Efficacy of functional electrical stimulation in ameliorating inactivity-induced atrophy in a fast hindlimb muscle in rat

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Abstract

The amount and pattern of electromechanical activity, i.e., activation and loading, are important determinants in the homeostatic regulation of skeletal muscle. The purpose of this study was to examine the effects of inactivity on muscle atrophy, and to determine the optimum electromechanical stimulation paradigm for maintaining muscle mass using a wireless implantable microstimulator (BION® Microstimulator). The model of spinal cord isolation (SI) was used to create a baseline of near zero activity to which known amounts and patterns of activation could be imposed. Beginning 2 days after SI surgery, the BION® implanted leg received daily electromechanical stimulation to induce isometric contractions. All stimulation protocols resulted in a total of 1 to 4 min of activity in a 24-hr period. After 30 days, the same amount of high-load, short-duration stimulation delivered in two vs. one bout per day was more efficacious in ameliorating muscle atrophy (72 and 79% of control, respectively). At a shorter training duration (5 days), the frequency, number, and intensity of stimulation were adjusted to produce an optimum paradigm, one that preserved 99% of the relative muscle mass. These results indicate that as little as 1 to 4 min of patterned high-load isometric contractions per day in an otherwise inactive muscle can preserve a normal homeostatic state and thus prevent muscle atrophy, and that distributing the same amount of activity in two sessions/day was more effective than in one session/day.

1. INTRODUCTION

Skeletal muscle mass is closely dependent on the level of activation and mechanical loading. As seen in models of spaceflight and spinal cord injury, a reduction in the level of neuromuscular activity results in muscle atrophy.[1,2] In contrast, an increase in activation and loading as occurs with functional overload and electrical stimulation under loaded conditions can induce hypertrophy. However, the models used in these studies have unknown levels of background electromechanical activity, making it difficult to parcel out its role in determining muscle mass. Spinal cord isolation (SI) is a model that virtually eliminates neuromuscular activity while maintaining the integrity of the motoneuron-muscle connection.[7] Therefore, this model provides a baseline of near zero activity to which known amounts and patterns of electromechanical loading can be imposed.

The purpose of this study was to use the SI model to determine 1) the effects of inactivity on muscle mass, and 2) the most efficacious electromechanical stimulation paradigm for maintaining the mass of an otherwise inactive fast hindlimb extensor muscle. Furthermore, these studies tested the feasibility of using a relatively non-invasive wireless implantable microstimulator (BION® Microstimulator) as a rehabilitative strategy for debilitating neuromuscular conditions.

2. METHODS

Adult female Sprague-Dawley rats (221 ± 2 g body weight) were assigned randomly to a normal control (Con), SI-Sham, or SI stimulated (SI-Stim) group. The SI surgery procedure is a modification[3,7] of the original protocols of Tower.[7] The SI model involves mid-thoracic and high-sacral complete spinal cord transections plus bilateral deafferentation between the two transection sites and results in near inactivity in the motoneurons in the isolated region and in the associated musculature. A wireless device (BION® Microstimulator[5], Alfred Mann Foundation) designed for functional electrical stimulation was implanted in SI-Stim rats in the thigh along the sciatic nerve in one leg. SI-Sham rats were implanted with a non-functional BION®.

During stimulation training of SI-Stim rats, the BION® implanted leg was secured in a cast (ankle at 120°, knee at 90°) to optimize isometric contractions of the plantarflexors and dorsiflexors. A force transducer embedded in the plantar surface of the cast enabled monitoring and quantification of the torque at the ankle produced by each stimulation. SI-Stim rats received daily stimulation training beginning two days after surgery (unless otherwise specified). One cycle of stimulation consisted of a 1 or 4-sec isometric contraction delivered every 30 sec for 5 min, followed by 5 min of rest (Fig. 1). A series of stimulation regimes were tested to determine the optimum paradigm for maintaining muscle mass by adjusting the stimulation duration, frequency, and number of stimulation cycles delivered each day (Table 1). The medial gastrocnemius (MG), a predominantly fast ankle extensor and knee flexor, was then removed, trimmed of fat and connective tissue and weighed (wet weight) after 5 and 30 days of training. All procedures were approved by the UCLA Chancellor's Animal Research Committee and followed the American Physiological Society Animal Care Guidelines.

One-way analyses of variance followed by Tukey post hoc tests were performed to determine statistical significance ($P \leq 0.05$).

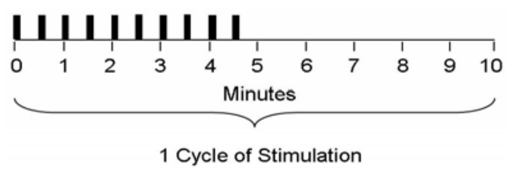

Figure 1: Illustration of one cycle of stimulation. Over a 10 min period, isometric stimulations (rectangular tick marks) were delivered once every 30 sec for 5 min, followed by 5 min of rest.

SI-Stim Protocol #	1	2	3	4	5	6	7
Total Days of Stim	30	30	5	5	5	5	5
Frequency (Hz)	100	100	100	50	100	100	100
Stim Duration (sec)	1	1	1	4	1	4	4
Cycles/Session	6	3	3	3	6	3	3
Sessions/Day	1	2	2	2	2	2	2
Total Stim Duration/Day (sec)	60	60	60	240	120	240	240

Table 1: The parameters of each stimulation protocol (SI-Stim 1 to 7).

3. RESULTS

There were no statistically significant differences in the muscle mass and physiological properties among the SI-Sham or the contralateral non-stimulated MG muscles of the SI-Stim groups. Thus, data from these muscles were combined and are presented as the SI group. The absolute and relative MG masses are summarized in Figure 2.

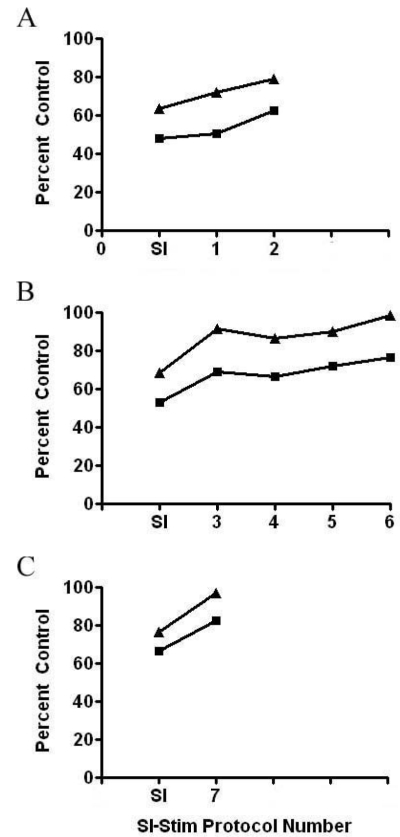

Figure 2: Absolute (square) and relative (triangle) MG masses of SI (SI surgery alone) and each stimulation protocol (SI-Stim) as a percent of normal control. (A) 30 day SI and SI-Stim 1 to 2. (B) 7 day SI and SI-Stim 3 to 6. (C) 6 day SI and SI-Stim 7. All SI-Stim data are significantly different from SI, except for the absolute mass of SI-Stim 1.

After 30 days of SI, the absolute and relative MG masses were 53 and 64% of Con, respectively (Fig. 2A). Daily bouts of high-frequency, short-duration stimulation delivered in a single 6-cycle session (SI-Stim 1) maintained relative MG mass to 72% of Con. The same stimulation paradigm delivered in two 3-cycle sessions with a 9-hr interval between sessions (SI-Stim 2) further ameliorated relative MG mass to 79% of Con.

After 7 days of SI, the relative MG mass was 68% of Con. Using the most effective stimulation protocol from the 30-day studies (SI-Stim 2), training for 5 days (SI-Stim 3) also had a significant impact on the relative MG mass (maintained at 92% of Con) (Fig. 2B).

Five days of long-duration, submaximal contractions (SI-Stim 4: 50 Hz, 4 sec duration) or doubling the total number of cycles (SI-Stim 5: two 6-cycle sessions) resulted in relative MG masses that were 87 and 90% of Con. Increasing the high frequency duration (SI-Stim 6: 100 Hz, 4 sec duration) maintained relative MG mass at 99% of Con. Beginning this stimulation training 1 day post surgery (SI-Stim 7, all previous protocols were initiated 2 days post surgery) preserved MG mass to 97% of Con (Fig. 2C). All stimulation protocols resulted in a total of 1 to 4 min of activity within any 24-hr period.

4. DISCUSSION AND CONCLUSIONS

In the present study, the role of activity-dependent factors on muscle mass was examined using the SI model. In the SI model of inactivity, all ascending, descending, and peripheral input to the motoneurons that innervate the hindlimb is eliminated while maintaining the integrity of the motoneuron-muscle connection. The near zero baseline level of activity in SI muscles allowed us to impose and determine the effects of known amounts and patterns of electromechanical activation.

Long-term inactivity (30 days) resulted in marked atrophy (64%) of the MG, a fast plantarflexor muscle. Daily bouts of high-load, short-duration isometric contractions ameliorated the loss in muscle mass during the 30-day period. Delivering the stimulation protocol in two sessions per day was more efficacious in maintaining muscle mass than through a single session of activation. This most likely reflects protein regulatory dynamics that require some activation at minimum intervals throughout the day to maintain homeostasis.

Inactivity-induced atrophy occurs in two phases. The first 8 to 10 days of inactivity is marked by a rapid rate of atrophy. This is followed by a slower rate of atrophy until a new steady state is reached.[4] Thus, maintaining muscle mass during the rapid atrophy phase may be critical for long-term preservation. In an effort to determine the most efficacious stimulation paradigm for maintaining muscle mass, we varied stimulation frequency (100 and 50 Hz), duration (1 and 4 sec), and number of cycles (2 sessions of 3 and 6 cycles) delivered per day during the first 7 days of inactivity. The most efficacious stimulation paradigm (100 Hz, 4 sec stimulation duration delivered in 2 sessions of 3 cycles) maintained relative MG mass at 99% of control. Therefore, as little as 4 min of electromechanical activity per day in an otherwise inactive muscle is sufficient to maintain the relative mass of a predominantly fast hindlimb muscle.

The results of this study suggest that brief periods (a total of 1 to 4 min over a 24-hr period) of patterned high-load isometric contractions is an efficacious rehabilitative strategy to reduce, and possibly eliminate, the atrophy of normally innervated fast muscle in the rat. The present study also highlights the importance of the pattern of electromechanical activity, and not only the total amount delivered, i.e., distributing the same amount of activity in two sessions/day was more effective than in one session/day. From a clinical perspective, the results demonstrate the feasibility of using the wireless implantable BION microstimulator on a very brief but daily basis over a period of weeks to counter the loss of skeletal muscle mass associated with debilitating neuromuscular conditions, such as occurs after spinal cord injury.

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