

**GISLAINE DOS SANTOS SILVA**

**EFEITO DE SESSÃO REPETIDA DE EXERCÍCIO EXCÊNTRICO  
NA EXPRESSÃO DE GENES PRÓ E ANTI-INFLAMATÓRIOS  
NO MÚSCULO ESQUELÉTICO**

Dissertação apresentada ao Programa de Pós-Graduação em Ciências Morfofuncionais do Instituto de Ciências Biomédicas da Universidade de São Paulo, para obtenção do Título de Mestre em Ciências.

Área de Concentração: Ciências Morfofuncionais

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## RESUMO

Silva GS. Efeito de sessão repetida de exercício excêntrico na expressão de genes pró e anti-inflamatórios no músculo esquelético. [Dissertação (Mestrado em Ciências Morfofuncionais)] São Paulo: Instituto de Ciências Biomédicas, Universidade de São Paulo; 2015.

Diante da importância da integridade muscular para a manutenção da qualidade de vida e para a prática de atividade física, torna-se cada vez maior o interesse na investigação de estratégias capazes de maximizar a resposta regenerativa muscular. Neste contexto, o estudo do possível efeito “protetor” gerado por sessão repetida de exercício excêntrico na musculatura esquelética ainda é pouco explorado. As contrações excêntricas causam lesão muscular por meio do desarranjo dos filamentos dos sarcômeros, o que leva à ruptura do sarcolema, aumento da concentração de  $Ca^{+2}$  intracelular com subsequente ativação de proteólise. Por outro lado, uma sessão repetida de exercício excêntrico acarreta na redução da lesão muscular com conseqüente atenuação da dor e da perda de força muscular. Esse fenômeno, uma espécie de mecanismo protetor, é conhecido como “efeito da carga repetida”. Visando contribuir para o melhor entendimento do efeito da carga repetida sobre a musculatura esquelética, em especial, a expressão de genes pró e anti-inflamatórios após o dano promovido pelo exercício excêntrico, camundongos selvagens foram submetidos a uma ou duas sessões de exercício excêntrico e os músculos sóleos foram avaliados após 24 horas e 3 dias. A análise histológica mostrou fibras musculares lesadas nos músculos sóleos de animais submetidos à 1ª sessão, com considerável atenuação no número de fibras musculares lesadas nos animais submetidos à 2ª sessão. Em geral, a expressão gênica das citocinas pró-inflamatórias (TNF- $\alpha$ , iNOS e IL-1 $\beta$ ) e anti-inflamatórias (IL-10, YM1, Fizz-1 e Arginase-1) foi alta em 24 h após a 1ª sessão, e reduziu aos níveis do grupo controle em 24 h após a 2ª sessão de exercício excêntrico. Nossos resultados sugerem que o aumento da expressão gênica dessas citocinas pró e anti-inflamatórias no tecido muscular antes da 2ª sessão pode ter contribuído para a atenuação da lesão muscular detectada em 24 h após a 2ª sessão de exercício excêntrico.

**Palavras-chave:** Músculo esquelético. Citocinas inflamatórias. Exercício excêntrico. Efeito da carga repetida. Expressão gênica.

## ABSTRACT

Silva GS. Repeated session effect of eccentric exercise in expression of pro and anti-inflammatory genes in skeletal muscle. [Masters thesis (Morphofunctional Science)] São Paulo: Instituto de Ciências Biomédicas, Universidade de São Paulo; 2015.

Considering that muscle integrity is crucial for maintenance of quality of life and for physical activity, there is an increasing interest in the investigation of strategies able to maximize the muscle regenerative process. In this context, the study of the possible protective effect generated by repeated eccentric exercise bout on skeletal muscle is still poorly explored. The eccentric contractions cause muscle injury through the disarray of sarcomere filaments, which leads to sarcolemma disruption, increase in the intracellular calcium concentration with subsequent activation of proteolysis. Moreover, one repeated eccentric exercise bout cause attenuation of muscle damage and consequent pain relief and loss of muscle strength. This phenomenon, a protective mechanism, is known as "repeated bout effect". In order to contribute to a better understanding of the repeated bout effect on skeletal muscle, in particular, the expression of pro and anti-inflammatory genes after injury caused by eccentric exercise, wild-type mice were subjected to one or two bouts of eccentric exercise and soleus muscles were evaluated after 24 hours or 3 days. Histological analysis showed damaged myofibers in soleus muscles from animals subjected to the 1<sup>st</sup> bout, with considerable attenuation in the number of injured myofibers from animals subjected to the 2<sup>nd</sup> bout. In general, the gene expression of pro-inflammatory (TNF- $\alpha$ , iNOS and IL-1 $\beta$ ) and anti-inflammatory (IL-10, YM1, Fizz-1 and Arginase-1) cytokines was high at 24h post the 1<sup>st</sup> bout, and reduced to the control group levels at 24 h after two bouts of eccentric exercise. Our results suggest that the increased gene expression of these pro and anti-inflammatory cytokines in muscle tissue before the 2<sup>nd</sup> bout might contribute to the attenuation of muscle injury detected in 24 h after the 2<sup>nd</sup> bout of eccentric exercise.

**Keywords:** Skeletal muscle. Inflammatory cytokines. Eccentric exercise. Repeated bout effect. Gene expression.

# 1 INTRODUÇÃO

## 1.1 Características gerais do músculo esquelético

O músculo esquelético é innervado e vascularizado, representando em torno de 50% da massa do corpo humano (Karagounis, Hawley, 2010; Yusuf, Brand-Saberi, 2012). Composto por fibras musculares e tecido conjuntivo (epimísio, perimísio e endomísio), o músculo esquelético atua como um arcabouço de sustentação do esqueleto, permitindo a movimentação coordenada do corpo (Bassel-Duby, Olson, 2006; Keynes, Aidley, 2001; Powers, Howley, 2005). Além disso, o músculo esquelético participa do metabolismo servindo de reserva de substratos (por exemplo, glicogênio e proteína) e auxilia na homeostase da temperatura corpórea (Donati et al., 2013; Zanou, Gailly, 2013).

Cada fibra muscular contém diversos núcleos periféricos e miofibrilas que são estruturas alongadas compostas por sarcômeros, unidades funcionais contráteis do músculo esquelético (Junqueira, Carneiro, 2008; Keynes, Aidley, 2001; Lieber, 2002; Tajsharghi, 2008). Os sarcômeros são constituídos principalmente por filamentos de actina e miosina (Clark et al., 2002; MacIntosh et al., 2006), os quais sobrepostos garantem o padrão estriado ao longo da fibra muscular esquelética.

Além das características mencionadas, a musculatura esquelética apresenta alta plasticidade frente à ausência ou à presença de estímulos (Harridge, 2007; Karagounis, Hawley, 2010; Pette, 2001), podendo atrofiar (Jackman, Kandarian, 2004), hipertrofiar (D'Antona et al., 2006; Zanou, Gailly, 2013), promover a conversão de tipos de fibras musculares (Tiidus, 2008) ou se regenerar após uma lesão (Jarvinen et al., 2005; Quintana et al., 2015). Dentre os possíveis estímulos lesivos como, por exemplo, a exposição à cardiotoxina (CTX) (Liu et al., 2014), ao cloreto de bário (Hong et al., 2012), à queimadura (Diaz et al., 2015) e às lacerações (Jarvinen et al., 2005), decidimos focar no estresse mecânico promovido pelo exercício excêntrico (Armstrong et al., 1983; Touchberry et al., 2012).

## **6 CONCLUSÃO**

Considerando que as citocinas pró e anti-inflamatórias possuem importantes funções na resposta imune e recuperação de tecidos lesados, nossos resultados sugerem que a elevação dos níveis de expressão gênica de citocinas pró e anti-inflamatórias contribuiu para a atenuação da lesão muscular detectada após a repetição de uma sessão de exercício excêntrico.

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