Hypoxic Swimming Exercise Increases Catalase Enzyme Activity in Trained Swimmers

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Abstract. Swimming is a carried out activity, out excessively causes the body's level of free radicals to rise. If the produced free radicals are not addressed, they will be harmful to the body. The body produces antioxidants in the form of the enzyme catalase as one of its defenses against free radicals. An enzyme called catalase may decompose hydrogen peroxide (H2O2) molecules into water and oxygen (O2). This research aimed to ascertain how swimming action affects the catalase enzyme's activity. 10 pupils who were tested for their ability to swim sprints provided samples at To distinguish between groups of trained and inexperienced swimmers, a 200 m distance was covered in 4 min. By using the spectrophotometric method to measure the catalase enzyme activity, the results revealed that the difference between the trained and untrained groups was greater for the catalase enzyme activity group and that swimming exercise had an effect on the catalase enzyme activity (p 0.05).

Keywords: Hypoxic Swimming Exercise · Catalase enzyme · hydrogen peroxide · free radicals

1 Introduction

Sport is an important activity to maintain health, one of which is swimming. Swimming is a sport without the force of gravity (*Non-Weight Barring*) which involves all the muscles of the body moving which triggers blood flow to the heart and breathing. Swimming is categorized as aerobic physical exercise [1]. Swimming activities that are carried out as fast as possible (*sprint*) to move and move places in a short time are known as anaerobic physical exercises. In this condition, energy is obtained from the phosphagen ATP-PC and lactic acid, followed by the use of O_2 [2].

The physical exercise aims to maintain and improve physical fitness [3]. However, excessive physical exercise will cause damage to body cells, including hypoxia. Hypoxia is often experienced by athletes during training, one of which is swimming training [4]. Hypoxia can cause an increase in free radicals and *Reactive Oxygen Species* (ROS) which are reactive to cell membranes [5]. The hypoxic state will result in an adaptation

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response to maintain the body's homeostasis to ensure the availability of oxygen in the brain [6]. However, hypoxia that occurs chronically can increase the production of free radicals in the body to increase and suppress enzyme action [7].

Free radicals are molecules with unpaired and unstable electrons that are present in the body [8]. Free radicals can exist in the body due to the side effects of the process of oxidation and cell metabolism, including the burning of cells during breathing, sports or engaging in exercise that is maximal or excessive, inflammation, or exposure to pollution outside the body. There are quite a lot of free radicals, but one of the most abundant in the body is *reactive oxygen species* (ROS) [9].

Increased levels of ROS in the body will cause damage to lipids, proteins, and DNA. One of the triggers for causes of chronic disease and inflammation is the occurrence of oxidative stress, the body's defense system will inhibit the process of free radical formation when there is an imbalance between the production of oxidants and antioxidants in the body [10].

Antioxidants are compounds that can overcome oxidative damage caused through free radicals [11]. Antioxidants are classified according to the source into endogenous and exogenous antioxidants. Exogenous antioxidants come from sources external to the body. Such as from food and beverages, while endogenous antioxidants come from within such as superoxide Dismutase in the body, *Glutathione Catalase (CAT) and Peroxidase (Gpx)* [12].

Catalase enzyme isone of the enzymes that is efficient in breaking down or converting O2 and H2O2 are formed. One of the free radicals that might lead to damage is H2O2. Oxidative stress. One catalase molecule can convert millions of H_2O_2 to H_2O and O_2 in seconds [13]. Halliwell's research reports that at high H_2O_2 concentrations the catalase enzyme works so it can be concluded that in the first hypoxia conditions there was high H_2O_2 production by MnSOD [14]. Changes in catalase activity occur due to exposure to hypoxia as a prooxidant and the highest catalase activity occurs in the blood [15] As previously explained, the catalase enzyme is known to have the ability to ward off free radicals that occur in the body after physical activity. However, in sprint swimming training activities, there is no known effect on catalase activity in humans. For this reason, a study will be conducted with the title of In trained swimmers, hypoxic swimming exercise boosts catalase enzyme activity.

2 Method and Materials

2.1 Materials

The components were an alcohol pad, student blood samples (5 ml), h2o2, pbs (phosphate buffered saline), aquades.

The tools used were a spectrophotometer, centrifuge, autoclave, beaker glass, sample storage refrigerator, icebox, micropipette, tips (10 μL, 200 μL, and 1000 μL), 1.5 mL microtube, vacutainer, syringe.

2.2 Methods

2.2.1 Sample

Sampling was obtained from 10 students who took swimming specialization courses which were then divided into two groups based on swimming speed (sprint) with a distance of 200 m for 4 min. Screening results were obtained from both the trained and untrained groups.

2.2.2 Examination of Catalase Enzyme Activity

The catalase activity was examined using the Mates method. The instrument used was a UV-vis spectrophotometer with a wavelength of 210nm. The examination was carried out by mixing 975 μL of H_2O_2 reagent and 25 μL of plasma. After mixing the reaction will run, t_0 is 30 s after mixing, t_1 is 1 min after t_0.

2.2.3 Measurement of Catalase Enzyme Activity

Measuring the activity of the enzyme catalase is carried out with the formula:

$$Catalase\ activity\left(\frac{U}{\mu L}\right) = \frac{(\Delta s - \Delta b)\Delta t}{[H2O2] \times Vsample} \times Dilution\ factor$$

2.2.4 Data Analysis

The difference in catalase enzyme activity between the trained and untrained groups was carried out by using the independent sample T test with a significant difference of $p < 0.05$. All data were analyzed using the SPSS Version 25 application.

3 Result and Discussion

Based on the results of the research conducted, it was found that there were differences in the activity of the catalase enzyme in the treatment group between the untrained and trained swimming groups, as described in Table 1 and Fig. 1.

Based on Table 1 and Fig. 1, shows that there is a difference in the average value of catalase enzyme activity in the untrained and amateur swimming teams. The average activity of the enzyme catalase in the trained group was 0.249 unit/μL and the trained group was 0.676 unit/μL. This indicated a difference in catalase enzyme activity between the untrained and untrained swimming groups. Furthermore, the data were analyzed by using the independent sample T-test to see the effect effects of swimming on the catalase enzyme's activity of each treatment group. After the test was carried out, significant results were obtained at ($p < 0.05$), so it can be concluded that the swimming activity had an impact on the catalase enzyme activity.

Specifically, swimming is a sport that demands more complex muscle performance [10]. Therefore, it is necessary to do repetitive and structured physical exercises [3]. When the body does physical activity such as swimming, it responds at a systemic and

Table 1. Catalase Enzyme Activity

Group	Sample	Catalase Enzyme Activity
Untrained	A3	0.224
	A6	0.241
	A8	0.257
	A9	0.269
	A10	0.252
Trained	A1	0.610
	A2	0.622
	A4	0.550
	A5	0.612
	A7	0.644

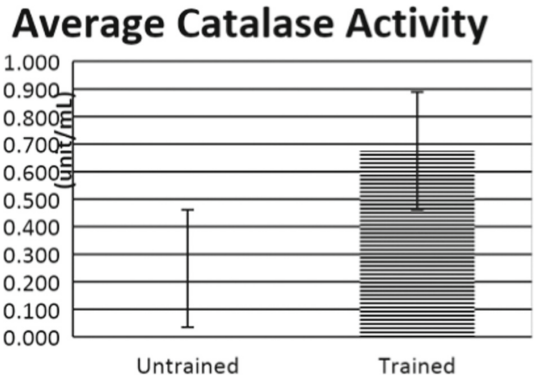

Fig. 1. Average catalase enzyme activity

cellular level [16]. When swimming, the body will need more O_2 troops in the tissue because of an increase in the body's free radical count. According to parwata a free radicals are molecules, atoms that have 1 or more unpaired electrons in their outer shell so they are very reactive to the body [9]. Based on research Thirumalai showed results that intense and excessive swimming exercise can increase the production of ROS in muscles [17] and according to Lima swimming that is done until fatigue also causes an increase in ROS in the liver [18].

The human body has several mechanisms to defend against free radicals, one of which is the presence of antioxidants. Antioxidants produced by the body are known as endogenous antioxidants which can be in the form of enzymes and one of them is the catalase enzyme [11]. Catalase functions as a catalyst enzyme for thedecomposition reaction using hydrogen peroxide (H_2O_2). Hydrogen peroxide can diffuse into and penetrate cell membranes so that it can cause damage to cells. Hydrogen peroxide in the body

comes from the process of electron transport in the mitochondria which reduces O_2 by accepting electrons and accepting electrons and the O_2^- dismutase reaction catalyzed by superoxide dismutase.

When swimming sprints, the catalase enzyme activity will increase to ward off body's presence of free radicals. As one of the radicals freeprevents enzymes, the catalase enzyme activity will increase when oxidative stress occurs. In Fig. 1, various catalase enzyme activities in the trained a group of much greater than within the uneducated group. This is consistent with the study of Clarkson and Thomson which stated that in trained subjects the levels of erythrocyte enzyme activity (superoxide dismutase, glutathione peroxidase, and catalase) were higher than in untrained subjects. Irregular exercise increases free radicals that are greater than antioxidants, resulting in oxidative stress [19]. The higher the intensity of physical exercise, the higher the body's metabolic rate and the more it shifts to the anaerobic metabolism area and increases lactic acid level [20]. This is in accordance with Syahrastani's research which states that exercise intensity affects cellular adaptation to free radicals [16].

4 Conclusion

Swimming activity has an impact on the activity of the catalase enzyme.

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