Hepatoprotective effects of beetroot juice at maximum physical activity

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Abstract. The purpose of this study was to determine the protective effects of beetroot juice treatment on hepatic oxidative stress at maximum physical activity. This type of research is an experimental study with a randomized control group pretest-posttest design research. The study was conducted at the Universitas Negeri Medan Stadium and Universitas Sumatera Utara Faculty of Medicine Laboratory. The sample was 30 students of Sports Science who met the criteria. The Pretest is done by checking the levels of alanine aminotransferase, aspartate aminotransferase, and malondialdehyde. Furthermore the sample was divided into 2 groups (P1 = 15, P2 = 15). During the 30-day training program, the P1 group was given 250 ml beetroot juice once a day, whereas P2 was given as a control. Then all samples performed a maximal physical activity using a bleep test, and again examined levels of alanine aminotransferase, aspartate aminotransferase, and malondialdehyde. The results showed beetroot juice could reduce levels of malondialdehyde, alanine transaminase, and aspartate transaminase at maximum physical activity.

1. Introduction

Regular physical exercise will have beneficial effects in the prevention of various diseases such as diabetes mellitus, hypertension, cancer, obesity, osteoporosis, and premature death [1]. Besides having a positive impact on the body, physical exercise also has a negative impact, especially heavy physical activity. Heavy physical activity, as well as exhausting exercise, can increase oxidative stress, which causes an imbalance between the body's oxidation system and antioxidant enzymes. Increased free radicals triggered by strenuous physical activity can cause damage to many parts of the cell such as protein, DNA, and cell membranes by stealing their electrons through a process called oxidation [2].

During maximum physical activity, some organs such as liver, kidneys, and other organs will experience hypoxia and ischemia because of the higher amount of oxygen consumption in the working muscles. After a physical activity is complete, blood flow will return to normal through the reperfusion process. In the reperfusion process, free radicals are produced, which will damage the cell membrane through the reaction of lipid peroxidation [3]. The formation of radicals in the liver causes deep fat peroxidation membrane inside the cell. Some research results indicate heavy physical activity can cause an increase in malondialdehyde (MDA) levels in the liver [4], a decrease in enzymatic antioxidant levels in liver tissue [4,5] which results in liver damage characterized by increased levels of alanine transaminase (ALT) and aspartate transaminase (AST) [6-10].

In the human body there are endogenous antioxidants that protect the body from the influence of free radicals. Free radicals formed during physical activity will be neutralized by the elaboration between the detained system between antioxidant enzymes and a number of non-enzyme antioxidants [10,11].

One alternative natural ingredient that has antioxidant content is red beet (Beta vulgaris L). Red beet is known to have betalain content which is a compound that has a very high antioxidant that is able to neutralize free radicals [12]. Other compounds in red beets (Beta vulgaris L) that function as antioxidants are betain [13], vitamin C, carotenoids, phenolic acids such as and flavonoids [14,15,16]. Phenolic acid compounds in red beets include ferulic acid, caffeic acid, p-coumaric acid, syringic acid, and vanillic acid [17]. Strong antioxidant power and many types of antioxidants in beetroot so that it needs to be investigated the effect of beetrooting during exercise on ALT and AST levels when performing maximum physical activity.

2. Methods

2.1. *Subjects*

Research subjects were 30 people with criteria of having a good VO2max level, male sex, age 20-22 years, having a good Body Mass Index (BMI), not smoking, not taking supplements and antioxidants two weeks before and during the study, willing to be the subject of research. Before testing, subjects gave their written informed consent to participate after the experimental procedures, associated risks, and the benefits of participation were explained.

2.2. *Study design*

The study used 30 students who met the criteria. All athletes performed hematological examinations to measure ALT and AST levels. Then the subjects were divided into two groups (P1 = 15, P2 = 15). During the 30-day training program, the P1 group was given one-day beetroot juice at a 300 ml dose two hours before undergoing the exercise program, while the P2 group was given a placebo. Then all athletes perform the maximum physical activity by doing a bleep test. Furthermore, hematologic measurements were performed again to measure ALT and AST levels.

2.3. *Blood Samples Collection.*

Blood samples (5 ml) were collected from the subject's forearms at 09.00 (during fasting) after the subject had maximum physical activity. Samples were centrifuged in 3000 rotations for 10 minutes to separate the serum which was then stored at -80°C until analysis.

2.4. *Biochemical Analyses.*

Serum MDA analysis was Enzyme-linked immunosorbent assay (ELISA) colorimetric method conducted by were analyzed by using MDA assay kit according to the manufacturer's instruction. The results were expressed as nmol/ml. The level of ALT and AST were measured by spectrophotometer.

2.5. *Statistical Analyses*

All data analysis was conducted using SPSS 19 software. All data were analyzed using paired two-tailed t-test and unpaired two-tailed t-test.

3. Result

3.1. *The Effect of Giving Beetroot Juice on Alanine Transaminase and Aspartate Transaminase Levels*

The results of the study the effect of beet juice administration during exercise on AST and ALT levels in maximum physical activity are shown in Figures 1 and 2. Figure 1 showed the AST levels of the control group and the experimental group increased significantly (p = 0,000). The control group increased by 16.74%, while the experimental group increased by 6.33%. Statistical test results using the independent t-test showed that there were significant differences between the control group and the experimental group AST levels post-test (p = 0,000).

Figure 1. Pre and Post-Test Change of AST (mean±SD)

Figure 2 showed the ALT levels of the control group and the experimental group increased significantly (p = 0,000). The control group increased by 13.77 %, while the experimental group increased by 7.69 %. Statistical test results using the independent t-test showed that there were significant differences between the control group and the experimental group ALT levels post-test (p = 0,000).

Figure 2. Pre and Post-Test Change of ALT (mean±SD)

3.2. *The Effect of Giving Beetroot Juice on on malondialdehyde (MDA) level*

Figure 3 showed the MDA levels of the control group and the experimental group increased significantly (p = 0,000). Statistical test results using the independent t-test showed that there were significant differences between the control group and the experimental group MDA levels post-test (p = 0,000).

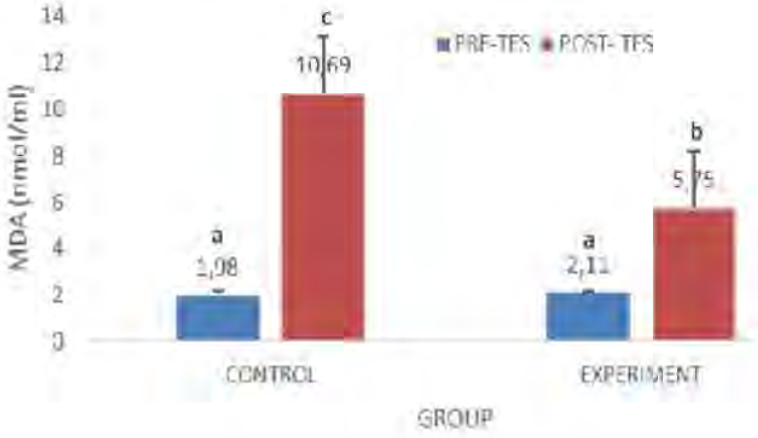

Figure 3. Pre and Post-Test Change of MDA (mean±SD)

4. Discussion

Liver is the largest organ in the human body and key organ of metabolism, including glycogen storage, decomposition of red blood cells, plasma protein synthesis, and detoxification. The liver plays a role in sports physiology. Physical exercise increases blood flow in working skeletal muscle, meanwhile blood flowing to the liver [18] and venous portal will decrease [19]. During high intensity exercise, hepatic microcirculation is regulated by endothelin-1. Then, the decrease in blood to the liver often causes liver damage [20] which is characterized by an increase in AST and ALT levels [21].

As shown in Figure 1 and 2 showed the AST and ALT levels of the experimental group were significantly lower than the control group ($p<0.05$). Decreasing AST and ALT levels in the experimental group in this study due to antioxidant activity and antioxidant content in beetroot juice can reduce MDA levels. It has been known that beetroot juice has a betalain content, which is a compound that has a very high antioxidant that is able to neutralize free radicals [12]. Other compounds in red beets (Beta vulgaris L) that function as antioxidants are betain [13], vitamin C, carotenoids, phenolic acids such as and flavonoids [14,15,16]. Phenolic acid compounds in red beets include ferulic acid, caffeic acid, p-coumaric acid, syringic acid, and vanillic acid [17]. Betain was found to exert its antioxidant activity via two mechanisms. One mechanism involves scavenging Reactive Oxygen Species (ROS) in cells via up-regulation of endogenous nonenzymatic antioxidant defenses. The other inhibits ROS generation by isolating cells from the oxidative stress inducer [22]. Phenolic compounds also increase the activity of antioxidant enzymes, thus indirectly affecting the concentration of harmful oxygen radicals in the living cell. The results were in agreement with Sadeek, [23] who reported that, treatment with beetroot (oral dose of 8ml/kg/day/rat) for 28 days significanly ($p<0.05$) restored the enzyme activities of the liver AST, ALT and ALP to normal level.

Maximum physical activity increases ROS production, which consequently attacks membrane lipids and results in the formation of lipid peroxidation products. Increased lipid peroxidation after complete exercise has been noted in several studies [24]. Malondialdehyde (MDA), one of the end products of polyunsaturated fatty acid peroxidation, has been widely investigated in exercise studies as a marker of oxidative stress [25]. As shown in Figure. 3, MDA content from the experimental group was significantly lower than the control group ($p <0.05$). Lower MDA levels in the experimental group compared to the control group were due to antioxidant content and the ability to scavenge free radicals in beetroot juice . MDA content from the experimental group was significantly lower than the control group ($p <0.05$). The lower MDA level in the experimental group compared to the control group was due to antioxidant content and the ability to take free radicals in beetroot juice. The results of this study are supported by research conducted by Olumese and Oboh [26] who reported

administration of beetroot juice can reduce MDA levels due to the administration of carbon tetrachloride (CCl4). These results indicate that beet juice effectively reduces lipid peroxidation due to increased free radicals at maximum physical activity

5. Conclusion

In conclusion, maximum physical activity is caused changes in liver function and pathology characterized by increased levels of AST, ALT and MDA. Beetroot juice supplementation during the exercise program can reduce AST, ALT and MDA levels at maximum physical activity

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