Insulin regulated aminopeptidase / AT_4 receptor deficiency is both cardio- and vaso-protective in angiotensin II-infused mice

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Chronic treatment with the hexapeptide 3-8 fragment of Angiotensin (Ang) II, known as Ang IV, mediates vasoprotective effects in the ApoE KO mouse model of atherosclerosis and we propose that these effects are mediated by the binding of Ang IV to the AT4 receptor, now identified as insulin-regulated aminopeptidase (IRAP), inhibiting the catalytic activity of this enzyme. Therefore the objective of this study was to investigate whether IRAP deficiency confers a protective phenotype under condition of cardiovascular stress.

Using the Ang II-infusion model of hypertension, male IRAP-deficient ($IRAP^{-/-}$) mice and their WT littermate controls (~12 weeks of age) were treated with either Ang II (800 ng/kg/day) or saline subcutaneously *via* osmotic mini-pumps for 4 weeks. Ang II-treated WT and $IRAP^{-/-}$ mice had a significant increase in systolic blood pressure (141 ± 4 mmHg and 149 ± 8 mmHg, respectively; n=8-11, P<0.01) compared to vehicle treated mice, with a concomitant increase in HW:BW ratio. Ang II-infused WT mice had impaired endothelium-dependent vasorelaxation (Rmax: 44.2% ± 6.0; n=7) compared to saline-treated WT mice (Rmax: 69.5 ± 7.3; n=6, *P*<0.01). Interestingly Ang II-infused $IRAP^{-/-}$ mice showed no evidence of endothelial dysfunction (Rmax: 63.9% ± 5.1; n=6). Ang II-induced endothelial dysfunction was correlated with a reduction in eNOS immunostaining in Ang II-infused WT mice that was prevented in $IRAP^{-/-}$ mice. Ang II-infusion evoked cardiac fibrosis in WT mice with an increase in collagen deposition (3.41 ± 0.33 %; n=5; *P*<0.001) compared to saline treated WT mice (0.86 ± 0.10%; n=5). Excitingly, $IRAP^{-/-}$ mice were protected against development of increased cardiac interstitial fibrosis (0.65 ± 0.04%; n=5; *P*<0.001) when treated with Ang II for 4 weeks.

In conclusion this study has shown that $IRAP^{-/-}$ mice possess both vaso- and cardio-protective phenotype under a condition of cardiovascular stress and thus highlights the importance of targeting the IRAP/AT_4R in cardiovascular disease.