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Effect of Maximal Aerobic and Anaerobic Exercise on Blood Zinc and Copper Levels of Male Athletes

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This study was carried out with 48 healthy male participants. They were separated into 3 equal groups with average age, body weight and heights as follows: Group 1 (aerobic group) with 22.69 ± 1.92 years, 80.69 ± 2.89 kg and 181.50 ± 4.00 cm; Group 2 (anaerobic group) with 21.56 ± 1.90 years, 80.94 ± 3.30 kg and 181.69 ± 2.50 cm and Group 3 (control group): 22.0 ± 1.32 years, 79.19 ± 3.41 kg and 181.63 ± 2.87 cm. The differences in blood zinc levels before and after aerobic and anaerobic loading (1 and 2 groups) were found to be statistically significant ($p < 0.01$). The post maximal aerobic exercise blood copper levels showed a statistically decrease compared to pre exercise levels ($p < 0.01$). This difference was found to be statistically insignificant for the anaerobic group (group 2) ($p > 0.01$). The difference between the pre and post test values of the control group also showed no statistically significant change as regards to both metals (group 3) ($p > 0.01$). In conclusion, it could be stated that aerobic exercises have more effects on reducing the blood zinc and copper level of athletes.

Key Words: Aerobic and anaerobic exercise, Blood, Zinc and copper levels, Male athletes.

INTRODUCTION

Zinc is a micronutrient important for the immune and reproduction systems, taste, skeleton build up and intestinal functions[1]. Zinc status is highly effective on physical performance. The zinc levels of the physically active people were observed to be low. The deficiency of zinc causes anorexia, loss of weight, exhaustion, loss of strength and osteoporosis[2]. Low zinc levels were reported to cause an increase in the viscosity of blood and decrease in performance[3]. However there is no consensus on the correlation between zinc levels and the exercise. Some workers report that the zinc level decreases with the exercise taken[4,5] while others say the opposite[6].

Copper is also one of the essential micro nutrients for the body. Copper deficiency causes the Menke disease and some hereditary disorders[7]. Copper deficiency manifests itself as a decrease in plasma concentrations. Copper is a co-factor for more than 30 enzymes in the body. Although some workers claim that the copper level would increase with exercise[4] there isn't any agreement on the correlation of copper level and the exercise as in the case for zinc. However, it is thought that large amount of copper ions is transferred to the drinking water from the copper ducts used to carry it. This may partly explain the vagueness in the change of copper levels.

EXPERIMENTAL

All the reagents used were in analytical grade. The solutions used for standard addition process were prepared from 0.1 M $Cu(NO_3)_2$ and $Zn(NO_3)_2$ (Merck) stock solutions. The buffer solution was prepared from acetic acid (Riedel de Häen) and NaOH (Merck). Nitric acid used for digestion process was in spectroscopic grade and all the samples were prepared using deionized water (16.8 MΩ)

The physical, physiological tests and measurement of age, body weight and height: The ages of the participants were recorded as years and the height was measured with a metric scale with naked feet and the weights were determined with the participants wearing only a short (Table-1). The accuracy of the scale was ± 0.01 kg. All the measurements were taken 1 h prior to the study.

TABLE-1
AVERAGE AGE BODY WEIGHT AND HEIGHTS, $MaxVO_2$, 100 m FREE STYLE SWIMMING OF THE PARTICIPANTS

Variable	N	Arithmetic mean ($\overline{X}$)	SS	Minimal Maximal
Age (years)	Aerobic group (16) 1.GROUP	22.69	1.9200	20.00-26.00
	Anaerobic group (16) 2.GROUP	21.56	1.9000	18.00-25.00
	Control group (16) 3.GROUP	22.00	1.3200	20.00-25.00
Height (cm)	Aerobic group (16) 1.GROUP	181.50	4.0000	174.00-190.00
	Anaerobic group (16) 2.GROUP	181.69	2.5000	176.00-186.00
	Control group (16) 3.GROUP	181.63	2.8700	176.00-186.00
Body Weight (kg)	Aerobic group (16) 1.GROUP	80.69	2.8900	76.00-87.00
	Anaerobic group (16) 2.GROUP	80.94	3.3000	76.00-87.00
	Control group (16) 3.GROUP	79.19	3.4100	71.00-84.00
$MaxVO_2$ (mL kg/dk)	Aerobic group (16) 1.GROUP	47.20	2.1500	43.9-50.8
100 m free style swimming (s)	Anaerobic group (16) 2.GROUP	1.12	0.0386	1.06-1.17

Selection of the participants: The study was carried our with the voluntary participation of 46 healthy male University students who has a regular sport habits but do it only at a fitness level. The participants were separated into three groups and given a copper and zinc free diet 1 week prior to the physical loading. They were adequately briefed about the importance of the study in order to increase their motivation. They were also informed about the rules to be obeyed throughout the study and asked to sign a voluntary participation form.

The anthropometric features of the participants were determined prior to the study and 5 mL of blood were taken from each participant at rest position. The participants were given 15 min resting period and then the 1st group (aerobic) was subjected to 20 m shuttle runs to force their aerobic limits. There were 5 mL of bloods taken from the participants at exhaustion just after the shuttle runs. The 2nd group (anaerobic) were made to swim 100 m free-style with maximum effort. There were 5 mL bloods taken from the participants after free-style swimming. The 3rd group (control group) was not subjected to any physical activity and the blood samples were taken at rest position in 1 h periods (Table-2).

TABLE-2
COMPARISON OF THE ZINC AND COPPER LEVELS OF THE PARTICIPANTS BEFORE AND AFTER THE MAXIMAL EXERCISE

Trace element	N	Resting ($\overline{X}$)	SS	After maximal exercise ($\overline{X}$)	SS	t.	p
Zinc (ppm)	Aerobic group (16) 1.Group	0.257	± 0.076	0.026	± 0.015	11.870	0.000* $p < 0.01$
	Anaerobic group (16) 2.Group	0.215	± 0.048	0.207	± 0.047	8.677	0.000* $P<0.01$
	Control group (16) 3.Group	0.225	± 0.033	0.223	± 0.031	1.541	0.144 $p > 0.01$
Copper (ppm)	Aerobic group (16) 1.Group	0.429	± 0.125	0.298	± 0.071	3.682	0.002* $p < 0.01$
	Anaerobic group (16) 2.Group	0.308	± 0.134	0.308	± 0.131	- 0.094	0.927 $p > 0.01$
	Control group (16) 3.Group	0.310	± 0.104	0.308	± 0.105	1.045	0.313 $p > 0.01$

20 Meter Shuttle run and determination of max VO_2: MaxVO_2 values which indicate cardiovascular efficiency and aerobic capacity were determined in mL kg/dk with 20 m shuttle runs. The test results were determined from maxVO_2 evaluation chart[8]. The exhaustion levels of the participants at the end of the test were determined from their heart beat rates.

100 M free style swimming: The participants were made to swim 100 m free-style in a 25 m semi Olympic swimming pool with under the supervision of a trainer and a doctor. Their exhaustion levels were determined from their heart beat rates at the end of the free style swimming.

Collection of the blood samples: There were venous 5 mL blood samples taken from the lefts arms of the participants at rest position with the use of heparinized syringes by professional nurses under the supervision of a doctor before the start of the study. The blood route was kept open with a cut down catheter for the extraction of further blood after the tests. The participants were subjected to 20 m - shuttle run (group 1) and 100 m maximum effort free style swimming (group 2) after a 15 min resting period. There were two 5 mL blood samples taken from the participants one just after the test and one after 1 h of rest after the test. (Group 3) or the control group was not subjected to any sort of physical activity

The samples taken were numbered, centrifuged and kept in a deep freeze. The samples were then analyzed as regards to copper and zinc contents in the laboratories of the Chemistry Department of Gazi University.

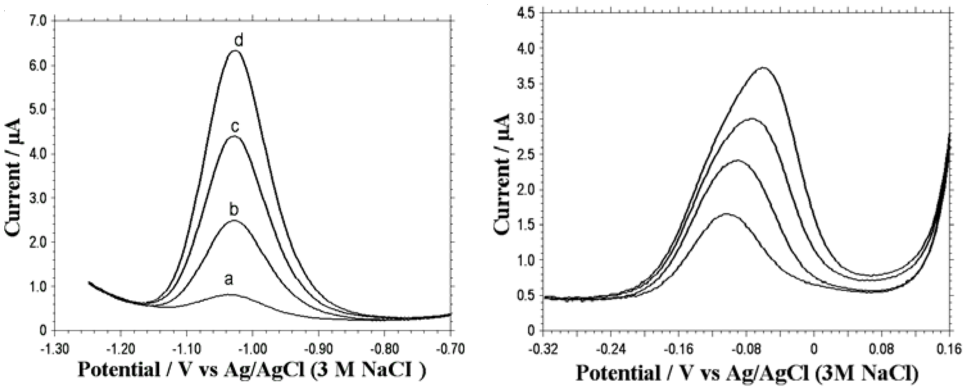
Digestion of the samples: The digestion procedure was carried out in Berghof/ Microwave Digestion System MWS-3 speedwave apparatus after taking 1 mL of blood and adding 2.5 mL HNO_3 on it. The microwave was kept at 160 °C for 5 min, 190, 100 and 80 ºC for 10 min each. The digested samples were diluted to 10 mL with the addition of deionized water (16.8 MΩ).

Voltammetric procedure: The trace elements analyses of the samples were carried out by the use of square wave stripping voltammetry. The electrochemical analyses were performed computer controlled CHI660B model potentiostat. The working electrode was 100 µm capillary hanging mercury electrode (BAS CGME) and the counter and reference electrodes were a Pt wire and Ag/AgCl (3 M NaCl). The residual oxygen in the system was removed by purging argon gas with spectrophotometric purity. The peak potentials for Cu^{2+} and Zn^{2+} were -0.10 V and -1.05 V (Ag/AgCl).

Analytical procedure: 0.5 mL of the samples which had been previously made up to 10 mL with deionized water was taken into the cell and 2 mL acetic acid-acetate buffer was added to them. The solution was stirred for 2 min before the stripping process. The stripping analysis is based upon cathodic deposition of the related metal ion and anodic stripping with a linear potential scan[9-11]. The voltammograms obtained after three standard additions of 20 µL 10^{-4} M Cu and Zn solutions were superimposed upon each other. The resulting voltammograms were displayed in Fig. 1. The increase in the Cu and Zn peaks at -0.10 V *vs.* -1.05 V was evaluated according to standard addition method. The procedure was repeated 3 times for each sample and the average of the amount of copper and zinc in each sample was computed. The blank study revealed that the amount of metal coming from nitric acid and deionized was negligible. In any case the amount of metal found in the blank sample was subtracted from the amount obtained for the actual samples.

Statistical analysis of data: The statistical analyses of the data were carried out with the use of 11.0 SPSS statistical software. The pre and post test values constituted the dependent and different study groups constituted the independent variables of the study. The mean values of the body weights and heights ($\overline{X}$) were given in the first sub-problem of the study and the evaluations were made according to this average point.

The testing of the second sub problem of the study in other words the testing of the difference between the pre and post tests grades of the participants was carried out by the use of paired simple t-test. The analyses of the third sub problem of the



Zn Cu

Fig. 1. Square wave anodic stripping results: (a) 0.5 mL sample + 2 mL acetic acid-acetate (b) addition of 20 mL 10^{-4} M Cu^{2+} and Zn^{2+}. (c) 40 mL 10^{-4} M Cu^{2+} and Zn^{2+}. (d) addition of 60 mL 10^{-4} M Cu^{2+} and Zn^{2+} voltammograms.

study in other words the variability of the pre test and post test Zn and Cu values of the participant according to the groups were performed with one way variance analysis (ANOVA). Also Tukey-HSD multiple comparison test was employed to determine between which groups this statistical difference existed. All these statistical analyses were carried at a reliability level of $\alpha = 0.01$.

RESULTS AND DISCUSSION

The change in the zinc status in blood after the maximal aerobic and anaerobic exercise (Group 1 and 2) was statistically significant ($p < 0.01$) (Tables 3 and 4). There was a marked statistically decrease in pre and post exercise copper values in the group who was subjected to maximal aerobic exercise (group 1). However, this difference was not of statistical significance in the anaerobic group (Group 2) ($p > 0.01$).

TABLE-3
CORRELATION OF $MaxVO_2$ AND PRE- AND POST-EXERCISE Cu AND Zn LEVELS OF GROUP 1 (AEROBIC GROUP)

Correlation	Aerobic group			
	Pre-exercise Zn	Post-exercise Zn	Pre-exercise Cu	Post-exercise Cu
$MaxVO_2$	r = 0.146	r = 0.228	r = 0.326	r = 0.114

There was also no statistically significant change in both metals for the control group (Group 3). When we look at the ANOVA results for zinc levels according to group variable there was a difference between group 1 and 2 and group 1 and 3 (Tables 5 and 6). Same trend also exists for the copper values (Tables 7 and 8).

TABLE-4
CORRELATION OF 100 m FREE STYLE SWIMMING TIMES AND PRE- AND POST-EXERCISE Cu AND Zn LEVELS OF GROUP 2 (ANAEROBIC GROUP)

Correlation	Anaerobic group			
	Pre-exercise Zn	Post-exercise Zn	Pre-exercise Cu	Post-exercise Cu
100 m free style swimming times	r = 0.776	r = 0.106	r = - 0.230	r = - 0.244

TABLE-5
ANOVA RESULTS OF PRE EXERCISE ZINC VALUES WITH GROUP VARIABLE

Group	N	$\bar{X}$	S	F	P
1	16	0.257	0.076	2.419	0.100
2	16	0.215	0.048		
3	16	0.225	0.033		
Total	48	0.232	0.057		

TABLE-6
ANOVA RESULTS OF POST EXERCISE ZINC VALUES WITH GROUP VARIABLE

Group	N	$\bar{X}$	S	F	P	Difference Tukey
1	16	0.026	0.015	162.443	0.000	1-2 1-3
2	16	0.208	0.047			
3	16	0.223	0.031			
Total	48	0.152	0.096			

TABLE-7
ANOVA RESULTS OF PRE EXERCISE COPPER VALUES WITH GROUP VARIABLE

Group	N	$\bar{X}$	S	F	P	Difference Tukey
1	16	0.429	0.125	5.162	0.010	1-2 1-3
2	16	0.308	0.134			
3	16	0.310	0.104			
Total	48	0.349	0.132			

TABLE-8
ANOVA RESULTS OF POST EXERCISE COPPER VALUES WITH GROUP VARIABLE

Group	N	$\bar{X}$	S	F	P
1	16	0.298	0.071	0.046	0.955
2	16	0.308	0.131		
3	16	0.308	0.105		
Total	48	0.305	0.103		

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