Inherited Hypercalciuric Syndromes: Dent's Disease (CLC-5) and Familial Hypomagnesemia With Hypercalciuria (Paracellin-1)

By Stephen J. Knohl and Steven J. Scheinman

Dent's disease and familial hypomagnesemia with hypercalciuria and nephrocalcinosis (FHHNC) are inherited diseases in which hypercalciuria, nephrocalcinosis, and renal failure are prominent features. Dent's disease resembles a Fanconi syndrome, with impaired reabsorption in the proximal tubule; FHHNC, with urinary loss of magnesium and calcium, is associated with impaired cation transport in the thick ascending limb of Henle's loop. Gene mapping in families and positional cloning led in both cases to identification of the responsible gene. Dent's disease is associated with mutations that disrupt function of a voltage-gated chloride channel, CLC-5, expressed in subapical endosomes of the proximal tubule and in other nephron segments. Impaired function of this channel disturbs reabsorption of filtered proteins, as well as other transport functions of the proximal tubule, and leads, apparently indirectly, to hypercalciuria and renal failure. FHHNC results from mutations in paracellin-1, a tight-junction protein that appears to be important in conducting or regulating paracellular cation transport. Impaired function of paracellin-1 leads specifically to urinary losses of magnesium and calcium, but because transcellular transport is intact these patients do not have hypokalemia or salt wasting. Identification of both genes represent triumphs of a genetic approach to solving problems of pathophysiology.
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ADVANCES IN MOLECULAR genetics have made it possible to unravel the physiology of syndromes that had seemed unsolvable puzzles. Two diseases of renal transport, both with hypercalciuria, represent striking successes of this approach. Physiologic observations in patients with Dent's disease (X-linked nephrolithiasis) and familial hypomagnesemia with hypercalciuria and nephrocalcinosis (FHHNC) pointed to a primary defect in the proximal tubule in the former and in the thick ascending limb of Henle's loop in the latter, but it was only with cloning of the responsible genes and identification of the gene products that the physiology of these disorders has begun to be understood. Table 1 summarizes the clinical differences between these 2 syndromes.

DENT'S DISEASE

The hallmarks of this disease are hypercalciuria and low-molecular weight proteinuria. The fully expressed syndrome includes proximal tubular reabsorptive failure resembling a Fanconi syndrome, nephrolithiasis, nephrocalcinosis, deterioration in renal function, and rickets. Different clinical characteristics predominated in initial descriptions of the disease from different geographic areas, prompting a variety of descriptive names such as X-linked nephrolithiasis, but this is now recognized as a single disease with a single name, Dent's disease, in honor of Charles Dent who in 1964 described 2 unrelated male children with hypercalciuria, rickets, and proximal tubular dysfunction. Because this is an X-linked recessive trait, all daughters of an affected male are heterozygous (carriers) for the condition, 50% of sons from a female carrier will be affected, and male-to-male transmission is impossible.[1,2]

Mutations in the *CLCN5* gene on the X chromosome are associated consistently with this disease.[3] *CLCN5* was identified through gene mapping and positional cloning in affected families.[3,4] It encodes a voltage-gated chloride channel, CLC-5, that had not been known previously. CLC-5 is a member of a channel family that also includes CLC-0, the major chloride channel in human muscle that is mutated in congenital myotonia; CLC-Kb, the basolateral chloride channel in the renal medullary thick ascending limb that is mutated in a large subset of patients with Bartter's syndrome; and CLC-7, expressed at the ruffled surface of osteoclasts that facilitates acidification of the mineralization front of bone and that is mutated in congenital malignant osteopetrosis.[5] CLC-5 is expressed predominantly in kidney, although low levels of expression can be found in other tissues including intestine. Along the nephron, CLC-5 is expressed in the subapical endosomes of the prox-

Table 1. Clinical Differences Between Dent's Disease and FHHNC

	Dent's Disease	FHHNC
Inheritance pattern	X-linked recessive	Autosomal recessive
Mutated gene (protein)	*CLCN5* (CLC-5)	*PCLN1* (paracellin-1)
Age at presentation	Infancy or childhood	Infancy
Major clinical features	LMW proteinuria and other proximal solute wasting	Hypomagnesemia
	Polyuria	Polyuria
	Microscopic hematuria	Infantile siezures
	Hypercalciuria	Hypercalciuria
	Nephrocalcinosis	Nephrocalcinosis
	Nephrolithiasis	Nephrolithiasis
	Renal failure	Renal failure
	Rickets/osteomalacia	Ocular abnormalities
Onset of renal failure	Adolescence; end-stage renal disease by 4th decade	Early childhood; end-stage renal disease often by late adolescence
Laboratory abnormalities		
Serum		
Calcium	Normal	Normal
Magnesium	Normal	Low
Uric acid	Normal	High
Intact PTH	Normal or low	High
$1,25(OH)_2D$	Normal or high	High
Hypercalciuria	Present	Present
Hypermagnesuria	Absent	Present
Low-molecular weight proteinuria	Always present	—
Nephrocalcinosis	Present	Present

imal tubule, and in endosomes in the cells of the medullary thick ascending limb of Henle's loop. It also is expressed in apical and subapical regions of α-intercalated cells of the collecting duct.[6,7] Although its function in these distal segments remains poorly understood, the role of CLC-5 in proximal tubules has come into clearer focus.

In the proximal tubule, CLC-5 colocalizes with the proton-adenosine triphosphatase in subapical endosomes that are a critical part of the reabsorptive apparatus for proteins.[6,7] Filtered proteins (including those of low-molecular weight as well as albumin) adsorb to the receptor megalin on the apical surface of the proximal tubular epithelial cells. When endosomes bud off this surface, the first step in degrading the adsorbed proteins requires acidification of the endosomal lumen, and this is achieved through the activity of the vacuolar proton adenosine triphosphatase. It has been known for a decade that acidification of the endosomes requires dissipation of the charge by a protein kinase A–regulated chloride conductance,[8] and this appears to be the function of CLC-5, although regulation by protein kinase A has not been documented.

Impaired function of megalin-dependent protein uptake would lead to urinary loss of proteins. About 50% to 70% of the urinary protein in patients with Dent's disease is comprised of low-molecular weight proteins, and the remainder is albumin.[9] This pattern is present from infancy (A. Norden, personal communication), indicating that the albuminuria is not a nonspecific consequence of glomerular sclerosis because renal insufficiency does not begin in the first decade of life. Thus, Norden et al[9] used measurements of urinary excretion of proteins in patients with Dent's disease without renal insufficiency to derive an in vivo estimate of glomerular sieving coefficients. Patients with Dent's disease excrete 0.5 to 2.0 g of protein per day, and are not clinically nephrotic. The most useful screening tests for Dent's disease are measurement of β2-microglobulin levels, which are widely available in clinical laboratories, and of retinal-binding globulin, which is available less often but is more sensitive and specific.[10]

Further evidence for proximal tubular dysfunction in Dent's disease is the wasting of glucose, amino acids, and phosphate, which have in common that they are reabsorbed through the activities

of sodium-dependent transporters. Physiologic regulation of the activities of these transporters involves recycling between endosomes and the apical cell surface,[11,12] and it has been shown that when acidification of subapical endosomes is inhibited, membrane trafficking is impaired.[13] In 2 mouse models in which *CLCN5* expression is prevented by targeted disruption of the gene, proximal tubular endocytosis is disturbed,[12,14] and in one of these knockouts trafficking of the sodium-dependent phosphate cotransporter NaPi2 is altered.[12] Glycosuria, aminoaciduria, and phosphaturia are variable among patients with Dent's disease, and can be intermittent. It is plausible to speculate that these defects may vary with fluctuations in the state of endosomal congestion, perhaps with variations in the load of proteins presented for reabsorption.

As further evidence of abnormal endosomal function, Piwon et al[12] have shown that their CLC-5 knockout mice have reduced expression of megalin in proximal tubular cells, and very low levels of megalin in the urine. Urinary megalin excretion also is decreased in patients with Dent's disease.[15]

Hypercalciuria is a hallmark of Dent's disease, and is the major risk factor for stone formation and nephrocalcinosis because these patients excrete normal quantities of oxalate, citrate, uric acid, and other stone risk determinants.[10,16] The degree of hypercalciuria in adolescents and adults resembles that in patients with idiopathic hypercalciuria, approximating 4 to 6 mg/kg body weight. In patients with Dent's disease at these ages, hypercalciuria is diet-dependent and often can normalize with dietary calcium restriction (which is not recommended because it could exacerbate bone demineralization). Infants and young children, however, exhibit heavier degrees of hypercalciuria in the range of 10 to 12 mg/kg, and the hypercalciuria may persist on fasting.[10]

In 2 mouse models of CLCN5 inactivation, hypercalciuria is diet-dependent,[17,18] as in the patients. However, in one mouse model in which *CLCN5* expression is eliminated completely through targeted disruption of the gene, a small degree of hypercalciuria persists on a very low calcium intake. This latter model resembles the persistent hypercalciuria seen in young children. It is not clear whether this persistent hypercalciuria results from a renal leak or a primary tendency to impaired bone mineralization. In patients with Dent's disease, hypercalciuria[10] responds to a thiazide diuretic, as in idiopathic hypercalciuria.[19] This is consistent with the fact that the CLC-5 chloride channel is not expressed in the thiazide-sensitive cortical distal tubule.[6,7]

Serum calcium levels are normal, and in some patients tend toward the upper limit of the normal range.[10,16,20,21] In patients with normal creatinine clearances, measurements of intact parathyroid hormone tend to be low, and levels of 1,25 dihydroxyvitamin D often are increased.[1] This is more consistent with a pattern of absorptive hypercalciuria rather than a primary renal calcium leak. It is not yet known whether the relatively high levels of 1,25-dihydroxyvitamin D result from hypophosphatemia or represent a dysregulation of activity of the 1α-hydroxylase as a consequence of CLC-5 inactivation.

Parathyroid hormone is among the filtered low-molecular weight proteins that are found in great excess in the urine of knockout mice[12] and humans with Dent's disease.[9] Jentsch and colleagues have speculated that the inhibition of phosphate transport and the stimulation of 1-hydroxylation of vitamin D are consequences of activation of parathyroid hormone receptors on the apical surface of cells of the late proximal tubule.[12]

Renal failure occurs in about two thirds of patients with Dent's disease. Renal function may begin to decline in the teenage years and reach end stage by the fourth decade of life, but it is not clear why some patients are spared renal failure even into old age. Prominent histologic features include tubular atrophy and interstitial fibrosis, and there are varying degrees of glomerular sclerosis. Basement membranes appear normal on electron microscopy and immune deposits have not been identified on immunofluorescence.[16,20,21] The etiology of the dysfunction is unclear. Sayer et al[22] showed, in cultured collecting duct cells in which CLC-5 expression is prevented, that exposure to calcium oxalate crystals results in cellular engorgement with crystals. However, in anecdotal series, the severity of nephrocalcinosis does not appear to correlate with the degree of renal failure.[1,16] Norden et al[9] have documented increased urinary excretion of bioactive peptides including hormones and cytokines including parathyroid hormone, insulin-like growth factor 1, and monocyte chemoattractant protein 1, and proposed that high lumenal

levels of cytokines may promote tubulointerstitial fibrosis. In a preliminary report, Cebotaru et al[23] recently described that renal failure can be seen if CLC-5 knockout mice are followed-up to a sufficient age, with histologic evidence of interstitial inflammation and fibrosis, and tubular atrophy. In this report, the knockout mice also had increased tissue levels of messenger RNA encoding transforming growth factor $\beta1$, a profibrogenic cytokine.

About one fourth of patients with Dent's disease have rickets, which often presents in infancy. As with renal failure, this feature does not correlate with the nature of the mutation, and can vary even within families in which multiple affected males all share the same mutation.[1] It is not clear that the occurrence of rickets can be explained by the degree of hypophosphatemia, which usually is mild. The role, if any, of CLC-5 in bone, and of any possible modifying genes, is not known.

FAMILIAL HYPOMAGNESEMIA WITH HYPERCALCIURIA AND NEPHROCALCINOSIS

FHHNC was first described in 1972, and identified as the *Michelis-Castillo syndrome*.[24] FHHNC is a rare entity transmitted in an autosomal-recessive fashion presenting at birth. Prominent features include renal wasting of magnesium and calcium associated with development of nephrocalcinosis by early childhood. As would be expected in patients with a renal calcium leak (and in contrast to Dent's disease), serum levels of parathyroid hormone are high in patients with FHHNC.[24-26] Hypomagnesemia can be severe, with neonatal seizures. Polyuria or urinary infections often are presenting complaints. Renal function declines progressively and many patients reach end-stage by the teenage or young adult years. Most patients have hyperuricemia.[24] In addition to these renal problems, a range of ocular abnormalities are seen in some patients.[24-26]

In elegant work from Lifton's laboratory, the disease gene was mapped in 12 families to a region on the long arm of chromosome 3 (3q27) (as is often helpful in mapping autosomal-recessive diseases, most of these families exhibited consanguinity). Through positional cloning, a gene (*PCLN1*) was identified that encoded a protein with homology to the claudin family of tight-junction proteins; the protein was named paracellin-1 (also known as claudin 16). Claudins are membrane proteins with

4 transmembrane domains and 2 extracellular loops that are presumed to play an important role in the integrity of the tight junction. Paracellin-1 is expressed in the tight junctions of the thick ascending limb of Henle's loop in humans[27]; in the rat its expression has been detected in the thick ascending limb as well as the distal convoluted tubule and collecting duct.[28] Functional studies have not been performed, but it is speculated that paracellin might be a paracellular ion channel, or might be involved in regulating paracellular cation conductance. In either case, paracellin-1 would represent the first protein shown to participate in renal paracellular transport.

Mutations in *PCLN1* segregated with disease in these families and were not polymorphisms[27]; this has now been confirmed by others.[24,29] Consistent with the recessive nature of inheritance of this disease, most of the reported patients have been homozygous for a mutation or compound heterozygotes with 2 different mutations in *PCLN1*.[24,27,29] Heterozygotes have been reported with mild, asymptomatic hypomagnesemia,[29] hypercalciuria, or even clinically overt disease.[24]

Most of the reported mutations in patients with FHHNC are missense mutations that predict substitution of a single amino acid, and these occur within the transmembrane domains and the 2 extracellular domains of the protein. Thus, it is plausible that these mutations would interfere with the function of the paracellular barrier and lead to abnormalities of transport in the thick ascending limb, a major site for reabsorption of calcium and magnesium. Unlike the distal convoluted tubule where the bulk of calcium and magnesium reabsorption occurs across the epithelial cell, cation transport in the thick limb is largely paracellular, driven by the lumen-positive electrical potential.

Maintenance of the positive charge in the lumen depends on proper functioning of the transcellular transport pathway in the cells of the thick ascending limb, including the bumetanide-sensitive Na-K-2Cl cotransporter, the apical potassium channel (ROMK), and the basolateral chloride channel CLC-Kb with its associated protein barttin. Defective function of genes encoding these transport proteins result in Bartter's syndrome, in which hypercalciuria is a universal but secondary consequence and hypomagnesemia, when it occurs, usually is mild.[30,31]

It is instructive to contrast the findings in patients with FHHNC, in whom transcellular transport is normal but paracellin is mutated, with those in patients with Bartter's syndrome, whose inherited defect impairs transcellular transport. Hypokalemic metabolic alkalosis, the hallmark of Bartter's and Gitelman's syndromes, is absent in patients with FHHNC. Both syndromes can present in infancy, but whereas children with Bartter's syndrome often have salt wasting and volume depletion, children with FHHNC suffer with symptomatic hypomagnesemia but do not have clinically significant salt wasting.[29] Serum levels of renin and aldosterone, which are high in Bartter's syndrome,[32] are normal in FHHNC.[29] Blanchard et al[29] studied the responses to furosemide in 2 unrelated patients with FHHNC who were homozygous for mutations in *PCLN1*. Administration of furosemide produces no natriuresis in patients with Bartter's syndrome, in which transcellular transport in the thick ascending limb of Henle already is impaired. In contrast, furosemide produces a natriuresis in patients with FHHNC that is comparable with the response in control subjects, but unlike the control subjects, furosemide did not increase excretion of calcium or magnesium in patients with FHHNC.

In preliminary studies described so far only in an abstract, mice with targeted disruption of the *PCLN1* gene exhibit most of the features of the human FHHNC syndrome. These knockout mice have hypomagnesemia with inappropriate urinary magnesium wasting, hypercalciuria, nephrocalcinosis, renal insufficiency, a urinary concentrating defect, and normal salt conservation.[33]

Inadequate urinary acidification was one of the features identified by Michelis,[34] but this was probably not a direct effect of the genetic defect but rather the consequence of medullary interstitial damage.[34]

Other findings in FHHNC remain, for the moment, unexplained. Hyperuricemia is present in the majority of patients with this syndrome,[24-26] but the mechanism is not known. No paracellular secretory process for uric acid has been described or even proposed. Abnormalities of the eye occur in a significant minority of patients. These include nystagmus, severe myopia, macular colobomata, and tapetoretinal degeneration.[36] It is not known whether *PCLN1* is expressed in the eye, and what its function there might be.

CONCLUSION

Dent's disease and FHHNC are both syndromes of hypercalciuria, nephrocalcinosis, and renal failure, for which the pathophysiologic bases certainly would not be so well understood without the success of the genetic studies described earlier. In both cases, identification of families made possible linkage analysis and then positional cloning, which led to the discovery in both diseases of a novel gene. The discoveries of CLC-5 and paracellin-1 have made possible in short order a more complete understanding of disease mechanisms and of normal renal tubular physiology, and also have allowed investigators to frame the right questions for further research. There are other inherited diseases associated with nephrolithiasis for which the molecular basis is known, particularly primary hyperoxaluria and cystinuria.[36] Exciting progress recently has been made in identifying genes for several other inherited syndromes of hypomagnesemia.[38-40] However, the path to understanding complex traits such as idiopathic hypercalciuria, the most common risk factor for kidney stones, will be much more difficult because in idiopathic hypercalciuria there are multiple mechanisms, probably reflecting polymorphisms in multiple genes. Nevertheless, there is every reason to expect that such obstacles will be overcome and that molecular genetics will continue to teach us about the pathophysiology of monogenic and polygenic diseases.

REFERENCES

1. Scheinman SJ: X-linked hypercalciuric nephrolithiasis: Clinical syndromes and chloride channel mutations. Kidney Int 53:3-17, 1998

2. Scheinman SJ, Thakker RV. X-linked nephrolithiasis/Dent's disease and mutations in the ClC-5 chloride channel, in Econs MJ (ed): Genetic Aspects of Osteoporosis and Metabolic Bone Disease. Totowa, NJ, Humana Press, 1999

3. Lloyd SE, Pearce SHS, Fisher SE, et al: A common molecular basis for three inherited kidney stone diseases. Nature 379:445-449, 1996

4. Scheinman SJ, Pook MA, Wooding C, et al: Mapping the gene causing X-linked recessive nephrolithiasis to Xp11.22 by linkage studies. J Clin Invest 91:2351-2357, 1993

5. Jentsch TJ, Stein V, Weinreich F, et al: Molecular structure and physiological function of chloride channels. Physiol Rev 82:503-568, 2002

6. Gunther W, Luchow A, Cluzeaud F, et al: ClC-5, the chloride channel mutated in Dent's disease, colocalizes with the proton pump in endocytotically active kidney cells. Proc Natl Acad Sci U S A 95:8075-8080, 1998

7. Devuyst O, Christie PT, Courtoy PJ, et al: Intra-renal and

subcellular distribution of the human chloride channel, CLC-5, reveals a pathophysiological basis for Dent's disease. Hum Mol Genet 8:247-257, 1999

8. Bae HR, Verkman AS: Protein kinase A regulates chloride conductance in endocytic vesicles from proximal tubule. Nature 348:637-639, 1990

9. Norden AG, Lapsley M, Lee PJ, et al: Glomerular protein sieving and implications for renal failure in Fanconi syndrome. Kidney Int 60:1885-1892, 2001

10. Reinhart SC, Norden AGW, Lapsley M, et al: Characterization of carrier females and affected males with X-linked recessive nephrolithiasis. J Am Soc Nephrol 5:1451-1461, 1995

11. Pfister MF, Hilfiker H, Forgo J, et al: Cellular mechanisms involved in the acute adaptation of OK cell Na/Pi-cotransport to high- or low-Pi medium. Pflugers Arch 435:713-719, 1998

12. Piwon N, Gunther W, Schwake M, et al: ClC-5 Cl⁻ channel disruption impairs endocytosis in a mouse model for Dent's disease. Nature 408:369-373, 2000

13. Presley JF, Mayor S, McGraw TE, et al: Bafilomycin A1 treatment retards transferrin receptor recycling more than bulk membrane recycling. J Biol Chem 272:13929-13936, 1997

14. Wang SS, Devuyst O, Courtoy PJ, et al: Mice lacking renal chloride channel, CLC-5, are a model for Dent's disease, a nephrolithiasis disorder associated with defective receptor-mediated endocytosis. Hum Mol Genet 9:2937-2945, 2000

15. Norden AG, Lapsley M, Igarashi T, et al: Urinary megalin deficiency implicates abnormal tubular endocytic function in fanconi syndrome. J Am Soc Nephrol 13:125-133, 2002

16. Wrong O, Norden AGW, Feest TG: Dent's disease: A familial proximal renal tubular syndrome with low-molecular-weight proteinuria, hypercalciuria, nephrocalcinosis, metabolic bone disease, progressive renal failure, and a marked male predominance. QJM 87:473-493, 1994

17. Luyckx VA, Leclercq B, Dowland LK, et al: Diet-dependent hypercalciuria in transgenic mice with reduced CLC5 chloride channel expression. Proc Natl Acad Sci 21:12174-12179, 1999

18. Silva IV, Cebotaru V, Wang H, et al: The CLC-5 knockout mouse model of Dent's disease has renal hypercalciuria and increased bone turnover. J Bone Miner Res 18:615-623, 2003

19. Raja KM, Schurman S, D'Mello RG, et al: Responsiveness of hypercalciuria to thiazide in Dent's disease. J Am Soc Nephrol 3:2938-2944, 2002

20. Frymoyer PA, Scheinman SJ, Dunham PB, et al: X-linked recessive nephrolithiasis with renal failure. N Engl J Med 325:681-686, 1991

21. Igarashi T, Hayakawa H, Shiraga H, et al: Hypercalciuria and nephrocalcinosis in patients with idiopathic low-molecular-weight proteinuria in Japan: Is the disease identical to Dent's disease in United Kingdom? Nephron 69:242-247, 1995

22. Sayer JA, Carr G, Pearce SH, et al: Disordered calcium crystal handling in antisense CLC-5-treated collecting duct cells. Biochem Biophys Res Commun 300:305-310, 2003

23. Cebotaru V, Devuyst O, Wang H, et al: The CLC-5 knockout mouse model of Dent's disease develops progressive renal failure. J Am Soc Nephrol 13:282A-283A, 2001 (abstr)

24. Weber S, Schneider L, Peters M, et al: Novel paracellin-1 mutations in 25 families with familial hypomagnesemia with hypercalciuria and nephrocalcinosis. J Am Soc Nephrol 12:1872-1881, 2001

25. Torralbo A, Pina E, Portoles J, et al: Renal magnesium wasting with hypercalciuria, nephrocalcinosis and ocular disorders. Nephron 69:472-475, 1995

26. Praga M, Vara J, Gonzalez-Parra E, et al: Familial hypomagnesemia with hypercalciuria and nephrocalcinosis. Kidney Int 47:1419-1425, 1995

27. Simon DB, Lu Y, Choate KA, et al: Paracellin-1, a renal tight junction protein required for paracellular Mg resorption. Science 285:103-106, 1999

28. Weber S, Schlingmann KP, Peters M, et al: Primary gene structure and expression studies of rodent paracellin-1. J Am Soc Nephrol 12:2664-2672, 2001

29. Blanchard A, Jeunemaitre X, Coudol P, et al: Paracellin-1 is critical for magnesium and calcium reabsorption in the human thick ascending limb of Henle. Kidney Int 59:2206-2215, 2001

30. Estevez R, Boettger T, Stein V, et al: Barttin is a Cl-channel beta-subunit crucial for renal Cl- reabsorption and inner ear K+ secretion. Nature 414:558-561, 2001

31. Simon DB, Bindra RS, Mansfield TA, et al: Mutations in the chloride channel gene, CLCNKB, cause Bartter's syndrome type III. Nat Genet 17:171-178, 1997

32. Simon DB, Lifton RP: The molecular basis of inherited hypokalemic alkalosis: Bartter's and Gitelman's syndromes. Am J Physiol 271:F961-F966, 1996

33. Lu Y, Choate KA, Wang T, et al: Paracellin-1 knock-out mouse model of recessive renal hypomagnesemia, hypercalciuria and hephrocalcinosis. J Am Soc Nephrol 12:756A, 2001 (abstr)

34. Michelis MF, Drash AL, Linarelli LG, et al: Decreased bicarbonate threshold and renal magnesium wasting in a sibship with distal renal tubular acidosis. Metab Clin Exper 21:905-920, 1972

35. Rodriguez-Soriano J, Vallo A: Pathophysiology of the renal acidification defect present in the syndrome of familial hypomagnesaemia-hypercalciuria. Pediatr Nephrol 8:431-435, 1994

36. Benigno V, Canonica CS, Bettinelli A, et al: Hypomagnesaemia-hypercalciuria-nephrocalcinosis: A report of nine cases and a review. Nephrol Dial Transplant 15:605-610, 2000

37. Scheinman SJ: Genetics of nephrolithiasis. Semin Nephrol 19:381-388, 1999

38. Meij IC, Koenderink JB, van Bokhoven H, et al: Dominant isolated renal magnesium loss is caused by misrouting of the Na+, K+-ATPase gamma-subunit. Nat Genet 26:265-266, 2000

39. Walder RY, Landau D, Meyer P, et al: Mutation of TRPM6 causes familial hypomagnesemia with secondary hypocalcemia. Nat Genet 31:171-174, 2002

40. Schlingmann KP, Weber S, Peters M, et al: Hypomagnesemia with secondary hypocalcemia is caused by mutations in TRPM6, a new member of the TRPM gene family. Nat Genet 31:166-170, 2002