# AN INVESTIGATION OF THE EFFECTS OF PROLONGED DANCING XERCISES ON SOME HAEMATOLOGICAL PARAMETERS

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**ABSTRACT**

**The Objectives:** The present study was to designed to examine the effects of exhausting dancing exercises on some haematological parameters.

**The Methods:** Male [n=8, age 21.38 ± 2.32 yr, body mass 66 ± 3.81 kg, height 174 ± 2.67 cm] and female [n=8, age 20.88 ± 2.42 yr, body mass 52 ± 4.40 kg, height 163.9 ± 3.98 cm] folk dancers performed prolonged dancing exercises during 3 hours until exhausting; their heart beats were 150-170 beat/minute. Blood samples were taken before and immediately after exercise and analyzed for haematological parameters. Mann-Whitney U tests used for analyzes between males and females, and Wilcoxon's signed-rank test used for differences within groups.

**The Results:** After acute exhausting dancing exercise a leukocytosis was found with a significant increase in neutrophil , lymphocyte, monocyte and platelet counts both in men and women ($p<0.01$). When comparing to the men, women had lower RBC ($p<0.01$), HGB($p<0.001$), HCT($p<0.001$), and NEUT($p<0.05$) levels before and immediately after exercise. But after exercise MCH ($p<0.05$) and WBC ($p<0.01$) levels also showed significant differences between men and women.

**Conclusions:** Long duration exercise induces oxidative stress, tissue damage and inflammation. This might reflect enhanced immune system activities, such as; alteration in the circulation quantity of white blood cells and subsets. Also the inflammatory effect of exercise in tissues may be another stimulant. The present study demonstrated that exercise leads an increase in white blood cell accompanied by an increase in platelet counts.

**Keyword:** Prolonged Exercise, Immune System, Leukocyte, Neutrophil, Platelet, Exhaustion

## Introduction

Athletes frequently encounter muscle soreness and damage after long duration or exhausting exercise (D. Thompson, L. Mcnaughton, 2001; F. H. Fu, and D. A. Stone, 2001). Structural damage to the contractile elements of the muscle fibers occurs after high-intensity or prolonged exercise in which eccentric muscle contractions are used. Increases in muscle damage markers both within the injured muscle and in the blood can act as attractants, initiating the inflammatory process (J. H. Wilmore, D. I. Costill, and W. L. Kenney, 2008; C.M. Deaton, and D.J. Marlin, 2003; P.M. Clarkson, M.J. Hubal, 2002; H. Kuipers, 1994). This inflammation-like immune response is necessary for muscular regeneration and adaptation to physical exercise (Z. Radak, H.Y. Chung, E. Koltai, A. W. Taylor, S. Goto, 2008; P. Tauler, A. Aguilo, N. Cases et al, 2002).

Immune response to exercise depends on the intensity and duration of the exercise. Exercise causes an increase in the muscle and plasma levels of cytokines involved in acute inflammatory responses during and after exercise (J.H. Wilmore, D.I. Costill, and W.L. Kenney, 2008; H. Bruunsgaard, H. Galbo, K.J. Halkjaer, 1997; L. Hoffman-Goets, 1996).

Circulating white blood cells represent a population participate in ongoing tissue surveillance, repair and adaptation. (Tauler,P., Aguilo,A., Cases,N., Sureda,A., Gimenez,F.,Villa,G. et al.,2002). The leukocytes increase in number as a result of exercises that induce muscle soreness. In contrast to moderate activity prolonged or strenuous exercise causes numerous changes in immunity that reflect physiological stress. (S. A. Plowman, and D. L. Smith, 2008; A. Moreira, R.A. Kekkonen, L. Delgado et al 2007; D.B. Pyne, 1994)

During exercise mononucleated cells in muscle are activated by the muscle damage, providing the chemical signal to circulating inflammatory cells such as neutrophils. Neutrophils invade the injury site and release cytokines, which then attract and activate additional inflammatory cells. Neutrophils possibly also release oxygen free radicals that can damage cell membranes (Wilmore, J.H., Costill,D.I., and Kenney, W.L., 2008). Muscle soreness, decreased immune response, vulnerability after long duration exercise or eccentric exercise have been associated with oxidative stress (M. Lamprecht, J. Greilberger, K. Oettl, 2004).

The neutrophil concentration in the bloodstream increases during and after exercise, whereas the level of monocytes only raised during exercise. Large increases in neutrophil numbers are evident for 2-6 hr following high-intensity exercise (S. A. Plowman, and D. L. Smith, 2008; E.W. Petersen and B.K. Pedersen, 2002; L. Hoffman-Goets,1996).

As well, long term exercise results in increased secretion of cortisol, which also could lead to immunosuppresion since cortisol plays a role in maintaining the neutrophilia and lymphopenia after prolonged exercise. Circulating total leukocyte, neutrophil and monocyte counts remained high for 3 hours after exercise, but some immune cell counts decreases below baseline at this stage of recovery. The immunosuppression depends on boths intensity and duration of the exercise that has been undertaken (Z. Radak, H.Y. Chung, E. Koltai, et al, 2008; Natale, V. M., Brenner, I. K., Moldovenau, A. I. et al, 2003). It has been reported that leucocytosis and neutrophilia may be sustained for a few hours during the recovery period; however, the lymphocytosis and elevations in lymphocyte subsets are typically followed by a significant depression during recovery (Y. Chen, S. H. Wong, C. Wong et al, 2008).

The basal level of circulating leukocytes is rapidly increased by physical activity. Despite the increase in neutrophil number following prolonged or high-intensity exercise, there is an evidence that neutrophil function may not respond uniformly. It has been reported that prolonged exercise is associated with enhanced neutrophil function. In contrast, moderate exercises has no effect on immune response (S. A. Plowman and D. L. Smith, 2008; L. T. Mackinnon, 2000; L. Hoffman-Goets, 1996).

An acute bout of prolonged exercise is associated with transient changes in a number of aspects of circulating neutrophil function, including degranulation, the concentration of free radicals in skeletal muscle and oxidative burst (N.C. Bishop, N.P. Walsh and G.A. Scanlon, 2003; J.R. Poortmans, 2003). There is a functional interaction between platelet and neutrophils. It is emphasized that neutrophils regulate platelet activation in atherosclerosis and thrombosis pathogenesis by tissue factor and free oxygen radicals. And the platelets plays a role in inflammatory responses. (A. Yakaryılmaz, 2005).

In this study, we researched the effects of prolonged exhausting dancing exercises on some haematological parameters.

**Material and Methods**

**Subjects:**

Sixteen folk dancer who had not participated any exercise for at least 48 hours, volunteered to participate as subjects after all procedures were explained. Written informed consent was obtained from all subjects prior to their participation and all subjects reported to be nonsmokers, and not taking any vitamin/mineral supplements. Subjects didn't have any health-related problems that might affect the parameters measured. Subjects were asked to refrain from consuming alcohol, caffeine and drugs for 2 days prior to all blood draws.

**Table 1. The physical parameters of the subjects**

| | Subjects (M ± SD) | |
|---|---|---|
| **Variables** | **Females (n=8)** | **Males (n=8)** |
| **Age**(years) | 21.38 ± 2.33 | 20.88 ± 2.42 |
| **Height** (cm) | 163.9 ± 3.98 | 174.0 ± 2.67 |
| **Weight** (kg) | 52.38 ± 4.45 | 66.34 ± 3.69 |
| **IMC** ($kg/m^2$) | 19.48 ± 1.26 | 21.90 ± 0.69 |

IMC, body mass index; M, average; SD, standard deviation; n, number of subjects.

**Exercise Protocol:**

Folk dancers performed prolonged dancing exercises during 3 hours until exhausting; their heart beats were 160-170 beat/minute during high intensity periods and 130-140 beat/minute during low intensity and recovery periods. The exercise applied like an interval training method until exhaustion. Heart rate was monitored by using a Polar coded transmitter, recorded continuously.

**Blood Sampling and Laboratory Procedures:**

Blood samples were drawn pre-exercise and immediately post-exercise in order to analyze the haematological parameters. Approximately 5 mL blood was obtained from a forearm antecubital vein using Vacutainer tubes.

**Data Analysis:**

In order to determine if participants experienced prolonged aerobic exercise would report different count of blood cells compared to pre-exercise, Mann-Whitney U test was performed for analyzes between males and females, and Wilcoxon's signed-rank test was used for differences within groups.

**Results**

Haematological parameters measured before exercise protocol, and male and female subjects are shown in Table 2. It was found that the male values for the red blood cells (RBC), the haematocrit (HCT), the neutrophil (NEUT) and the hemoglobin (HGB) content were significantly higher than the corresponding female values ($p<0.01$ , $p<0.001$, $p<0.05$ and $p<0.001$, respectively).

**Table 2. Haematological parameters of females and males before exercise protocol**

| | Subjects (M ± SS) | | Mann-Whitney U |
|---|---|---|---|
| **Variables** | **Females** | **Males** | **Z** |
| **RBC** (K/uL) | 4.59 ± 0.24 | 5.35 ± 0.30 | 3.151** |
| **HGB** (g/dL) | 12.16 ± 1.89 | 15.68 ± 0.80 | 3.361*** |
| **HCT** (%) | 38.93 ± 4.59 | 47.61 ± 2.12 | 3.361*** |
| **MCV** (fL) | 84.86 ± 9.03 | 89.09 ± 2.62 | 0.630 |
| **MCH** (pg) | 26.54 ± 3.97 | 29.34 ± 1.14 | 1.890 |
| **WBC** (K/uL) | 7.15 ± 2.16 | 8.54 ± 1.27 | 1.115 |
| **NEUT** (K/uL) | 3.94 ± 1.26 | 5.07 ± 1.05 | 0.046* |
| **LYM** (K/uL) | 2.52 ± 0.73 | 2.74 ± 0.50 | 0.528 |
| **MONO** (K/uL) | 0.56 ± 0.23 | 0.56 ± 0.16 | 0.833 |
| **EO** (K/uL) | 0.11 ± 0.07 | 0.14 ± 0.06 | 0.243 |
| **BASO** (K/uL) | 0.03 ± 0.01 | 0.02 ± 0.01 | 0.959 |
| **PLT** (K/uL) | 236.3 ± 53.8 | 273.1 ± 66.9 | 0.279 |

$p<0.05$*, $p<0.01$**, $p<0.001$***

The mean corpuscular hemoglobin (MCH) ($p<0.05$) and the white blood cell counts (WBC) ($p<0.01$) also showed significant differences between men and women in addition to RBC ($p<0.001$) , HGB ($p<0.001$), HCT ($p<0.001$) and NEUT ($p<0.01$) values (Table 3).

**Table 3. Haematological parameters of females and males after exercise protocol**

| | Subjects (M ± SS) | | Mann-Whitney U |
|---|---|---|---|
| **Variables** | **Females** | **Males** | **Z** |
| **RBC** (K/uL) | 4.61 ± 0.22 | 5.43 ± 0.31 | 3.366*** |
| **HGB** (g/dL) | 12.18 ± 1.84 | 15.96 ± 0.90 | 3.371*** |
| **HCT** (%) | 38.85 ± 4.31 | 48.05 ± 2.67 | 3.361*** |
| **MCV** (fL) | 84.31 ± 9.03 | 88.64 ± 3.11 | 0.737 |
| **MCH** (pg) | 26.44 ± 3.98 | 29.45 ± 1.15 | 2.001* |

| | | | |
|---|---|---|---|
| **WBC** (K/uL) | 11.59 ± 3.32 | 15.79 ± 1.96 | 2.521** |
| **NEUT** (K/uL) | 6.93 ± 2.26 | 10.07 ± 1.58 | 2.626** |
| **LYM** (K/uL) | 3.64 ± 1.30 | 4.40 ± 0.92 | 1.050 |
| **MONO** (K/uL) | 0.89 ± 0.27 | 1.16 ± 0.35 | 1.680 |
| **EO** (K/uL) | 0.10 ± 0.08 | 0.13 ± 0.09 | 0.264 |
| **BASO** (K/uL) | 0.03 ± 0.02 | 0.03 ± 0.02 | 0.163 |
| **PLT** (K/uL) | 279.3 ± 67.8 | 340.0 ± 85.3 | 1.472 |

p<0.05*, p<0.01**, p<0.001***

After acute exhausting dancing exercise a leukocytosis was found with a significant increase(p<0.01) in neutrophil , lymphocyte, monocyte and platelet counts in men (Table 4) and in women (Table 5).

**Table 4. Haematological parameters of females before and after prolonged dancing exercise**

| Variables | Subjects (M ± SS) Pre-exercise | Post-exercise | Wilcoxon Z |
|---|---|---|---|
| **RBC** (K/uL) | 4.59 ± 0.24 | 4.61 ± 0.22 | 0.848 |
| **HGB** (g/dL) | 12.16 ± 1.89 | 12.18 ± 1.84 | 0.284 |
| **HCT** (%) | 38.93 ± 4.59 | 38.85 ± 4.31 | 0.070 |
| **MCV** (fL) | 84.86 ± 9.03 | 84.31 ± 9.03 | 1.192 |
| **MCH** (pg) | 26.54 ± 3.97 | 26.44 ± 3.98 | 0.424 |
| **WBC** (K/uL) | 7.15 ± 2.16 | 11.59 ± 3.32 | 2.521** |
| **NEUT** (K/uL) | 3.94 ± 1.26 | 6.93 ± 2.26 | 2.521** |
| **LYM** (K/uL) | 2.52 ± 0.73 | 3.64 ± 1.30 | 2.521** |
| **MONO** (K/uL) | 0.56 ± 0.23 | 0.89 ± 0.27 | 2.524** |
| **EO** (K/uL) | 0.11 ± 0.07 | 0.10 ± 0.08 | 0.710 |
| **BASO** (K/uL) | 0.03 ± 0.01 | 0.03 ± 0.02 | 1.890 |
| **PLT** (K/uL) | 236.3 ± 53.8 | 279.3 ± 67.8 | 2.527** |

p<0.05* , p<0.01**, p<0.001***

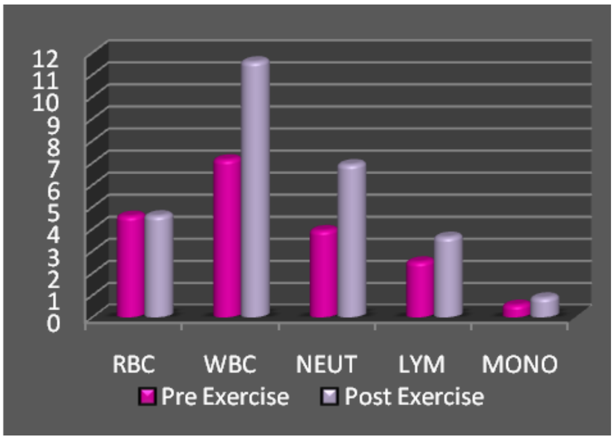


**Figure 1.** Some haematological parameter of females before and after prolonged exhausting exercise. (RBC, red blood cells; WBC, white blood cells; NEUT, neutrophils; LYM, lymphocytes; MONO, monocytes.). There was no significant difference in RBC between pre-exercise and post-exercise values (p>0.05). However, a significant increase was observed in white blood cells and its subsets (p<0.01).

**Table 5. Haematological parameters of males before and after prolonged dancing exercise**

| Variables | Subjects (M ± SS) Pre-exercise | Post-exercise | Wilcoxon Z |
|---|---|---|---|
| **RBC** (K/uL) | 5.35 ± 0.30 | 5.43 ± 0.31 | 1.682 |
| **HGB** (g/dL) | 15.68 ± 0.80 | 15.96 ± 0.90 | 1.689 |
| **HCT** (%) | 47.61 ± 2.12 | 48.05 ± 2.67 | 1.123 |
| **MCV** (fL) | 89.09 ± 2.62 | 88.64 ± 3.11 | 1.183 |
| **MCH** (pg) | 29.34 ± 1.14 | 29.45 ± 1.15 | 0.949 |
| **WBC** (K/uL) | 8.54 ± 1.27 | 15.79 ± 1.96 | 2.521** |
| **NEUT** (K/uL) | 5.07 ± 1.05 | 10.07 ± 1.58 | 2.524** |
| **LYM** (K/uL) | 2.74 ± 0.50 | 4.40 ± 0.92 | 2.521** |
| **MONO** (K/uL) | 0.56 ± 0.16 | 1.16 ± 0.35 | 2.524** |
| **EO** (K/uL) | 0.14 ± 0.06 | 0.13 ± 0.09 | 0.703 |
| **BASO** (K/uL) | 0.02 ± 0.01 | 0.03 ± 0.02 | 1.511 |
| **PLT** (K/uL) | 273.1 ± 66.9 | 340.0 ± 85.3 | 2.524** |

p<0.05* , p<0.01**, p<0.001***

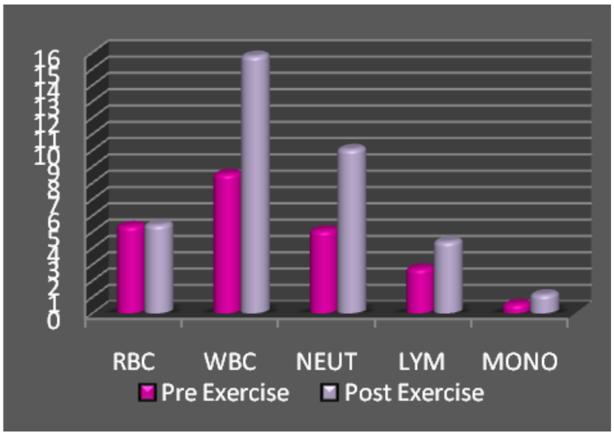


**Figure 2.** Some haematological parameter of males before and after prolonged exhausting exercise. (RBC, red blood cells; WBC, white blood cells; NEUT, neutrophils; LYM, lymphocytes; MONO, monocytes.). There was no significant difference in RBC between pre-exercise and post-exercise values (p>0.05). However, a significant increase was observed in white blood cells and its subsets (p<0.01).

The platelets and the blood immune cells increased noticeably after prolonged exhausting exercise in study group (males and females), significantly (p<0.001). But there was no statistically significant difference in other blood cell parameters such as RBC, HGB and HCT (Table 6).

**Table 6. Some haematological parameters of males and females before and after prolonged exercise**

| Variables | Subjects (M ± SS) Pre-exercise | Post-exercise | Wilcoxon Z |
|---|---|---|---|
| **RBC** (K/uL) | 4.96 ± 0.47 | 5.01 ± 0.50 | 1.706 |
| **HGB** (g/dL) | 13.91 ± 2.29 | 14.06 ± 2.40 | 1.560 |
| **HCT** (%) | 43.26 ± 5.66 | 43.45 ± 5.88 | 0.907 |
| **WBC** (K/uL) | 7.84 ± 1.86 | 13.69 ± 3.41 | 3.516*** |
| **NEUT** (K/uL) | 4.50 ± 1.26 | 8.50 ± 2.49 | 3.517*** |
| **LYM** (K/uL) | 2.63 ± 0.62 | 4.02 ± 1.16 | 3.517*** |

| | | | |
|---|---|---|---|
| **MONO** (K/uL) | 0.56 ± 0.19 | 1.02 ± 0.33 | 3.517*** |
| **PLT** (K/uL) | 254.8 ± 61.7 | 309.6 ± 80.8 | 3.520*** |

p<0.05* , p<0.01**, p<0.001***

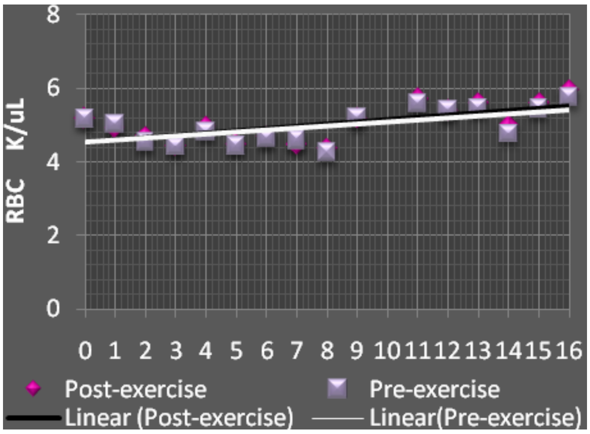


**Figure 3.** Distribution of red blood cell counts before and after prolonged exercise in study group. (RBC, red blood cells). There was no significant differences in RBC values after exercise (p>0,05).

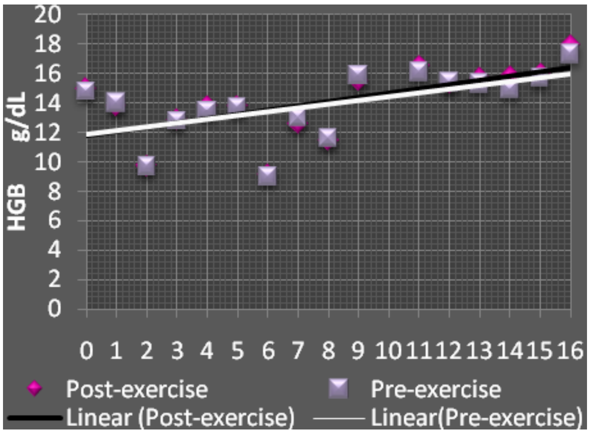


**Figure 4.** Distribution of hemoglobin before and after prolonged exercise in study group(HGB,hemoglobin). There was no significant differences in HGB values after exercise (p>0,05).

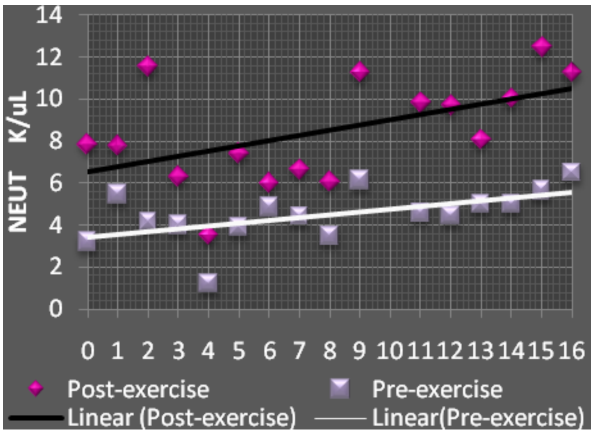


**Figure 5.** Distribution of neutrophils before and after prolonged exercise in study group(NEUT, neutrophils). NEUT increased significantly after exercise (p<0.001).

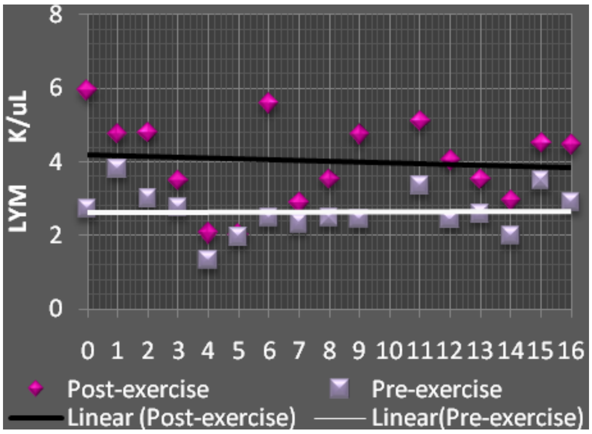


**Figure 6.** Distribution of lymphocytes before and after prolonged exercise in study group(LYM,lymphocytes). LYM increased significantly after exercise (p<0.001).

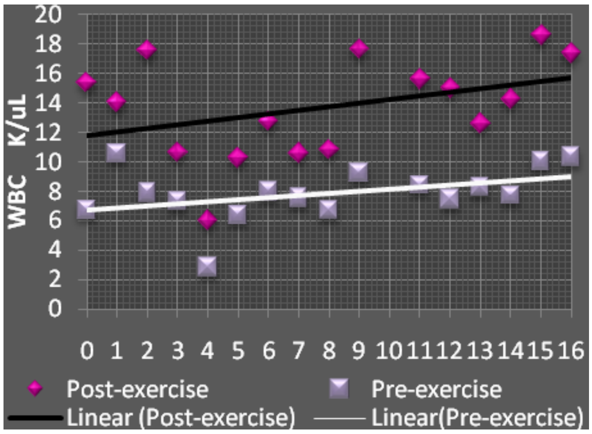


**Figure 7.** Distribution of white blood cell counts before and after prolonged exercise in study group. (WBC, white blood cells). WBC increased significantly after exercise (p<0.001).

## Discussion

The aim of the study was to examine the effects of prolonged exhausting exercises on some haematological parameters.

Changes in immune response to exercise have been reported consistently. Researchers declined that during and immediately after prolonged or heavy physical exercise, the total number of WBC in peripheral blood samples increases (D. G. Rowbottom, K. J. Green, 2000; H. Gabriel and W. Kindermann, 1997; D. C. Nieman, 1997; S. Shinkai, S. Shore, P. N. Shek, R. J. Shephard, 1992).

Before the experiment, we analyzed blood for haematological parameters if there is a difference between men and women subjects. We found that males has higher RBC, HGB and HCT values than females (p<0.01, p<0.001 and p<0.001, respectively). The results indicate that there is no significant difference in WBC, LYM, MONO, BASO and PLT values between men and women.

R. D. Telford, R. B. Cunningham (1991) measured 706 nationally ranked athletes in 12 sports for HGB, HCT, RBC and WBC. They reported that the blood variables was significantly greater in males ($p<0.01$), with the exception of the WBC, which was greater in females ($p<0.01$).

Prolonged or strenuous exercise has been consistently reported to result in an increase of the leukocyte counts (M. G. Nikolaidis, A. Z. Jamurtas, 2009; P. J. Robson, A. K. Bllannin, N. P. Walsh, L. M. Castell, M. Cleeson, 1999; L. Hoffmann-Goetz, 1998; M. Gleeson, A. K. Blannin, D. A. Sewell, R. Cave, 1995; J. S. Wang, C.J. Jen, H.C. K., et al, 1994). In agreement with other investigators, prolonged exercise caused an increase in immune blood cells and platelets counts both in men and women, in our study. Collaterally, progressive increase neutrophil, lymphocyte and monocyte concentrations after 10 km running exercise were noted by Y. Chen, S.H. Wong, C. Wong et al. (2008). In support of our data, researchers reported that prolonged periods of intense exercises results in great neutrophilia and important lymphopenia ( A. Moreira, R.A. Kekkonen, L. Delgado et al , 2007; N. Cases, A. Agulio, P. Tauler, et al. 2005). Similarly, E.W. Petersen and B.K. Pedersen (2002) reported that the neutrophil concentration in the bloodstream increases during exercise and continues to increase post-exercise. The lymphocyte concentration increases during exercise but falls below pre-exercise values following intense long-duration exercise, although not after moderate exercise.

As reviewed by L.T. Mackinnon (2000), the immune function may show differences with exercise type, duration and the intensity. Recent evidences suggest that prolonged periods of intense exercises induced a significantly greater increase in circulating counts for total leukocytes, neutrophils, lymphocytes and natural killer cells. In contrast, moderate exercise training has no effect on immune function (J.C., Quindry, W.L. Stone, J. Ki and C.E. Broeder, 2003)

Our comparison of men and women consistent with previous reports by showing no significant effect of gender on changes in WBC, NEUT, LYM, MONO and PLT values in response to exercise (W. B. Timmons, M.J. Hamadeh, M.C. Devries and M.A. Tarnopolsky, 2005; E.W. Petersen and B.K. Pedersen ,2002). Moreover, N. M. Moyna, G. R. Acker, K. M. Weber et al (1996) reported that progressive incremental exercise increases circulating leukocyte counts and alteration in the number of circulating leukocytes are independent of gender and fitness. Also, W. B. Timmons, M.J. Hamadeh, M.C. Devries and M.A. Tarnopolsky (2005) reported that immunological responses to exercise were similar between men and women. Conformably, P. M. Clarkson, M. J. Hubal (2002) reported that in contrast to the animal literature, which displays that females experience less damage than males, research using human studies suggest there is no difference between men and women.

J.S.Wang, C.J. Jen, H.C. K., et al (1994) demonstrated that the intensity of acute exercise is important factor affecting blood platelet function. They reported an increase of %18.2 after moderate exercise and %24.7 after strenuous exercise in platelet counts of physically active healthy men. D.C. Nieman, D.A. Henson, C.S. Sampson et al demonstrated that exhaustive resistance exercise to muscular failure applied on males, results in a very similar immune response to that associated with intense endurance exercise.

Concurring with several other authors (I,Singh, H. Quinn, M. Mok et al,2006; J. S. Wang, 2004),we found a significant increase in platelet counts after prolonged exhausting exercise. J. S. Wang (2004) concluded that intense exercise promotes the extent of shear-induced platelet aggregation in men and elevation of circulating norepinephrine may be related with this augmentation during intense exercise (H. Ikarugi, T. Taka, S. Nakajima et al, 1999, P.T, Larsson, N.H., Wallen, and P.Hjemdahl, 1994).

High-intensity exercise causes tissue damage, production of stress hormones such as epinephrine and norepinephrine, and increases in the function and quantity of various immune cells. The increased mobilization of leukocytes into the circulation associated with an increased neuroendocrine response. (V.M. Natale, I.K. Brenner, A.I. Moldoveanu et al, 2003; O. Ronsen, ,B.K. Pedersen, T.R. Oritsland, et al, 2001). Previous studies have shown that the increases in leukocyte numbers are related mainly to plasma norepinephrine concentrations in moderate exercises, but with more intense exercise epinephrine concentrations assume a major importance (I. Brenner, P.N. Shek, J. Zamecnik and R.J. Shephard, 1998)

Previous researches suggesting that prolonged exercise activating several components of the inflammatory response. In our study, we observed an augmentation of the exercise-induced increase in total leukocyte, neutrophil, lymphocyte, monocyte and platelet counts independent of gender, concurring with similar studies.

**Conclusion**

In conclusion, these findings suggest that prolonged intense exercise leads an increase in white blood cells and subsets, accompanied by an increase in platelet counts. The immunological responses to prolonged exercise were similar between men and women. This observation confirms the results from studies in humans.

**Acknowledgments**

This article is dedicated to the memory of Prof. Dr. Yaşar SEVİM.

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