Investigating the role of the *Spp1* gene in cardiac hypertrophy and fibrosis utilising normotensive and spontaneously hypertensive rat strains.

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Abstract

Background: A quantitative trait locus (QTL) for left ventricular mass index (LVMI) was previously identified on chromosome 14 in an F2 cross of the stroke-prone spontaneously hypertensive (SHRSP) and Wistar Kyoto (WKY) rat strains. Congenic strains (SP.WKY$_{Gla}$14a and WKY.SP$_{Gla}$14a) were constructed on both SHRSP and WKY genetic backgrounds respectively to confirm the LVMI QTL. Microarray gene expression profiling identified *Spp1* as a candidate gene for the increased LVMI phenotype. *Spp1* expresses the osteopontin phosphoprotein and plays a role in cardiac fibrosis and hypertrophy.

Aim: In this study, we aimed to investigate the role of *Spp1* in the development of cardiac hypertrophy and fibrosis using transaortic constriction (TAC) in CRISPR Cas9-generated SHRSP *Spp1* knockout (KO) and wildtype (WT) rats, WKY and WKY.SP$_{Gla}$14a congenic strains.

Methods: Transaortic-constriction (TAC) or sham-surgery was performed in 15-week-old, male rats. Thereafter, 8-weeks of phenotypic analysis was conducted, using tail-cuff plethysmography for BP and echocardiography for cardiac parameters. Ex-vivo histological examination of hearts was conducted for measurement of cardiac fibrosis.

Results TAC surgery had no impact on BP in any of the rat strains. TAC surgery induced a hypertrophic response as evidenced by increased LVMI in WKY and WKY.SPGla14a strains ($p<0.05$ and $p<0.001$, respectively). Strain-specific differences in echocardiography parameters (cardiac output, stroke volume and relative wall thickness) in sham-operated rats were generally negated in TAC-operated rats. Histological analysis revealed significantly increased perivascular fibrosis in all TAC-operated strains ($p<0.05$). Interstitial and perivascular fibrosis in WKY.SPGla14a rats were significantly increased compared to all other strains post-TAC ($p<0.05$ and $p<0.0001$, respectively).

Conclusions TAC successfully induced hypertrophic responses in normotensive genetic background rat strains, whereas *Spp1* WT and KO rats exhibited pre-existing hypertrophy, potentially limiting further cardiac remodelling. Preliminary findings suggest that elevated early-life *Spp1* expression in SHRSP rats correlates with increased susceptibility to cardiac hypertrophy and fibrosis in adulthood.