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# The effect of 4-month cycling trainings performed on elite cyclists on some immunological parameters

Fulya ERANTGIL<sup>1</sup>, Mehibe AKANDERE<sup>2</sup>

<sup>1</sup>İstanbul Rumeli Universty, Vocational College, İstanbul,Turkey <sup>2</sup>Selçuk University Faculty of Sport Science, Konya,Turkey Address Correspondence to F.Erantgil e-mail: fulya.erantgil@hotmail.com

#### Abstract

The aim of this study was to investigate some immunological parameters, which showed differences in elite cyclists as a result of four months of chronic cycling trainings. The experimental group in this study consisted of 15 elite level trained cyclists between the ages of 17-19 who participated in international competitions. A regular training program was applied to the cyclists belonging to the experiment group for 6 days a week for 4 months. Prior to four-month chronic training, blood samples of cyclists were taken from the elbow vein. This procedure was repeated four months later. The control group consisted of individuals who did not regularly exercise and their ages were equivalent to the cyclists. No training program was applied to the control group. The data obtained in the study were analyzed statistically with the SPSS computer package software after being transferred to the electronic medium. In order to determine the homogeneity of the obtained data, "Single Sample Kolmogorov-Smirnov" test was performed, and the data was found to show normal distribution. "Independent t" test was used to determine differences between groups, while the analysis of intra-group measurements was performed by "Paired t" test. Significance level was determined as P<0.05. Immunoglobulins and leukocyte parameters were measured in the study. In the obtained findings, as a result of the 4-month chronic cycling training in the experimental group, statistically significant difference was found in leukocyte, monocyte%, neutrophil, neutrophil%, lymphocyte, lymphocyte% values before and after training, whereas, monocyte, eosinophil, eosinophil%, basophil and basophil% parameters did not display any significant difference (P>0.05). Significant differences were observed in IgA, IgG, and IgE parameters, whereas no significant difference was found in IgM parameter (P<0.05). When the leukocyte counts of the experimental group and the control group were examined, no significant difference was observed in the lymphocyte, monocyte, monocyte%, eosinophil, eosinophil%, basophil parameters, while a statistically significant difference was detected between leukocyte, neutrophil, neutrophil%, lymphocyte% and basophil% values (P<0.05). When the immunological parameters belonging to both groups were examined, it was found that there was a significant difference in the parameters of IgG, IgG, and IgM between the control and experimental group (P<0.05). This was not the case for IgE parameter (P>0.05). In conclusion, some leukocyte and immunoglobulin parameters were increased in the presence of chronic training. This increase is due to the reaction of the immune system to the training.

Keywords: exercise; immunoglobulin; leukocyte

### INTRODUCTION

After 1980, the body left being a research topic of biology, and its existence in socialization has come to the forefront with various studies. Various sciences have been influential on bodily development and transformation, the literature has been reviewed and analyzed, and the information that these studies have developed has been continually improved (15). When the human body is closely examined, it can be realized that it is a wonderful entity that possesses specific abilities. With regular exercises, physical and physiological capacities develop significantly. In previous studies, it has been stated that exercise has positive effects on physiological, psychological, physical and motor characteristics, and the immune system (13). In this context, the immune system is in our lives in all

scientific or non-scientific, written and visual publications. When the topic is this current or up-todate, the question "Is the immunological system or the immune system known enough?" comes to mind at first (6). The presence of mechanisms that take place in the immune system may render our body vulnerable when interacting with metabolic risk factors. It is assumed that the immune system is a developmental process based on evolution, originating from the need to recognize 'the self' when the cells came together to form the organism in primitive multi-celled living organisms, and transformed itself during the evolutionary process into a system that recognizes foreign materials and organisms, and protects the organism from the outside (35). When a living or nonliving object enters the body, there are many layers that protect it.

As the first layer, skin is the first physical barrier that provides protection against possible microbes entering the body (1). Immune system cells are found in blood and lymph circulation under normal conditions. When faced with an infectious agent, the natural system is activated first, followed by T and B lymphocytes (15). When the natural immune system components form the first line of defense that meets microorganisms when they penetrate the epithelial surface of the body (30).

Leukocytes are the mobile elements of the system responsible for the body's protection. They are partially formed in the bone marrow (granulocytes, monocytes, and a few lymphocytes). Following their formation, they are transported through blood to different body parts where they can be useful. The real importance of leukocytes is that most of them are specifically transported to areas of severe infection and inflammation, thus providing a rapid and strong defense against any possible infectious agent (17).

B and T lymphocytes, which are lymphocyte subtypes responsible for cellular and humoral immunity, and immunglobulins and interleukins, which contain soluble factors, are constantly interacting with each other (20). Lymphocytes are also divided into two main groups: T lymphocytes and B lymphocytes. T cells constitute 70% of blood lymphocytes, and B cells constitute 20-25% (3).

Immunoglobulins act as both receptor and effector molecules. As receptors, they recognize toxins, viruses, and other foreign antigens on the surface of pathogenic organisms. They inactivate foreign antigens after reacting to stimuli or they contribute to their elimination (40).

## EXERCISE and IMMUNE SYSTEM

In recent years, current research on the effects of chronic training on the immune system has gained impetus. The general perception here is that the immune system of athletes and other individuals engaging in physical activity develop as a result of chronic training (28).

Over the course of history, there have been studies on how exercise affects blood values. In fact, exercise is also important in terms of hematological pathology as much as it affects blood values with its type and intensity (23).

Exercise is known to cause stress on the human organism. This stress generally has various physiological and metabolic effects. Differences in blood values are the first of these observed effects (18). Can see that moderate regular exercise reduces the risk of infection. It is a widespread observation in athletes that small discomforts such as respiratory tract infections and colds can harm exercise performance. There are a variety of behavioral, nutritional and educational strategies to limit the risk of infection and to limit immunosuppression associated with exercise (14).

An intense exercise and competition schedule can lead to immunological damage in athletes, which increases susceptibility to infections. This immune dysfunction due to exercise, is often caused by the immunosuppressive formation of stress hormones such as adrenaline and cortisol, which increase during exercise (9).

Athletes often concentrate on their workouts for a few days or weeks at certain times of the season. With the increased performance in this period, the over-consolidation may lead to a reduction in performance and transition to a light training period. Many studies in recent years have examined the short-term effect of intensive training on immune function while resting, and immunoendocrine responses of endurance training. These studies show that; lymphocyte proliferation, antibody synthesis, NK cytotoxic cell activity, T lymphocyte CD4, CD8 ratios, and leukocyte functions, including monocyte and neutrophilia, depend on the increasing training and well-trained status of athletes (26).

## MATERIALS and METHODS

15 elite level, trained cyclists participating in international competitions, with an average age of 17,66±0,81 years, participated in the study as the experimental group.

The experimental group of cyclists in the study consisted of elite athletes of TOHM (Turkish Olympic Preparatory Centers) who will successfully represent our country. It was deemed appropriate to discuss with the trainers of the athletes and obtain information. The trainers and the athletes were informed about the study one week before the blood collection. The significance and purpose of the study was explained to the participants. Athletes were also informed about the measurements and procedures to be performed. For the study, written consent of the athletes was obtained and the study was carried out under medical supervision. Before the training period of 4 months, at 8:00 am, the blood collection procedure was carried out by a nurse at the elbow venue and 10 cc under the supervision of a doctor before the training of the athletes.

A regular training program was applied to the cyclists in the experimental group in medium and sub-maximal intensity for 6 days a week for 4 months. The trainers of the athletes were interviewed on the training program, and information was collected regarding the appropriate training method to be followed. Athletes made cycling training in road and bicycle room and they made running training on the running track, they had fitness training program at the gym.

15 males with a sedentary lifestyle, with no habit of regular exercise in the past, with an average age of 18,13±0,91 years, voluntarily participated in the study as the control group.

The control group included in the study did not have any chronic illness, metabolic disorder, and disability that could affect their immune system in their life history and health examination. Considering the age range and gender of the healthy sedentary control group, attention was paid to keep these characteristics close to the experimental group. No training program was applied to the control group.

The statistical evaluation of the findings was performed by SPSS computer package software. The arithmetic mean and standard deviations of the obtained parameters were calculated. The "Single Sample Kolmogorov-Smirnov" test was performed to determine the homogeneity of the data, and it was determined that the data showed normal distribution. "Independent T" test was used to determine the differences between the groups, and "Paired T" test was used for the analysis of intragroup measurements. Differences at P<0.05 level were accepted as significant.

## **FINDINGS**

Table 1. Measurements of Pre-Test and Post-Test Leuco	ocyte Parameters of the Experiment Group
	cyte i ulunicielo of the Experiment Group.

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Parameters	Timing	Ν	Mean±SD	Т	Р
WBC(K/µL)	Pre Test	15	7,00 ±1,55	0 1 9 2	0,047*
	Post Test	15	8,01 ±1,74	-2,183	
Neutrophils(K/µL)	Pre Test	15	$3,27 \pm 1,18$	2 020	0,009*
	Post Test	15	$4,77 \pm 1,75$	-3,029	
$\mathbf{N}_{\mathbf{r}}$	Pre Test	15	45,76 ± 6,96	2 790	0,002*
Neutrohpils (%)	Post Test	15	$58,12 \pm 9,78$	-3,789	
	Pre Test	15	$2,09 \pm 0,43$	2.017	0,009*
LYM(K/µL)	Post Test	15	$2,48 \pm 0,46$	3,017	
I VM (%)	Pre Test	15	$42,40 \pm 6,73$	2 010	0,002*
LYM (%)	Post Test	15	$32,15 \pm 8,09$	3,910	
MONO(V/I)	Pre Test	15	$0,60 \pm 0,17$	0,946	0,360
MONO(K/µL)	Post Test	15	$0,57 \pm 0,13$	0,946	
MONO (%)	Pre Test	15	$8,63 \pm 1,82$	2,355	0,034*
MONO (%)	Post Test	15	$7,63 \pm 1,65$	2,333	
EQC(V/I.)	Pre Test	15	$0,16 \pm 1,38$	1 011	0,246
EOS(K/µL)	Post Test	15	$0,05 \pm 0,02$	1,211	
EOS (%)	Pre Test	15	$2,40 \pm 1,22$	1.022	0,074
	Post Test	15	$1,69 \pm 1,38$	1,932	
BASO(K/µL)	Pre Test	15	$0,05 \pm 0,02$	0,959	0,354
	Post Test	15	$0,05 \pm 0,02$	0,959	
	Pre Test	15	$0,78 \pm 0,28$	1.025	0,075
BASO (%)	Post Test	15	0,66 ±0,28	1,925	
*P<0,05					

 Table 2. Pre-Exercise and Post-Exercise Immunoglobulin Parameters of the Experimental Group.

Parameters	Timing	Ν	Mean±SD	Т	Р
IgA mg/ml	Pre Test	15	124,73 ± 37,92	-3.422	0,004*
	Post Test	15	177,26 ± 67,33	-3,422	
IgG mg/ml	Pre Test	15	1009,26 ±120,24	-8,747	0,000*
	Post Test	15	1270,33 ±218,67	-0,747	
IgM mg/ml	Pre Test	15	80,00±33,22	-1.930	0,074
	Post Test	15	106,26±70,66	-1,930	
IgE mg/ml	Pre Test	15	30,85±14,38	4 279	0,001*
	Post Test	15	69,50±43,40	-4,278	

\*P<0,05

Table 3. Comparison Of Leukocyte Parameters Of The Pre-Exercise Experimental Group And Control Group

Parameters	Group	Ν	Mean±SD	Т	Р
WBC(K/µL)	Experiment Group	15	7,00±1,55	-2,804	0,009*
	Control Group	15	8,77±1,87	-2,004	
	Experiment Group	15	3,27±1,18	2 1 9 (	0.004*
Neutrophils (K/µL)	Control Group	15	5,01±1,74	-3,186	0,004*
Neutrophils (%)	Experiment Group	15	45,76±6,96	0.470	0,002*
	Control Group	15	56,06±9,12	-3,473	
LYM (K/µL)	Experiment Group	15	2,09±0,43	0 512	0,612
	Control Group	15	2,80±0,57	0,512	
I VM (0/)	Experiment Group	15	42,40±6,73	3,272	0,003*
LYM (%)	Control Group	15	33,06±8,76		
MONO(K/µL)	Experiment Group	15	0,60±0,17	-1,901	0,068
	Control Group	15	0,72±0,18		
MONO(9/)	Experiment Group	15	8,63±1,82	0,636	0,530
MONO (%)	Control Group	15	8,28±1.06		
EQC(V/L)	Experiment Group	15	0,16±1,38	-0,157	0,876
EOS(K/µL)	Control Group	15	0,17±0,10		
EOS (%)	Experiment Group	15	2,40±1,22	0,886	0,383
	Control Group	15	2,00±1,29		
BASO(K/µL)	Experiment Group	15	0,05±0,02	1,112	0,276
	Control Group	15	0,04±0,01		
BASO (%)	Experiment Group	15	0,78±0,28	2 704	0,012*
	Control Group	15	0,57±0,12	2,704	
*D<0.0E					

\*P<0,05

Table 4. Comparison Of İmmunoglobulin Parameters Of The Pre-Exercise Experimental Group And Control Group.

control Group.					
Parameters	Group	Ν	Mean±SD	Т	Р
IgA mg/ml	Experiment Group	15	124,73 ± 37,92	-3,646	0,001*
	Control Group	15	184,80 ± 51,31	-3,646	0,001
IgG mg/ml	Experiment Group	15	1009,26±120,24	1 200	0.000*
	Control Group	15	1266,73±204,29	-4,206	0,000*
IgM mg/ml	Experiment Group	15	80,00 ± 33,22	2 591	0,015*
	Control Group	15	122,40 ± 54,27	-2,581	
IgE mg/ml	Experiment Group	15	30,85 ± 14,38	0.102	0.95(
	Control Group	15	31,93 ± 17,82	-0,183	0,856

## DISCUSSION AND CONCLUSION

The aim of this study was to investigate some immunological parameters, which showed differences in elite cyclists as a result of four months of chronic cycling trainings. 15 elite cycling athletes with a mean age of 17.66±0.81 and 15 sedentary individuals with a mean age of 18.13±0.91 voluntarily participated in the study.

In their study, Tomar and Antony (43) analyzed the increase in total leukocyte and basophil counts of male subjects who did not exercise, as a result of the 16-week football training conducted. Another important detail in the study was the decrease in monocytes and eosinophils as a result of the training, but no significant change in neutrophils and lymphocytes was observed. In contrast to the leukocyte increase observed after the 16-week football training in this study, Lal's (25) study showed a decrease in the leukocyte ratio of the experimental group as a result of 8-week yoga training.

In a study conducted by Patlar (37), it was found that the leukocyte values measured postexercise increased significantly compared to preexercise values, and that there was no difference between the leukocyte values measured immediately after exercise and the values measured two hours after exercise. Similarly, Ali et al. (2) found that there were no significant changes in monocyte, eosinophil and basophil values between the measurements taken right after the exercise and two hours after the exercise.

According to Brines et al. (5), neutrophil is a parameter with a more significant response to exercise compared to the leukocyte movement in blood. An increase in the amount of neutrophil was observed depending on the intensity and severity of the exercise. Exercises have short-term and longterm effects on neutrophil function. Generally, neutrophils are induced by chemical toxins, oxidative burning and phagocytosis in medium severity and intensity exercise. Accordingly, and according to his study, Mackinnon (28) reported that chronic intensive training was proven in many studies to exert many changes that could suppress the immune system, but that moderate chronic training may have a slight positive effect on the development of the immune system. Pedersen et al. reported that immunological functions (38) developed in regular and mild aerobic exercise in mice, whereas prolonged or intense exercise suppressed immune response.

Ünal (45) found significant increases in leukocytes as a result of 8 weeks of aerobic exercises. Walsh et al. (46) observed an increase in lymphocyte count (lymphocytosis) during and immediately after exercise. This increase comes as a process that depends on the duration and intensity of the exercise. The increase is greater in T lymphocytes than the increase in B lymphocytes. The number of lymphocytes then falls below the pre-exercise count, reaching the pre-exercise state within 24 hours. Ersöz et al. (12) found significant increases in leukocyte counts as a result of 6 weeks of moderate exercise in their studies on sedentary young people. This increase is explained by several mechanisms. According to Pyne (39), first of these mechanisms is the amount of epinephrine that increases with exercise causes the neutrophils in the margination pool to enter circulation through demargination. The other mechanism defined by Severs et al. (41) is the acceleration of the release from the neutrophil reservoir pool in the bone marrow due to stress, muscle damage, and increased temperature caused by exercise. In these studies, the severity, duration, and intensity of exercise directly influence leukocyte mobilization (p>0.05, Table 1).

Accordingly, Gleeson et al. (16) found a significant decrease in IgA, IgG parameters during intensive training or competition periods in elite swimmers, depending on the rest periods, whereas a significant increase in the transitional periods. IgM values were not significantly different in both periods. Djken et al. (11) argue that high intensity and strong physical exercise does not lead to the development of IgA, IgM and IgG regardless of whether or not the individuals actively exercise. In their study, Mackinnon et al. (27) found that there was a significant decline in NK cell activity and saliva IgA levels persisting over a long period of time after exercise, following 2 hours of cycling in racing cyclists. In the study of Kale (21), 12-14% increase in IgG and IgA levels was observed after high severity exercise, and it was reported that this increase was only 7% in IgG after a short distance sprint (100 m) in sprinters. In their studies, McKune and Smith (32) have shown that serum immunoglobulin (IgA, IgM, IgG) consistency can be affected by hormonal secretions and exercise (p>0.05, Table 2).

Based on their study on 23-year-old sedentary individuals and athletes, Chiang et al. (8) evaluated some of the immune system parameters of athletes during intense training period to be suppressed compared to sedentary individuals, while immune system parameters of the athletes during the rest period displayed a stronger reaction. In their study, Moorty and Zimmerman (33) found that leukocyte and lymphocyte counts were not different in individuals undergoing long-term training compared to sedentary individuals, but maximal exercises were found to cause leukocytosis and lymphocytosis in both trained and sedentary individuals. On the other hand, sub-maximal exercises are known to create no difference in trained athletes but are known to cause leukocytosis and lymphocytosis in sedentary individuals. According to Devries and Hosh (10), exercise can

lead to an increase in the number of leukocytes, or circulating white blood cells. Although this effect is temporary, it takes up to 24 hours for the number of white blood cells to return to the normal level after an intense exercise. Moreover, the leukocytosis brought on by exercise can also be related to the intensity of exercise. It has been observed that the functions of the immune system are repressed and cannot function due to the effect of stress hormones such as cortisol and epinephrine after heavy exercise, and upper respiratory tract infections are commonly seen in individuals who are subjected to this type of heavy training program (4). In this sense, the effects of exercise on health and especially on the immune system have been intensely studied in the last 20 years and many studies have been carried out on this subject. The results from these studies have shown that exercise may suppress the immune system as an acute and chronic stressor in athletes, although the clinical link is not yet fully demonstrated (7). In general, after a high-intensity exercise, the total leukocyte count increases by 50-100%. This increase is mainly caused by neutrophils and lymphocytes, with a minor contribution of monocytes. After a long exercise such as a marathon, the increases can reach 200-300% (24). However, within 30 minutes after exercise, lymphocyte counts fall below the pre-exercise level, remain at this level for 3-6 hours, and neutrophil counts continue to increase (42). A moderate exercise leads to less leukocytosis, lymphocytosis, neutrophilia, and lymphocytopenia (22). (p>0.05, Table 3).

In a study conducted on 24-year-old male students, McDowell et al. (31) found a significant increase in IgA and IgG levels after 3 days a week, 20 minutes of intensive exercise for 8 weeks, while no significant difference was found in Ig values as a result of 8-week chronic exercise (60% VO2max). Özgürbüz et al. (36) found that the IgG and IgM parameters of the middle age group performing aerobic exercise was significantly higher than that of similar age sedentary group. Madden and Felten (29) reported that the serum immunoglobulin levels of athletes during intense training periods were significantly lower than sedentary individuals.

Nieman et al. (34) found that athletes undergoing intense training periods were twice as likely to have respiratory tract infections as compared to athletes undergoing low-intensity training programs. Mackinnon (28) reported that during intensive training periods, serum immunoglobulin levels of athletes were lower than those of individuals who do not do any sports, also reporting that acute and chronic exercises had greater effects on mucosal immunglobulin concentrations (IgA, IgG, IgM) relative to total protein secretions. Tyede et al. (44) suggest that the functions of the immunity system are stimulated by a moderate intensity exercise, but severe and longlasting exercises suppress the immune system. Iriadam and Ozbek (19) reported that the immune systems of the athletes participating in their study did not respond significantly to short-term exercises, but demonstrated significant responses that boosted the immune system to long-term exercises, suggesting that long-term exercises could activate the immune system or protect the body from infections (p>0.05, Table 4).

In this study, the pre-test and post-test values of the experimental group were compared, and as a result of intensive training, some measured parameters were observed to increase. This increase is the response of the immune system following suppression induced by intense training. Based on the interpretations of the findings obtained in this study, the results were discussed, and some recommendations were made according to these results. These recommendations are:

Immune parameters can be studied with a low intensity exercise group.

This type of study can be done by comparing different exercise groups.

Acute exercise types can be applied to the same group.

Different immunological parameters such as cytokines, T- and B- lymphocytes can be examined.

The same study can be conducted on a population consisting of females.

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