MECHANICAL PROPERTIES ANALYSES OF THE GASTROCNEMIUS MUSCLE ELECTRICAL STIMULATED AFTER IMMOBILIZATION

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Abstract: This study evaluated some mechanical properties of the gastrocnemius muscle immobilized and submitted to neuromuscular electrical stimulation (NMES) through Russian current. Thirty-two (32) Wistar female rats were used, divided into four groups: control (CG), immobilized (IG); immobilized and later free mobilization (IFG) immobilized and later NMES (NMESG). Mechanical experiment of longitudinal traction was carried out to attain the mechanical properties. Results: maximum load 33.68±3.58N (CG), 18.36±4.43N (IG), 25.54±2.88N (IFG) and 28.25±5.88N (NMESG). Maximum elongation 18.37±0.49x10^{-3}m (CG), 10.84±2.11x10^{-3}m (IG), 15.82±1.71x10^{-3}m (IFG) and 17.08±2.30x10^{-3}m (NMESG). Stiffness 1555.14±222.43N/m (CG), 2193.60±847.90N/m (IG), 1715.25±137.25N/m (IFG) and 1654.80±262.28N/m (NMESG). The study suggests that the NMES is not capable to reestablish the muscle mechanical properties after the immobilization.

Keywords: Skeletal muscle, immobilization, electrical stimulation.

INTRODUCTION:

The muscle is the motor of the human body element, which brings into action the corporal segments in a spontaneous or reflective way. The function of the skeletal muscle depends on intact proprioceptive activity, motor innervation, mechanical load and articular mobility. The muscle is the most mutable among the biological tissues and it responds to normal or altered commands with both morphological and functional adjustments (Appell, 1986).

Muscular tissue holds a major incidence of traumas because of the practice of sports causing muscular injuries through direct or indirect impact contusions (Crisco, 1994; Hurme, 1991). Muscular activities of those who practice sports are studied by many specialists, in particular those individuals who are responsible for maintaining the athletes osteomyoarticular structure in excellent condition and thus enabling them to have better physical, technical and tactical capability (Lopes, et al.,1995).

The induced alterations by the immobilization of a segment are not restricted to the muscle, since innervation and circulation damages, bony atrophy, ligamentous alterations, edema and articular rigidity are commonly observed (Booth, 1977). It is important to verify these properties because they are common in injuries of the athletes.

Referring to the mechanical properties of the skeletal muscle we can describe: the rigidity as being the strength of deformity, as estimated from the length-tension curve, load *versus* lengthening; the load in maximum limit as being the highest value of strength reached by the muscle, and the lengthening in maximum limit being the major deformity reached by the muscle (Loitz et al., 1989).

The neuromuscular electrical stimulation (NMES) consists in the administration of therapeutic electrical stimulus on the muscle, throughout the whole nervous system (Järvinen, Einola, Virtanem, 1992). This technique, whenever applied on the intact peripheral neuromuscular system, is capable of producing potentials of actions on nerves and muscles which have been not characterized from those emitted by the regular nervous system (Fournier,1983).

The Russian current is a cord of mean frequency, sinusoidal alternated (two-phase) at 2.500 Hz and it is capable of producing deeper levels of muscular contractions and beside, it amplifies the strength, being more indicated on muscle treatment with preserved innervation (Fournier,1983).

Athletes need to improve their performance and they lose it when are immobilized. Russian current could be an alternative during immobilization period. Then, the aim of this experimental study was to evaluate the effects of neuromuscular electrical stimulation on gastrocnemius muscle immobilized of rat.

MATERIALS AND METHODS:

Thirty-two (32) female Wistar rats were utilized, then divided into 4 groups: Control (CG, n=8), these animals were exempted from any intervention; Immobilized (IG, n=8) they had their posterior right members immobilized during 14 days; Immobilized/Liberated (IFG, n=8), they remained immobilized for 14 days and afterwards released for 10 consecutive days; Immobilized/Electrical stimulated (NMESG, n=8) they were immobilized and after this period of time they were submitted to NMES of mean frequency during 10 consecutive days.
The immobilization of the right posterior member of the animals was accomplished using a plastered machine including the pelvis, hip and knee in total extent and the articulation of the ankle in plantar flexion. The animals were previously anaesthetized with hydrochloride of ketamine (50 mg/kg) and xylazine (15 mg/kg). The plaster was replaced whenever needed. The animals remained immobilized for two weeks (14 consecutive days) and separated in plastic cages with free access to water and food.
For the therapeutic intervention by NMES, a Russian current generator was utilized with a complete cycle including an *on cycle* of 6 seconds was used, including the *rise* of 2 seconds, the *decay* of 2 seconds and the *off cycle* of 13 seconds, for a total of 10 contractions. The current was transmitted to the animal through the cables and electrodes (carbon silicon, dimension 5x3 cm and pen developed in a minor scale, to local stimulation of the gastrocnemius muscle).
To the mechanical experiment of the gastrocnemius muscle, an universal experimental machine, EMIC® DL-3000 model was utilized. It belongs to the Research Laboratory of Odontological Material from Universidade de Uberaba Uberaba and is equipped with a 50 kgf load capacity cell Kratos®.
The machine was connected to a micro-computer with eqquiped a softwear able to detect the values of the loads and stretching during the mechanical experiments. The velocity of longitudinal traction was set at 10 mm/min. The gastrocnemius muscle was dissected preserving its origin (femur) and insertion (caucaneum) which allowed, through the use of a special apparatus, the fixation of the bone-tendon-muscle-tendon-bone complex to the experimental machine. For each enlargement of the load applied to the muscle, a resultant value of the stretching was attained, permitting the construction of the graphic, load *versus* lengthening, through the Microsoft Excel 2003® program. ANOVA test was utilized for the simultaneous analyses of the groups and the "Tukey-Kramer" test was utilized for comparison between the groups, to adjust p=0.05 for Type 1 error.

RESULTS

Table 1 Means mechanical properties evaluation in gastrocnemius muscle

	CG	IG	IFG	NMESG
Maximum Load	33,68 ± 3,58	18,36 ± 4,43■;●;♦	25,54 ± 2,88■;▲	28,25 ± 5,88▲
Maximum Elongation	18,37 ± 0,49	10,84 ± 2,11■;●;♦	15,82 ± 1,71■;▲	17,08 ± 2,30▲
Stiffness	1555,1 ± 222,43	2193,6 ± 847,90■	1715,3 ± 137,29	1654,8 ± 262,28

■: p<0.05 (compared with CG); ▲: p<0.05 (compared with IG);
●: p<0.05 (compared with IFG); ♦: p<0.05 (compared with NMESG).

DISCUSSION:

The muscle adjusts itself in accordance with the stimulus it undergoes, when it is immobilized in a short-term position it shows greater atrophy on the muscular fibers, a larger number of

crossed bridges, reduction in the number of sarcomeres and consequent diminishment of the muscular length (Williams, 1987).
It was observed in this study that the stiffness property of the group submitted only to immolibization in short-term position demonstrated higher values when compared to the other groups. Amongst some of the factors that could interfere, altering the muscle mechanical properties, would be an enlargement in the quantity of the connective tissue produced by the matrix extracelular in the muscular fibers. The immobilization for a period of 14 days would not be able to supply this augmentation of the connective tissue that would induce mechanical alteration (Mattiello-Sverzut et al., 2006). The alterations present in our results could be related to other causes, which have not been evidenced on the evaluation of the mechanical properties, but they could be elucidated through a histomorphological evaluation.
Taking into consideration the alterations of muscular structures and functions due to the immobilization, the muscular plasticity demonstrates the capacity in the rehabilitation of its original properties independent if they are derived or not from external stimulations. In the present study we conduced a free motion type of treatment, since Järvinen et al., (1977), declared that the muscular mechanical properties can be reestablished through realization of any type of exercise and electrical stimulation with Russian current, because such stimulation by Russian current of modulated medium frequency at 50Hz is capable of reestablishing the skeletal muscle properties (Carvalho, 2004).
Järvinen, (1993) and Kujala, (1997), stated that active mobility can help in disappearance of edema, in the increase of extensiveness and the organization of myofibrils. In our study it confirmed that the free movement is able to increase the limits to prolongate and in agreement rigidity the cited authors above. We also support the findings of Mattiello-Sverzut et al., (2006), who through morphological analyses, have reported decreases in cellular transformations such as, cellular reactive activity diminishment. Demonstrating that the animals belonging to the Free Immobilized Group, could show minor loss of fibers type I and II in diameter compared to those from the Immobilized Group.
The muscular staple fibers of fast contraction (type II) are enlisted to add muscular force and rapidity to the movement, these answer to the frequencies in the band of 50-150 Hz. The staple fibers of type I (slow contraction) are the first to become active and have a tetanic frequency of 20-30 Hz (Low; Reed, 2001). This way, in order to reestablish the muscular strength lost at the moment of the immobilization procedure, we chose to utilize a 50 Hz, frequency, because in accordance with what Carvalho (2004) declares, after a period of 14 days of immobilization the electrical stimulation of the muscle at such frequency reestablishes some mechanical properties of the muscle as for example, stiffness, resilience, load and elongation, at its maximum limit. This could be important in treatment of athletes.
Mattiello-Sverzut et al., (2006) declared that the neuromuscular stimulation could be able to generate hypertrophy to the type II fibers, such fibers should be preferentially electrical stimulated at 50Hz frequency. In our study we are able to confirm that the stimulation through the use of Russian cable at 50 Hz frequency is capable of bringing back to the after-immobilized muscle its maximum load properties, maximum elongation and resilience. However, this improvement did not reach control levels, suggesting a time period of longer than 10 days of electrical stimulation through the use of Russian current are necessary.

CONCLUSION:

This study suggests that the NMES through the Russian current following determined protocol, is not capable of reestablishing the mechanical properties of the muscle after the immobilization at control level, although when comparing just the liberation, this technique shows better results. Russian current could be a specific alternative to immobilized athletes but the period of stimulation must be more than 10 days. We suggest more studies to verify this correct period to improve the performance of athletes and to decrease sequels of immobilization.

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