Protective effect of *ex-vivo* reconditioning with PerSorb on kidney tubular mitochondrial dysfunction induced by ischemia-reperfusion injury

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Background and aim

Ischemia-reperfusion injury (IRI) represents the main cause of Delayed Graft Function (DGF) in the transplanted kidney, in particular using organs from ECD and DCD. Mitochondrial dysfunction (MD) represents an important mechanism of AKI enhancing the biological processes of fibrosis and cellular senescence and finally leading to early graft failure with the need of dialysis.

The aim of this study is the evaluation of MD in perfused pig kidneys at different time points in the presence or absence of the PerSorb adsorbing device.

Methods

3 en-block kidneys from pigs were procured: from each block, one kidney was perfused for 2 or 4 hours with the PerLife system (Aferetica SRL, Bologna, Italy), with or without the PerSorb cartridge, and the other one was used as experimental control. The following data were collected: temperature, arterial flow, arterial pressure (with initial target set at 60 mmHg), vascular resistance. Biochemical analyses were performed on proteins extracted from bioptic samples collected hourly, frozen, sonicated and processed by extraction of the mitochondrial components.

In particular, the evaluation of mitochondrial activity was performed on enzymes of the Krebs cycle, electron transport chain and on the efficiency of ATP synthesis: oxidation and loss of membrane potential were also evaluated as parameters of MD.

Results

Ex-vivo perfusion with the PerLife system, indipendently from the presence of PerSorb, showed excellent results in terms of arterial temperatures and pressures, as well as a significant reduction in vascular resistance (Fig. 1).

PerLife was able to improve the activity of the relevant enzymes of the Krebs cycle (TCA) citrate synthase, alpha-ketoglutarate, succinate and malate dehydrogenase (Fig. 2) and oxidative phosphorylation (OXPHOS), suggesting an improvement in mitochondrial function particularly evident after 4 hours of perfusion. Consistently, *ex-vivo* treatment increased the activity of enzymes of the electron transport chain along with increased intracellular ATP levels (Fig. 3). Moreover, MD (free radicals, mitochondrial reagents of thiobarbituric acid and loss of membrane potential) appeared significantly reduced in perfused kidneys vs. controls, with a further enhancement in the presence of the adsorptive cartridge PerSorb (Fig. 4).

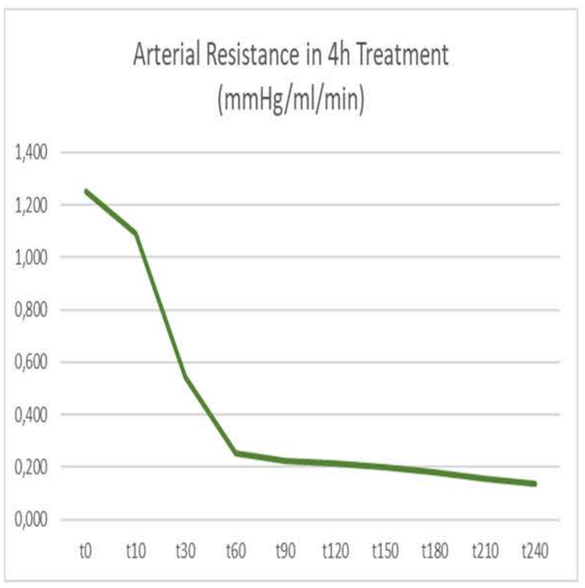

Fig. 1

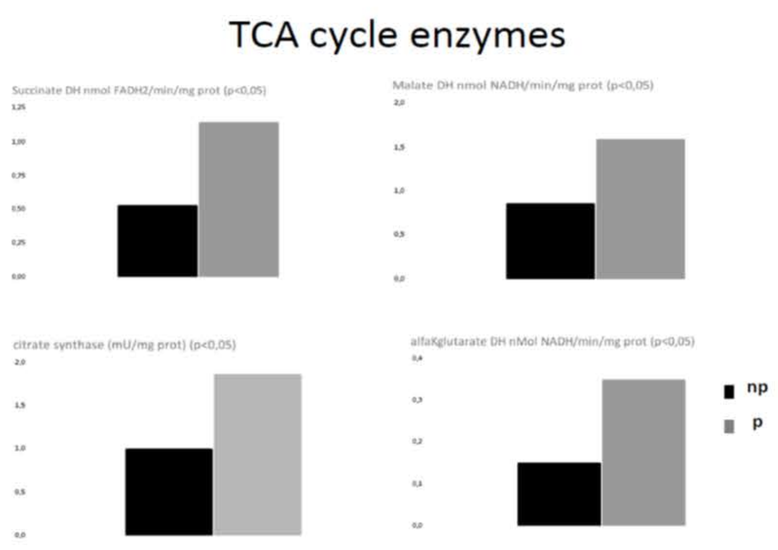

Fig. 2

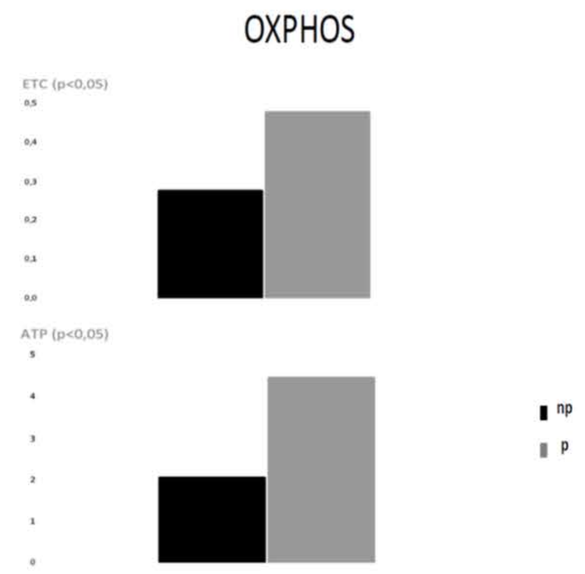

Fig. 3

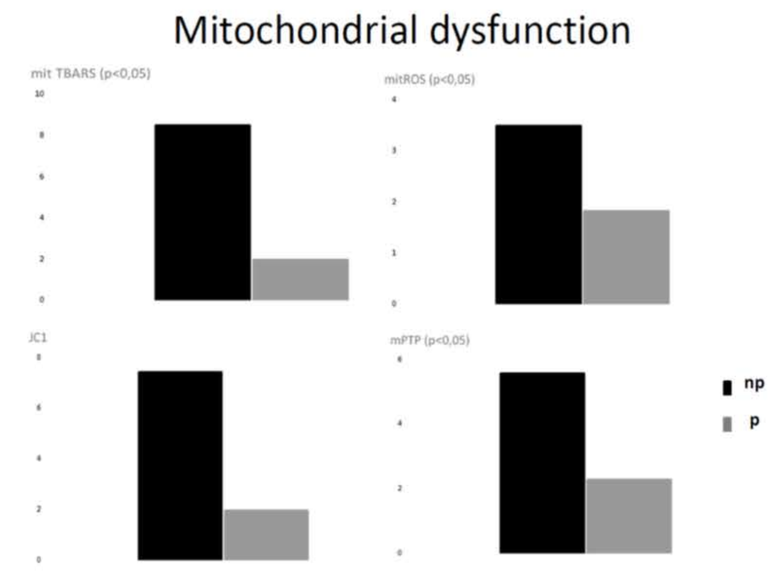

Fig. 4

Conclusion

The results of this preliminary study showed that kidney reconditioning with the PerLife system and in particular using PerSorb significantly restored the homeostasis of the mitochondrial network with a possible limitation of IRI and promoting tubular regeneration. The integration of the PerLife system with drugs sustaining mitochondrial function or inhibiting senescence as well as the potential use of mitochondrial transplantation may further improve ischemic kidney injury limiting DGF and graft function.