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Short-term effects of the Extract of Melissa Officinalis Supplements on Serum Creatine Kinase levels and extent of perceived Soreness after Aerobic exercise in a Negative Slope

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Abstract

The purpose of this study was to investigate the effects of downhill running short of Melissa officinalis (MO) supplementation on serum creatine kinase and the amount of perceived soreness (pain) in the male athletes. For this purpose, 20 male athletes were randomly divided into two groups of 10 (supplement and placebo). Level of total serum creatine kinase, level of understanding thigh bruising and swelling in the ground state before and after the supplementation was measured. After collecting the data, ANOVA for repeated evaluation of time series and the Bonferroni post hoc test was significant, and t-test was used to evaluate difference between-groups. Meaningful level p>0.05 has been used. The findings of the study were that total serum creatine kinase levels were increased less in the supplement group than placebo. The amount of perceived soreness was lower in the supplement group, although the difference between groups was observed in the swelling of the thigh. But in both groups compared to the ground state of the thigh was swollen. Generally, it is concluded that the herbal supplement can affect on reduce swelling, bruising, elevated serum creatine kinase levels and perception of fatigue.

Keywords: Melissa officinalis, creatine kinase, delays fatigue, perceived soreness, eccentric activity.

Introduction

Running downhill like coming down from the mountain or stairs with eccentric contractions cause more muscle soreness than other muscle contractions[1,2]. The main feature of this type of additional pressure is contraction of muscle fibers, connective tissue and the muscle damage while doing these activities. Although eccentric contractions of metabolism requires less energy than other activities, this kind of damage to small contraction of skeletal muscles, is also larger inflammatory response, oxidative stress than the other contractions[3].

Delayed muscle soreness is an unpleasant state of feeling which associated with pain spasms, stiffness, weakness, muscle[2]. DOMS usually 24 to 72 hours after a bout of activity reached its peak and eventually disappears after 5 to 7 days[4]. Several theories expressed to explain the mechanism of DOMS including lactate accumulation, inflammation, muscle spasm, muscle injury, damage connective tissue and increase muscle temperature[5,6]. A common feature of most of these theories is that the production of free radicals increases during DOMS[4-6].

Exercise, especially aerobic exercise increases the oxygen consumption of cells, so muscle cells can produce more energy. The result is an increase in oxygen, creating reactive molecules that is called free radicals[4,8]. Free radicals can combine with other molecules that are capable of causing oxidative stress and damage in this case is more in extroverted aerobic activity[9] Different strategies have been conducted in order to reduce DOMS, which include: Massage, Cryotherapy, Ultrasound, anti-inflammatory drugs such as aspirin and the use of artificial and natural supplements[10,11]. Bloomer et al in their review showed that Supplementation didn't measure and 2 weeks before aerobic exercise, vegetables and fruits reduce oxidative stress in men and women which act like vitamins C and E supplementation[12]. Besides fruits and vegetables, scientists have been studied antioxidant herbs like saffron, cinnamon and green tea[13,14]. Melissa officinalis is an herb that has anti-oxidation properties. Rostami et al investigate the antioxidant effect of Melissa officinalis, vitamin C, and concluded that have a similar effect with vitamin C and suggested that it be named as natural antioxidants[15]. Melissa officinalis is an aromatic plant of the family Lamiaceae, mainly grows in central and southern Europe, North Africa, the Mediterranean and the northern part of the country. And in previous studies the impact of antioxidant nutrients has been investigated on radiology staff[16], of aluminum workers[17] and patients with liver fat[18] and noticeably that there isn't any study on athletes about the effect of water plant. Therefore, the aim of this study was to investigate the effects of downhill running (on treadmill for 30 minutes with an intensity of 65% peak aerobic power and the negative slope of 5/8 grade) and short-term supplementation of MO (5/1 g twice daily as T-Bag and 14 days) on serum creatine kinase and rate of perceived soreness (pain) on athletic.

Methodology

Subjects: This study was approved by the Research Ethics

Committee of the University Borojerd and quasi-experimental design was used. The subjects consisted of 20 male athletes who had regular exercise. They were not Smokers and didn't have the medical history of cardiac, renal, hepatic, and physical and announced the lack of sensitivity to the blood taking or not stated MO. Subjects then were ready to cooperate voluntarily by filling consent form. Thus, after introducing the whole subject, objectives and methodology of research, healthy subjects were selected by taking measurements of age, weight, body mass index, body fat percentage, exercise history and no history of previous injuries due to health questionnaires. The sample size was based on previous studies, was determined for each group in the level of significance (alpha or Type I error) of five percent and one (beta or Type II error) 2/0 Mdkal using software to version 10.0.2.0. In each group 10 patients randomly were replaced into two groups, supplementary training (MO) and placebo.

Data collection: Before starting the test, the objectives, the details and risks of implementing the activities described for subjects and then they written consent was obtained from them. In this session, participants' height was measured to an accuracy of 1 cm / 0 by meter height built which made in Iran. Subjects' body fat was measured by the thickness of subcutaneous fat layer in thoracic, abdominal, and femoral measurements using calipers Lafayette Building America. , and by replacing the equations for estimating body fat percentage, which was estimated by Jackson Pollock[19,20]. Baseline blood samples for the study of the underlying indices were taken from a vein, ten days before the eccentric aerobic exercise in elbow crutch. A second blood sample was measured immediately after the exercise protocol, as well as measures the perception of fatigue and circumference around thigh (before supplementation). A week before the exercise protocol, the subjects were measured in aerobic and anaerobic power. Also, early circumference around thigh was measured in both groups. Each of the supplement and placebo groups were used respectively, MO (daily 5/1 mg per kg of body weight, MO) and dextrose (day 5/1 mg per kg of body weight CSP) for 14 days.

After 14 days, subjects protocol run again immediately after that, a third blood sample taken from the subjects also perceived fatigue group and the activity of serum CK. Before each blood sampling, participants were asked to complete the 24-hour dietary recall. In addition, they were asked to avoid taking any medication, smoking, antioxidant supplements and anti-inflammatory supplements such as ibuprofen, ginger during research.

Eccentric exercise: Each of the subjects ran on a treadmill for 30 minutes with an intensity of 65% maximal oxygen consumption and 5/8- steep grade (15%). Baseline heart rate of each individual after 10 minutes of rest (sitting) were recorded with a stethoscope polar .Also, the maximum heart rate during the exercise test was recorded Bruce treadmill through the display device.

On the other hand to control the exercise intensity of 65% maximum heart rate Karonen method was used. Prior to conducting protocol, for warm-up, subjects performed a 5-minute stretch and then 3 minutes running on a treadmill with no tilt.

After this stage, the treadmill speed and incline to reach target heart rate (65% heart rate reserve) was grown in two minutes. Each participant approached with an intensity of 65% heart rate reserve and 15 percent negative slope ran on a treadmill for 30 minutes. Heart rate, incline and speed up the treadmill exercise test was controlled by the researcher[2].

Measurement of hematologic parameters and variables: All research procedures were performed in standard conditions with relative humidity 55%, temperature 25°C between 8 am to 10 am, In all subjects, baseline blood samples were taken at the rate of five millimeters. Then the samples were placed to clot formation for 30 min at temperature 22 to 25 degrees. Then serum was separated by centrifuging and kits Pars was measured total creatine kinase enzyme activity using autoanalyzer (Alcyon 300).

Measurement of perceived muscle soreness: Questionnaire for measuring perceived muscle soreness (verbal descriptive - Talag) were used as subjects in the seven-state as follow:

1-No Pain, 2 – mild unknown pain -3- little pain 4- more than minor pain 5 -painful 6-very painful - 7 unbearable pain highlights the scale of zero to 12 and are recorded based on subjects' words[21].

Methods of statistical analysis: After calculating data and index of serum and nonserum levels DOMS, the general characteristics of the subjects and the research data in the form of charts and tables using the software EXCEL2007 cross was studied .Then hypothesis (after approval of Normality data: Komologroph Smirnov test results and homogeneity of variances), inferential statistics, repeated ANOVA followed by Bonferroni test was used to compare differences in time-series.

Also, the effect between groups and within groups determined (two-way analysis) and t –test used if there was any significant between-groups . 0.05 significance level for all tests has been used by SPSS software under Windows version 17. Also, the rate of effectiveness of each of the independent variables were determined by using eta squared.

Results and Discussion

The characteristics of subjects are presented in table-1 and other details are given in table-2 and figure-1,2.

Levels of total serum creatine kinase: Total serum creatine kinase analysis of variance (the measure of group differences) suggests that both short-term supplementation of MO and

running downhill changes affect total serum creatine kinase (table-2) Bonferroni post hoc test results indicate that exercise causes a significant increase in total serum creatine kinase in both groups. Although total serum creatine kinase levels after short-term supplementation of MO and running downhill relative increase is less than placebo.

In any case, the increase is significant in both groups (figure-1) It should be noted that the mean and range of total serum creatine kinase after exercise protocol implementation MO were significantly lower in the group receiving supplements than placebo. So that the share effect (eta squared) in the group receiving supplements of MO was 997/0 and998/0 in placebo (table-2 and figure-1). So, we can say that short-term supplementation of MO significantly increased the relative total serum creatine kinase prevents male athletes after running downhill.

Perceived muscle soreness: Understanding fatigue analysis of variance (the measure of group differences) suggests that both short-term supplementation of MO and running downhill changes affect perceived muscle soreness (table-2) Bonferroni post hoc test indicated the levels of both groups increased roughly equal immediately after running downhill without using Supplementation significantly (figure-2) It should be noted that the average perceived muscle soreness immediately after exercise and supplementation protocol implementation in MO significantly lower in the group receiving placebo, and thus the scope of these changes were greater for the latter group. So that the share effect (eta squared) in the placebo group was 858/0, and the complement of MO was 788/0. According to the results of the independent t-test and range of perceived muscle soreness, it can be say that short-term supplementation MO prevented significantly relative increased to male athletes perceived muscle soreness after downhill running (table-2 and figure-2).

Discussion: This study suggests that running downhill and short-term supplementation of creatine kinase are effective on changing measure total serum MO over the three periods and short-term supplementation of MO significantly prevented increased total serum creatine kinase relative male athletes immediately after downhill running. Total serum creatine kinase is an indirect indicator in muscle injury. Significant increase in total serum CK activity after eccentric muscle is identified[10,11].

Table-1
Mean and standard deviation of anthropometric and physiological

Indexes under study	Group	Mean	SD
Age(year)	MO	15.70	1.25
	placebo	15.90	1.20
Weight(kg)	MO	61.85	9.103
	placebo	58.10	8.949
Height(cm)	MO	176.10	5.405
	placebo	174.10	5.446
Index of body mass	MO	20.589	2.394
	placebo	20.559	2.308
Fat(percent)	MO	11.607	0.247
	placebo	11.711	0.130
Maximum of consumed Oxygen	MO	50.00	4.761
	placebo	49.80	2.936

Simpson et al study subjects ran during 30 min in a gradient of 18 - percent and creatine kinase levels were measured before and after exercise. Results showed a significant increase in total serum creatine kinase activity after 24 hours prior to the activity[22]. According to Schwann enzyme release, the rupture Sarkvlm causes float freely between the muscle fibers of other cell contents. Collagen and protein metabolic products are released into the extracellular space.

Table-2
Serum total creatine kinase level in two groups and level of perceived soreness(mean ± SD)

Variables	Groups	Basic rate	After activity(Before supplementation)	After activity(after supplementation)
creatine kinase (U/I)	MO	91/90±10/51	161/88±12/26†	185/78±13/59*‡†
	placebo	90/75±8/22	164/53±8/2†	204/33±9/43‡†
level of perceived soreness	MO	------	1/87±1/02	1/11±0/65*‡
	placebo	------	1/89±0/81	1/84±0/76

Represents a significant difference between the group ($p<0.05$). †: represents a significant difference compared to the base case ($p<0.05$). ‡: denotes significant difference. Significant difference compared to the active state before supplementation (05/0 P <).

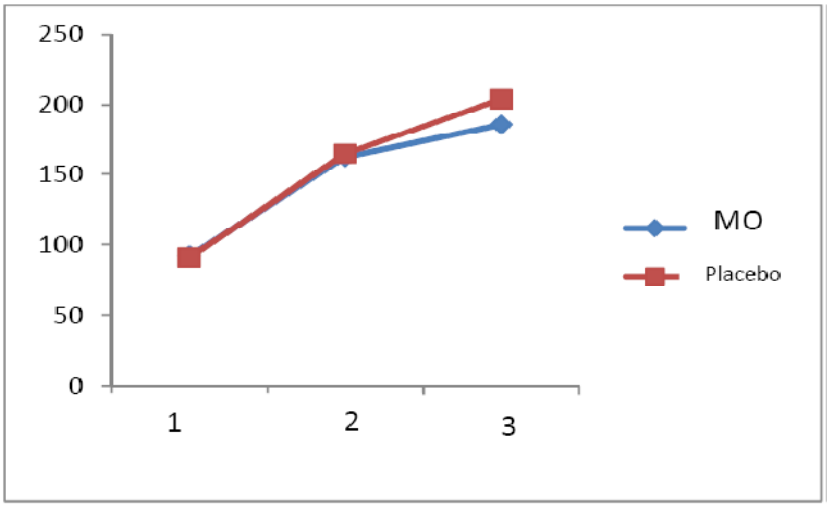

Figure-1
Changes in total serum creatine kinase group and placebo

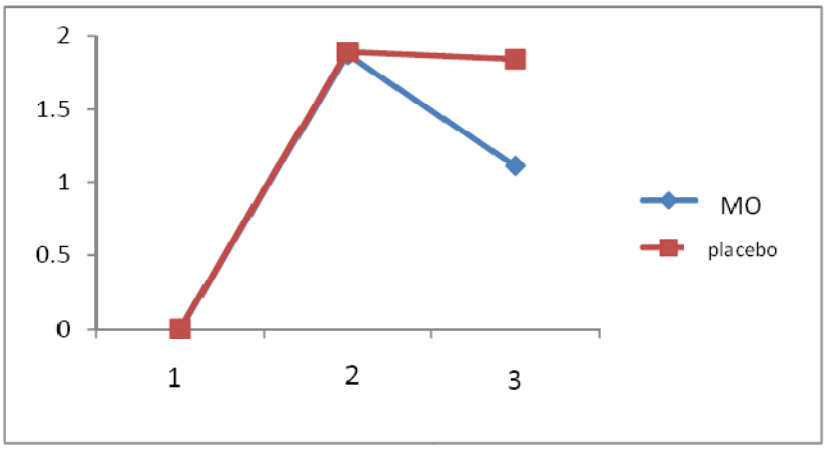

Figure-2
Changes in perceived muscle soreness and placebo group in different stages

Eccentric muscle activity duration and intensity effect on membrane permeability and degradation of blurry[23]. It has established extremely high oxidation potential of hydroxyl and is very active in biological systems occurs mainly from the decomposition of hydrogen peroxide. It also established a foundation to produce other secondary membrane lipid peroxidation collectively participate in the launch[24]. Thus, it can be conclude that MO by removing free radicals and the body's anti-oxidation capacity decreases peroxidation of membrane lipids and loss of membrane phospholipid damage, therefore, prevents leakage and total serum CK enzyme penetration into the extracellular fluid.

So far, many studies on the antioxidant and anti-inflammatory effects of this plant was carried out in patients with laboratory animals, for example Rostami et al examined the antioxidant effect of Melissa officinalis in vitamin C and concluded that a similar effect with vitamin C and K and proposed that it can be named it as natural antioxidants[15]., But, there is no study on reducing muscle soreness from exercise, this was the first study to investigate the effect of MO supplementation on serum creatine kinase that can be checked immediately after eccentric exercise. This study also showed that short-term supplementation of both MO and running down hill affect on muscle soreness perception changes. The rates of both groups significantly increased immediately after downhill running. However, short-term supplementation significantly prevented increased relative MO male athletes perceived muscle soreness after downhill running. Adenosine as a major factor in inflammatory pain induced damage increases. Adenosine receptors are present in most tissues.

It has been shown that the activity of adenosine receptor gene expression increase in human Eccentric nearly six-fold. It has been suggested, many of the actions of adenosine receptor blocking drugs linked to cognitive MO. Adenosine A1 and A2a

receptors in the sensory neurons located in the skeletal muscles, stimulates pain receptors and adenosine receptors MO block reduces pain in muscles[25]. Also, it has been shown that MO combined with indomethacin that inhibits the production of prostaglandins[26]. As mentioned, this is the first study to investigate the effect of Melissa officinalis extract which was performed on muscle soreness, and more research is needed to determine the possible effects.

As noted in the introduction, Melissa officinalis is an aromatic plant of the family Lamiaceae. Melissa was one of the world's most important medicinal plants due to certain aromatic compounds which found in taste, the pharmaceutical industry, health and functional food is abundant[20]. And because of its content (anti-oxidant, anti-inflammatory, etc.), is faster possibly through the prevention and rehabilitation of many of DOMS .As seen in this review MO is a short-term supplementation beneficial effect on cell damage (increased serum creatine kinase), and also, to understand the extent of the bruising and swelling has reduced. Due to the limitations of the study, the researcher was unable to measure creatine kinase levels 24 to 48 hours later. Therefore, more research is needed to determine the precise effect of MO with different doses and periods when it is necessary .To determine whether supplementation of the plant can cause inflammation and oxidative stress, and reduced the soreness caused by the Eccentric exercise. The study identified factors on inflammation, oxidative damage and soreness such as leukocytes, lactate dehydrogenase and necessary malondialdehyde.

The practical findings: This was the first study to investigate the efficacy of short-term supplementation on serum creatine kinase and review delayed soreness MO. And the general conclusion is that this herbal supplement can reduce swelling, bruising, elevated serum creatine kinase levels which may affect the perceived soreness .But before any nutritional prescribed athletes to further explore must be performed on the biochemical approach (measurement of enzymes and other inflammatory agents) and running (prevents loss of strength and range of motion)

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