Cystinuria

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Summary: Cystinuria is an inherited disorder characterized by the impaired reabsorption of cystine in the proximal tubule of the nephron and the gastrointestinal epithelium. The only clinically significant manifestation is recurrent nephrolithiasis secondary to the poor solubility of cystine in urine. Although cystinuria is a relatively common disorder, it accounts for no more than 1% of all urinary tract stones. Thus far, mutations in 2 genes, SLC3A1 and SLC7A9, have been identified as being responsible for most cases of cystinuria by encoding defective subunits of the cystine transporter. With the discovery of mutated genes, the classification of patients with cystinuria has been changed from one based on phenotypes (I, II, III) to one based on the affected genes (I and non–type I; or A and B). Most often this classification can be used without gene sequencing by determining whether the affected individual's parents have abnormal urinary cystine excretion. Clinically, insoluble cystine precipitates into hexagonal crystals that can coalesce into larger, recurrent calculi. Prevention of stone formation is the primary goal of management and includes hydration, dietary restriction of salt and animal protein, urinary alkalinization, and cystine-binding thiol drugs.
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Cystinuria is a genetic disorder of the reabsorption of cystine and dibasic amino acids (ornithine, arginine, and lysine) in the proximal tubule of the nephron and epithelial cells of the gastrointestinal tract. Only impaired cystine transport results in nephrolithiasis, however, because the dibasic amino acids are relatively soluble in urine; an increase in their tubular concentration fails to form crystals in the acidic environment of the distal nephron. Cystinuria was first described by Wollaston[1] in 1810 after he extracted a large bladder stone from one of his patients. At the time, Wollaston[1] believed that the crystals had the chemical properties of an oxide and that the stone had originated from the bladder wall, hence he referred to its composition as *cystic oxide*. Although it later was shown that this particular crystal is neither an oxide nor secreted from the bladder, the amino acid when isolated was named *cystine* in recognition of this historical discovery. In recent years, cystinuria has been attributed to mutations in 2 genes: SLC3A1 (solute carrier family 3A1) and SLC7A9. More than 150 such mutations in these 2 genes have been described, accounting for the defective transport of cystine in individuals with cystinuria.

EPIDEMIOLOGY

Although cystinuria accounts for only 1% of all renal calculi, it is one of the most common inherited genetic diseases. Although not as common as polycystic kidney disease or cystic fibrosis in Caucasians, with prevalences of 1:4,500 and 1:3,000, respectively, the average worldwide prevalence of cystinuria is about 1:7,000. There is significant population variation in the prevalence of cystinuria, ranging from 1:2,500 among Libyan Jews to 1:100,000 persons in Sweden.[2] In the United States, cystinuria occurs in approximately 1:15,000 adults. These figures are likely an underestimate of the true prevalence of cystinuria because many genotypic cystinurics with incomplete

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penetrance fail to develop a stone in their lifetime and therefore escape detection. To underscore this point, the Quebec neonatal screening program reported persistent cystinuria in 562 cases per million infants screened, which is 7 times higher than the number of clinically diagnosed cystinurics in the adult population of Quebec.[3]

PHENOTYPIC CLASSIFICATION

The original classification system of cystinuria was based on phenotypic expression because it was developed long before the genetic defects of the disorder were identified. Three phenotypic subtypes of cystinuria emerged that were defined by the amount of urinary cystine excretion of the patient's parents (obligate heterozygotes for the dysfunctional genes).[4] Patients with type I cystinuria, an autosomal-recessive disorder, have I/N parents (N for normal allele) who both are silent carriers of the disease because they excrete cystine in the normal adult range (0-100 μmoles of cystine per gram of creatinine). In addition, type I patients show no increase in plasma cystine levels after an oral cystine load, suggesting intestinal uptake also is defective.

Both parents (obligate heterozygotes) of patients of what was called type II and type III cystinuria have markedly increased amounts of urinary cystine excretion (>900 μmoles of cystine per gram of creatinine in type II parents, 100-900 μmoles of cystine per gram of creatinine in type III parents, with some overlap seen). Unlike their type I/N counterparts, type II/N parents will on rare occasion form cystine stones given their increased levels of cystine excretion. They can therefore be considered to have an autosomal-dominant disorder, or one that is incompletely recessive. Intestinal cystine transport does occur in patients with these subtypes, unlike in type I cystinurics, although at a reduced rate compared with normal controls.

As the identification of affected genes emerged, it became clear that the historical classification scheme lacked utility. Mutations in only 2 genes, SLC3A1 and SLC7A9, have been held responsible for all 3 phenotypic subtypes of cystinuria. To date, all mutations of SLC3A1 confer a type I phenotype whereas mutations in SLC7A9 can cause all 3 phenotypic subtypes.[5] Dello Strologo et al[6] showed that 14% of defective SLC7A9 homozygotes had parents with normal urinary cystine excretion. Under the phenotypic classification scheme, these patients would be classified as phenotypical type I. In an effort to incorporate what is now known about the culprit genes, Dello Strologo et al[6] proposed a genotypic classification scheme based on the gene affected by the mutation. In this new classification system, type A cystinuria is caused by mutations in both inherited alleles of SLC3A1 and type B cystinuria is the result of 2 mutations in the inherited alleles of SLC7A9. Type A cystinurics have a recessive disorder and therefore would correspond to type I cystinuria under the old classification system. Type B would include types II (incompletely dominant) and III; in some cases a mutation with mild consequences has even accounted for a type I phenotype in the heterozygote parents. Occasionally investigators use a nomenclature that instead includes types I and non–type I patients. The variables that account for the difference in cystine excretion between heterozygotes and homozygotes with different mutations in SLC7A9 are not yet known. The A/B classification system also allows for the classification of cystinurics who have inherited one mutation in SLC3A1 from one parent and one mutation in SLC7A9 from the other parent. These patients would be referred to as *type AB*. Using this classification system, the International Cystinuria Consortium (ICC) has reported that type A (MIM 220100), type B (MIM 600918), and type AB account for 38%, 47%, and 14% of cystinurics in their registry, respectively.[7]

GENETICS

The first defective gene known to cause cystinuria was identified in 1994 by Calonge et al.[8] By using linkage analysis, SLC3A1 was located on the short arm of chromosome 2 (2p16.3-21). SLC3A1 encodes "related to $b^{0,+}$ amino acid transporter" (rBAT) and the mutation of this gene exclusively manifests in the type A phenotype. The characteristics of the cystine transporter include those of a transporter studied in

mouse blastocysts (b), which transport neutral (0) and dibasic (+) amino acid transport (denoted by the superscripts).

Normally, the SLC3A1 gene product is translated into a 685-amino acid heavy subunit of the cystine transporter with 4 transmembrane domains.[9] To date, 103 SLC3A1-specific mutations have been identified, including chromosomal rearrangements, deletions and insertions, and missense, nonsense, and frameshift mutations.[10] The most common mutation of SLC3A1 is M467T, which codes for amino acid threonine instead of methionine at position 467. This mutation accounts for 26.4% of cystinuria cases worldwide.[10] To emphasize the population-specific nature of these mutations, M467T accounts for 40% of abnormal genes in Mediterranean patients with cystinuria, whereas this mutation is relatively rare in Quebec.[11,12]

After the discovery of SLC3A1, it was noted that non–type I patients with cystinuria possessed wild-type sequences of the SLC3A1 gene. This implied that another gene product must be defective in type B patients with cystinuria. In 1999, the ICC, again using linkage analysis, was able to identify a second cystinuria locus, SLC7A9, on the long arm of chromosome 19 (19q12-13).[13] The normal SLC7A9 gene encodes $b^{0,+}$ AT, a 487-amino acid light subunit of the cystine transporter with 12 membrane-spanning domains. More than 60 mutations of SLC7A9 have been identified to date.[10] Worldwide, the most common mutation of SLC7A9 is G105R, a substitution of arginine for glycine at position 105. According to the ICC registry, G105R accounts for 27.4% of non–type I mutations.[10] As with SLC3A1, population variation is considerable with SLC7A9 mutations. In Libyan Jews, the most common mutation is V170M, not G105R, in which a substitution of valine for methionine occurs at position 170.

PATHOPHYSIOLOGY

In normal individuals, greater than 99% of the amino acids, including cystine, filtered by the glomerulus are reabsorbed by the end of the proximal tubule. However, in cystinuria, the fractional excretion of cystine can increase to greater than 100% of the filtered load, implying some tubular cystine secretion. Dent et al[14] and Palacin et al[15] were the first to hypothesize that cystinuria was the result of a defective amino acid transport system. Subsequently, 2 transport systems appear to be responsible for cystine reabsorption (Fig. 1).[2,15,16] The high-affinity–low-capacity system mediates the uptake of cystine, as well as the dibasic amino acids, and is found almost exclusively in the apical membrane of the third segment (S3) of the proximal tubule. Approximately 10% of the filtered cystine is reabsorbed through this transporter. The S3 location of the high-affinity–low-capacity system coincides with the location of the majority of the gene product of SLC3A1, the rBAT protein.

The bulk of cystine reabsorption, about 90% of the filtered load, occurs in the S1 and S2 segments of the proximal tubule through a low-affinity–high-capacity transport system.[17] The $b^{0,+}$AT protein, encoded by SLC7A9, is found in all 3 segments of the proximal tubule with a gradient of expression highest in S1 where the low-affinity–high-capacity transporter resides to lesser amounts in S2 and then S3 segments. Given the relatively proximal location of $b^{0,+}$AT expression, some investigators in the past had implied its possible role in low-affinity–high-capacity transport; however, it has been shown that $b^{0,+}$AT associates exclusively with rBAT. Their coexpression results in high-affinity cystine reabsorption as in the S3 segment of the proximal tubule.[16,18] Thus far, the low-affinity–high-capacity system has yet to be characterized and its identity remains unknown. Of note, rBAT does not exclusively form heterodimers with $b^{0,+}$AT, a fact that is suggestive of the existence of a yet unidentified light subunit that binds to rBAT. The creation of SCL7A9 knockout mice may help further elucidate the roles of both gene products in the high- and low-affinity transport systems.[19]

The relationship between transport system and gene product may shed some light on the phenotypic expression seen in some heterozygotes.[15] SLC3A1 heterozygotes (type I/N or A/N), with loss of half the complement of normal rBAT, have a defect in the high-affinity–low-capacity system, which means they still are able to reabsorb 90% of the filtered cystine load by

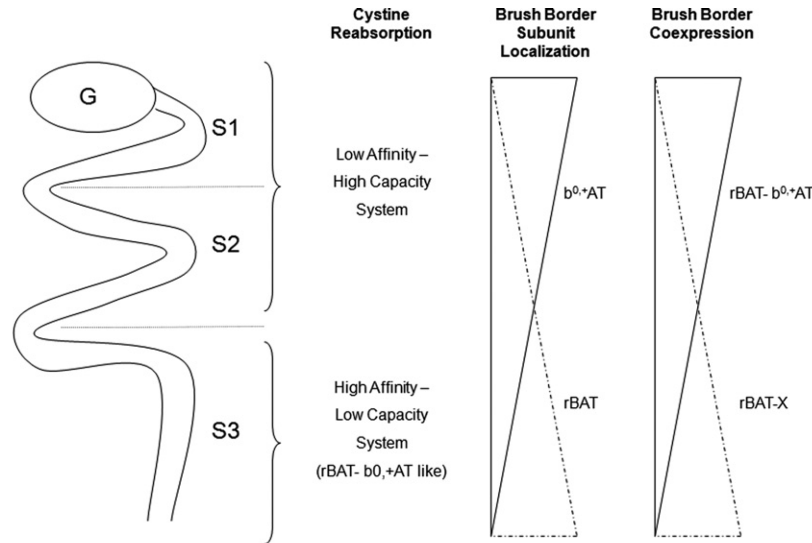

Figure 1. Proposed cystine transport systems. The high-affinity–low-capacity system is localized primarily to the S3 segment of the renal proximal tubule whereas the low-affinity–high-capacity system is found in the S1 and S2 segments. The SLC7A9 gene product $b^{0,+}$AT is expressed in all 3 segments of the proximal tubule, with predominant expression in the S1 and S2 segments. Its expression in the apical membrane occurs only where rBAT is expressed. The SLC3A1 gene product rBAT is expressed almost exclusively in the S3 segment of the proximal tubule. It has been shown that tubular cell coexpression of rBAT and $b^{0,+}$AT results in high-affinity cystine transport. However, the inverse gradient of expression of the 2 gene products is suggestive of independent roles as well that have yet to be characterized. Attention has been called to the possibility that rBAT binds to an as yet unidentified light component, given that rBAT is not coexpressed exclusively with $b^{0,+}$AT. However, no gene other than SLC3A1 and SLC7A9 has been identified as causing cystinuria so it is possible that rBAT simply is overexpressed in S3 and not heterodimerizing with an unidentified light component. G, glomerulus; X, unidentified light subunit. Data from Palacin et al[15] and Fernandez et al.[16]

the more proximal low-affinity–high-capacity system and as a result can stay in the normal range of cystine excretion. Conversely, defects in SLC7A9 gene product affect the high-affinity–low-capacity and possibly the low-affinity–high-capacity systems. These heterozygotes (B/N), with the loss of half the normal complement of $b^{0,+}$AT, lose their ability to reabsorb filtered cystine throughout the tubular lumen. Their excretion of cystine is much higher than A/N heterozygotes and usually is above the normal range of cystine excretion. Some of these heterozygotes rarely may have stones, constituting a dominant, or incompletely recessive, phenotype.

These descriptions correlate well with observations in the corresponding knockout mouse models. Animals heterozygote for mutation in SLC7A9 have abnormal urine cystine excretion but do not have cystine stones, whereas heterozygotes for mutations in SLC3A1 do not have increases in urinary cystine excretion.[20,21]

TRANSCELLULAR CYSTINE TRANSPORT

The transport of cystine and dibasic amino acids occurs across the apical membrane of the proximal renal tubule cell in a sodium-independent manner unlike other amino acids and solutes such as glucose (Fig. 2). Sodium-dependent neutral amino acid transporters present at the apical and basolateral membranes maintain a membrane potential that favors the transport of cystine and the dibasic amino acids into the cell. Intracellular cystine is reduced to 2 molecules of cysteine that then exit the cell across the basolateral membrane. The metabolism of cystine within the tubular cell helps to maintain a favorable concentration gradient across the apical membrane.

The cystine transporter is a heterodimer composed of 2 subunits linked by a disulfide bridge.[22] The heavy subunit rBAT is the glycosylated protein produced by the SLC3A1 gene sequence whereas the light subunit $b^{0,+}$AT is encoded by SLC7A9. Although the heterodimer

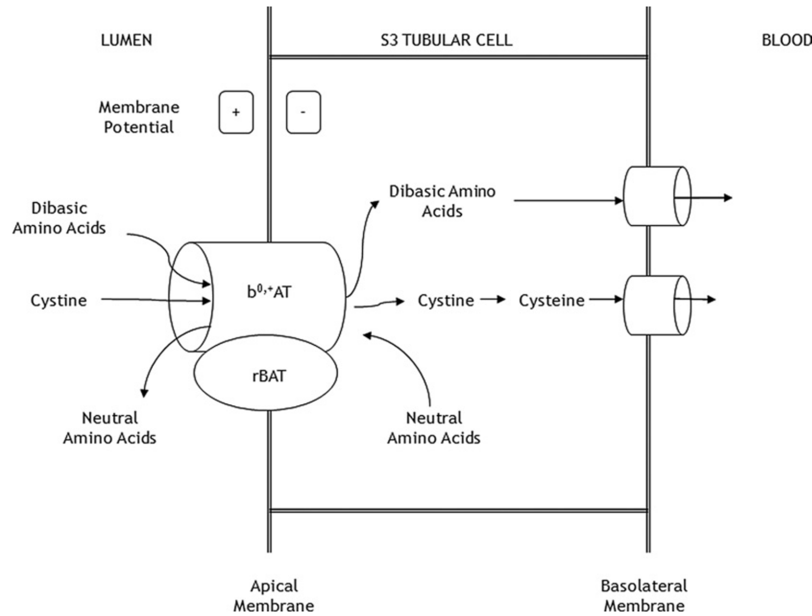

Figure 2. Normal cystine and dibasic amino acid transport in the S3 segment of the proximal tubule. After cystine is filtered across the glomerular capillary membrane into the lumen of the proximal tubule it is reabsorbed across the apical membrane of the S3 segment of the proximal tubule. This transport is mediated by the amino acid transporter that is affected in cystinuria. The transporter is a heterodimer composed of 2 subunits, the protein products of SLC3A1 (rBAT, the heavy component) and SLC7A9 ($b^{0,+}AT$, the light component). Sodium-dependent neutral amino acid transporters on the luminal and basolateral sides of the cell maintain a membrane potential that favors cystine and dibasic amino acid transport into the cell from the proximal tubule (not shown). Cystine and dibasic amino acid reabsorption is accompanied by an efflux of neutral amino acids with a stoichiometry of 1:1. Inside the proximal tubule cell, cystine is reduced to 2 molecules of cysteine, which exit across the basolateral membrane.

is the functional unit, in vivo the heterodimers oligomerize into functioning tetramers.[23] Other amino acid transporters are constructed similarly with SLC3- and SLC7-family components, constituting the class of heteromeric amino acid transporters (HATS). The SLC7A9 gene product $b^{0,+}AT$ has been proposed to be the catalytic (ie, transporting) subunit. It confers specificity as to which amino acids the protein transports. The heterodimer mediates the electrogenic uptake of cystine and dibasic amino acids in exchange for efflux of neutral amino acids in a stoichiometry of 1:1.[24] The role of rBAT in cystine transport has been elucidated by Palacin et al,[15] who reported that rBAT is required to bring $b^{0,+}AT$ to the apical cell membrane. In the absence of $b^{0,+}AT$, rBAT remains confined to the endoplasmic reticulum after its synthesis and is not glycosylated properly.[13] Reconstitution of $b^{0,+}$ AT in liposomes show that it is a functional cystine transporter in the absence of rBAT. rBAT may have a modulatory role on $b^{0,+}$ AT activity.

The trafficking function of rBAT may explain the phenotypic presentation of heterozygotes. A/N, heterozygotes, have normal catalytic subunits while having a reduced number of chaperone subunits. However, overexpression of rBAT in comparison with $b^{0,+}$ AT implies that these heterozygotes may have normal functioning cystine transporters at the apical membrane.[10] On the other hand, B/N heterozygotes have functioning chaperone subunits but defective catalytic subunits. Although the transporters are reaching the apical membrane, half of them are not able to reabsorb cystine effectively, thereby increasing the amount of cystine excreted.

The gastrointestinal implications of impaired absorption fail to manifest themselves clinically because serum cystine levels are comparable with normal people. Adequate cystine absorption is maintained by the ability of the small intestine to absorb oligopeptides containing cystine. The metabolism of methionine to cystine also helps to maintain normal plasma cystine availability.

CLINICAL MANIFESTATIONS

Although cystine stones account for 1% of all renal calculi, cystinuria is notable for the frequency of recurrence and the large size of the stones. In the pediatric population, cystine stones account for 6% to 10% of all cases of nephrolithiasis.[25] Furthermore, it has been shown that 50% of type A patients with cystinuria will form a stone in the first decade of life and another 25% form stones in their teenage years.[26] Differences in the heterogeneity of age of onset of stones have not been explained. The risk of stone formation is high in the pediatric age group with 82.9% of symptomatic patients experiencing a stone in the first 2 decades of life.[6] Poorly managed patients have a risk of forming one new stone per patient-year and requiring one urologic procedure every 3 years.[27] Dello Strologo et al[6] showed that stone recurrence has a predilection for males, with one new stone occurring every 3 years versus females who experience stone formation every 5 years. Sex did not play a role, however, in the age of onset according to this study. To further risk stratify, Dello Strologo et al[6] showed that the rate of recurrent stone formation was the same in type A and B cystinuria. A lower incidence of stone formation has been shown only in type AB cystinurics compared with types A and B.[24]

The only clinically significant manifestations of cystinuria are related to stone formation. Stones may present with nausea, flank pain, hematuria, and recurrent urinary tract infections. In rare but extreme circumstances, cystinuria can progress to acute or chronic renal failure requiring dialysis. In one study, as many as 17% of patients with cystinuria had evidence of a decreased glomerular filtration rate.[6] Compared with a calcium oxalate stone-forming cohort, cystinurics were almost 5 times as likely to have undergone a nephrectomy.[28]

Cystine crystals have a classic flat hexagonal appearance in urine specimens of patients with cystinuria (see the article by Asplin in this issue, p. 99). The presence or absence of crystalluria and the volume of cystine crystals correlate with stone activity and predict stone recurrence.[29] The stones formed are usually pale yellow in color with a waxy texture (Fig. 3).

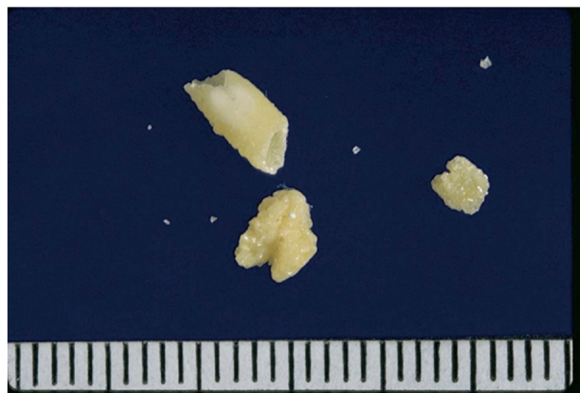

Figure 3. Cystine stones. The hollow, cylindric structure at upper left was voided spontaneously by a patient 24 hours after the removal of a ureteral stent.

Cystine stones frequently coalesce to form large branching staghorn calculi. Renal tissue has been obtained from patients who have undergone urologic interventions to remove stones.[30] These biopsies reveal that the ducts of Bellini are plugged with cystine crystals. The ductal lining often is denuded with inflammatory and fibrotic changes of the surrounding interstitium. Many intermedullary collecting ducts are dilated with intraluminal hydroxyapatite (calcium phosphate) crystals. The specific pathophysiologic role of these apatite components is uncertain. Over time, the cycle of inflammation and fibrosis can lead to nephron dysfunction and chronic kidney disease as evidenced by an increased serum creatinine concentration in 5.8% of patients with cystinuria.[28]

DIAGNOSIS

All patients with an episode of nephrolithiasis before the age of 30 or a strong family history of recurrent nephrolithiasis should be screened for cystinuria. Urinalysis, the initial test of choice, may reveal the presence of the pathognomonic hexagonal cystine crystals, seen in 25% of pediatric patients with cystinuria.[31] A rapid qualitative screening test with sodium cyanide-nitroprusside is used commonly to diagnose cystinuria. Cyanide reduces cystine to cysteine, which then binds nitroprusside causing a reddish/purplish color change. Sodium cyanide-nitroprusside can detect cystine levels of 75 mg/L or greater, with a sensitivity of 72% and a specificity of 95%.[32] False positives have

been seen in patients with Fanconi syndrome, homocystinuria, and acetonuria, or patients taking sulfa drugs, ampicillin, or N-acetylcysteine. This test also may detect children who are heterozygotes for SLC7A9. In the first 1 to 2 years of life, the renal tubules continue to mature and cystine transport is suboptimal, so these children may excrete cystine at the homozygote level.[33] Therefore, it is preferred that the test be performed at 2 years of age or older, when heterozygotes excrete cystine at levels comparable with their adult counterparts. A 24-hour urine collection often is required to confirm the diagnosis. The normal rate of cystine excretion is 30 mg/d. Homozygotes usually excrete more than 300 mg/d of cystine; heterozygotes excrete either normal amounts (A/N) or abnormal, if intermediate, amounts (B/N).

Imaging can be used to visualize cystine stones, although it is more helpful in guiding treatment than diagnosing the disorder. On plain films, cystine stones are radiopaque, if variably so, because of the density provided by their disulfide bond; however, they often are less dense than calcium stones. Helical computed tomography (CT) is more sensitive and may be the test of choice in patients with high stone burdens because it is difficult to delineate multiple stones on plain films. Renal ultrasound is another effective imaging modality. Although less sensitive than CT, renal ultrasound has the benefits of lower cost and avoidance of repeated radiation exposure.

TREATMENT

There have been few advances in the management of cystinuria over the past 15 years. Stone prevention is accomplished using methods aimed at reducing the absolute amount of cystine excreted in the urine and maintaining the large amount of excreted cystine in a soluble state. Typically, the initial medical management involves increasing oral fluid intake to decrease urine cystine concentration to less than 250 mg/L (about 1 mmol/L), with at least 3 L/d of urine output. This requires oral intake of more than 3 to 4 L/d. Cystine concentrations greater than 250 mg/L increase the risk of stone formation as the saturation point of urine is approached. One study showed that hyperdiuresis could reduce the formation of stones in two thirds of patients with cystinuria.[14] Maintaining high flow rates through all 24 hours each day should be emphasized. This includes consuming large fluid volumes at bedtime to decrease nocturnal aggregation of crystals.

The pH of urine also will alter the absolute level of saturation with increasing cystine solubility as pH increases. However, until urinary pH is 7.0 or greater, the effect of pH on solubility is minimal.[34] Dent et al[14] showed that the solubility of cystine in urine is approximately 250 mg/L up to a pH level of 7.0, but solubility of cystine increases to 500 mg/L or more with a pH level of 7.5 or greater. Therefore, another approach of medical management is to increase cystine solubility through urinary alkalinization using supplementation with an oral base.[23] Potassium citrate, sodium citrate, and sodium bicarbonate are used commonly. Citrate is an organic anion and its metabolism by the liver consumes protons and generates bicarbonate. This results in an overall reduction in renal proton excretion and bicarbonaturia that effectively alkalinizes the urine. The potassium salt formulation is preferred to sodium salts because sodium restriction reduces the amount of cystine excreted in the urine.[35,36] Although the transport mechanism by which this occurs is unclear, higher sodium intake correlates with higher cystine excretion so that sodium restriction is advised.[37,38]

A reduction in dietary consumption of cystine and methionine (the amino acid precursor for cystine) by limiting animal protein products has been recommended in an effort to reduce the total cystine load filtered and excreted by the nephron. Restriction of animal protein intake does reduce cystine excretion acutely.[39] Higher urinary urea nitrogen excretion, a surrogate of animal protein ingestion, correlates with higher cystine excretion.[38] However, even if all dietary amino acid intake is eliminated, cystine is produced endogenously through normal metabolic activity. Therefore, there is always some baseline glomerular filtration of cystine presenting to the proximal tubule for reabsorption. The restriction of animal protein, the major dietary source of protons, also serves to decrease renal proton excretion, increasing

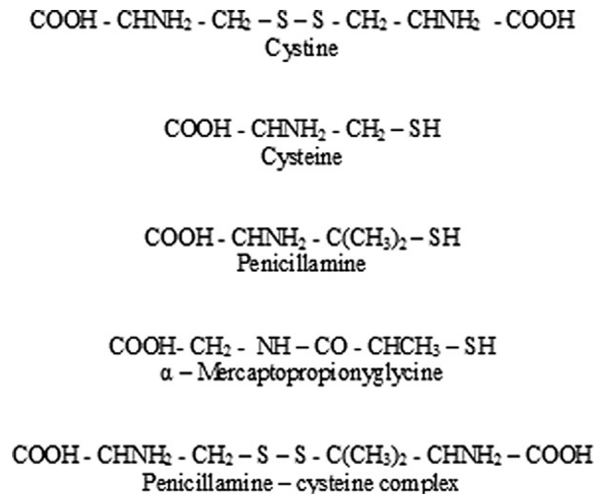

Figure 4. Chemical structures of cystine, cysteine, cystine-binding thiol agents, and a thiol-cysteine complex. The 2 thiol (-SH) containing drugs used for the treatment of cystinuria are penicillamine and α-MPG or tiopronin. Both agents reduce cystine's disulfide bond, creating the more soluble cysteine-drug complex. Only the penicillamine-cysteine complex is shown here.

urine pH or reducing the need for exogenous base intake. Furthermore, a reduction in animal protein consumption often is accompanied by an increase in fruit and vegetable ingestion, which are high in potential base in the form of organic anions. This too will lead to the alkalinization of urine by reducing renal proton excretion and increasing bicarbonaturia. Despite animal protein restriction being associated with higher urine pH and lower cystine excretion, the efficacy of dietary protein restriction to prevent stone recurrence has never been shown in a randomized controlled trial.

CYSTINE BINDING THIOL DRUGS

If patients are refractory to the medical management mentioned thus far (hydration, alkalinization, and dietary sodium and animal protein restriction), thiol-containing agents can be used. The 2 agents commonly used are d-penicillamine and α-mercaptopropionylglycine (α-MPG), also known as *tiopronin*. These drugs work by reducing the disulfide bond in cystine, yielding 2 molecules of cysteine. The thiol group of the drug combines with cysteine to form a soluble cysteine-drug product (Fig. 4). The cysteine-penicillamine complex is up to 50 times more soluble than cystine.[40] Side effects, similar for both agents, limit their use. The reported adverse reactions include allergy, nausea, vomiting, fever, rash, diarrhea, arthralgias, leukopenia, thrombocytopenia, proteinuria (secondary to membranous nephropathy), and systemic lupus erythematosus–like syndromes. Penicillamine has been shown to be somewhat more culpable, which is why α-MPG is the more commonly used agent. In one study, discontinuation of therapy secondary to adverse reactions was as high as 69.4% in the penicillamine group versus 30.6% in the α-MPG group.[41] Captopril, an angiotensin-converting enzyme inhibitor, contains a thiol group that has been shown to increase cystine solubility in vitro. However, studies of efficacy in patients with cystinuria with captopril have been equivocal and variable.[42,43] Captopril excretion in the urine may not be adequate to cause meaningful changes in cystine supersaturation.[38] It may, however, given its potential effect, constitute a preferred agent for the management of hypertension in cystinuria.

D-penicillamine use in the SLC7A9 knockout mouse is associated with a significant reduction in cystine excretion and stone mass measured by radiograph densitometry of kidneys.[19] This animal model may serve as an important means of evaluating the efficacy of drug therapy for the treatment of cystinuria.

Many artifacts in measuring cystine excretion have prevented the measurement of cystine excretion per se from being clinically useful. More acidic urine leads to cystine precipitation and failure to accurately measure the excreted amino acid. One way to circumvent this problem is to be sure to alkalinize the collected urine adequately after voiding to ensure measurement of all excreted cystine.[44] Another problem is that most assays fail to distinguish between cystine and cysteine-drug complex when thiol drugs are used. The use of a solid-phase assay for cystine supersaturation can distinguish effectively between cystine and the cysteine-drug complex to guide treatment choice and drug dosing more effectively.[38,45] Cystine supersaturation decreases as expected in the presence of thiol drugs.[46] Such measurements may represent an important improve-

ment in care because the technique has become commercially available recently, although it has not been shown prospectively to better correlate with stone activity than more conventional tests yet.

Overall, using aggressive medical management, including hyperdiuresis, oral alkalinizing agents, and cystine-reducing thiol agents, Barbey et al[27] were able to decrease the incidence of stone formation by 78% and reduce the need for urologic procedures by 52% for patients with cystinuria enrolled in this study. However, complying with medical management recommendations can be difficult. Pietrow et al[47] showed that only 15% of medically managed patients were able to maintain therapeutic success as defined by urine cystine concentrations of less than 300 mg/L.

Decisions regarding surgical intervention largely are dependent on the size and location of a stone in the urinary tract, regardless of composition. Stones that are larger than 5 mm spontaneously pass in less than 50% of cases and staghorn caliculi will not pass without urologic intervention. Extracorporeal shockwave lithotripsy (ESWL), popular and widespread in the treatment of calcium stones, has been effective for some cystine stones less than 1.5 cm in diameter.[48,49] However, many cystine stones, because of their homogeneous molecular structure, are denser and less easy to fragment by ESWL and should be treated with ureteroscopy or percutaneous nephrostolithotomy.[50] Patients' prior experience with ESWL should guide selection of urologic modality. For cystine stones larger than 1 cm in diameter that have failed ESWL and retrograde uteroscopy, percutaneous nephrostolithotomy is the standard of care. Some investigators have distinguished between rough and smooth cystine stones, suggesting that the former may be more susceptible to fragmentation with ESWL.[51] If a rough versus a smooth texture can be distinguished reliably by helical CT scanning, the choice of urologic intervention may be made more rationally in the future.

CONCLUSIONS

As techniques in molecular genetics and biology evolve, so does our understanding of this fascinating subset of nephrolithiasis. Cystine stones have been described for nearly 200 years, but it is only in the past decade that we have begun to understand the genetic mutations and transport defects involved. Our understanding is not complete, however, because researchers have not been able to identify culprit gene mutations in some patients. This suggests that there are mutations in unidentified genes coding for cystine transport systems or for inhibitors or promoters of cystine crystallization. Interestingly, even with better descriptions of the pathophysiology of cystinuria, new treatment options have not emerged. Medical and surgical management have been largely unchanged over the past decade. Hydration and alkalinization followed by thiol-reducing agents are the mainstays of preventative care. With the identification of the responsible genes, however, targeted gene delivery to renal tubule cells makes gene therapy a promising alternative management strategy in the future.

REFERENCES

1. Wollaston WH. On cystic oxide: a new species of urinary calculus. Philos Trans R Soc Lond. 1810;100:223-30.
2. Palacin M. Cystinuria. In: Scriver CR, Beaudet AL, Sly WS, et al, editors. The molecular and metabolic bases of inherited disease. New York: McGraw-Hill; 2001. p. 4909-32.
3. Scriver CR, Clow CL, Reade T, et al. Ontogeny modifies expression of cystinuria genes: implications for counseling. J Pediatr. 1985;106:411-6.
4. Rosenberg LE, Downing S, Durant JL, et al. Cystinuria: biochemical evidence for three genetically distinct diseases. J Clin Invest. 1966;45:365-71.
5. Leclerc D, Boutros M, Suf D, et al. SLC7A9 mutations in all three cystinuria subtypes. Kidney Int. 2002;62:1550-9.
6. Dello Strologo L, Pras E, Pontesilli C, et al. Comparison between SLC3A1 and SLC7A9 cystinuria patients and carriers: a need for new classification. J Am Soc Nephrol. 2002;13:2547-53.
7. Rogers A, Kalakish S, Desai RA, et al. Management of cystinuria. Urol Clin North Am. 2007;34:347-62.
8. Calonge MJ, Gasparini P, Chillaron J, et al. Cystinuria caused by mutations in rBAT, a gene involved in the transport of cystine. Nat Genet. 1994;6:420-5.
9. Goodyer P, Boutros M, Rozen R. The molecular basis of cystinuria: an update. Exp Nephrol. 2000;8:123-7.
10. Font-Llitjos M, Jiminez-Vidal M, Bisceglia L, et al. New insights into cystinuria: 40 new mutations, genotype-

phenotype correlation, and digenic inheritance causing partial phenotype. J Med Genet. 2005;42:58-68.
11. Saadi I, Chen XZ, Hediger M, et al. Molecular genetics of cystinuria: mutation analysis of SLC3A1 and evidence for another gene in the type I (silent) phenotype. Kidney Int. 1998;54:48-55.
12. Horsford J, Saadi I, Raelson J, et al. Molecular genetics of cystinuria in French-Canadians: identification of 4 novel mutations in type I patients. Kidney Int. 1996; 49:1401-6.
13. Feliubadalo L, Font M, Purroy J, et al. Non-type I cystinuria caused by mutations in SLC7A9, encoding a subunit ($b^{0,+}$AT) of rBAT. Nat Genet. 1999; 23:52-7.
14. Dent CE, Friedman M, Green H, et al. Treatment of cystinuria. BMJ. 1965;5432:403-8.
15. Palacin M, Fernandez E, Chillaron J, et al. The amino acid transport system $b^{0,+}$ and cystinuria. Mol Membr Biol. 2001;18:21-6.
16. Fernandez E, Carrascal M, Rousaud F, et al. rBAT-b(0,+) AT heterodimer is the main apical reabsorption system for cystine in the kidney. Am J Physiol. 2002;283:F540-8.
17. Silbernagl S. The renal handling of amino acids and oligopeptides. Physiol Rev. 1988;68:911-1007.
18. Chairoungdua A, Segawa H, Kim J, et al. Identification of an amino acid transporter associated with the cystinuria-related type II membrane glycoprotein. J Biol Chem. 1999;274:28845-8.
19. Font-Llitjos M, Feliubadalo L, Espino M, et al. SLC7A9 knockout mouse is a good cystinuria model for antilithiasic pharmacological studies. Am J Physiol. 2007; 293:F732-40.
20. Feliubadalo L, Arbones ML, Manas S, et al. SLC7A9-deficient mice develop cystinuria non-I and cystine urolithiasis. Hum Mol Genet. 2003;12:2097-108.
21. Peters T, Thaete C, Wolf S, et al. A mouse model for cystinuria type I. Hum Mol Genet. 2003;12: 2109-20.
22. Verrey F, Jack DL, Paulsen I, et al. New glycoprotein-associated amino acid transporters. J Membr Biol. 1999;172:181-92.
23. Fernandez E, Jimenez-Vidal M, Calvo M, et al. The structural and functional units of heteromeric amino acid transporters. J Biol Chem. 2006;281:26552-61.
24. Chillaron J, Estevez R, Mora C, et al. Obligatory amino acid exchange via systems $b^{0,+}$AT-like and y^{+}L-like. J Biol Chem. 1996;271:17761-70.
25. Milliner DS, Murphy ME. Urolithiasis in pediatric patients. Mayo Clin Proc. 1993;68:241-8.
26. Goodyer P, Saadi I, Ong P, et al. Cystinuria subtype and the risk of nephrolithiasis. Kidney Int. 1998;54: 56-61.
27. Barbey F, Joly D, Rieu P, et al. Medical treatment of cystinuria: critical reappraisal of long term results. J Urol. 2000;163:1419-23.
28. Assimos DG, Leslie SW, Ng C, et al. The impact of cystinuria on renal function. J Urol. 2002;168: 27-30.
29. Daudon M, Cohen-Solal F, Barbey F, et al. Cystine crystal volume determination: a useful tool in the management of cystinuric patients. Urol Res. 2003; 31:207-11.
30. Evan AP, Coe FL, Shao Y, et al. Renal crystal deposits and histopathology in patients with cystine stones. Kidney Int. 2006;69:2227-35.
31. Thomas JC, DeMarco RT, Donohoe JM, et al. Pediatric ureteroscopic stone management. J Urol. 2005;174: 1072-4.
32. Finocchiaro R, D'Eufemia P, Celli M, et al. Usefulness of cyanide nitroprusside test in detecting incomplete recessive heterozygotes for cystinuria: a standardized dilution procedure. Urol Res. 1998;26:401-5.
33. Boutros M, Vicanek C, Rozen R, et al. Transient neonatal cystinuria. Kidney Int. 2005;67:4438.
34. Nakagawa Y, Coe FL. A modified cyanide nitroprusside method for quantifying urinary cystine concentration that corrects for creatinine inference. Clin Chim Acta. 1999;289:57-68.
35. Pak CY, Fuller CJ. Assessment of cystine solubility in urine and of heterogeneous nucleation. J Urol. 1983; 129:1066-70.
36. Jaegar P, Portmann L, Saunders A, et al. Anticystinuric effects of glutamine and of dietary sodium restriction. N Engl J Med. 1986;315:1120-3.
37. Rodriguez LM, Santos F, Malaga S, et al. Effect of low sodium diet on urinary elimination of cystine in cystinuric children. Nephron. 1995;71:416-8.
38. Goldfarb DS, Coe FL, Asplin JR. Urinary cystine excretion and capacity in patients with cystinuria. Kidney Int. 2006;69:1041-7.
39. Rodman JS, Blackburn P, Williams JJ, et al. The effect of dietary protein on cystine excretion in patients with cystinuria. Clin Nephrol. 1984;22:273-8.
40. Lotz M, Bartter FC. Stone dissolution with D-penicillamine in cystinuria. BMJ. 1965;5475:1408-9.
41. Pak CY, Fuller C, Sakhaee K, et al. Management of cystine nephrolithiasis with alpha-mercaptopropionylglycine. J Urol. 1986;136:1003-8.
42. Sloand JA, Izzo JL. Captopril reduces urinary cystine excretion in cystinuria. Arch Intern Med. 1987;147: 1409-12.
43. Chow GK, Streem SB. Medical treatment of cystinuria: results of contemporary clinical practice. J Urol. 1996;156:1576-8.
44. Nakagawa Y, Asplin J, Goldfarb DS, et al. Clinical use of cystine supersaturation measurements. J Urol. 2000;164:1481-85.
45. Coe FL, Clark C, Parks JH, et al. Solid phase assay of urine cystine supersaturation in the presence of cystine binding drugs. J Urol. 2001;166:688-93.
46. Dolin DJ, Asplin JR, Flagel L, et al. Effect of cystine-binding thiol drugs on urinary cystine capacity in patients with cystinuria. J Endourol. 2005;19: 429-32.
47. Pietrow PK, Auge BK, Weitzer AZ, et al. Durability of the medical management of cystinuria. J Urol. 2003; 169:68-70.

48. Chow GK, Streem SB. Contemporary urological intervention for cystinuric patients: immediate and long term impact and implications. J Urol. 1998;160:341-4.
49. Kachel TA, Vijan SR, Dretler SP. Endourological experience with cystine caliculi and a treatment algorithm. J Urol. 1991;145:25-8.
50. Rudnick DM, Bennett PM, Dretler SP. Retrograde renoscopic fragmentation of moderate-size (1.5-3.0 cm) renal cystine stones. J Endourol. 1999;13:483-5.
51. Kim SC, Hatt EK, Lingeman JE, et al. Cystine: helical computerized tomography characterization of rough and smooth calculi in vitro. J Urol. 2005;174:1468-70.