Metabolic Demand of Fast Rhythms in Isolated Working Hearts and Langendorff Perfused Hearts

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Background

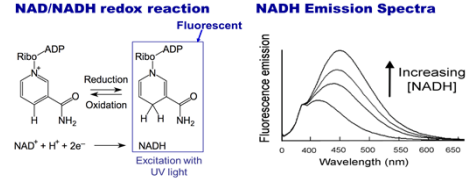
Accurate metabolic studies of the heart require that the heart perform work within the context of preload and afterload pressures, a feature unique to bi-ventricular (bi-V) working heart preparations[1,2]. The objective of this study was to compare differences in the metabolic demand of fast rhythms in isolated bi-V working hearts and non-working Langendorff[3] perfused hearts. Hearts from New Zealand white rabbits were connected to a bi-V working heart system and perfused with modified Krebs-Henseleit solution[4] at 37°C. Preload and afterload pressures were set at physiological values. An epicardial monophasic action potential electrode was used to monitor electrical activity while hearts were paced at cycle lengths of 300, 200, and 150ms. Fluorescence of NADH (fNADH) was imaged to monitor the redox state of epicardial tissue.

Understanding metabolic differences will aid in isolated heart studies of arrhythmias caused by ischemia and reperfusion.

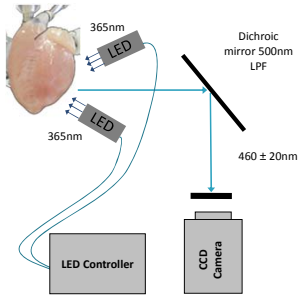
Indication of metabolic state using fluorescence

NADH is the reduced form of nicotinamide adenine dinucleotide (NAD^+). NAD^+ is a co-enzyme that carries electrons from one reaction to another in the electron transport chain. In absence of oxygen in the cells, NADH accumulates in the mitochondria.

NAD/NADH redox reaction **NADH Emission Spectra**



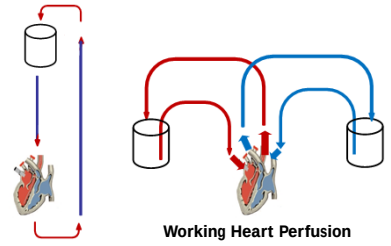
Methods for fNADH Imaging



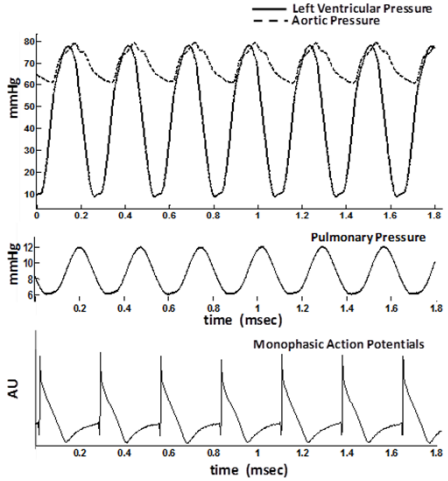
Imaging components include a CCD camera (80% quantum efficiency at 460nm), dichroic mirror (500nm, Chroma 500dclp), 365nm LEDs, and emission filter (460±20nm, Chroma ET460/40).

To image fNADH, the epicardium was illuminated with dual 365nm LED sources. Emitted light was passed through a 500nm low-pass filter and then band-pass filtered (460±20nm) before acquisition with a CCD camera (2 fps). Heart perfusion was then switched to non-working Langendorff mode and the pacing and imaging protocol was repeated. Changes in fNADH per unit time were measured and compared using N-way ANOVA tests.

Heart Perfusion Systems

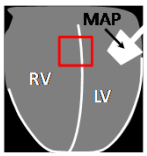


The system provides for both Langendorff and working heart perfusion. Three pumps are able to supply flow to the heart and four chambers establish preload and afterload pressures.

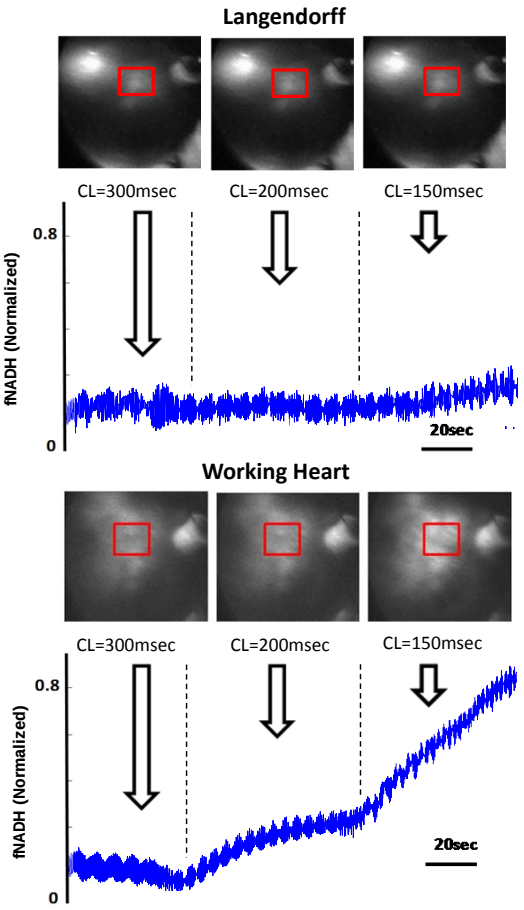


Top: Left ventricular pressure (solid line) and aortic pressure (dotted line). **Middle:** Pulmonary pressure. **Bottom:** Representative monophasic action potentials. The signal is aligned with pressures shown in top and middle panels.

Results : NADH Kinetics

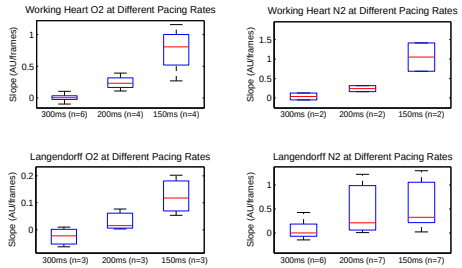


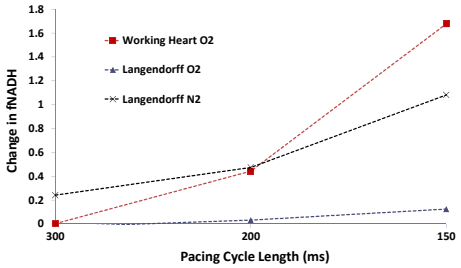
Ventricles are stimulated to contract at 3 different rates (300, 200, and 150 msec). A monophasic action potential (MAP) electrode is placed on the anterior LV epicardium to confirm activation rate and to measure electrical activity.



Top: A cartoon of the field of view (left) and three fNADH images are shown. The region of interest for the fNADH signal in the bottom panel is indicated by the red box. The tip of the monophasic action potential (MAP) electrode is seen to the right of the region of interest. **Bottom:** Average fNADH for the region of interest indicated by the red box in the top panel.

Results : Statistics





N-way ANOVA

fNADH increased:	p-value
During shorter CLs for both perfusion modes	0.001
Up to 5x higher at CL=150ms vs. other CLs	0.05
Higher and faster in working heart vs. Langendorff setups	0.007

Conclusions

Fast rhythms elevate the redox state and, concomitantly, metabolic demand in both bi-ventricular working and Langendorff heart preparations. However, elevations are much more rapid for working heart preparations, indicating that the time course of electrical alterations during acute ischemia and reperfusion could be very different between working and non-working heart studies.

References

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3. Langendorff O. Untersuchungen am uberlebenden Saugethierherzen [Investigations on the surviving mammalian heart]. *Arch Gesante Physiol*. 1895; 61: 291-332.

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Acknowledgements

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