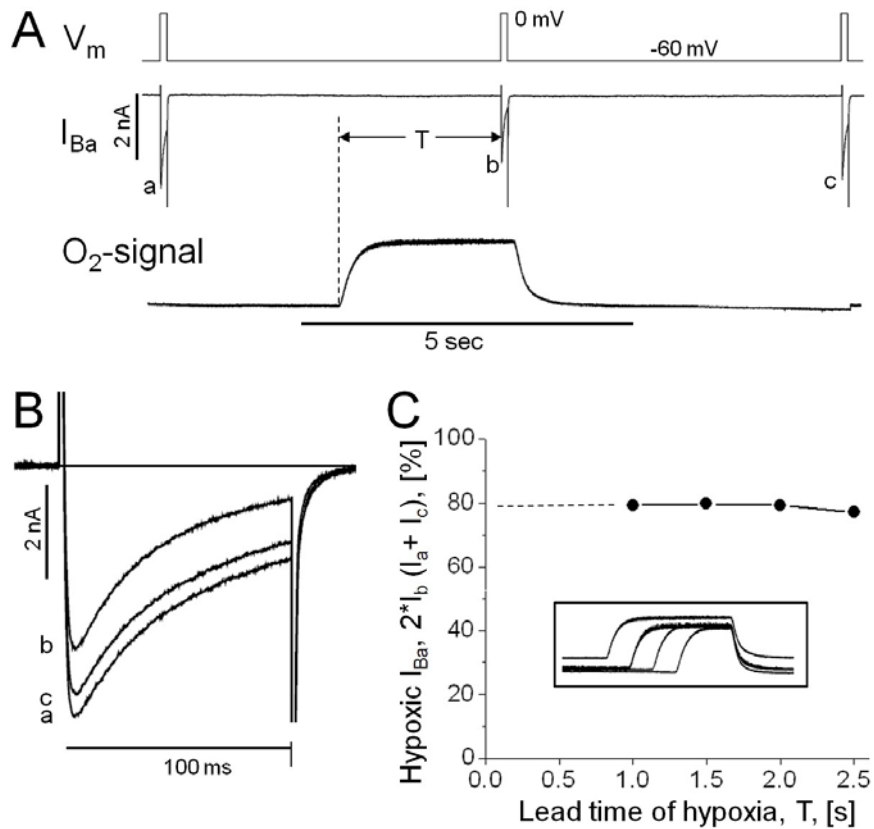


SUPPLEMENTAL DATA

Hypoxic regulation of cardiac Ca^{2+} channel: possible role of heme-oxygenase.

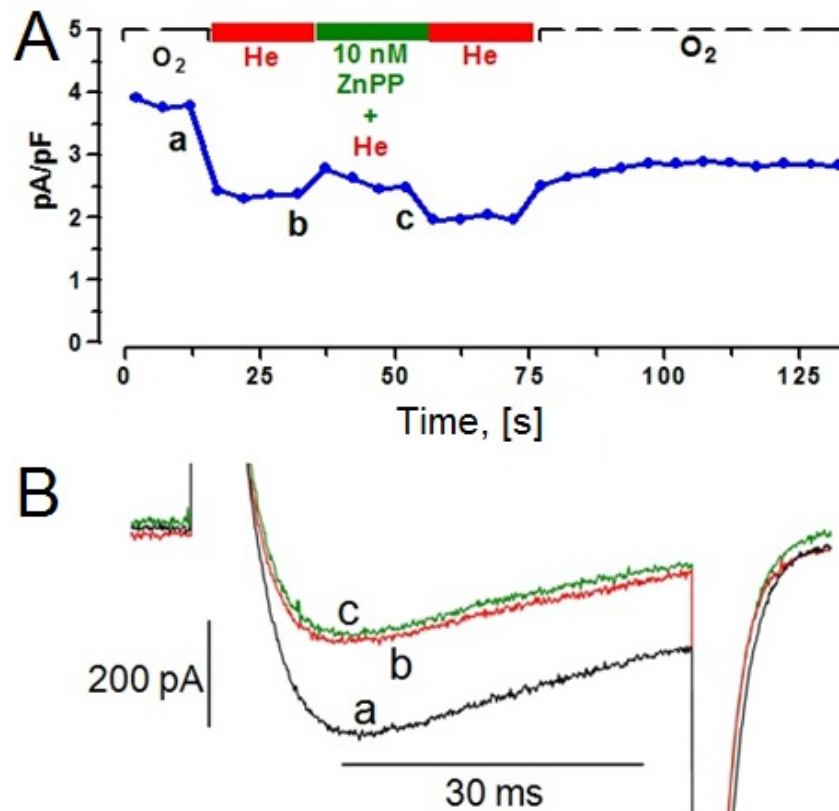
by

Angelo O. Rosa, Shahrzad Movafagh, Lars Cleemann, **and** Martin Morad



Supplemental Figure 1. Simultaneous measurements of I_{Ba} and O_2 signal in a voltage-clamped rat ventricular cardiomyocyte. A: Recording of I_{Ba} and O_2 -signal during 3 consecutive voltage-clamp depolarizations from -60 to 0 mV. The oxygen tension in the immediate vicinity of the voltage-clamped cells was measured by plating the cells onto cover slips that have been coated with a thin layer containing a ruthenium based O_2 sensor imbedded in a matrix of organic silicates (Xiong *et al.*, 2010). The ruthenium compound was excited with a 470 nm diode laser using the cover slip as a light guide and the O_2 -dependent fluorescence was measured by collecting light (>510 nm) originating from the immediate vicinity of the voltage-clamped cell. The recorded O_2 -signals was generated by

actuating electromagnetic valves that switched to a hypoxic solution with a lead time of 2.5 sec (T) before the second depolarizing pulse and switching back to a fully oxygenated solution 200 ms after the termination of this pulse. The Ba^{2+} currents recorded before (a), during (b), and 5 sec after (c) the brief period of hypoxia are shown on an expanded time scale in panel B. Panel C shows that I_{Ba} was suppressed ~21% with hypoxic lead times ranging from 1 to 2.5 sec. The inset shows O_2 signals corresponding to lead times (T) of 2.5, 2, 1.5 and 1 sec. The I_{Ba} in hypoxic solution was normalized relative to the values measured before and after ($2 \cdot I_b / I_a + I_c$).



Supplemental Figure 2. The heme oxygenase inhibitor ZnPP had little additional effect on I_{Ba} after it had already been suppressed by hypoxia. The hypoxia was introduced by switching from superfusing solution that was equilibrated O_2 to one that was equilibrated with He. A: Time course of the peak value of I_{Ba} measured every 5 sec in a voltage-clamped rat ventricular cardiomyocyte. B: Sample tracings of I_{Ba} measured at the indicated times (a, b, and c).

Reference

Xiong Y, Zhu D, Chen S, Peng H & Guan Y. (2010). A fiber-optic evanescent wave O_2 sensor based on Ru(II)-doped fluorinated ORMOSILs. *J Fluoresc* **20**, 269-274.