SPECIFIC FORCE OF THE RAT EXTRAOCULAR MUSCLES, LEVATOR AND SUPERIOR RECTUS, MEASURED IN SITU

Gordon S. Lynch, Paul Gregorevic, David A. Williams and Bartley R. Frueh*, Department of Physiology, The University of Melbourne, Victoria, 3010, Australia

Extraocular muscles (EOM) are characterised by their faster rates of contraction and their higher resistance to fatigue relative to limb skeletal muscles. One of the most perplexing issues in muscle physiology is why EOM generate significantly lower specific forces \([sP_o, \text{force per muscle cross-sectional area (CSA), kN/m}^2]\) than skeletal muscles. In previous studies the examination of EOM contractility was generally performed on isolated muscles, *in vitro*. It is possible that during the intricate dissections required for *in vitro* investigation that surgical trauma results in damage directly to the EOM and this contributes to the disparity in \(sP_o\) values between EOM and skeletal muscles. In this study, we have re-examined the issue of whether EOM produce lower \(sP_o\) than skeletal muscles. Specifically, we have investigated the force producing capacity of the levator palpebrae superioris (levator) and superior rectus muscles, from the rat, *in situ*. We compared the values for absolute force \((P_o)\) and \(sP_o\) with those for muscles studied *in vitro*. We tested the null hypothesis, that the \(sP_o\) for EOM obtained *in situ* would not be different from that of limb skeletal muscles. A corollary to our primary hypothesis was that \(P_o\) and \(sP_o\) for EOM obtained *in situ* and *in vitro* would not be different.

For the evaluation of EOM function *in situ*, Sprague-Dawley rats (250-450 g) were anaesthetised deeply with sodium pentobarbitone of \((60-80 \text{ mg kg}^{-1}, \text{i.p.})\) such that they did not respond to tactile stimuli throughout the procedures. During the intricate dissection procedures, nerve and blood supply to either the levator or the superior rectus muscle remained intact. The EOM were attached to a force transducer and the cranial nerves exposed for direct stimulation. After determination of optimal muscle length \((L_o)\) and stimulation voltage, a full frequency-force relationship was established for each muscle. In separate experiments, the levator and superior rectus muscles were ‘excised for evaluation of isometric contractile function *in vitro*, using methods described in detail elsewhere (Lynch et al., 2000). Animals were killed by cardiac excision whilst still anaesthetised.

Maximum \(P_o\) for the levator and superior rectus muscles was \(177 \pm 13 \text{ mN}\) and \(280 \pm 10 \text{ mN}\), respectively. For the calculation of specific force, a number of rat levator and superior rectus muscles were partially digested in a 20% nitric acid-based solution in order to isolate individual muscle fibres. Muscle fibre lengths \((L_f)\) were expressed as a percentage of overall muscle length, allowing a mean \(L_f\) to \(L_o\) ratio to be used in the estimation of muscle CSA. Mean \(L_f/L_o\) was determined to be 0.38 for the levator muscle and 0.45 for the superior rectus muscle. The \(sP_o\) for the rat levator and superior rectus muscles measured in situ, was \(275 \text{ kN/m}^2\) and \(280 \text{ kN/m}^2\), respectively. These values are within the range of \(sP_o\) values commonly reported for rat skeletal muscles. Furthermore, \(P_o\) and \(sP_o\) for the levator and superior rectus muscles measured *in situ* were significantly higher \((P < 0.001)\) than \(P_o\) and \(sP_o\) for these muscles measured *in vitro*.

The results indicate that the force output of intact EOM differs greatly depending on the mode of testing. *In situ* evaluation yields higher forces such that \(sP_o\) values are similar to those for limb muscles. Most skeletal muscles develop similar forces *in situ* and *in vitro*, whereas EOM generate far less force in all studies performed *in vitro*. Although *in vitro* evaluation of EOM contractility will continue to reveal important information about this group of understudied muscles, the lower \(sP_o\) values of these preparations should be recognised as being significantly less than their true potential. We conclude that EOM are not intrinsically weaker than skeletal muscles.


W.K. Kellog Eye Center, Department of Ophthalmology, The University of Michigan, Ann Arbor MI 48109-0714, USA