Inherited Disorders of Renal Magnesium Handling

DAVID E. C. COLE and GARY A. QUAMME
Department of Laboratory Medicine & Pathobiology, University of Toronto, Toronto, Ontario, and
Department of Medicine, University of British Columbia, Koerner Pavilion, University Hospital, Vancouver,
British Columbia, Canada.

Abstract. The genetic basis and cellular defects of a number of primary magnesium wasting diseases have been elucidated over the past decade. This review correlates the clinical pathophysiology with the primary defect and secondary changes in cellular electrolyte transport. The described disorders include (1) hypomagnesemia with secondary hypocalcemia, an early-onset, autosomal-recessive disease segregating with chromosome 9q12-22.2; (2) autosomal-dominant hypomagnesemia caused by isolated renal magnesium wasting, mapped to chromosome 11q23; (3) hypomagnesemia with hypercalciuria and nephrocalcinosis, a recessive condition caused by a mutation of the claudin 16 gene (3q27) coding for a tight junctional protein that regulates paracellular Mg$^{2+}$ transport in the loop of Henle; (4) autosomal-dominant hypoparathyroidism, a variably hypomagnesemic disorder caused by inactivating mutations of the extracellular Ca$^{2+}$/Mg$^{2+}$-sensing receptor, Casr gene, at 3q13.3-21 (a significant association between common polymorphisms of the Casr and extracellular Mg$^{2+}$ concentration has been demonstrated in a healthy adult population); and (5) Gitelman syndrome, a recessive form of hypomagnesemia caused by mutations in the distal tubular NaCl cotransporter gene, SLC12A3, at 16q13. The basis for renal magnesium wasting in this disease is not known. These inherited conditions affect different nephron segments and different cell types and lead to variable but increasingly distinguishable phenotypic presentations. No doubt, there are in the general population other disorders that have not yet been identified or characterized. The continued use of molecular techniques to probe the constitutive and congenital disturbances of magnesium metabolism will increase the understanding of cellular magnesium transport and provide new insights into the way these diseases are diagnosed and managed.

Control of magnesium homeostasis resides principally within the nephron of the kidney (1). Approximately 80% of the total plasma magnesium (0.65 to 1.20 mM) is filtered through the glomerulus. Five to 15% of the ultrafiltrable magnesium is reabsorbed by the convoluted and straight portions of the proximal tubule. The cortical segment of the thick ascending limb of the loop of Henle plays a major role in the determination of magnesium reabsorption, as it accounts for approximately 70% of magnesium conservation (Figure 1). Ten to 15% of the filtered magnesium is delivered distally from the loop of Henle, of which 70 to 80% is reabsorbed by the distal tubule. There is no evidence for significant magnesium absorption beyond the distal convoluted tubule so that it plays an important role in determining the final urinary excretion (1). There are no reports of significant magnesium secretion in any of the tubule segments that compose the nephron, so that control of renal magnesium homeostasis involves changes in reabsorption. Overall, less than 5% of the filtered magnesium normally appears in the urine.

The purpose of this review is to discuss the familial disorders of renal magnesium reabsorption that lead to urinary magnesium wasting. Inborn errors of renal magnesium handling tell us much about the mechanisms that the kidney normally uses to conserve magnesium. The genetic basis and cellular defects of a number of primary magnesium wasting diseases have been elucidated over the past decade, whereas others remain to be identified and there are undoubtedly others that have not yet been recognized. In this review, we attempt to correlate the clinical pathophysiology with the primary defect and secondary changes in cellular electrolyte transport. We begin, however, with a brief overview of normal renal magnesium handling; more detailed reviews have been published elsewhere (1,2).

Proximal Tubule

In the adult, the proximal tubule reabsorbs only 10% of the filtered magnesium, whereas the fractional reabsorption of sodium and calcium is normally in excess of 70% (1). However, in the neonate, the proximal tubule reabsorbs approximately 70% of the filtered magnesium—similar to that for sodium and calcium (2). Leleivre-Pegorier et al. (2) reported that the permeability of the proximal tubule changes during development so that less magnesium is reabsorbed in the proximal tubule of the adult. This maturation in segmental handling of magnesium must be taken into consideration when assessing renal magnesium handling in children relative to that in adults.

Loop of Henle

De Rouffignac and colleagues showed (3) that the cortical segment of the thick ascending limb (cTAL) reabsorbs approximately 70% of the filtered magnesium, whereas the medullary
segment (mTAL) does not absorb magnesium. They further reported that transepithelial magnesium absorption is passive, moving from lumen to the interstitial space through the paracellular pathway (Figure 2). This is consistent with the earlier observations of Shareghi and Agus (4). The driving force for magnesium movement is the positive luminal transepithelial voltage (3). Any influence that alters transepithelial voltage or the permeability of the paracellular pathway will alter magnesium reabsorption in the cTAL (3). The voltage in the loop is determined by the rate of Na-K-Cl cotransport and active sodium absorption. Changes in their transport rates will affect the transepithelial voltage and thus magnesium absorption. The permeability of the paracellular pathway is determined by electrostatic charges of proteins that compose this route (1). Although there seems to be some selectivity in the paracellular pathway, changes in permeability, i.e., proteins that compose the pathway, would be expected to alter sodium and calcium as well as magnesium (1,3,5). Recently, a paracellular protein, “paracellin-1” or “claudin 16,” has been identified in the TAL (6). The claudin 16 gene encodes a protein of 305 amino acids that form the tight junction of the paracellular pathway. Simon and colleagues (6) proposed that claudin 16 is involved in controlling magnesium and calcium permeability of the paracellular pathway in the cTAL.

A large number of hormones stimulate magnesium reabsorption in the loop (Table 1). All of these hormonal responses are mediated by changes in both transepithelial voltage and paracellular permeability (1). As indicated by the diversity of the hormones, the responses are mediated by different receptor-signaling pathways that change transepithelial voltage and paracellular structure.

**Table 1.** Coordinate controls of magnesium transport in the loop of Henle and distal tubule

<table>
<thead>
<tr>
<th>Peptide hormones</th>
<th>Thick Ascending Limb</th>
<th>Distal Tubule</th>
</tr>
</thead>
<tbody>
<tr>
<td>parathyroid hormone</td>
<td>Increase (7)</td>
<td>Increase (15)</td>
</tr>
<tr>
<td>calcitonin</td>
<td>Increase (1)</td>
<td>Increase (8)</td>
</tr>
<tr>
<td>glucagon</td>
<td>Increase (7)</td>
<td>Increase (8)</td>
</tr>
<tr>
<td>arginine vasopressin</td>
<td>Increase (7)</td>
<td>Increase (8)</td>
</tr>
<tr>
<td>β-adrenergic agonists</td>
<td>Increase (1)</td>
<td>Increase (8)</td>
</tr>
<tr>
<td>isoproterenol</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Prostaglandins, PGE₂</td>
<td>Decrease (8)</td>
<td>Increase (16)</td>
</tr>
<tr>
<td>Insulin</td>
<td>Increase (1)</td>
<td>Increase (15)</td>
</tr>
<tr>
<td>Mineralocorticoids</td>
<td></td>
<td></td>
</tr>
<tr>
<td>aldosterone</td>
<td>Increase (8)</td>
<td>Increase (8)</td>
</tr>
<tr>
<td>Vitamin D 1,25(OH)₂D₃</td>
<td></td>
<td>Increase (8)</td>
</tr>
<tr>
<td>Magnesium restriction</td>
<td>Increase (1)</td>
<td>Increase (8)</td>
</tr>
<tr>
<td>Hypermagnesemia</td>
<td>Decrease (8)</td>
<td>Decrease (8)</td>
</tr>
<tr>
<td>Hypercalcemia</td>
<td>Decrease (8)</td>
<td>Decrease (8)</td>
</tr>
<tr>
<td>Extracellular volume expansion</td>
<td>Decrease (1)</td>
<td>Increase (1)</td>
</tr>
<tr>
<td>Metabolic acidosis</td>
<td>Decrease (8)</td>
<td>Decrease (8)</td>
</tr>
<tr>
<td>Metabolic alkalosis</td>
<td>Increase (8)</td>
<td>Increase (8)</td>
</tr>
<tr>
<td>Phosphate-depletion</td>
<td>Decrease (8)</td>
<td>Decrease (8)</td>
</tr>
<tr>
<td>Potassium-depletion</td>
<td>Decrease (8)</td>
<td>Decrease (8)</td>
</tr>
<tr>
<td>Diuretics</td>
<td></td>
<td></td>
</tr>
<tr>
<td>furosemide</td>
<td>Decrease (8)</td>
<td>No effect (8)</td>
</tr>
<tr>
<td>amiloride</td>
<td>No effect (8)</td>
<td>Increase (8)</td>
</tr>
<tr>
<td>chlorothiazide</td>
<td>No effect (8)</td>
<td>Increase (8)</td>
</tr>
</tbody>
</table>

The specific studies are referenced in the review articles (1,8).
Hypermagnesemia and hypercalcemia have long been known to cause an increase in urinary magnesium and calcium excretion (8). Massry et al. (9) elevated serum magnesium levels in thyro-parathyroidectomized dogs and determined the urinary magnesium excretion rates with elevated filtered magnesium. Using clearance studies, they showed that elevated filtered magnesium concentration was initially associated with increases in magnesium reabsorption until an apparent tubular reabsorptive maximum, Tm, was attained, beyond which any additional filtered magnesium was excreted in the urine. The cellular basis for these results is the extracellular Ca\(^2+\)/Mg\(^2+\)-sensing receptor (Casr) present in the basolateral membrane of the TAL that inhibits salt transport and passive Mg\(^2+\) and Ca\(^2+\) absorption (10,11). Although the apparent Tm has been used in the clinical assessment of renal magnesium conservation, it is not easily interpreted because it is a receptor-mediated, not a transport-dependent, phenomenon (12). The magnesium-loading test is a more appropriate approach to assess magnesium balance (13,14).

A number of influences affect renal magnesium conservation, including metabolic acidosis, potassium depletion, and hypophosphatemia (8). Although the cellular mechanisms are poorly understood, these influences alter passive magnesium reabsorption in the loop by changing the transepithelial voltage or the permeability of the paracellular pathway. Furosemide diminishes the luminal positive voltage by virtue of its effects on the Na-K-Cl cotransporter. Because of this, acute usage of furosemide leads to enhanced distal delivery and increased urinary magnesium excretion (8). However, with chronic furosemide therapy, urinary excretion usually returns to near control levels. This is likely due to enhanced reabsorption in the proximal tubule, loop, and distal tubule. Thus, prolonged furosemide use does not often lead to renal magnesium wasting.

**Distal Tubule**

Magnesium transport within the distal convoluted tubule (DCT) is transcellular and active in nature (Figure 3). Magnesium enters the cell through selective channels across the apical membrane, driven by the transmembrane negative electrical potential (8). Magnesium entry across the apical membrane is the rate-limiting step in transepithelial reabsorption, and many of the hormonal and nonhormonal controls act at this site. Cellular Mg\(^{2+}\) is actively extruded at the basolateral membrane, possibly by a sodium-dependent exchange mechanism (8).

A large number of influences regulate magnesium transport within the DCT (Table 1). Most of these controls are similar to those of the TAL. However, the cellular mechanisms are different, as the influences within the DCT act through changes in active magnesium transport. Renal cells respond very sensitively to decreased magnesium availability caused by dietary restriction, intestinal malabsorption, or excessive renal excretion by increasing transport rates (8). The change in magnesium transport is rapid, sensitive, and selective for magnesium. This response is a genomic effect involving transcriptional/translational control and de novo protein synthesis, possibly by the formation of new transporters or channels (1). This adaptive response of transport rates in the DCT provides for the selective renal magnesium conservation. Elevated extracellular Mg\(^{2+}\) or Ca\(^{2+}\) inhibits fractional magnesium transport in superficial rat distal tubules through activation of the Casr (17). These studies clearly indicate that hypermagnesemia and hypercalcemia per se can modify hormone regulation of magnesium transport within the distal tubule, leading to increased urinary magnesium excretion. Metabolic acidosis, hypokalemia, and phosphate depletion inhibit active Mg\(^{2+}\) transport in the DCT (8). Experimental and clinical data suggest an association among these diseases (18). Our evidence indicates that these three influences have different actions on cellular magnesium transport so that the three disturbances may act in an additive manner to compromise renal magnesium conservation.

The distal diuretics amiloride and chlorothiazide increase Mg\(^{2+}\) transport in DCT cells (8). Although amiloride has clearly been shown to be a magnesium-sparing diuretic, chronic chlorothiazide usage may lead to renal magnesium wasting (8). The cellular mechanisms for the chronic chlorothiazide effects are unclear but may involve hypokalemia, which can jeopardize renal magnesium conservation (8).

**Inherited Disorders of Renal Magnesium Handling**

**Primary Inherited Disorders of Renal Magnesium Handling**

A number of inherited magnesium wasting diseases that likely have their basis in defective magnesium transporters have been described (Table 2).
Hypomagnesemia with Secondary Hypocalcemia. Hypomagnesemia with secondary hypocalcemia (HSH) is an autosomal-recessive disorder that manifests in the newborn period and is characterized by very low serum magnesium and low calcium concentrations (19–24) (Table 3). Patients usually present before 6 mo of age with neurologic symptoms of hypomagnesemic hypocalcemia, including tetany, muscle spasms, and seizures (25,26). In older children with inadequate magnesium control, clouded sensorium and disturbed speech are often seen and choreoathetoid movements have been described. The hypocalcemia is secondary to parathyroid failure and peripheral parathyroid hormone (PTH) resistance as a result of magnesium deficiency (27). Hypoparathyroidism is occasionally present and is corrected only with normalization of plasma magnesium (28).

The disease primarily is due to defective intestinal magnesium absorption and may be fatal unless treated with high oral intakes (19,20,29–31). Walder et al. (32) reported that HSH is an autosomal recessive disease and showed by genetic linkage studies that the gene segregates to chromosome 9 (9q12–9q22.2) (Table 3). They suggested that the candidate gene codes for a receptor or ion channel involved in active intestinal magnesium absorption. As passive intestinal transport is normal, the disease can be controlled with high oral magnesium supplements, although the acute presentation may be more rapidly corrected with intramuscular or intravenous magnesium therapy. Renal magnesium conservation has been reported to be normal in most studies, suggesting that the kidney responds appropriately to low circulating magnesium levels by reabsorbing fractionally greater amounts of filtered magnesium. In some cases, however, there may be a renal leak, which manifests in the inability of oral supplements to normalize sufficiently the serum magnesium or the hypomagnesemic symptoms (33). We speculate that the renal leak may be due to altered Mg$^{2+}$ entry into DCT cells (Figure 3). Although intramuscular or intravenous magnesium supplementation may be required, continuous nocturnal nasogastric infusion of magnesium may be an effective alternative (34). Whether HSH is genetically heterogeneous remains to be seen, but additional studies to address renal tubular magnesium absorption as a function of plasma concentration and filtered magnesium in patients with well-delineated intestinal defects clearly are warranted.

**Infantile Isolated Renal Magnesium Wasting (Dominant).** Hypomagnesemia as a result of isolated renal magnesium loss is an autosomal-dominant condition associated with a few symptoms other than chondrocalcinosis (35). Patients always have hypocalciuria and variable but usually mild hypomagnesemic symptoms (36). Meij et al. (37) reported that the disorder maps to chromosome 11q23 in two large Dutch families. Database searches of the linkage region have failed to identify candidate genes, but Meij et al. speculated, from our experimental studies, that the mutation may lie in the distal tubule (Figure 3) (8,37).

**Infantile Isolated Renal Magnesium Wasting (Recessive).** There is evidence for a variant form of hypomagnesemia that is more consistent with isolated renal magnesium loss with autosomal-recessive inheritance. Meij et al. excluded linkage to any known previously reported loci indicating a distinct disease (I.C. Meij, personal communication, April 1999). The patients also have variable symptoms, but they usually have normal urinary calcium excretion (38,39). Because the epithelial transporters of magnesium have not been conclusively delineated, it is unclear at what level the tubule magnesium absorption is affected.

Because both dominant and recessive inheritance have been reported, it is likely that a number of familial renal magnesium wasting diseases exist. As the distal tubule reabsorbs 10 to 15% of the filtered magnesium (80 to 95% of that delivered to it), one would expect that if the primary reabsorptive channel were affected, then renal magnesium conservation would be severely compromised and lead to marked hypomagnesemia. Alternatively, there may be separate magnesium transporters within the distal tubule under separate genetic control. This is a fertile ground for further genetic studies.

**Idiopathic Hypermagnesiuria**

Although idiopathic hypercalciuria is an established clinical entry, the notion that a similar state of idiopathic hypermagnesiuria may occur in some fraction of the general population has not been examined extensively. The calcium-losing state is a familial disorder associated with increased urinary calcium oxalate and calcium hydrogen phosphate supersaturation, frequently leading to stone formation (40). Current evidence is that this disease is genetic in origin (41). By selective inbreeding, Bushinsky (40) was able to establish a colony of hypercalciuric stone-forming rats. These animals routinely develop kidney stones on normal dietary calcium intake. In rats, the stones are more likely to be brushite (calcium hydrogen phosphate) then calcium oxalate, which is more common in humans. The magnesium content of the stones is not elevated, and
abnormal renal magnesium handling is not thought to be part of the condition. Bushinsky and colleagues showed that the defects that lead to hypercalciuria include increased intestinal calcium absorption and bone resorption associated with increased intracellular vitamin D receptor concentrations (40). The molecular links to the associated defect of renal calcium reabsorption are still the subject of intense investigation (42). On the basis of responses to furosemide and chlorothiazide, these investigators suggested that the renal defect is localized to the TAL of the loop of Henle (42), but the molecular basis of the inherited defect is not known (42). Because the rats have normal renal magnesium homeostasis, it is reasonable to conclude that the defect is located more distally where calcium and magnesium are separately controlled (8).

An increasingly popular approach to the genetics of disease is the identification of candidate loci for quantitative traits. Like height or arterial BP, serum and urine magnesium concentrations lie along a continuum and can be analyzed as quantitative traits.

<table>
<thead>
<tr>
<th>feature</th>
<th>hypomagnesemia and secondary hypocalcemia</th>
<th>isolated dominant hypomagnesemia (infantile)</th>
<th>isolated recessive hypomagnesemia (infantile)</th>
</tr>
</thead>
<tbody>
<tr>
<td>inheritance</td>
<td>AR</td>
<td>AD</td>
<td>AR</td>
</tr>
<tr>
<td>gene</td>
<td>?</td>
<td>?</td>
<td>?</td>
</tr>
<tr>
<td>chromosome</td>
<td>9q12–22.2</td>
<td>11q23</td>
<td>?</td>
</tr>
<tr>
<td>serum Mg²⁺</td>
<td>↓ ↓</td>
<td>↓</td>
<td>↓</td>
</tr>
<tr>
<td>serum K⁺</td>
<td>N</td>
<td>N</td>
<td>N</td>
</tr>
<tr>
<td>serum Ca²⁺</td>
<td>↓</td>
<td>N</td>
<td>N</td>
</tr>
<tr>
<td>blood pH</td>
<td>N</td>
<td>N</td>
<td>N</td>
</tr>
<tr>
<td>urine K⁺</td>
<td>N</td>
<td>N</td>
<td>?</td>
</tr>
<tr>
<td>urine Mg²⁺</td>
<td>N or ↑</td>
<td>↑</td>
<td>↑</td>
</tr>
<tr>
<td>urine Ca²⁺</td>
<td>N or ↑</td>
<td>↓</td>
<td>?</td>
</tr>
<tr>
<td>onset</td>
<td>neonatal</td>
<td>childhood</td>
<td>childhood</td>
</tr>
<tr>
<td>growth</td>
<td>N</td>
<td>N</td>
<td>?</td>
</tr>
<tr>
<td>rickets</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>chondrocalcinosis</td>
<td>–</td>
<td>+</td>
<td>?</td>
</tr>
<tr>
<td>other abnormalities</td>
<td>–</td>
<td>(Seizures)</td>
<td>–</td>
</tr>
<tr>
<td>omim#</td>
<td>602014</td>
<td>154020</td>
<td>248250</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>feature</th>
<th>hypomagnesemia, hypercalciuria, and nephrocalcinosis</th>
<th>autosomal dominant hypoparathyroidism</th>
</tr>
</thead>
<tbody>
<tr>
<td>inheritance</td>
<td>AR</td>
<td>AD</td>
</tr>
<tr>
<td>gene</td>
<td>PCLN1</td>
<td>Casr</td>
</tr>
<tr>
<td>chromosome</td>
<td>3q27</td>
<td>3q13.3–21</td>
</tr>
<tr>
<td>serum Mg²⁺</td>
<td>↓</td>
<td>↓ or N</td>
</tr>
<tr>
<td>serum K⁺</td>
<td>N</td>
<td>N</td>
</tr>
<tr>
<td>serum Ca²⁺</td>
<td>N</td>
<td>↓</td>
</tr>
<tr>
<td>blood pH</td>
<td>N or sl. ↓</td>
<td>N</td>
</tr>
<tr>
<td>urine K⁺</td>
<td>?N</td>
<td>N</td>
</tr>
<tr>
<td>urine Mg²⁺</td>
<td>↑</td>
<td>↑</td>
</tr>
<tr>
<td>urine Ca²⁺</td>
<td>↑ ↑</td>
<td>↑</td>
</tr>
<tr>
<td>onset</td>
<td>infancy</td>
<td>infancy</td>
</tr>
<tr>
<td>growth</td>
<td>↓ ↓</td>
<td>N</td>
</tr>
<tr>
<td>rickets</td>
<td>+</td>
<td>–</td>
</tr>
<tr>
<td>chondrocalcinosis</td>
<td>–</td>
<td>+</td>
</tr>
<tr>
<td>other abnormalities</td>
<td>ocular anomalies</td>
<td>–</td>
</tr>
<tr>
<td>omim#</td>
<td>603959</td>
<td>601198</td>
</tr>
</tbody>
</table>

AR, autosomal recessive; AD, autosomal dominant; ↑, increased; ↑ ↑, markedly increased; sl. ↑, slightly increased; N, normal; +, present; –, absent; other abnormalities in parentheses are atypical features; OMIM# refers to the Online Mendelian Inheritance in Man database reference number (http://www3.ncbi.nlm.nih.gov/omim/).
Genetic studies of quantitative traits are difficult to perform because family members usually cannot be divided neatly into affected and unaffected categories as can be done with mendelian (single gene) disorders. Moreover, most quantitative traits are specified by more than one gene (polygenic inheritance), resulting in the failure of traditional linkage analysis to detect the effects of any single locus without prohibitively large population sampling. In an effort to define the genetic basis of magnesium homeostasis, Henrotte and colleagues (43) established inbred lines of mice by selecting for high and low plasma magnesium levels. Mice of the hypomagnesemic line have inappropriately high urinary magnesium excretion relative to the extracellular levels (44). The hypomagnesemic, hypermagnesiuric mice have normal calcium homeostasis, suggesting a selective tubular defect of magnesium reabsorption. Genetic analysis of these mice indicated that both histocompatibility (H2)-related and H2-unrelated to loci were significant determinants of extracellular and intracellular magnesium content in the mice (44). The hypomagnesemic, hypermagnesiuric mice have normal calcium homeostasis, suggesting a locus controlling a selective tubular pathway of magnesium reabsorption. Parallel studies in humans are sparse. Classically, comparison of monozygotic with dizygotic twins has been a useful measure of genetic contributions to the variability of quantitative traits, although the results frequently are subject to overinterpretation. Analysis by Henrotte indicates that serum magnesium is a genetic trait (45), a conclusion further supported by quantitative analysis of sib-pair data (46). No human locus has been clearly implicated as a determinant of magnesium homeostasis, but common functional polymorphisms of those genes whose complete loss causes inherited hypomagnesemia are obvious possibilities. We recently described strong associations between the common polymorphism of the Casr gene and extracellular calcium concentrations (47,48), making it an attractive candidate in this regard.

Clinically, such genetic determinants should be useful in the delineation of gene-environment interactions in diseases characterized by abnormalities of magnesium homeostasis (49). For example, in the presence of a genetic polymorphism that predisposes to enhanced renal magnesium conservation and elevated resting blood level, a patient may be more likely to experience toxic effects of magnesium administration—during tocolytic therapy, for example. Conversely, an individual with a polymorphism that predisposes to increased renal magnesium loss and lower blood levels may be abnormally sensitive to the hypomagnesemic effects of drugs or to the hypomagnesemic states induced by alcoholism or diabetes. Thus, genetic analysis of magnesium concentration as a quantitative trait will benefit from and probably contribute to our understanding of the less common inherited disorders that cause hypomagnesemia, as well as the more common acquired conditions characterized by low magnesium or renal magnesium wasting.

### Hypomagnesemia with Hypercalciuria and Nephrocalcinosis

A distinct syndrome of hypomagnesemia with hypercalciuria and nephrocalcinosis (HHN) has been described. The HHN syndrome is an autosomal-recessive disorder that is characterized by renal magnesium wasting resulting in persistent hypomagnesemia, marked hypercalciuria leading to early nephrocalcinosis (50–55). It is distinguished from other conditions by the absence of infantile hypocalcemic tetany and normal plasma potassium (52). Also characteristic is the multisystem involvement; the most distinctive features are ocular abnormalities including severe myopia, nystagmus, and chorioretinitis. Hearing impairment, tetany, seizures, chondrocalcinosis, rickets, arterial hypertension, and gouty arthritis all have been reported (52,53). The hypomagnesemia is unresponsive to magnesium administration. Renal transplantation corrects the abnormal magnesium and calcium homeostasis.
handling and normalizes serum magnesium and calcium (53). Rodríguez-Soriano et al. (56) postulated that absorption of magnesium and calcium in the loop of Henle is abnormal because chlorothiazide corrects the hypercalciuria, although it was only variably effective in raising the plasma magnesium. Using positional cloning, Simon et al. (6) identified a human gene, claudin 16 (CLDN16; paracellin-1 [PCLN-1]), that codes for a tight junctional protein located in the paracellular pathway of the TAL. Mutations in this gene presumably result in abnormal permeability of the paracellular pathway leading to decreased magnesium and calcium reabsorption (Figure 2). The marked increase in urinary calcium predisposes to renal stone formation and its sequelae. Accordingly, these patients may require early transplantation.

**Inherited Disorders Associated with Abnormal Extracellular Mg\(^{2+}\)/Ca\(^{2+}\) Sensing**

**Autosomal Dominant Hypoparathyroidism.** The Casr plays an important role in controlling calcium and magnesium transport in both the loop and the distal tubule (10,17). Both activating and inactivating mutations of the Casr have been described and are now well characterized (57). Activating mutations (autosomal dominant hypoparathyroidism) are dominant and present clinically as isolated hypocalcemic hypoparathyroidism. Associated hypomagnesemia may be observed in up to half of the patients (58,59). Because the mutant parathyroid and kidney Casr has a lower set-point for plasma Ca\(^{2+}\) and Mg\(^{2+}\), PTH secretion and renal calcium and magnesium reabsorption are suppressed and the disease is characterized by inappropriately low serum PTH and increased calcium and magnesium excretion. Elevated urinary calcium may lead to nephrolithiasis despite increased magnesium excretion. The hypomagnesemia is usually asymptomatic, but significant deficiencies have been reported (59). Some of this variability is due to the heterogeneity of the activating mutations and a corresponding variability in set-point displacement for serum Ca\(^{2+}\) and Mg\(^{2+}\) concentrations.

**Familial Hypocalciuric Hypercalcemia and Neonatal Severe Hyperparathyroidism.** Familial hypocalciuric hypercalcemia (60–63) and neonatal severe hyperparathyroidism result from inactivating heterozygous and homozygous mutations, respectively (57,64,65). Renal excretion of calcium and magnesium is reduced, which leads to hypercalcemia and sometimes hypermagnesemia (60–63). Defective extracellular Casr likely leads to inappropriate absorption of calcium and magnesium in the TAL (10) and magnesium transport in the distal tubule (17). A knockout mouse model displays all of the characteristics of familial hypocalciuric hypercalcemia and neonatal severe hyperparathyroidism, providing experimental evidence to support this idea (66).

Although the human mutations indicate that magnesium regulation is not the primary function of the Casr gene, the possibility exists that specific defects of the sensing mechanism may affect renal magnesium balance. Bapty et al. (17) observed that the sensitivity of divalent sensing may be greater for extracellular Mg\(^{2+}\) than Ca\(^{2+}\) in immortalized mouse DCT cells. The effects of extracellular Ca\(^{2+}\) and Mg\(^{2+}\) were not additive. Whether this is due to selective receptors or ion-specific effects on the same receptor is not clear. The ligand-binding and signal transduction properties of the Ca\(^{2+}\)/Mg\(^{2+}\)-sensing receptor protein are still to be elucidated.

Mehrotra et al. (67) described a 44-yr-old male with marked hypomagnesiuric hypermagnesemia and hypercalciuria but nor-
mocalcemia that they speculated was due to abnormal Ca\(^{2+}/\)Mg\(^{2+}\) sensing. However, the presence of progressive renal failure, the concommitant hypokalemic metabolic alkalosis, and the absence of confirmatory molecular data make an inherited defect of the Casr in this patient less likely.

**Hypomagnesemia Associated With Abnormal NaCl Transport**

Gitelman syndrome and Bartter syndrome are two autosomal-recessive disorders of renal electrolyte transport that have been associated with hypokalemia as a result of renal potassium loss, chloride-resistant metabolic alkalosis, and elevated plasma renin and aldosterone levels but normal BP (Table 2) (68,69). In Gitelman syndrome, hypomagnesemia is a distinctive feature (68), whereas there is doubt as to whether disordered magnesium metabolism is ever significantly abnormal in patients with true Bartter syndrome (70,71).

**Gitelman Syndrome.** Patients with Gitelman syndrome often present in late childhood with a hypokalemic metabolic alkalosis and low serum magnesium, which may be asymptomatic or may be severe enough to cause hypomagnesemic tetany (72,73). Patients are not polyuric or polydipsic but have hypocalciuria and usually show renal magnesium wasting (74). The absence of nephrocalcinosis, which may be best defined by renal ultrasound, therefore is an important diagnostic feature.

Skeletal problems are occasionally observed in children with Gitelman syndrome. Some children without severe renal defects will present with growth retardation as a result of rickets. The other causes of renal rickets can be rapidly excluded by characterizing the abnormality of renal magnesium handling. Bettinelli et al. (75) described a variant form of Gitelman syndrome with intermittent electrolyte abnormalities but severe growth failure. The two affected children were unrelated but had growth hormone deficiency and partial vasopressin insufficiency associated with an empty sella on intracranial imaging. Pituitary screening may be warranted in patients in whom growth failure is a prominent feature. Gitelman syndrome is also characterized by chondrodysplasia (76,77) and rarely rhabdomyolysis that is secondary to severe hypokalemia (78), but the long-term outlook for these patients is good (79). Treatment with oral magnesium corrects the magnesium deficit but not the metabolic alkalosis (80), and careful management of the potassium wasting is important. The relationship of these skeletal abnormalities to magnesium wasting and hypomagnesemia is unclear.

Patients with Gitelman syndrome fail to respond to chlorothiazide, leading to the prediction that the renal defect is in the DCT (81). Simon et al. (82) showed that Gitelman syndrome families are genetically linked to a locus at 16q13 and identified causative mutations in the chlorothiazide-sensitive NaCl cotransporter expressed in the DCT (NCC/SLC12A3). As chlorothiazide enhances calcium reabsorption in this nephron segment, the hypocalciuria of Gitelman syndrome is readily explained (71). The reasons for renal magnesium wasting are unknown (8). We have shown that chlorothiazide stimulates Mg\(^{2+}\) uptake in mouse DCT cells by mechanisms similar to those that increase Ca\(^{2+}\) entry (8). One would predict, therefore, that hypomagnesuria should be the prevailing phenotype, rather than increased renal magnesium excretion. Reilly and Ellison (83) recently postulated another way to explain magnesium wasting of patients with Gitelman syndrome. They suggested that the absence of claudin 16 expression may somehow allow magnesium secretion via the paracellular pathway, thus leading to increased urinary excretion. The notion is that Gitelman syndrome converts some DCT cells that are predominantly electroneutral cells to cells that reabsorb Na\(^{+}\) in an electronegrogenic manner. As discussed by these authors, the cells are also responsive to the actions of aldosterone. Accordingly, the combination of the dominance of electronegrogenic ion transport pathways, the stimulation by aldosterone, and the increased Na\(^{+}\) concentration all favor electronegrogenic Na\(^{+}\) reabsorption that greatly increases the magnitude of the transepithelial voltage (83). The luminal negative voltage drives magnesium secretion. We do not favor this explanation as our earlier micropuncture studies failed to detect any Mg\(^{2+}\) secretion in micropерfused distal tubules (8). By deleting the gene coding for the NaCl cotransporter, Schulteis and colleagues (84) developed a mouse model of Gitelman syndrome. These mice show all of the cardinal features of Gitelman syndrome, including renal magnesium wasting, so this knockout model may be useful in delineating the molecular physiology of this condition.

**Bartter Syndrome.** Patients with infantile Bartter syndrome characteristically present in infancy with a urinary concentrating defect, polyhydramnios, failure to thrive, and fasting hypercalciuria leading to medullary nephrocalcinosis (85). Those with classic Bartter syndrome present in childhood with features of water and salt depletion, including polydipsia, polyuria, and episodes of dehydration. Clinical features may include growth retardation, developmental delay, nephrocalcinosis, and hydrenephrosis as a result of impaired water clearance (25,71). Patients fail to respond normally to furosemide, suggesting to investigators that this disease was due to defective loop function. Using family linkage studies, Simon et al. (86–88) delineated three genetic defects that form the basis for distinguishing three distinct physiologic phenotypes in these patients. Type I Bartter syndrome is due to defective Na-K-C1 cotransport (NKCC2 gene), characterized by severe hypokalemia in addition to the above (86). Type II Bartter syndrome is associated with mutations in a potassium channel (ROMK gene, 30 pS K\(^{+}\) channel) activity (87). As this K\(^{+}\) channel is also present in the cortical collecting duct and is involved in potassium secretion in this segment, hypokalemia is less severe in this form of the disease. Type III Bartter syndrome is based on a mutation in the basolateral membrane chloride channel (CICNKB) (88). Like type I patients, type III patients have severe hypokalemia, but unlike Type I and Type II phenotypes, nephrocalcinosis is not observed.

Additional phenotypes have been reported, and genetic evidence for further heterogeneity has been presented (70,88–92). A form of infantile Bartter’s syndrome that is consistently associated with sensorineural deafness has been described in some Bedouin families (89). Although there is no evidence that the renal defect differs from that observed in isolated Bartter’s syndrome, genetic linkage to chromosome 1p31 tends to exclude any of the three transport defects identified to date (90), despite the proximity to the CICNKB gene.
(1p36) of type III Bartter syndrome (88). Speculation by Brennan et al. (90) that another Na-K-Cl transporter is involved is supported by the recent identification of a specific sensorineural deafness syndrome in knockout mice that lack a second, more widely expressed Na-K-Cl cotransporter gene (91).

It has been suggested that up to 30% of patients with Bartter syndrome may have hypomagnesemia as a result of renal magnesium wasting. However, it is clear that some of these cases represent a form of Gitelman syndrome, and others may be another variant of hypokalemic metabolic alkalosis. In addition, hypomagnesemia does not reliably segregate to any of the three types of Bartter syndrome defined by molecular studies, suggesting that other circumstances may affect renal magnesium absorption. Supporting this notion is evidence that magnesium balance is normal in a Na-K-Cl cotransport knockout mouse mimicking type I Bartter syndrome (93). Chronic use of furosemide is sometimes normal in a Na-K-Cl cotransport knockout mouse mimicking type I Bartter syndrome (93). Chronic use of furosemide is sometimes associated with hypomagnesemia as a result of excessive urinary magnesium excretion, but it is not universal (8). Thus, it is not apparent why most individuals with Na-K-Cl cotransport defects should be comparatively free of renal magnesium wasting. It may be that the loop and distal tubule adapt to conserve magnesium in this disorder (1,8).

Conclusion

In summary, the molecular and genetic basis of various inherited disorders that affect renal magnesium handling are being delineated. These conditions affect different nephron segments and different cell types, which leads to variable but increasingly distinguishable phenotypic presentations. Undoubtedly, there are other disorders in the general population that have not yet been identified or characterized. The continued use of molecular techniques to probe the constitutive and congenital disturbances of magnesium metabolism will increase our understanding of cellular magnesium transport and provide new insights into the way these diseases are diagnosed and managed.

Acknowledgments

We thank Iwan C. Meij for reading parts of this review. This work was supported by research grants from the Medical Research Council of Canada (MT-5793) and from the Kidney Foundation of Canada to G.A. Quamme and from NSERC and Dairy Farmers of Canada to D.E.C. Cole.

References

12. Rude RK, Ryzen E: TmMg and renal Mg threshold in normal man and in certain pathophysiologic conditions. Magnesium 5: 273–281, 1986


