Supporting information for:

Mass spectrometry imaging rapidly identifies ischemic injury in renal tissue.

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Figure S1

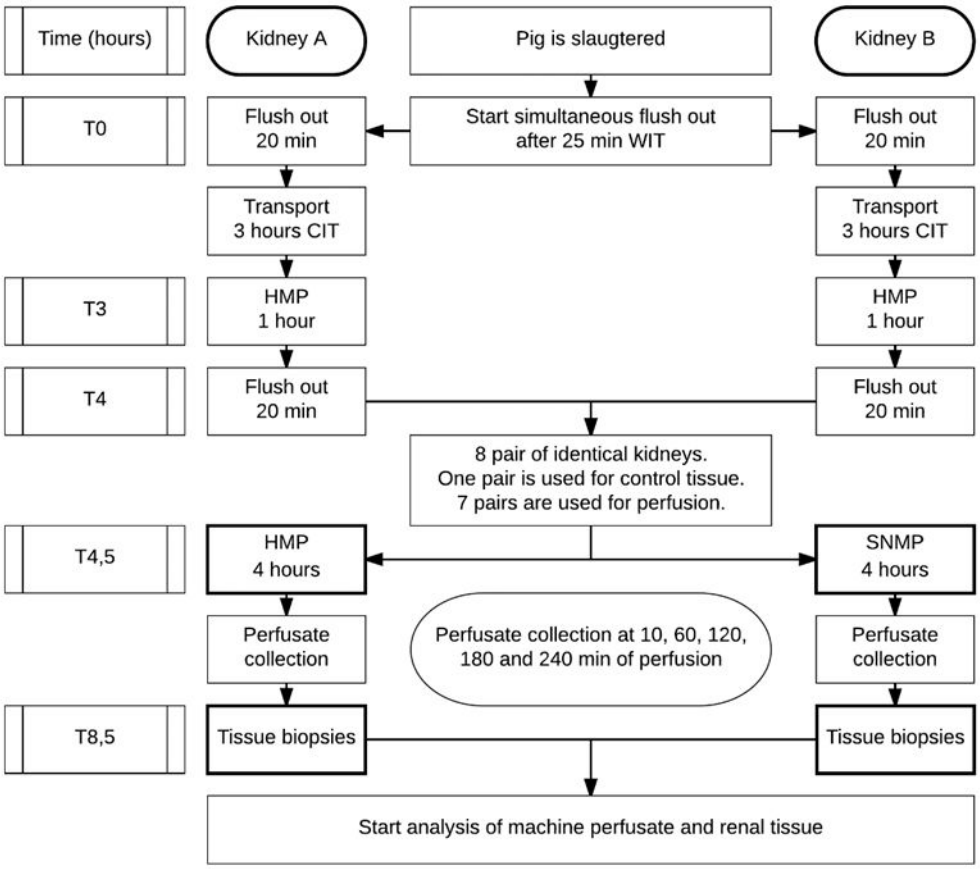
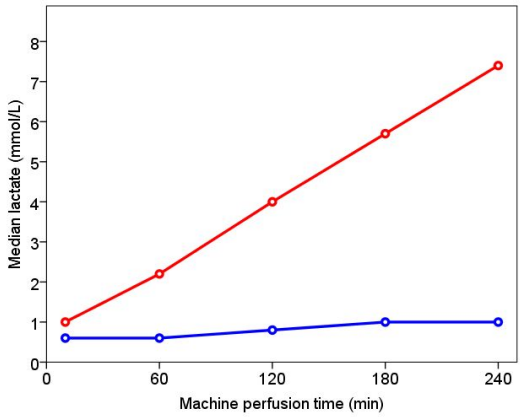
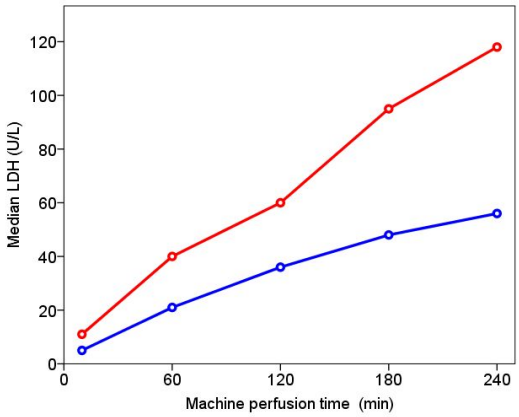


Figure S1: Vertical time-line in hours of the study set-up using 8 pigs. A pig is slaughter and its kidneys randomly assigned in either of two groups (kidney A or B). Twenty five minutes after death kidneys are simultaneously flushed using cold solutions and put in cold storage. After 3 hours of cold ischemia time, simultaneous HMP is started for 1 hour after which another flush out follows. One pair of kidneys is directly biopsied, serving as control tissue. The remaining 7 pair of kidneys are assigned in a HMP (cold ischemia) and SNMP (warm ischemia) group for a subsequent 4 hours of perfusion, in which perfusate is sampled. After perfusion, biopsies are taken and analysis of machine perfusate (lactate and LDH) and renal tissue (histopathologic examination and MALDI-MSI) is performed.

Figure S2





1a 1b

Figure S2: Renal flush out of lactate and LDH during machine perfusion. Red lines resemble flush out of SNMP kidneys with warm ischemia, the blue line flush out in HMP kidneys with cold ischemia. 1a: After four hours there is a significant difference in median lactate concentration between groups (median lactate: HMP vs. SNMP = 1.0 (IQR, 0.9-1.1) vs. 7.4 (IQR, 5.2-9.0), P=.018). 1b: After four hours there is a significance difference in LDH concentration between groups (median LDH: HMP vs. SNMP = 56 (IQR, 41-62) vs. 118 (IQR, 61-130), P=.028).

Figure S3

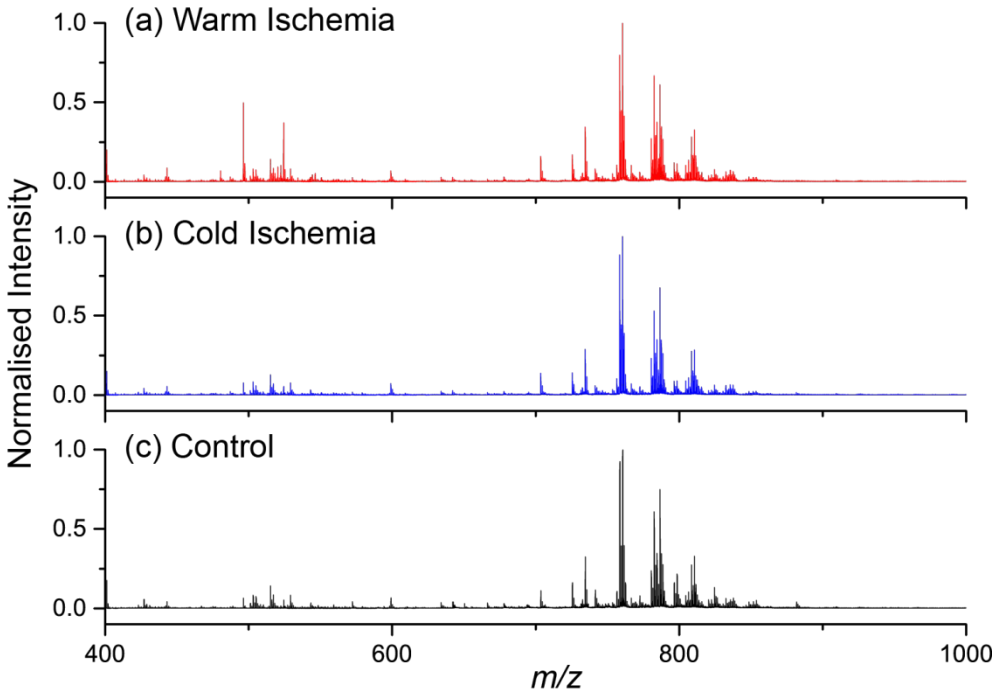


Figure S3: Average positive ion spectra from warm ischemia (a), cold ischemia (b) and control (c) tissues.

Figure S4

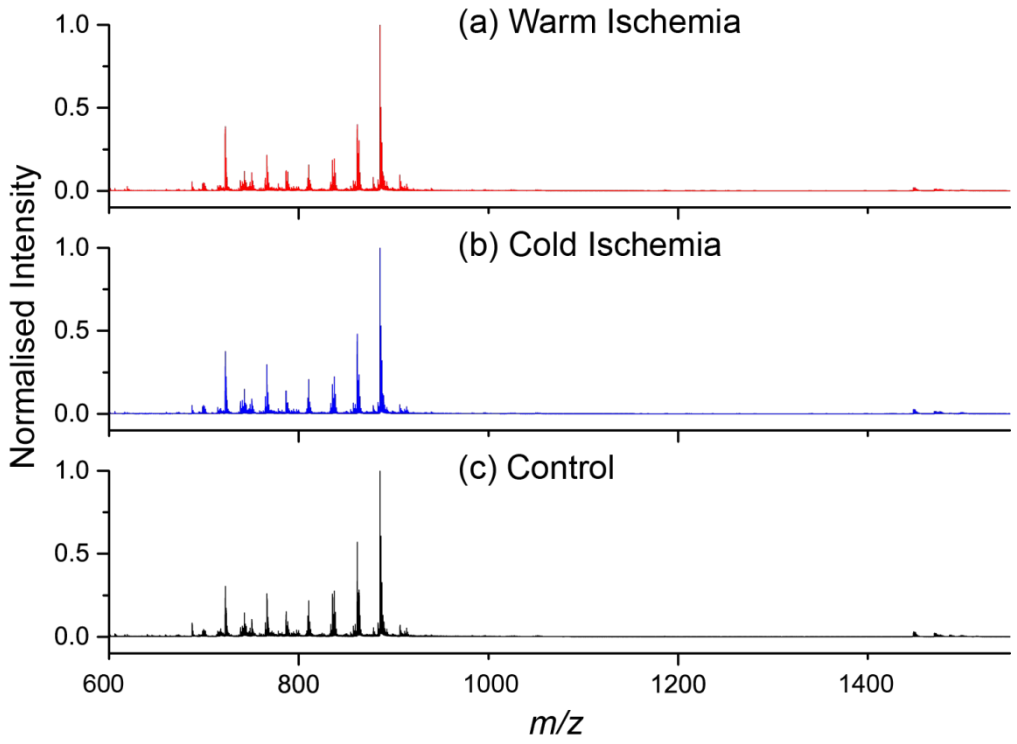


Figure S4: Average negative ion spectra from warm ischemia (a), cold ischemia (b) and control (c) tissues.

Figure S5

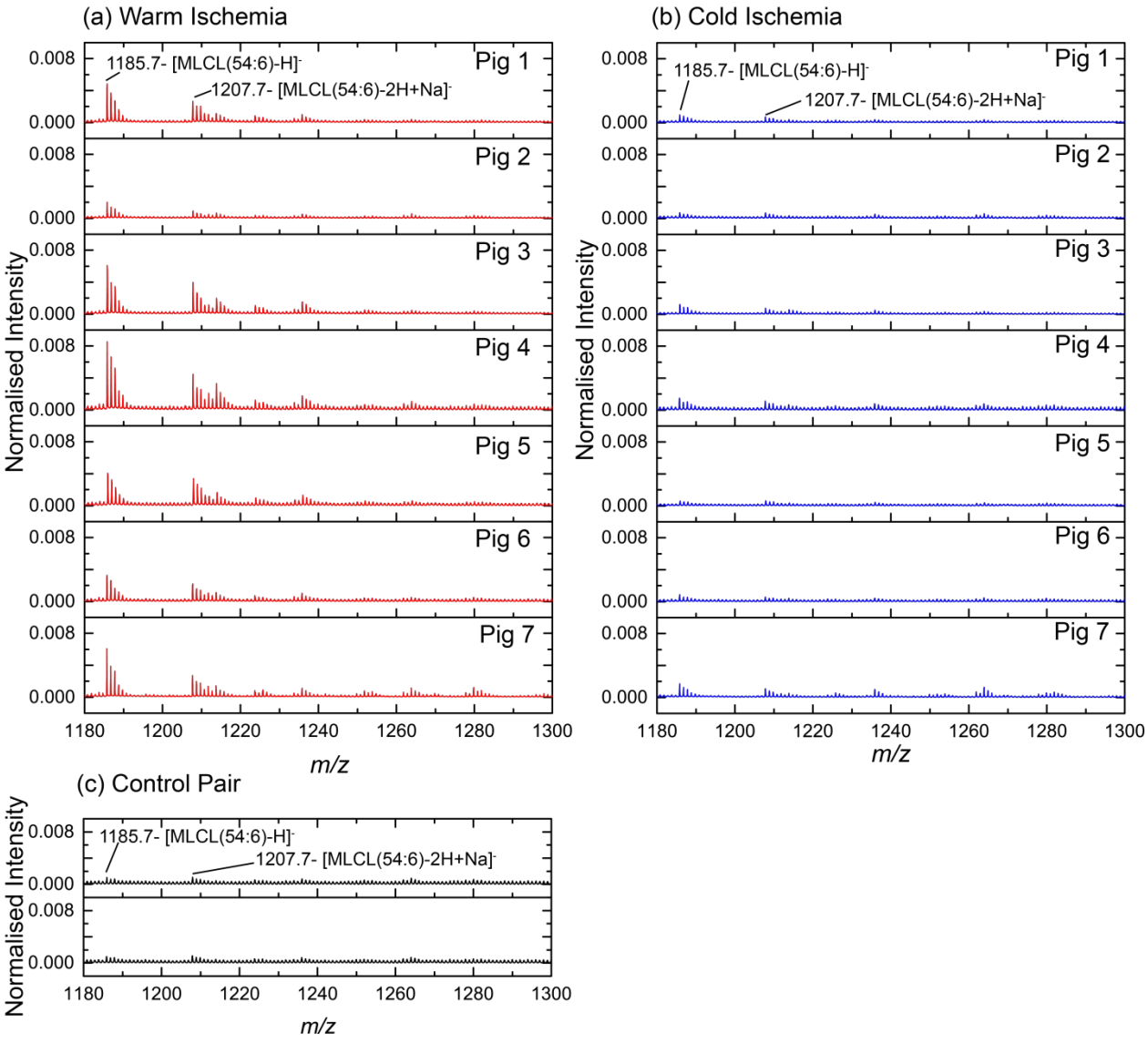


Figure S5: Average negative ion MALDI mass spectra acquired from each individual warm ischemia (a), cold ischemia (b) and control (c) tissue in the *m/z* range 1180-1300. Data show the elevation of MLCL(54:6)-related ion signals in the case of warm ischemia.

Figure S6

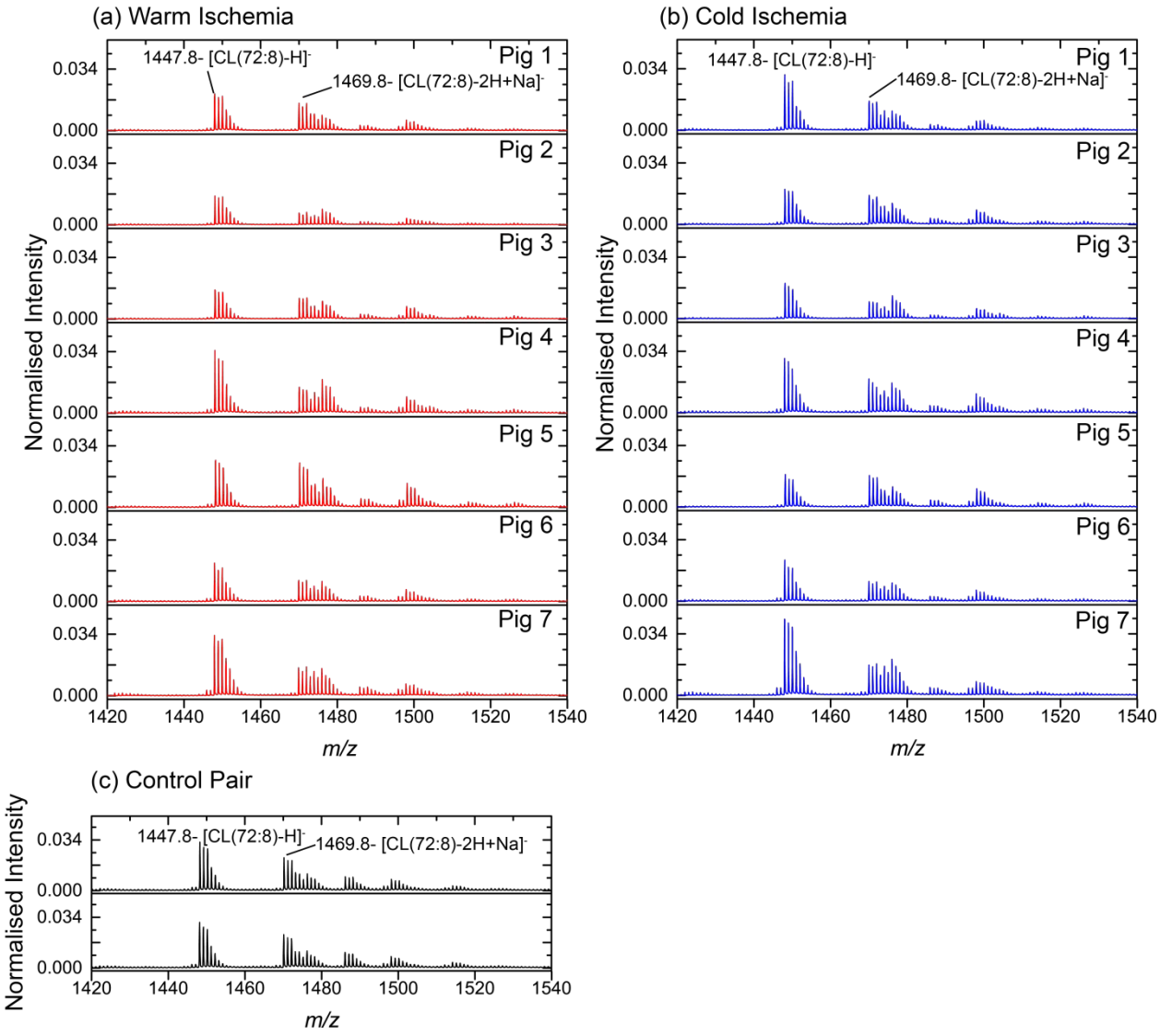


Figure S6: Average negative ion MALDI mass spectra acquired from each individual warm ischemia (a), cold ischemia (b) and control (c) tissue in the *m/z* range 1420-1540. Data show the signal intensity of cardiolipins-related signals (mostly originating from CL(72:8)) across each tissue.

Figure S7

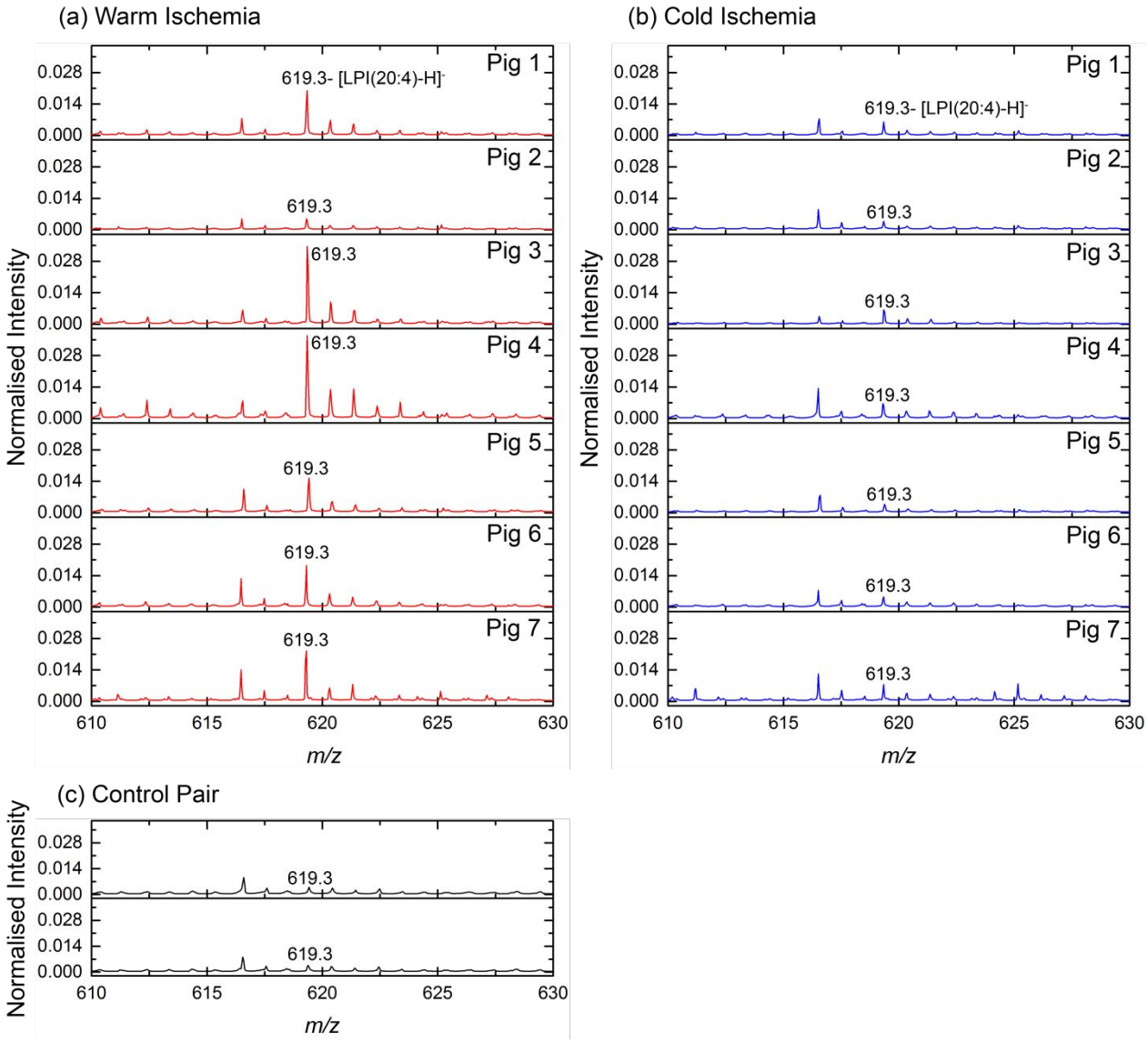


Figure S7: Average negative ion MALDI mass spectra acquired from each individual warm ischemia (a), cold ischemia (b) and control (c) tissue in the *m/z* range 610-630. Data show the elevation of [LPI(20:4)-H]$^-$ signal in the case of warm ischemia.

Figure S8

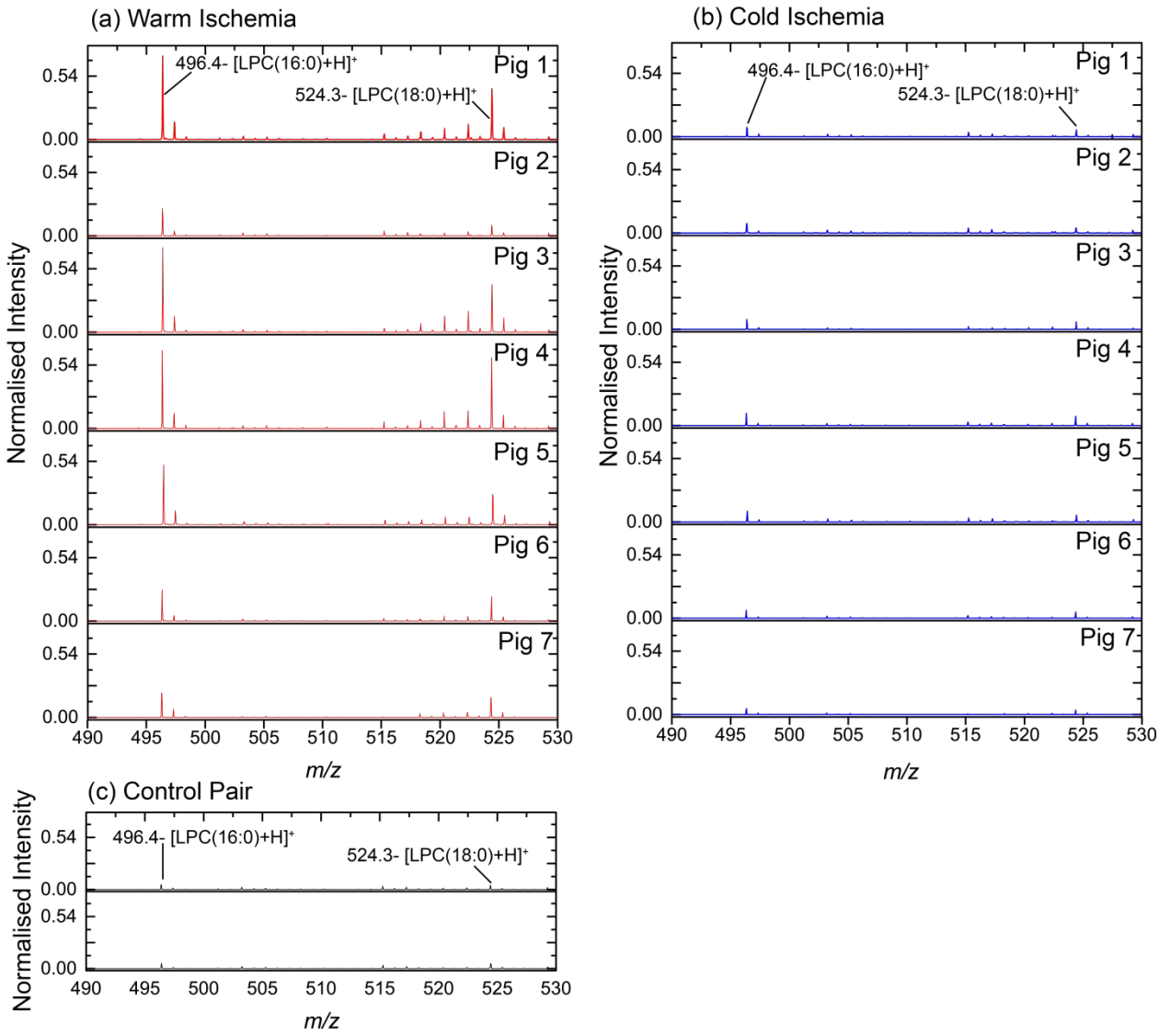


Figure S8: Average positive ion MALDI mass spectra acquired from each individual warm ischemia (a), cold ischemia (b) and control (c) tissue in the *m/z* range 490-530. Data show the elevation of lysophosphatidylcholine signals in the case of warm ischemia.