# Oxidative stress biomarker response to concurrent strength and endurance training

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**Abstract.** The purpose of this study was to investigate the effects of concurrent training on oxidative stress biomarkers in judokas, as well as to compare the effects of such training on performance characteristics in relation to usual training programs. A total number of 14 male judokas were divided into two groups: experimental (E) and control (C). Over 12 weeks, subjects from E group were included into specially designed training composed of concurrent strength and endurance training and perfecting of specific judo techniques. Subjects from C group were included into the same strength training and perfecting of specific judo techniques, but did not have any endurance training. The investigation protocol consisted of Wingate test for the upper extremities, estimation of maximum oxygen uptake, the assessment of body composition, special judo fitness test and determination of selected markers of oxidative stress. The results obtained suggest that usual training program pattern had no effects on oxidative stress levels in C group subjects, while concurrently performed training for strength and endurance induces the increases in anaerobic power and maximal oxygen uptake, but also affects oxidative stress biomarkers. A significant increase in erythrocyte malondialdehyde and plasma catalase can be considered negative effects of this training program.

**Key words:** Oxidative stress — Concurrent training — Judo — Power

**Abbreviations:** ROS, reactive oxygen species; MDA, malondialdehyde; CAT, catalase; $VO_{2peak}$, maximal aerobic uptake; SJFT, special judo fitness test.

## Introduction

Free radicals, chemical species containing one or more unpaired electrons that are capable of independent existence, are produced in all living cells. The majority of free radicals produced *in vivo* are oxidants, which are capable of oxidizing a range of biological molecules, including carbohydrates, amino acids, fatty acids and nucleotides. Generation of reactive oxygen species (ROS) is a normal process in the life of aerobic organisms. Under physiological conditions, these deleterious species are mostly removed by the cellular antioxidant systems, which include antioxidant vitamins, protein and non-protein thiols, and antioxidant enzymes. Muscular exercise increases the production of free radicals and other forms of ROS (Alessio et al. 2000). The risk of oxidative stress with exercise depends on exercise intensity and the participant's state of training (Radovanovic and Rankovic 2004). It is widely assumed that oxidative stress is detrimental to exercise performance, but there is little experimental evidence to support this (Sachdev and Davies 2008).

Judo is characterized by short duration, high-intensity, intermittent exercise followed by a period of constant pulling, pushing, lifting, grappling and gripping movements in preparation for the next explosive effort. As a result, judo is often considered to be an explosive sport which demands great anaerobic strength and capacity, accom-

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panied by a well developed aerobic system. A high level of physical fitness and strength, with good fatigue tolerance, are necessary preconditions for competitive success (Pulkkinen 2001). From the aspect of exercise physiology, competitive success depends significantly on the ability of the judokas to, within their own weight category, achieve higher levels of anaerobic capacity and manifest great muscle strength with quick recovery between successive matches (Borkowski et al. 2001).

In the past decade, concurrent strength and endurance training has received much attention as a form of training. Many of previous investigations have examined similar variables including maximal aerobic uptake ($VO_{2peak}$), anaerobic power, isotonic and isokinetic strength, and body composition during concurrent training (Dolezal and Potteiger 1998; Leveritt et al. 1999). Moreover, they have demonstrated that the impact of concurrent training appears to be more determinable to potential strength gains and not to aerobic power (Rahnama et al. 2007). Additionally, after concurrent strength and endurance training, investigators have noted positive changes in body composition (decreases in fat mass and body fat percentage) (Garcia-Lopez et al. 2007; Rahnama et al. 2007).

The purpose of this study was to investigate the effects of concurrent training on oxidative stress biomarkers in judokas, as well as to compare the effects of such training on performance characteristics in relation to usual training programs during the preparatory period in judokas.

## Materials and Methods

### *Subjects*

A total number of 14 male judokas, with several-year-lasting sport experience, participated in the investigation. They were divided into two groups: experimental (E, $n = 7$) and control (C, $n = 7$). Over 12 weeks, subjects from E group were included into specially designed training program composed of concurrent strength and endurance training and perfecting of specific judo techniques. Subjects from C group were included into the same strength training and perfecting of specific judo techniques, but did not have any endurance training. The whole investigation was carried out during preparatory period, before the start of competition period.

The purpose of the study was explained to the subjects and all gave their informed consent. None of the participants suffered any illness for at least two weeks before the study. They were not taking any medication known to affect hormonal or metabolic responses to exercise. In the present study, however, we did not assess the dietary intake of the subjects.

### *Experimental design*

#### *Strength training*

The 12-week whole-body strength training was carried out, under supervision, three times per week. The program included three to five exercises for the main muscle groups of the body. Mainly, machine exercises were used throughout the training period. All exercises were performed using concentric muscle action, followed by eccentric action. The loads were determinate according to the one-repetition maximum (1RM) method. The intensity ranged from 60 to 85% of the 1RM. The number of sets in each exercise increased and the number of repetitions decreased during the training program.

#### *Endurance training*

Endurance training consisted of running twice a week. All the training sessions were supervised and a heart rate monitor was used. Subjects were engaged in a 30 min training session divided into four loading intervals: 10 min under aerobic threshold, 5 min between aerobic-anaerobic thresholds, 5 min above the anaerobic threshold and again 10 min under aerobic threshold. The focus of training was to improve maximal endurance in a 30-min session.

#### *Laboratory testing*

The anaerobic capacity parameters (peak power and mean power) were determined by the "all-out" 30-s anaerobic Wingate test (Inbar et al. 1996). This test is known to be a highly reliable and valid test of anaerobic power. For the purpose of this test, an arm cycle ergometer (Monark, Sweden) equipped with an electronic measuring device with a display was used. The setting up of the equipment and the subjects' warm-up was carried out according to standard protocols. Data registration was carried out with the help of a specially designed computer program on the basis of the standards devised by the author of the test and the published technical description of a system for registering data by means of a computer (Inesta et al. 1995).

$VO_{2peak}$ was estimated by a method of extrapolation (American College of Sports Medicine 2006) after a standardized submaximal test on the leg cycle ergometer (Kettler, Germany) and arm cycle ergometer (Monark, Sweden) along with telemetric monitoring of heart function (Polar, Finland). The testing was carried out at least 24 h prior the execution of the "all-out" Wingate test. All laboratory testing were carried out in the morning hours; in a room where the temperature was 21–23°C, and the humidity was 55–60%, so that the microclimatic conditions conformed to the standards for functional laboratory testing (Winter et al. 2007).

The percentage of fat mass was measured by bioelectrical impedance analysis (see in References: National Institute of Health Technology 1996) and the device BF300 (Omron, Japan) was used. Data regarding percentage of fatty tissue were read off the display with an accuracy of 0.1%.

*Field test*

The special judo fitness test (SJFT) (Franchini et al. 1998) was performed in a training gym, at least 6 h after Wingate test, in the following sequence: two *Uke* judokas in the same weight category and of similar height were positioned at a distance of 6 m from each other, while the tested subject, *Tori*, stood in the middle between them. When the command *Hajime* was given, the *Tori* was required to run up to one of the *Ukes* and perform an *Ippon-seoi-nage* throw, followed by the same type of throw on the second *Uke*. This procedure was repeated for 15 s (series A), after which the *Matte* command was given, followed by a 10-s break. Series B and series C followed on after procedures was repeated for 15 s (series A), after which the *Matte* command was given, followed by a 10-s break. Series B and series C followed on after a second and third 10-s break. The heart rate was measured after 1 min rest, which followed immediately on the series A, B and C throws. The SJFT index was calculated according to the following equation: $(HReff + HRres) \cdot (A + B + C)^{-1}$ where HReff and HRres are the heart rate following the effort, and 1 min after the test, respectively, and A + B + C is the total number of throws effected in series A, B and C. A lower index indicated better results.

*Blood sample preparation and analysis*

Venous blood samples were taken using ethylenediaminetetraacetic acid as an anticoagulant. Blood samples obtained from brachiocephalic vein at rest at the beginning of investigation, and after 12 weeks were analyzed for the determination of selected markers of oxidative stress – malondialdehyde (MDA), catalase (CAT), carbonyl and sulphydryl group assay for determination of modified proteins and total antioxidant status. Blood markers of oxidative stress were determined by standardized spectrophotometry techniques (Goth 1991; Levine et al. 1994; Koracevic et al. 2001).

*Statistical analysis*

Depending on a statistical marker, measurement scale, type of distribution, and number and size of samples, the following tests were used: Student's *t*-test, Mann-Whitney U test, the Wilcoxon rank sum test, and the Wilcoxon test for paired samples. In order to process the results of the study, the SPSS statistical program for Windows (Release 10.0; Chicago, IL, USA) was used. Statistical significance was set at $p = 0.05$ for all statistical analyses.

## Results

The results of study are presented in Tables 1, 2 and 3.

**Table 1.** Subject physical characteristics along 12-week experimental training period

| | E ($n = 7$) | C ($n = 7$) |
|---|---|---|
| Age (years) | 23 ± 1.5 | 22 ± 1.8 |
| Sport experience (years) | 13 ± 4.2 | 12 ± 3.7 |
| Body height (cm) | 178.4 ± 6.18 | 176.9 ± 7.44 |
| Body weight (kg) | | |
| Pre-training | 75.3 ± 11.2 | 74.1 ± 10.5 |
| Post-training | 72.6 ± 9.9 * | 72.8 ± 9.7 |
| Body fat content (%) | | |
| Pre-training | 9.08 ± 3.86 | 8.82 ± 4.08 |
| Post-training | 7.86 ± 4.04 * | 8.4 ± 3.64 |

Values are means ± SD; * significant difference ($p < 0.05$) from corresponding pre-training value; E, experimental group; C, control group.

**Table 2.** Subjects performance characteristics along 12-week experimental training period

| | E ($n = 7$) | C ($n = 7$) |
|---|---|---|
| Peak power ($W \cdot kg^{-1}$) on arm ergometer | | |
| Pre-training | 9.82 ± 1.66 | 9.44 ± 1.82 |
| Post-training | 11.78 ± 1.8 * | 12.34 ± 1.94 * |
| Mean power ($W \cdot kg^{-1}$) on arm ergometer | | |
| Pre-training | 7.16 ± 0.96 | 7.31 ± 1.08 |
| Post-training | 8.54 ± 1.1 * | 8.98± 1.22 * |
| $VO_{2peak}$ ($ml \cdot kg^{-1} \cdot min^{-1}$) on arm ergometer | | |
| Pre-training | 46.52 ± 6.67 | 48.32 ± 6.22 |
| Post-training | 50.86 ± 5.92 * | 47.66 ± 5.84 |
| $VO_{2peak}$ ($ml \cdot kg^{-1} \cdot min^{-1}$) on leg ergometer | | |
| Pre-training | 51.24 ± 7.38 | 50.28 ± 6.6 |
| Post-training | 54.58 ± 6.96 * | 51.98 ± 7.08 |
| SJFT index | | |
| Pre-training | 15.86 ± 2.32 | 15.41 ± 2.08 |
| Post-training | 13.24 ± 1.75 * | 13.58 ±1.91 * |

Values are means ± SD; * significant difference ($p < 0.05$) from corresponding pre-training value. E, experimental group; C, control group.

**Table 3.** Oxidative stress biomarkers along the study period

| | E ($n$ = 7) | C ($n$ = 7) |
|---|---|---|
| Erythrocyte MDA ($\mu mol \cdot l^{-1}$) | | |
| Pre-training | 14.74 (10.69–16.81) | 13.86 (9.56–16.12) |
| Post-training | 19.18 (15.17–27.21)* | 15.62 (10.06–21.88) |
| Plasma CAT ($IU \cdot l^{-1}$) | | |
| Pre-training | 8.57 (4.08–14.72) | 7.21 (3.65–13.24) |
| Post-training | 25.66 (10.38–44.20)* | 9.44 (5.56–18.28) |
| Sulphydryl group ($\mu mol \cdot l^{-1}$) | | |
| Pre-training | 211.10 (183.80–258.01) | 191.06 (170.12–216.46) |
| Post-training | 246.28 (235.97–353.64) | 205.73 (186.49–217.31) |
| Carbonyl group ($\mu mol \cdot g^{-1}$) | | |
| Pre-training | 0.60 (0.53–1.18) | 0.71 (0.6–0.98) |
| Post-training | 0.89 (0.55–1.11) | 0.82 (0.66–1.06) |
| Total antioxidant status (%) | | |
| Pre-training | 74.65 (47.08–94.53) | 59.91 (37.66–82.29) |
| Post-training | 84.55 (83.80–86.43) | 62.67 (40.02–88.26) |

Values are Me (25–75. percentil); * significant difference ($p < 0.05$) from corresponding pre-training value (Wilcoxon test); E, experimental group; C, control group.

## Discussion

Strenuous exercise is characterized by an increased oxygen consumption and disturbance of intracellular prooxidant-antioxidant homeostasis. Under physiological conditions, these deleterious species are mostly removed by the cellular antioxidant systems, which include antioxidant vitamins, protein and non-protein thiols, and antioxidant enzymes. Some conditions associated with intense exercise, such as local tissue hypoxia or elevated tissue temperatures, could also contribute to reactive oxygen production (Alessio et al. 2000). The evidence that muscle conditioning results in upregulation of antioxidant defenses (Gomez-Cabrera et al. 2008) also suggests a close relationship between reactive oxygen and contractile activity. Therefore, there appears to be a significant role for reactive oxygen in normal muscle physiology. However, a number of conditions may lead to an imbalance of oxidant production and antioxidant defense, and these, presumably, do create conditions of oxidant stress (Bloomer et al. 2005). To defend against ROS, muscle cells contain complex cellular defense mechanisms to reduce the risk of oxidative injury. Two major classes of endogenous protective mechanisms (enzymic and non-enzymic) work together to reduce the harmful effects of oxidants in the cell (Sachdev and Davies 2008). The mechanisms of exercise-induced oxidative stress are not well understood. There is no evidence that this affects sporting performance in the short term, although it may have longer term, not necessarily detrimental, health consequences.

Strength and endurance training, when performed independently, induces different functional and structural adaptations with little overlap between them. Strength training typically results in the increase in muscle mass and muscle power. In contrast, endurance training induces increases in maximal oxygen uptake and metabolic adaptation that lead to an increased exercise capacity (Hennessy and Watson 1994). In many sports, a combination of strength and endurance trainings is performed simultaneously which may lead to a potential interference in strength development making such a combination seemingly incompatible. It is now clear that, at molecular level, different forms of exercise induce antagonistic intracellular signaling mechanism that, in turn, could have a negative impact on muscle's adaptive responses to this particular form of training (McCarthy et al. 1995).

The effects of long-term exercise on steady-state dynamics of enzymatic antioxidant defense system are not clear, and there is an evident lack of studies focused on combat sports. The magnitude of oxidative damage may be related to the power of the pro-oxidant attack (intensity and duration of physical exercise) and capacity of the individual exerciser's antioxidant system (Bloomer et al. 2005; Degoutte et al. 2006).

The results of the conducted investigation showed that in the C group subjects there was a significant increase in the examined values of anaerobic capacity parameters, peak and mean power, after 12 weeks' strength training (Figures 1 and 2). In this group of subjects, $VO_{2peak}$ value did not change significantly after 12 weeks (Figures 3 and 4). A methodical plan aimed to improve the performance of specific judo techniques, performed under the supervision of experienced trainers, resulted in a greater number of throws made during SJFT. With a somewhat lower value of

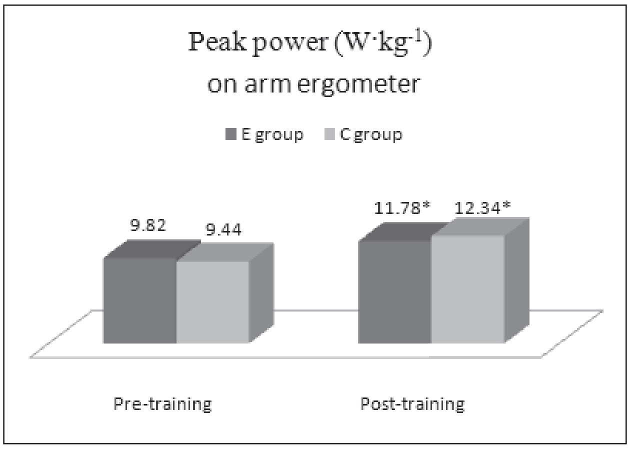


**Figure 1.** The anaerobic capacity parameter peak power in experimental (E group) and control (C group) subjects. The levels of peak power were determined as described in Materials and Methods. Values are means ± SD; * significant difference ($p < 0.05$) from corresponding pre-training value.

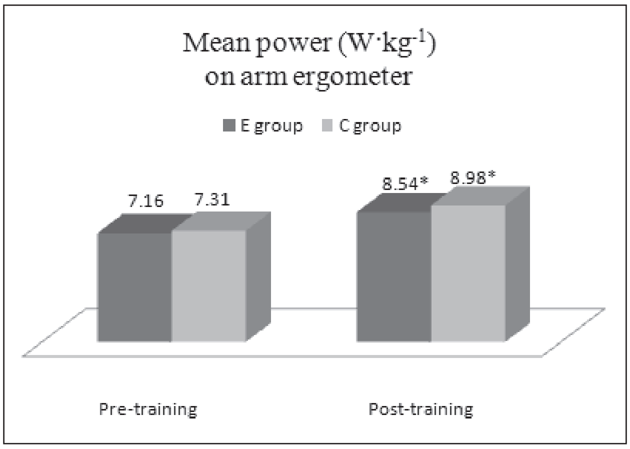


**Figure 2.** The anaerobic capacity parameter mean power in E group and C group. The levels of mean power were determined as described in Materials and Methods. Values are means ± SD; * significant difference ($p < 0.05$) from corresponding pre-training value.

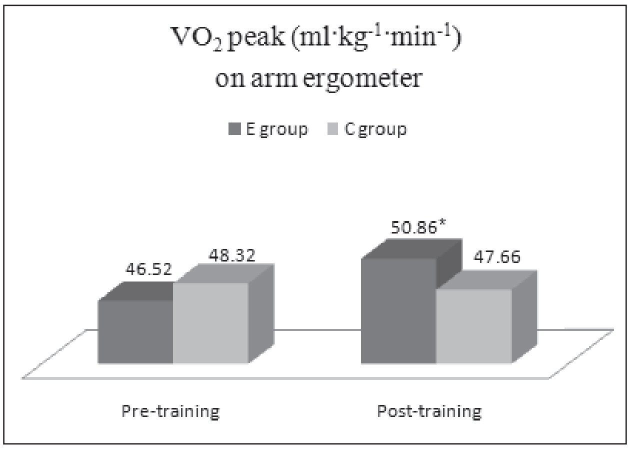


**Figure 3.** Maximal aerobic uptake ($VO_{2peak}$) estimated after a standardized test on the arm cycle ergometer in E group and C group. The levels of $VO_{2peak}$ were determined as described in Materials and Methods. Values are means ± SD; * significant difference ($p < 0.05$) from corresponding pre-training value.

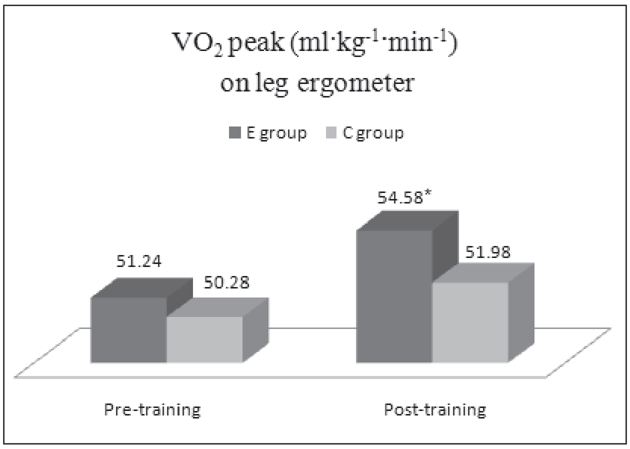


**Figure 4.** Maximal aerobic uptake ($VO_{2peak}$) estimated after a standardized test on the leg cycle ergometer in E group and C group. The levels of $VO_{2peak}$ were determined as described in Materials and Methods. Values are means ± SD; * significant difference ($p < 0.05$) from corresponding pre-training value.

the heart frequency, the overall SJFT index was lower, and the achievement was better. The comparison of values of the mentioned index in the C group subjects showed that the increase was above the threshold for statistical significance (Figure 5). The changes in all examined oxidative stress markers showed no statistical significance (Table 3). The explanation for this finding can be found in the fact that the subjects, although chronologically young, had a long experience with the similar pre-competitive preparation programs. The applied program differed by its higher intensity and duration of strength training as well as longer overall duration of daily trainings from common trainings conducted throughout the year. It can be concluded that this training program pattern had no effects on oxidative stress levels in well-trained young judokas, and that the body's natural antioxidant defenses responded adequately to increases in 12-weeks training program.

In the E group subjects, 12-weeks' concurrent training resulted in a statistically significant increase in the values of anaerobic capacity parameters (Figures 1 and 2). The

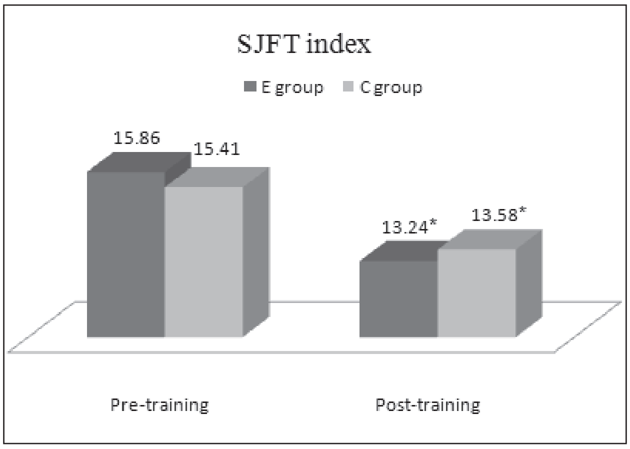


**Figure 5**. The special judo fitness test (SJFT) index in E group and C group. SJFT index was calculated as described in Materials and Methods. Values are means ± SD; * significant difference ($p < 0.05$) from corresponding pre-training value.

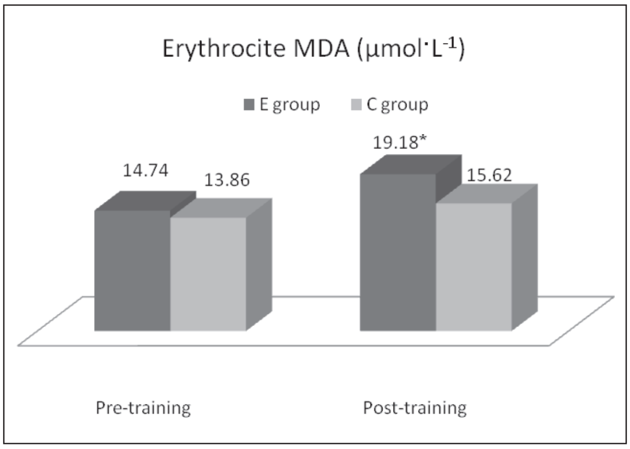


**Figure 6.** Marker of oxidative stress erytrocite malondialdehyde (MDA) in E group and C group. The levels of different oxidative stress biomarkers were determined as described in Materials and Methods. Values are Me (25–75. percentil). * significant difference ($p < 0.05$) from corresponding pre-training value (Wilcoxon test).

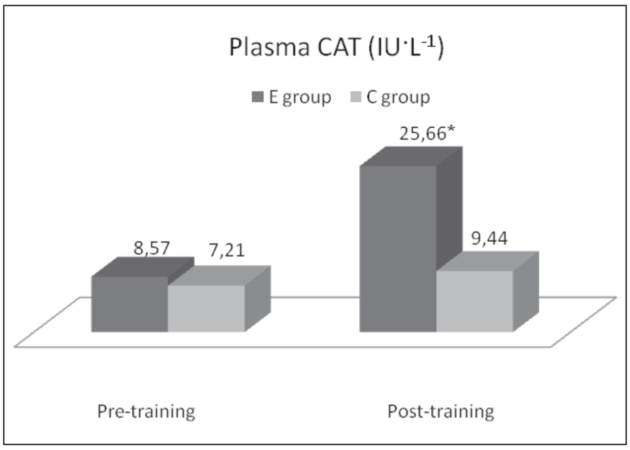


**Figure 7.** Marker of oxidative stress plasma catalase (CAT) in E group and C group. The levels of different oxidative stress biomarkers were determined as described in Materials and Methods. Values are Me (25–75. percentil). * significant difference ($p < 0.05$) from corresponding pre-training value (Wilcoxon test).

comparison of results obtained between groups showed that at the beginning and end of the investigation there were no statistically significant differences in the examined parameters. However, the values of anaerobic capacity parameters were higher in the C group subjects after 12 weeks. The above mentioned points out that the concurrent training resulted in somewhat smaller effects of strength training in the E group subjects. $VO_{2peak}$ values were statistically significantly higher after 12-weeks' training (Figures 3 and 4), pointing out to the effectiveness of concurrent training on the development of endurance. SJFT index values were statistically significantly lower after 12-weeks' preparatory period (Figure 5). Such a result is attributed to the improvement in performances of specific judo techniques, the same as in the C group subjects, as well as to a significantly lower heart frequency during the test which is considered to be another effect of endurance training. After both 12 weeks' training programs we noticed statistically significant changes in body composition of subjects involved in study, decreased of body weight and body fat content (Table 1). A smaller percentage of fatty tissue in elite judokas is thought to enable better metabolic adaptation to different technical-tactical demands during the match (Kim et al. 1996). The comparison of the examined markers of oxidative stress showed that the values of erythrocyte MDA (Figure 6) and plasma CAT (Figure 7) were statistically significantly increased after 12-weeks' training. We consider the results obtained, especially if compared to the results in the C group, a consequence of simultaneously applied strength and endurance training.

The findings of similar investigation (Bloomer et al. 2005; Rahnama et al. 2007; Garcia-Lopez et al. 2007) of the application of concurrent training in other sports may indirectly confirm our conclusions. Other investigations that studied simultaneous training for the development of endurance and muscle power in a long-time period (Hennessy and Watson 1994; McCarthy et al. 1995) indicated the possibility of a decrease in physical abilities in athletes with a several-year-lasting training experience. The abilities that require demonstration of power, i.e. large muscle power and speed, are the most susceptible to the "incompatibility" with the above-mentioned programs for large-extent and high-intensity trainings to which elite athletes are subjected to. The successful combination of training depends on many factors such as the athlete's genetic potential, length of training experience, current physical preparation form, intensity and extent of training, optimal periodization, nutrition and supplementation etc.

The results obtained suggest that concurrently performed training for strength and endurance induces the increase in anaerobic power and maximal oxygen uptake, but also affects oxidative stress biomarkers. A significant increase in erythrocyte MDA and plasma CAT can be considered negative effects of this training program. Since certain biomarkers of oxidative stress are increased after functionally effective specially designed judo training, future research may investigate the methods of reducing macromolecule oxidation, possibly through the use of antioxidant supplementation.

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