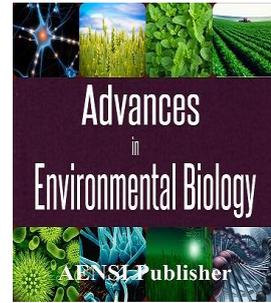




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The Effects of Severe Interval Activities on the Cytokines among Male Soccer Players

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ABSTRACT

This research was done for surveying on the adopting effects of a selected periodic exercise on the safety plasma cytokines among male soccer players that was done after an exhaustive exercise bout. An exercise planning was tested close to the exhaustion in this research to survey on this effects activity on the cytokines & safety cellules. The static sample was 44 persons from male soccer players in the first division League of country & they were place in the two groups; experimental & controlled groups. The experimental group were in the mean weight 77.25±6.99 Kg, height 181.45±2.71 Cm & Oxygen consumption maximum 34.18±2.75 (ml/Kg in minute). Controlled group were in the mean weight 79.69±3.82 Kg, height 179.81±4.82 Cm & Oxygen consumption maximum 34.1±3.79 (ml/Kg in minute). For the first, subjects did exhaustive activity. Then the subjects divided in to the 2 groups. Experimental group exercised for periodic 8 weeks activities. After 8 weeks, two groups did periodic activities again, after & before each activity in two steps, blood samples were taken. Mean, tables were used in the descriptive static part & variance analysis test & continuum measurement in the deductive static part. The findings of research showed that doing a course exercise after periodic activity after exhaustive activity didn't cause to the changes in IL1, TNF density, but changed the number of safety cellules & CD8, CD4 anti-gens. Selective periodic activity caused to the strengthen & positive adoption in the innate immune system cells in the experimental group as decreasing in the number of safety cellules & CD4 in experimental was lower than controlled group & increasing safety cellules & CD8 in experimental group is higher than controlled group

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INTRODUCTION

Sport activities make changes regarding to the time of implementation, intensity or kind of contraction involved in activity, production & proliferation of cellular & non-cellular components (cytokine) of the immune system & they are a subset of constituent [1]. Sometimes caused to increasing or decreasing function, activity & mediated proliferation of cells & molecules [2].

Gabriel (1993) reported the changes of number of leukocytes in less & more than 45 minutes [3] also the results of researches showed that light exercise to 0.85 anaerobic threshold & less than two hours, in response to hormonal & immune cell density makes minor adjustments [4]. Activity with different maximum oxygen using (65%, 30%, 75%) in different times (30 minutes, 60 minutes, 120 minutes) showed that the most increasing of CD4 decreasing in the most longest activities (120 minutes) is obvious (5). In the other research that was done by Kendal & *et al* [22]. Increasing the T & CD4 cellules were seen in the readiness persons, but CD8 had meaningful increasing among groups that had lower fitness. Maximum sport activity affects on the subcategory of lymphocytes & 3IL1. Same researches were done on the 11 cyclists. The meaningful increasing showed in CD4, CD8, IL1 & meaningful decreasing was showed in CD4/CD8 [6. 7]. He reported that severe exercise caused to the increasing the inflammatory, proinflammatory & triggering cytokine responses of IL6, TNF & against receiver of IL1 in blood [8]. Other results showed that doing the severe activity to 100% aerobic threshold & its continuum to many times be more changed than light activity & monocytes caused to more IL6, TNF & IL1 secretion [6]. A study was done on the gymnasts & the researchers found that immune system

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function decreased among practiced heroes [9], but several & long time researches showed that there was inverse association between physical activity & inflammation signs. In this research pointed that regular activity has inflammation effect & caused to the anti-inflammatory [4] & then caused to modulate the immune system [10]. Doing the moderate activity in 5 days of week as 30-45 minutes caused to the appropriate changes in immune system & decrease 25-50 percents of daily disease than in-active group [3]. Fleg (2005) showed that sport activity has anti-inflammatory effect & also showed that 6 months regular exercises caused to the decreasing cytokines; TNF & IL1 [4]. Glessen (2007) showed that the effects of anti-inflammation of sport activity on the immune system. The research was done on the 8 men that 5 of them periodic exercise 8 hours in a week & the results showed that IL6 was increased in respondent to exercise. Although the immune system evaluation was not successes in all studies, for example; TNF & IL1 cannot be survivable after matron competition (1986) & other severe activities [10, 11]. After 2:30 hours on the treadmill 75% maximum oxygen usage was seen increasing the IL6 inflammation cytokines among heroes, but other cytokines such as; TNF, IL1 didn't have any significant change. Regarding to the controversies [4] needs to for increasing healthy, selected the best exercise plan. Each researcher, studied on the immune system part (innate, acquired & humoral) & sometimes surveyed on the immune cellules or in-immune cellules. In the last researches used men subjects & lower researchers studied on the effects of periodic activities on women immune system. Regarding to the effects of hormones, specially, cortisol, testosterone & prolactin on the changes of immune system [2, 4, 8] & paying attention to the effects of sport activity on the men was done different researches [3, 4, 6, 7]. The men immune system is surveyed than periodic plan in this research.

Football is a periodic sport with high intensity that players suffered psychological pressures during training & competition. Information is low in about responses of football players' immune system & most of researches were surveyed on the effects of severe of sport activity on the immune system, activities such as; long-term & continuum activities or using short-term protocol. So the goal of this research is determining the effects of severe periodic activity on the cytokines among men football players & the researcher tried to answer this question whether severe periodic activity affects on the cytokines among men football players or not?

Methodology of research:

Static method:

This research is practical & it is as semi-experimental with the pre-test & post-test plan. In this research was used static methods; descriptive, mean & tables. First, specified the nature of distribution by using Clomogrov-Smirnov test. Then mean of each dependent variables between two experimental & controlled groups. The p-value was determined as $\alpha \leq 0.05$ & also was used SPSS version 19 & Excel for calculating & reported the information.

Samples:

For selecting the subjects were reported the goal of research & the method of implementation of it to the officials participating teams in the First Division championship in Isfahan. 67 persons of them selected & then tested. 44 persons of them were placed r&omly in the two controlled & experimental groups after completed specific sports-medicine questionnaire. 24 hours before began the research, personal characteristics of subjects were measured & recorded; weigh, height & body mass index by medical scale & the percentage of subjects' fat was measured by caliper.

Research protocol:

It should be noted that the participants were in the pre-season tournament & Copenhagen test was done after 8 weeks of continuous exercise in the players in readiness step & in this time controlled group did daily activity. Before doing protocol was taken blood samples from two groups. Then experimental group implemented the protocol & controlled group did not any activity. Immediately 3, 24 hours after final of protocol was taken blood samples. Copenhagen soccer test was used to simulate a football match. This test included two 45 minutes courses with 15 minutes rest among them, which designed by football activity model. Also the maximum of oxygen usage, heart rate & the time of implementation of exhaustive activity were recorded.

Periodic activity plan was selected as first heart rate reserve all participants were determined by the Karvonen method;

- 1- Determining the maximum of heart rate by using equation; age-22-maximum of heart rate
- 2- Determining the resting heart rate
- 3- Subtract resting heart rate from maximum heart rate (heart rate reserve) & 60% & 80% of resting heart rate; add to two numbers to obtain the limitation of heart goal. Regarding to the participants conditions in terms of heart rate reserve percentage was specified intensity & during time of activity & periodic planning

Blood sampling: first; anti-cubital vein of the right h& of the subjects were taken 9 ml blood sampling in the sitting status. Then, blood serum samples of isolated erythrocytes were stored at -80°C by using centrifuges. Cytokines IL1, TNF was surveyed by human's kits by ELISA method & also the numbers of immune cellules were surveyed by flowcytometry in laboratory.

Blood sampling analysis & method of blood measurement;

Cytokines IL1 & TNF (non-cellules safety variables) & CD4, CD8 (cellules variables) were surveyed in this research. Measuring the IL1 & TNF were done by laboratory kits, auto analyzer cobs mira (Roch) with OD scale or MedSystem Bender, made in Ostrich based on provided protocol in kit. The sensitivity of TNF kit was 2.3 Pg on ml & sensitivity of IL1 was 0.3 Pg on ml. Evaluation of changes of CD4 & CD8 were done by Flowcytometric analysis of three colors. Flowcytometric analysis was done by Pas model made in Norway in DAKO Company with the scale of number & percentage of cellules in 10000 cellules. For surveying the CD4 of Fluorescin isothiocyante & for surveying on the CD8 of Phycoerinthrin that react with cellular level of anti-genes specially. Other measurement tools included; interior questionnaire for awareness of general conditions of participants' healthy, medical scale model Seca made in Germany, Caliper Yagami made in Japan was used for determining the percentage of fat as Jackson & Polack 's method, the Rotary used for separate the plasma & serum of electric treadmill machine model T9700 HRT used for Boros's test made in America.

Findings of research:

The personal characteristics of subjects was seen in table 1 & 2 in pre-test & post-test (before start the research & in the final of research after 8 weeks periodic activity among experimental group)

Table 1: demographic characteristics of controlled & experimental groups (mean \pm st&ard deviation).

Row	Experimental group	Controlled group
Age (year)	21.60 \pm 1.7	24.60 \pm 4.71
Height (Cm)	181.46 \pm 2.73	179.46 \pm 4.73
Weight (Kg)	79.27 \pm 7.01	76.27 \pm 3.01
Fat (percentage)	14.99 \pm 0.8	12.769 \pm 0.48
Maximum of usage oxygen (ml on Kg minute)	33.98 \pm 2.76	36.98 \pm 3.76
The index of body mass (Kg on height square)	22.01 \pm 2.49	21.41 \pm 1.29
Maximum of heart rate in periodic activity (number in minute)	181 \pm 12	186 \pm 5

Table 2: Static comparison variables of personal characteristics.

row	Experimental		controlled	
Weight (Kg)	0.001	1.000	-1.000	0.351
Fat (percentage)	-1.065	0.351	1.964	0.90
The maximum of oxygen usage (ml on Kg on minute)	-6.662	0.001**	0.505	0.627
Body mass index (Kg on height square)	0.200	0.984	-0.823	0.438
Heart rate (number in minute)	3.054	0.014*	0.848	0.425

$P \leq 0.05$ * is meaningful

$P \leq 0.001$ * is meaningful

Regarding to the above results & obtained number value of t, the maximum oxygen usage & heart rate in subjects was showed significant difference in controlled group subjects ($p=0.001$, $p=0.14$, $p=0.017$). Mean & st&ard deviation of IL1, TNF,, CD4, CD8 of blood samples of each group, after 8 weeks periodic activity (before & after doing second test of exhaustive activity) & after 8 weeks periodic activity (before & after second exhaustive test) in table 3. Periodic activity was used as sport activity that caused to the changes in immune system, so can be surveyed on the effects of selective periodic activity.

Table 3: the static of immune system variables before & after exercise.

stage	Group	Mean \pm st&ard deviation			
		IL1 (pg/ml)	CD4 Number in 10000	CD8 Number in 10000	TNF (pg/ml)
Before exercise	The results of pre-test	0.001	32.10 \pm 5.36	22.8 \pm 5.10	0.22 \pm 0.59
		1.96 \pm 5.49	30.99 \pm 2.98	20.49 \pm 3.11	0.42 \pm 0.61
	The results of post-test	0.001	26.7 \pm 5.03	22.98 \pm 4.71	0.98 \pm 2.33
		0.001	24.26 \pm 3.21	20.22 \pm 3.1	0.001
After exercise	The results of pre-test	0.001	44.69 \pm 6.7	19.99 \pm 2.58	0.001
		0.001	43.62 \pm 4.1	19.29 \pm 2.1	0.12 \pm 0.34
	The results of post-test	0.001	38.28 \pm 6.51	21.6 \pm 2.68	0.001
		0.001	33.71 \pm 4.1	19.51 \pm 2.06	0.001

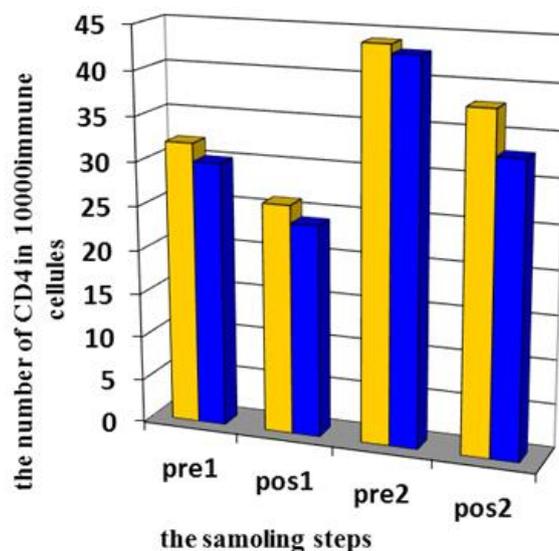


Fig. 1: CD4 cellules changes in controlled & experimental groups.

As we seen in the figure 1 after doing severe activity at the first of research, CD4 was decreased in the two groups. In the final of 8 weeks, CD4 was increased but after doing sever activity in each group was along with decreasing. The meaningful of this change was specified in table (4).

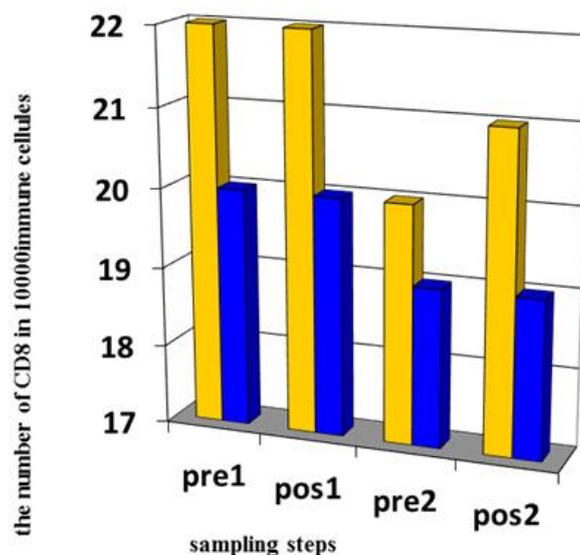


Fig. 2: The CD8 cellules changes in controlled & experimental groups.

As the figure 2 showed; after doing severe activity at the first of research, the CD8 in controlled group was increased & it was decreased in controlled group. After 8 weeks, CD8 was decreased in two groups, but after severe activity, it was increased. Significant was specified in table 4. After obtaining mean of research variables, the obtained data was surveyed by variance analysis test & continuum measurements. The results of test were showed in table (4).

Table 4: The results of variance analysis test in continuum measurements.

variable	F	P
IL1	1.19	0.31
TNF	0.55	0.64
CD4	3.69	0.02*
CD8	4.77	0.007*

P<0.05* was meaningful

P<0.001** was meaningful

Discussion & Conclusion:

The results of research showed that 8 weeks selected exercises didn't have meaningful effects of the density of IL1 that it was same as Pedersen (2000), Suzuki (2004) & Starkie (2001) [10, 15,14,13]. Some of researchers reported that after doing exercise planning on subjects, IL1 changes can be evaluated & surveyed [10, 11], Starkie & Ostrowski (1998) were not seen in IL1 changes in plasma [14, 24]. Of course,

Most of severe periodic activity & severely damaging eccentric contraction didn't make any changes in IL1 [17, 11, 18, 20]. The effects of sport activity were not visible on the immune system, but one of the physiological benefits of sport activity was, decreasing the IL1 production [4]. The various results can be related to the kind of physical activity, intensity & time of exercise. Other probability was a break of myofibrils & muscle protein breakdown [10]. Nemeth of IL1 changes can be known as the results of metabolic responses [20]. The paradox is due to the periodic activity, because this research was used periodic activity increasingly, like a marathon workouts, intense, long & do not be repeated & against of eccentric contractions, with no tissue damage & destruction to the myofibrils [17]. Controlling the nutrition was not done in this research, but the subjects were not faced to the decreasing the energy resources decreasing & also didn't do any metabolic motivation. Also the subjects had regular physical activity & surveying of fitness & maximum of oxygen usage & their adaptation to exercise, because immune system for non-athletes & non-active & sedentary persons showed more reaction. Regarding to the significant of t-value of heart rate, time of exhaustive activity & maximum of oxygen usage in controlled group & can be said periodic activity had effects. Due to the fact, if evaluation of IL1 in muscle or urine or using the human kits of IL1 measurements with higher sensitivity were used in the nano grams per milliliter, so can be surveyed the cytokines changes. The results of research showed that, selective periodic activities didn't have any significant effect on TNF changes, the findings Derent (1995), Starkie (2001) showed the same results [2, 11, 15]. Nemeth (2002) & Febrio (2002) stated that these cytokines were visible just after long term sports or sports that hurt to the skeletal muscles [18, 4, 11, 20]. &row's (2003) thesis showed that TNF can be sensitive glycogen is sensitive to very low resources that visible in most of severe competitions [23] the results of Febrio (2002) showed that contractile activity continued for several hours didn't caused to the TNF increasing significantly [11]. Dernet (1995) cannot be found the TNF after periodic exercise [14, 15] Gorzi (2006) didn't see any significant effects on TNF changes after 10 weeks periodic exercises [21]. The exercising methods & subjects' characteristics were obtained of effective factors on results. It seems that low of activity intensity make a kind of positive adaptation in subjects that the level of cytokines & their changes decrease. Because the subjects did sport regularly, & have not been seen significant changes in the levels of cytokines. Tissue damage are due to heavy activity, catabolic response, production of reactive oxygen species after exercises, over exercising syndrome & hypertension, increased cytokine TNF follows that probably occurred in this study.

After 8 weeks selected periodic exercises, happened decreasing the CD4 in experimental group & increasing the CD8. The results of this research is same as Naiman (1999) & Pederson (2000).It has been reported that exercise of moderate intensity exercise, five days a week for 30 to 45 minutes, make appropriate changes in the immune system [22, 23]. Nelson (1996) showed that increasing the CD3 & CD4 was made by the effects of rowing exercise with maximal exercise & ergometer [10]. Pool & Axford (2001) believed that CD8 was increased based on changes in lymphocyte sub-categories. Lankester (2005) saw the decreasing of CD4 & CD8 based on activity [22, 5]. Increasing of CD4 & CD8 during the exercise the recruitment of lymphocytes is all subcategories, but fall after activity & recovery to the first status [24, 25]. It looks intense exercise suppresses the immune system & reduce the CD4 [18]. This reduction may be temporary & depends to the amount of physical fitness [5]. Maybe showed that respond to an intense activity, t-lymphocytes subcategory of the circulatory system, the two-phase response which indicates that the increase in the number of cells at the time t & decreased activity when they return to the initial state [22]. Significant decreasing CD4 & significant increasing CD8 were signs that can be seen in healthy individuals & athletes, after the workout. The basic elements of the immune system in healthy individuals who have regular activities, there are not many changes. But the heroes, who are affected by intense exercise, have lower levels of lymphocytes. Due to various factors affecting the assessment of cytokines & immune cells such as the intensity & duration of activity, time to return to the initial state, subjects, physical fitness, sensitive kits, methods of evaluation (peripheral blood, urine, biopsy), the above results show that exercise can be said to further strengthen, stimulate the immune system in the experimental group were less consistent.

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