UROKINASE PLASMINOGEN ACTIVATOR-INDUCED CARDiac FIBROSIS PROCEEDS BY AN ANGIOTENSIN-INDEPENDENT MECHANISM.

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OBJECTIVE Cardiac fibrosis is a common finding in end stage heart disease and likely contributes to worsening function. Increased plasmin activity is found in human hearts with fibrosis. We have generated mice with macrophage-targeted overexpression of urokinase (SR-uPA+/o mice) that develop cardiac fibrosis and macrophage accumulation by 15 weeks of age. Treatment with angiotensin II receptor blockers (ARB) decreases cardiac fibrosis in several mouse models. Since renin is a plasmin substrate, we hypothesized that uPA-induced cardiac fibrosis is mediated in part by the renin-angiotensin system. To test this hypothesis, we treated SR-uPA+/o mice with the ARB losartan and analyzed hearts for fibrosis and macrophage accumulation.

METHODS SR-uPA+/o mice and nontransgenic littermates were administered either 10 mg/kg/d losartan in drinking water or no treatment from 5 weeks of age (a time-point with no cardiac fibrosis) to 13.5 weeks of age, and were sacrificed at 15 weeks. Hearts were fixed, embedded, and sectioned. Fibrillar collagen was detected with picrosirius red staining, and percent fibrosis in the mid-ventricle was quantified using digital image analysis. Macrophages were detected using a Mac-3 antibody, and average macrophage density was determined by manual counting of 20 high-power (400X) fields per heart.

RESULTS Contrary to our initial hypothesis, SR-uPA+/o mice receiving losartan treatment had no significant difference in fibrosis compared to SR-uPA+/o mice receiving no treatment (12.9 ±6.7% vs 7.9 ±5.6%, n=8, P=0.126 by student’s t-test). However, the losartan group had a 2-fold increase in cardiac macrophages compared to the untreated group (212±175 vs 102±81 Mac-3 positive cells/mm², n=8, P=0.05 by one-way ANOVA). Untreated and losartan-treated nontransgenic control mice showed no fibrosis and minimal macrophage accumulation.

CONCLUSIONS In SR-uPA+/o mice, treatment with losartan did not decrease cardiac fibrosis, indicating that uPA-induced cardiac fibrosis proceeds through an angiotensin-independent pathway. Surprisingly, losartan treatment significantly increased cardiac macrophage density in SR-uPA+/o hearts. Further investigation of the mediators of ARB-induced increases in cardiac macrophage accumulation may illuminate mechanisms of angiotenin inhibitor escape in end-stage heart disease.