

Differences in the Grade of Inflammation in the Liver Tissues of Old White Female Mice (*Mus Musculus*) due to Weight Bearing and Non-Weight Bearing Exercise

Muhammad Alim Ananto^{1*}, Irfiansyah Irwadi², Alphania Rahniayu³

¹ Faculty of Medicine, Universitas Airlangga Surabaya, Indonesia

² Department of physiology, Universitas Airlangga Surabaya, Indonesia

³ Department of Pathology, Universitas Airlangga Surabaya, Indonesia

*Corresponding author: alimananto@yahoo.com

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ABSTRACT

Old age causes a decrease in quality of life. Exercise inhibiting the decline in quality of life, however exercise can cause oxidative stress that causes tissue injury including in the liver. There are two types of exercise, weight-bearing and non-weight bearing exercise. It is unknown the type of exercise causes the least injury. This study aimed to analyze the differences in the inflammatory response shown by the degree of inflammation in the liver tissue of old female white mice who performed weight-bearing and non-weight bearing exercise. Thirty-six mice (*mus musculus*) were divided into 3 groups, control group (12), weight-bearing treatment group (12) and non-weight bearing treatment group (12). The weight bearing treatment consisted of running for 30 minutes, while the non-weight bearing treatment consisted of swimming for 30 minutes. Euthanized on the last day of 4th week. Microscopic evaluation of the degree of inflammation of the liver tissue was carried out according to the Batts and Ludwig method. Statistical analysis used the Kruskal Wallis method to determine whether there were differences in the degree of liver inflammation between groups. Mean value of inflammation degree in control group: $1,50 \pm 0.552$. Mean value of inflammation degree in weight-bearing exercise group: $1,92 \pm 0.515$. Mean value of inflammation degree in non-weight bearing exercise group: $1,92 \pm 0.669$. The Kruskal Wallis statistical test showed the P value 0.139. The results showed that there was no significant difference in the degree of inflammation in the mice liver after receiving weight-bearing and non-weight-bearing exercise treatments.

I. BACKGROUND

In aging there is a process of degeneration, a decrease in general physical condition and the emergence of chronic diseases that reduce the quality of life. Along with the aging process, the decline in general physical condition and coupled with decreased physical activity in the elderly, causes an increase in a person's chances of suffering from chronic diseases. Daily activities, regardless of whether a person does physical work, or sports, often result in skeletal muscle injury and this with muscle performance, increased tissue edema, inflammation and degeneration/necrosis as well as pain (Baker, B. A., 2017). Various components of the body in the elderly experience unavoidable degeneration. Changes in the body of the elderly include a decrease in neurological function, muscle mass and strength, the immune system, etc (Amarya S., Singh K., 2018).

The process of degeneration due to aging can be inhibited through exercise activities. A good quality of life in the elderly can be achieved if it is supported by proper exercise. Inappropriate exercise will cause a chemical response that can have a negative impact on the body. In the general public, especially the elderly, do not understand the various types of exercise, the benefits and negative impacts they have on the body (Simioni C., et.al. 2018).



Sport is a physical activity that will trigger the body's system response to improve general health conditions. Exercise is considered a preventive therapy to prevent cognitive and memory decline in aging.⁴ In addition, many tissues, organs, and body systems are affected by exercise (Rueggsegger G. N., Booth F. W. 2018), so it can be a solution to improve the physical condition and quality of life of the elderly. There are two types of exercise programs, namely weight bearing and non-weight bearing. Weight bearing exercise is a type of strength training to build physical strength and increase skeletal muscle mass by utilizing the force of gravity to generate pressure generated by muscles through concentric contractions or eccentric contractions, while non-weight bearing exercise is a type of strength training to build physical strength and strength. increase the mass of the skeletal muscles without utilizing the force of gravity to generate the pressure generated by the muscles through concentric contractions or eccentric contractions. Both of these exercises are very beneficial for increasing muscle mass and body strength, but if done incorrectly it can have a bad impact. Physical activity such as exercise will produce free radicals, including Reactive Oxygen Species (ROS). These free radicals have an impact on causing oxidative stress that causes oxidative damage to proteins, fats and nucleic acids. This causes damage to various tissues, including the liver (Simioni C., etc. 2018., Pizzino, G., etc. 2017., and Kawamura Takuji, Isao Muraoka 2018). Oxidative stress in the long term will be responded to by the body as an inflammatory injury (Kumar Vinay, Abdul K. Abbas, Jon C. Aster. 2018).

The liver is a large glandular tissue in the human body. The liver plays an important role in metabolic processes so that if pathological changes appear in a person's body, it will appear in the condition of the liver. High and long-lasting oxidative stress can cause tissue injury, including liver tissue. Injury to liver tissue can be observed microscopically and can be assessed using a grading method based on Batts and Ludwig (Goodman, Zachary D. 2007).

Until now, not many studies have studied the correlation between exercise activity and the level of inflammation in liver tissue, especially in the elderly. To solve this problem, a study is needed to find out how the different degrees of inflammation in the liver tissue of old female white mice are treated with different weight-bearing and non-weight-bearing exercise treatments as an illustration of what kind of exercise is appropriate for the elderly.

II. METHODS

The research design used was an experimental study with mice (*Mus musculus*) as experimental animals. The approach used in this study is a randomized post-test only control group design. Experimental animals were divided into 3 groups, consisting of 1 control group and 2 treatment groups and then given treatment according to the group. The weight bearing treatment in this study was in the form of running using a treadmill at a speed of 21 cm/second, at 13.00 PM, with duration 30'/day, 6 days/week, in 4 weeks. The non-weight bearing treatment in this study was swimming without weights, at 13.00 PM, with duration 30'/day, 6 days/week, in 4 weeks. Independent variables are weight bearing exercise treatment and non-weight bearing exercise treatment. The dependent variable is degree of inflammation. The controlled variables are experimental animal variants, age of experimental animals, and the sex of the experimental animal. The inclusion criteria were *mus musculus* mice, female, aged 7-8 months, looked active and healthy. The exclusion criteria were mice with disabilities, mice died during adaptation.

There are several evaluation systems for chronic hepatitis, one of which is the Batts and Ludwig scoring system. In general, this scoring system assesses grades and stages, similar to other scoring systems. Grade indicates the degree of inflammation and hepatocellular injury, which eventually progresses to fibrosis. Grade is determined based on the results of the assessment of periportal interface hepatitis and parenchymal injury (Goodman, Zachary D. 2007).

III. RESULTS AND DISCUSSION

The grading was carried out on the results of observations of the degree of inflammation that appeared in the sample area. The following are microscopic examination data.

Table 1. Data grading the degree of inflammation in the liver of mice.

Group	Grade 1	Grade 2	Grade 3	Grade 4
Control	6	6	-	-
Weight bearing	2	9	1	-
Non-Weight bearing	3	7	2	-

The average level of inflammation seen in liver tissue was assessed using the Batts and Ludwig grading method in the control group, the weight bearing group and the non-weight bearing group. The results show that there is no significant difference in the degree of liver inflammation.

- Statistical test comparing the degree of inflammation of the control group and the non-weight bearing group was performed using the Kruskal-Wallis H statistical test. The P value was 0.119. The value is > 0.05 , it can be concluded that there is no significant difference in the degree of inflammation in the liver of experimental animals in the control group and the non-weight bearing group.
- Statistical comparison of the degree of inflammation in the weight-bearing group and the non-weight-bearing group was performed using the Kruskal-Wallis H statistical test. The P value was 0.972. The value is > 0.05 , it can be concluded that there is no significant difference in the degree of inflammation in the liver of experimental animals in the weight-bearing group and the non-weight-bearing group.

Currently, it is known in research conducted by Kawamura (2018) that regular exercise can affect/inhibit the level of oxidative stress caused by acute exercise by increasing antioxidant levels. In fact, several studies have reported that exercise if carried out regularly and in the long term can reduce oxidative stress due to acute exercise. In accordance with the treatment in this study where weight bearing and non-weight bearing exercise treatments were given routinely and regularly 6 days/week within a week, a period of 4 weeks in accordance with the minimum treatment time mentioned in the study conducted by Garatachea (2015). This is thought to be the reason why in this study the liver of experimental animals did not find a significant difference in the degree of inflammation in weight-bearing and non-weight-bearing sports due to the mechanism of antioxidant release.

It is also mentioned in a study conducted by Bejma J. (2000) that the level of oxidant production in the liver is approximately 10 times higher than in other organs where the aging process plays a significant role in increasing oxidant production in the liver under basal conditions. This could also be the reason why there was no significant difference in the degree of inflammation between the weight-bearing and non-weight-bearing groups, because the aging process, which produces high levels of oxidants in the liver, allows long-lasting inflammatory injury to occur. In this study, experimental mice with the age of 7 months were included in the old age, so it was suspected that both groups had parenchymal injury which then caused a bias in the examination results.

It was stated in a study conducted by Chung (2006) that in the aging process, the inflammatory response in the body becomes too reactive and can even cause tissue damage that causes pathological conditions, corroborated by other studies which state that the production of oxidants in the liver is 10 times higher than in other organs. Based on the literature, it could be the reason why the level of inflammation in the liver of mice after being treated with weight-bearing and non-weight-bearing exercise did not have a significant difference.

IV. CONCLUSION

Based on this research it can be concluded that: There was no significant difference in the degree of inflammation in the liver of experimental mice after receiving weight-bearing and non-weight-bearing exercise treatments.

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