Facilitating donation after circulatory death in heart transplantation: effects of cardioprotective reperfusion strategies on mitochondria

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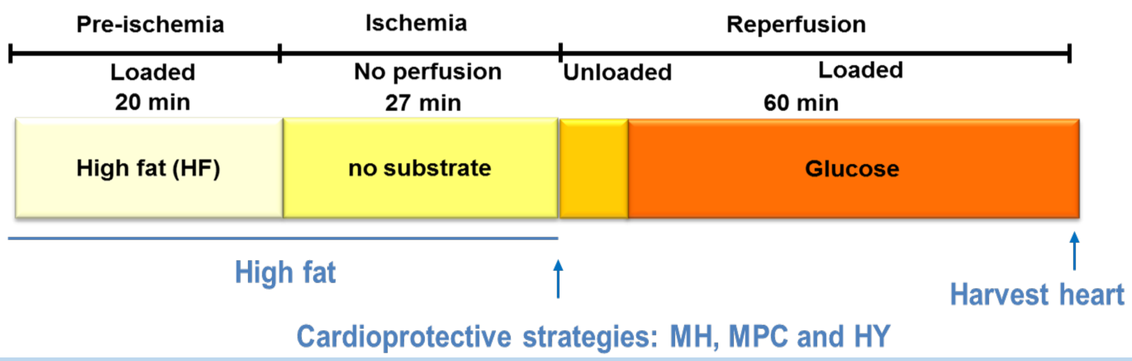
BACKGROUND AND AIM

Donation after circulatory death (DCD) could aid in reducing the shortage of donor hearts. However, concerns persists regarding donor heart integrity following warm ischemia after the obligatory circulatory arrest. Our laboratory has recently identified three reperfusion strategies that improve hemodynamic recovery of ischemic hearts: mild hypothermia (**MH**), mechanical post-conditioning (**MPC)** and hypoxia (**HY)**[1]. Considering that mitochondria are key players in ischemia/ reperfusion injury, we hypothesized that mitochondrial changes may underlie the mechanism of action of these cardioprotective strategies.

METHODS AND PROTOCOL

Experiments were performed using an isolated, working rat heart model of DCD. Hearts of male rats were submitted to 20 min basal perfusion, 27 min global ischemia (37°C) and 60 min reperfusion. No intervention hearts (**Control**; n=8) were compared to hearts in which strategies were applied at the beginning of reperfusion: **MH**= 30°C, 10 min (n=8); **MPC**= 2x30 seconds ischemia/reperfusion (n=8); **HY**= no O_2, 2 min (n=7). An additional group of hearts, not submitted to global ischemia, but perfused for 104 min (**Sham**, n=8) was included. Ventricular tissue was stored at -80°C at the end of the perfusion, and subsequent analysis of mitochondrial function and mass parameters, as well as oxidative stress were performed.



RESULTS

1 Reperfusion strategies significantly improve post-ischemic contractile recovery

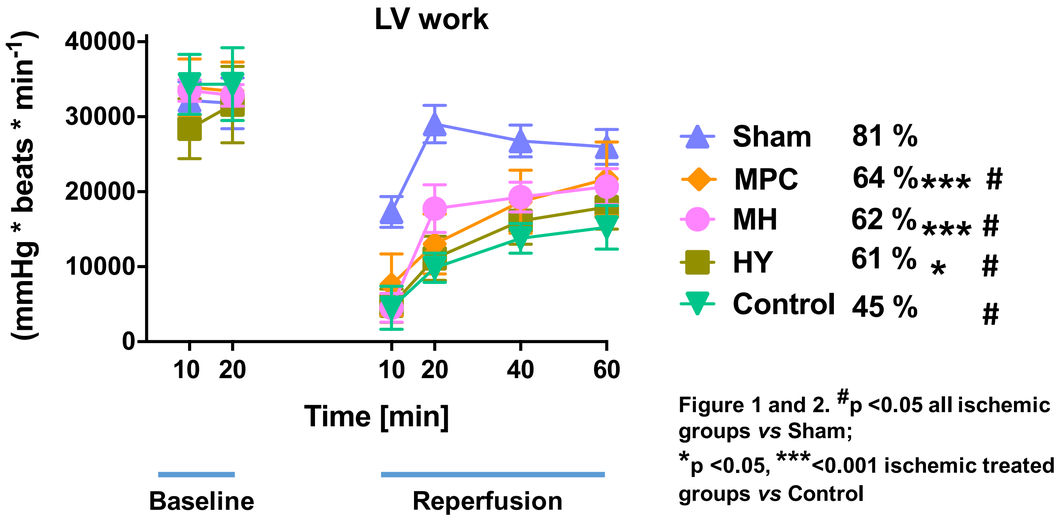
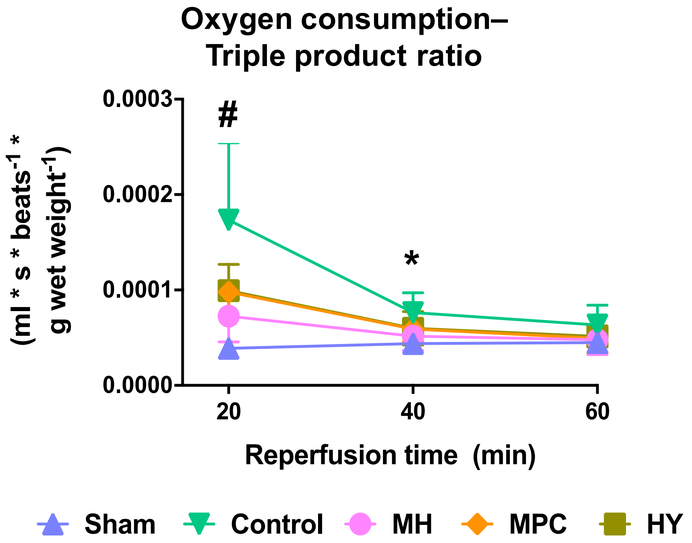


Figure 1 and 2. #p <0.05 all ischemic groups *vs* Sham;
*p <0.05, ***<0.001 ischemic treated groups *vs* Control

Left ventricle (LV) work: heart rate-developed pressure product

2 All strategies rapidly normalize cardiac oxygen efficiency



Triple product (T): LV work-dP/dtmax product

3 No strategy prevents protein carbonylation

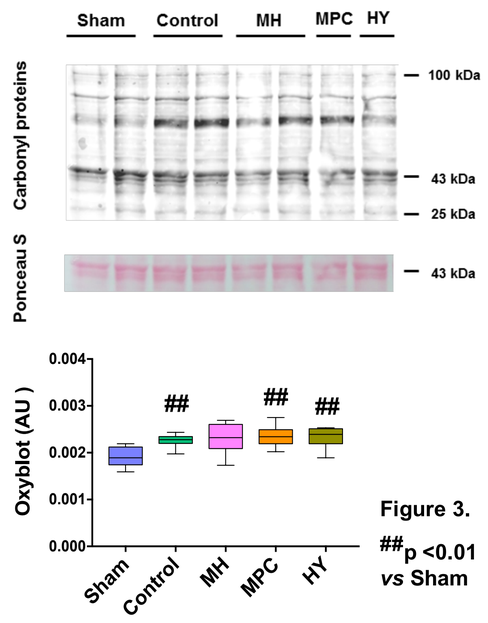


Figure 3. ##p <0.01 *vs* Sham

4 Strategies differentially modify mitochondrial function-related parameters

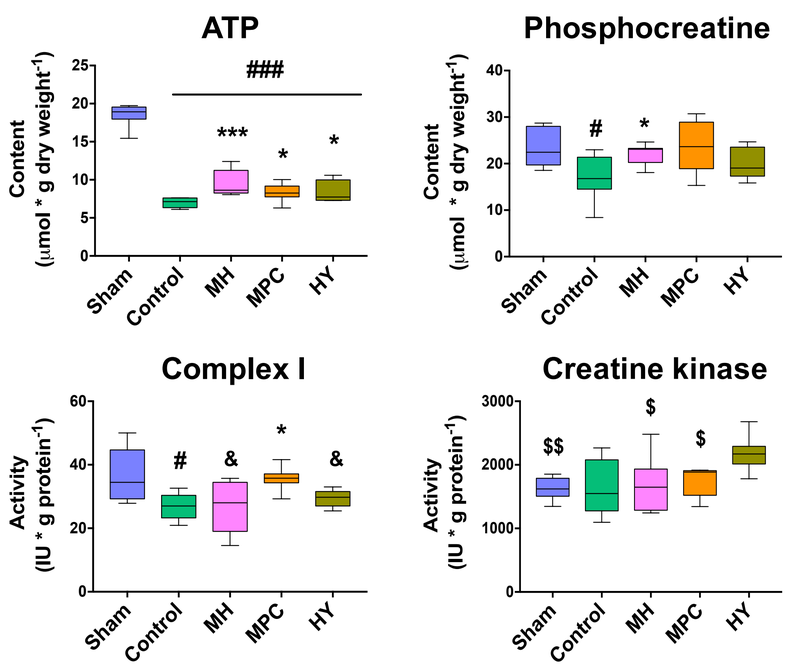


Figure 4. #p<0.05, ###<0.001 *vs* Sham; *<0.05, ***<0.001 *vs* Control; $$<0.01, $<0.05 *vs* HY; &<0.05 *vs* MPC

5 Mitochondrial DNA content tends to be normalized with MPC

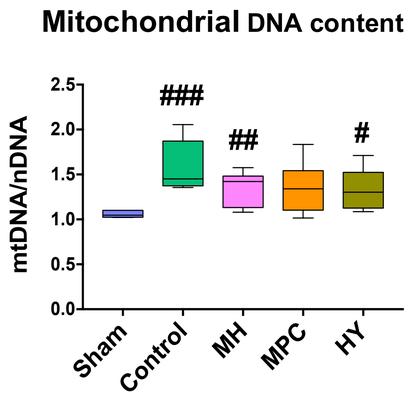


Figure 5. #p<0.05, ##p<0.01, ###<0.001 *vs* Sham

mtDNA, mitochondrial DNA; nDNA, nuclear DNA

6 60 min reperfusion: correlations profile between mitochondrial parameters and cardiac recovery differs with each strategy

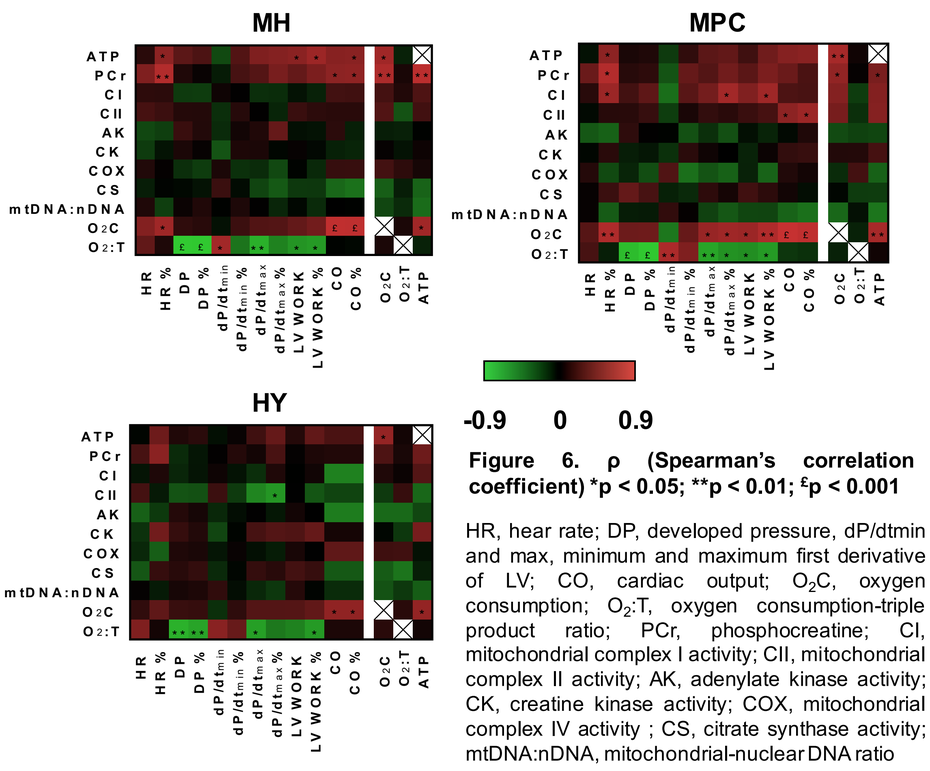


Figure 6. ρ (Spearman's correlation coefficient) *p < 0.05; **p < 0.01; £p < 0.001

HR, hear rate; DP, developed pressure, dP/dtmin and max, minimum and maximum first derivative of LV; CO, cardiac output; O_2C, oxygen consumption; O_2:T, oxygen consumption-triple product ratio; PCr, phosphocreatine; CI, mitochondrial complex I activity; CII, mitochondrial complex II activity; AK, adenylate kinase activity; CK, creatine kinase activity; COX, mitochondrial complex IV activity ; CS, citrate synthase activity; mtDNA:nDNA, mitochondrial-nuclear DNA ratio

CONCLUSION AND PERSPECTIVES

The cardioprotective reperfusion strategies, MH, MPC and HY, all appear to modify energy production and/or transfer, albeit through differing mechanisms. Further investigation into reperfusion-induced effects on mitochondria should aid in the identification of new therapeutic targets to optimize graft recovery and facilitate DCD heart transplantation.

[1]Farine E et al. Frontiers in Physiology (2016)

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Project funding: European Society of Cardiology, Swiss Heart Foundation, and Swiss National Science Foundation