Detection of microvascular flowmotion within skeletal muscle *in vivo*

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Blood flow in resting muscle undergoes rhythmical variation (flowmotion) such that at any one time only a proportion of the capillaries that supply nutrients to the muscle myocytes are perfused. Alterations to this rhythmical blood flow can lead to an overall increase or decrease in nutrient supply to muscle by altering the number of capillaries that are perfused at any one time depending upon the contribution of the differing frequencies that occur. Flowmotion within skeletal muscle *in vivo* is difficult to determine and has depended upon invasive studies with implanted Laser Doppler flow (LDF) probes. This study reports the novel use of real-time contrast-enhanced ultrasound (RT-CEU) to determine flowmotion in rat muscle *in vivo*.

Anaesthetized rats were infused with phospholipid microbubbles and the microvascular flow monitored by RT-CEU with a Philips iU22 ultrasound machine using a L9-3 transducer on the upper thigh. Analysis of regions of interest were performed by QLab (Philips) and wavelet analysis (Matlab) was used to determine the contribution by endothelial (0.006-0.02Hz), neurogenic (0.02-0.06Hz) and myogenic (0.06-0.3Hz) vasomotion. RT-CEU data were compared to simultaneous LDF data collected from subcutaneous tissue with surface probes (VP1-HP, Moor Instruments) and muscle tissue with implantable probes (MP4s, Moor Instruments).

Both RT-CEU and LDF data demonstrated that muscle and subcutaneous tissues displayed flowmotion with a strong neurogenic rhythmical variation. Phentolamine treatment abolished the neurogenic flowmotion and lead to a recruitment of microvascular flow to the same extent as muscle contraction. RT-CEU provides a new minimally invasive technique that can be used to determine *in vivo* flowmotion within skeletal muscle of experimental animals and humans. This technique can provide novel information about defects in the vascular regulation in muscle during pathological conditions associated with poor nutrient delivery such as insulin resistance.