Renal Oxidative Stress in Renin-Angiotensin-Aldosterone Model of Hypertension

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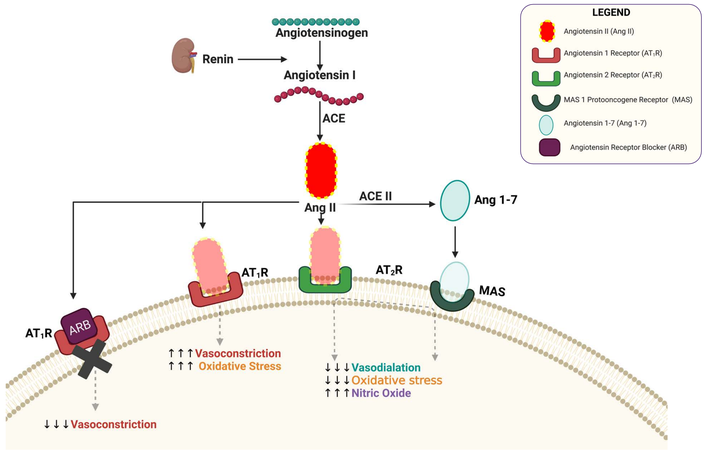
Abstract

Hypertension is a polygenic condition in which high blood pressure leads to cardiovascular complications, cerebral damage, and kidney failure. The Renin-Angiotensin-Aldosterone System (RAAS) model of hypertension, (mREN2)27 transgenic rodent, triggers complex signaling pathways and cellular processes. It is characterized by overexpression of mouse Ren-2^{d} gene in brain and adrenal gland, with a reduction in kidney renin. The main pressor component of the RAAS is Angiotensin II (AngII) and it exists in many local organs and tissues, including the nephrons. AngII signals the production of reactive oxygen species (ROS) that serves as the epicenter for intracellular signaling pathways and contributes to the hypertensive episode. Despite the research contributions over the years, the role of circulating angiotensin peptides in genetically predisposed hypertensive episodes and their lasting effects on inflammation and immune response remains unclear. We *hypothesized* that in the (mREN2)27 model of hypertension, there will be imbalance in the expression of inflammatory responses and ROS modulators. The *results* show that there are significant increases in gene expression for ROS modulators such as Heme Oxygenase-1 (HO-1), Nuclear Factor Erythroid-2-Related Factor 2 (Nrf2), and Receptor for Advanced Glycation End Products (RAGE). Further, gene expression for inflammatory responses to Transforming Growth Factor Beta (TGF-β), and Nuclear Factor Kappa B (NF-κB) are significantly higher in (mREN2)27 compared to normotensive rodents. The generation of ROS exhibits numerous inflammatory properties that provoke the ultimate disruption to renal physiology. The upregulation of these modulators and inflammatory responses suggests kidney stress due to prolonged activation of the RAAS which may lead to chronic kidney disease (CKD) in this hypertensive model. These findings underscore the potential to identify therapeutic targets aimed at mitigating CKD.

Introduction



Angiotensin II pathway diagram and the mechanism of action for candesartan, which is an AT_1 R blocker.

Objective

The present study investigates the intracellular gene expression for ROS and proinflammatory modulators in renal tissue of the (mREN2)27 transgenic hypertensive rodent.

Methods

RNA Isolation and Preparation
- Extracted RNA from kidney tissues of both hypertensive (mRen2)27 and normotensive (HnSD) rats
- Assessed the purity and concentration of the RNA

Complementary DNA Preparation
- Converted the extracted RNA to complementary DNA (cDNA) using reverse transcription.

Real-Time Polymerase Chain Reaction
- Prepared qRT-PCR reaction mix including cDNA, gene-specific primers, and a master mix containing DNA polymerase, and SYBR Green fluorescent dye

Results

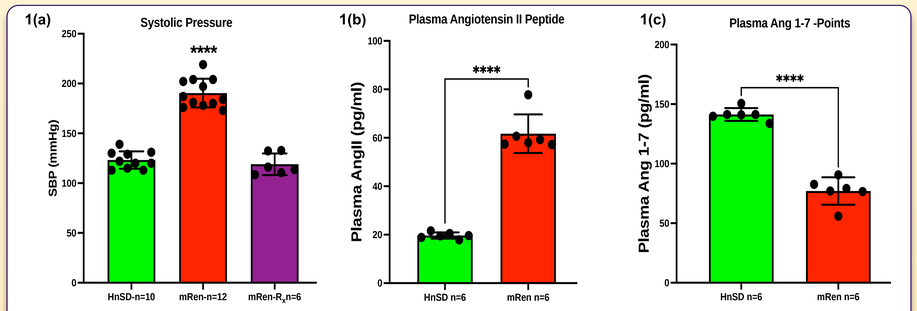


Figure 1: Baseline Hypertensive Biomarkers. (**a**) **Systolic Blood Pressure** was significantly higher in (mREN2)27 rats compared to HnSD (****p<0.0001). Candesartan (CandRx) lowered blood pressure to normal, indicating Ang **II** effects mediated via AT_1 receptor sub-type. A significantly higher level of circulating (**b**) **Plasma Angiotensin II Peptide** and lower levels (**c**) **Plasma Ang 1-7 –Points** in (mREN2)27 rats compared to HnSD (****p<0.0001; ****p<0.0001)

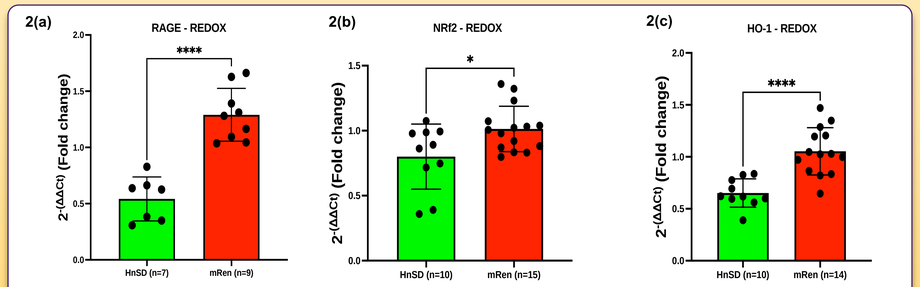


Figure 2: RT-PCR expression of mRNAs in REDOX. Relative gene expressions of (**a**) **RAGE**, (**b**) **NRf2**, and (**c**) **HO-1**. mRNA for (**a-c**) is significantly higher in renal tissue of (mREN2)27 transgenic hypertensive animal compared to HnSD normotensive control, ****p<0.0001; ****p<0.0001; and *p<0.05 respectively.

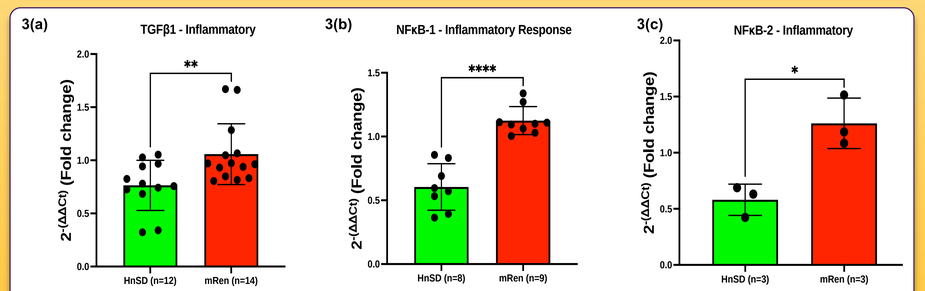
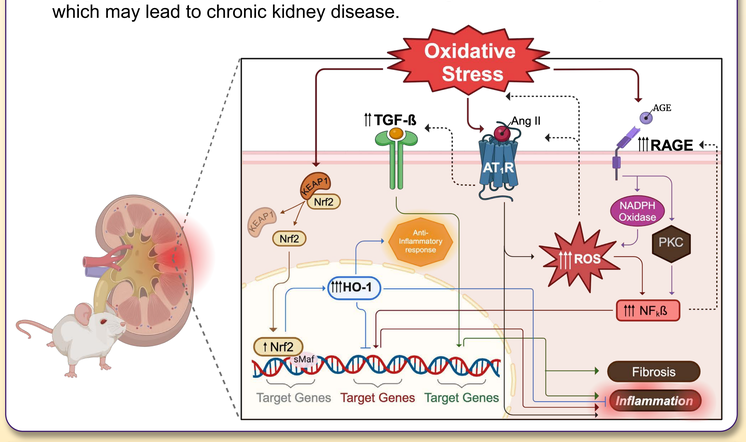


Figure 3: RT-PCR expression of mRNAs in Proinflammation. Relative gene expressions of (**a**) **TGFβ1**, (**b**) **NFkB-1**, and (**c**) **NFkB-2**. mRNA for (**a-c**) is significantly higher in renal tissue of (mREN2)27 transgenic hypertensive animal compared to HnSD control. ****p<0.0001; **p<0.01; and *p<0.05 respectively.

Conclusions

- Systolic Blood pressure was normalized by an Angiotensin Receptor Blocker (ARB), Candesartan via AT_1R mediation.
- Increase level of AngII but a decrease in AngII metabolite, (Ang 1-7), indicating a diminution in the protective arm of AngII.
- There is upregulation of mRNA expression for ROS and inflammatory responses suggesting kidney stress may be due to prolonged activation of systemic RAAS which may lead to chronic kidney disease.



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