AEROBIC TRAINING PREVIOUS TO NERVE COMPRESSION: MORPHOMETRY ANALYSIS OF MUSCLE IN RATS

EXERCISE AND SPORTS SCIENCES

ORIGINAL ARTICLE

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ABSTRACT

Introduction: Sciatica stems from compression of the sciatic nerve and results in pain, paresthesia, decreased muscle strength and hypertrophy. Exercise is recognized in the prevention and rehabilitation of injuries, but when performed in overload, it may increase the risk of injury and subsequent functional deficit. Objective: To evaluate effects of aerobic training prior to an experimental model of sciatic pain concerning morphometric parameters of soleus muscles of rats. Materials and methods: 18 rats were divided into three groups: sham (dip, 30 seconds), regular exercise (swimming, 10 minutes daily) and progressive aerobic training (swimming, progressive time from 10 to 60 minutes daily). After six weeks of exercise, rats were subjected to the experimental model of sciatic pain. On the third day after injury, they were killed and their soleus muscles were dissected, weighed and processed for histological analysis. The analyzed variables were: muscle weight, cross-sectional muscle area and mean diameter of muscle fibers. Results: Statistically significant difference was observed for all groups when compared to control muscle and that submitted to sciatic injury. Intergroup analysis showed no statistically significant difference for any of the variables. Conclusion: Both regular physical exercise and aerobic training had no preventive or aggravating effects on the consequences of functional inactivity after sciatica.

Keywords: aerobic exercise, primary prevention, sciatic nerve, skeletal muscle.

INTRODUCTION

Sciatic pain, commonly named sciatica, is also found as radiculopathy, lumbosacral radicular syndrome, radicular pain and compression or irritation of nervous root, originates from the compression of the sciatic nerve and is a symptom referred by the trajectory of this nerve and its branches, distributing by the respective dermatomes and myotomes[1-4]. It is estimated that 40% of the world population will experience it in any time of their lives; however, approximately 1% will present motor or sensitive deficit[2-4], being also one of the main causes for absences, representing an economical and health problem[2]. Its etiological characterization includes syndrome of the piriformis muscle[2,3], besides tumor processes, many congenital mechanical, trauma, degenerative, functional conditions, inflammatory and metabolic conditions involving vertebral articulations[1-8].

Commonly, sciatic pain is followed by low back pain due to the compressing of this region, in a situation known and differentiated as lumbosciatica[5,6]. Other clinical characteristics derived from sciatic compression include paresthesia[7], decrease of muscular strength [4,7], claudication[6], and consequent muscular hypotrophy[3], leading hence to functional deficit [2].

The muscle fibers are equipped with high capacity of physiological and biochemical adaptation according to the stimuli to which they are submitted, reflecting in size, metabolism and type of muscle fiber[9-11]. Besides the positive physiological responses due to the posture maintenance, athletic performance and injury repair, the muscle loses mass and function in disuse situations[9,11], as the sciatica consequences already mentioned above.

Physical exercise promotes adaptations to the muscular oxidative metabolism, as increase in the number and size of mitochondria, increase in enzymes expression and activity of the energetic metabolism of biochemical ways, increase in capacity of storage of energetic substrates and in protein synthesis[10]. In a few weeks of training alterations in the sinapses of the motor units and cross-sectional area individually in each fiber can be observed[9].

Musculoskeletal and mechanical overloads in the sports activities may trigger disc degeneration which lead to nervous compression, causing nervous irritation and pain, consequently leading to functional deficit[12,13]. On the other hand, physical training is used in rehabilitation of these conditions, so that muscular rebalance and return to function can occur[12]. However, there is still some questioning about the advantages of physical activity in prevention or minimization of the sciatica consequences[14].

Therefore, the aim of the present study was to evaluate the effects of an aerobic training in water medium, previous to a sciatica experimental model, concerning morphometric parameters of soleus muscles in rats.

MATERIALS AND METHODS

Study and sample characterization

18 male Wistar rats, mean weight 413 ± 49 g and 14 ± 2 weeks

old, obtained from the Central Animal Facility of the State University of Western Paraná were used. This study was according to the International Guidelines of Ethics in Animal Experiment [15] and was approved by the Ethics in Animal Experiment and Practical Classes Committee (CEEAAP/Unioeste) under protocol 68/09. The animals were grouped and kept in polypropylene cages in the animal facility of the Laboratory of Study of Injuries and Physiotherapeutic Resources under controlled environmental conditions, with a 12-hour light/dark cycle, temperature at 23ºC ± 2ºC and water and food *ad libitum*.

The animals were randomly divided in three groups:

Simulation Group (SG, n = 6) – was submitted to a 30-second period in a tank with heated water, during six weeks, representing a sedentary group concerning physical activity;

Regular Exercise Group (REG, n = 6) – performed daily aerobic exercise (swimming) of 10 minutes. for six weeks, representing a group practitioner of regular physical activity;

Progressive Training Group (PTG, n = 6) – performed daily progressive swimming sessions for six weeks, starting with tem minutes of swimming on the first week, 20 minutes on the second week and successively until completing 60 minutes of aerobic physical activity on the sixth week, representing a progressive aerobic training group.

On the day following the last day of aerobic exercise performance (swimming), all animals were submitted to a sciatica experimental model and on the third day, after injury induction, they were anesthetized and euthanized by guillotine decapitation, for isolation of the right and left soleus muscles of each animal. These muscles were weighed on an analytical scale (Shimadzu®). Subsequently, the muscles were stained in Bouin and stored in alcohol 70%[16] and then followed the routine histological processing[17] for analysis in light microscope.

The soleus muscle was chosen due to its easy access and predominance of type I fibers; that is, fibers which use aerobic metabolism as main energy source[10], in agreement with the aim of the chosen exercise.

Protocol of the aerobic exercise

The aerobic exercise (swimming) was performed in a sturdy plastic oval tank with capacity of 200 liters and depth of 60 centimeters, with water kept at controlled temperature of 31 ± 1°C (Incoterm® thermometer), during six weeks of experiment. Each group performed respectively exercise time as previously mentioned, being five consecutive days of training followed by two recovery days in all weeks of the research. After swimming, all animals were dried in a cotton towel and placed in their boxes.

Sciatica experimental model

The neuropraxy experimental model in the sciatic nerve occurred with the animals being previously anesthetized with an intraperitoneal injection of xilazine (4 mg/kg) and ketamine (35 mg/kg) combination. After having been anesthetized, they were submitted to tricotomy in mean third of the right thigh and local hygienization with polyvinylpyrrolidone-iodine (Povidine®). Subsequently, a parallel incision to the fibers of the biceps femoris muscle was made with a scalpel with number 10 blade, exposing hence the sciatic nerve. The sciatic nerve was compressed through four nodes at distinct regions, one milliliter away from each node, using chromed 4.0 Catgut, based on model originally described by Bennet and Xie in 1988[18]. The fasciae and cutaneous tissue were then sewed with suture needle and thread, with two stitches and skin was again hygienezed with polyvinylpyrrolidone-iodine.

Histological processing of the muscle tissue

The dissected soleus muscles, after having been weighed, were stained in Bouin for 24 hours at room temperature. After that period, they were stored in alcohol 70ºGL at 4ºC[16] until the histological processing in paraffin, which followed conventional protocol of histological routine. 7 µm cuts were obtained in CUT 4055 rotating microtome (Olympus®), totalizing five cuts per mount, which were stained with hematoxylin and eosin (HE)[17].

Shot and analysis of the histological images

The mounts were analyzed by light microscope (Olympus®) with attached camera (DCE-s) and 40 x objective for digitalization of images of the transversal cuts of the muscle fibers[19]. Afterwards, with the use of the image-Pro-Plus 3.0 program, area and mean diameter of 100 fibers per muscle were assessed[20].

Statistical analysis

The data were validated comparing the results obtained of the soleus muscles of the left hind limb (control) and right (submitted to sciatic compression), among animals from the same experimental group, using paired Student's *t* test and one-way ANOVA for intergroup comparison, with Tukey post-test, being $p < 0.05$ considered significant.

RESULTS

Intragroup analysis

Based on the analysis of the muscle weight parameter, as well as of the transversal histomorphometric parameters (transversal area section and mean diameter of the muscle fibers of the soleus muscles), statistically significant difference for all groups was observed when the left (control) and right (experimental, submitted to sciatic injury) were compared, as demonstrated in table 1; that is, neither regular physical exercise nor progressive training with aerobic exercise were sufficient to minimize or eliminate the deleterious effects of the nervous compression.

Intergroup analysis

The results demonstrated in the analysis between the different experimental groups (SG, REG and PTG) did not reveal statistically significant difference for any of the analyzed variables when the morphometric parameters values of the soleus muscles of the right hind limb after the nervous compression are observed (table 1).

DISCUSSION

Light physical activity, or even progressive exercise, many times used as therapy for muscular rebalance and functional return[12], was not useful in the progressive study. However, it was observed that despite the absence of significant results concerning prevention

Table 1. Means and standard deviation of the muscular weight, cross-sectional area and mean diameter of the fibers variables, according to the experimental group, compared between right soleus (R) and left soleus muscle (L) values.

Muscular weight (in g)			
	R	L	P value
SG	0.1158 ± 0.01508	0.1542 ± 0.02614	0.0211
REG	0.1036 ± 0.01278	0.1853 ± 0.02561	0.0001
PTG	0.1124 ± 0.01262	0.1805 ± 0.02108	0.0009
Cross-sectional area (in μm^2)			
	R	L	P value
SG	580.9 ± 96.78	974.2 ± 213.1	0.0021
REG	650.5 ± 108.9	1142 ± 135.3	0.0074
PTG	586.9 ± 135.4	1125 ± 188.9	0.0014
Mean diameter of the fiber (in µm)			
	R	L	P value
SG	20.54 ± 2.083	27.46 ± 3.260	0.0214
REG	22.14 ± 1.535	30.26 ± 2.514	0.0018
PTG	20.79 ± 2.286	29.45 ± 2.493	0.0018

of muscle hypotrophy, for the groups which performed physical exercise, there was not worsening of any of the characteristics when compared with the simulation group; that is to say, physical exercise in the used protocols did not produce positive or negative effects on the muscular tropism in animals with hinder nervous compression.

Contractile activity is the main factor for development, maturation and maintenance of the structures of the muscle fibers [21] and, in disuse situations, muscular mass loss is observed[9,11]. This disuse may occur as consequence of the loss of function derived from paresthesia[2] and pain, caused by the nervous compression[2,4].

In a few weeks of resistance training, individual alterations in the cross-sectional area can be observed in each muscle fiber[9]. Swimming was chosen because besides being an aerobic activity with increase of heart and respiratory rate, due to its viscosity property, which offers resistance to movements in any direction [22], contributing for a muscular resistance training.

Regular exercise promotes antioxidant protection of the muscle cells[23,24]. Venojärvi *et al.*[25] observed that both regular physical activity and intense exercise increase proteins which contribute to the restoration of protein homeostasis of the muscle fibers, without affecting the oxidative stress or antioxidante protection. Moreover, the expression of these proteins may have therapeutic effects and contribute to the protection against muscular atrophy and degeneration in periods of disuse. However, in this study it was not possible to verify such prevention benefits of regular exercise and aerobic training, over muscular atrophy consequent of sciatic pain. Probably, the type of training chosen, resistance, had not been sufficient to generate increase of muscle mass in order to minimize atrophy by disuse due to the sciatica. Therefore, further research involving resistance exercises with load inclusion is suggested.

Nevertheless, Stafford *et al.*[2] report that regular walking and running seem to cause double of the incidence of sciatic pain. Aerobic physical exercise, in this investigation performed in aquatic environment, despite having not obtained significant results in prevention of hypotrophic characteristics (muscle weight, crossed-sectional area and mean diameter of the muscle fibers), was not connected with any losses either, to these same characteristics. It is worth mentioning that in aquatic environment, there is less impact of the structures and less stress to the muscle fibers, comparing to the activities performed on the ground, due to the thrust, viscosity and water heating properties[22]. Such features may have protected the muscle fibers from oxidative stress which would be aggravating to the degradation signs subsequent to nervous compression.

Muscle contraction is one of the main factors for activation of protein synthesis [10,26]. A insulin muscular growth factor, the IGF-1 is positively regulated by physical activity[27-29], and during intense exercise, the most circulating part of this factor is produced by the muscle, and effectively used by it[26]. Increase in the IGF-1 levels may result in hypertrophy of the muscular tissue through signs which stimulate proliferation, differentiation and fusion of satellite cells[28,29]. Probably, the way the exercise was applied in this investigation did not allow stimulation of this growth factor production to the extent to generate hypertrophy which would compensate the damage caused by the muscular disuse.

Goldspink[26] also describes that the IGF-1, when produced from the growth hormone, is the main regulator in the development of muscular mass during childhood; however, in adulthood, when produced by the muscle during exercise, it becomes the most important factor for muscular mass maintenance. Moreover, IGF-1 plays an important role in the prevention of muscular atrophy, since in response to injuries, the satellite cells stimulate increase of the IGF synthesis, leading them to proliferation and differentiation, forming myoblasts which blend to generate new fibers[29].

A study developed by Garcia *et al.*[30], using training methodology similar to in this investigation, presented in the morphometric analysis of soleus of rats submitted to daily training that, after it, some muscle fibers presented initial and complete stages of the phagocytosis process, angular fibers, atrophic, centered nuclei and sarcolemma loss, which indicate the onset of degeneration. Although an analysis at the cellular level has not been done in this study, it is possible that it had occurred after the proposed training, added to the damage derived from the nervous comprehension.

Despite the specific care about study extrapolations with animals, it can be inferred that the sequelae on muscular tropism for a short training period, does not differ between trained and untrained individuals. Moreover, it can be stressed that the lack of positive results in the present study concerning aerobic training in prevention of muscular degeneration by the nervous compression may be attributed to the lack of intervals between the interventions with training, which were daily performed, five consecutive days, with interval of only two days and again, five days of training; since in studies [21,31,32] demonstrated that longer intervals between mechanical stimulations on the muscle fiber are more favorable to muscular growth, that is, a great limitation presented by the exercise protocol used. Another limitation to be mentioned was the training period of six weeks, which may have contributed to the absence of effects, indicating hence that further research with longer period of interval exercises is needed.

CONCLUSION

In the present study, neither regular physical exercise nor aerobic training such as swimming, according to the used protocol, produced prevention effects, not even aggravating, on the muscular consequences of functional inactivity caused by sciatic pain derived from experimental nervous compression.

All authors have declared there is not any potential conflict of interests concerning this article.

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