

# The Contribution of Titin to Striated Muscle Shortening

Joseph Schuster

Dr. Kerry McDonald Ph.D Thesis Supervisor

## Abstract

Striated muscle shortens under various loads (i.e., performs work), which allows movement of the skeleton and pumping of blood throughout the circulatory system. The amount of work that a muscle can do is determined by the speed at which it shortens against a given load. One sarcomeric protein hypothesized to assist muscle shortening speed is the giant protein titin. Titin is a 3-4 MDa protein that spans the sarcomere from Z line to M line and is thought to both bind actin in the thin filament and proteins of the thick filament. Titin contains extensible regions that are thought to provide passive restoring forces, with the size of these extensible regions being the primary difference between titin isoforms. The larger the extensible regions, the lower the passive force for a given increase in muscle length. The extension of titin, and consequent increase in passive force, may assist in muscle shortening by bearing some of the load normally carried by cross-bridges. To test this hypothesis that titin assists shortening in an isoform dependent manner three muscle types were studied: slow-twitch skeletal muscle fibers (with the largest titin isoform), fast-twitch skeletal muscle fibers (intermediate-sized titin isoform), and cardiac myocytes (smallest titin isoform). Single permeabilized skeletal muscle fibers or cardiac myocyte preparations were attached between a force transducer and a position motor and changes in muscle length were monitored during shortening against varied loads during submaximal  $\text{Ca}^{2+}$  activations. Force-velocity relationships were obtained before and after a mild trypsin treatment that has been shown to cleave titin with no apparent effects on other sarcomeric proteins. Although cleavage of titin did not affect isometric force or loaded shortening velocity in any of the three types of striated muscle, titin cleavage did alter the pattern of force re-development by markedly reducing the extent of a transient overshoot in isometric force that typically occurs after muscle re-stretch in all three muscle types. These results imply that titin strain has a negligible effect in determining striated muscle loaded shortening velocity but does appear to modulate the number of force-generating cross-bridges following muscle stretch.