Polycystin: From structure to function

Autosomal dominant polycystic kidney disease (ADPKD) is a multisystem disease characterized by the progressive development of numerous fluid-filled cysts in the kidney, leading to chronic renal failure. Predominant tubular epithelial cell abnormalities associated with ADPKD include abnormal maturation, excessive proliferation, and fluid secretion. Since polycystin 1 and polycystin 2 have been cloned, more recent studies have focused on defining the mechanism through which structural alterations in these genes give rise to the tubular epithelial cell abnormalities characteristic of ADPKD. Polycystin 1 has 9 to 11 transmembrane domains, a large amino terminal extracellular tail that appears to be involved in cell-cell and cell-matrix interactions, and a cytoplasmic carboxy terminus that contains a number of motifs that may interact with the cytoskeleton and play an important role in intracellular signaling [1]. In vitro expression studies have shown that the cytoplasmic domain of polycystin 1 contains sites that are phosphorylated by protein kinase A and c-src [2] and may be involved in the protein kinase Cα dependent and c-Jun N-terminal kinase dependent activation of transcription factors, including AP-1 [3].

Despite these advances, the mechanisms underlying excessive tubular epithelial cell proliferation in ADPKD have not been defined. In this issue of Kidney International, Yamaguchi and colleagues provide evidence that cyclic AMP (cAMP) may play a critical role as a second messenger in a signaling pathway leading to tubular epithelial cell proliferation and fluid secretion in ADPKD [4]. In tubular epithelial cells derived from normal human kidney (NHK), agonists of the cAMP signaling system block basal and growth factor-mediated activation of the extracellular signal-regulated protein kinase (ERK) pathway and inhibit proliferation. In contrast, Yamaguchi et al report that agonists of the CAMP signaling system stimulate proliferation of tubular epithelial cells derived from patients with ADPKD. The stimulatory effect of CAMP on proliferation of ADPKD cells was associated with activation of the ERK-pathway. Both protein kinase A (PKA) inhibitors and MEK (mitogen-activated, ERK-activating kinase) inhibitors blocked the proliferative response of ADPKD cells to CAMP. However, receptor tyrosine kinase inhibitors failed to block cAMP-mediated proliferation of tubular cells from ADPKD patients. These studies indicate that CAMP agonists stimulate PKA-mediated activation of ERK, at a locus distal to receptor tyrosine kinase in ADPKD cells, but not in NHK cells.

The basis for this fundamental difference in the proliferative response of ADPKD and NHK cells to CAMP agonists, and how this difference may be related to structural alterations in the polycystin molecules raises a number of questions that need to be addressed by further study:

(1) Is the CAMP-PKA pathway abnormally activated in ADPKD? To demonstrate that the proliferative response of ADPKD cells to CAMP agonists is relevant to our understanding of the pathophysiology of polycystic kidney disease, it will be necessary to show that the CAMP-PKA pathway is indeed activated. There is some indirect evidence in support of this activation. In the murine pcy/pcy model of PKD, cystogenesis is associated with a progressive increase in renal cAMP content [5]. Cysts from human ADPKD patients contain micromolar concentrations of ATP and metabolites of ATP, presumably released from cyst lining epithelial cells [6]. In theory, this concentration of ATP is sufficient to activate renal epithelial cell purinergic receptors, increasing secretion of fluid into the cyst lumen. However, further studies are needed to determine whether intracellular activation of the CAMP-PKA pathway occurs in ADPKD.

(2) If the CAMP-PKA pathway is indeed activated in ADPKD, how does this relate to structural abnormalities in the polycystin 1 and/or polycystin 2 gene products? Cystogenic epithelium in ADPKD is often described as having a “fetal-like” phenotype, characterized by persistent expression of genes expressed during embryogenesis, by an increased rate of proliferation, and by abnormal polarity of ion transporters and growth factor receptors. It is certainly possible that structural abnormalities in the polycystin molecules give rise to a wide variety of defects in cell-cell and/or cell-matrix interactions during embryogenesis, and may thereby alter the pattern of gene expression associated with normal tubular epithelial cell growth and development. The precise mechanism by which these developmental alterations lead to increased CAMP-PKA signaling needs to be defined. Another possibility is that structural defects in and/or aberrant expression of polycystin in tubular epithelial cells...
gives rise to alterations in intracellular cAMP metabolism in ADPKD. Intracellular cyclic nucleotide levels are largely regulated through catabolic pathways mediated by a large family of phosphodiesterases (PDEs) [7]. Even partial inhibition of PDEs may give rise to a manyfold increase in cAMP-PKA signaling. Recent studies, using PDE isoform-specific inhibitors, have demonstrated that mesangial cells contain functionally compartmentalized pools of cAMP that regulate disparate cellular functions, including mitogenesis and reactive oxygen species generation. Activation of the cAMP-PKA pathway in ADPKD may therefore be secondary to a deficiency in PDE production and/or compartmentalization. Even though it may be possible to relate deficiencies in PDE production and/or localization to structural abnormalities in polycystin, it will be more difficult to explain the fundamental difference in cAMP-PKA mitogenic signaling between ADPKD cells and NHK cells, as observed by Yamaguchi and colleagues, on this basis.

(3) Are targets of the cAMP-PKA signaling pathway different in ADPKD versus NHK cells? In many cell types, activation of growth factor receptors by ligand triggers activation of the low molecular weight G protein Ras, with subsequent activation of Raf-1, followed by MEK-1 and ERK. In mesangial cells and many other cell types, cAMP-mediated PKA activation apparently leads to phosphorylation of Raf-1 at an inhibitory site, preventing the activation of MEK-1 [8]. Dr. Yamaguchi and colleagues hypothesize that developmentally acquired or up-regulated proteins in tubular epithelial cells from ADPKD patients circumvent the normal inhibition of Raf-1 by increased levels of cAMP. Rap-1 and/or B-Raf are postulated intermediates in this aberrant cAMP-PKA signaling pathway leading to MEK-1 activation, rather than inhibition, in ADPKD [9]. Additional studies are needed to determine whether Raf-1 and/or B-Raf are differentially expressed or have increased function in ADPKD cells, but not NHK cells. It is possible that polycystin or proteins regulated by polycystin during development may play a critical role in cAMP-PKA signaling by binding and localizing effector molecules near the cell membrane. These potential interactions could be addressed by co-immunoprecipitation analysis.

(4) Can the cAMP-PKA pathway be modulated to prevent cystogenesis in ADPKD? If hyperactivity of the cAMP-PKA pathway triggers susceptible tubular epithelial cells to accelerated proliferation, fluid hypersecretion, and ultimately cystogenesis, it may be possible to inhibit this pathologic response by decreasing intracellular cAMP levels. In vivo transfection of constructs that overexpress isoforms of PDE may be one method to accomplish this modulation. Feasibility of using PDE overexpression to attenuate cAMP-mediated responses has been demonstrated using in vitro systems [10].

The studies by Yamaguchi et al. provide indirect evidence that polycystin may play a key role in the organization and integration of signals that regulate a variety of processes, including proliferation and secretion. Studies to determine whether components of the cAMP-PKA signaling pathway directly interact with polycystin and to determine how these interactions may be altered may provide important insights into the pathophysiology of ADPKD. These studies may provide the basis for targeting the cAMP-PKA pathway through “signal transduction pharmacotherapy” to inhibit tubular epithelial cell mitogenesis and secretion, thereby retarding the rate of cystogenesis in patients with ADPKD.

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REFERENCES