## Effect of high-intensity intermittent exercise on the contractile properties of human type I and type II skeletal muscle fibres

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Reactive oxygen species (ROS) molecules generated within muscle fibres in both exercise and pathological conditions can greatly affect muscle function. *In vitro* studies previously showed that the alteration in the redox state of the contractile apparatus can be biphasic and dependent on both exposure time and dosage (Mollica *et al.*, 2012). The *in vivo* environment of exercising skeletal muscle is complex and little is known about the extent to which specific oxidants affect function and whether different fibre types are affected similarly. This highlights the need for *in vivo* exercise studies in order to verify the role of ROS on the modulation of the contractile apparatus following fatiguing contractions.

The present study examined the contractile apparatus properties of mechanically-skinned fibres obtained from fresh biopsies of the *vastus lateralis* muscle in seven healthy and active subjects, comprising four males and three females  $(27 \pm 8 \text{ yr})$  prior to (PRE) and following (POST) a high-intensity intermittent cycling exercise which consisted of a series of 15 s maximal efforts produced every 3 min. The maximal efforts were completed on a custom-built bike ergometer and against a constant external resistance. Between each maximal effort, participants cycled at 80 rpm and 15% of the maximal power predicted at this cadence. The PRE and POST biopsies were obtained approximately 10 min prior to and ~1 min following the exercise, respectively. All procedures were approved by the Human Research Ethics Committees at Victoria University and La Trobe University. Muscle samples were taken using a Bergstrom biopsy needle following the injection of a local anesthetic (1% lidocaine) into the skin and fascia.

We assessed whether there were any differences between the PRE and POST muscle samples in regard to specific force and  $Ca^{2+}$ -sensitivity in type I and type II fibres, and the effects of reversible oxidative modification. Individual mechanically-skinned muscle fibres were exposed to a sequence of heavily buffered solutions at progressively higher free [ $Ca^{2+}$ ] to determine their force- $Ca^{2+}$  relationship. To examine whether reversible oxidative modification of the contractile proteins occurs during exercise, some skinned fibres were treated for 5 min with 10 mM DTT, a potent reducing agent. Finally, a subset of muscle fibres were subjected to S-glutathionylation by successive treatments with 2,2'-dithiodipyridine (DTDP) and glutathione (GSH). Western blotting was used to determine fibre type.

Following the exercise, the Ca<sup>2+</sup>-sensitivity was significantly decreased in type I fibres (-0.06 pCa unit) but not in type II fibres (-0.01 pCa unit). Specific force significantly decreased after the exercise in type II fibres (-18%), but no significant alteration was found in type I fibres (+5%). Treatment with the reducing agent DTT significantly increased by ~4% specific force only in type II fibres from POST. Importantly, DTT treatment in POST type II fibres induced a 2 fold larger decrease in Ca<sup>2+</sup>-sensitivity compared to PRE, likely indicating a higher level of S-glutathionylation of fast troponin I (TnI-f) following exercise. DTT treatment had no significant effect in type I fibres. These finding suggest that the decrease of cycling power output following the exercise may have been due in part to a decrease of Ca<sup>2+</sup>-sensitivity in type I fibres and a decrease of specific force production in type II fibres. Furthermore, it appears that S-glutathionylation of TnI-f resulting from exercise-induced ROS production helps maintain the Ca<sup>2+</sup>-sensitivity of the contractile apparatus in type II fibres, with likely beneficial effects on performance.

Mollica JP, Dutka TL, Merry TL, Lamboley CR, McConell GK, McKenna MJ, Murphy RM, Lamb GD. (2012) *Journal of Physiology*, **590**: 1443-1463.