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# Development of diagnostic and therapeutic drugs for salt-sensitive hypertension and hyponatremia in elderly

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Summary

Identification of gain-of-function mutation in epithelial sodium channel (ENaC) in Liddle's syndrome patients, a hereditary form of salt-sensitive hypertension, indicates the importance of the sodium reabsorption through the kidney in the pathogenesis of salt-sensitive hypertension. In 1997, Vallet et al isolated a channel-activating protease (CAP-1), a trypsin-like serine protease, from A6 cell line and demonstrated that co-expression of CAP-1 and ENaC in *Xenopus* oocytes increased ENaC activity. We isolated a serine protease prostasin, a mammalian CAP-1 homologue, from rat kidney cDNA library and demonstrated that co-expression of prostasin and ENaC increased the amiloride-sensitive sodium current in *Xenopus* oocytes. We also found that aldosterone increases sodium reabsorption through ENaC by increasing the expression of prostasin. Recently a serine protease inhibitor, protease nexin-1 (PN-1), was identified as an endogenous inhibitor for prostasin activity. Therefore, we hypothesized that PN-1 may regulate sodium reabsorption in the kidney by reducing prostasin activity, and that expression of PN-1 was regulated by TGF-β1 or aldosterone, like prostasin. Expression of PN-1 substantially decreased Prostasin-induced $I_{Na}$ by approximately 68% in oocytes. Treatment of M-1 cells with 20 ng/ml TGF-β1 significantly increased mRNA and protein expression of PN-1 by 5.6 ± 0.3 and 3.8 ± 0.5 fold respectively, whereas administration of $10^{-6}$ M aldosterone markedly decreased mRNA and protein expression of PN-1 to 61.9 ± 6.0 and 53.7 ± 6.7 % respectively. In addition, knockdown of PN-1 expression by siRNA in M-1 cells resulted in the increase in $I_{eq}$ by 1.8 fold. Our study indicates that PN-1 could have a natriuretic role by inhibiting prostasin activity and suggests the possibility that aldosterone and TGF-β reciprocally regulate the expression of PN-1 in renal epithelial cells contributing to salt retention or natriuresis, respectively by an additional mechanism. PN-1 could represent a new factor that contributes to regulation of ENaC activity in the kidney.