

## THE EFFECTS OF ACTIVE AND INACTIVE RECOVERY AFTER EXERCISE HIIT ON HSP72 AND PLASMA CORTISOL RESPONSE

Jafar Goudarzi\*<sup>1</sup> and Dr. Amir Hossein Barati<sup>2</sup>

<sup>1</sup>Master of Exercise Physiology, University of Tehran Shahid Rajaii, Iran.

<sup>2</sup>Sport Medicine Faculty Member at The University of Tehran Shahid Rajaii, Iran.

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### \*Corresponding Author

**Jafar Goudarzi**

Master of Exercise

Physiology, University of  
Tehran Shahid Rajaii, Iran.

### ABSTRACT

The purpose of this research has compared to the effect of active and inactive recovery of after one time High Intensity Interval Training (HIIT) on the level of Heat Shock Protein72 (HSP72) and cortisol of plasma. Statistical population was all 18-20 years old soccer player men in the south of Tehran city. The available population was 50 players of ansar soccer club. The study sample was include of 16 soccer players with an average age of  $18.9 \pm 0.5$  (years), weight  $70.1 \pm 1.7$  (kg), height  $176 \pm 2.2$  (cm), body mass index  $22.4 \pm 1$  (kg/m<sup>2</sup>) and maximal oxygen consumption  $51.3 \pm 2.5$  (milliliters per kilogram of

body weight) that were selected from the above mentioned population randomly. The subjects in one group were participated in tow active and inactive recovery tests. In the first stage of the test, blood samples at stages before the exercise, immediately after and 90 minutes after recovery of inactive recovery from ante brachial vein of Non-dominant hand. In the second one which was done one week after the first one the blood sampling was done as the some as the first step but with the active recovery. Analysis of research findings by Kolmogorov-Smirnov test and repeated measure analysis and Bonferroni post hoc test at  $P \leq 0.05$  significance level was performed using the SPSS 22 software. The results showed that the active recovery than inactive recovery on plasma cortisol response of alpha level ( $P \leq 0.05$ ) have a significant positive effect.

**KEYWORDS:** HSP72, Plasma cortisol, Soccer players, HIIT, Active recovery, Inactive recovery.

## INTRODUCTION

One of the most interested subjects for sports science experts is the quality of performing proficiencies as well as steady readiness for attending competitions.<sup>[1]</sup> HIIT<sup>1</sup> is one of the noteworthy sports activity protocols for physiological scholars, which includes sports activity frequentation in addition to very low intensity active rest reprises.<sup>[2]</sup> Physical fatigue due to body work, especially sports activities, is able to affect many parts and functions of the immunity system.<sup>[3]</sup> Metabolic waste materials aggregation due to body work stress and sports activity, results in cellular destruction and damage in many tissues and consequently may endamage the sportsman activity and causes drop in sportive performance.<sup>[4]</sup> HSP<sup>2</sup> molecules are of those key players in preserving intracellular organelles. The operation of these proteins is effective on a wide range of cellular activities such as controlling cellular signals, Protein fold, Immune responses regulation and Apoptosis.<sup>[5]</sup> Heat Shock Protein has different types of Small Heat Shock Protein, 40, 60, 70, 90 and 110.<sup>[6]</sup> Intensive physical or psychic pressure activates Pituitary–Adrenal and results in increased level of ACTH<sup>3</sup> and Cortisol hormones.<sup>[7]</sup> Central temperature changes while sports activity, affects the amount of Plasma Cortisol and a direct relationship between central temperature increment and Cortisol amount increment has been observed.<sup>[8]</sup> Cortisol is able to instigate the HSP72<sup>4</sup> response.<sup>[9]</sup> Witham *et. al.* (2006) observed that increase in HSP72 response is accompanied by increase in Cortisol response. These findings show that Cortisol is an important intermediate in response to HSP72.<sup>[10]</sup> Increase in the amount of HSP72 in blood, on one hand indicates more damage and therefore higher involvement of immunity system, and on the other hand is an index of preservation of tissues and cells against damages caused by different stresses.<sup>[11]</sup> Intensive daily sports activities may have a cumulative suppression effect on the immunity system.<sup>[12]</sup> The level of heat shock proteins could be brought up as a strong index of Aterosklerosis.<sup>[5]</sup> Fast accumulation of HSP mRNA<sup>5</sup> happens after heat shock and so when physiological conditions return to normal, the amount of HSP mRNA decreases.<sup>[13]</sup> Hence, if after the sports activity, the temperature increment would be able to rapidly return to physiologic temperature, then the steadiness of mRNA may fade and consequently HSP would decreases.<sup>[14]</sup> Within the period of returning to the initial state, different metabolic trends occur in the body which is all as important as the activity period trends and are all acting along with retrieving the lost energy and storing it.<sup>[15]</sup> Esfarjani *et. al.* (1390), expressed that performing 10 minutes of active recovery in comparison with inactive recovery and massage, is more effective on next performance of the runners.<sup>[16]</sup> Will *et. al.* (2008) expressed that inactive method is more effective than active recovery.<sup>[17]</sup> Since few

researches have been performed on the effect of recovery on HSP and also the existing results pertaining active and inactive recovery are contradictory, the scholar has accomplished this research.

1. High Intensity Interval Training
2. Heat Shock Protein
3. Adrenocorticotrophic Hormone
4. Heat Shock Protein 72
5. Mesenge Ribo Nucleic Acid
6. Kolmogorov–Smirnov
7. Nano-gram/milliliter
8. Nano-gram/deciliter
9. micro-gram/deciliter

## METHODOLOGY

This research was semi-experimental and performed in form of a field research in two stages with a group. The statistical population of the research included all 18 to 20 years old football players living in south of Tehran. The available population was Ansar football team consisting of 50 persons and among 29 volunteers for participation in the exam, 16 persons were randomly selected after initial tests and experiments. One week before the first stage of experiment, anthropometrical indexes, fat percentage, VO<sub>2</sub>max and BMI were measured. 48 hours before research and after 10 to 12 hours of fasting, sample bloods were taken from all triable subjects. Angiocath was used in order to prevent stress in subjects and venesection was performed from non-dominant hand. In the first stage after 2 hours from breakfast, 6 cc of blood was taken from subjects and warming-up was accomplished for 20 minutes. Breakfast consisted of 2 palm size pieces of Sangak bread, cheese as much as a match box and a 150 cc glass of tea containing 5 grams of sugar also. Afterwards, HIIT exercise was performed and immediately after stopping the exercise, 6cc of blood was taken from all subjects again, then inactive recovery was implemented by sitting on the ground for 10 minutes which was followed by 6 cc venesection one more time. 90 minutes after inactive recovery 6cc venesection was done once more. In the second stage, which was one week after the first one, all steps of the first stage were repeated, however the difference was replacing inactive recovery by active. Active recovery was consisting of 5 minutes running with 40% of the maximum heartbeat and 5 minutes of tensional warm-up movements mainly in lower parts of the body, all in all in 10 minutes. In the first stage temperature was 38°C and humidity was 3 while in the second stage temperature was 38°C and humidity was 4.

**HIIT protocol**

In this research, half exercise protocol was used. The implementation method was such that the players dribbled the first 10 cones through a helix path and jumped from 30 cm high barriers. Afterwards, next cones were passed through a helix form again while players were controlling the ball, moving backwards and then finally returning toward the starting point. Activity periods were consisting of four periods – each one four minutes – which were separated by 3 minutes of rest from each other. Exercise intensity was equal to 90% to 95% of the maximum heartbeat for each player and the inter-periodical active recovery was containing 65% to 75% of the maximum heartbeat.<sup>[18]</sup>

**The method of determining blood variables**

6 cc of blood was taken from each subject from which, 4 cc was taken in clots tube and 2 cc in EDTA contained tube for Plasma preparation. Clots tubes remained in the environment temperature for 20 minutes for coagulate process completion and then centrifuged for 10 minutes at 2500 rpm in order to separate serum from clot. Serum was separated by sampler afterwards. EDTA contained tubes were centrifuged at 3000 rpm for 10 minutes and then Plasma was removed from them. Separated serum and Plasma were transferred to laboratory within tubes which were positioned in ice-including containers and then related experiments were performed on them. Method of determining serum's HSP72 was ELISA and was performed by HSP72 measurement kit (model Mbs9302902), made in Canada and method of determining Plasma Cortisol also was ELISA and was performed with Sigma Cortisol measurement kit, made in United States.

**Statistical methods of research**

All data were analyzed with the use of SPSS software version 22. K-S<sup>1</sup> test was used for data natural distribution, while Repeated Measures and Bonferroni methods were utilized for data statistical analysis. A meaningful level ( $P \leq 0.05$ ) was considered and all diagrams were drawn in Excel 2010.

**RESULTS**

In this section, with the use of descriptive statistics, different variables are presented in terms of table and diagram and in the form of mean values  $\pm$  Standard deviation for different stages of the research on the studied group.

**Table 1 – Anthropometrical specifications of the subjects (M±SD).**

Amount	Variable
16	Number
176±2.2	Height(cm)
70.1±1.7	Weight(Kg)
22.4±1	BMI (kg/m <sup>2</sup> )
18.9±0.5	Age (year)
51.3±2.5	Vo2max(ml/min.kg)

According to table 2, it was shown in Bonferroni test that:

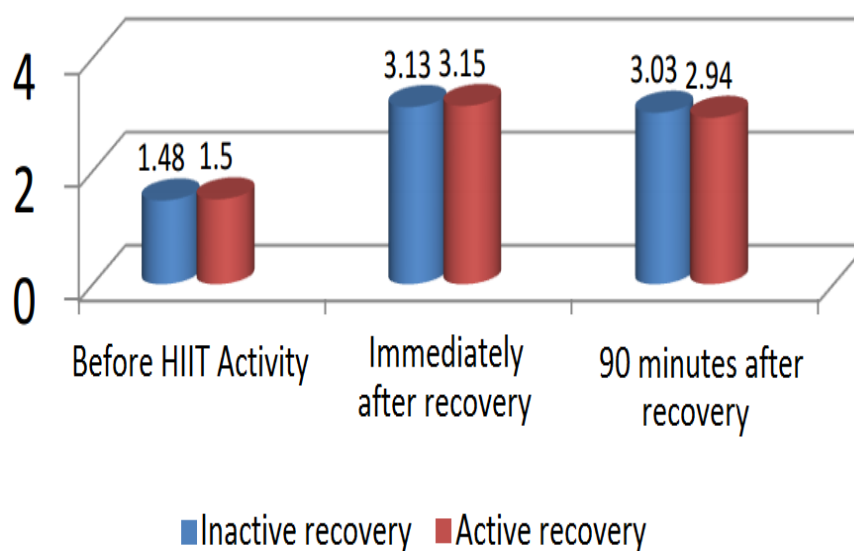
1. Immediately after active recovery, 90 minutes after inactive recovery and 90 minutes after active recovery at Alfa level of 0.001, there is a meaningful difference between pre-HIIT stage in inactive recovery and the immediate stages after inactive recovery.
2. Immediately after active recovery, 90 minutes after inactive recovery and 90 minutes after active recovery at Alfa level of 0.001, there is a meaningful difference between pre-HIIT stage in active recovery and the immediate stages after inactive recovery.
3. 90 minutes after inactive recovery and 90 minutes after active recovery at Alfa level of 0.05, there isn't a meaningful difference between the immediate stage after inactive recovery and immediate stage after active recovery.
4. There is a meaningful difference between the immediate stage after active recovery and 90 minutes after inactive recovery at Alfa level of 0.001, however this difference is not meaningful between the immediate stage after active recovery and 90 minutes after active recovery at Alfa level of 0.05.
5. There is no meaningful difference between 90 minutes after inactive recovery and 90 minutes after active recovery at Alfa level of 0.05.

**Table 2 - Bonferroni test method for HSP72 (ng/ml<sup>1</sup>).**

	Before HIIT inactive recovery	Before HIIT active recovery	Immediately after inactive recovery	Immediately after active recovery	90 minutes after inactive recovery	90 minutes after active recovery
Before HIIT inactive recovery		0.034**	1.66**	1.68**	1.55**	1.54**
Before HIIT active recovery	0.05		1.62**	1.65**	1.52**	1.50**
Immediately after inactive recovery	0.001	0.05		0.021	0.104	0.122
Immediately after active recovery	0.001	0.001	0.05		0.128**	0.146

90 minutes after inactive recovery	0.001	0.001	0.05	0.001		.018
90 minutes after active recovery	0.001	0.001	0.05	0.05	0.05	

\*\* Meaningful at  $P \leq 0.001$ .



**Fig. 1** The amounts of HSP72 in two stages of active and inactive recovery (ng/dl<sup>1</sup>).

According to table 3 it was shown in Bonferroni test that:

1. Immediately after active recovery, 90 minutes after inactive recovery and 90 minutes after active recovery at Alfa level of 0.001, there is a meaningful difference between pre-HIIT stage in inactive recovery and the immediate stages after inactive recovery.
2. Immediately after active recovery, 90 minutes after inactive recovery and 90 minutes after active recovery at Alfa level of 0.001, there is a meaningful difference between pre-HIIT stage in active recovery and the immediate stages after inactive recovery.
3. 90 minutes after inactive recovery and 90 minutes after active recovery at Alfa level of 0.05, there isn't a meaningful difference between the immediate stage after inactive recovery and immediate stage after active recovery.
4. There is a meaningful difference between the immediate stage after active recovery and 90 minutes after inactive recovery at Alfa level of 0.001, however this difference is not meaningful between the immediate stage after active recovery and 90 minutes after active recovery at Alfa level of 0.05.

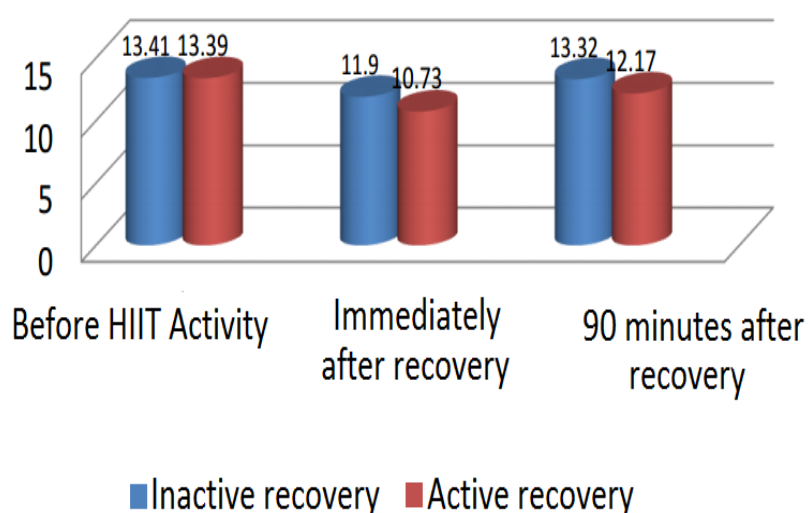
5. There is no meaningful difference between 90 minutes after inactive recovery and 90 minutes after active recovery at Alfa level of 0.05.

**Table 3 - Bonferroni test method for Plasma Cortisol ( $\mu\text{g/dl}^1$ ).**

	Before HIIT inactive recovery	Before HIIT active recovery	Immediately after inactive recovery	Immediately after active recovery	90 minutes after inactive recovery	90 minutes after active recovery
Before HIIT inactive recovery		0.379	1.62*	2.16*	0.113*	1.31
Before HIIT active recovery	0.05		1.24	2.23*	0.491	0.935
Immediately after inactive recovery	0.05	0.05		0.988	1.73	0.312
Immediately after active recovery	0.05	0.05	0.05		2.72**	1.29
90 minutes after inactive recovery	0.05	0.05	0.05	0.001		1.42
90 minutes after active recovery	0.05	0.05	0.05	0.05	0.05	

\* Meaningful for  $P \leq 0.05$ .

\*\* Meaningful for  $P \leq 0.001$ .



**Fig. 2 The amounts of Plasma Cortisol in two stages of active and inactive recovery ( $\mu\text{g/dl}$ ).**



## DISCUSSION AND CONCLUSION

Heat Shock Proteins are involved in consorting molecules which play an important role in the immunity system. It's not strange that because of wide publication and homology between different types, HSP would be the provider of target antigens as the immunity response. Being exposed to antigen HSPs may convert the immunity response to anti-host antigens and instigate autoimmune disease. These types of chaperons are immunologically strong and can cause pathogenicity in the tissue and besides can play role in infecting, autoimmune disease, idiopathic disease, arthritis and Atherosclerosis.<sup>[5]</sup> After HIIT activity there was a meaningful negative effect on HSP72 between active and inactive recovery. This meaningfulness in both stages of the experiment was because of increase in HSP72 in comparison with its level before HIIT activity. This increase could be resulted from glycogen storage reduction, blood PH increment, temperature increment after bodily activity or also the occurring changes in Cortisol hormone.<sup>[19]</sup> The results in Bonferroni test was such that Immediately after active recovery, 90 minutes after inactive recovery and 90 minutes after active recovery at Alfa level of 0.001, there was a meaningful difference between pre-HIIT stage in inactive recovery and the immediate stages after inactive recovery and this meaningfulness showed that HSP72 was increased in the aforesaid stages in comparison with the pre-HIIT stage in inactive recovery. Also, immediately after active recovery, 90 minutes after inactive recovery and 90 minutes after active recovery at Alfa level of 0.001, there was a meaningful difference between pre-HIIT stage in active recovery and the immediate stages after inactive recovery. The results showed that, there is a meaningful difference between the immediate stage after active recovery and 90 minutes after inactive recovery at Alfa level of 0.001 and it shows that HSP72 was higher immediately after active recovery compared with 90 minutes after inactive recovery. Also, HSP72 showed more reduction in 90 minutes after active recovery compared with 90 minutes after inactive recovery, however wasn't statistically meaningful. The results of this hypothesis were along the same lines of the researches performed by Kohandel (2005), Pik *et. al.* (2005) and Nesing *et. al.* (2004).<sup>[20, 21, 22]</sup> They expressed that there is no meaningful difference between active and inactive recovery and their results are not compatible with James *et. al.* (2008), Kashef (1380) and Oshi *et. al.* (2003).<sup>[23,24,25]</sup> James *et. al.* (2008) found in their studies of omitting blood lactate through three active recovery programs, respectively with intensities of 50%, 100% and 150%, lactate threshold is higher than inactive recovery. The reason for this non-alignment could be the measured blood factor or difference in exercises. Kashef (1380) expressed that CO<sub>2</sub> pressure, HB and Bicarbonate ion have meaningful differences in both recovery stages, and it shows that active recovery



(walking and running) has a meaningful positive effect in comparison with inactive recovery (sitting). The reason of non-alignment of his research with the present hypothesis could be the measured blood factors as well as venesection intervals. Oshi *et. al.* (2003), performed a 60 minutes of temperature increasing (42°C) and then had a 4 hours recovery and found out that HSP72 has increased up to twofold. The reason of his non-alignment with this hypothesis could be the temperature at which the experiments have been held, because temperature could be really effective on HSP72 changes. Cortisol is an steroid hormone which is secreted from adrenal cortex and is explanatory of catabolic and stressful states. Long-time exercise causes Plasma Cortisol condensation and Cortisol increment, increases not only protein metabolism but also Gluconeogenesis in liver, gastrointestinal tract and kidney. Meanwhile, as a stress index, Cortisol results in HSP72 increase.<sup>[26]</sup> Stress has an increasing role in Cortisol level increment. High amount of these hormones in long-term could be harmful for arteries and heart.<sup>[27]</sup> Repeated Measures test was such that after HIIT activity, there is a meaningful positive effect on Plasma Cortisol at alfa level of 0.001 and this meaningfulness was because of lower level of Plasma Cortisol immediately after recoveries and also 90 minutes after recoveries, in comparison with its level in pre-HIIT activity and the reason of Cortisol reduction could be exercise intensity and Plasma volume reduction. In Bonferroni test it was such that there was a meaningful difference between pre-HIIT stages in inactive recovery and immediately stages after inactive as well as active recovery at alfa level of 0.05 and it shows that Plasma Cortisol immediately after inactive as well as active recovery has reduced in comparison with pre-HIIT stages in inactive recovery. Again, there is a meaningful difference between pre-HIIT activity in active recovery and immediately after active recovery at alfa level of 0.05 and it shows that the level of Plasma Cortisol has reduced immediately after active recovery in comparison with pre-HIIT activity stage in active recovery. The results also show that there was a meaningful difference between immediately after active recovery and 90 minutes after inactive recovery at alfa level of 0.001 and it means that Plasma Cortisol has been lower immediately after active recovery in comparison with 90 minutes after inactive recovery and it represents more effectiveness of active compared with inactive recovery. The results of this hypothesis is aligned with the research of James *et. al.* (2008), Esfarjani *et. al.* (1390) and Kashef (1380). They expressed that there is a meaningful difference between active and inactive recovery and actually active recovery is more useful<sup>[16,24,25]</sup> which is not aligned with Gaeni *et. al.* (1384) and Tabrizi *et. al.* (1398).<sup>[28,29]</sup> Gaeni *et. al.* (1384) showed that from the aspect of blood lactate changes caused by heavy exhaustive exercise, there is no meaningful difference between the active and inactive

recovery program in minutes 5 and 12. The reason of non-alignment of their research with the existing hypothesis could be the type of measure blood factor, recovery times and also difference in exercise protocol. Tabrizi et. al. (1389) expressed in their research that there is no meaningful difference on immunity factors, between active and inactive recovery. The reason of non-alignment of their research with the existing hypothesis could be the type of exercise protocol as well as measure blood factors. Active recovery can cause blood keep flowing at skin surface and this way central heat would be dissipated. One of the reasons because of which Cortisol increases is heat which could be reduced through active recovery. Based on the results of the existing research, post-HIIT recovery could be effective on Plasma Cortisol as well as HSP72. In all stages, HSP72 and Plasma Cortisol were respectively higher and lower than their pre-HIIT levels. There was no meaningful difference between active and inactive recovery on HSP72 response, however HSP72 showed reduction in 90 minutes after active compared with inactive recovery, which was not statistically meaningful though. There was a meaningful difference between active and inactive recovery for Plasma Cortisol response. So it could be warily said that active recovery is more effective than inactive recovery.

## REFERENCES

1. Wilmore Jack H, Costill, David L (1994). Physiology of sport and exercise.
2. Translation: Ziya moeini (1998). Volume I and II. Publisher mobtakeran.
3. Hemati nafar, mohammad, mohammad reza kordi, servat chopani, sirous chobineh, reza qarakhanloo (2013). The impact of high-intensity interval training (HIIT) on plasma levels of adiponectin, insulin resistance and sensitivity young men disabled. *mjlh Journal of Zanjan University of Medical Sciences.*, Dec; 21(84): 1-12.
4. Ali aqa nejad, hamid, mahdiyeh mola nouri shamsi (2010). Exercise release of cytokines from the skeletal muscle with an emphasis on IL6. *Journal of Endocrinology and Metabolism Iran.*, Dec; 12(2).
5. Antonio, J and Stout, J (2001). Sport supplements, Lippincott Williams and Wilkins, 116-117.
6. Nour bakhsh, Fatemeh, Sasan Rezaei (2012). Heat shock protein. First Edition. Publisher Andishe sara.
7. Kim Kee-bum. Kim Mun-Hee Kim and Lee Dong-Jun (2004). The effect of exercise in cool, control and hot environments on cardioprotective HSP70 induction. *J Physio Anthropol Appl Human. Sci.*, 23(6): 225-230.

8. Williams RH (1994). Textbook of Endocrinology. 3<sup>rd</sup> ed. Philadelphia: W.B Saunders Company.; 245-66. 11.
9. Peter A H, Mark P B, Robert G M, Erica S C, Hackney AC (2010). Relationship between change in core temperature and change in cortisol and TNF- $\alpha$  during exercise. Journal of Thermal Biology.; 35: 348–353.
10. Rohleder N, Wolf JM, Kirschbaum C (2003). Glucocorticoid Sensitivity in humansinter individual differences and Acute Stress effects. The International Journal on the Biology of Stress, 6: 207-2.
11. Whitham M, Walker GJ, Bishop NC (2006). Effect of caffeine supplementation on the extracellular heat shock protein72 response to exercise. J Appl Physiol, 101: 1222-7.
12. Febbraio MA, Steensberg A, Walsh R, Koukoulas I, Van Hall G, Saltin B, et al (2002). Reduced glycogen availability is associated with an elevation in HSP72 in contracting human skeletal muscle. J Physiol, 538(Pt 3): 911-7.
13. Sari-Sarraf V, Reilly T, Doran DA, Atkinson G (2008). Effects of Repeated bouts of soccer-specific intermittent exercise on salivary IgA. Int J Sports Med, 29: 366-71.
14. Mounier N, Arrigo AP (2002). Actin cytoskeleton and small heat shock proteins: how do they interact? Cell Stress Chaperones, 7(2): 167-176.
15. Murlasits Z, Cutlip RG, Geronilla KB, Rao KM, Wonderlin WF, Alway SE (2006). Resistance training increases heat shock protein levels in skeletal muscle of young and old rats. Exp Geronto, 41(4): 398-406.
16. Green, H.J (1997). Mechanisms of muscle fatigue in intense exercise. J Sports Sci, 15(3): 247-56.
17. Esfarjani, Fahimeh, Farzaneh Taqiyan, Ali Mohammad Forouzan (2011). Comparing the effects of three types of cooling program on changes of blood lactate and subsequent implementation of runners. Research in Sport Sciences. Dec; 9: 41-52.
18. Vaile, J., Halson, S., Gill, N., Dawson, B (2008). Effect of Cold Water Immersion on Repeat Cycling Performance and Thermoregulation in the Heat. Journal of Sport Sciences., 26: 431-40.
19. Qare daqi, Nima, Mohammad reza Kordi, Abbass ali Gaeini (2013). The effect of four weeks of intermittent aerobic exercise extreme (Houph) of VO<sub>2</sub>max, T<sub>max</sub> and v VO<sub>2</sub>max Iran's football club. Life Science Sports. Dec; 17: 47-57.
20. Yamada P, Amorim F, Moseley P, Schneider S. (2008). “Heat Shock Protein 72 Response to Exercise in Humans”. Sports Med., 38(9): 715-733.

21. Nething K, L. Wang, Y. Liu, W. Lormes & J. M. Steinacker (2004). Blunted HSP70 response to acute exercise in welltrained skeletal muscle. *Med Sci Sports Exerc*, 36: S318.
22. Peake JM, Suzuki K, Hordern M, Wilson G, Nosaka K, Coombes JS (2005). Plasma cytokine changes in relation to exercise intensity and muscle damage. *European journal of applied physiology*, 95(5-6): 514-21.
23. Kohandel M, Hojat Sh, Kkasbparast M, a comparison of 3 programs of recovery with lactic acid in blood of boy student athletes after a severe and tire some activity in sports. *Jahesh J*, 2005; 1: 37-42.
24. Oishi Y, K. Taniguchi, H. Matsumoto, A. Ishihara, Y. Ohira & R.R. Roy (2003): Differential responses of HSPs to heat stress in slow and fast regions of rat gastrocnemius muscle. *Muscle Nerve*, 28: 587-594.
25. Kashef, Majid (2001). The effects of active and passive recovery on enzymes and blood gases in young athletes. *Olympic Journal*. Dec; 19: 29-38. In Persian.
26. James, D., et al. (2008). Intensity of exercise recovery, blood lactate disappearance and subsequent swimming performance. *J Sports Sci*, (26): 29-34.