anasthetised but not subject to ischemia. DIOHF (10 mg/kg iv) or vehicle (50% DMSO) was injected intravenously 5 minutes before reperfusion. mPTP opening was measured by monitoring mitochondrial Ca2+ retention capacity. Oxygen consumption of isolated mitochondria was measured in the presence of 5 mM and malate (5 mM) with a Clark-type electrode and ROS generation was assessed by measuring the rate of H2O2 production detected by amplex red.

Results: Treatment of sham rats with DIOHF significantly increased the concentration of Ca2+ required to stimulate mPTP opening (sham 87±6.8; sham-DIOHF 120±8.9 M). This was accompanied by an increase in state 3 oxygen consumption (sham 352±35; sham-DIOHF 572±21 nmol O2/min/mg protein) and a significant increase in H2O2 release (sham 0.028±0.002; sham-DIOHF 0.019±0.002 nmol/mg/mg protein). IR significantly decreased the concentration of Ca2+ required to stimulate mPTP opening (IR 44±5 μM), decreased state 3 oxygen consumption (IR 232±15 nmol O2/min/mg protein) and increased H2O2 release (IR 0.034±0.001 nmol/mg/mg protein) compared to sham. Treatment with DIOHF prevented IR-induced changes in mPTP opening (IR+DIOHF 78±7 μM), state 3 oxygen consumption (IR+DIOHF 375±30 nmol O2/min/mg protein) and H2O2 release (IR 0.028±0.002 nmol/mg/mg protein) so that there was no difference compared to sham.

Conclusions: In normal rats DIOHF inhibits mPTP opening and decreases mitochondrial ROS production. Importantly, DIOHF administration before reperfusion prevents IR-induced mPTP opening, impairment of state 3 respiration and increased oxygen consumption. The beneficial actions of DIOHF on mitochondria are likely to make a major contribution to its cardioprotective actions.

P1862 I BENCH Remote perconditioning limits the infarct size in anasthetized rabbits after stimulation of STAT5, independently of the RISK and SAFE pathways

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Introduction: We have previously confirmed that remote Perconditioning (PerC), induced by occlusions of the carotid artery of short duration, is a strong stimulus for the reduction of the infarct size in anesthetized rabbits, independently of its timing. However, effective algorithms in PerC and their mechanisms are still obscure.

Purpose: In the present study, we sought to test whether the RISK, SAFE and STS pathways are involved in the mechanism of the infarct size reduction in PerC.

Materials and methods: Anasthetized rabbits were subjected to 30-min myocardial ischemia (issc) and 10 minutes of reperfusion (rep). PerC with 4 cycles of 1 min issc-1min rep was applied at different time points of myocardial iscc by ligation of the carotid artery in 15 groups: Control, PerC-A applied at 29.30 min of iscc, PerC-B applied at 23.30 min of issc and ended at 30 sec of rep, PerC-C applied at 22.30 min and ended at 29.30 min of iscc, and PerC-D applied at 30 sec and ended at 7.30min of rep. The groups Control-W, PerC-A-W, PerC-B-W, PerC-C-W and PerC-D-W underwent the same interventions with co-administration of the PI3 inhibitor wortmannin (W); the groups Control-AG, PerC-A-AG, PerC-B-AG, PerC-C-AG and PerC-D-AG groups underwent the same interventions with co-administration of the selective STAT3 inhibitor Ag490. The infarct (i) to risk (R) ratio was estimated in %. In a second series of experiments, tissue samples from the ischemic area of myocardium were collected at the 10th minute of rep for P13k, GSK3). ERK1/2, STAT3 and STAT5 assessment.

Results: IR was reduced in all PerC groups (22.7±2.1% in PerC-A, 23.8±3.2% in PerC-B, 29.3±3.7% in PerC-C, 31.6±3.7% in PerC-D, 22.5±4.5% in PerC-A-W, 32.0±6.1% in PerC-B-W, 27.9±3.8% in PerC-C-W, and 30.5±3.3% in PerC-D-W compared with 47.0±2.5% in the Control and 47.6±2.4% in the Control-W group (P<0.05). Ag490 did not abrogate protection from PerC: %IR was 22.0±3.1% in PerC-A-AG, 16.6±3.0% in PerC-B-AG, 26.2±2.2% in PerC-C-AG and 16.3±2.2% in PerC-D-AG, respectively, with 47.2±5.7% in the Control and 39.7±3.8% in the Control-AG group (P<0.05). The phosphorylation of P13k, GSK3 and ERK1/2 was significantly higher in the PerC groups compared with the Control and W groups. No STAT3 phosphorylation was observed in any group. From the staining of STAT5 phosphorylation was significantly higher in PerC compared with the Control and W groups.

Conclusion: Remote perconditioning is effective in limiting the infarct size independently of the RISK and SAFE pathways. STAT5 stimulation seems to be the main trigger of cardioprotection.