

Example Abstract:

The effect of protein X knockdown on muscle mass and muscle fiber size in male and female mice

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(Introduced by Josephine Blogs, University of Melbourne).

Introduction: Skeletal muscles are fundamentally important for movement, breathing, heat generation and whole-body metabolic control. As such, the loss of muscle mass has major implications for physical activity, metabolic health and quality of life. It was recently shown that protein X is required for muscle cell differentiation *in vitro* [1] and that the abundance of protein X is markedly reduced in muscles from aged mice [2]; however, the role of protein X in mature skeletal muscle remains to be determined.

Methods: All experimental procedures were approved by the Hawkins Laboratory Animal Research Ethics Committee. Under anaesthesia (2-4% isoflurane inhalation), adeno-associated viral (AAV) vectors expressing shRNA against protein X mRNA were injected into tibialis anterior muscles (or scramble control shRNA injected into the contralateral muscle) of 8 wk old female and male C57Bl/6J mice. Muscles were collected under anaesthesia (2-4% isoflurane inhalation) 4 wk post-injection, weighed and portioned for Western blotting and for sectioning for immunofluorescence staining with laminin and myosin heavy chain isoforms for muscle fibre cross-sectional area (CSA) and muscle fibre type proportion analyses, respectively.

Results: Western blot analysis revealed that protein X abundance was reduced by 79% at 4wk post-shRNA AAV injection. Reduced protein X levels were associated with a 23% reduction in basal muscle mass compared to control muscles and with a 19% reduction in muscle fibre CSA. There was change in muscle fiber type proportions and the reduction in muscle fibre CSA was greatest in type 2B fibres. There were no differences between male and female mice.

Conclusion: These data show that protein X is necessary to maintain basal skeletal muscle mass and muscle fibre size in adult mice. Given that protein X is reduced in aged muscle [2], increasing protein X may be a potential therapeutic strategy for attenuating age-related muscle atrophy.

References:

1. Johnson *et al.* (2020) Protein X regulates differentiation of mouse myogenic C2C12 cells. *J Bio Chem.* 300: 1000259-1000263.
2. Brooks *et al.* (2019) Protein X is downregulated in aged skeletal muscle. *Natural Comm.* 20: 300000-300006.

Or, if space limited:

1. Johnson *et al.* (2020) *J Bio Chem.* 300: 1000259-1000263. **2.** Brooks *et al.* (2019) *Natural Comm.* 20: 3000000-3000006.