Chronic Cyclosporine Treatment Does Not Reduce Total LV Collagen and Fibrosis in Mini-Swine with Heart Failure

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ABSTRACT

Cardiac extracellular matrix remodeling is a pathological process that may negatively affect the mechanical properties of the heart in patients with heart failure with preserved ejection fraction (HFpEF). The remodeling process is partially regulated by the loss of cardiomyocytes through cell death pathways mediated in part by the mitochondria. Our laboratory previously showed that low intensity interval exercise training attenuates mitochondrial dysfunction, characterized by increased mitochondrial permeability transition (MPT). Conventional treatments have failed to improve the prognosis of HFpEF patients, and there is a critical need for generating novel treatment options for those diagnosed with the disease. Therefore we hypothesized that a reduced, non-immunosuppressive dose of the drug cyclosporine (CsA; a general cyclophilin inhibitor) would block MPT via inhibition of cyclophilin D, a key component of the MPT pore, and attenuate the development of HFpEF via inhibition of cell death pathways and subsequent fibrotic myocardial remodeling.

METHODS

**Cyclosporine Treatment**

In the presence of existing LV hypertrophy (six weeks post-surgery), animals began Cyclosporine treatment. Animals were dosed 2 mg/kg/day for a duration of 14 weeks.

**Groups**

- Control non-banded (CON); n=5
- HFpEF non-treated (HF); n=5
- HFpEF treated with CsA (HF-CsA); n=5

**Aortic Banding Procedure**

LV hypertrophy/HF was induced by aortic banding. The aortic band was placed on the ascending aorta proximal to the brachiocephalic artery. A systolic trans-stenotic gradient of 70 mmHg (73 ± 2 & 74 ± 1 mmHg for HF & HF-CsA, respectively, P = nonsignificant [NS]) was achieved while maintaining a distal peripheral vascular MAP of 90 mmHg (93 ± 1 & 90 ± 1 mmHg for HF and HF-CsA, respectively, P = NS) under anesthesia using phenylephrine (1–3 g·kg−1·min−1 iv) at a heart rate (HR) of 100 beats/min (100 ± 5 and 107 ± 2 beats/min for HF and HF-CsA, respectively, P = NS).

**Histology and immunohistochemistry**

Cross-sections of LV were formalin fixed, embedded in paraffin, and immunohistochemistry stained for the assessment of fibrosis and collagen. Brachiocephalic Artery sections of LV were visualized using Masson's trichrome stain, and total collagen was visualized using Picrosirius red staining with previously established methods. Fibrosis and collagen were quantified from 4 separate fields/animal using Image-Pro Plus analysis software (version 6.2, MediaCybernetics, Bethesda, MD) and expressed as the percent area stained and density of the stain.

**RESULTS**

- **Figure 1.** Cyclosporine does not decrease total LV fibrosis measured as percent area (A) or measured as density of stain (B) in HEPF animals (*P<0.05 vs. HF & HF-CsA).
- **Figure 2.** A, CsA does not decrease total LV collagen measured as percent area. B, CsA does decrease LV collagen density. (*P<0.05 vs. HF & HF-CsA; †P<0.05 vs. CON & HF-CsA).

**CONCLUSION**

Chronic cyclosporine treatment does not decrease total LV collagen or fibrosis in a mini-swine model of HFpEF.

Our results suggest cyclosporine is not a viable therapeutic treatment for HF.

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