Expression of Na+/K+ ATPase Isoforms and Na+/Ca2+ Exchanger Activity in Cardiac Myocytes of Ren-2 Transgenic model of Hypertension

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Abstract

Introduction: The Renin Angiotensin Aldosterone System (RAAS) is composed of various reactions in the regulation of blood pressure and plays crucial roles in cardiovascular physiology and pathophysiology. Classically, renin cleaves liver-derived angiotensinogen (AGT) into angiotensin I (Ang I), a decapeptide [Ang 1–10] which is then further processed by angiotensin-converting enzyme (ACE) into angiotensin II (Ang II) [Ang 1–8]. Ang II is known to physiologically regulate blood pressure and is a key player in hypertension. Ang II causes vasoconstriction and production of aldosterone leading to retention of sodium (Na+) and water resulting in cardiovascular dysfunction. Objective: To study Ang II receptor mediation and the expression of intramembranous transporter of Na+ and Ca2+ in cardiomyocytes and correlate activities of different Na+/K+ ATPase isoforms with the (mRen2)27 transgenic phenotype. Methods: The left ventricles of 12–15-week-old (mRen2)27 transgenic and Hanover-Sprague Dawley (HSD) rats were isolated where protein was used for SDS PAGE and Western blotting analysis. Results: showed a significant increase in the Mean Arterial BP in (mRen2)27 transgenic rats but no change in pulse rate compared to HSD control. There was a significant protein expression for Ang II receptor sub-type 1 (AT, R) in (mRen2)27 when compared to control normotensive rodent. There were no differences in Na+/K+ ATPase α-1 isoform in both strains but a significant diminution in α-2 isoform in (mRen2)27 transgenic hypertensive rodents, suggesting an increase in intracellular Na+ and Ca2+ concentrations in cardiomyocytes through the Na-Ca exchanger system. Conclusion: The finding suggests an increased AT, R receptor protein, a diminished Na+/K+ ATPase α-2 isoform expression and augmented intracellular Na+ and Ca2+ concentrations in (mRen2)27 transgenic hypertensive rodents, all of which may contribute to an increase in cardiac contractility, cardiac output, and sustained blood pressure.

Discussion

• The main objective is to identify the pathways directly responsible for the increase in contractility of the heart and hypertension in genetic form of hypertension, using the (mRen2)27 transgenic model.
• The AT, R sub-type is significantly present in hypertension compared to control, HSD, suggesting a possible source Ang II receptor mediation of the increased cardiac contractility.
• There was no change in the expression of ATPase α-1 isoform in (mRen2)27 and HSD, but the ATPase α-2 isoform was significantly lowered in (mRen2)27 suggesting that the Na+/K+ ATPase pump activity is diminished, leading to an increase in intracellular Na+ as well a possible increase in Ca2+ concentrations. The latter is due to the reversal of sodium–calcium exchanger and a consequence in membrane depolarization that then leads to increased cardiac contractility and cardiac output as well as high blood pressure.
• This research is a part of a larger goal to seek the intracellular targets for genetic form of hypertension for therapeutic purposes.

Results

Figures 1: Blood Pressure Measurements. Tail-cuff (non-invasive) was used to record blood pressure for five consecutive days in 12–15-week-old Rodents. Systolic and Mean arterial BP was significantly higher in (mRen2)27 rats when compared to HSD rats (*p<0.001; n=12). There was no change in the pulse rate.

Figures 2: AT-1 Expression in Heart

• There was a significant increase in the amount of AT, R in (mRen2)27 as compared to HSD at 12.5 weeks old. The mediation of the ventricular contractility is thought to be via AT, R in (mRen2)27 compared to HSD. *p<0.001; n stands for total number left ventricular muscle tissues used in the analysis in each group.

Figures 3: Na+/K+ ATPase isoforms in the ventricular myocytes. There was no difference ATPase α-1 isoform in both strains but a significant decrease in α-2 isoform in (mRen2)27 transgenic rodents compared to HSD, suggesting an increase in intracellular Na+ and Ca2+ concentrations through the Na-Ca exchanger. *p<0.05, n stands for total number left ventricular muscles used in the analysis in each group.

Methods

(a) Isolation of left ventricular muscle from the HnSD and mRen227, (b) Protein isolation from the HnSD and mRen227 heart (c) SDS PAGE and Western blotting analysis of Na+/K+ ATPase isoforms (α-1 & α-2), Na+/Ca2+ activity and protein expression.

Scheme: Na+/K+ ATPase, Ca2+ Exchanger – Cadium is removed from cells by Na+/K+ basic mechanisms: 1) an ATPase-dependent Ca2+ pump that actively removes calcium from the cell 2) the sodium-calcium exchanger. During ventricular systole when the myocytes are depolarized, Ca2+ enters the cell through this exchanger. In contrast, during ventricular diastole when the cells are repolarized, Ca2+ leaves the cell through the exchanger. An increase in intracellular sodium concentration leads to an increase in intracellular calcium concentration through this exchange. This has important physiological implications including when the activity of the Na+/K+ ATPase is decreased.

Conclusion


Reference

• An introduction to the intracellular targets for genetic form of hypertension for therapeutic purposes.

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