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by Deshinta Fitrianti Syahida

Submission date: 19-Apr-2023 09:54PM (UTC+0800)

Submission ID: 2069325386

File name: 16._Differences_in_left_ventricular_wall.pdf (237.81K)

Word count: 3108

Character count: 17127

How to Cite:

Syahida, D. F., Susilo, I., & Wiyasihati, S. I. (2022). Differences in left ventricular wall thickness in aerobic weight-bearing exercise and non-weight-bearing exercise in elderly *Mus Musculus*. *International Journal of Health Sciences*, 6(S6), 1–8.
<https://doi.org/10.53730/ijhs.v6nS6.9151>

Differences in left ventricular wall thickness in aerobic weight-bearing exercise and non-weight-bearing exercise in elderly *Mus Musculus*

Deshinta Fitrianti Syahida

Student of Medical Faculty of Universitas Airlangga, Surabaya, Indonesia

Email: deshinta.fitrianti.syahida-2017@fk.unair.ac.id

Imam Susilo

Anatomical Pathology Department of Medical Faculty of Universitas Airlangga/
Soetomo General Hospital, Surabaya, Indonesia

Email: imam-susilo@fk.unair.ac.id

Sundari Indah Wiyasihati

Department of Medical Physiology and Biochemistry, Medical Faculty, Universitas Airlangga, Surabaya, Indonesia

Email: sundari-i-w@fk.unair.ac.id

Abstract--Due to the process of aging, the function of human organs and muscles decreases. Exercise is a solution to improve organ and muscle function. But it all must be in proportion. So far, aerobic exercise is an exercise solution. This study aims to test the differences between the effects of aerobic weight-bearing exercise and aerobic non-weight-bearing exercise as a sports solution for the elderly. The experimental laboratory used a completely randomized post-test design using 30 *mus musculus* samples with three actions, namely 10 *mus musculus* that were not exercised, 10 *mus musculus* with aerobic weight-bearing exercise and 10 *mus musculus* with aerobic non-weight-bearing exercise. This process is carried out for 6 weeks with a frequency of exercise 5 times a week, and processed statistically using the statistical product and service solution (SPSS) program. There were significant differences between control group and both aerobic weight-bearing exercise and aerobic non-weight-bearing exercise with both p-value being <0.05. However, aerobic weight-bearing exercise and aerobic non-weight-bearing exercise resulted in an enlargement of the ventricle sinistra but there was no significant difference between groups due to the post hoc Mann Whitney test, with p-value >0.05. Furthermore, there were no significant differences between the two groups' weight-bearing and non-weight-bearing aerobic exercise.

Keywords---*Aerobic weight-bearing exercise; Aerobic non-weight-bearing exercise; Old age.*

Introduction

Sport is an activity carried out to train one's body (Arovah, 2016). The purpose of exercise is to improve health and improve physical fitness (Agus, 2012). Aerobic exercise can improve body composition, such as body fat, bone health, muscle mass, and increase endurance, muscle mass and muscle strength, and flexibility so that the elderly are healthier and fitter and reduce the risk of falling (Pribadi, 2015). The benefits of aerobic exercise include prolonging life, healthy lungs, heart, muscles, and bones, making the elderly more independent, preventing obesity, reducing anxiety and depression, and gaining higher self-confidence.

Aerobic exercise can maintain health, but it should be done in the morning, and not exceeding the maximum heart rate, which is between 60-70% of the maximum heart rate (Pribadi, 2015). Aerobic sports include activities that make a person bear his own body weight (weight-bearing), such as walking or activities that do not directly support his own body weight (non-weight-bearing), such as cycling, swimming. Whether or not an exercise program is useful for the elderly also depends on the program being implemented. We recommend that the exercise program that is run must meet the concept of Frequency, Intensity, Time, Type (FITT) (Kurnianto, 2015).

Based on research conducted by Aziz in 2020, the heart rate of athletes is predominantly aerobic and anaerobic with a lower value in predominantly aerobic athletes, thus proving that when athletes do aerobic exercise (endurance), the oxygen demand is high and constant. Then there is an increase in preload and volume which becomes the main stimulus for ventricular adaptation in the form of hypertrophy and an increase in ventricular wall, thereby increasing the effectiveness of heart pumping or stroke volume, which in turn will reduce heart rate in predominantly aerobic athletes compared to anaerobic (Aziz, 2020).

Hypertrophy is divided into two, namely pathological hypertrophy and physiological hypertrophy. However, hypertrophy due to aerobic exercise is a functional physiological hypertrophy characterized by normal heart structure and function (Darsana, 2019). According to Rahima in 2015, in this situation there may also be changes in the levels of Brain Natriuretic Peptide (BNP) in the heart muscle which results in changes in the histological size of the heart muscle (Rahima, 2015). Another thing that causes hypertrophy is the PI3K (P110 α)-Akt, (IGF-1), and insulin signaling pathways (Darsana, 2019). Therefore, to find out more about the effect of which aerobic exercise is better, this study was made to compare the effect of aerobic weight-bearing exercise and aerobic non-weight-bearing exercise. This study was conducted on experimental muscles of the *mus musculus*, to determine the difference in the size of the left ventricular wall thickness in aerobic weight-bearing exercise and non-weight-bearing exercise of the elder *mus musculus*.

Materials and Methods

This research focuses on experimental laboratory utilizing randomized post test design, accomplished in two months January 2021 to February 2021 in the Animal Trials in the Veterinary Faculty of University of Airlangga. Samples used in this research are white female mice (*mus musculus*), 30 of them, divided into 3 groups: Control (K0), first group given aerobic weight-bearing exercise (K1) and second group given aerobic non weight-bearing exercise intervention (K2). At the end of the sixth week, 27 mice are left out of 30 mice, 3 died in the process, one in each group. Data extraction are accomplished by surgery to retrieve mice heart.

Samples are then stained by HE stain and analyzed with type CX 20 microscope to observe the thickness of left heart ventricle. This research has been approved ethically (No. 77/EC/KEPK/2021).

Each and every mice grouped have been given different intervention. First group which is the control unit are just given sustenance *ad libitum* without exercise. Second group are given treadmill exercise with 0o inclination with velocity of 21 cm/second. First session had a duration of 30 minute, frequency 5x/week done in 6 weeks (Souza, 2007). Third group (aerobic non-weight-bearing) mice are given swimming exercise without any weights. Fisrt session had a duration of 30 minutes, 5 times per week done in 6 weeks (Darsana, 2019).

After all process are completed, data extraction is analyzed statistically with statistical product and service solution (SPSS) version 16.0 for Windows. Data extracted are analyzed with descriptive analysis to determine the mean, standard deviation in each group. Data normality are then tested using kolmogorov-smirnov, and homogeneity are testes using Lavene test to determine ANOVA test. If ANOVA test does not meet homogeneity assumption and normality, the alternative test using kruskal wallis non parametric test shall be used. Kruskal Wallis non parametric test gives us a value of $p < 0.05$, which means there is a significant difference between three groups. Post hoc Mann Whitney test are then executed and gives us $p < 0.05$ which means there is significant difference between two groups.

Results and Discussions

These are the histological findings in left ventricle of the mice's heart after interventional methods.

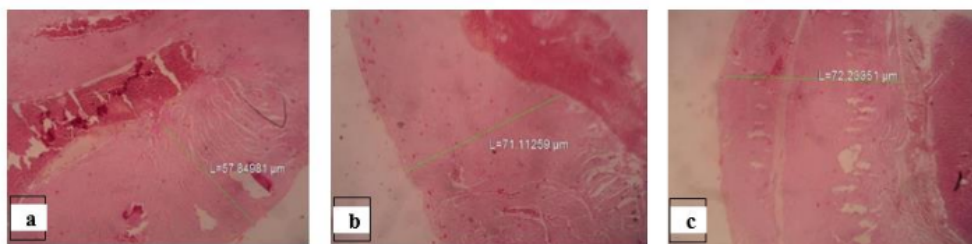


Figure 1. Histological finding of left ventricle hipertrophy. a) Control groups; b) Treatment group 1 (aerobic weight-bearing exercise); c) Treatment group 2 (aerobic non-weight-bearing exercise)

From extracted data, descriptive analysis from left ventricle are established and resulted in data as follows.

Table 1. Characteristics of data in accord of left ventricular in each group.

| Groups | nMyocardium thickness + SD (μm) | mean Minimum thickness (μm) | Maximum thickness (μm) |
|-----------|--|--|-------------------------------------|
| K0 | 965,67 + 3,15 | 57,85 | 68,28 |
| K1 | 970,53 + 0,42 | 69,78 | 71,11 |
| K2 | 971,27 + 1,14 | 69,84 | 72,40 |

Data shows that Left ventricular thickness in *mus musculus* in control groups (without exercise) yields a value of 65.67 μm , standard deviation is about 3.15 μm . minimum value is 57.85 μm and maximum value 68.28 μm .

Left ventricular thickness in *mus musculus* in aerobic weight-bearing exercise yielded data 70.53 μm , standard deviation is as 0.42 μm . Minimal value is as 69.78 μm and maximum value 71.11 μm .

Left ventricular thickness in *mus musculus* in aerobic non weight-bearing exercise yielded 71.27 μm mean value, standard deviation is about 1.14 μm , minimal value is 69.84 μm and maximum value is 72.40 μm .

Table 2. Mean thickness data, normality test, and homogeneity in each group.

| Group | n | Mean + SD (μm) | Normality test (Kolmogorov Smirnov) | Homogeneity test (Lavene Test) |
|-------|---|-----------------------------|-------------------------------------|--------------------------------|
| K0 | 9 | 65,67 + 3,15 | 0,064 | |
| K1 | 9 | 70,53 + 0,42 | 0,074 | 0,030 |
| K2 | 9 | 71,27 + 1,14 | 0,007 | |

Data in three groups are tested for its normality and distribution using Kolmogorov Smirnov method and yielded $p > 0.05$ which means data of left ventricular thickness had normal distribution. Homogeneity variant of Lavene test

yielded $p=0.03$ ($p<0.05$) which means the variant size of left ventricular in each group is not homogenic, thus one-way ANOVA test are unable to be executed. Alternatively Kruskal Wallis test can be executed and it's data is as follows:

Table 3. Results of Kruskal Wallis in left ventricular thickness

| Test Statistics ^{a,b} | Thickness value |
|--------------------------------|-----------------|
| Kruskal-Wallis H | 17.746 |
| Df | 2 |
| Asymp. Sig. | .000 |

a. Kruskal Wallis Test
b. Grouping Variable: Group

Based on table 3, we retrieved p-value of 0.000. Our findings are lower than the significance parameter of 5%, which means H_0 is denied. In conclusion, there are difference in adaptation between three groups of mice with different interventions: control, aerobic weight-bearing exercise, and aerobic non-weight-bearing exercise if we view the left ventricular thickness. Post hoc Mann Whitney test is used to yield the difference of detail, viewed in table 4.

Table 4. Results of post hoc Mann Whitney in left ventricular thickness

| Sample 1-Sample 2 | Test Statistic | Std. Error | Std. t Statistic | Tes Sig. | Adj. Sig. ^a |
|-------------------|----------------|------------|------------------|----------|------------------------|
| K0-K1 | -12.333 | 3.742 | -3.296 | .001 | .003 |
| K0-K2 | -14.667 | 3.742 | -3.920 | .000 | .000 |
| K1-K2 | -2.333 | 3.742 | -.624 | .533 | 1.000 |

From the table output above, it shows that K0 with K1 and K0 with K2 possess difference due to p-value less than the alpha (5%). K1 and K2 intervention does not show difference due to p-value more than alpha (5%). In conclusion, there are difference between control and aerobic weight-bearing exercise and there are difference as well in control and aerobic non-weight-bearing exercise, but there are no difference between aerobic weight-bearing exercise group and aerobic non-weight-bearing group if we analyze the left ventricular thickness on either group.

Effect of aerobic weight-bearing exercise

Mice group with aerobic intervention using treadmill with 15 centimeter/minute for 45 minutes in 8 weeks shows that there are increases of BNP in plasma ($p=0.002$) compared to the before state (Soya, 2007). BNP gene expression causes physiological hypertrophy, which manifests as the thickening of heart muscle and renewal of muscle fibers, acting positively in heart endurance (Maurellet, 2008).

That statement is also backed up by other research that shows after 6 weeks of running, there are light hypertrophy in left ventricle due to physiological response (Phul, 2015). This statement is supported from a study conducted by Opie in 2008 that shows that running could cause eccentric hypertrophy (Opie, 2008). Data yielded through our research concludes that there are significant hypertrophic effects between mice that was in the aerobic weight-bearing group and control, which yielded data of left ventricular hypertrophy of 4.86 μm in which mice that are not exercising is 65.67 μm and mice that are given aerobic weight-bearing exercise is 70.53 μm . This data is supported as well from the post hoc Mann Whitney test that yielded data 0.001 which means there are difference between control and aerobic weight-bearing exercise groups.

Effect of aerobic non-weight-bearing exercise

Aerobic non-weightbearing exercise induced heart muscle adaptation, including hypertrophic effects and renewal of muscle fibers (Medeiros, 2004). In a study using mice as well, intervened by swimming for 4 weeks, there are marked increase in the mass of left ventricle, ratio, weight, and its thickness (Derumeaux, 2008). This statement is supported from a study conducted by Opie in 2008 that shows that swimming could cause eccentric hypertrophy (Opie, 2008). The author, through strenuous data concluded that there are difference between aerobic non-weight-bearing and mice that did not exercise at all, that is 5.6 μm , whereas mice that did not exercise has ventricular thickness of 65.67 μm and mice that are given aerobic non-weight-bearing about 71.27 μm . This data is supported by post hoc Mann Whitney test that yielded 0.000 which means there are difference between control and aerobic non-weight-bearing exercise.

The difference between aerobic weight-bearing exercise and non-weight-bearing exercise

Exercise increases muscle strength, muscle mass, muscle perfusion, and velocity of nerve to muscle (Pribadi, 2015). Aerobic exercise is shown to fix body composition, such as body fat, bone health, muscle mass, and resistance to physical exercise, which means better and healthier heart. Athletes heart rate that are predominantly aerobic and anaerobic with lower score that athletes that are aerobic which shows that when athletes exercise aerobically (endurance) oxygen demand increases (Pribadi, 2015). Preload and volume become stimulus for left ventricular physiological hypertrophy and ventricular wall dilatation such that it increases effectivity in which it increases heart rate in athletes predominantly aerobic compared anaerobic exercise (Aziz, 2020). This is also supported by other studies that shows that after cardio exercise there are light hypertrophy in left ventricle as a physiological response by increasing ventricular diameter (Phul, 2015). In animal trials studies using mice intervened to physical exercise, it shows that there are marked increase in left ventricular mass, ratio of left ventricular mass, and mice weight and the increase of muscle wall relative thickness (Derumeaux, 2008). Based on studies conducted by Yandriyani in 2018, endurance training athletes causes an increase in dynamic load and the heart responses as eccentric hypertrophy or physiological hypertrophy. This could be caused by the increase in cardiac output and stroke volume (Yandriyani, 2018).

Based on post hoc Mann Whitney test there are p-value of 0.533 which can be concluded that there are no difference between aerobic weight-bearing exercise and aerobic non-weight-bearing exercise if we view the thickness of the left ventricular muscle of the test subjects. There are several possibility could factor in this conclusion; similar oxygen demand, similar preload increase, and duration of exercise sessions which are too short.

Conclusion

The conclusion of this study is that aerobic weight-bearing exercise causes changes in left ventricular wall thickness in the *mus musculus* of old age, aerobic non-weight-bearing exercise results in changes in left ventricular wall thickness in the *mus musculus* of old age. However, there was no difference between the aerobic weight-bearing exercise group and the aerobic non-weight-bearing exercise group when viewed from the thickness of the left ventricular wall in the *mus musculus* of old age.

Acknowledgments

The authors would like to thank the Laboratory of Experimental Animals, Faculty of Veterinary Medicine, Universitas Airlangga, Surabaya for providing a place for the research process, the Department of Anatomical Pathology Laboratory of Dr. Soetomo Academic General Hospital Surabaya for helping to provide a place to process research data, our gratitudes to the Universitas Airlangga for the financial aid that were given to us.

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