

**Original Article** 

# Effects of a Selected Training Program on Motor Performance and Serum Levels of IL-4 and IL-6 in Elite Water Polo Players

Seyed Mojtaba Hoseini D (PhD Candidate) Department of Physical Education, Neyshabur Branch, Islamic Azad University, Neyshabur, Iran

Amene Barjaste Yazdi Department of Physicsl education, Neyshabur Branch, Islamic Azad University, Neyshabur, Iran

Rambod Khajei D (PhD) Department of Physical education, Neyshabur Branch, Islamic Azad University, Neyshabur, Iran

Amir Rashid Lamir () (PhD) Department of Exercise Physiology, Faculty of Sport Sciences, Ferdowsi University of Mashhad, Mashhad, Iran

**Corresponding author:** Amene Barjaste Yazdi **Tel:** +989151514174

Email: barjaste.a7@gmail.com

Address: Islamic Azad University, Neyshabur, Iran

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#### Abstract

**Background and objectives:** In recent years, exercise immunology studies have focused on key components of immune function, such as immune cells, immunoglobulins, and messenger molecules, as well as the effects of environmental and exercise factors. The aim of this study was to evaluate effects of 8 weeks of selected training on serum levels of interleukin (IL)-4, IL-6, and motor performance in elite water polo players.

**Methods:** For this purpose, 20 male water polo players (mean age: 23 years) were randomly divided into an experimental group (n=10, selected water polo training) and a control group (n=10, traditional water polo training). The selected water polo training was performed for 8 weeks, 6 sessions per week. The intensity of training was controlled using maximum heart rate (80-100%) and increased by 5% every week. Water polo performance tests included a record of 50 meters swimming, 100 meters high swimming, 200 meters' breaststroke,  $6 \times 50$  meters swimming, and 400 meters swimming. To evaluate the serum levels of IL-6 and IL-4, venous blood (5 ml) was taken in the pretest and posttest stages. The serum levels of IL-4 and IL-6 were measured by sandwich enzyme-linked immunosorbent assay. Mean values were compared using the t-test, at significant level of 0.05.

**Results:** The selected water polo training significantly changed IL-4 levels in elite water polo players (p<0.05). However, IL-6 levels and performance of the subjects did not change significantly.

**Conclusion:** The selected swimming exercises could relatively improve swimming stagnation of water polo players. However, due to the limited knowledge in this regard, there is a need for further research.

Keywords: <u>Anti-Inflammatory Agents</u>, <u>Interleukin-6</u>, <u>Interleukin-4</u>, <u>Water Sports</u>.

## **INTRODUCTION**

In recent years, many studies have focused on exercise immunology and key components of immune function, such as immune cells, immunoglobulins, glutamine, and cytokines, as well as the influence of environmental, nutritional, and exercise factors (1). Cytokines are soluble agents in the immune system. In general, these proteins are divided into two major groups: proinflammatory [interleukin (IL)-18, IL-1 beta, and IL-6] and antiinflammatory (IL-4, IL-6, and IL-10) cytokines. Proinflammatory cytokines are involved in the development and progression of inflammation, while anti-inflammatory cytokines are secreted in response to inflammation to limit and reverse the inflammatory processes (2). On the other hand, IL-6 is a dual-function cytokine that has a different effect on cell types due to the nature of its receptors (3). It is mainly secreted by immune cells, vascular endothelial cells, adipocytes, and muscle fibers, and its receptor is present in various cells, including most leukocytes, hepatocytes, adipocytes, and epithelial cells (4).

Skeletal muscles have the capacity to express several cytokines, including IL-6, IL-8, and IL-15, collectively called myokines (5). These are in fact cytokines produced by muscle cells that link exercise to inflammation (6). Myokines facilitate several cellular responses to exercise, such as angiogenesis, suppression of proteolysis, and regulation of muscle glycogen (7). Induction of IL-6 and IL-4 secretion following exercise may be due to skeletal muscle injury and inflammation (8).

Some studies indicated that strength training increases muscle mass and contractile proteins, which in turn increases muscle strength and capillary damage. Exercise programs with different intensities and durations affect cytokine levels (9). There are conflicting findings about the appropriate exercise intensity to achieve the anti-inflammatory effects of physical activity (10). Exercise with moderate or high intensity/duration has a greater anti-inflammatory effect compared with low intensity exercise (11). The relationship between intermittent exercise and IL-4 and IL-6 changes has not been studied extensively. and only nutritional and pharmacological interventions on patients have been emphasized upon (12). In the present study, we investigate response of inflammatory factors to intense intermittent training in water polo athletes, without nutritional and pharmacological considerations. This study evaluates effects of 8 weeks of selected training on serum levels of IL-4 and IL-6 in elite water polo athletes.

## MATERIALS AND METHODS

This was a practical and quasi-experimental study with a pretest-posttest design. The statistical population included all water polo players in Mashhad, Iran. Inclusion criteria included age of 20-25 years, at least 4 years of experience in continuous training, and participation in the national water polo premier league. Overall, 20 athletes were purposefully and accessibly selected and divided into a selected water polo exercise group (n=10) and a traditional water polo exercise group (n=10). After explaining the objectives and methods of the research, written consent was taken from all participants. It should be noted that all subjects were healthy at the time of the study and were not on any medication.

Water polo performance tests included 50meter swimming record, 100-meter high swimming, 200-meter breaststroke,  $6\times50$ meter swimming, and 400-meter swimming. Selected water polo exercises were performed for 8 weeks (<u>Table 1</u>). The Karvonen method was used as a percentage of the maximum heart rate (80-100%). In addition, the intensity of training was increased by 5% every week (<u>13</u>).

To evaluate the serum levels of IL-6 and IL-4, 5 ml of venous blood were drawn. After separating serum, the levels of IL-6 and IL-4 were measured by sandwich enzyme-linked immunosorbent assay (ELISA) method, using commercial kits (ab46027 and ab46087, Eastbiopharm Company, USA). In this study, The Kolmogorov-Smirnov test was performed to assess normality of data, and t-test was performed to compare mean values at a significant level of 0.05.

# RESULTS

<u>Table 2</u> shows the characteristics of the participants in the study groups.

Day of the week	General warmup	Special warmup	Main exercise	Cool down	
Saturday	warm-up exercises, 5 minutes jogging	200 meters free, 4×50 meters mixed, 200 meters mixed feet	10 × 100 meters swim with 3 minutes rest, swim head up with the ball, work with the ball, shoot, and pass	2×200 meters chest crawl, 2×100 meters crawl back with frog legs	
Sunday	warm-up exercises, 5 minutes running	200 meters free, 4×50 meters mixed, 4×50 meters mixed feet	6 × 200 meters free swimming with 4.5 minutes rest, 10 × 50 meters swimming with 1.5 minutes rest, work with the ball	400 meters chest crawl, 100 meters crawl back	
Monday	warm-up exercises, move the water polo shot with the cache in different directions	4×50 meters mixed, 4×50 meters mixed feet	4 × 15 (minutes) water polo game (without restarting from the middle of the field)	400 meters freestyle	
Tuesday	warm-up exercises	4×50 meters mixed, 4×50 meters mixed feet	5× 400 meters (with insole + twin) chest crawl, 5 × 400 meters (50 butterflies + 300 chest crawl + 50 butterflies) with fin	400 meters freestyle	
Wednesday	warm-up exercises, 5 minutes running	4×50 meters mixed, 200 meters mixed feet	3×15 (minutes) shallow water polo game without dangerous errors without water polo rules, rescue swimming with z- shaped ball and attack and counter-attack by shooting	200 meters chest crawl, 100 meters crawl post, 100 meters feet	
Thursday	warm-up exercises, 5 minutes running	4×50 meters mixed, 100 meters mixed legs	(100-200-300-400 meters) breast crawl, (100-200-300-400) meters breast crawl, work with the ball, shoot, practice team tactics	200 meters chest crawl, 100 meters crawl post, 100 meters feet	
Friday		Official game with	ith complete rules		

Table 1- Details of the selected water polo training program

Given the normality of data, parametric tests were used. Results of the independent and paired t-tests are presented in <u>Table 3</u>. Based on the results of the paired t-test, the level of IL-4 and IL-6 increased significantly

in the experimental group compared to the control group. The selected water polo training did not change the swimming records of 50 meters, 100 meters, 200 meters, and 400 meters in experimental subjects.

	Group	Number	Age (years)	Height (cm)	Weight (kg)
1	Selected water polo exercises	10	$23.17\pm2.51$	$182.8\pm 6.32$	$91.2\pm6.32$
2	Traditional water polo exercises	10	$23.02\pm2.67$	$181.1\pm5.58$	$88.6 \pm 5.96$

Given the normality of data, parametric tests were used. Results of the independent and paired t-tests are presented in Table 3.

Variable	Group	Pretest	Posttest	Changes intergroup		Changes intergroup	
				t value	Significance level	t value	Significance level
IL-6 (pg/ml)	Experimental	70.14±2.19	20.71±2.41	4.871	0.062	34.4	*0.00
	Control	22.14±4.32	22.49±4.48	1.922	0.087		
IL-4 (n/gl)	Experimental	19.05±12.32	31.88±10.62	12.345	0.00	0.640	*0.00
	Control	32.18±10.32	13.73±8.60	0.246	0.811		
50 m record	Experimental	82 .1.02±72	27.76±0.78	6.29	0.001	1.32	0.21
	Control	0.53±10.29	28.53±0.52	2.09	0.09		
100 m (sec)	Experimental	3.48±61.62	59.87±2.55	6.04	0.001	1.29	0.22
record	Control	1.89±00.64	62.16±1.60	3.37	0.02		
100/200 m (sec) record	Experimental	5.22±00.161	158.71±3.68	1.48	0.18	1.060	0.31
	Control	3.60±16.164	164.33±5.39	0.09	0.92		
400 m (sec) record	Experimental	13.30±85.358	353.00±13.49	4.46	0.004	0.34	0.74
	Control	8.67±00.362	356.83±6.73	3.18	0.02		
6 ×50 m (sec) record	Experimental	2±71.34.49	33.85±2.47	2.52	0.045	0.99	0.32
	Control	2.07±50.35	34.66±1.50	2.71	0.042		

Table 3-Comparison of intragroup and intergroup changes of variables in elite water polo players

Based on the results of the paired t-test, the level of IL-4 and IL-6 increased significantly in the experimental group compared to the control group. The selected water polo training did not change the swimming records of 50 meters, 100 meters, 200 meters, and 400 meters in experimental subjects.

#### DISCUSSION

The present study examined the level of IL-4 and IL-6 in water polo athletes. In this regard, a previous study showed that 12 weeks of swimming training reduced IL-6 in mice (14). The mentioned study reported that 8 weeks of endurance swimming training increased IL-10 and decreased IL-6 in heart tissue (15). In our study, 8 weeks of selected training had no effect on motor performance of elite water polo players. In this regard, a precious study investigated the effect of 4 weeks of swimming training beta-alanine and supplementation on repetitive speed performance in 22 elite male water polo players (16). Before and after receiving the supplement, the subjects performed repetitive speed performance tests in 30 minute-sessions with an interval of 30 minutes (17).

The results showed no significant difference between the experimental and control groups as a result of the first iterative speed performance. However, this difference was statistically significant as a result of the second iterative speed performance test. Another study investigated effects of using sodium bicarbonate and beta-alanine on 100-meter freestyle swimming in 30 elite male swimmers (18). Subjects were divided into four groups: placebo, beta-alanine supplementation, sodium supplementation, bicarbonate and betaalanine-sodium bicarbonate supplementation. The intervention lasted 2 weeks, and the subjects performed 100-meter swims three times. The results showed no difference between the groups in the first 100-meter swim, but after the second and third swims, lactate levels in the beta-alanine and betaalanine-sodium bicarbonate supplementation groups increased compared with the control group. In the mentioned study, the subjects were not elite swimmers, but they were used in many research sources, the athlete's progress at the professional level reaches a functional plateau, which at this stage is very difficult to improve performance. In other words, the stagnation of swimmers in the selected training group increased slightly compared to the control group, although it was not statistically significant.

Other important factors in achieving these results include the nature of the sport of water polo and its basic required skill i.e. swimming  $(\underline{19})$ . In addition to physical fitness, the technical performance of the swimmer also plays a major role in improving overall

performance.

## CONCLUSION

Overall, the results showed that 8 weeks of selected water polo exercise could significantly change plasma IL-4 level of elite water polo athletes. Regular aerobic exercise appears to reduce this by reducing sympathetic stimulation and increasing anti-inflammatory cytokines. On the other hand, the exercises could not significantly change IL-6 levels. However, due to the limited knowledge in this regard, there is a need for further research.

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# DECLARATIONS

#### funding

This research received no specific grant from any funding agency in the public, commercial or not-for-profit sectors.

### Ethics approvals and consent to participate

Written consent was taken from all subjects prior to participation in the study.

# **CONFLICT OF INTEREST**

The authors declare that there is no conflict of interest regarding publication of this article.

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