BALANCE AND COORDINATION TRAINING IMPROVES SENSORIMOTOR FUNCTIONS AND SCIATIC NERVE REGENERATION AFTER PERIPHERAL NERVE INJURY MODEL

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Numerous therapeutic interventions have been tested to enhance functional recovery after peripheral nerve injuries, and various forms of exercise training have shown beneficial effects in muscle and nerve function-related parameters in animal models of nerve injury [1,2,3]. The present study was designed to compare the effects of balance and coordination training versus motor control training on sensorimotor recovery, and to carry out a morphometric analysis of the sciatic nerve and soleus muscle fibers after nerve injury through sciatic nerve crushing.

The experiment was performed on 23 three-month-old, male Wistar rats. Animals were randomly divided in four groups: (1) sham-operated rats, without sciatic crush and unexercised, sham (SH, n = 5); (2) rats with sciatic crush and unexercised, sedentary (SE, n = 6); (3) rats with sciatic crush and motor control training (MC, n = 6); and (4) rats with sciatic crush and balance and coordination training (BC, n = 6). For the surgical procedures, animals were anesthetized using ketamine and xylazine and the right sciatic nerve was exposed through a skin incision. The nerve crush injury was performed with 1 mm hemostatic forceps for 30 seconds. Before the surgical procedures, animals were adapted for 5 days in each training program protocol. The motor control training program was adapted from Kleim et al. [4] and was performed on a flat obstacle-free runway 100 cm long and 8.5 cm wide ending in a dark box. Each rat of this group crossed this flat obstacle-free runway 25 times. For balance and coordination training program [4,5,6], animals were required to traverse 5 different elevated obstacles per day, such as suspension bridges, rope bridges, parallel bars, etc. (each 100 cm long) ending in a dark box, each rat of this group crossed these obstacles 25 times. The motor control, such as the balance and coordination training program, comprised 5 sessions per week for 4 weeks. Two days after the sensorimotor tests, the animals were transcardially perfused with saline solution, followed by 0.5% glutaraldehyde and 4% paraformaldehyde in a 0.1 M phosphate buffer (pH 7.4, PB) at room temperature. The right soleus muscles were dissected, small samples of the central part of each muscle were selected and two short segments of the right sciatic nerves were excised. The materials were fixed by immersion in the same fixative solution and postfixed in phosphate-buffered OsO4, dehydrated in ascending alcohol and acetone, and embedded in araldite. Semithin sections (1µm) were stained with 1% toluidine blue in 1% sodium tetraborate. Images were captured, digitalized and processed with Image Pro Plus software 6.0. For morphometric evaluation of the muscle, a set of 5 images was chosen using random sampling of one slice, twenty different muscle fibers of each image were selected. Morphometric measurements of the soleus muscle included the averaged muscle fiber area (μm^2) [7]. For the nerve evaluation, both proximal and distal portions of the right sciatic nerves were separately analyzed, a set of 8 images was chosen using random sampling of one slice of each portion. Morphometric measurements of the sciatic nerve included the (1) averaged myelinated fiber area (μm^2); (2) average myelin sheath thickness (µm); (3) averaged myelinated fiber diameter (µm); (4) averaged axon diameter (µm) of the myelinated fiber and (5) *g* ratio (the quotient axon diameter/fiber diameter, a measure of the degree of myelination). In the analyses of the morphometric parameters, the mean area of muscle fibers was larger in the animals performing coordination and balance exercise than in the sedentary and motor control groups (Fig. 1). Concerning the average myelin sheath thickness and *g* ratio of the distal portion of the sciatic nerve, the balance and coordination group showed significantly better values than the sedentary and motor control groups (Fig. 2). The findings indicate that balance and coordination training improves sciatic nerve regeneration after an experimental traumatic injury, as trained animals showed a greater degree of myelinated fiber maturation, so as to make it possible to revert/prevent soleus muscle atrophy and improve performance in sensorimotor tests after 4 weeks of training.

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References:

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Figure 1 - Digitized images of transverse-semithin sections (1 µm) obtained from central part of right soleus muscle after 4 weeks of specific exercise training. A: central part of soleus muscle of the SH group. B: central part of soleus muscle of the SE group. C: central part of soleus muscle of the MC group. D: central part of soleus muscle of the BC. This images show that the structure of the muscle fibers in the BC group and in the SH group were similar, presenting most of the large muscle fibers, with polygonal shape and little conjunctive tissue between them. The fibers muscle of SE and MC groups are shorter and rounded and had much conjunctive tissue between them. SH = sham; SE = sedentary; MC = motor-control trained; BC = balance and coordination-trained. Scale bar = 50 µm.

Figure 2 - Digitized images of transverse-semithin sections (1 µm) obtained from regenerating sciatic nerves after 4 weeks of specific exercise training. A and B: proximal and distal portions, respectively, from normal nerves of the SH group. Note the bimodal fiber spectrum, the large and small myelinated fibers and small space existent between the fibers. C and D: proximal and distal portions, respectively, from regenerating nerves of the SE group. Note the small differences between the proximal portions of SE and SH groups; however, the distal portion of SE group show the predominance of small-diameter thin myelin sheath fibers, increase in endoneurial connective tissue between the nerve fibers and degeneration debris in this portion of the nerve. E and F: proximal and distal portions, respectively, from regenerating nerves of the MC group. Note the small differences between the proximal portions of MC and SH groups. At the distal portion, myelinated fibers appear to be similar to the SE; with predominance of small-diameter thin myelin sheath fibers, increase in endoneurial connective tissue between the nerve fibers and degeneration debris in this portion of nerve. G and H: proximal and distal portions, respectively, from regenerating nerves of the BC group. The proximal portion of the BC group appears to be similar to the others groups; nevertheless, in the nerve fibers of the distal portion, note that the myelin sheath thickness appear to be more large than those of the SE and MC groups. Mf = myelinated nerve fiber; Sc = Schwann cell; * (asterisk) = endoneurial connective tissue; Dd = degeneration debris; BV = blood vessel; SH = sham; SE = sedentary; MC = motor-control trained; BC = balance and coordination-trained; P = proximal portion; D = distal portion. Scale bar = 20 µm.

Figure 1

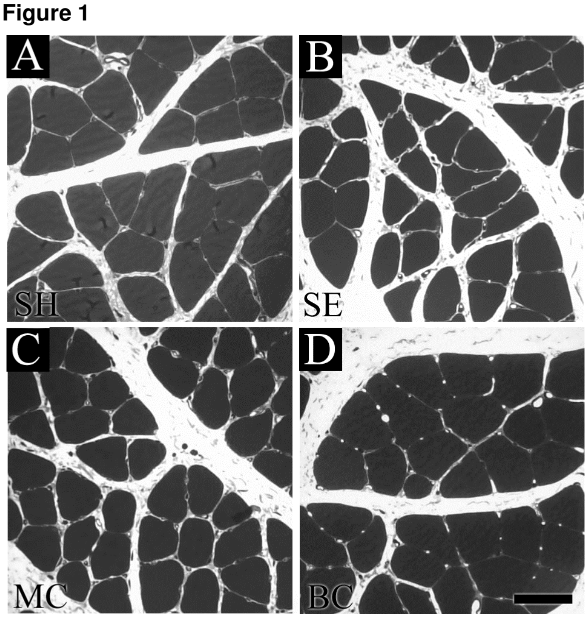

Figure 2