USP10: the nexus between nexin and vasopressin

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FROM THE TIME ELECTROPHYSIOLOGICAL tools became available to renal physiologists, tissues have been bombarded with a variety of hormones and pharmacological compounds to investigate their role in “salt and water transport.” The investigations into the actions of antidiuretic hormone (ADH) on a variety of tissues from frog skin and toad bladder to rabbit kidney tubules have consistently demonstrated the rapid action of ADH (vasopressin) to increase transepithelial sodium transport. Over time these early investigations were confirmed in cell model systems. The translocation of epithelial sodium channels (ENaC) to the apical membrane to increase channel density appears to account, in large part, for the increase in sodium transport. While it has been documented that the cytoskeleton plays a role in this process, the mechanisms remained largely undescribed. There has been a figurative black box linking the binding of vasopressin to its V2 receptor, activation of adenylate cyclase, and production of PKA, with the increase in ENaC density observed at the apical surface of tissues and cells.

In the work presented by Boulkroun and colleagues (1), the lid of the black box has been opened to offer a glimpse into the complexities of the signaling processes involved. The study not only the induction of vasopressin-dependant protein expression but goes on to associate one of the induced proteins to machinery which may regulate ENaC surface density. In this way, the authors uncover the cell’s ability to rapidly alter the expression of proteins in response to external signals and translate that expression into action. A deubiquitinating enzyme (DUB), USP10, was found to be rapidly induced by the application of vasopressin. This component of the study was carried out in a cell line derived from mouse cortical collecting ducts and natively expressing ENaC and its accessory and regulatory proteins. Recently, two other DUBs, UCH-L3 and USP2-45, have been demonstrated to deubiquitinate ENaC in the same cell line (2, 4). As expected, exogenous expression of USP10 with ENaC in a model cell line increased ENaC surface density. However, unlike the action of UCH-L3 and USP2-45, USP10 did not alter ENaC surface density by changing the level of ubiquitination of ENaC itself.

By taking their study a step further, the authors sought to identify the mechanism of action of this vasopressin-induced DUB. Employing a yeast two-hybrid screen, they were able to fish out USP10’s interacting partner from the cellular morass and link the DUB to sorting nexin 3 (SNX3). SNX3 appears to be a target for USP10 while ENaC is not, and it is through SNX3 that ENaC is regulated. What this sets up is a complicated interaction of a DUB with a trafficking protein which regulates ENaC by yet to be defined mechanisms. This results in the increase in ENaC density at the cell membrane and an increase in sodium transport by vasopressin, which was described so many years ago in toads and frogs. Clearly, there are other players in this pathway, other DUBs which act on ENaC directly or on proteins which regulate ENaC, but the lid has been opened. The genomic and proteomic approaches being undertaken in this kind of study give cell physiologists new tools with which to work, to take our understanding of salt and water regulation into a new era. While the web of interacting proteins promises to be extensive, we have the tools to begin identifying the components of these hormonal signaling cascades.

The current work also reinforces the emerging significance of deubiquitination in the regulation of trafficking and expression of membrane proteins such as ion channels and receptors (3). It is well known that ubiquitination of proteins mediates their targeting to proteosomes or lysosomes for degradation, but ubiquitination is a fully reversible posttranslational modification reminiscent in its diversity of phosphorylation. DUBs are a widely diverse family of proteins, actually five families of proteins with over 80 members, which act as proteases to cleave the isopeptide bonds linking ubiquitin to substrate proteins (5). The substrate specificity of some DUBs is known, and in many cases, as with ENaC, more than one DUB may act on the ubiquitinated protein at varying sites in the cell. Moreover, regulatory proteins, such as SNX3 described here, or members of the endosomal sorting complex, or even ubiquitin ligases such as Nedd4-2 itself, are subject to ubiquitination. All these targets clearly represent sites where regulation of substrate, in this case ENaC expression, may occur. A particularly interesting aspect of this study, as with the studies with USP2-45, is the suggestion that regulatory DUBs themselves may be the targets of hormonal regulation. This study offers not only a novel and interesting association of vasopressin-induced proteins and ENaC regulation but also a view of the possibilities this approach affords and a suggestion of what we can expect to see in years to come.

REFERENCES


