LOCALIZATION OF THE SODIUM-ACTIVATED SODIUM CHANNEL (NaX) IN THE RAT KIDNEY.

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Male Sprague-Dawley (SD) rats subjected to high salt intake (HS) and chronic angiotensin II (Ang II) infusion show exaggerated increases in proteinuria and renal cortical number of n-tyrosine residue proteins, a marker of oxidative stress without additional increases in systolic blood pressure. The combination of HS with chronic Ang II infusion exacerbates proteinuria and albuminuria to a greater extent than Ang II infusion alone. It remains unclear how increased dietary salt triggers specific signaling pathways that may contribute to greater oxidative stress and further development and progression of hypertension and kidney injury. Recent studies have described a sodium sensor in the brain, lung, pregnant uterus and heart, which is thought to be in the family of the voltage-gated sodium channel, which operates as a sensor of sodium concentration in the interstitial fluid bathing the cells. The overall objective of this study is to identify and characterize the presence of the sodium-activated sodium channel (NaX) in the kidney.

To achieve this aim total RNA was extracted from SD rat kidney cortex, medulla and two types of rat kidney cells, cultured immortalized proximal tubule cells (rPT) and isolated cells from inner medullary collecting ducts (IMCD). Total RNA from brainstem and organum vasculosum of the lamina terminalis (OVLT) were used as positive controls. By semi-quantitative PCR, the presence of the 251 bp bands was observed in the samples from both kidney cortex and medulla, IMCD cells as well as in the positive controls. The absence of a band in rPTC indicated that the immortalized cell lost the capacity to express the NaX. The protein expression of the NaX was determined by Western blot and immunocytovhemistry. Protein extracts derived from homogenate and total membrane fraction of SD rat kidney cortex revealed a single band around 110-160 KDa in both preparations. The enrichment in total membrane fractions (3 times) was parallel to the enrichment of the (Na⁺+K⁺)-ATPase suggesting that the NaX is a membrane-resident protein as expected for a channel. Immunofluorescence staining on acetone fixed kidney pole frozen sections showed the presence of the NaX mainly in the luminal membrane of the proximal tubules. These data demonstrate the presence of a NaX in the luminal membrane side of kidney proximal tubule cells; thus suggesting that it has a physiological role in the control of the local extracellular Na⁺ concentration.

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