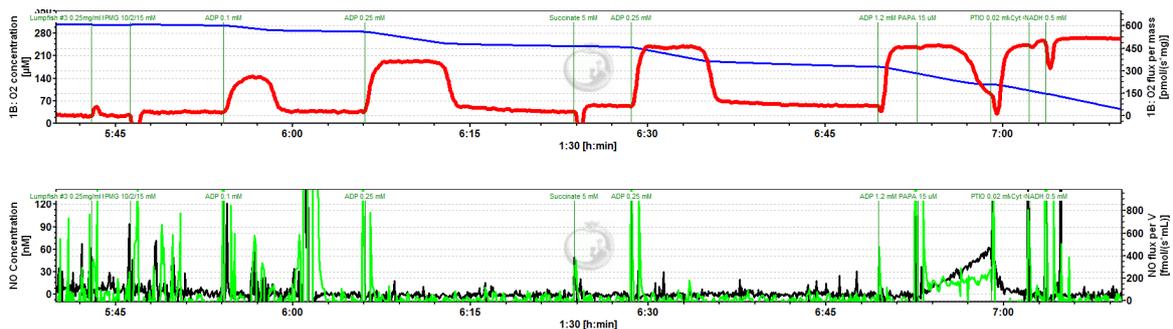


Supplementary Figure S1

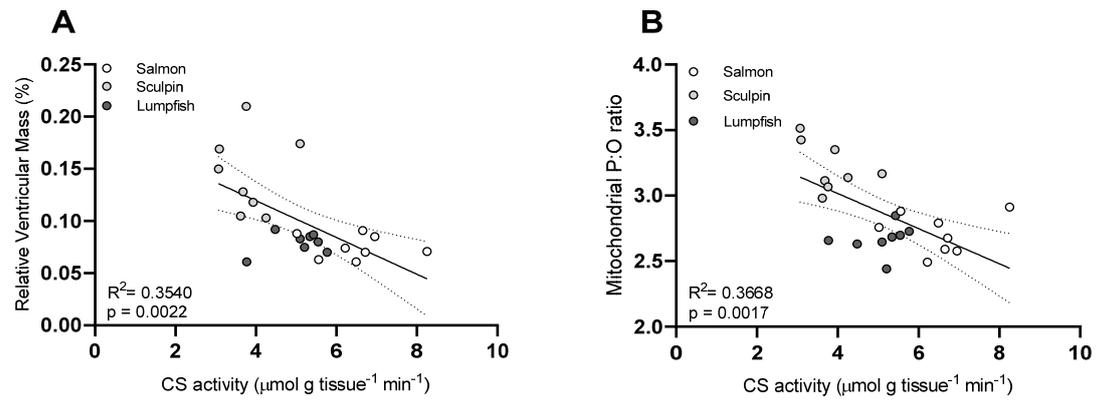
Supplementary Figure S1. Representative recording of Experiment 1. The figure shows the simultaneous measurement of O₂ consumption (flux) and ROS release rate (flux) during OXPHOS (State 3) and leak (State 4) respiration in the presence of substrates of complex-I alone (pyruvate, malate and glutamate) and complexes-I+II (+ succinate). O₂ concentration (in μM) is represented by the blue line, O₂ flux [i.e., mitochondrial respiration rate (in pmol O₂ (s*mg)⁻¹)] by the red line, H₂O₂ concentration (in μM) by the black line and H₂O₂ flux [i.e., mitochondrial ROS release rate (in pmol H₂O₂ (s*mg)⁻¹)] by the green line. Events and concentrations of chemical used are labeled on the recording. Figure shows data from cardiac mitochondria (0.25 mg mL⁻¹ chamber) of lumpfish incubated at 12°C.

Supplementary Figure S2



Supplementary Figure S2. Representative recording of Experiment 2. The figure shows the measurement of mitochondrial respiration during OXPHOS (State 3) in the presence of substrates of complexes-I+II (pyruvate, malate, glutamate and succinate, PMGS) and excess ADP; following the initiation of NO production (using the NO donor PAPANONOate); after PTIO was added to the chamber to scavenge NO and re-establish OXPHOS respiration; and finally after cytochrome c and NADH were added to examine mitochondrial integrity. This protocol was used to determine the sensitivity of mitochondrial respiration to NO (i.e., its IC₅₀). O₂ concentration (in μM) is represented by the blue line, O₂ flux [i.e., mitochondrial respiration rate (in pmol O₂ (s*mg)⁻¹)] by the red line, NO concentration (in μM) by the black line and NO flux (in fmol NO (s*mg)⁻¹) by the green line. Events and concentrations of chemical used are labeled on the recording. Figure shows data from cardiac mitochondria (0.25 mg mL⁻¹ chamber) of lumpfish incubated at 12°C.

Supplementary Figure 3



Supplementary Figure S3. Relationships between ventricular citrate synthase (CS) activity, and relative ventricular mass (RVM) and mitochondrial efficiency (P:O ratio), of Atlantic salmon, short-horned sculpin and lumpfish. The solid line represents the overall linear pairwise regression and the dotted lines represent the 95% confidence limits of the fitted line. Symbols represent individual paired values. Values are means \pm s.e.m. (N=8 per group). A $P < 0.05$ was considered significant.