# Skeletal muscle contraction time is an important factor in the muscle damage response in kickboxing athletes

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# Abstract

Background and Study Aim:	Combat sports athletes strive to improve their muscle performance, however, they also habitually attempt to reduce their body-mass before the competition voluntarily. The cognitive purpose of this study was the knowledge about combined effects of voluntary body-mass loss and the influence of exercise induced muscle damage (EIMD) markers on the mechanical muscle properties in real-life settings.			
Materials and Methods:	Ten elite kickboxing athletes (all males, age 22.1 $\pm$ 4.1 years) were tested at the beginning of the tapering riod (t-1) and two days before the competition (t-2). Muscle mechanical properties (contraction time <i>Tc</i> ; maximum amplitude of radial displacement <i>Dm</i> ) of the biceps femoris (BF), vastus lateralis (VL) and vastus r dialis (VM) of each athlete's dominant leg were assessed using tensiomyography, while blood and urine sa ples were collected to address biochemical response and hydration status.			
Results:	Body-mass decreased by $-1.3\%$ , plasma volume decreased $-0.9 \pm 1.5\%$ , while EIMD markers decreased by $-74.4\%$ and $-29.4\%$ (p<0.05) for creatine kinase (CK) and lactate dehydrogenases (LDH), respectively. <i>Tc</i> of the VL and BF decreased by $-22.2\%$ and $-9.9\%$ (p<0.001), respectively, while the <i>Dm</i> decreased only in BF by $-20.7\%$ (p<0.001). This study also found a moderate correlation between the average <i>Tc</i> of all three muscles and the CK activity (r <sub>s</sub> = $-0.70$ ; p=0.03).			
Conclusions:	Apparently, after a tapering period that was paralleled by gradual body-mass loss, kick boxers decreased their EIMD markers and improved their contractile muscle performance. According to present correlation findings, Tensiomyography assessment could be used as a surrogate method to denote local muscle fatigue. However, these data also suggest that the CK activity decreases were lower in athletes with lower averaged <i>Tc</i> , an indirect measure of myosin heavy chain type-I proportion.			
Key words:	blood analysis • combat sports athletes • contractile characteristics • hydration • muscle function • training load			
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#### Tensiomyography (TMG) -

a surrogate diagnostic tool to screen for local muscle fatigue in combat athletes, especially during the intense preparation period. Notably, these results underline the importance of systematic monitoring of mechanical muscle properties prior to competition.

# Exercise-induced muscle

damage (EIMD) – usually associated with muscle fatigue, increased plasma levels of muscle proteins, muscle fibre damage, inflammation and impairments in muscle functioning.

Biceps – noun any muscle formed of two parts joined to form one tendon, especially the muscles in the front of the upper arm biceps brachii and the back of the thigh biceps femoris. Triceps (NOTE: The plural is biceps.)

**Biceps brachii** – *noun* the muscle at the front of the upper arm that bulges when contracted [dic].

**Creatine** – *noun* a compound of nitrogen found in the muscles, produced by protein metabolism and excreted as creatinine creatine kinase [37].

Creatine kinase – noun an enzyme that breaks down phosphocreatine into creatine and phosphoric acid, releasing energy creatine monohydrate creatine monohydrate [37].

A neuromuscular *adjective* is referring to both nerves and muscles [37].

**Vastus lateralis** – *noun* one of the four muscles that form the quadriceps vastus medialis [37].

**Vastus medialis** – *noun* one of the four muscles that form the quadriceps [37].

**Muscle function** – noun the smooth expansion or contraction of muscles in the body to create movement [37].

**Combat sport** – *noun* a sport in which one person fights another, e.g. wrestling, boxing and the martial arts [37].

#### Combat sports - are

competitive contact sports with one-on-one combat. Determining the winner depends on the particular contest's rules. In many combat sports, a contestant wins by scoring more points

# INTRODUCTION

It is generally accepted that combat sports athletes strive to improve their muscle performance, however, they also habitually attempt to reduce their body-mass before the competition voluntarily. The integration of such deleterious, physically stressful factors may promote the exercise-induced muscle damage (EIMD). The EIMD is usually associated with muscle fatigue, increased plasma levels of muscle proteins, muscle fibre damage, inflammation and impairments in muscle functioning [1]. Currently, invasive blood sampling is the most frequently reported diagnostic tool used to monitor the EIMD and denote muscle fatigue in athletes [2]. In this context, Magal et al. [3] showed that muscle soreness after eccentrically induced EIMD response was positively related to fibre type distribution (r = 0.51, p=0.04), whereas there was no correlation between fibre type distribution and creatine kinase (CK) activity. This may be due to a greater recruitment of type II muscle fibres in the highintensity eccentric exercise that is commonly used in controlled EIMD studies. Furthermore, type II fibres have been found to be more susceptible to disruption compared to type I, indicating that athletes with a greater proportion of type II muscle fibres might also facilitate higher EIMD responses in other models for inducing the EIMD response [3-5].

Nowadays, tensiomyography (TMG) offers reliable [6, 7], as well as valid [8] insight into intrinsic muscle belly contractile capacity through non-invasive and selective neuromuscular functional assessments of different contractile properties, including muscle tone amplitude [9] and fibre-type distribution [8]. In other words, TMG detects the mechanical response (lateral thickening in millimetres) of skeletal muscle belly to single twitch stimulation. Mechanical response summates muscle belly bulk movement (shift of the muscle mass) and muscle fibre lateral oscillations and thickening. From TMG mechanical response several contractile are estimated from where it has been demonstrated that contraction time (Tc) alone, or in linear combination with delay time and half-relaxation time, estimates myosin heavy chain type-I (MHC-I) proportion in vastus lateralis (VL), with standard error of estimate of 6.08% [8].

Under well-controlled conditions, Hunter and authors [10] observed an association between EIMD markers and skeletal muscle contractile

properties of the biceps brachii muscle, extracted by TMG and found a decrease in TMG amplitude and increase in contraction time during EIMD. Prolongation of TMG assessed contraction time in fatigued biceps femoris (BF) muscle was confirmed by García-Manso and authors [11], who observed increased TMG amplitude from 65.1±22.1 to 77.4±28.5 ms after an Ironman competition. Conversely, Giovanelli et al. [12] found no change in TMG amplitude of the VL muscle following an uphill Etna marathon. Notably, the information regarding the association of EIMD and contractile capacity of skeletal muscle in conjunction with voluntary body-mass loss via acute dehydration in combat sports is limited. Although TMG has been used in a very few fatigue studies, the exact mechanisms explaining TMG indicators alterations remain to be investigated. Notably, it has been recently indicated that whole-body fluid deficit(s) facilitates declines in muscle contractile capacity, extracted by TMG in elite wrestlers of Spain [13]. Moreover, yet, the study above may have overemphasised the role of the whole-body fluid deficit detected via bioelectric impedance readings in the context of measuring actual whole-body fluid fluctuations [14], especially as their conclusions were based on a single time-point measurement before a competition. Indeed, the cross-section study designs could mask/or exacerbate muscle function decrements related to actual body fluid deficit(s).

To best of our knowledge, no study has assessed the adjustment of skeletal muscle contractile capacity to vigorous pre-competition training protocols in striking combat athletes. Therefore, the primary aim of the present study is to reexamine the influence of voluntary body-mass reduction on the contractile capacity of lower limb muscles. Also, to examine the applicability of non-invasive TMG analysis as a surrogate method to screen local/peripheral muscle fatigue, this project investigated the association between EIMD serum markers and contractile capacity of lower limb muscles in highly trained kickboxing athletes following real-life settings.

The cognitive purpose of this study was the knowledge about combined effects of voluntary body-mass loss and the influence of exercise induced muscle damage (EIMD) markers on the mechanical muscle properties in real-life settings.

than the opponent or by disabling opponent [38]

**Training load –** "A simple mathematical model of

training load can be defined

as the product of qualitative and quantitative factor. This

whenever the quantitative factor is called 'workload

volume' or 'training volume'

reasoning may become unclear

# MATERIAL AND METHODS

# Participants

Ten elite kickboxing athletes (all males, age 22.1 ±4.1 years; 1.86 ±0.09 m in body height, 83.8 ±16.1 kg in body mass and 13.5 ±4.2% estimated the percent of body-fat), current national champions of Croatia from different weight divisions volunteered to participate in this study. On average, they had  $6.5 \pm 2.8$  years of kick-box training experience, with  $13.5 \pm 2.6$  hours of training per week. During the previous competitive season, no participant had displayed any history of renal disease or musculoskeletal injuries according to their team medical staff. The authors' Institutional Research Ethical Board (University of Split Research Ethics No. 2181-205-02-05-15-008), by the Helsinki Convention, approved this study before any data collection. Written informed consent was obtained from each athlete before any data collection.

# **Experimental design**

Before any data collection, simple instructions were given to athletes and coaches regarding weight-management practice. To remove possible confusing factors that could obscure the associations between hypohydration and skeletal muscle contractile capacities, athletes were asked to avoid hyperthermia exposure, as suggested by Mora-Rodríguez et al. [15]. The present study design did not interfere with the sequence of exercises, training load or intensity throughout the investigation, which included two training sessions lasting ~90 minutes each day. According to the head coach, sport-specific training activities were structured in accordance with previous studies describing a traditional kickboxing training [16]. More precisely, the athletes' distribution of workload throughout the investigation was dominantly focused on kickboxing specific drills that were manipulated to generate peak performance at the official competition and to promote short-term goals, bearing in mind the acknowledgements summarised by Hoffman and authors [17]. Next, to eliminate possible confounding factors of weight-loss habits and supplement consumption, a validated questionnaire on weight management and its associated protocols was given to all kickboxers [18]. Based on their previous weight management history (e.g., time-span from previous weight-loss experience, supplement consumption, dietary restriction patterns) ten kickboxers were selected for inclusion in the study, whilst two were excluded for not meeting pretesting guidelines, as they reported previous supplement consumption. Athletes were not provided with any information regarding the time-frame to reach their desired body-mass, but they were instructed to withdraw from caffeine ingestion and to maintain their normal diet at least one week before the first data collection.

According to self-reported diet logs, to reduce body mass, kick-boxers used both passive (i) (reduction of carbohydrates/fluid intake) and active methods (ii) (additional training) to accomplish pre-competition body mass following protocols established elsewhere [19]. Athletes reported to the laboratory between 07:30-08:30 h, at least 24 h after their last training, and they were tested on two different occasions by the same researchers: two weeks (t-1) and two days before the major international competition (t-2). The temperature and relative humidity within the testing facility ranged from 14-17°C and ~40% for all trials, respectively.

# Anthropometric measurements

Body mass was determined using the Tanita BC 520 scale (Tokyo, Japan), with the athletes barefooted and wearing only dry underwear. Percent body-fat was estimated via the sum of three skinfolds (subscapular, abdominal, triceps) using Harpenden calliper (Crymych, UK) described in the literature by Lohman [20], according to recommendations for combat athletes [19].

#### Urine sampling

Athletes provided a first-morning urine specimen in agreement with the American College of Sports Medicine's (ACSM) hydration testing guidelines [21] to determine urine specific gravity (Usg) values (AtagoPal-10s refractometer, Tokyo, Japan), which provides accurate readings to the 0.001 unit. The refractometer was calibrated with distilled water before use. A glass pipette was used to apply the urine sample to the instrument, which provided digital results, whilst the urine sampling was performed by the same, qualified staff.

# Hematological and biochemical measurements

Blood sampling was performed by a qualified nurse. Blood samples (210 mL) were drawn via venipuncture from an antecubital vein of the athlete's left arm and analysed using an automated haematology analyser (ADVIA 120, Siemens, HealthCare), including haemoglobin, hematocrit, erythrocytes, mean corpuscular volume, mean

units have been adopted as measures, i.e. the number of repetitions, kilometres, tons, kilocalories, etc. as well as various units of time (seconds, minutes, hours) (...) As in the real world, nothing happens beyond the time, the basic procedure of improvement of workload measurement should logically start with separation of the time factor from the set of phenomena so far classified together as 'workload volume' (...) Due to the fact that the heart rate (HR) is commonly accepted as the universal measure of workload intensity. the product of effort duration and HR seems to be the general indicator of training load defined as the amount of workload. It is useful in analyses with a high level of generality. (...)In current research and training practice, the product of effort duration and HR was referred to as conventional units', or further calculations have been made to convert it into points." [39. p. 238].

corpuscular haemoglobin, mean corpuscular haemoglobin concentration. Additional laboratory processing of the blood samples was handled by the same, experienced laboratory staff. The reliability of estimated haematology concentration values was CV<1%. Blood markers of muscle damage (CK and LDH), and electrolytes (sodium, potassium, and calcium) were determined photometrically (ADVIA 2400; Siemens Healthcare). The results showed satisfactory reliability for EIMD markers [CV<4.9%, for lactate dehydrogenases (LDH) and CK, respectively]. Change in plasma volume ( $\Delta$  PV) was calculated according to the suggestion of Dill and Costill [22], while hematocrit values were multiplied by 0.96 and then 0.91 to correct for trapped plasma and the venous-to-whole blood hematocrit excess.

#### **Tensiomyography measurements**

Radial muscle-belly displacement of the knee extensor muscles (VL and vastus medialis VM) and the knee flexor and hip extensor muscle (BF) of the dominant leg was assessed with TMG. All measurements were performed under isometric conditions in relaxed pre-defined positions: for VL and VM, in a supine position with the knee angle set at 30 deg. flexion (where 0 deg. represents the extended joint); for BF, in a prone position with the knee angle set at 5 deg. flexion. Foam pads were used to support the joints. To assure high signal-to-noise ratio and high reliability, TMG sensor was applied 0.2 N/cm<sup>2</sup> pre-tension on the muscle belly before the measurement was performed [23]. In response to an electrically induced twitch, the oscillations of the muscle belly were recorded on the surface of the skin using a sensitive digital displacement sensor (TMG-BMC, Ljubljana, Slovenia), at the 1kH sampling frequency. A single maximal monophasic 1-millisecond electrical impulse in duration was used to induce maximal muscle belly radial displacement. Carbon rubber electrodes (5 x 10 cm, Medicompex, Switzerland) were placed proximally (anode) and distally (cathode) over the muscle belly, three centimetres to the sensor tip. When necessary, the measuring point and electrode positions were adjusted to obtain maximal radial displacement (Dm) of the muscle belly. The current output was adjustable in the range of 0-110 mA on 0 to 1000  $\Omega$ . The maximal stimulation amplitude was achieved gradually by increasing the amplitude of electrical impulse from threshold to the lowest amplitude needed to achieve highest Dm in each muscle after no further increase was observed in Dm.

Lastly, two maximal twitch responses were saved for further analysis. This methodology was previously used in similar papers [24, 25] and other reliability studies on TMG measurement resolution [6, 7]. To minimise the effects of fatigue and potentiation, rest periods of minimum 10 s were given between each stimulation. From each TMG response the peak Dm was calculated as the peak value of the TMG parameters; delay time (Td) as the length of time between the electrical impulse and when the response reached 10% of Dm; Tc as the time response increased from 10% Dm to 90% Dm; half relaxation time (Tr) as the time response fell from 90% Dm to 50% Dm (as discussed by Šimunič et al. [9]). Variables selected from the TMG response were basal due to its correlation to muscle composition [8] and the maximum amplitude of radial displacement [9]. The results showed satisfactory reliability for TMG assessment, which was CV<4.2%. This amount was consistent with previous reports on of reliability of the TMG assessments [6, 7].

#### Statistics

The data were analysed data using descriptive statistics for the distribution of variables (means, ± standard deviations). All observed variables, aside from LDH and CK, passed the Shapiro-Wilk's normality test. Student's paired t-tests were applied to establish differences between trails. For the non-normally distributed data, the non-parametric Wilcoxon's test was applied. The coefficient of variation, which indicates within subject variation (CV%) was calculated as described in the literature by Hopkins [26]. The homogeneity of variances was tested with the Levine's test. Cohen's d effect sizes (ES) for identified significant differences were determined, and Spearman's correlation coefficients were calculated between the percentage of change in CK activity and the TMG contraction time, which were then averaged for VL, VM and BF muscles. Percentage of change from t-1 to t-2 in all dependent variables was calculated as illustrated in the Equation 1. Reliability of the weight-management questioner was evaluated with Spearman's R for ordinal variables, whilst the percentage of equally responded queries was calculated for nominal variables. Statistical significance was accepted at p<0.05.

Equation 1:

$$\% \Delta = \frac{(\text{Final measurement (t2)} - \text{Basline measurement (t1)})}{\text{Baseline measurement(t1)}} x \ 100$$

Table 1. Basic anthropometric characteristics and urine specific gravity readings (data are presented as mea
and $\pm$ ; % $\Delta$ percentage of change from baseline values; ES (d) effect size; denotes *different from t-1 p<0.001).

Variable	Body height (m)	Body mass (kg)	Body fat (%)	Usg (g∙mL <sup>−1</sup> )
t-1	1.86 ±0.9	83.8±16.1	13.5 ±4.2	$1.020\pm\!0.005$
t-2	-	82.7 ±16.0*	13.2 ±4.3	$1.028 \pm 0.003$
%Δ	-	-1.3	-0.3	0.8
ES (d)	-	0.1	0.1	1.6

Table 2. Red blood cell count parameters, plasma concentrations of electrolytes and EMID markers

Variable	t-1	t-2	%Δ	ES (d)
Erythrocytes (pL)	4.87 ±0.3	4.86 ±0.2	-0.2	
MCV(fL)	89.1 ±3.8	88.9 ±3.7	-0.2	0.1
MCH (pg)	29.8 ±1.04	30.1 ±1.3	1.1	0.3
MCHC (g/dL)	32.9 ±6.5	32.5 ±5.0	0.5	0.1
Hb (g/dL)	14.5 ±0.5	14.6 ±0.5	0.6	0.1
Hct (%)	43.1 ±1.2	43.2 ±1.8	0.1	0.1
Δ PV (%)			-0.9 ±1.5	
LDH(U/L <sup>-1</sup> )	262 ±7	184 ±19.4 <sup>×</sup>	-29.4	1.4
CK(U/L <sup>-1</sup> )	600 ±7	154 ±71.8*	-74.4	0.9
Sodium (mmol/L)	140 ±1.7	141 ±1.5	0.3	0.3
Potassium (mmol/L)	4.3 ±0.4	4.5 ±0.5	5.9	0.7
Calcium (mmol/L)	2.3 ±0.1	2.4 ±0.1	1.8	0.6

Notes: Data are presented as mean and  $\pm$ ;  $\% \Delta$  percentage of change from baseline value; ES (d) effect size; Erythrocytes concentration; **MCV** mean corpuscular volume; **MCH** mean corpuscular hemoglobin; **MCHC** mean corpuscular hemoglobin concentration; **Hb** hemoglobin concentration; **Hct**% hematocrit; **%PV** plasma volume change; **LDH** lactate dehydrogenases concentration; **CK** creatine kinase concentration; \*different from t-1 (p<0.001); ¥ different from t-1 (p<0.002)

# RESULTS

### Study reliability issues

Intra-assay reliability results for the Usg demonstrated a CV<1%. Reliability analyses of the weight-cutting questionnaire confirmed the appropriate reliability of the testing. Spearman's R was ( $r_s = 0.90$ , p<0.05; habitual weight-loss patterns), whilst percentage of equally responded queries ranged from 95% (competitive ranking) to 100% (weight-class category). The consistency of the test-retest answers was 90 to 100%, evidencing appropriate reliability.

# TMG and blood analysis data

In Table 1, body-mass decreased by -1.3% (p<0.001, d=0.1), and Usg readings increased by 0.8±0.5% (p=0.001, d=1.6) over the course of the study. In Table 2, PV decreased by  $-0.9\pm1.5$ , and EIMD markers significantly decreased over the course of this study by -74.4% and -29.4% for CK and LDH, respectively (CK: p<0.001, d=0.9; LDH: p<0.002, d=1.4), while gradual weightloss did not result in profound hematological response. In Figure 1, the results demonstrated that *Tc* values of the BF and VL significantly



Figure 1. Individual adjustments of the lower limbs muscles contractile properties extracted by TMG assessment (Tc contraction time; Dm maximum amplitude radial displacement; denotes \* different from t-1 p<0.001).



Figure 2. Association between % CK activity and average contraction time for all lower limb muscles (solid and dash line represents fitted linear model, with 95% confidence intervals; %CK percentage of change in creatine kinase concentration; Tc contraction time).

Week 1	Monday	Tuesday	Wednesday	Thursday	Friday	Saturday	Sunday
Morning (8:00-9.30h)	8-km run; 3x3 SB	Muscle endurance development	10-km jogging	Sport-specific strength training	5-km interval run; 3x3 SB	Rest	6-mile jogging
Evening (18:00- 19:30h)	6Rx3.30min SSD; 3Rx3min SS	10Rx3min SSD; 3Rx3min HBW	3Rx3min SS 3x3HBW	6Rx3min; Pad work 3R 3min - SS.	8Rx3 min SSD; 3Rx3min HBW		3Rx3.30minSSD; 3Rx3min HBW
Week 2							
Morning (8:00-9.30)	8-km run; 3x5 SB	5-km interval run; 3x3 SB	8-km jogging	Sport-specific strength training	Muscle endurance development	Rest	¥
Evening (18:00-19:30)	6Rx3 min SSD; 3Rx3min SS	6Rx3 min SSD; 3Rx5min HBW	Rest	6Rx3 min SSD;3x3HBW	4Rx3 min SSD; 3Rx3min HBW		

#### Table 3. General training data during the tapering period.

Note  $\neq$  Day 1 of the investigation;  $\neq$  end of the investigation period and the investigation in parallel; Round; **SB** shadow boxing; **SSD** sport specific drills; **HBW** heavy-bag workout; **SS** sparring session.

decreased by -22.2% and -9.9%, respectively (BF: p<0.001, d=1.3; VL: p<0.001, d=0.9), while *Dm* values in BF decreased by -20.7% (p<0.001, d=0.7). There were no differences in the *Dm* of the VL (NS: p=0.13, d=0.3); and *Tc* or *Dm* of the VM (NS: p=0.23 and p=0.61, respectively). In Figure 2 Spearman correlational analysis showed a moderate association between the averaged *Tc* for all lower-limb muscles and the percentage of change in CK activity ( $r_s$ = -0.70; p=0.030) but not in LDH ( $r_s$ = 0.42, p=0.271).

# DISCUSSION

Contraction time was inversely correlated with the percentage of change in CK activity (r = -0.70; p=0.03) indicating TMG contraction time assessment as a surrogate method to denote local muscle fatigue. Surprisingly, kickboxing athletes failed to reach the >3% hypohydration threshold suggested by Savoie and authors [27] losing, on average, only -1.3% of their body mass over the course of the investigation, which was probably due to the real-life scenario approach and the exclusion of hyperthermia exposure. Contrary to our study design, 93% of elite German combat athletes reported thermal stress (e.g., sauna exposure) as a weight-reduction method to meet the desired body-mass [19]. In this context, we were able to observe the combined influence of <3% gradual body-mass loss and an increased concentration of EIMD markers

on the contractile properties of skeletal muscles.

Namely, a significant decrease in the Dm in BF (-20.2%, p<0.001) was observed, while in VL, only a decreasing trend was noted (-14.4%, p=0.13). A much greater decrease was reported by Hunter et al. [10], who showed a -31% Dm decline after eccentrically-induced EIMD response in the biceps brachii; however, they reported comparable CK levels (~600 U/L<sup>-1</sup>) to kickboxing athletes in this study. Hunter et al. [10] explained Dm decline with the shortening of muscle fibres as a result of EIMD, in agreement with the observation of Nosaka et al. [28], who suggested that muscle adapts rapidly to excessive training aiming to prevent further muscle damage by increasing muscle stiffness through several mechanisms. The findings of Hunter et al. were established using exclusively isolated eccentricmuscle contractions, under well-controlled conditions, while results presented encompassed the involvement of a much boarder span of muscle contraction types (such as stretch-shortening contraction type for striking and kicking) throughout pre-competition preparation.

A more realistic explanation was outlined by Garica-Manso et al. [29] who reported ~20% *Dm* decrement in biceps brachii muscle, following high load and volume dynamic exercise protocol. Albeit, Garica-Manso et al. [29] suggested that *Dm* decrement is instrumental to the muscle fatigue development, owing to the magnitude

of the load when performing repetitive contractions. Changes in BF muscle observed in the present study are in line with a recent report on volleyball players that highlighted alterations in BF muscle stiffness when performing different muscle contractions (isometric, isokinetic and vertical jumps) [30]. Correspondingly, wholebody, structured dynamic exercise translates into the *Dm* decrement of the lower-limb muscles in these elite combat athletes, especially during the tapering period.

Recently, Raeder et al. [31] confirmed the postulate above by demonstrating that the maximal radial displacement amplitude (Dm) of the VM is affected by excessive whole-body strength training causing Dm to decrease from 9.0  $\pm$ 2.1 to 7.7 $\pm$ 1.7 mm. Likewise, decreased values of the Dm readings were instrumental to eight weeks of plyometric training, in a randomised controlled trial of Zubac and Šimunič [24] who reported a significant reduction in Dm values (by 15-30%), in three lower-limb skeletal muscles (e.g., BF, gastrocnemius lateralis and medialis).

The Tc decreased in VL (-9.9%) as well as in the BF (-20.7%), which was also in opposition to the study of Hunter et al. [10], where the Tc of the biceps brachii increased by ~33%. The reasons for this difference are multifactorial. Hunter et al. [10] investigated the same muscle (e.g., biceps brachii) as it was used to trigger EIMD by eccentric contractions, whereas present study used the real-life intensive whole-body exercise. Because of the comparable CK responses observed in this study, it seems that the muscle damage and membrane disruptions were more concentrated in the elbow flexors of a single arm. More precisely, eccentric exercise damages primarily type II fibres, whereas whole-body exercise does not [32]. Participants in both studies were athletes (weightlifters vs kickboxers); however, kickboxers experience more habitual contractions than weightlifters, which could also contribute to differing Tc adaptations.

Research summarised by Kent-Braun et al. [33] proposed that an excessive rate of ATP turnover (subsequent to anaerobic energy demands) further promote the CK activity in the blood. Indeed, metabolic requirements of high-level kickboxing rely on anaerobic pathways to fuel energy demands during intense preparation [34], leading to increased CK activity. Previously, Detanico et al. [35] documented that CK activity increased by ~50%, even 48-h post-judo training session. Importantly, kickboxing athletes had an elevated concentration of EIMD serum markers in their blood already at t-1, which possibly mirrors the response to muscle damage from previous mesocycles, as training volume and intensity were reduced by ~30% during the tapering period. Moreover, Clarkson and authors [2] suggested that muscle damage occurred through both metabolic and mechanical mechanisms, while the time to peak CK response occurred 4 days post-exercise.

Meanwhile, other hypotheses indicate that greater increases in serum CK activity after EIMD were associated with a shorter Tc, a measure of estimated MHC-I proportion. More precisely, deceased Tc values indicate higher fast-twitch muscle fibre proportion. For example, the estimated VL MHC-I proportion (extracted from the linear combination of contraction time, delay time and half-relaxation time) decreased by -8.2% (p=0.041) in parallel with decreased Tc, as a result of eight weeks of plyometric training [24]. Also, Macaluso et al. [36] reported micro trauma of predominantly type II fibres following a single bout of plyometric training. Increased CK levels and damaged fibres around the sarcolemma and the sarcomere (at the site of the Z-disc) were found. Substantial metabolic demands during excessive exercise may be coupled with a structural disadvantage (e.g., thinner and arguably weaker Z-discs), subsequently resulting in an increased risk of greater damage to type II muscle fibres [28]. The correlation between averaged Tc of all investigated muscles, previously found to be related to type I fibres [8], and the CK level was significant ( $r_{c}$  = -0.70, p=0.03). This finding poses a new research question investigating interactions between skeletal muscle composition and EIMD responses. Nevertheless, from an applied perspective present findings are in agreement with García-Manso et al. [11], who recommended TMG as a simple, non-invasive and selective diagnostic tool to screen local muscle fatigue following real-life competitive scenario.

# CONCLUSIONS

Apparently, after a 2-week tapering period followed by gradual body mass loss; kickboxers decreased their EIMD and improved their muscle contractile performance. Adjustments in contractile indicators of the lower-limb skeletal muscles coupled with this association could have a substantial effect on training planning *per se*. Striking combat sports athletes could reduce the risk of muscle injuries and improve athletic performance by detecting changes in cross-bridge kinetics and muscle belly amplitude response. Therefore, present findings imply that the TMG assessment during the tapering period is a viable tool to selectively screen and control skeletal muscle fatigue, but also to evaluate adjustments in contractile capacity. Importantly, it seems that the CK activity decrease was lower in combat sports athletes with lower averaged *Tc*, an indirect measure of MHC-I proportion. However, further research is encouraged as the total numbers of athletes analysed were low, and these results should be confirmed by direct assessments of muscle composition.

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