Effects of a 4-Week Moderate-Intensity Swimming Exercise Increase Serotonin Levels in Mice (Mus musculus)

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Abstract

Serotonin is a neurotransmitter that important in maintaining the integrity of body functions, regulating mood, emotion, sleep, appetite, and regulating memory function. However, changes in serotonin and its receptors are associated with the development of anxiety, depression, and psychiatric disorders, such as schizophrenia. Exercise is suggested as a treatment for several psychiatric disorders. The study aims to prove the effect of moderate-intensity swimming exercise for 4 weeks on increasing serotonin levels in male mice (Mus musculus). The study was a true experiment with research design from a randomized control group of posttest-only designs using 20 male mice (Mus musculus), eight weeks of age, weighing 30-35 grams and randomly divided into two groups, that is CTL (n = 10, control group), MIE (n = 10, moderate-intensity swimming exercise). Moderate-intensity swimming exercise is carried out with a frequency of 3 times/a weeks for 4 weeks. Measurement of serotonin levels using the Enzyme-Linked Immunosorbent Assay (ELISA) method. Data is analyzed using the Independent Samples T-Test with a significant level ($p \leq 0.01$). The results showed a significant difference in serotonin levels between MIE and CTL (100.61±20.08 vs. 57.06±5.13 ng/mL ($p \leq 0.001$)). Based on the results of the study concluded that moderate-intensity swimming exercise increases serotonin levels.

Keyword: Serotonin, moderate-intensity swimming exercise, mental health care

1. Introduction

Serotonin is an important neurotransmitter in emotional processing (Harmer et al., 2008) and memory function regulator in the hippocampus (Haider et al., 2006). It maintains the integrity of body functions and influences the physiological and metabolism of the body (Hassan & Amin, 2011). In addition, it controls mood, emotion, sleep, and appetite, and regulates memory and motivation (Susser et al., 2016). Also regulates several metabolic processes that affect the concentration of glucose in the blood (Longo & Matson, 2014). Several studies have confirmed that serotonin contributes to the development of hyperglycemia, may result of increased levels of adrenal catecholamines, it is caused by the hypoglycemic properties of serotonin (5-hydroxytryptamine; 5-HT) that affect the process of glucose utilization to the peripheral tissue cellular, such as muscle and fat (Kondro et al., 2015). Serotonin and its receptors regulate nearly all brain functions, and the dysregulation of the serotonergic system is involved in the pathogenesis of many psychiatric and neurological disorders (Smith et al., 2020).

A decrease in serotonin levels impacts increases health disorders, such as depression (Hemat-Far et al., 2012), cognitive function decline, and increase anxiety (Pusponegoro, 2007). It also causes a decrease in neuron hippocampus and cortex, neuronal maturation, and impaired formation of dendrites and synapses (Whitaker-

Azmitia, 2001). Decrease in circulating serotonin levels in the brain may lead hyperphagia and weight gain (Lam, et al., 2010), that risk to obesity (Nam et al., 2018). Impaired serotonin and its receptors are associated with the developing anxiety, depression, and psychiatric disorders, such as schizophrenia (Lin et al., 2013). Therefore, it takes the right ways to prevent lower serotonin levels.

Exercise can modulate circulating serotonin levels both in the blood and in the brain, depending on the brain region, and is influenced by the intensity and duration of exercise (Lin et al., 2013). Treadmill exercise performed at moderate intensity for 4 weeks significantly lowers serotonin levels without affecting the metabolism of 5-HT in the hippocampus (Chen et al., 2008). In contrast, seven days of high-intensity treadmill exercise significantly increased serotonin levels in the hippocampus (Chennaoui et al., 2000). Furthermore, moderate-intensity treadmill exercise for 4 weeks did not affect changes in serotonin levels in the amygdala (Chen et al., 2008). The results of a study conducted by Flora et al. (2016), reported that swimming exercises with an intensity of 65%-75% HRmax for 30 minutes significantly increased serotonin levels. Matsunaga et al. (2021) study also reported that swimming exercises for 4 weeks significantly increased serotonin levels in Wistar-type mice. However, how the fundamental effects of exercise intensity and duration on serotonin levels have not been exploited.

Based on before, the purpose of this study was to prove the effects of moderate-intensity swimming exercises performed for 4 weeks on increased serotonin levels in male mice (Mus musculus).

2. Materials and Methods

The study was a true experiment with research design from a randomized control group of posttest-only designs using 20 male mice (Mus musculus), eight weeks of age, weighing 30-35 grams and randomly divided into two groups, that is CTL (n = 10, control group), MIE (n = 10, moderate-intensity swimming exercise). The study was conducted at the Animal Biochemistry Laboratory, Faculty of Medicine, Airlangga University for 5 weeks consisting of one week of acclimatization and four weeks of exercise intervention. Mice are placed in standard conditions (light-dark cycle: 12 h/12 h, temperature: 23 ± 1 °C, humidity: 50 ± 10%) with food and water ad libitum (Daniele et al., 2017). The study follows the principles of animal welfare published by the European Convention for the Protection of Vertebrate Animals. The Health Research Ethics Commission of the Faculty of Medicine, Universitas Airlangga at Surabaya has approved all applied procedures in this study.

Moderate intensity swimming exercise is done with a weight of 5% of total body weight (Guerreiro et al., 2015). Exercise is carried out for 70% of the maximum swimming time with a frequency of 3 times/week for 4 weeks (Matsunaga et al., 2021; Pranoto et al., 2020). Swimming exercises are carried out on a glass tub with a length of 100 x 40 x 60 cm and a water depth level of 30 cm. At the time of moderate-intensity swimming exercise intervention, the water temperature is maintained in the range of 30-32 °C.

Weight measurements are taken before exercise (pretest) and after 4 weeks of exercise (posttest) using the Digital Harnic HL-3650 Heles scale (scale 0-5 kg). A blood sample is collected from the left ventricle of the squeak as much as 1 mL. Blood collection is done 2x24 hours after moderate-intensity swimming exercise intervention for 4 weeks. Blood is checked for 15 minutes at a speed of 3000 rpm. The serum is separated and stored at a temperature of – 80 °C for the analysis of serotonin levels. Measurement of serotonin levels using the Enzyme-Linked Immunosorbent Assay (ELISA).

Data analysis using software statistics packet for social science (SPSS) version 17. Normality tests use the Shapiro-Wilk test. Meanwhile, difference-tests use the Paired Samples T-Test and the Independent Samples

T-Test. All data is displayed with Mean ± Standard Deviation (SD). All statistical analyses use significant levels ($p \leq 0.01$).

3. Results

Based on the results of the study showed that there was an alteration in weight between before and 4 weeks after moderate-intensity swimming exercise in each group that can be seen in Figure 1.

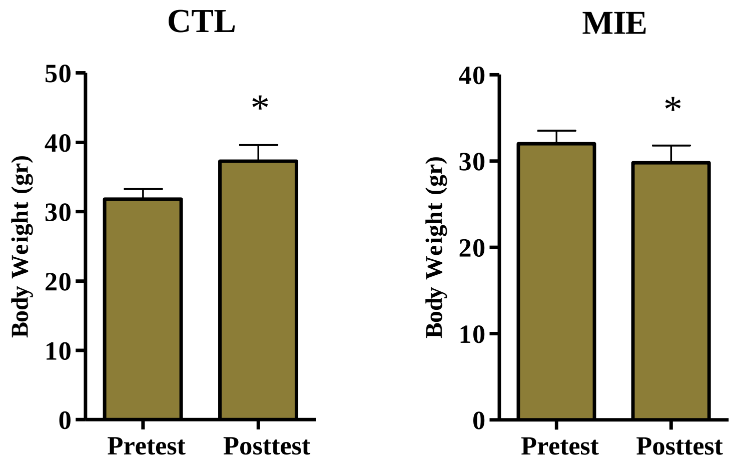

Fig. 1. Weight between before and after intervention in both groups
Note: CTL: Control group; MIE: Moderate-intensity swimming group. (*) Significant vs. Pretest ($p \leq 0.001$)

Based on Fig 1 shows that there is an increase in body weight between pretest and posttest on CTL, while MIE shows a weight loss between pretest and posttest. The results of the Paired Samples T-Test analyst showed a significant difference in weight gain between pretest and posttest at CTL (31.80±1.48 vs. 37.30±2.31 gr ($p \leq 0.001$)), while in MIE showed a significant loss in weight between pretest and posttest (32.00±1.49 vs. 29.80±1.99 gr ($p \leq 0.001$)). The results of an analysis of the serotonin levels between CTL and MIE are presented in Fig 2.

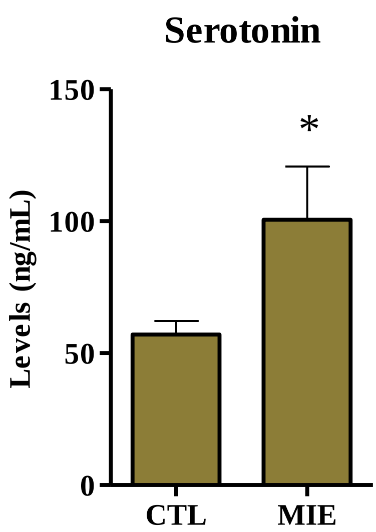

Fig. 2. Serotonin levels between the control group and moderate-intensity swimming exercise
Note: CTL: Control group; MIE: Moderate-intensity swimming group. (*) Significant vs. CTL ($p \leq 0.001$)

Based on the results of the study presented in Figure 2 shows that serotonin levels in MIE are higher compared to CTL. Independent Samples T-Test analysis showed significant differences in serotonin levels between MIE and CTL (100.61±20.08 vs. 57.06±5.13 ng/mL ($p \leq 0.001$)).

4. Discussion

This study aims to prove the effect of moderate-intensity swimming exercise conducted 3 times/week for 4 weeks on the increase in serotonin levels in male mice (Mus musculus). Based on the results showed that serotonin levels in MIE are higher compared to CTL (Figure 2). Independent Samples T-Test analysis shows significant differences in serotonin levels between MIE and CTL. The results are in line with the study by Park et al. (2020) which reported that swimming exercise with an intensity of 5% of body weight significantly increased serotonin expression. Also, research conducted by Furqaani et al. (2018) reported that moderate-intensity aerobic exercise conducted for 4 weeks significantly increased serotonin levels in the brain in male Wistar rats (Rattus norvegicus). A study by Gerin et al. (2008), concluded that exercise significantly increases serotonin levels by 41%. 3 weeks of swimming exercise also significantly increased serotonin levels in Sprague-Dawley-type mice (Dremencov et al., 2017). Daniele et al. (2017), also reported that exercise has been associated with increased serotonin levels. Therefore, the increase in serotonin levels in moderate-intensity swimming exercises is probably due to the effects of exercise. Exercise leads to increased activity of the sympathetic nervous system (SNS) (Clarke et al., 2014). Increased SNS activity stimulates increase epinephrine for the mobilization of fats and proteins (Sugiharto, 2012), increasing the amino acid tryptophan (Clark &Mach, 2016). Increased amino acid tryptophan leads to increased serotonin levels (Boroumand & Hashemy, 2015).

Serotonin is a neurotransmitter synthesized from amino acid tryptophan, that is transported through the blood-brain barrier by serotonin receptors then hydroxylated by the action of tryptophan hydroxylase. Increased levels of amino acid tryptophan in blood circulation support an increase in the concentration of amino acid tryptophan in the central nervous system (CNS), therefore any condition that increases the amino acid tryptophan in the blood circulation will induce an increase in serotonin biosynthesis in the CNS (Strüder et al., 2001). Elevated serotonin levels may play a role in emotion processing (Harmer et al., 2008), the regulation of memory function in the hippocampus (Haider et al., 2006). In addition, increased serotonin levels can also play a role in maintaining the integrity of body functions including regulating metabolism in the body (Hassan &Amin, 2011), regulating food intake and body weight (Lam et al., 2010). Another role of serotonin is as a regulator of mood, emotions, sleep, and appetite, and regulates memory and motivation (Susser et al., 2016). Serotonin also plays a role in regulating several metabolic processes, which can affect the concentration of glucose in the blood (Longo & Matson, 2014). Several confirmed that serotonin may contribute to the development of hyperglycemia, which is most likely the result of increased levels of adrenal catecholamines, which is caused by the hypoglycemic properties of serotonin (5-hydroxytryptamine; 5-HT) can affect the process of glucose utilization by peripheral tissue cells, such as muscle and fat (Kondro et al., 2015). Serotonin and its receptors are also important in the regulation of nearly all brain functions, and dysregulation of the serotonergic system involves in the pathogenesis of many psychiatric and neurological disorders (Smith et al., 2020). Therefore, the results of this study may support the proposition that exercise is effective in relieving anxiety and protecting the brain from wild stress (Greenwood & Fleshner, 2008; Greenwood et al., 2007; Greenwood et al., 2003).

5. Conclusion

Overall, our results showed that moderate-intensity swimming exercise conducted 70% of the maximum swimming time with a frequency of 3 times/week for 4 weeks lost weight and increased serotonin levels.

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