Osmotic compression improves force production in skinned muscle fibres of the rat and human

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The skinned fibre is a powerful tool for investigating skeletal muscle function. However, one possible drawback with the technique is that the skinned fibre normally swells when placed in artificial physiological solution, because high molecular weight molecules normally present in the cytoplasm are washed out, thereby allowing the myofibrils to swell appreciably (Fryer & Stephenson, 1996; Godt & Maughn, 1981). This swelling can be prevented by maintaining the normal osmotic compression by including long-chain polymers in the bathing solution. However, the importance and effects of such osmotic compression on muscle fibre function are poorly understood. In this study, mechanically skinned fibres, prepared from rat and human skeletal muscle, were osmotically compressed by addition of Dextran T500 or polyvinylpyrrolidone (PVP-40) to the bathing solution. Solutions and procedures were otherwise similar to those described previously (Lamb & Stephenson, 1994; Posterino *et al.*, 2000).

All animal experiments were approved by the La Trobe University Animal Ethics Committee. Sprague-Dawley rats were killed by overdose of isoflurane (4% vol/vol) in a glass chamber, and then *extensor digitorum longus* and *soleus* muscles were removed by dissection. All procedures on human subjects were approved by the Human Research Ethics Committees at Victoria University and La Trobe University. After injection of a local anesthetic (1% lidocaine) into the skin and fascia, a sample from the *vastus lateralis* muscle was taken using a Bergstrom biopsy needle.

In the absence of any Dextran or PVP-40, skinned fibre diameter increased ~16% upon transfer from paraffin oil to the solution, but was returned to near its *in situ* size by the presence of either 4% Dextran or 3% PVP-40. This restoration potentiated both tetanic force (~7.4% and ~11.4% in PVP-40 and in Dextran, respectively) and depolarization-induced force (~9.8% and ~5.8% in PVP-40 and in Dextran, respectively) in rat type II fibres. PVP-40 and Dextran had different effects on myofibrillar Ca^{2+} sensitivity, as indicated by the pCa_{50} ($-\log_{10}[Ca^{2+}]$ at half-maximal force). The compression by 4% Dextran increased pCa_{50} (~0.07 pCa units shift in type I and type II fibres), whereas there was no change with 3% PVP-40. In human fibres, myofibrillar Ca^{2+} sensitivity was also increased by Dextran-induced compression (~0.15 and ~0.07 pCa units shift in type I and type II fibres, respectively). Ca^{2+}-induced maximal force was slightly increased in type II fibres of the rat (~2.8% and ~4.1% in PVP-40 and in Dextran, respectively), but not in type I fibres. Human type I fibres displayed ~5.8% increase in Ca^{2+}-induced maximal force, but not type II fibres. These results show that the restoration of normal osmotic compression improves myofibril function and/or increases Ca^{2+} release from the sarcoplasmic reticulum, resulting in significant potentiation of force production.

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