This, too, shall pass—like a kidney stone: a possible path to prophylaxis of nephrolithiasis? Focus on “Cholinergic signaling inhibits oxalate transport by human intestinal T84 cells”

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Nephrolithiasis (kidney stones) has a lifetime prevalence in the developed world of ~5% in females, and up to 12% in males, with recurrence in 50% of cases. Normal human urine is supersaturated with respect to the calcium oxalate ion pair, and roughly two thirds of kidney stones are composed predominantly of calcium oxalate. Oxalate input derives from dietary oxalate absorbed across the gut mucosa and from hepatic oxalate biosynthesis, but is a terminal metabolite that cannot be metabolized except by oxalate or oxalyl-CoA decarboxylases of certain bacteria of the gut lumen (23). The body must match excretion to input to avoid retention of toxic levels of oxalate. This is accomplished by renal glomerular filtration and renal proximal tubular secretion of oxalate into the urine, as well as by intestinal enterocyte secretion of oxalate for fecal excretion.

Elevated urinary excretion of calcium (hypercalcuria) and of oxalate (hyperoxaluria) are independent risk factors for oxalate nephrolithiasis. Calcium oxalate precipitation in the supersaturated urine of hyperoxaluric and/or hypercalciuric patients can be attenuated by maximizing oral fluids and increasing urinary citrate excretion, but strategies to reduce the load of oxalate delivered to the kidney are few. Although hepatic oxalate biosynthetic pathways are known, drugs to block these pathways are not available. Reduction of dietary oxalate intake is difficult to sustain, while dietary oxalate bioavailability and net enteric oxalate absorption are highly variable. Inhibitors of enteric mucosal oxalate influx by still undefined pathways remain unavailable. However, the major oxalate secretory pathway of the enterocyte has been defined by recent discoveries of hyperoxaluria (5, 13) and nephrolithiasis in mice genetically deficient in the apical membrane oxalate/Cl⁻ exchanger Slc26a6 (13) or genetically deficient in the basolateral membrane oxalate/sulfate exchanger Slc26a1 (4). Urolithiasis in these models has been attributed to loss of the major oxalate secretory pathway of the enterocyte, accompanied by reduced fecal oxalate excretion leading to increased plasma oxalate concentration (hyperoxalemia) and, consequently, to increased renal oxalate load reflected by hyperoxaluria (1, 4, 5, 13).

These observations suggest that stimulation of fecal oxalate excretion or metabolism of oxalate in the gut by bacterial oxalate decarboxylases (23) should reduce the risk of nephrolithiasis by reducing urinary oxalate excretion. The efficacy of this latter strategy is under study by dietary supplementation with preparations of Oxalobacter formigenes or other oxalate-degrading bacteria that constitute part of normal gut flora, but are lost in 25% or more of adults (20). Results indicate efficacy in a mouse model of primary hyperoxaluria type I, as well as in wild-type mice (10), but results in humans remain thus far mixed (12, 15, 20).

Enhanced luminal bacterial oxalate degradation should synergize with stimulation of oxalate secretion into the gut by Slc26a6-mediated oxalate/Cl⁻ exchange across the enterocyte luminal membrane. But direct activators of Slc26a6 have not been identified. In the current issue of the American Journal of Physiology-Cell Physiology, Hassan et al. (8) present evidence that suggests the therapeutic possibility of antagonizing physiological downregulation of Slc26a6 to increase enteric oxalate secretion. These investigators reported in 2007 (9) that oxalate uptake into Xenopus oocytes expressing murine Slc26a6 oxalate/Cl⁻ exchanger polypeptide was reduced 90% by the protein kinase C (PKC) activator, phorbol 12-myristate-13-acetate (PMA), and that the reduced oxalate uptake was entirely attributable to reduced oocyte surface expression of Slc26a6. Hassan et al. now demonstrate in polarized filter-grown monolayers of T84 human colon carcinoma cells that DIDS-sensitive oxalate uptake consistent with oxalate/Cl⁻ exchange is mediated by Slc26a6 in the apical membrane (8). The authors’ conclusion is supported by siRNA knockdown experiments in this venerable experimental model of hormone-regulated chloride secretion, until recently considered refractory to multiple techniques of gene transfer. This experiment confirms the central importance of Slc26a6 to transepithelial oxalate secretion by polarized human intestinal cells first demonstrated in Caco-2Bbe1 colon carcinoma cell monolayers by Freeel et al. (6).

Figure 1 presents a cellular model for muscarinic inhibition of oxalate/Cl⁻ exchange by Slc26a6 in T84 cells, integrating the data of Hassan et al. (8). In this model, carbachol stimulation of basolateral (and, surprisingly, apparently apical) M₃ muscarinic receptors works through Gα₁q/11 to activate phospholipase C. The resulting diacylglycerol activates and leads to translocation of PKCβ and (nominally downstream) activation of e-Src. These and undefined, subsequent steps lead to reduced apical membrane abundance of Slc26a6, reflecting either enhanced endocytic internalization and/or slowed exocytic delivery to the apical membrane. The proposed role of PKCβ in this downregulation of Slc26a6 relied in part on the use of rotterlin as a (controversial) kinase inhibitor (8), but the role of PKCα has been substantiated by experiments with kinase-dead PKCδ in Xenopus oocytes (18) and by adenoviral shRNA knockdown in T84 cells for a different ion transporter (22).
Can pharmacological inhibition of physiological muscarinic downregulation of intestinal oxalate secretion actually increase fecal oxalate excretion, and secondarily decrease urinary oxalate excretion? The observed inhibitory effect of carbachol on oxalate uptake is only 35% in T84 cell monolayers. The SLC26A6+/− mouse exhibits normal plasma and urine [oxalate], despite parallel 50% reductions in trans-intestinal epithelial oxalate secretion and mucosal SLC26A6 polypeptide (1), suggesting considerable reserve of intestinal oxalate secretion in the mouse. Part of this reserve might be contributed by other, less active candidate oxalate secretory transporters of the enteric mucosa, SLC26A2 and SLC26A3 (11), both inhibited by PKCδ in Xenopus oocytes (18).

However, as wild-type mice are highly refractory to nephrolithiasis, mutant mice are not a faithful model of the human disorder. One explanation may be the remarkably different apparent affinities for extracellular Cl− of human SLC26A6 (62 mM) and mouse SLC26A6 (8 mM) (2; see Ref. 6 for an alternate view). Since luminal [Cl−] falls to 30 mM in human ileum (22) and to 10 mM in mouse colon (21), human enteric oxalate secretion by SLC26A6 may show greater susceptibility to low luminal [Cl−]. Thus, the common hypofunctional SLC26A6 polymorphism V206M (2, 16) might contribute to idiopathic hyperoxaluria in hypercalciuric individuals (3), although not to the extreme hyperoxaluria of primary hyperoxaluria patients with hereditary hepatic oxalate overproduction (16).

If stimulation of intestinal SLC26A6 activity could decrease urinary oxalate excretion in appropriate clinical settings, are drugs available to do so? The functional predominance of intestinal M3 receptors in vivo remains controversial (7). The rare use of anti-cholinergics in the care of renal stone patients has been limited to the M1 antagonist scopolamine, to relax ureteric hyperperistalsis in the setting of acute obstruction. Nonetheless, M3 antagonists in current clinical use merit consideration. Inhaled M3 blockers such as tiotropium, aclidinium, and glycopyrrolate are used to treat bronchoconstriction and chronic obstructive pulmonary disease (17). The data of Hassan et al. showing activity of apical carbachol on highly polarized T84 cells (8) suggest that the low (2–3%) oral bioavailability of tiotropium might be a virtue for potentially selective anticholinergic stimulation of enteric oxalate secretion. The low gastrointestinal absorption could serve to minimize the potentially serious side-effects of glucose intolerance and central obesity (14, 19).

Hassan and colleagues (12, 13) have shown us that inhibition of physiological muscarinic downregulator signaling can enhance enteric oxalate secretion. Additional downregulatory pathways might be demonstrated and inhibited. Therapy based on this mechanism should redirect the burden of oxalate excretion away from the kidney, which is highly susceptible to oxalate toxicity in the forms of nephrolithiasis and nephrocalcinosis, towards the gut, in which oxalate is better tolerated and can be metabolized. Reducing the urinary burden of oxalate excretion, in conjunction with probiotic luminal bacterial oxalate degradation, may yet soften the sharp bite of the old adage, “This, too, shall pass—like a kidney stone.”

DISCLOSURES
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AUTHOR CONTRIBUTIONS
REFERENCES


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