GDNF'S EXPRESSION IN SLOW- AND FAST-TWITCH MUSCLE FIBERS ARE DEPENDENT ON EXERCISE INTENSITY

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ABSTRACT

Glial cell-line derived neurotrophic factor (GDNF) is the most potent factor for motor neuron survival and supports and maintains the neuromuscular system. GDNF's expression in skeletal muscle has been shown to be altered following exercise in as little as two weeks. Following low-intensity exercise, GDNF has been shown to increase in slow-twitch skeletal muscle, while decreasing in fast-twitch skeletal muscle. GDNF appears then to be expressed in an activity-dependant manner. **PURPOSE**: It is our aim to test whether a higher intensity exercise can alter GDNF's expression in both slow- and fast-twitch muscle fibers. **METHODS**: Male Sprague-Dawley rats (4 weeks old) were housed individually with a resistance wheel (RW, n=5), a free wheel (FW, n=4), or without a wheel (CON, n=5). The R-RUN group had 120g of resistance added to the wheel for the duration of the study. All wheel running was voluntary and continuously recorded for two weeks. **RESULTS**: GDNF protein content increased significantly in the recruited fast-twitch Plantaris (PLA) muscles following RW (6.3±1.0, p<0.05) and FW (6.8±2.1, p<0.05) running compared to controls (2.3±0.4, p<0.05). GDNF protein content also increased in the recruited slow-twitch Soleus (SOL) muscles following FW (4.1±0.9, p<0.05) running compared to controls (1.1±0.7, p<0.05). GDNF protein content and the distance traveled was positively correlated in SOL (r=.760, p<0.01, n=14), while GDNF protein content and velocity were positively correlated in PLA (r=.690, p<0.01, n=14) for all groups. **CONCLUSION**: We show that higher intensity exercise (higher velocity and shorter bouts) can increase GDNF in the recruited fast-twitch muscle fibers. We conclude that regulation of GDNF expression is activity dependent and that the intensity of the exercise can alter its expression differently in slow and fast twitch muscle fibers.

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METHODOLOGY

Subjects & Training

- 4 week-old Sprague Dawley rats
  - Randomly assigned to control, resistance wheel (RW), or free wheel (FW)

Skeletal Muscle

- Soleus (SOL; slow twitch)
- Extensor Digitorum Longus (EDL; fast twitch)
- Plantaris (PLA; fast twitch)

Visualization of GDNF & NMJ

- Antibodies raised against GDNF & alpha-bungarotoxin

Visualization of Skeletal Muscle Fibers

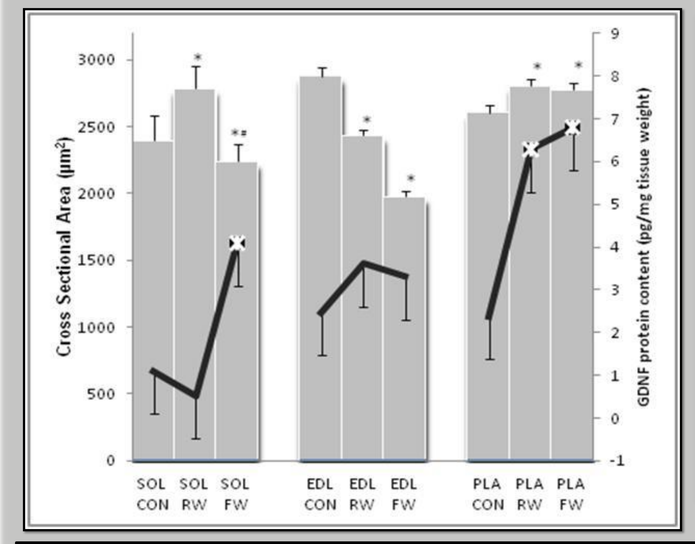
- Average Cross Sectional Area (CSA)
  - Antibodies raised against MHC (I, IIa, IIx, IIb)
  - CSA measured for 125-150 random EDL and SOL fibers (3 animals/group) using widefield microscopy

Quantification of GDNF protein content

- ELISA

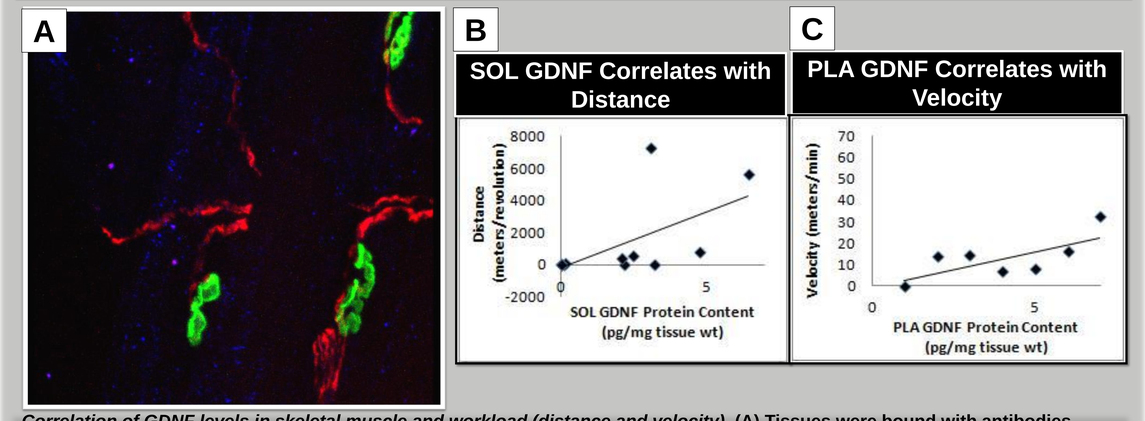
RESULTS

GDNF PROTEIN CONTENT INCREASES IN RECRUITED MUSCLES FOLLOWING HIGH INTENSITY VOLUNTARY RUNNING



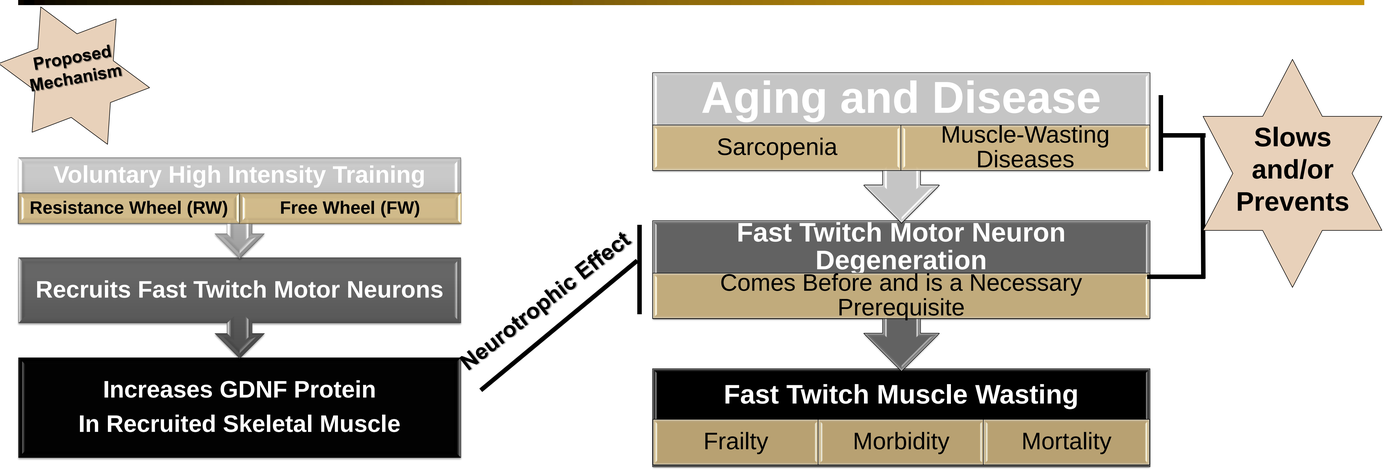
GDNF protein content increases in recruited muscles. **Quantification of GDNF was detected with ELISA. Cross sectional area (CSA) was measured in 125-150 random SOL, EDL, and PLA fibers that were captured by widefield microscopy and analyzed using ImageJ software. GDNF protein content was significantly increased in all recruited fibers, evidence by CSA positive adaptations. FW and RW exercise was able to increase the CSA of the PLA muscle as well as increase GDNF protein content. FW exercise decreased the CSA of the SOL muscle as well as increased GDNF protein content. The EDL was not recruited in FW and RW exercise and had no effect in GDNF changes.**

GDNF PROTEIN CONTENT CORRELATES WITH DISTANCE AND VELOCITY



Correlation of GDNF levels in skeletal muscle and workload (distance and velocity). **(A) Tissues were bound with antibodies raised against GDNF (blue) to visualize protein localization, alpha-bungarotoxin (green) to visualize end plates and neurofilament (red) to visualize motor neurons. (B) Levels of GDNF protein (pg/mg tissue wt.) in SOL tissues were positively correlated with distance (r=.760 P<0.01, n=14). (C) Levels of GDNF protein (pg/mg tissue wt.) in EDL tissues were positively correlated with velocity (r=.690 P<0.01, n=14). Tissues were processed for GDNF protein content using ELISA.**

INTRODUCTION AND PROPOSED MECHANISM



PURPOSE

It is our aim to test whether a higher intensity exercise such as voluntary wheel running (FW and RW) can alter GDNF's expression in both slow and fast twitch muscle fibers.

CONCLUSIONS

- Voluntary high intensity exercise altered GDNF protein content in recruited fast twitch skeletal muscle fibers
- Activity can regulate GDNF's expression differently in slow and fast twitch fibers depending on various workload factors