, affordable, and locally available natural anti-inflammatory agent also aligns with the Sustainable Development Goals (SDGs), particularly SDG 3 (Good Health and Well-being). By supporting faster recovery and reducing the discomfort that discourages physical activity, while lowering dependence on pharmacological agents whose long-term use carries documented health risks, such an agent can contribute to safer exercise participation and healthier communities, especially in resource-limited settings where red ginger is widely accessible.

This study aims to analyze the effect of red ginger extract on reducing the number of plasma neutrophils after eccentric exercise in untrained individuals through careful monitoring and analysis pre-intervention and post-intervention. This study used red ginger extract because it has higher anti-inflammatory activity than white ginger (Aksono et al., 2022). Neutrophils were chosen as a marker of inflammation because neutrophils are one of the markers of the inflammatory response in EIMD (Stožer et al., 2020), with an increase in blood levels occurring more quickly, namely between 1-24 hours after eccentric exercise (Peake et al., 2017).

RESEARCH METHOD

Study design

This study is an experimental interventional study with control groups and randomization (Aziza, 2023). The design of this study is a pre test-post test control-group design, namely the sample is divided into 2 groups, control and experiment, where both groups will undergo a pre test and post test, but only the experimental group is given treatment (Creswell & Creswell, 2018).

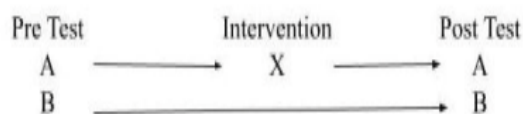


Figure 1. Research design

Participants

The sample in this study were students of the Medical Faculty of Universitas Negeri Surabaya. The sample collection technique in this study used purposive sampling, namely determining the sample with certain considerations (Sugiyono, 2007). The consideration in this study was students who were not trained or did not exercise regularly because in untrained people, skeletal muscles are less able to store glycogen so that they stimulate a faster inflammatory response (Brown et al., 2015). The sample size in this study was 30 students of the Faculty of Medicine, Universitas Negeri Surabaya, divided into 2 groups, namely 15 people as the control group and 15 people as the experimental group with the inclusion criteria in this study as follows:

- a) Active students of the Faculty of Medicine, Universitas Negeri Surabaya;
- b) Do not exercise regularly according to WHO recommendations, namely not doing moderate intensity aerobic exercise for 150-300 minutes and/or 75-150 minutes of high intensity aerobic exercise for one week (World Health Organization, 2022);
- c) Willing to be a research subject by filling in the consent form

The exclusion criteria in this study are as follows:

- a) Have a history of injury that can interfere with the course of the study;
- b) Resign as a student of the Faculty of Medicine, Universitas Negeri Surabaya when the study is taking place;
- c) Sick when the study is taking place;
- d) Experiencing an injury during the study

Research research instrument

The instruments used in this study were 10 cc syringe, 3ml Plain Blood Tube, 3ml EDTA K3 Blood Tube, Stopwatch, Red ginger extract solution using 4g of red ginger powder, placebo, hematology analyzer SYSMEX Type XP-100 Method consist of participants, instrument and procedures, data analysis.

Procedure

Blood sampling was performed before the subjects performed eccentric exercise and then analyzed to see the basal neutrophil levels. After that, the subjects were asked to do push ups and squats as much as they could. The experimental group was given ginger extract solution, while the control group was given a placebo. Blood sampling was performed again 2 and 18 hours after intervention of red ginger extract. Subjects were not allowed to drink anything other than water for 24 hours before the study was conducted until after the second blood sampling.

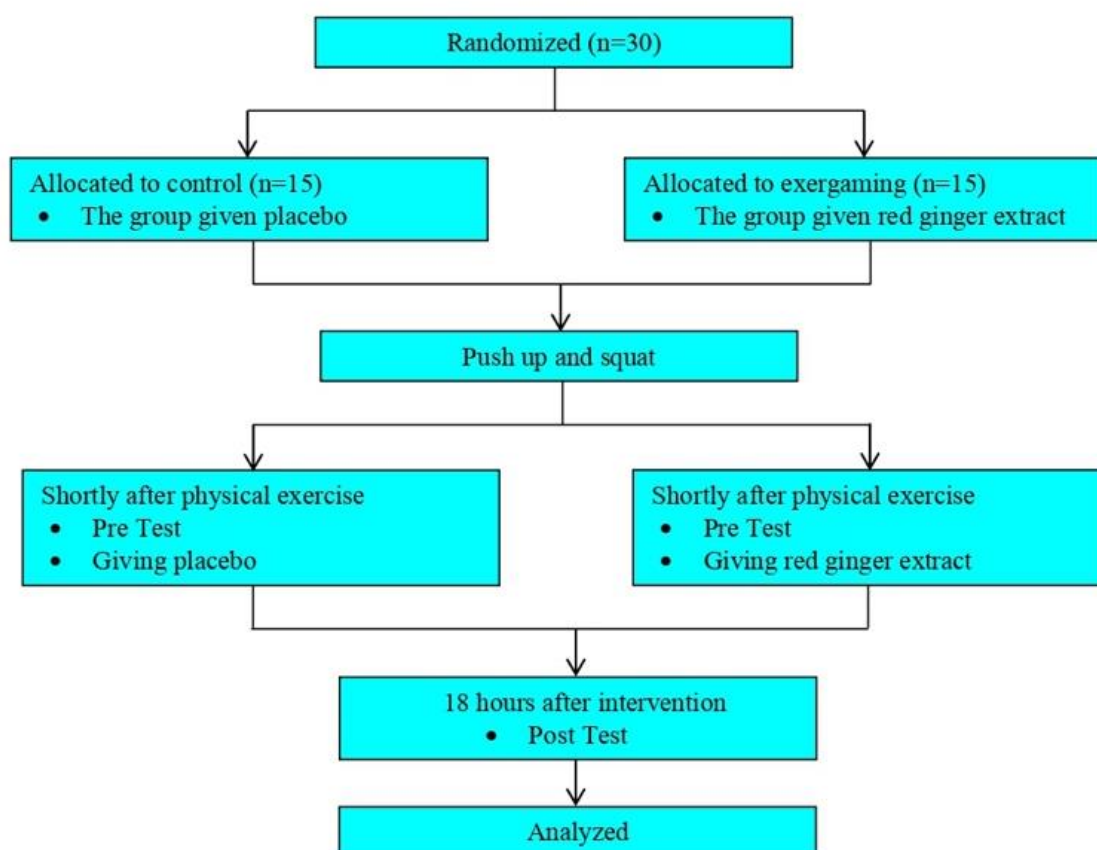


Figure 2. The procedure flowchart

Statistical analysis

Data in the form of laboratory test results were documented and then analyzed using SPSS version 26. Data normality testing was performed before conducting a t-test. The results of these tests were interpreted to see the comparison of blood neutrophil levels in the two groups and then conclusions were drawn.

RESULTS AND DISCUSSION

Results

Table 1. Number of neutrophils based on gender

Gender		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	Male	15	50.0	50.0	50.0
	Female	15	50.0	50.0	100.0
	Total	30	100.0	100.0	

Table 2. Number of neutrophils based on age

Age		Frequency	Percent	Valid Percent	Cumulative Percent
Valid	18	21	70.0	70.0	70.0
	19	8	26.7	26.7	96.7
	20	1	3.3	3.3	100.0
	Total	30	100.0	100.0	

Table 3. Test of normality

Tests of Normality		Kolmogorov-Smirnov ^a			Shapiro-Wilk		
		Statistic	Df	Sig.	Statistic	Df	Sig.
Pre Test Control		.148	15	.200*	.949	15	.509
Post Test Control		.166	15	.200*	.914	15	.157
Pre Test Experiment		.192	15	.143	.942	15	.408
New Post Test Experiment		.158	15	.200*	.946	15	.457

*. This is a lower bound of the true significance.

a. Lilliefors Significance Correction

Table 4. Number of neutrophils in control group

Paired Samples Statistics						
		Mean	N	Std. Deviation	Std. Mean	Error
Pair 1	Pre Test Control	5.1667	15	1.74833		.45142
	Post Test Control	5.0600	15	1.65478		.42726

Table 5. Paired t-test in control group

Paired Samples Test Paired Differences										
			Mean	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference		t	df	Sig. (2-tailed)
						Lower	Upper			
Pair 1	Pre Control	Test - Post Test Control	.10667	1.78464	.46079	-.88164	1.09497	.231	14	.820

Table 6. The correlation between variables in the control group

Paired Samples Correlations			
		N	Correlation Sig.
Pair 1	Pre Test Control & Post Test Control	15	.451 .091

Table 7. Number of neutrophils in experimental group

Paired Samples Statistics					
		Mean	N	Std. Deviation	Std. Error Mean
Pair 1	Pre Test Experiment	6.1933	15	1.33816	.34551
	Post Test Experiment	4.7600	15	1.40499	.36277

Table 8. Paired t-test in experimental group

Paired Samples Test Paired Differences										
			Mea n	Std. Deviation	Std. Error Mean	95% Confidence Interval of the Difference		t	df	Sig. (2- taile d)
						Lower	Upper			
Pair 1	Pre Experiment Post Experiment	Test - Test	1.43 333	.71880	.18559	1.03528	1.83139	7.723	14	.000

Table 9. The correlation between variables in the experimental group

Paired Samples Correlations			
		N	Correlation Sig.
Pair 1	Pre Test Experiment & Post Test Experiment	15	.864 .000

The normality test using Kolmogorov-Smirnov and Shapiro-Wilk (Table 3) showed that all variables had a Sig. value > 0.05, which means that the data is normally distributed. Therefore, parametric tests can be used for further analysis. The result of data analysis based on gender (Table 1) and age (Table 2) showed that there was no significant difference in the number of neutrophils between male and female either before or after eccentric exercise.

This indicates that the inflammatory response to eccentric exercise is relatively uniform between the two sexes. Although some studies have shown that hormonal factors, such as estrogen in female can have a protective effect on inflammation and muscle damage. The results of the study based on gender in this study did not show a significant difference.

The majority of respondents in this study were in the age range of 18-20 years. Further analysis showed that there was no significant difference in changes in the number of neutrophils between different age groups. This may be due to the fact that

the age range used in this study was quite narrow, making it impossible to see the greater variation in inflammatory responses that may occur in older or younger individuals.

The results of the analysis of changes in the number of plasma neutrophils in the control group (without red ginger extract) (Table 4) obtained an average pre test result = 5.1667 with an SD value = 1.74833, and an average post test = 5.0600, with an SD value = 1.65478. The results of the Paired T-Test (Table 5) obtained Sig. (2-tailed) = 0.820 p value > 0.05 means not significant. From these results, it is interpreted that there is no significant change in the number of neutrophils after eccentric exercise in the control group. In the experimental group (with red ginger extract) (Table 7) the average pre test result was 6.1933, with an SD value = 1.33816 while the average post test result was 4.7600, with an SD value = 1.40499. Paired T-Test results (Table 8) obtained Sig. (2-tailed) = 0.000 p value < 0.05, it can be concluded that there is a significant decrease in the number of neutrophils after eccentric exercise in the experimental group.

The results of the correlation between variables in the control group (Table 6), the pre test-post test correlation was 0.451 (p = 0.091) meaning a weak and insignificant correlation. In the experimental group (Table 9), the pre test-post test correlation was 0.864 (p = 0.000) meaning a strong and significant correlation. These results indicate that changes in the number of neutrophils in the experimental group have a strong relationship with the treatment given, namely red ginger extract.

Discussion

The results of the study in the experimental group with a p value < 0.05 showed that administration of red ginger extract had a significant effect in reducing the number of plasma neutrophils after eccentric exercise, and can be interpreted as a decrease in the inflammatory response due to exercise-induced muscle damage (EIMD). Exercise-Induced Muscle Damage (EIMD) is a condition that occurs due to intensive physical activity, especially after eccentric exercise, which causes microscopic damage to muscle fibers. One of the main signs of EIMD is increased neutrophil infiltration, which occurs within 1-24 hours after exercise (Peake et al., 2017). This increase in neutrophils aims to clean damaged tissue and induce muscle regeneration. However, if inflammation is excessive, it can prolong recovery time and reduce physical performance.

In the control group, the p value > 0.05 means there was no significant change in the number of neutrophils. In the control group (not given ginger extract solution) showed that eccentric exercise caused inflammation that persisted without any intervention. In contrast, in the experimental group given red ginger extract, there was a significant decrease in the number of neutrophils (p < 0.05), indicating that red ginger was able to inhibit the inflammatory response after exercise.

This indicates that red ginger can reduce the effects of excessive inflammation and accelerate the recovery of muscle tissue damaged by eccentric exercise. The eccentric exercise carried out in this study triggered an inflammatory response characterized by an increase in the number of neutrophils in the control group. This indicates that eccentric exercise causes muscle damage that triggers the activation of the innate immune system. In the experimental group, administration of red ginger extract significantly reduced the number of neutrophils, indicating that red ginger was able to reduce the inflammatory response due to EIMD. This effect is likely due to the ability of active compounds in red ginger to suppress the activation of inflammatory pathways,

thereby accelerating the muscle recovery process and reducing the duration of unwanted inflammation.

These results support previous findings that controlling excessive inflammation can help accelerate muscle recovery without inhibiting the tissue regeneration process. Therefore, consuming red ginger extract after strenuous exercise can be an effective nutritional strategy in optimizing muscle recovery while reducing discomfort due to prolonged pain and inflammation.

Research conducted by Alshibani et al. (2022) showed that ginger had similar effectiveness compared to NSAIDs in reducing pain in postoperative periodontitis patients. Another study showed that there was no significant difference in pain relief between ginger and mefenamic acid, but fewer side effects occurred in subjects given ginger (Ankita et al., 2020), so it can be considered safe for short-term effect in young, untrained individuals. Overall, ginger can be used as recovery regiment in post because it can reduced muscle pain in Delayed Onset Muscle Soreness (DOMS) (Hoseinzadeh et al., 2015), included post eccentric exercise (Matsumura et al., 2015). Many substances released from muscle during exercise including several inflammatory cytokines that have been suggested to play a role in muscle hyperalgesia (Mizumura & Taguchi, 2024). Red ginger inhibits the synthesis of inflammatory mediators by inhibiting the expression of cyclooxygenase (COX)-1, COX-2, and 5-lipoxygenase (5-LO) enzymes, the expression of inducible nitric oxide synthase (iNOS) that would produce nitric oxide, which is mediated by the attenuation of nuclear factor kappa B (NF- κ B). 6-gingerol in ginger inhibits 12-O-tetradecanoyl phorbol acetate 13 (TPA)-induced ornithine decarboxylase (ODC) activity and inflammation (Wilson, 2015).

Red ginger (*Zingiber officinale* var. *Rubrum*) contains gingerol, shogaol, and zingerone which have anti-inflammatory properties through inhibition of the NF- κ B pathway, which is a key regulator of inflammation (Zhang et al., 2022). Previous studies have also shown that ginger supplementation can reduce the production of IL-6, TNF- α , and CRP, which play a role in systemic inflammation (Rondanelli et al., 2020). In addition, gingerol and shogaol are known to inhibit the production of prostaglandins and leukotrienes which are the main mediators of inflammation.

In this study, a significant decrease in neutrophils in the experimental group indicated that the active compounds in red ginger were able to reduce inflammation by suppressing neutrophil recruitment, which is in line with previous studies on the anti-inflammatory effects of ginger. These results further strengthen the understanding that red ginger not only works as an antioxidant but also has an important role in modulating the immune system and inflammation after strenuous exercise. This study shows that red ginger extract supplementation can significantly reduce the number of neutrophils after eccentric exercise. This effect is in line with research stating that ginger has the ability to suppress the release of pro-inflammatory cytokines and reduce oxidative stress, which are often associated with inflammation due to muscle injury. In addition, the results of this study also showed significant short-term effects in young, unprotected individuals. This group of subjects tends to be more susceptible to muscle damage and higher inflammatory responses after eccentric exercise, because physiological adaptation to intense physical activity has not yet formed.

A significant decrease in the number of neutrophils in the group receiving red ginger extract indicates that this supplementation can help reduce acute inflammation in the early phase after exercise. This finding is important because it can increase comfort and

motivation to exercise in beginner individuals, as well as reduce the risk of injury due to excessive inflammation.

The results of this study have important implications in the field of sports and rehabilitation. For athletes, red ginger supplementation can be used as a natural strategy to accelerate muscle recovery after intense exercise and prevent excessive inflammation that can interfere with athlete performance. For the general public, red ginger can help reduce muscle pain after exercise, which can increase sports participation rates and reduce the risk of long-term injury due to prolonged inflammation. The potential application of the results of this study in the context of sports physiotherapy is also worth highlighting explicitly. Eccentric training, although effective in increasing muscle strength, is widely known as a major trigger for Delayed Onset Muscle Soreness (DOMS), which is muscle pain that appears 24 to 72 hours after exercise due to microscopic damage to muscle fibers. This condition can interfere with performance, reduce exercise motivation, and prolong recovery time, especially in individuals who are just starting an exercise program or returning after a period of rest. The results of this study indicate that red ginger as a natural anti-inflammatory agent is effective in reducing the number of neutrophils after eccentric exercise, which is an indicator of the acute inflammatory response to muscle damage. This suggests that red ginger supplementation has the potential to be used as part of a non-pharmacological recovery strategy in sports physiotherapy.

The limitations of this study only measure the number of neutrophils as a marker of inflammation. Other parameters such as IL-6, TNF- α , or CRP which are also markers of inflammation can provide a more comprehensive understanding of the anti-inflammatory mechanisms of red ginger. This study can be continued by examining the long-term effects of red ginger administration, as well as comparing its effects with pharmacological anti-inflammatories such as NSAIDs to determine its effectiveness in managing inflammation after physical exercise.

CONCLUSION

Fundamental Finding : Eccentric exercise increased plasma neutrophil counts, reflecting inflammation due to muscle damage. The control group showed no significant change, indicating that inflammation persists without intervention, whereas the experimental group given red ginger extract showed a significant decrease in neutrophil counts. This demonstrates that red ginger extract is effective in suppressing the acute post-exercise inflammatory response. **Implication :** Red ginger has potential as a natural, accessible anti-inflammatory supplement to accelerate muscle recovery after intense physical exercise. This may benefit both athletes and the general public by improving comfort, supporting exercise participation, and reducing reliance on NSAIDs, thereby contributing to SDG 3 (Good Health and Well-being). **Limitation :** This study measured only the number of neutrophils as a marker of inflammation. Other parameters such as IL-6, TNF- α , or CRP were not assessed, the sample was limited to 30 untrained students within a narrow age range, and only short-term effects were observed. **Future Research :** Future studies should examine additional inflammatory markers (e.g., IL-6, TNF- α , CRP) for a more comprehensive understanding of the anti-inflammatory mechanisms of red ginger, evaluate longer-term effects, and compare red ginger with pharmacological anti-inflammatories such as NSAIDs to determine its relative effectiveness in managing post-exercise inflammation.

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AUTHOR CONTRIBUTIONS

In accordance with the Contributor Roles Taxonomy (CRediT): **R.P. Nevangga, A.P. Bakti, N.S.R. Pembayun, and D.A.T. Rachman** contributed to conceptualization, research and sampling methodology, and data analysis; **U. Asyrofa and K. Sahani** contributed to manuscript writing and revision. All listed authors have reviewed and approved the final version of this submission.

CONFLICT OF INTEREST STATEMENT

The authors confirm that there are no conflicts of interest, either financial or personal, that may have influenced the content or outcome of this study.

ETHICAL COMPLIANCE STATEMENT

This manuscript complies with research and publication ethics. The authors affirm that the work is original, conducted with academic integrity, and free from any unethical practices, including plagiarism.

STATEMENT ON THE USE OF AI OR DIGITAL TOOLS IN WRITING

The authors acknowledge the use of digital tools, including AI-based technologies, as support during the writing stages of this article. All outputs generated with digital assistance were critically evaluated and revised to ensure that academic rigor and ethical standards were upheld. The final responsibility for the manuscript rests entirely with the authors.

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