A chronic angiotensin II infusion mouse model of hypertension-induced cardiac fibrosis

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BACKGROUND & AIM

Development of novel pharmaceutics for hypertension-induced cardiac disease depends on animal models that can reliably reproduce patient phenotypes, including the full spectrum of cardiac fibrosis. Chronic infusion with angiotensin II (AngII) in mice has become the most widely used preclinical model and symptoms include hypertension, cardiac fibrosis, cardiomyocyte death, and dilated cardiomyopathy.

Here we present a mouse model of hypertensive cardiac remodeling, left ventricular dysfunction and fibrosis induced by chronic AngII infusion.

METHODS

Male C57BL6/N mice were administered saline vehicle (n=10) or Ang II (1.5 mg/kg/day, n=12) for 5 weeks using subcutaneous osmotic minipumps (Alzet 2006). Echocardiography was performed in study week 4. At termination, plasma was collected for biochemistry and the heart was sampled for histology and RNA sequencing.

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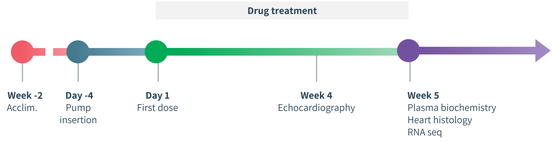
1 Study design and study groups



Group	Animal	Gender	Number of animals	Treatment	Administration route	Dosing concentration
1	C57Bl/6N	Male	10	**Vehicle**	Osmotic pump (2006 Alzet)	-
2	C57Bl/6N	Male	12	**AngII**	Osmotic pump (2006 Alzet)	**1.5 mg/kg/day**

Figure 1. Study outline.

2 Chronic AngII infusion causes cardiac hypertrophy

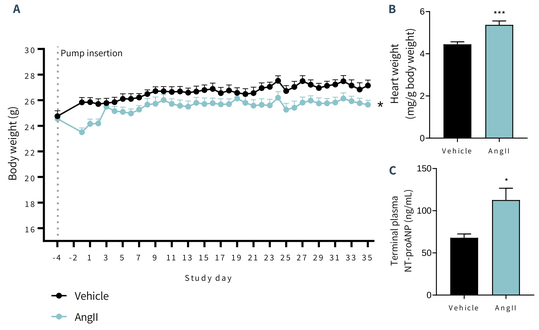


Figure 2. Body weight, heart weight and plasma NT-proANP. Osmotic minipumps were inserted on study day -4. **(A)** Body weight. **(B-C)** Relative heart weight and plasma N-terminal pro-atrial natriuretic peptide (NT-proANP) levels at termination (study day 35-36). Mean + S.E.M. *p<0.05, ***p<0.001 vs. Vehicle (Dunnett's test one-factor linear model).

3 Chronic AngII infusion causes cardiac fibrosis

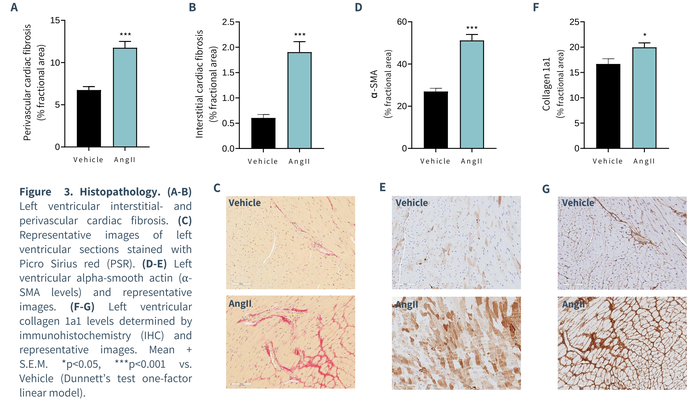


Figure 3. Histopathology. (A-B) Left ventricular interstitial- and perivascular cardiac fibrosis. **(C)** Representative images of left ventricular sections stained with Picro Sirius red (PSR). **(D-E)** Left ventricular alpha-smooth actin (α-SMA levels) and representative images. **(F-G)** Left ventricular collagen 1a1 levels determined by immunohistochemistry (IHC) and representative images. Mean + S.E.M. *p<0.05, ***p<0.001 vs. Vehicle (Dunnett's test one-factor linear model).

4 Chronic AngII infusion causes LV dilation and cardiac dysfunction

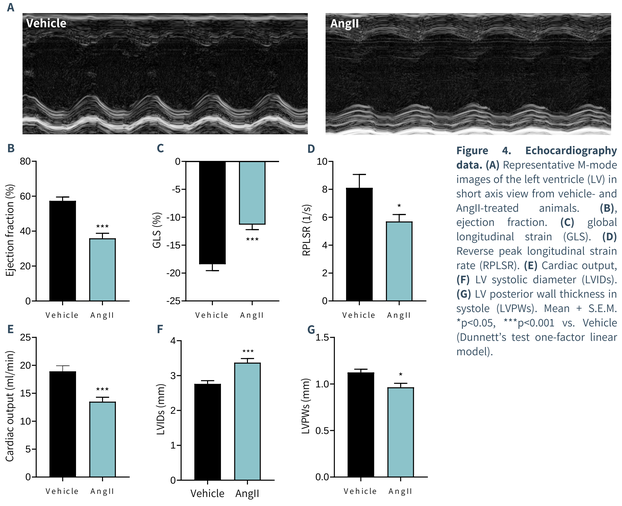


Figure 4. Echocardiography data. (A) Representative M-mode images of the left ventricle (LV) in short axis view from vehicle- and AngII-treated animals. **(B)**, ejection fraction. **(C)** global longitudinal strain (GLS). **(D)** Reverse peak longitudinal strain rate (RPLSR). **(E)** Cardiac output, **(F)** LV systolic diameter (LVIDs). **(G)** LV posterior wall thickness in systole (LVPWs). Mean + S.E.M. *p<0.05, ***p<0.001 vs. Vehicle (Dunnett's test one-factor linear model).

5 RNA sequencing

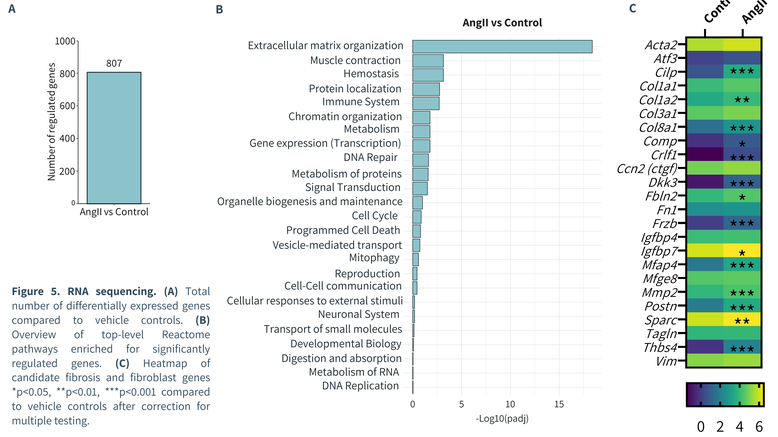


Figure 5. RNA sequencing. (A) Total number of differentially expressed genes compared to vehicle controls. **(B)** Overview of top-level Reactome pathways enriched for significantly regulated genes. **(C)** Heatmap of candidate fibrosis and fibroblast genes *p<0.05, **p<0.01, ***p<0.001 compared to vehicle controls after correction for multiple testing.

CONCLUSION

- Chronic AngII infusion causes perivascular and interstitial fibrosis.
- Chronic AngII administration promotes eccentric hypertrophy reflected by increased heart weight and left ventricular dilation.
- These changes result in impaired cardiac remodling indicated by systolic and diastolic dysfunction along with decreased cardiac output.
- RNA sequencing revealed significant upregulation of fibrosis and fibroblast gene expression markers in the model.
- The chronic AngII mouse model is suitable for evaluating drug effects on hypertension-driven cardiac remodeling and fibrosis.

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