

Effect of Preconditioning by Light Load Eccentric Exercise Versus Heat on Markers of Muscle Damage in Collegiate Males

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Background: Delayed onset muscle soreness (DOMS) occurs following unaccustomed or intense bouts of exercise. Previous research has demonstrated that that preconditioning with low load exercise or heat relieves muscle soreness. However, actively warming up the muscles with exercise may be an effective alternative.

Objectives: This study compared preconditioning effect of light load eccentric exercise and heat using microwave diathermy on markers of muscle damage in collegiate males.

Patients and Methods: Thirty six sedentary collegiate males were randomly assigned into two experimental groups: 10% Eccentric exercise group (10% ECC group) and Microwave diathermy group (MWD group). Both the groups performed 30 repetitions of maximal eccentric exercise (Max-ECC) of the elbow flexors. The 10% ECC group performed light load eccentric exercise (10% ECC), five sets, six repetitions using a dumbbell set at 10% of maximal voluntary isometric contraction strength (MVC), 2 days prior to Max-ECC. The MWD group received heat using microwave diathermy (150 watts, 20 minutes) one day prior to Max-ECC. Changes in MVC, range of motion (ROM), upper arm circumference, soreness, were assessed before and 24-72 hours after whereas serum creatine kinase activity and lactate dehydrogenase (LDH) activity up to 48 hours following the Max-ECC were compared between groups by a mixed model ANOVA.

Results: No significant difference ($P > 0.05$) were found between the groups for changes in all variables post Max-ECC, except for LDH activity showing significant interaction effect ($P = 0.04$).

Conclusions: Preconditioning with light load eccentric exercise and heat using microwave diathermy treatment did not differ in their effects on muscle damage markers after Max-ECC. However, with time their effects on LDH activity were found to be different.

Keywords: Light; Exercise; Heat; Muscle

1. Background

Performing an unaccustomed exercise causes symptoms characterized by muscle weakness, stiffness, soreness and swelling (1). It is known that eccentric exercise (ECC) produces these symptoms greater than concentric exercise (2), and the symptoms are often used as markers of muscle damage (1, 3). Other studies of exercise-induced muscle injury have reported increase in plasma level of intracellular muscle proteins (i.e. creatine kinase (CK), myoglobin, lactate dehydrogenase (LDH) and aspartate aminotransferase (AST) (4)) as markers of injury. Eccentric exercise induces ultra-structural muscle damage within sarcomere? Leading to membrane damage and failure of excitation contraction coupling pathway (5, 6). It was suggested that exercise induced muscle damage was attenuated with heat shock proteins (HSPs) (7). Heat preconditioning induces (HSPs), which protect cells by promoting synthesis of and controlling the degradation of muscle proteins (5, 8). Heat induced HSP 72 protects skeletal muscle against impairments of excitation contraction coupling (9). It has been reported that an

elevation of HSP expression peaked between 24 and 48 hours following heat where the core temperature of rats was maintained around 41°C for 30 minutes (10). A study (11) documented 24 hours prior thermal preconditioning (immersion in water at 41.5 ± 0.5°C for 20 minutes) has attenuated exercise induced muscle injury in mice. It was reported previously (12) that heat preconditioning using microwave diathermy (150 Watts, 20 minutes) increased the temperature of vastus lateralis to over 40°C and after 1 day resulted in the release of several HSPs (HSP 90, HSP 72, HSP 27) in human skeletal muscles. One day prior heat preconditioning using microwave diathermy (150 watts, 20 minutes) against exercise induced muscle damage was found to be significantly effective in the faster recovery of maximal voluntary isometric contraction strength (MVC), associated with a smaller decrease in range of motion (ROM) with no effect on the upper arm circumference increase and CK activity post exercise (13, 14). Soreness on extension was also found to be reduced with microwave diathermy treatment (13). At first, it was

found that 2 days prior 10% ECC that does not result in any changes in muscle damage markers, was effective in inducing faster recovery of MVC, ROM and reduced muscle soreness following submaximal eccentric exercise (15), suggesting that the light eccentric exercise preconditioned the muscle against a damaging exercise bout. Other studies (16, 17) extended the previous findings and reported the prophylactic effects of 10 % ECC against Max-ECC. These studies resulted that 10% ECC has induced faster recovery of MVC, ROM, soreness, upper arm circumference and CK activity after Max ECC performed 2 days (16) or 7 days (17) later.

2. Objectives

Previous researches have already mentioned that both 10% ECC and heat using microwave diathermy precondition the muscles for subsequent damaging exercise bout, but no study has focused on the comparison of their effects. Therefore, purpose of the present study was to compare their preconditioning effect and to establish which one of them would be more beneficial in reducing muscle damage.

3. Patients and Methods

3.1. Subjects

Thirty six sedentary collegiate males (age 19-26 years) from Jamia Millia Islamia University, India, participated in the study. Ethical approval was granted by Institutional Human Ethical Committee and the subjects were given written informed consent. The subjects were explained the purpose and methodology and possible risks of the study. None of the subjects were involved in regular resistance training for at least 12 months prior to this investigation and all of them were free from any musculoskeletal disorder of upper extremities. The sample size was estimated using data of changes in maximal isometric strength from a previous study (14) in which a similar heat preconditioning using microwave was performed and the subjects from same population were used and eighteen subjects ($n = 18$) per group were shown to be necessary based on the effect size of (d) 1.27, alpha level of 0.05 and a power ($1-\beta$) of 0.95.

3.2. Procedure and Protocol

A comparative, parallel group, prospective study design, random sampling was done for allocation of subjects into two experimental groups ($n = 18$ each group). Both the experimental groups performed Max-ECC of the elbow flexors with their non-dominant arm. 10% ECC group performed a bout of light load eccentric exercise of the elbow flexors of the non-dominant arm 2 days prior to maximal eccentric exercise (Max-ECC). Microwave diathermy (MWD) group received microwave diathermy treatment 1 day prior to Max-ECC. In both the groups,

MVC, ROM, upper arm circumference, soreness were measured immediately before and 24-72 hours following the Max-ECC. Whereas serum CK and LDH activity were assessed immediately before and 24-48 hours following the Max-ECC. All of the measures were taken from the exercised arm and blood samples were taken from the contralateral arm.

MVC: Subjects were positioned supine on the couch. The non-dominant arm was positioned with the shoulder abducted 30° , the elbow flexed 90° and forearm supinated (18) and the strength was measured with the help of a strain gauge. The subjects were asked to perform two maximal isometric contractions for 3 seconds each with 30 second rest in between the efforts. The average of the two MVC values was used for the determination of the exercise load (15).

Max-ECC: Dumbbell weighing each of individual's MVC strength at 90° elbow flexion was used. For the exercise of the elbow flexors, subjects were seated on a standard arm curl bench with the shoulder flexed at 45° and the forearm in a supinated position. The Max-ECC protocol described below was used in the previous study (19). Subjects were instructed to lower the dumbbell from an elbow flexed (50°) to elbow extended position (170°) in 4-5 seconds keeping the velocity as constant as possible ($30^\circ/\text{second}$). After each eccentric contraction, the examiner removed the load and the arm was returned to the starting position without load. The movement was repeated every 45 seconds for 30 repetitions. **10% ECC:** Dumbbell set at 10% of MVC strength at 90° elbow flexion. The dumbbell weight was adjusted by sticking small lead bars (100 g each) to the dumbbell with tape. The 10% ECC consisted of five sets of six eccentric contractions, in which the subjects were instructed to lower the dumbbell from elbow flexed (90°) to an elbow fully extended position (0°) in 3 seconds. The dumbbell was removed by the therapist at the extended position and the arm was returned to the start position without load (15, 19). The interval between contractions was 10 seconds and a 2 minute rest between the sets was given (16). **Heat preconditioning:** The non-dominant upper arm was exposed to heat preconditioning using microwave diathermy set at 150 watts for 20 minutes (13, 14). Subjects in the sitting position with the treatment arm relaxed and stretched on a bench placed in front of the body (13).

3.3. Dependent Variables

MVC: As explained previously, MVC was measured at 90° elbow flexion using strain gauge. Two measurements for maximal effort were taken and the average of the two values was used for further analysis (15).

ROM: ROM of the elbow joint was determined as the difference between the elbow joint angles of maximal flexion (FANG) and extension (EANG). The FANG was measured at maximal elbow flexion, and the EANG was measured at maximal elbow extension. A Universal plastic goniometer was used for the measurement. To ensure

Table 1. Comparison of Demographic Data and Baseline Dependent Variables Between Groups ^{a,b}

Variable	10% Eccentric Exercise Group	Microwave Diathermy Group	P Value ^c
Age, y	22.00 ± 2.30	22.22 ± 2.10	0.8
Height, m	1.70 ± 0.05	1.70 ± 0.01	0.8
Weight, kg	63.66 ± 8.7	63.55 ± 2.4	1
BMI, kg/m ²	21.93 ± 2.87	21.95 ± 3.20	1
MVC, kg	13.26 ± 1.14	12.90 ± 2.07	0.5
Range of motion, degrees	128.28 ± 3.41	125.83 ± 4.29	0.07
Upper arm circumference, cm	24.18 ± 1.98	24.79 ± 2.96	0.5
Serum creatine kinase, IU/L	53.55 ± 13.49	60.33 ± 14.72	0.2
Serum lactate dehydrogenase, IU/L	275.76 ± 18.45	282.44 ± 20.77	0.3

^a Abbreviations; BMI, body mass building; MVC, maximal voluntary isometric contraction strength.

^b Data are presented as Mean ± SD.

^c Independent t-test.

Table 2. Summary of Mixed Model Analysis of Variance Results of Preconditioning Between Groups

Source	P Value
Maximal voluntary isometric contraction strength, kg	
Group (G)	0.5
Time (T)	< 0.001
Interaction (G×T)	0.08
Range of motion, degrees	
Group (G)	1
Time (T)	< 0.001
Interaction (G×T)	0.06
Upper arm circumference, cm	
Group (G)	0.5
Time (T)	< 0.001
Interaction (G×T)	0.4
Soreness on extension, cm	
Group (G)	0.4
Time (T)	< 0.001
Interaction (G×T)	0.7
Creatine kinase activity, IU/L	
Group (G)	0.9
Time (T)	< 0.001
Interaction (G×T)	0.08
Lactate dehydrogenase activity, IU/L	
Group (G)	0.1
Time (T)	< 0.001
Interaction (G×T)	0.04

that measurements were taken from the same point each time, a semi-permanent ink pen was used to mark a point over the proximal apex of the deltoid, the axis of rotation of the elbow, the styloid process and dorsal tubercle of the radius (13). Upper arm circumference: Upper

arm circumference was measured using a Gulik constant tension tape. Measurements were taken at 3, 5, 7, 9 and 11 cm proximal from elbow crease of cubital fossa and the mean value of the five sites was used for further analysis (15). Blood markers: Approximately 2 mL of venous blood was drawn from an antecubital vein of the dominant arm (non-exercised arm) by a standard venepuncture technique using a disposable needle. The blood was then centrifuged to separate the serum. The serum CK and LDH activity was monitored by a spectrophotometer at 340 nm set wavelength using a CK KIT and LDH KIT respectively. Muscle soreness: Subjects were asked to indicate their pain level on a 10 cm visual analogue scale (VAS) while their elbow flexors were being extended (13).

3.4. Statistical Analysis

Data were assessed by Shapiro-Wilk test for the normality of the distribution scores. The demographic characteristics and the baseline dependent variables prior to Max-ECC were compared between groups by an independent t-test. Changes in the dependent variables over time following Max-ECC were compared between groups by a mixed model ANOVA. Also, Bonferroni test was employed as post hoc analysis to locate the time points having significant difference. Significance level was set at $P < 0.05$. The data were presented as mean (Standard deviation).

4. Results

Shapiro-Wilk test indicated that dependent variables in both groups were normally distributed. No significant difference for the demographic characteristics and baseline dependent variables existed between groups (Table 1). No significant relation between group and time was found in case of MVC, ROM, upper arm circumference, soreness on extension and CK activity for changes from 0 to 72 hours post Max ECC (Table 2). However in case of LDH activity, a significant relation between group and time was found for changes from 0 to 48 hours post Max-ECC (Table 2 and Figure 1).

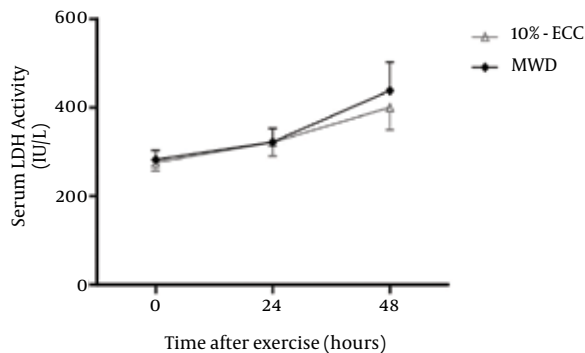


Figure 1. The Serum Lactate Dehydrogenase Activity for Both the Group

5. Discussion

The results of the present study demonstrated that the preconditioning effects of light load eccentric exercise and heat using microwave diathermy treatment were similar on the markers of muscle damage (MVC, ROM, upper arm circumference, soreness, CK activity) except for the changes on LDH activity; following Max-ECC (Table 2). The present study is the first study comparing these 2 experimental groups as well as highlighting their effects on LDH activity. In the present study, MVC strength decreased up to 48 hours in a similar manner in both groups with a peak reduction of 34.1% in 10% ECC group and 29.4% in MWD group (Table 3). Research has demonstrated that calcium release from the sarcoplasmic reticulum is impaired in injured muscles. Eccentric contractions have also been shown to alter the structure of T-tubules. Both of these changes would be responsible for the impairment of excitation-contraction coupling (4). Moderately intensive eccentric exercise results in strength losses of 30-50%, while intense protocols reduce strength by 50-70%, with

recovery times of 7-10 days, extending to several weeks (20). ROM decreased up to 48 hours in a similar manner in both groups, with a peak reduction of 16.34° in 10% ECC group and 12.94° in MWD group (Table 3). It has been suggested that an increase in the number of contracted fiber segments related to either an increase in resting cytosol calcium levels or ultrastructural damage may be responsible for ROM reduction (21). Significant increase in the muscle soreness was observed in both groups with a peak value at 48 hours post exercise (Table 3) which is in line with the other previous studies (13, 15-17). The primary mechanisms underlying the delayed onset muscle soreness (DOMS) sensations include swelling and subsequent increased pressure within the muscle that activates resident mechanoreceptors (nociceptors). The chemical changes responsible for DOMS include increased histamine, bradykinins and prostaglandins that follow the infiltration of inflammatory cells. These substances activate the polymodal nociceptors that are sensitive to chemical signals (20). Peak value of soreness on extension at 48 hours post exercise for 10% ECC is 3 cm, which was also reported previously as 2.5 cm (15) and for MWD it was also 3cm (Table 3), as similarly reported previously (13). After 48 hours, both the groups recovered MVC with similar rates (10% ECC = 11%, MWD = 11%) as well as a similar amount of increase in the ROM (10 % ECC = 4°, MWD = 6°), statistically not significant and recovery of soreness started in both groups. Both the groups showed increment in the upper arm circumference up to 72 hours following Max-ECC in a similar manner. Magnetic resonance imaging has demonstrated intracellular edema in muscle tissue after eccentric exercise (22). The amount of increase in upper arm circumference from pre exercise value to 72 hours post exercise in 10% ECC was 8 mm which is quite near to previous studies (16) reporting approximately 5 mm and in MWD the increase was 7 mm which is quite near to previous studies (14) reporting approximately 10 mm.

Table 3. Post Hoc Analysis of the 10% ECC Group and Microwave Diathermy Group ^{a,b}

Time, h	Group	MVC, kg	ROM, degrees	UAC, cm	SOE, cm	CK, IU/L	LDH, IU/L
0	10% ECC	13.26 ± 1.14	128.28 ± 3.41	24.18 ± 1.98	0	53.55 ± 13.49	275.76 ± 18.45
0	MWD	12.90 ± 2.07	125.83 ± 4.29	24.79 ± 2.96	0	60.33 ± 14.72	282.44 ± 20.77
24	10% ECC	9.49 ± 1.51	118.06 ± 5.51	24.70 ± 2.00	2.17 ± 0.62	100.80 ± 10.17	321.61 ± 31.38
24	MWD	10.21 ± 1.77	118.06 ± 3.72	25.30 ± 2.87	1.94 ± 0.73	96.77 ± 18.23	322.27 ± 30.45
48	10% ECC	8.71 ± 1.52	111.94 ± 5.69	25.01 ± 2.10	3.00 ± 0.49	128.85 ± 14.98	400.06 ± 50.35
48	MWD	9.06 ± 1.97	112.89 ± 3.01	25.52 ± 2.90	3.00 ± 0.59	128.27 ± 20.39	438.44 ± 64.19
72	10% ECC	9.69 ± 1.38	116.39 ± 6.33	24.96 ± 2.02	1.89 ± 0.58	-	-
72	MWD	10.43 ± 1.56	118.00 ± 3.38	25.46 ± 2.84	1.72 ± 0.67	-	-
0 vs. 24 post hoc	10% ECC	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
0 vs. 24 post hoc	MWD	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
24 vs. 48 post hoc	10% ECC	0.3	< 0.001	< 0.001	0.002	< 0.001	< 0.001
24 vs. 48 post hoc	MWD	0.049	< 0.001	0.02	< 0.001	< 0.001	< 0.001
48 vs. 72 post hoc	10% ECC	0.004	0.001	1.0	< 0.001	-	-
48 vs. 72 post hoc	MWD	< 0.001	< 0.001	1.0	< 0.001	-	-

^a Abbreviations: CK, creatine kinase activity; LDH, lactate dehydrogenase activity; MVC, maximal voluntary isometric contraction strength; ROM, range of motion; SOE, Soreness on extension; UAC, upper arm circumference.

^b Data are presented as Mean ± SD.

Both the groups showed significant serum CK and LDH elevations up to 48 hours post exercise, consistent with a previous study (23). When the exercise intensity is within the normal range of metabolism, the muscle tissue is exercised without marked changes in membrane permeability. However, when the exercise intensity exceeds this permissible range, the membrane permeability temporarily changes, resulting in CK release from the active muscle. The boundary of this permissible range is its break point (24). From 0 to 48 hours, for CK activity, 10% ECC showed an increase of 75.3 IU/L and MWD showed an increase of 67.94 IU/L (Table 3), (statistically not significant). For LDH activity, the amount of increase in 10% ECC group and in MWD group was 124.3 IU/L and 156 IU/L (Table 3 and Figure 1), respectively which was found to be statistically significant (Table 2). One limitation of the study was that analysis of LDH activity was done only up to 48 hours so further analysis is recommended. The present study concluded that 2 days prior light load eccentric exercise or 1 day prior heat using microwave diathermy had similar effects on muscle damage markers after Max-ECC. Therefore both of them can be used interchangeably as a preventive measure against muscle damage in clinical settings depending upon the availability of the equipment, therapist's skill or knowledge and client's preference. However with time, their effects on LDH activity were found to be different. Further studies are therefore recommended to establish a more detailed analysis of the effects on changes in LDH activity by comparison up to several days post exercise.

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Authors' Contributions

Sumbul Zaheer: Concept and design, Acquisition of data, Data analysis and interpretation, Manuscript preparation, Approval of the article; Jamal Ali Moiz: Concept and design, Data analysis and interpretation, Manuscript preparation, Critical revision of the manuscript, Approval of the article; Mohammad Yaqoob Shareef: Concept and design, Approval of the article; Ejaz Hussain: Concept and design, Critical revision of the manuscript, Approval of the article.

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References

- Clarkson PM, Nosaka K, Braun B. Muscle function after exercise-induced muscle damage and rapid adaptation. *Med Sci Sports Exerc.* 1992;**24**(5):512-20.
- Lavender AP, Nosaka K. Changes in fluctuation of isometric force following eccentric and concentric exercise of the elbow flexors. *Eur J Appl Physiol.* 2006;**96**(3):235-40.
- Ebbeling CB, Clarkson PM. Exercise-induced muscle damage and adaptation. *Sports Med.* 1989;**7**(4):207-34.
- Linkous DH, Flinn JM, Koh JY, Lanzirotti A, Bertsch PM, Jones BF, et al. Evidence that the ZNT3 protein controls the total amount of elemental zinc in synaptic vesicles. *J Histochem Cytochem.* 2008;**56**(1):3-6.
- Koh TJ. Do small heat shock proteins protect skeletal muscle from injury? *Exerc Sport Sci Rev.* 2002;**30**(3):117-21.
- Warren GL, Ingalls CP, Lowe DA, Armstrong RB. What mechanisms contribute to the strength loss that occurs during and in the recovery from skeletal muscle injury? *J Orthop Sports Phys Ther.* 2002;**32**(2):58-64.
- McHugh MP. Recent advances in the understanding of the repeated bout effect: the protective effect against muscle damage from a single bout of eccentric exercise. *Scand J Med Sci Sports.* 2003;**13**(2):88-97.
- Naito H, Powers SK, Demirel HA, Sugiura T, Dodd SL, Aoki J. Heat stress attenuates skeletal muscle atrophy in hindlimb-unweighted rats. *J Appl Physiol (1985).* 2000;**88**(1):359-63.
- Febbraio MA, Steensberg A, Fischer CP, Keller C, Hiscock N, Pedersen BK. IL-6 activates HSP72 gene expression in human skeletal muscle. *Biochem Biophys Res Commun.* 2002;**296**(5):1264-6.
- Selsby JT, Dodd SL. Heat treatment reduces oxidative stress and protects muscle mass during immobilization. *Am J Physiol Regul Integr Comp Physiol.* 2005;**289**(1):R134-9.
- Nonaka K, Akiyama J, Tatsuta N, Iwata A. Thermal Preconditioning Attenuates Exercise-induced Muscle Injury in Mice. *J Phys Ther Sci.* 2011;**23**(3):409-11.
- Ogura Y, Naito H, Tsurukawa T, Ichinoseki-Sekine N, Saga N, Sugiura T, et al. Microwave hyperthermia treatment increases heat shock proteins in human skeletal muscle. *Br J Sports Med.* 2007;**41**(7):453-5.
- Nosaka K, Muthalib M, Lavender A, Laursen PB. Attenuation of muscle damage by preconditioning with muscle hyperthermia 1-day prior to eccentric exercise. *Eur J Appl Physiol.* 2007;**99**(2):183-92.
- Saga N, Katamoto S, Naito H. Effect of heat preconditioning by microwave hyperthermia on human skeletal muscle after eccentric exercise. *J Sports Sci Med.* 2008;**7**(1):176-83.
- Lavender AP, Nosaka K. A light load eccentric exercise confers protection against a subsequent bout of more demanding eccentric exercise. *J Sci Med Sport.* 2008;**11**(3):291-8.
- Chen HL, Nosaka K, Chen TC. Muscle damage protection by low-intensity eccentric contractions remains for 2 weeks but not 3 weeks. *Eur J Appl Physiol.* 2012;**112**(2):555-65.
- Chen TC, Tseng WC, Huang GL, Chen HL, Tseng KW, Nosaka K. Low-intensity eccentric contractions attenuate muscle damage induced by subsequent maximal eccentric exercise of the knee extensors in the elderly. *Eur J Appl Physiol.* 2013;**113**(4):1005-15.
- Bohannon RW, Lusardi MM. Modified sphygmomanometer versus strain gauge hand-held dynamometer. *Arch Phys Med Rehabil.* 1991;**72**(11):911-4.
- Chen TC, Nosaka K, Sacco P. Intensity of eccentric exercise, shift of optimum angle, and the magnitude of repeated-bout effect. *J Appl Physiol (1985).* 2007;**102**(3):992-9.
- Sayers SP, Hubal MJ. Histological, chemical, and functional manifestations of muscle damage. In: Tiidus PM editor. *Skeletal Muscle Damage and Repair*. Champaign: Human Kinetics; 2008.
- Proske U, Morgan DL. Muscle damage from eccentric exercise: mechanism, mechanical signs, adaptation and clinical applications. *J Physiol.* 2001;**537**(Pt 2):333-45.
- Ploutz-Snyder LL, Nyren S, Cooper TG, Potchen EJ, Meyer RA. Different effects of exercise and edema on T2 relaxation in skeletal muscle. *Magn Reson Med.* 1997;**37**(5):676-82.

23. Naiya A. Biochemical markers an indirect method for evaluating Delayed Onset Muscle Soreness among recreational athletes. *Int J Biol Med Res*. 2012;**3**(2):1624–6.
24. Totsuka M, Nakaji S, Suzuki K, Sugawara K, Sato K. Break point of serum creatine kinase release after endurance exercise. *J Appl Physiol* (1985). 2002;**93**(4):1280–6.