Role of a novel vasopressin receptor, Vp in intrarenal sodium transport and its pathophysiological significance

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Summary

Recently we reported that a new AVP receptor (Vp) in early proximal tubule (S_1) using a cytosolic free calcium measurement. The physiological significance of Vp was an inhibition of the ATP-consuming on transport system based on changes in cellular ATP. In order to clarify further the functional significance of Vp, we have tried the molecular cloning of Vp. Although rat kidney cDNA library was screened with binding assay and panning method using COS-1 cell expression system, no positive clone was identified. Thus, we undertook PCR cloning. In order to collect highly Vp expressed cell as a template, we have screened cloned S1 cells derived from SV-40 large T antigen transgenic mice using cytosolic calcium measurement, resulting in the non-cloned cells as the most stably Vp-expressed cells. Degenerative primer was constructed according to the conserved amino acid sequence of AVP receptor family. PCR reaction was processed under various conditions. However, only cDNA for V_1 and V_2 have been cloned by PCR, suggesting that Vp may have a low homology with AVP receptor family. Therefore, we have started the expression cloning system using Xenopus oocytes for the molecular cloning of Vp.