

## Effect of 8 Weeks Endurance Training on Immune System Cell Changes with Recovery Period

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### ABSTRACT

**Objectives:** In this study for surveying the relation between exercise and immune cell, we examine the effect of Effect of 8 weeks endurance training on immune system cell changes with recovery period.

**Study design:** Experimental study

**Methods:** participant of this research including health and yang males that were randomized divided into two groups :( ETG) endurance training groups with 15 men; and (CG) 13 men in to control group. Different factor of anthropometric characteristics (i.e. age, weight & height) and also white blood cell (i.e. lymphocyte, neutrophils, monocytes) were experimented. In this study, subjects, runs on a treadmill for 15-30 min at 50 % - 70% maximal Heart rate for 8 weeks, with Venous blood sample was taken at pre, post and at 24- hours and 48-hours after exercise. For data analyze, we used of one -way using repeated-measurements ANOVA, in SPSS12. And also Significance was evaluated as  $P < 0.05$ . In addition, all values are expressed as mean  $\pm$  standard deviation.

**Results:** we found that lymphocyte level increased after 4<sup>th</sup> week and 48h-recovery, but decreased after 8<sup>th</sup> week (Mid-exercise), and 24h- recovery in exercise group. Also levels of Neutrophils and Monocytes decreased after after 4<sup>th</sup> week (Mid-exercise), post- exercise (8<sup>th</sup> week), 24h- recovery and 48h-recoveries in exercise group.

**Conclusion:** These data demonstrate that a period endurance exercise leads to Levels changes of leukocytes after exercise and recovery period, but these changes not significant.

**Key words:** Lymphocyte, Neutrophils, Monocytes, Endurance training.

### INTRODUCTION

Physical activity and Exercise training is a stressful stimulus that induces changes and adaptation in many organs such as skeletal system, endocrine system, pulmonary system, cardiovascular system, immune system and other organs. The immune system is a defense network that plays an important role in human.

Research on topic of exercise immunology area (physical activity and immune system function), approximately began from 1900. Numerous papers in this area were written.

Bryan et al. (2001) reported that more than 600 papers on exercise and immune system published in pub med

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until 2001.<sup>1</sup> And we fund about 3000 paper on exercise and immune system in pub med from 1900 to 2011. Our research in pub med showed that 524 paper of these papers was review paper that publication by famous researchers.

Recently different study demonstrated that, the relation between exercise and immune function was favorite area for researcher. Varieties of published data suggest that immune system function changed after physical activity. Immune system divided into subset, the innate immune and the adaptive immune. Malaguarnera et al (2008), demonstrating that, different componesent of immune system activated against pathogens and also act as the first defense.<sup>2</sup>

Regular physical activity is beneficial for general health. The protective and therapeutic effects of physical activity or exercise and training on several diseases (e.g. cardiac disease, diabetes, and hypertension) are well known.<sup>3</sup>

Type, intense and duration of exercise and physical activity effect on immune system and also alter the immune system response. In this area, Buttner et al. (2007) has suggested that intense training induce decrease in immune system components. Whereas this components increase after moderate exercise.<sup>3</sup>

Exercise has several effects on health in individual. These effect influenced by immune system component. Physical activity can changes immune system component. Evidence from several studies suggested that prolonged endurance training decreased lymphocyte number in circulation and immune cell function.<sup>4</sup>

We know exercise is important effector for immune function. Numbers of circulating immune cells (e.g. lymphocytes, monocytes, and neutrophils) increase after Intense and vigorous training.<sup>5-6</sup> Leukocytes subsets include lymphocytes, monocytes, and neutrophils. We know that Neutrophils is most important subsets of Leukocytes, because 50–60% of the total leukocyte composed of neutrophils. And also, Lymphocytes compose about 20-25% of all leukocytes.

Several organs such as endocrine, autocrine, or paracrine can influence on Leukocytes and increased Leukocytes subset.<sup>7</sup> We know neutrophils and other blood cell increased after exercise, In addition to, leukocyte function changed after increase in circulation immune cell numbers.<sup>8</sup> Evidence from several studies suggested that, generally increased neutrophil functions by moderate physical activity, whereas, prolonged and intense physical activity has negative effect on neutrophil.<sup>9</sup>

The number of lymphocytes may increase during and after physical activity.

Shephard et al. (1997) suggested that increased neutrophil count after physical activity. And also they showed that moderate and intense exercise can increase immune cell numbers. Result demonstrated that moderate exercise induce increase ranges from 20 - 60% in immune cell numbers, whereas, this increase by intense exercise was greater.<sup>10</sup>

Nowadays, surveying the effect of exercise training on immune function became interest topic, because, exercise training has important effect on health by increase in activity and recruitments of leukocytes.<sup>11</sup>

We therefore, interested to see the influence of exercise and recovery period on leukocyte changes after 8 week endurance training.

## MATERIALS AND METHODS

### Participants

To examine the role of endurance training on immune cell change, 28 student males Participate in our study.

Participant of this research including health and young student males between the ages 18 and 24years, were randomized divided into two groups :(ETG) endurance training groups with 15 men; and (CG) 13 men in to control group.

### Anthropometric Measurement

Anthropometric characteristics including: Height, weight, and body mass were measured using standard procedure before the beginning of the study for all participants.

### Research design

This study consisted of a four and eight week endurance training program with recovery period. In this study, subjects, endurance training groups exercised on treadmill 3 times weekly. They running on treadmill for 15-30 min at 50 % - 70% maximal Heart rate for 8 weeks, with pre, post and at 24 h and 48h after exercise, venous blood sample were assessments.

Baseline, anthropometric characteristics were measured and also resting blood samples were drawn for leukocytes subsets assessment.

The participant were randomized divided into two group: (ETG) endurance training group and (CG) control group. We also recorded heart rates from participant in endurance training group for Maximal heart rate calculates.

Before the beginning the training, all participant after a 10-h –fasted period at 10AM, were seated position for 10 min, and then blood sample were collected for the first time. Second blood samples were collected after 4<sup>th</sup> week, and third blood samples were collected after 8<sup>th</sup> week (end of exercise training) and also after 24 and 48- hour's recovery blood sample were collected.

### Exercise program

We use of the endurance training (8week) protocol. Endurance training group participated in endurance training (8week) protocol.

Endurance training (8week) protocol consisted of runs on a treadmill for 15-30 min at 50 % - 70% maximal Heart rate, 3 times weekly for 8 week as shown in Figure 1.

Endurance training includes:

We allowed to each Participant to warm up for 5 minutes, after warm up:

- 1) Week 1= 15 min run on a treadmill at 50 % maximal Heart rate
- 2) Week 2= 20 min run on a treadmill at 55 % maximal Heart rate
- 3) Week 3= 25 min run on a treadmill at 60 % maximal Heart rate
- 4) Week 4= 25 min run on a treadmill at 65 % maximal Heart rate

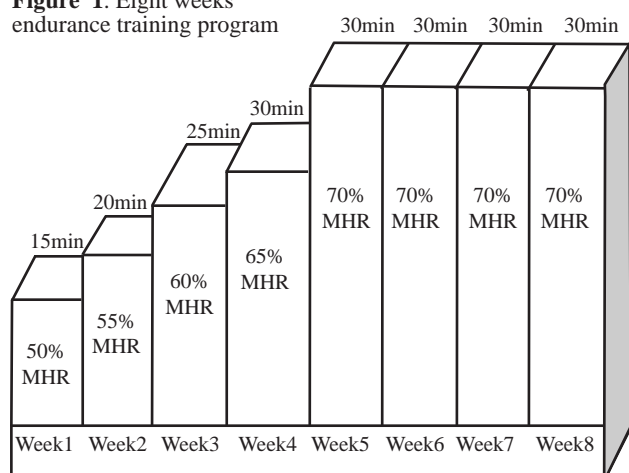
- 5) Week 5= 30 min run on a treadmill at 70 % maximal Heart rate
- 6) Week 6= 30 min run on a treadmill at 70 % maximal Heart rate
- 7) Week 7= 30 min run on a treadmill at 70 % maximal Heart rate
- 8) Week 8= 30 min run on a treadmill at 70 % maximal Heart rate

Maximal heart rate calculates with Karvonen method.<sup>12</sup>

HR target =% Intensity (HR max –HR rest) + HR rest

We selected an eight week endurance training protocol (running on treadmill) because Heesen et al (2003) reported that eight week endurance training can changes immune function.<sup>13</sup>

**Figure 1.** Eight weeks endurance training program



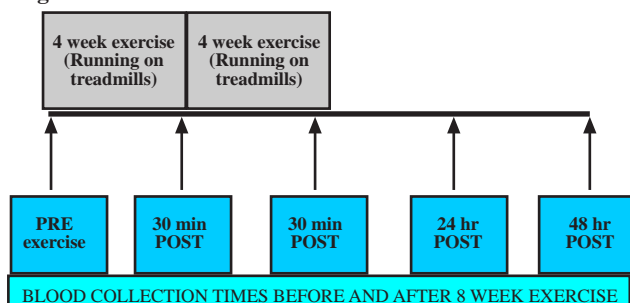
### Blood preparation Times

Subjects came to our experimental laboratory without taking breakfast.

Blood samples were collected at 10 AM. 5ml blood samples were collected at each time.

We collected blood sample at 5 time point: pre –exercise, post –exercise1 (after 4th- week), post –exercise2 (after 8th week), and also after 24 hour and 48- hour’s recovery, as shown in Figure 1.

**Figure 2.** Blood collection times before and after 8 week exercise



For data analyze, we used of one -way ANOVA with Repeated measures test in SPSS12 and also statically significance was accepted at p<0.05. All values are expressed as mean ± standard deviation. Then, he compared the means to see whether or not there is any significant difference between them.

## RESULTS

### Subjects

Table 1 summarizes anthropometric Results for the subjects.

**Table 1:** Anthropometric characteristics (Mean+SD)

	Age( yr)	Height(cm)	Weight( kg)	BMI( kg/m <sup>2</sup> )
Exercise group	21.1± 1.8	176.1± 6.4	65.5± 8.5	21.1±2.4
Control group	19.3±1.2	175.6 ± 6.1	57.8 ± 8.2	19.2±2.2

Values are means ± SE; n (exercise) = 15 & n (control) = 13 subjects; BMI=body mass index

### Leukocyte response to exercise

Table 2 summarizes the impact of exercise and recovery period on total leukocytes, lymphocytes, monocytes, and Neutrophils.

Levels of lymphocyte, monocytes, and Neutrophils changed from Pre-exercise to 48h- recovery.

We demonstrated Levels of leukocyte decreased after 4<sup>th</sup> week (Mid-exercise), post- exercise (8<sup>th</sup> week) and 24h- recovery, but this level increased after 48h-recovery. But in control group level of leukocyte decreased after 4<sup>th</sup> week (Mid-exercise) and increased after post- exercise (8<sup>th</sup> week), 24h- recovery and 48h-recovery.

The result showed that Levels of lymphocyte increased after 4<sup>th</sup> week (Mid-exercise) and after 48h-recovery. But decreased after 8<sup>th</sup> week (Mid-exercise), and 24h-recovery in exercise group, but level of lymphocyte increased after 4<sup>th</sup> week (mid- exercise) and decreased after post- exercise (8<sup>th</sup> week), 24h- recovery and 48h-recovery.

We also find that Levels of Neutrophils in exercise group decreased after 4<sup>th</sup> week (Mid-exercise), post-exercise (8<sup>th</sup> week), 24h- recovery and 48h-recovery. And also level of Neutrophils decreased in all situations in control group.

Data showed that that Levels of Monocyte decreased after 4<sup>th</sup> week (Mid-exercise), post- exercise (8<sup>th</sup> week), 24h- recovery and 48 h-recoveries in exercise group. But in control group this level increased after 4<sup>th</sup> week and decreased in other situation.

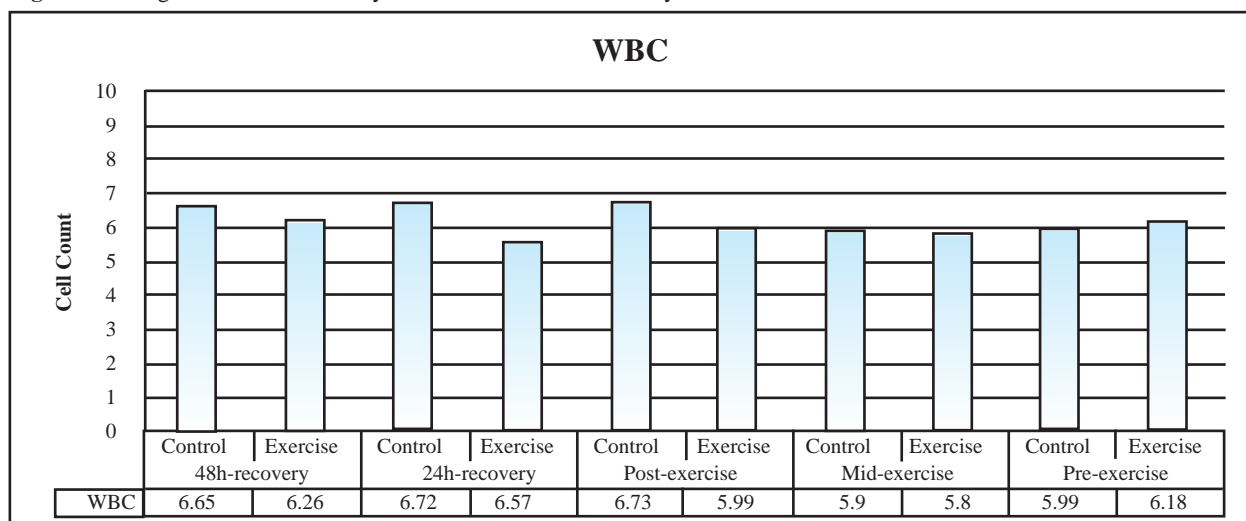
The changes in levels of leukocytes content did not differ in 28 subject under exercise (n=15) and control group (n=13) conditions after 4<sup>th</sup> week (Mid-exercise), post- exercise (8<sup>th</sup> week), 24h- recovery and 48 h-recoveries (Fig.3).

**Table 2.** Leukocyte response to exercise and recovery fase

Time / Variable	Pre-exercise	Mid-exercise	Post-exercise (8week)	24h-recovery	48h-recovery
<b>Total Leukocytes</b>					
Exercise	6.18 ± 0.94	5.8 ± 0.84	5.99 ± 0.97	5.57 ± 1.43*	6.26 ± 1.03**
Control	5.99 ± 1.15	5.9 ± 1.17	6.73 ± 2.67	6.72 ± 2.65	6.65 ± 2.46
<b>Total Lymphocytes</b>					
Exercise	2 ± 0.37	2.16 ± 0.63	1.9 ± 0.44	1.8 ± 0.43*	2.2 ± 0.67**
Control	2.07 ± 0.49	2.22 ± 0.48	1.77 ± 0.46	1.75 ± 0.47	1.76 ± 0.46
<b>Total Neutrophils</b>					
Exercise	3.48 ± 1.03**	3.03 ± 0.77	3.47 ± 0.93	3 ± 0.76*	3.41 ± 0.9
Control	4.14 ± 2.31	3.13 ± 0.91	3.59 ± 1.08	3.54 ± 1	3.51 ± 0.96
<b>Total Monocytes</b>					
Exercise	0.7 ± 0.24**	0.6 ± 0.21	0.61 ± 0.14	0.55 ± 0.12*	0.65 ± 0.14
Control	0.58 ± 0.31	0.6 ± 0.17	0.55 ± 0.13	0.55 ± 0.14	0.56 ± 0.13

\*the lowest level  
 \*\*the highest level

**Figure 3.** Changes in levels of leukocytes after exercise and recovery fase Cell count=  $x \cdot 10^9$



## DISCUSSION

We sought to investigate of effect of 8 weeks endurance training on immune system cell changes with recovery phase in 28 men. The primary finding of the present study was that immune cells number such as lymphocyte, Monocytes, and Neutrophils changed in the circulating blood after 30 min running on treadmill.

Our data revealed that level of some immune cell changed in the circulation after prolonged exercise. But, these changes in the circulating concentration or percentage of some immune cell after exercise not significant.

Our finding extend previous observations,<sup>14</sup> demonstrating that leukocytes recruitment in to active tissue after training (during the recovery phase).and also, Pizza et al (2002) reported that level of leukocytes (e.g. monocytes, neutrophils and lymphocytes) increased after exercise.<sup>15</sup>

The main findings of the study were that leukocytes concentration were not significantly changed following 4 and 8 week endurable training but lymphocyte level was highest changed after 48-h recovery.

The finding of our research is in contrast with previous work demonstrating an increased concentration and percentage of neutrophils. In addition to, several study showed that, immune system cell influenced by Type, intense and duration of exercise. For example, zar et al. (2010) reported that moderate endurance training increased the circulation neutrophils level, and also demonstrated that increase concentration on neutrophils was significant. In this study, subjects (men college judoists), 60 min at 60% maximal Heart rate running on treadmill. Blood samples were drawn pre-exercise and immediately post-exercise.<sup>16</sup>

Hovanloo et al.(2009), demonstrating that Neutrophil concentrations significant increased after running on treadmills (60 min at 60% maximal Heart rate), although running on treadmills with low-intensity (60 min at 45% maximal Heart rate) induce increase in Neutrophil concentrations, but this change not significant.<sup>17</sup>

Pedersen and coworker (2000) also examined the relationship between physical activity and leukocytes subset and observed that increased leukocytes levels during and after physical activity. Furthermore, they showed increase in lymphocyte level during physical activity, but after prolonged physical activity return to pre-value in base line.<sup>9</sup>

Circulating stress/inflammatory cells concentration (e.g. monocytes, neutrophils and lymphocytes subset: T, B, and natural killer (NK) cells) elevated by acute intense training. And also these cells increased during physical activity by increase in recruitment of leucocytes to the peripheral circulation. Whereas, in contrast with moderate physical activity, strenuous physical activity induce increase the circulation lymphocytes number.<sup>18</sup>

Stress hormone have also been studied in relation to exercise and immune cell change by researchers such as Helle et al (2000), who reported during acute physical activity, elevated level of adrenalin, noradrenalin and also cortisol induce increase in lymphocytes and neutrophils level in circulation.<sup>18-19</sup>

## CONCLUSION

In summary, we found that, levels of leukocytes (lymphocyte, monocytes, and Neutrophils), changed in response to 8 week endurance training and increased after 48-h recovery compare to post exercise. Post-exercise levels of lymphocyte, monocytes, and Neutrophils decreased in compare to base line level. Observed changed levels of leukocytes could be involved in the process of adaptation of human organism to physical training. This change, maybe, influenced of duration, intense and type of exercise.

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