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Changes in Liver Enzymes After the Implementation of Astrand and RAST Tests in Overweight Individuals

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ABSTRACT

Background & Objective: Overweight people are more vulnerable to developing fatty liver disease, moreover; they are more likely to have increased serum liver enzymes and lactate dehydrogenase than others. The aim of this study was to evaluate the changes in liver enzymes and *lactate* dehydrogenase after doing exhaustive aerobic and anaerobic exercises in active overweight male students.

Materials & Methods: In this semi-experimental study, 45 overweight male students were randomly divided into aerobic (n=15), anaerobic (n=15) and control (n=15) groups. In aerobic group: first, a study was performed on the Astrand aerobic test (including running on a barrel at speeds of 5 to 8 miles per hour with a gradient of 3 minutes, and after 3 minutes, every 2 minutes, a steep gradient of 2.5%, a constant speed, and a fatigue), the subjects of the anaerobic group performed the Rast anaerobic test (including 6 fast-paced repetitions at 35 meters distance and maximum severity, with a rest interval of 10 seconds in each repetition). The control group was without any intervention. The data were analyzed using one-way ANOVA and Bonferroni post hoc test.

Results: The results indicated a noteworthy increase in ALP and LDH values of the three groups in the aerobic and anaerobic exercise groups (P=0.3, P=0.6). However, although AST and ALT were increased by aerobic and anaerobic exercise, there was no significant difference between the three groups (P=0.02) (P=0.01).

Conclusion: There was an evident increase in the serum levels of ALP, AST, ALT and LDH as an indicator of liver damage in overweight people, which are more vulnerable to fatty liver disease. Therefore, according to the achieved results, aerobic exhausting exercise could be a more appropriate exercise for the overweight.

Keywords: Aerobic and Anaerobic exercise, Liver enzymes, LDH, Overweight

Introduction

In previous years, the effect of exercise on different body systems has been examined and its positive effects on the heart and breathing apparatus, nerve, bone, and muscle have been demonstrated **(1)**. Studies show that intentional and sudden blows to tissues can lead to impaired activity of plasma enzymes. Some studies have confirmed the association of muscle damage with the release of muscle enzymes **(1)**. Researchers demonstrated that aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP), and lactate dehydrogenase (LDH) are vital enzymes and indicate muscle damage **(2)**. The liver is one of the body's major organs and through various enzymes regulates hormones and metabolic activities of the body at rest, during exercise and recovery **(2,3)**. In normal circumstances, the liver and kidneys, respectively receive 27 and 22% of the amount of circulating blood, but blood flow to the liver and kidneys is reduced 5% and 3% as a result of a heavy exercise, respectively **(2)**. Long-term reduction in blood flow to the liver and kidneys may cause adverse consequences, including submaximal fatigue caused by ongoing activity. The liver is the most sensitive tissue to oxidative stress induced by exercise **(3)**. The plasma activity of the liver enzymes is influenced by the duration, intensity, type and manner of the exercise **(3,4)**. Panu Praphatsorn *et al.* (2010) studied the effect of running on a treadmill on an incline on serum levels of ALT and AST in rats of Sprague – Dawley, and the levels of both enzymes significantly increased immediately after the exercise **(5)**. Togashi *et al.* (2010), and Kim *et al.* (2008) have reported in human subjects and in some cases results overlapped and in some cases did not **(6,7)**. However, the best clinical evaluation of the liver is through examining the activity of liver enzymes, especially alanine aminotransferase (ALT), aspartate Alanine8 transaminase (AST), alkaline phosphatase (ALP) and lactate dehydrogenase (LDH), because if a liver cell is damaged transaminases increase in the serum **(4)**. AST and ALT are enzymes that catalyze the transfer of amino groups from aspartate and alanine to alpha-ketoglutarate. These specific enzymes are in the heart and kidney and the concentration of AST is more than ALT in the liver (7). Some studies have revealed that the high levels of liver enzymes like ALT, AST, ALP are associated with non-alcoholic fatty liver disease (NAFLD) (4,5). Therefore, physical activity is one of the preventive strategies, which reduces the risk of diseases such as non-alcoholic fatty liver, and diabetes, and its importance is getting more obvious every day by day (4). Saengsirisuwan *et al.*, Mashiko *et al.* and Clarkeon reported a significant increase in the levels of liver enzymes and LDH after exercise and sports competitions (8,9). Meanwhile, in their study Matsus *et al.* did not confirm a significant variation in these enzymes after a session of activity, and their results are not consistent with the results of the present study (10). Possible reasons for the contradiction between the previous research findings and the results of this study can be attributed to the individual differences as well as the type and duration of the physical activity.

Since 1990, due to the rapid changes that have been made in the Iranian diet, and due to the reduction in physical activity, a rapid increase in the body mass index, has been observed in Iran. With regard to the findings, the prevalence of fatty liver in the community is associated with the prevalence of obesity (4). The present study aimed to answer the fundamental question "does exhaustive training affect liver enzymes and lactate dehydrogenase in overweight people?”.

Materials and Methods

Firstly, the active overweight students of Shahid Chamran University of Ahvaz in 2015, who had a history of regular sports activities on average of one hour, three days a week were registered and were selected based on the sample size formula[1] (11). Inclusion criteria of the study included: the age range of 23-25 years, non-smoking, lack of insulin injection, lack of cardiovascular disease, hypertension, respiratory and musculoskeletal diseases and no history of recurrent hypoglycemia during exercise. After determining the maximum oxygen uptake and body composition assessment, 45 subjects were selected for the study. Participants were randomly divided into three groups: aerobic, anaerobic and control group. A week before the start of the experiment, the subjects completed a questionnaire about their health and performed Strand Test and Rast test to learn more (12,13). Also, participants were asked to avoid using black tea, juice, any pills or supplements and severe physical activity during the study period. The intensity of the exercises was controlled by a percentage of the maximum heart rate and using the Pulbar's pulse rate monitor. The maximum heart rate of subjects was calculated using the Karunen[2] equation for each person. Their VO2max was calculated using the Rocket Test. At all stages of the aerobic exercise Strand, the intensity of training was between 85-95% of maximum heart rate, which was calculated for each participant individually using a pulsar (Polar model made in Finland) that was installed on the subjects' chest area. At all stages of the Strand aerobic exercise, the intensity of training was between 85-95% of maximum heart rate, which was calculated for each participant individually. Descriptive statistics were used to determine the mean and standard deviation of each variable and the Shapiro Wilk was used to determine the normal distribution of data. According to the Shapiro Wilk test, the variables of height, weight, body mass index and maximum oxygen consumption were of normal distribution.

Immediately before and after a training session, 5mL blood samples from the brachial vein were taken from the subjects, at the time of fasting (to reduce heart rate), and then soft kinetic movements were performed. The serums were separated immediately by centrifugation with 3000 RPM for 15 minutes, and the samples were held in -20°C temperature until examination day. In the current study, aspartate aminotransferase, alkaline aminotransferase, alanine aminotransferase and lactate dehydrogenase levels were measured by ELISA and biochemical methods using Pars Azmoon kits and by GBC Australia's spectrophotometric device.

Subjects did warm-up exercises before and after the session that included 5 to 7 minutes of stretching and a soft movement and cool-down activities including 2 minutes walking slowly to reduce the heart rate and then soft stretching. At first, the aerobic training group was asked to perform the Astrand test. The Astrand test involves running on a treadmill at a speed of 5 to 8 miles per hour with zero incline for 3 minutes. After 3 minutes, the excess was 2.5% icline every 2 minutes and speed remains constant and the activity continued until the person reaches exhaustion (Table 1). Then the anaerobic group performed the RAST test. The RAST test consists of six quick repeated runs at a distance of 35 meters with maximum intensity with rest intervals of 10 seconds between each iteration (Table 2). Before starting the test, subjects warmed up for 5 minutes and records were recorded with photocells FTB-500 optical system (photocell) EXFO company in which the two pairs of photocells located near the start point and after 35 meters and the subjects stand in each iteration at a distance of 70 cm from the starting line and started to run full swing by hearing the sound of the machine. After passing through the optical sight, the machine stops and timer record was recorded. In order to remove the reaction time, the device was in adjustment mode so that the timer began to work after passing the first optical sight. The control group continued their daily activities without interference.

[1] *n=[(SD12 +SD22)×(Z1-a/2 +Z1-b)2]/D2*
[2] Maximum Heart Rate = 220-age

Table 1. **Strand test**

levels	Slope	Speed/miles	Km	m
Step 1	10	1.7	2.7	45
Step 2	12	2.5	4	47
Step 3	14	3.4	5.5	92
Step 4	16	4.2	6.8	113
Step 5	18	5	8	133
Step 6	20	5.5	8.8	147
Step 7	22	6	96	160

Table 2. **Rast test**

The distance went (m)	Return distance(m)	Total distance(m)	Repeat	Rest time between each repetition (s)
35	35	210	6	10

For intragroup changes, T-test was used, and one-way variance analysis and Bonferroni post hoc test were used to compare the groups. Data analysis was performed using SPSS software (SPSS Inc., Chicago, Ill., USA).

Results

The characteristics of the studied subjects are presented in Table 3. As specified in the table, they did not differ significantly both in terms of physical characteristics and physical fitness. In Table 4, the mean values of enzymes ALP, AST, ALT and LDH of the three groups are presented as a result of the exhaustive aerobic exercise which the results of the analysis of T-test showed that there was a significant increase in the average of ALP, AST, ALT and LDH enzymes in aerobic groups in the pre-test and post-test as an effect of the aerobic exercise. Considering AST and ALT levels of the anaerobic group in pre-test and post-test despite increasing, there was no significant change by the anaerobic training. The values of ALP and LDH in the anaerobic group showed a significant increase in pre-test and post-test after anaerobic training. In addition, based on the results of covariance analysis, the values of ALP and LDH of the three groups showed a significant increase in the aerobic and the anaerobic exercise sessions. But AST and ALT values between the three groups, despite the increase was not significant at the level by performing an aerobic and anaerobic exercise session. Also, the results of the Bonferron hunting test are presented in Table 5. Based on this test, the levels of AST and ALT were determined; the groups of the aerobic-control, the anaerobic-control and the aerobic-anaerobic groups in post-test had no significant difference in post-test. Followed by ALP and LDH values of aerobic-control groups, the anaerobic-control and the aerobic-anaerobic groups showed a significant difference in post-test.

Table 3. **Physical and anthropometric characteristics of subjects**

Characteristics of the subjects studied	Group			P-value
	Aerobic	anaerobic	Control	
Age (year)	23.58±0.22	25.18±2.22	24.18±0.3	0.78
Height (cm)	173.96±1.55	172.11±2.41	173.45±1.1	0.86
Weight (kg)	78.42±6.76	76.97±8.33	78.42±6.76	0.63
BMI(kg/m^2)	26.51±0.96	26.03±1.32	25.14±0.83	0.67
$VO2_{MAX}$(Ml / kg / min)	45.05±2.03	45.74±3.11	45.35±1.46	0.73

Table 4. **The changes of ALT, ALP, AST and LDH enzymes in each group due to aerobic and anaerobic activity**

Index	*Group*	*Pre-test*	*Post-test*	*P(inside the group)**	*P (between groups) **
ALT (IU)	aerobic	4.91±1.88	6.25±2.22	0.02	0.3
	anaerobic	4.33±2.34	7.66±2.05	0.04	
	Control	5.2 ± 2.2	5.3 ± 4.4	0.2	
AST (IU)	aerobic	17.25±5.2	18.83±6.71	0.05	0.6
	anaerobic	16.93±1.4	19.08±2.96	0.03	

Index	*Group*	*Pre-test*	*Post-test*	*P(inside the group)**	*P (between groups) **
	Control	15.3 ± 0.6	16.8 ± 0.8	0.1	
ALP (IU)	aerobic	145.33±36.68	153.97±41.79	0.02	0.02
	anaerobic	136.47±25.18	155.83±25.81	0.01	
	Control	148.5 ± 12.7	148.8 ± 19.3	0.06	
LDH (IU)	aerobic	139.55±3.56	140.2±3.95	0.01	0.01
	anaerobic	139.24±3.67	148.7±4.56	0.001	
	Control	139.3 ± 4.5	140.5 ± 5.7	0.07	

* Significant level ($P \leq 0.05$). T-test was used to check the intra-group variation and one-way ANOVA test was used for comparison between groups.

Table 5. Bonferroni post hoc test results between aerobic-anaerobic groups

Index	*Group*	x±Š	*P (between groups) **
ALT (IU)	Aerobic- Anaerobic	6.95±3.8	0. 3
	Aerobic- Control	5.91±1.28	0. 2
	Anaerobic- Control	5.03±2.31	0. 4
AST (IU)	Aerobic- Anaerobic	18.94±2.01	0. 3
	Aerobic- Control	17.99±2.2	0. 5
	Anaerobic- Control	16.55±1.07	0. 3
ALP (IU)	Aerobic- Anaerobic	154.07±3.05	0.001
	Aerobic- Control	150.29±38.41	0.02
	Anaerobic- Control	150.17±23.12	0.01
LDH (IU)	Aerobic- Anaerobic	144.01±6.87	0.01
	Aerobic- Control	139.50±3.13	0.01
	Anaerobic- Control	143.22±7.65	0.001

* Significant level ($P \leq 0.05$).

Discussion

The aim of this study was to evaluate the changes in serum liver enzymes and lactate dehydrogenase after the exhaustive aerobic and anaerobic exercises in overweight active boy students. The results of this study showed that exhaustive exercise activity significantly increased the levels of ALP and LDH in the aerobic-control. The aim of this study is to evaluate changes in liver enzymes and lactate dehydrogenase after exhaustive aerobic and anaerobic exercise in overweight active boy students. The results of this study showed that the exhaustive exercise activity significantly increased the levels of ALP and LDH in the aerobic-control, the anaerobic-control and the aerobic-anaerobic groups. Also, there was no significant difference between the AST and ALT levels of aerobic-control, anaerobic-control and aerobic-anaerobic groups in the post-test.

Most studies have proven that, most overweight people are more prone to fatty liver as well as the likely increase in liver enzymes and lactate dehydrogenase activity due to exhaustion than other people. These enzymes are distributed in many tissues of the body and have a higher concentration than that in the liver and are more considered as liver transaminase. If the tissue is damaged, the amount of these enzymes also increases **(10)**. Losing weight can be achieved through the exercise and a diet plan, which can lead to a significant improvement in serum enzymes and liver histology of patients with NAFLD **(3)**. Studies have shown that weight loss can reduce average BMI and serum ALT levels and reduce the liver fat refining and necrosis inflammation **(3)**. The results of research in this area are in some ways similar and in some ways contradictory. Younesian *et al.* (2014) also studied 2028 high school students and found that here is a significant relationship between the liver enzymes levels with weight, BMI and waist to hip ratio (WHR) **(14)**. Devaki and colleagues (2010), studied adult male rats and made them perform forced swimming for 15 minutes to 4 hours and observed no significant increase in serum levels of ALT and AST (15). The reason for antithetic results with the study of Devaki can be viewed on the type and duration of the exercise. The findings of the present study are based on the increase of liver enzymes and serum lactate dehydrogenase immediately after an aerobic and maximal anaerobic digestion activity meeting the results of Pantoja *et al.* (2009) and Pettersson *et al.* (2007) (16,17). The Pantoja research group showed a significant increase in alkaline phosphatase and plasma lactate dehydrogenase enzymes immediately after activity and examining the indices of hepatic enzymes in nine healthy men after three bending and opening movements of the elbows with a maximum intensity of 10 repetitions (16). Pettersson *et al.* (2007) also stated that an anaerobic weightlifting session at the age of 15 led to an increase in all liver enzymes for seven days after exercise (17). Eskendari *et al.* reported that serum AST and ALT increased after a 200-meter continuous run. The results of this research are consistent with the results of our research (18). Exercise on the liver is characterized by its positive effect on liver function. Exercise increases the oxidation and metabolism of fat throughout the body and burns the liver and fatty acids. The reason for the consistency of research with our research results is probably the type of exercise or its severity. On the other hand, the research team Fatouros *et al.* (2010) stated that following a 30-minute resistance anaerobic activity session in 17 healthy young men, the activity of liver enzymes and lactate dehydrogenase did not change much (19). Barquilha *et al.* also sought to investigate the effects of a repeat of the maximum chest pressure test on liver enzyme indices in 11 healthy subjects (8 males and 3 females), with the intermittent collection of blood samples 24, 48, and 6 days after the activity, reported that plasma keratin kinase activity increased significantly on day 6 when compared with before activity (20). Possible reasons for the contradiction between the findings of the published studies and the results of this study can be attributed to the individual differences in the response to keratin kinase to the level of health as well as to the type and duration of physical activity. That is probably the reason why research is consistent with the research is a type of exercise. Kratz and colleagues (2002), measure the amount of ALP and AST in marathon runners before and 24 hours after the end of the race, observed significant increase in these enzymes (21). In contrast, Kinoshita and colleagues (2003), investigate the relationship between vigorous activity and liver cell damage in male mice. They found that mice with 60 to 80 percent of maximum heart rate and running time 120 minutes did not show a significant increase in their liver enzyme values (22). The results of the study are antithetic with the results of this study. On the other hand, some research have also examined the gender factor. For example, Dervis *et al.* (2008), studied the effect of endurance exercise on liver enzymes in men and women. The results indicated that the enzyme concentration in groups does not change and gender has no effect on this subject (23). ALT, AST and ALP enzymes are involved in the metabolism of the liver. Therefore, the probability of long-term liver damage to cell membranes and endurance is high. In case, if the exercise is resistance, most energy of these activities is supplied through anaerobic and liver cells, especially those enzymes are not involved in energy production, then they will be less damaged (24). Thus, as can be seen, the duration and intensity of activity and exercise training increase as the level of liver enzymes involved in ATP production increases. According to the theory of intracellular enzyme release out through the cell membrane, the leak of AST and ALT into the blood may be high (25). Due to high resistance and endurance exercise, adaptations are created in the cell and stabilize membranes and decreases the release of the liver enzymes and lactate dehydrogenase in the blood (26). In general, according to the findings of this study, it can be said that due to aerobic and anaerobic exhaustive workout, there is a possibility that ALT, AST, ALP enzymes, and LDH will increase in overweight individuals; increase in these values due to aerobic training is less than in anaerobic exercise. Given the important effects of obesity, diabetes and metabolic syndrome, it is recommended to increase the risk of fatty liver, aerobic physical activity, and weight loss and diet. Also, there was no significant difference between the AST and ALT levels of aerobic-control, anaerobic-control and aerobic-anaerobic groups in the post-test.

Conclusion

The results of this study revealed that one session of exhaustive aerobic and anaerobic exercise increases the levels of liver enzymes ALT, AST, ALP and LDH. Overweight people are more prone to fatty liver disease. The increased levels of these enzymes are considered as an indicator of liver damage. Therefore, according to our results, exhausting aerobic exercise can be a more appropriate exercise for those with extra weight.

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Conflict of Interest

Authors declared no conflict of interests.

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