EXPANDED ABSTRACT

AVP-stimulated nucleotide secretion in perfused mouse medullary thick ascending limb and cortical collecting duct

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Summary : Extracellular nucleotides are local, short-lived signaling molecules that inhibit renal tubular transport via both luminal and basolateral P2 receptors (1, 2). Apparently, the renal epithelium itself is able to release nucleotides (3, 4). The mechanism and circumstances under which epithelia nucleotide release is stimulated remains elusive (5, 6). Here, we investigate the phenomenon of nucleotide secretion in intact perfused mouse medullary thick ascending limb (mTAL) and cortical collecting duct (CCD). The nucleotide secretion was monitored by a biosensor cell placed to register nucleotides in the tubular out-flow. $[Ca^{2+}]_i$ was measured simultaneously in the biosensor cells and the renal tubule with fluo-4. We were able to identify spontaneous tubular nucleotide secretion in resting perfused mTAL. This was seen as lively $[Ca^{2+}]_i$ oscillations in the nucleotide biosensor cells when the tubular outflow fluid engulfed the sensing cells. In mouse mTAL 10 nM AVP and dDAVP induced robust $[Ca^{2+}]_i$ oscillations, whereas AVP in the CCD induced large, slow and transient $[Ca^{2+}]_i$ elevations. Importantly, we identify that AVP/dDAVP triggers tubular secretion of nucleotides in mTAL. After addition of AVP/dDAVP the biosensor cells registered bursts of nucleotides originating from the tubular perfusate. The approximated tubular nucleotide concentration reached peak values of ~0.2-0.3 μM. A very similar response was observed after AVP stimulation of CCDs. Thus, AVP stimulated tubular secretion of nucleotides in a burst like pattern with peak tubular nucleotide concentrations in the low micromolar range. Luminal nucleotides are prone to activate luminal P2 receptors (1) which in turn are well described to inhibit AVP-augmented aquaporin-2-dependent water absorption (7) or ENaC-mediated Na^+ transport (8). Therefore, we speculate that local nucleotide signaling is an intrinsic feed-back element of hormonal control of renal tubular transport (9). J. Med. Invest. 56 Suppl. : 262-263, December, 2009

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