Royal Veterinary College
University of London

Protocol for precipitation of samples for Pyruvate and Lactate Ratios

For each sample (i.e. for pre-exercise and post-exercise), prepare 2 tubes each with 500 µl of ice cold 0.46 mol/L perchloric acid, keep cold at the patient's bedside on an ice pack. Collect blood into a lithium heparin tube and IMMEDIATELY pipette 100 µl of the blood sample into each of the perchloric acid tubes. Mix vigorously, transport to your laboratory on the ice pack. Centrifuge within 10 minutes at 4°C, 3000 rpm for 5 minutes. Remove the supernatant (top layer) and freeze supernatant in separate tubes. Store frozen and transport to the Neuromuscular Lab frozen (on dry ice). Please notify us of any shipment. Any delay in sample precipitation will result in rapid deterioration of the analyte level. Our method requires that the proportion and concentration of perchloric acid is strictly adhered to in order to produce reliable results. Manufacturers supply perchloric acid at a variety of strengths. Please prepare the working perchloric acid as specified below:

Stock perchloric acid Supplied by manufacturer	Preparation of 0.46 mol/L perchloric acid
60 % w/w (SG 1.54)	2.50 ml stock made up to 50 ml with distilled water
70 % w/w (SG 1.70)	1.94 ml stock made up to 50 ml with distilled water

Keep the working reagent in a plastic bottle at 4 °C.

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