Action potential mediated Ca^{2+} release in mechanically skinned fast-twitch muscle fibres of the rat is reduced by low [ATP] and by elevated [Mg^{2+}]

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The cytoplasmic [ATP] in human type IIX fibres has been shown to decline from 8 mM to ~1 mM during a 25 s bout of maximal cycling exercise[1], with total power output decreasing ~50%. Using a recently developed TBQ-BAPTA assay[2], we specifically examined whether action potential (AP)-mediated Ca^{2+} was reduced by low [ATP] and by raised free [Mg^{2+}] in the cytoplasm, to determine if this could cause reduced force output.

Long-Evans hooded rats were killed under deep anaesthesia (2% v:v fluothane), EDL muscles excised, single fibres mechanically skinned, connected to a force transducer and electrically stimulated (2 ms, 75 V cm^{-1} pulse) to produce twitch or tetanic (50 Hz) force responses. The K^{+}-HDTA bathing solution (containing 1 mM free Mg^{2+} and 8 mM total ATP) was altered appropriately (e.g. adding various [BAPTA]-50 m M TBQ and/or raised [Mg^{2+}], lowered [ATP] or adenosine).

TBQ-BAPTA assays revealed that AP-mediated Ca^{2+} release was significantly ($P<0.05$) reduced when: 1) [ATP] was lowered to 1 or 0.5 mM (86±4%; n=6, and 80±2%; n=21, control levels respectively), 2) free [Mg^{2+}] was raised to 3 mM (62±4%; n=4), and 3) adenosine (4 mM) and 1 mM ATP was present (54±4%; n=6).

These data suggest that: 1) ATP must be bound to the stimulatory site on ryanodine receptors for the dihydropyridine receptors (DHPR) to potently trigger Ca^{2+} release, 2) elevated free [Mg^{2+}] reduces DHPR-mediated Ca^{2+} release, and 3) weak ATP agonists such as AMP, ADP and adenosine exacerbate the reduction in Ca^{2+} release. Thus, these factors may underlie the reduction in Ca^{2+} release occurring during fatigue in type IIX fibres. Terminating Ca^{2+} release may help prevent complete exhaustion of ATP and the cellular damage that would ensue.

(1) Karatzaferi, C., de Haan, A., Ferguson, R.A., van Mechelen, W. & Sargent, A.J.(2001) Pflügers Archiv 442: 467-474.
(2) Posterino, G.S. & Lamb, G.D. (2003) Journal of Physiology 551.1: 219-237.