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The Effect of 12 Weeks of Continuous Aerobic Training on Hematological Parameters in Old Female Rats

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Abstract: The aim of this research was to compare the changes of hematological parameters in female rats after a period of continuous aerobic training. 32 female rats (21.5 months old, weight 322.75+8.14 gram) were randomly divided into experimental and control (pretest and posttest) groups. The subjects lived in individual cages (15 x 15 x 20 cm) in the following condition: temperature 22+2°, humidity 50+5% and cycle 12:12 hours. They were killed after 12 weeks of continuous aerobic training in the same condition. K-S test was used to examine the normality of the groups. T test was used to determine the difference between the two groups (P=0.05). The results showed that a period of such training significantly changes iron (t=5.495, P=0.001), hemoglobin (t=12.342, P=0.000), TIBC (t=9.169, P=0.000), %Hc (t=14.472, P=0.000), transferrin (t=4.395, P=0.003), MCH (t=7.301, P=0.000), MCHC (t=7.301, P=0.005), MCV (t=3.453, P=0.011), PLT (t=8.724, P=0.000), RBC (t=5.329, P=0.001) in experimental group. Generally, it can be stated that a period of continuous aerobic training increases the performance of hematological parameters which play a crucial role in cell respiration.

Key words: Aerobic Training % Hematological Parameters % Old Female Rats

INTRODUCTION

In the current century, sports, correct treatment and regular medical examination are the factors for a longer and more fruitful life. One of the serious problems people face worldwide is to provide health care for the elderly. In USA in 1990, 75 billiard dollars were expended in the health care and treatment of the elderly. Today in developed countries, hope to live longer be 71 years old for men and 78 years old for women. Although it is estimated that the people over the age of 65 will live another 16.4 years, they spend only 12 years of this time span healthily. Therefore, these people should be provided with health promotion measures. The reports show that those in a better sporting and nutritional condition are 10-20 biological years younger than those at their real age [1].

The researches show that sporting and physical activity positively affects the cardiovascular system. Type of physical activity (endurance, spring and resistance, interval, maximal and sub-maximal), the duration and intensity result in various reactions of hematological parameters and iron condition.

Most researches investigated the acute effect of a period of physical activity on the above parameters while few researches investigated the effects of a training period (adaptation to training) on subjects, especially old subjects, female subjects as well as menopausal subjects [2].

Peikkio *et al*. [3] conducted a research on Sprague Dawley rats. The rats enjoyed two types of diets (low iron diet: 6 ml/kg and high iron diet: 50 ml/kg) for two weeks. Next, they performed endurance training on a treadmill for one month. The results showed that low iron diet decreased hemoglobin (8 g/dl against 16 g/dl). It also significantly decreased cytochrome C, cytochrome oxidase, succinate oxidase and muscle oxidative enzymes. Therefore, the researchers suggested that endurance training along with a low iron diet can result in an acute anemia and can decrease the higher production of oxidative enzymes in skeletal muscles (which help a better performance of endurance activities).

Dressendorfer [4] conducted a research (named development of runners' anemia during 20-day road race: effect of iron supplements) on 15 runners and showed

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that 12 subjects significantly decreased their serum iron, hemoglobin and red blood cells after 10 days of the competition.

Schumacher *et al*. [5] conducted a research on 39 non-athletes, athletes and elite athletes to investigate the effect of various types of training and physical activity on serum iron, percentage of transferring saturation and ferritin concentration.The first and the second groups had the Bruce laboratory test as well as a 45- minute run with a constant speed and 70% VO2max. The third group had a long aerobic cycling. The results showed a significant increase in transferrin in athletes and non-athletes after both tests (Bruce test and a 45- minute run with 70% VO2max) while it did not change in the third group. In addition, the serum iron significantly increased in athletes (second group) after the first test and in non-athletes (first group) after the second test.

Michalis *et al*. [6] conducted a research named variation of soluble transferrin receptors and ferritin concentrations in human serum during recovery from exercise. 15 young male non-athletes cycled an ergometer (150-155 heart rates per minute) for 45 minutes. The blood samples were gathered and analyzed before, immediately after and 24 hours after the activity. The results showed that although the variation of transferrin receptors is much lowerthan ferritin receptors, variation of transferrin and ferritin is not significant.

Yu *et al*. [7] conducted a research named molecular analysis of increased iron status in moderately exercised rats. 30 Sprague Dawley female rats (age=5 weeks, weight=90-100 gr.) were divided into three groups: control, experimental (moderate exercise: 1.5 hours swimming daily) and experimental (intense and maximal exercise) and their serum iron, serum ferritin, blood hematocrit, divalent metal transport 1(DMT1), ferroprotein (FPN1), epithelium duodenum and hepcidin mRNA were investigated. The results showed that:

- The iron in the experimental group (moderate exercise) was higher than the other two groups. The iron changed with the intensity of the exercise.
- Hepcidin mRNA in the experimental group (moderate exercise) was lower than the normal level.
- Maximal and intense physical exercise decreased iron after 10 weeks of exercise while moderate exercise increased iron and resulted in more adaptability to exercise.
- Moderate exercise could increase the absorption of TIBC while intense exercise decreased iron homeostasis.
- Moderate exercise could increase iron through normalization of DMT1 and appearance of IRE and FPN1.

The effect of hepcidin in the liver may be through its effect on the natural activities of proteins involved in iron absorption [7]. With regard to the effect of antioxidant supplements, drugs, amino acids and various diets on hematological parameters and iron metabolism [8] and due to the fact that all these parameters can be controlled in human subjects, the researches which can determine the effect of exercise on these parameters seem essential.

As movement deficiency (due to aging) can decrease plasma volume and red blood cell volume [9], this research intends to investigate the effect of 12 weeks of continuousaerobic training on some hematological parameters and iron status in female rats and after menopause.

MATERIALS AND METHODS

This research is experimental. 32 rats (21.5 months old, weight: 322.75+8.14) were randomly selected from the rats in Iran Pastor Institute. The menopause was investigated as follows: the female rats were under Pastor Institute experts' supervision for several months so that the number of their birth-giving decreased and their fertility ended at the age of 18 months. The samples were transferred to the animal laboratory of Physical Education and Sport Science Faculty (University of Tehran) and adapted to the environment. They trained on a treadmill for rats (made in Physical Education and Sport Sciences Faculty) and were randomly divided into experimental group (pretest, mid-test and posttest) and control group (pretest, mid-test and posttest). The exercise protocol was a 12-week (5 training sessions per week) continuous aerobic run on a treadmill (from 12m/min. for 10-14 minutes in the first session to 22m/min. for 80 minutes in the last session). The blood samples of the two groups were gathered at the beginning and the end of the training protocol. Kolmogorov-Smirnov test was used to investigate the normal distribution of data, Microsoft Word to rank the data and to design the tables, t test to investigate the intra-group differences and repeated measures analysis and ANOVA to investigate the inter-group differences in pretests and posttests and LSD to determine the differences between the two groups ($P<0.05$).

RESULTS

Tables 1 and 2 show the female rats' characteristics in both groups.

Various hematological parameters were measured and the results were presented in Table 3:

Table 1: the female rats' characteristics in experimental and control groups

	Characteristic	
Group	Weight (gr.)	M+SD
Experimental and Control (Pretest)	322.75+8.14	21.5
Experimental (post test)	318.38+4.17	24.5
Control (post test)	323+2.83	24.5

Table 2: Statistical findings of hematological parameters in pretest and posttest

Hematological parameters		Tests	Mean	SD (+)	Mean Dif.	Lower	Upper
Fe mg/dl	Pre	Experimental and Control	322.75	28.424	10.049	296	367
	Post	Experimental	400.88	65.669	23.217	312	493
		Control	228.13	71.788	25.381	165	346
HB g/dl	Pre	Experimental and Control	13.613	0.2357	0.0833	13.4	14
	Post	Experimental	15.05	0.239	0.0845	14.8	15.4
		Control	13.525	0.1909	0.0675	13.3	13.8
TIBC mg/dl	Pre	Experimental and Control	365.63	4.207	1.487	356	369
	Post	Experimental	432.5	14.639	5.175	410	450
		Control	345	30.822	10.897	310	390
HCr %	Pre	Experimental and Control	38.713	0.2357	0.0833	38.4	39
	Post	Experimental	40.863	0.4104	0.1451	40.2	41.4
		Control	36.675	0.495	0.175	36	37.3
Trans mg/dl	Pre	Experimental and Control	118.88	1.458	0.515	117	121
	Post	Experimental	124.63	4.438	1.569	119	130
		Control	112.75	4.464	1.578	106	119
MCH pg	Pre	Experimental and Control	17.95	0.6803	0.2405	17.2	19.3
	Post	Experimental	19.088	0.2532	0.0895	18.8	19.5
		Control	17.438	0.5579	0.1972	17	18.5
MCHC g/dl	Pre	Experimental and Control	35.438	0.447	0.158	34.9	35.9
	Post	Experimental	37	0.2507	0.0886	36.5	37.2
		Control	36.575	0.0886	0.0313	36.5	36.7
MCV fl	Pre	Experimental and Control	52.1	0.7616	0.2693	51.1	53.7
	Post	Experimental	52.375	1.2992	0.4593	50.6	53.5
		Control	49.838	1.2455	0.4403	46.9	50.7
PLT ×1000/µl	Pre	Experimental and Control	599.63	159.134	56.262	344	798
	Post	Experimental	985.13	32.8	11.597	951	1026
		Control	622.13	113.572	40.154	512	801
RBC mi/µl	Pre	Experimental and Control	7.3587	0.11801	0.04172	7.26	7.52
	Post	Experimental	7.95	0.20007	0.07074	7.59	8.18
		Control	7.3313	0.17675	0.06249	7.11	7.65

Table 3: Inter-group and intra-group findings of hematological parameters in both groups

	Mean Dif.	SD (+)	Samples	T test	Sig.
Fe(mg/dl)					
Pretest – posttest (experimental)	25.53	72.22	8	-2.668	0.032*
Posttest (experimental) – posttest (control)	31.44	88.91	8	5.495	0.001*
HB(g/dl)					
Pretest – posttest (experimental)	0.1546	0.4373	8	-9.297	0.001*
Posttest (experimental) – posttest (control)	0.1236	0.3495	8	12.342	0.001*
TIBC(mg/dl)					
Pretest – posttest (experimental)	5.604	15.851	8	1.897	0.001*
Posttest (experimental) – posttest (control)	9.543	26.992	8	9.169	0.001*

Table 3: Continued

	Mean Dif.	SD (+)	Samples	T test	Sig.
HCR%					
Pretest – posttest (experimental)	0.189	0.5345	8	-11.377	0.001*
Posttest (experimental) – posttest (control)	0.2894	0.8184	8	14.472	0.001*
Trans (mg/dl)					
Pretest – posttest (experimental)	1.59	4.496	8	-3.617	0.009*
Posttest (experimental) – posttest (control)	2.702	7.643	8	4.395	0.003*
MCH (pg)					
Pretest – posttest (experimental)	0.207	0.5854	8	-5.496	0.001*
Posttest (experimental) – posttest (control)	0.226	0.6392	8	7.301	0.000*
MCHC (g/dl)					
Pretest – posttest (experimental)	0.2052	0.5805	8	-5.496	0.001*
Posttest (experimental) – posttest (control)	0.1048	0.2964	8	7.301	0.005*
MCV (fl)					
Pretest – posttest (experimental)	0.4048	1.1449	8	-0.679	0.519
Posttest (experimental) – posttest (control)	0.7348	2.0784	8	3.453	0.011*
PLT (x1000/µl)					
Pretest – posttest (experimental)	61.973	175.287	8	-6.22	0.001*
Posttest (experimental) – posttest (control)	41.608	117.686	8	8.724	0.001*
RBC (mi/µl)					
Pretest – posttest (experimental)	0.06351	0.17964	8	-9.309	0.001*
Posttest (experimental) – posttest (control)	0.1161	0.32839	8	5.329	0.001*

*shows the significant level and P=0.05

C A period of continuous aerobic training significantly increased Fe (mg/dl) after 12 weeks (P=0.032). Also, there was a significant difference in Fe between experimental and control group after 12 weeks of training (P=0.001) (Fig. 1).

C A period of continuous aerobic training significantly increased hemoglobin after 12 weeks (P=0.000).

Also, there was a significant difference in HB between experimental and control group after 12 weeks of training (P=0.000) (Fig. 2).

C A period of continuous aerobic training significantly increased TIBC (mg/dl) after 12 weeks (P=0.000). Also, there was a significant difference in TIBC between experimental and control group after 12 weeks of training (P=0.000) (Fig. 3).

C A period of continuous aerobic training significantly increased HCR% after 12 weeks (P=0.000). Also, there was a significant difference in HCR% between experimental and control group after 12 weeks of training (P=0.000) (Fig. 4).

C A period of continuous aerobic training significantly increased transferrin saturation % after 12 weeks (P=0.009). Also, there was a significant difference between experimental and control group after 12 weeks of training (P=0.003) (Fig. 5).

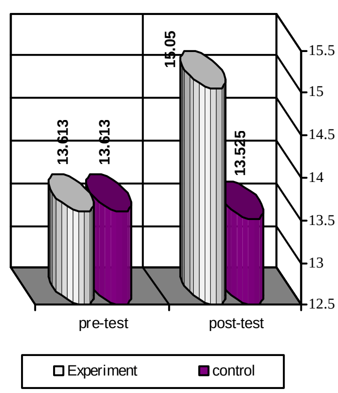

Fig. 1: Iron in experimental and control group

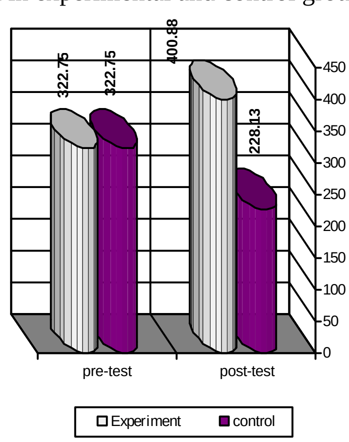

Fig. 2: HB in experimental and control group

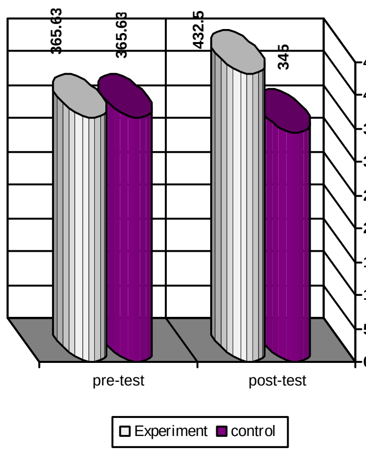

Fig. 3: TIBC in experimental and control group

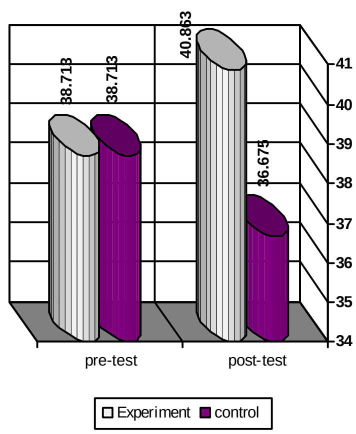

Fig. 4: HCR% in experimental and control group

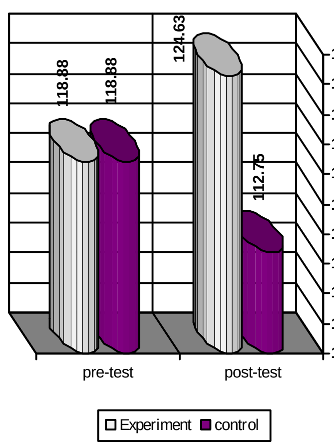

Fig. 5: Transferrin in experimental and control group

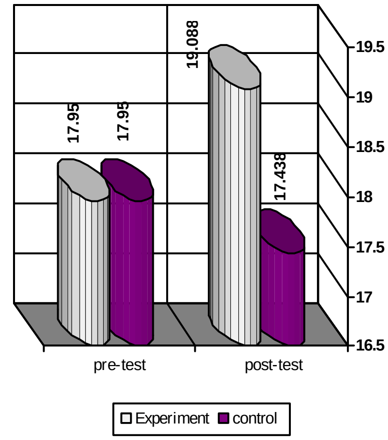

Fig. 6: MCH in experimental and control group

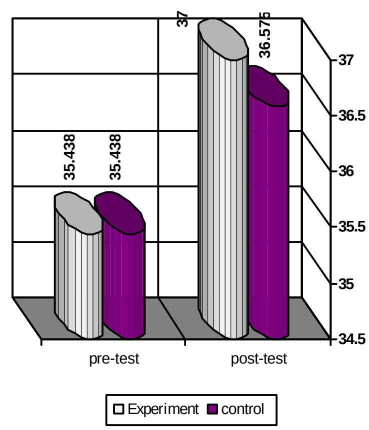

Fig. 7: MCHC in experimental and control group

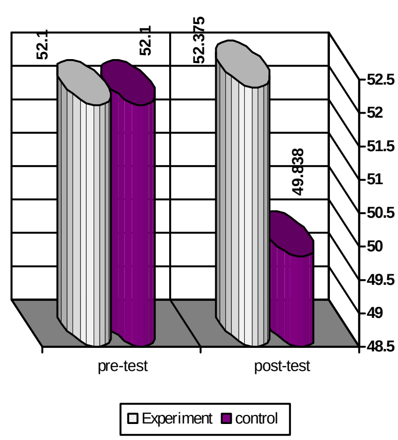

Fig. 8: MCV in experimental and control group

- A period of continuous aerobic training significantly increased MCH (pg) after 12 weeks (P=0.001). Also, there was a significant difference between experimental and control group after 12 weeks of training (P=0.000) (Fig. 6).

- A period of continuous aerobic training significantly increased MCHC (g/dl) after 12 weeks (P=0.000). Also, there was a significant difference between experimental and control group after 12 weeks of training (P=0.005) (Fig. 7).

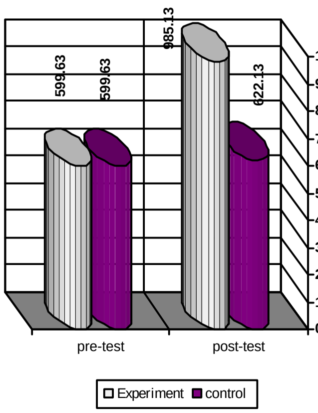

Fig. 9: PLT in experimental and control group

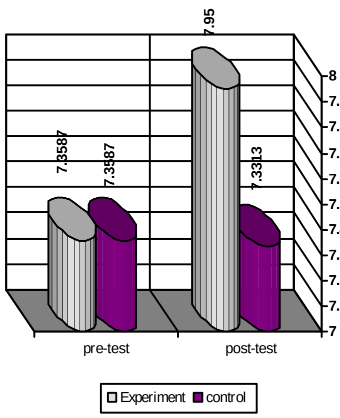

Fig. 10: RBC in experimental and control group

- A period of continuous aerobic training did not significantly increase MCV (fl) after 12 weeks between pretest and posttest (P=0.519). But, there was a significant difference between experimental and control group after 12 weeks of training (P=0.011) (Fig. 8).
- A period of continuous aerobic training significantly increased PLTx1000/µl after 12 weeks (P=0.000). Also, there was a significant difference between experimental and control group after 12 weeks of training (P=0.000) (Fig. 9).
- A period of continuous aerobic training significantly increased RBC after 12 weeks (P=0.000). Also, there was a significant difference in RBC between experimental and control group after 12 weeks of training (P=0.001) (Fig. 10).

DISCUSSION AND CONCLUSION

The results of the present research showed that hemoglobin significantly increased after a period of continuous aerobic training. Peikkio *et al*. [3] support this finding although they used two types of diets: low iron and high iron before the exercise training protocol. The results of the present research showed a significant increase in hemoglobin, HCR%, MCV, MCH and MCHC after a period of continuous aerobic training while Cordova *et al*. [11] made the rats to swim until they reached exhaustion and showed that although hemoglobin and HCR% significantly increased, MCV, MCH and MCHC did not significantly change.

Portmans [12] observed significant changes in transferrin and haptoglobin concentration immediately after 60 minutes of cycling (67% Vo2max) which supports the present findings.

Bourque *et al*. [13] conducted a research named "the effect of twelve weeks of endurance exercise training on iron status in healthy women" on 31 women aged between 23 and 43. They all had normal iron status. The training protocol consisted of 12 weeks, 3-4 sessions of running, walking and cycling per week (80% Vo2max). Energy expenditure was 150 kilocalories in the first week and it was 375 kilocalories in the ninth to twelfth week. The following parameters were measured: serum ferritin concentration, serum iron, transferrin concentration percent, TIBC, serum haptoglobin concentration as well as other hematological parameters at the end of week 2, 4, 8, 12. After 12 weeks, the results showed no significant change in serum ferritin concentration in the experimental group. No significant difference was observed in iron and serum transferrin concentration, TBIC and haptoglobin between the two groups. Generally, the results showed that 12 weeks of moderate aerobic training did not decrease the iron status and blood indexes in healthy women with natural ferritin (20 micrograms). These results do not support the present findings which show a significant difference in hematological parameters of the experimental group after 12 weeks of continuous aerobic training [13].

Schumacher *et al*. [5] conducted a research on athletes and healthy untrained individuals by Bruce test (70% Vo2max) and long-term cycling. Their results showed that the long-term cycling did not significantly change transferrin and serum iron in human subjects. The above findings support Michalis *et al*. [6], but are contrary to our present findings. It should be mentioned that Haas *et al*. [9] conducted a review named "iron deficiency and reduced work capacity" pointed to a difference in results between human and animal subjects. Yu *et al*. [7] found an increase in iron status and HCR% of Sprague Dawley female rats which supports our findings.

Finally, it can be stated that moderate continuous aerobic training can increase red blood cells as well as hematological parameters such as serum iron, HCR%, MCHC, MCH, MCV, TIBC and PLT, but an increase in plasma volume after endurance training should be considered as well.

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