Passive and active isometric stress of skinned muscle fibres from a rat model of chronic stroke

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Summary

Stroke is a leading cause of physical disability, but the extent to which post-stroke changes in skeletal muscle contribute is not known. Here we examine passive and active isometric stress of skinned fibres from paretic and non-paretic medial gastrocnemii in a rat model of chronic stroke. Our preliminary results show minimal changes in passive stress and lower active stress in fibres from paretic compared to non-paretic muscle. These early findings suggest that fibre-level changes may contribute to muscle weakness post-stroke.

Introduction

Stroke is a leading cause of physical disability worldwide [1], and much of the focus on understanding the source of post-stroke mobility deficits has been on nervous system impairments rather than potential changes in skeletal muscle. Most studies on post-stroke muscle changes focus on humans using indirect whole-muscle measures, limiting exploration of mechanisms behind functional deficits. The few studies using animal stroke models to obtain direct measures examined muscle only 2-4 weeks post-stroke [2,3], likely before muscle changes could fully progress. Therefore, the goal of this study was to examine the mechanical properties of skinned single fibres obtained from rats in the chronic phase of stroke.

Methods

We induced photothrombotic stroke in 12-week-old female Sprague-Dawley rats (n = 21) by shining a light on their intact skulls, activating an intravenously injected photoactive dye to damage the underlying brain tissue. We focused the light on the hindlimb motor cortex which resulted in deficits in the hindlimb on the side contralateral to the stroke lesion. We also induced sham stroke (n = 7) using the same procedures but without turning on the light.

We euthanized the animals and harvested their medial gastrocnemius muscles 18 weeks following surgery which corresponds to the chronic phase of stroke in rats [4]. Strips of the muscles were stored in rigor-glycerol [5] for 4 weeks to ensure sufficient skinning. For mechanical testing, we isolated single fibres in relaxing solution and attached one end to a force transducer and the other to a length controller. We set the initial fibre length to correspond with an average sarcomere length of 2.4 µm then held the fibres at this length or lengthened them to average sarcomere lengths of either 2.6, 2.8, or 3.0 µm. Fibres were held at this length until passive force plateaued, at which time they were transferred to and held in activating solution until the active force plateaued. We measured the steady-state passive and active force and calculated stress using the area approximated from the fibre diameter.

Results and Discussion

Our preliminary results from 3 animals with stroke (6 muscles, 17 fibres) showed no substantial changes in passive stress between paretic (contralateral to lesion) and non-paretic muscle (Figure 1; top). Passive stress in paretic muscle from children with cerebral palsy has been found to be higher at the fibre bundle level due to increased extracellular matrix (ECM) content and stiffness and lower at the myofibril level due to decreased titin content [6]. In our skinned fibres, higher stiffness of the ECM that is partially degraded with skinning could have balanced out a loss of titin and resulted in the minimal differences in passive stress.

We also found that active stress was lower in the paretic compared to non-paretic muscles on average (Figure 1; bottom), within each animal and each length. Loss of active force in paretic muscle is often assumed to be due to replacement of contractile tissue with fat or fibrosis [6]. Here we show that the force per unit area at the fibre level may be lower, which suggests that functional changes in remaining contractile tissue, possibly even the sarcomeres, may also contribute to weakness in paretic muscle affected by stroke.

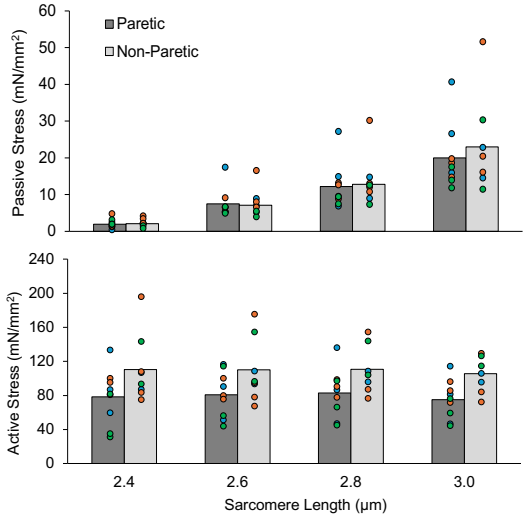

Figure 1: Passive (top) and active (bottom) fibre stress across sarcomere lengths. Bars show means of the fibre means for each muscle in each animal. Points show individual fibre values with each colour representing fibres from a different animal.

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References

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