CASE RECORDS of the MASSACHUSETTS GENERAL HOSPITAL

Case 21-2024: A 10-Month-Old Boy with Vomiting and Hypercalcemia

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PRESENTATION OF CASE

Dr. Andrew F. Cohen (Pediatrics): A 10-month-old boy was admitted to this hospital because of vomiting, weight loss, and hypercalcemia.

The patient had been well until 2 weeks before this admission, when projectile vomiting began to occur once every few days, typically after eating solid foods. The vomiting did not seem to bother the patient, and he immediately returned to his normal activity after each episode. During the subsequent 12 days, the vomiting became more frequent, occurring daily, and had begun to occur after drinking liquids as well.

Two days before this admission, the patient was evaluated in the pediatric primary care clinic of this hospital. On examination, the weight was 7.36 kg (0.4th percentile for his age) (Fig. 1). The patient appeared alert, attentive, and comfortable but thin. The abdomen was soft, without distention or tenderness, and there was no evidence of organomegaly or masses on palpation. The skin was diffusely dry with excoriations. Treatment with omeprazole was started.

Over the course of the next 2 days, the patient lost interest in eating solid food and preferred milk and water. He gagged at the sight of food. The frequency of the vomiting had increased, occurring after the patient ate or drank anything.

On the day of this admission, the patient refused to take any food by mouth. His parents brought him to the emergency department of this hospital. On arrival, his parents reported that in the previous 2 weeks, he had had normal activity, wet diapers, and daily soft stools. They also noted that he always seemed thirsty, and when given liquid, he would drink a lot. At times, he would even suck the water out of the washcloths that were used to bathe him.

The patient had been born after induction of labor at 38 weeks' gestation owing to maternal gestational hypertension. He had received routine newborn care and met normal developmental milestones. He maintained normal growth for his age until an evaluation at 9 months of age, when the weight was 7.44 kg (1.6th per-

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CME

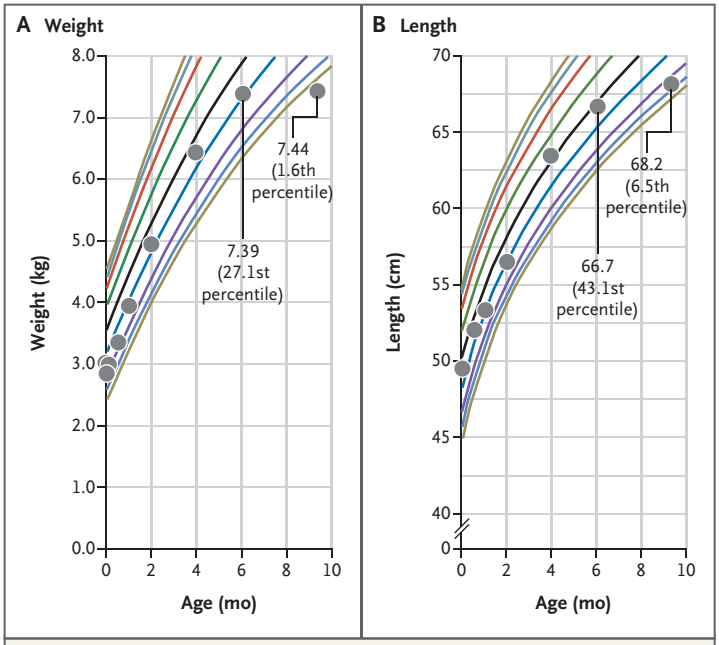

Figure 1. Growth Curves.

The growth curves show the patient's body weight (Panel A) and length (Panel B) relative to the average values for boys in his age group. The solid lines indicate growth percentile, and the gray dots indicate growth points before and at the time of this evaluation. The patient had shown normal growth until 9 months of age, when the rate of weight gain and length increase leveled abruptly. At 6 months of age, the weight was 7.39 kg (27.1st percentile) and the length 66.7 cm (43.1st percentile); at 9 months of age, the weight was 7.44 kg (1.6th percentile) and the length 68.2 cm (6.5th percentile).

centile), and the body length was 68.2 cm (6.5th percentile) (Fig. 1). At a routine evaluation when he was 6 months of age, the weight was 7.39 kg (27.1st percentile), and the length was 66.7 cm (43.1st percentile). The patient had a history of gastroesophageal reflux disease (GERD) and eczema. Medications included omeprazole; he was not receiving any vitamins or supplements. He had no known drug allergies, and immunizations were up to date.

The patient lived with his mother and father in a suburban area of New England. His father had GERD and melanoma, and cardiomyopathy had led to heart transplantation 9 years earlier. His mother was well. The patient's paternal grandmother had the Lynch syndrome and hyperparathyroidism. His maternal grandmother had diabetes and coronary artery disease, his maternal grandfather had hypertension and eczema, and his maternal uncle had the DiGeorge syndrome.

On examination, the temporal temperature was 36.1°C, the blood pressure 102/78 mm Hg, the pulse 140 beats per minute, and the respiratory rate 40 breaths per minute. The patient was alert and appropriately interactive. He appeared thin. The abdomen was soft, and there was no distension or tenderness. The muscle tone was normal.

The blood calcium level was 13.2 mg per deciliter (3.3 mmol per liter; reference range, 8.5 to 10.5 mg per deciliter [2.1 to 2.6 mmol per liter]). Other laboratory test results are shown in Table 1. Radiographs of the wrists and knees were normal. The dose of omeprazole was increased. Treatment with famotidine and intravenous fluids was started. The patient was admitted to this hospital.

During the subsequent two hospital days, the patient was fed formula, and emesis continued to occur. Additional laboratory test results were obtained (Table 1). Ultrasonography of the abdomen showed a normal pylorus and no evidence of intussusception. Medullary nephrocalcinosis was present in both kidneys. An upper gastrointestinal series showed intermittent spasm of the gastric pylorus without evidence of hypertrophic pyloric stenosis.

A diagnostic test was performed.

DIFFERENTIAL DIAGNOSIS

Dr. Christina Jacobsen: This previously healthy 10-month-old boy presents with vomiting and hypercalcemia. The clinical manifestations of hypercalcemia are similar regardless of age and include gastrointestinal symptoms such as constipation, anorexia, and vomiting.[1] In this patient, vomiting began to occur 2 weeks before admission, and a clinically significant worsening of vomiting developed in the 2 days before admission. This is a common presentation for hypercalcemia because vomiting leads to dehydration, which worsens the hypercalcemia, and ultimately results in more vomiting. Anorexia resulting from hypercalcemia could explain this patient's weight loss (Fig. 1). We know that the hypercalcemia had been relatively long-standing, given the finding of nephrocalcinosis.[2]

On hospital day 3, the patient's parathyroid hormone (PTH) level was 6 pg per milliliter, which is appropriately suppressed, considering the degree of hypercalcemia (Fig. 2).[4] This finding is helpful in developing a differential diag-

Table 1. Laboratory Data.*

Variable	Reference Range, This Hospital†	On Admission	Hospital Day 3
White-cell count (per μl)	6000–17,500	17,030	—
Hemoglobin (g/dl)	10.5–13.5	12.3	—
Hematocrit (%)	33.0–39.0	37.4	—
Platelet count (per μl)	150,000–400,000	494,000	—
Sodium (mmol/liter)	135–145	142	142
Potassium (mmol/liter)	3.4–5.0	4.2	4.1
Chloride (mmol/liter)	98–106	104	104
Carbon dioxide (mmol/liter)	22–27	22	21
Urea nitrogen (mg/dl)	5–20	16	7
Creatinine (mg/dl)	0.30–1.00	0.34	0.35
Glucose (mg/dl)	70–110	78	87
Magnesium (mg/dl)	1.7–2.4	—	2.4
Phosphorus (mg/dl)	4.5–6.7	—	3.6
Calcium (mg/dl)	8.5–10.5	13.2	12.2
Ionized calcium (mmol/liter)	1.14–1.30	—	1.47
1,25-Dihydroxyvitamin D (pg/ml)	24–86	—	37
25-Hydroxyvitamin D (ng/ml)	20–80	—	64
Thyrotropin (μIU/ml)	0.40–5.00	1.92	—
Parathyroid hormone (pg/ml)	10–60	—	6
Aspartate aminotransferase (U/liter)	9–80	34	—
Alanine aminotransferase (U/liter)	10–55	17	—
Alkaline phosphatase (U/liter)	122–469	190	—
Total bilirubin (mg/dl)	0.0–1.0	0.2	—
Albumin (g/dl)	3.3–5.0	4.6	—
Globulin (g/dl)	1.9–4.1	2.3	—

* To convert the values for urea nitrogen to millimoles per liter, multiply by 0.357. To convert the values for creatinine to micromoles per liter, multiply by 88.4. To convert the values for glucose to millimoles per liter, multiply by 0.05551. To convert the values for magnesium to millimoles per liter, multiply by 0.4114. To convert the values for phosphorus to millimoles per liter, multiply by 0.3229. To convert the values for calcium to millimoles per liter, multiply by 0.250. To convert the values for 1,25-dihydroxyvitamin D to picomoles per liter, multiply by 2.4. To convert the values for 25-hydroxyvitamin D to nanomoles per liter, multiply by 2.496. To convert the values for bilirubin to micromoles per liter, multiply by 17.1.

† Reference values are affected by many variables, including the patient population and the laboratory methods used. The ranges used at Massachusetts General Hospital are for adults who are not pregnant and do not have medical conditions that could affect the results. They may therefore not be appropriate for all patients.

nosis, since it indicates PTH-independent hypercalcemia.[3] Other laboratory test results included normal levels of 25-hydroxyvitamin D and 1,25-dihydroxyvitamin D.[2] On the basis of the patient's presentation and these laboratory findings, I will first explore possible endocrinopathies, including thyrotoxicosis, severe congenital hypothyroidism, adrenal insufficiency, and inborn errors of metabolism (Fig. 2).[1,3]

ENDOCRINOPATHIES

Thyroid disorders are thought to cause hypercalcemia through the increased turnover of bone.[5] Such disorders would not apply to this patient, who had a normal newborn screening and a normal alkaline phosphatase level with no signs of increased bone turnover. Adrenal insufficiency is also an unlikely cause, given that the patient had a prolonged period of vomiting with no evi-

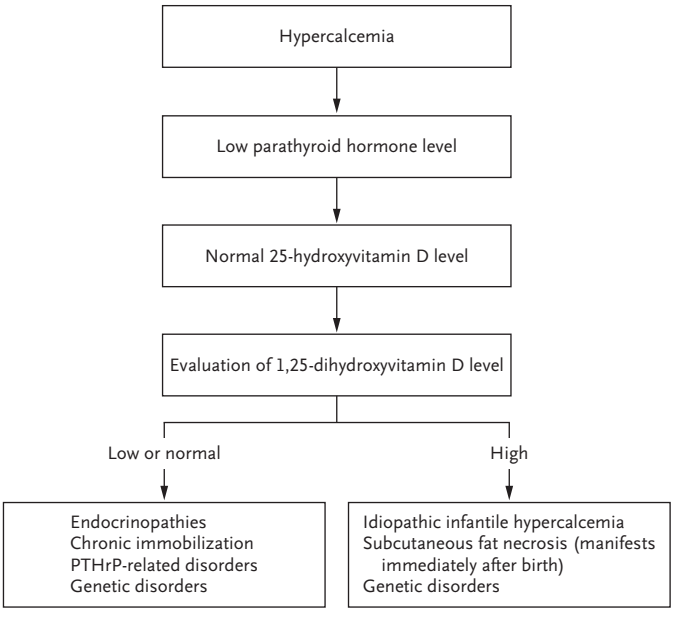

Figure 2. Differential Diagnosis for PTH-Independent Hypercalcemia.
Shown is a schematic diagram of the differential diagnosis for parathyroid hormone (PTH)–independent hypercalcemia, which has been simplified to include only diagnoses that would be appropriate for pediatric patients in the first year of life. After the patient is found to have hypercalcemia and a low PTH level, decision making is guided by the evaluation of the 25-hydroxyvitamin D and 1,25-dihydroxyvitamin D levels.[3] If the 25-hydroxyvitamin D level is high, vitamin D intoxication is included in the differential diagnosis. PTHrP denotes PTH-related peptide.

dence of hypotension and normal levels of electrolytes.[6,7] An inborn error of metabolism would be an extremely unlikely diagnosis in an otherwise normally developing infant who had been able to withstand a period of minimal oral intake.[8] Prolonged immobilization can also lead to hypercalcemia,[9,10] but there is no evidence that this occurred in this patient.

PTH-RELATED PEPTIDE DISORDERS

Although the PTH level in this patient was appropriately suppressed, the possibility of PTH-related peptide (PTHrP)–mediated hypercalcemia should be considered (Fig. 2).[11] PTHrP can be secreted by some tumors; however, the occurrence of such tumors would be extremely rare in a patient of this age. Types of PTHrP-secreting tumors that have been reported in young children include sarcomas (especially fibrosarcoma), acute lymphocytic leukemia, and familial pheochromocytoma.[3] No physical, laboratory, or radiographic evidence was found to support a diagnosis of any of these conditions in this patient. Congenital anomalies of the kidney and urinary tract (CAKUT) have also been reported to cause elevated levels of PTHrP.[12] This patient has no history of urinary tract infections or other findings that would indicate such an anomaly.

GENETIC DISORDERS

In very young patients, genetic disorders are found to be the cause of hypercalcemia more often than acquired disorders (Fig. 2).[3] Genetic disorders that can lead to hypercalcemia include hypophosphatasia, Jansen's metaphyseal chondrodysplasia, and the Williams syndrome.

Hypophosphatasia

Hypophosphatasia is a rare genetic disorder caused by variants in *ALPL*, the gene that encodes tissue-nonspecific alkaline phosphatase.[13] Expression of *ALPL* assists in the mineralization of bone. Both recessive and dominant variants in *ALPL* can cause hypophosphatasia, but only the severe infantile forms that are caused by recessive variants typically manifest in patients with hypercalcemia within the first year of life.[14] Hyperphosphatemia usually occurs with hypercalcemia.[13,14] This patient not only had a slightly low phosphorus level but also had normal findings on bone radiography with no evidence of mineralization defects, which would be expected in a patient with hypophosphatasia.

Jansen's Metaphyseal Chondrodysplasia

Jansen's metaphyseal chondrodysplasia is a rare skeletal disorder characterized by abnormal endochondral bone formation.[15] This disorder is caused by heterozygous (dominant) activating variants in the PTH–PTHrP receptor encoded by *PTH1R*.[16] Due to these variants, patients have hypercalcemia and hypophosphatemia as in hyperparathyroidism, but these changes occur despite reduced or undetectable PTH levels.[17] Short stature is also a common feature of this disorder, and patients typically have a normal birth weight.[17] Metaphyseal changes are observed on radiography, and bowing of the legs is seen soon after walking begins.[15] This patient had no signs of metaphyseal changes on radiography, so a diagnosis of Jansen's metaphyseal chondrodysplasia can be ruled out.

The Williams Syndrome

The Williams syndrome is one of several recurrent microdeletion syndromes, the most well-known of which is the 22q11 deletion syndrome. The cause of hypercalcemia in patients with the Williams syndrome is not well understood, but it is generally thought to be related to increased calcium absorption from dietary sources.[18] The Williams syndrome is caused by a 1.5- to 1.8-Mb deletion on chromosome 7 in the 7q11.23 region.[19] Patients typically present with intrauterine growth restriction and cardiac abnormalities, most commonly peripheral pulmonary stenosis or supravalvular aortic stenosis.[20] Developmental delay is also very common, including delayed milestones.[19] This patient did not have intrauterine growth restriction, no findings were consistent with cardiac disease on examination, and no evidence of developmental delay was present, which together make the Williams syndrome an unlikely diagnosis. However, it is always prudent to consider and rule out the Williams syndrome in cases of infantile hypercalcemia, given the difficulty of making the diagnosis on the basis of physical examination alone in very young children.[20]

IDIOPATHIC INFANTILE HYPERCALCEMIA

This patient had a 25-hydroxyvitamin D level of 64 ng per deciliter, which is in the upper end of the normal range and unusual for an infant living in New England, with limited sun exposure owing to the geographic location, and who is not receiving any supplementation.[21] Although it is rare to have a high-normal level in this region, the patient's level of 25-hydroxyvitamin D is certainly not in the range that would lead to hypercalcemia owing to the toxic effects of vitamin D.[22] In addition, although the level of 1,25-dihydroxyvitamin D was in the normal range, it could still be considered to be elevated in a patient with a suppressed PTH level and hypercalcemia (Fig. 2). PTH is required to activate the 1α-hydroxylase enzyme that produces 1,25-dihydroxyvitamin D.[21] Therefore, a "normal" level of 1,25-dihydroxyvitamin D in the absence of an elevated PTH level is unusual. When we expand the differential diagnosis to consider disorders that include a low level of PTH but an elevated level of 1,25-dihydroxyvitamin D, we are left with the diagnosis of idiopathic infantile hypercalcemia.[3]

Idiopathic infantile hypercalcemia has long been a clinical diagnosis of exclusion in infants when all other causes of hypercalcemia have been ruled out.[3] In the age of widespread genetic testing, research has shown that many of these cases are caused by genetic variants in the vitamin D pathway that increase calcium absorption.[23-25] Idiopathic infantile hypercalcemia can be caused by variants in several genes, but the most common is *CYP24A1*, the gene encoding the 24-hydroxylase enzyme.[23-25] 24-Hydroxylase is responsible for converting 25-hydroxyvitamin D and 1,25-dihydroxyvitamin D to 24,25-dihydroxyvitamin D and 1,24,25-trihydroxyvitamin D (calcitroic acid), respectively, both of which are inactive and can be excreted through the kidneys (Fig. 3).[21,22] This excretion prevents the toxic effects of vitamin D, including the main associated complications — hypercalcemia and nephrocalcinosis.[22] Recessive variants in *CYP24A1* have been shown to cause idiopathic infantile hypercalcemia.[24] Unsurprisingly, patients typically have higher-than-expected levels of vitamin D owing to the reduced ability to excrete 25-hydroxyvitamin D.[23] This scenario is certainly consistent with this patient's presentation. Patients with idiopathic infantile hypercalcemia typically present with hypercalcemia and nephrocalcinosis or calcium-containing renal stones.[25,26] Other findings on physical examination or dysmorphic features are absent, which is also consistent with the findings in this patient. Development is usually normal.[26] In most cases, there is a frankly elevated level of 1,25-dihydroxyvitamin D; however, inappropriately normal levels, in relation to the degree of hypercalcemia and appropriately suppressed PTH level, have been reported.[23-25] A diagnosis is usually made either by measuring the ratio of 25-hydroxyvitamin D to 24,25-dihydroxyvitamin D or by sequencing *CYP24A1*.[23]

DR. CHRISTINA JACOBSEN'S DIAGNOSIS

Hypercalcemia caused by *CYP24A1* variants.

DIAGNOSTIC TESTING

Dr. Harald Jüppner: On hospital day 3, the level of 25-hydroxyvitamin D was 64 ng per milliliter, and the level of 1,25-dihydroxyvitamin D (the

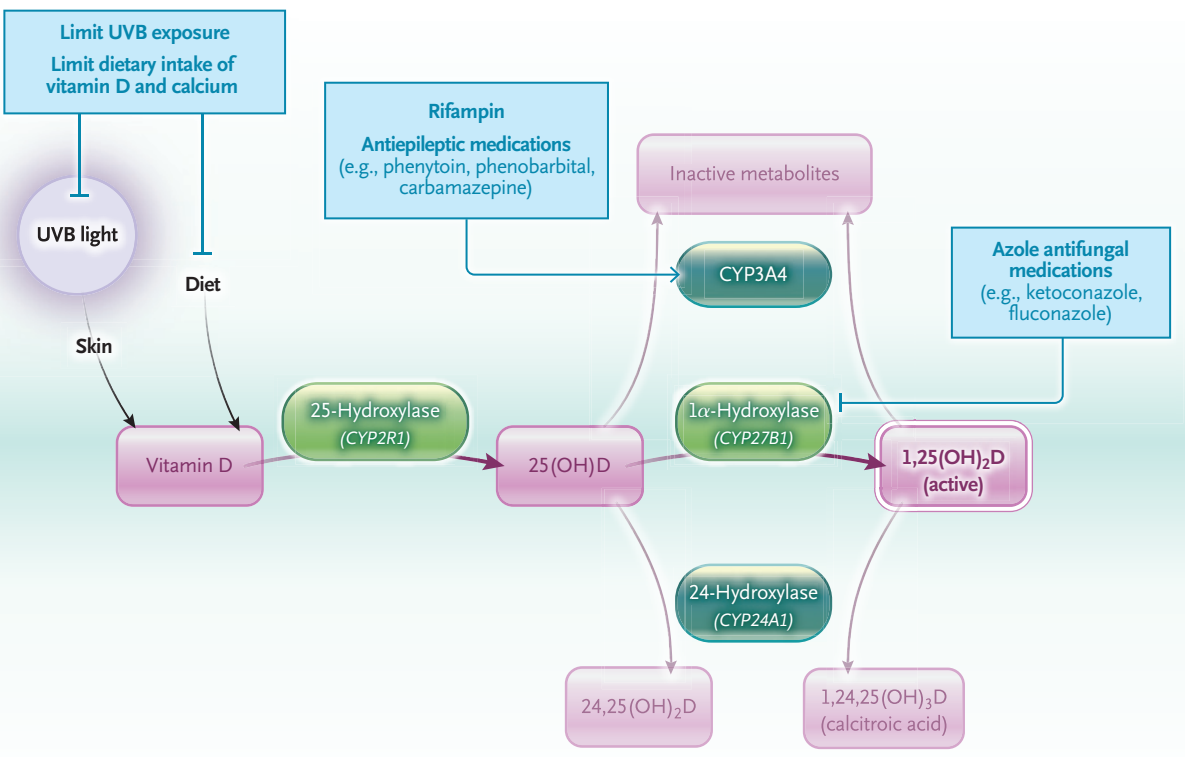

Figure 3. Metabolism of Vitamin D.

Vitamin D is converted to 25-hydroxyvitamin D (25[OH]D) by a 25-hydroxylase and then to the active form 1,25-dihydroxyvitamin D ($1,25[OH]_2D$) by a 1α-hydroxylase encoded by *CYP27B1*. Both 25-hydroxyvitamin D and 1,25-dihydroxyvitamin D can be inactivated either by the 24-hydroxylase enzyme encoded by *CYP24A1* or by oxidation by CYP3A4. Nonpharmacologic therapies for 24-hydroxylase deficiency include restricting dietary intake of calcium and vitamin D, as well as avoiding endogenous production of vitamin D through limiting exposure to ultraviolet B (UVB) light. Azole antifungal medications, such as ketoconazole and fluconazole, can reduce the level of circulating 1,25-dihydroxyvitamin D by inhibiting CYP27B1. Rifampin, certain antiepileptic medications, and other inducers of CYP3A4 can accelerate the breakdown of 25-hydroxyvitamin D and 1,25-dihydroxyvitamin D through stimulation of this alternative catabolic pathway. $1,24,25(OH)_3D$ denotes 1,24,25-trihydroxyvitamin D.

biologically active form of vitamin D) was 37 pg per milliliter, both of which were within normal limits. However, when the measurement was repeated 2 weeks later, the 1,25-dihydroxyvitamin D level was elevated (at 120 pg per milliliter).

Precursor 25-hydroxyvitamin D and 1,25-dihydroxyvitamin D undergo hydroxylation by 24-hydroxylase (CYP24A1) to generate 24,25-dihydroxyvitamin D and 1,24,25-trihydroxyvitamin D, respectively, thereby preventing excessive levels of 1,25-dihydroxyvitamin D. Only the level of 24,25-dihydroxyvitamin D is measured routinely and can therefore serve as an indicator of CYP24A1 bioactivity. On hospital day 5, the 25-hydroxyvitamin D level was again within normal limits, at 68 ng per milliliter, but the 24,25-dihydroxyvitamin D level had decreased to 0.4 ng per milliliter (reference range, 1.6±1)[27]; however, this result did not become available until 12 days after admission. The calculated ratio of 25-hydroxyvitamin D to 24,25-dihydroxyvitamin D was elevated (170 ng per ng; reference range, 7 to 35), which suggests that the patient's hypercalcemia might be a result of impaired metabolism of both 25-hydroxyvitamin D and 1,25-dihydroxyvitamin D. This possibility would be consistent with hypercalcemia due to biallelic variants in *CYP24A1*, since patients with this condition have reduced levels of 24,25-dihydroxyvitamin D, which result in elevated ratios

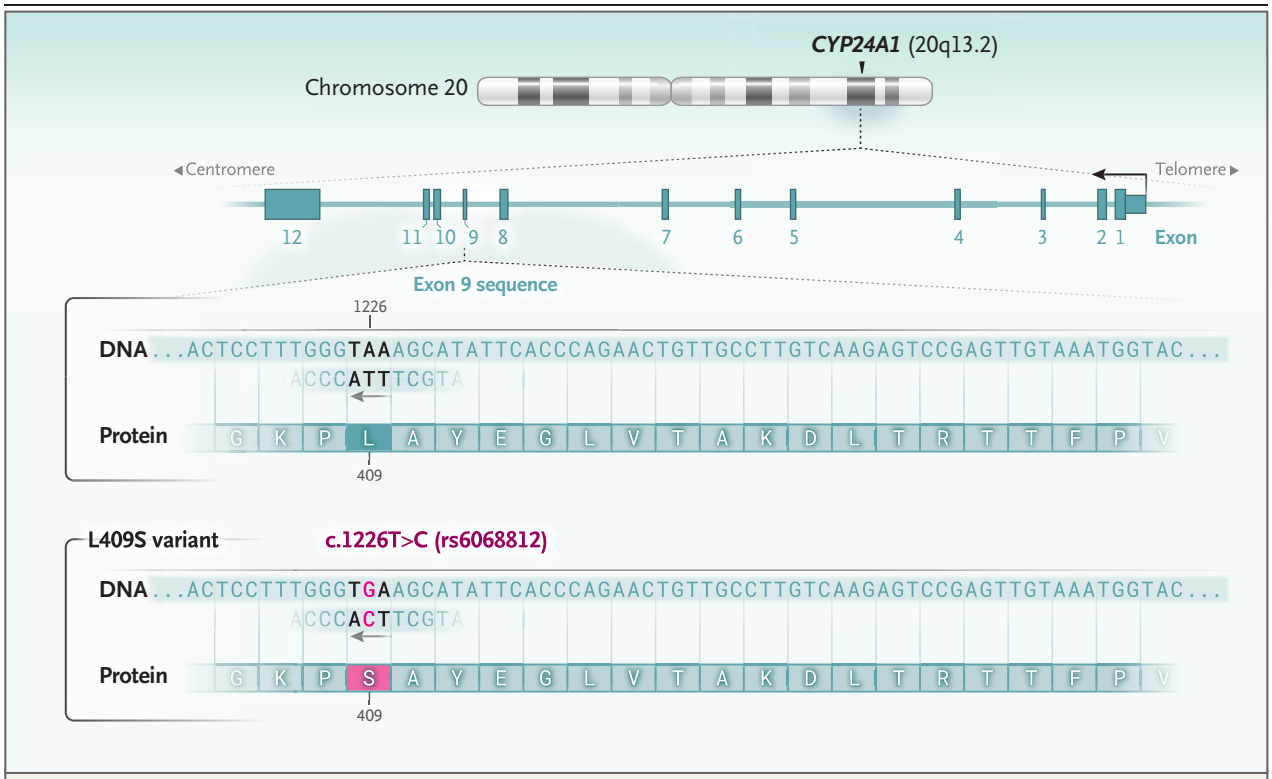

Figure 4. *CYP24A1*.

CYP24A1, the gene encoding the 24-hydroxylase enzyme, is located on the long arm of chromosome 20. It comprises 12 exons that are transcribed in the opposite direction (i.e., telomeric to centromeric). Thus, leucine (Leu, L) at position 409 is encoded by TTA on the complementary strand and the T-to-C change at nucleotide position c.1226 (rs6068812) that was identified in the patient generates codon TCA, which encodes serine (Ser, S).

of 25-hydroxyvitamin D to 24,25-dihydroxyvitamin D.[2] Likewise, hypercalcemia due to a *CYP24A1* variant impairs metabolism of 1,25-dihydroxyvitamin D into 1,24,25-trihydroxyvitamin D, which leads to increased levels of biologically active 1,25-dihydroxyvitamin D and, consequently, increased intestinal calcium absorption.[28]

Hypercalcemia should lead to reduced expression of *CYP27B1*, the gene encoding the 1α-hydroxylase that generates 1,25-dihydroxyvitamin D from its precursor 25-hydroxyvitamin D. However, on hospital day 3, the 1,25-dihydroxyvitamin D level of 37 pg per milliliter was inappropriately normal, considering the severity of hypercalcemia. Levels of 1,25-dihydroxyvitamin D within the normal range have been reported in other patients who had hypercalcemia with biallelic pathogenic variants in *CYP24A1*.[28,29] Furthermore, mice that are null for *Cyp24a1* have lower levels of 1,25-dihydroxyvitamin D than wild-type controls.[30] Thus, normal levels of the biologically active vitamin D metabolite do not rule out a biallelic pathogenic variant in *CYP24A1* in patients with hypercalcemia, as in this case. Once the hypercalcemia had abated in response to different medical interventions, the 1,25-dihydroxyvitamin D level had increased to 120 pg per milliliter, which is well above the reference range. These findings, combined with the elevated ratio of 25-hydroxyvitamin D to 24,25-dihydroxyvitamin D, made it plausible that the hypercalcemia in this patient was caused by biallelic inactivating variants in *CYP24A1*.

Consistent with this prediction, nucleotide sequence analysis of *CYP24A1* revealed a homozygous thymine-to-cytosine change at position c.1226 in exon 9, which changes a conserved leucine at amino acid position 409 to serine (L409S) (Fig. 4). The same variant, either homozygous or compounded with another pathogenic variant in *CYP24A1*, was previously reported in several other patients with hypercalcemia result-

ing from a 24-hydroxylase deficiency[29,31,32] On the basis of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology framework (i.e., previously established pathogenicity of the L409S variant), the biallelic *CYP24A1* variant that was identified in this patient was probably responsible for his severe hypercalcemia.

Previous cell-based studies have shown how 1,25-dihydroxyvitamin D is hydroxylated by the wild-type CYP24A1 enzyme to generate 1,24,25-trihydroxyvitamin D, which is then metabolized further.[29] This in vitro approach showed that different variants of the CYP24A1 protein, encoded by pathogenic *CYP24A1* variants identified in patients with hypercalcemia, failed to generate metabolites of 1,25-dihydroxyvitamin D. The L409S variant of *CYP24A1* showed some residual enzymatic activity, thus allowing the generation of small amounts of 1,24,25-trihydroxyvitamin D and other metabolites. However, the residual activity of the variant CYP24A1 enzyme was inadequate to sufficiently reduce the level of circulating 1,25-dihydroxyvitamin D and to prevent hypercalcemia.[29]

The parents of the patient have not yet been tested for the L409S-encoding variant in *CYP24A1*, but both parents are probably heterozygous carriers of the identified genetic defect. Therefore, 25% of their children would be predicted to be homozygous for the *CYP24A1* variant and, as a result, would be at risk for the development of severe hypercalcemia. Genetic counseling is therefore advised, and any future siblings of this patient should undergo genetic testing.

GENETIC DIAGNOSIS

24-Hydroxylase deficiency due to a homozygous, loss-of-function variant in *CYP24A1*.

DISCUSSION OF MANAGEMENT

Dr. Deborah M. Mitchell: The mainstay of the acute management of hypercalcemia is saline diuresis.[33] Historically, loop diuretic agents have been used to promote calciuresis, but this approach has fallen out of favor owing to the risk of volume depletion and paradoxical worsening of hypercalcemia.[34] Additional agents, including calcitonin and bisphosphonates, can be considered if saline infusion is insufficient to normalize the blood calcium level.[3]

This patient was initially treated with an infusion of normal saline containing 5% dextrose. In addition, his diet was transitioned to a specialized metabolic infant formula that contained a low amount of calcium and no vitamin D. By hospital day 7, the blood calcium level had decreased to 10.1 mg per deciliter (2.5 mmol per liter), and his symptoms had abated. Intravenous fluids were discontinued; however, within 4 days after this measurement was obtained, his calcium level had increased to 12.0 mg per deciliter (3.0 mmol per liter). One dose of zoledronic acid was then administered, which resulted in a decrease in the blood calcium level to 8.8 mg per deciliter. The patient was discharged to home with a plan for close follow-up.

The primary goal of therapy for patients with 24-hydroxylase deficiency is to prevent symptomatic hypercalcemia, which manifests with lethargy, decreased appetite, nausea and vomiting, constipation, and weight loss (Fig. 1),[3] as was seen in this patient. Hypercalcemia can lead to polyuria and polydipsia by inducing resistance to arginine vasopressin (formerly called nephrogenic diabetes insipidus).[35] In addition, it is critical to prevent or minimize hypercalciuria, since 24-hydroxylase deficiency confers a substantial risk of nephrolithiasis, nephrocalcinosis, and chronic kidney disease, which can progress to end-stage kidney failure.[31,36] Additional complications that may be associated with 24-hydroxylase deficiency include calcification of the vasculature and of the joint spaces.[37,38]

To date, targeted therapies for the long-term management of 24-hydroxylase deficiency are lacking. Given that the pathophysiology of this disorder involves elevations in the level of 1,25-dihydroxyvitamin D (the active form of vitamin D) that lead to increased gut absorption of calcium, nonpharmacologic therapies include limiting dietary intake of calcium and vitamin D and avoiding endogenous production of vitamin D through assiduous protection from sunlight.[39] It is also prudent to encourage good hydration in order to decrease the risk of recurrent formation of kidney stones.[40]

Overall, the patient did well with the treatment he received. His blood and urinary calcium levels were closely monitored, and levels outside

the normal range were rare. His care was complicated by a national shortage of infant formula that particularly affected specialty formula.[41] This shortage led to a transition back to regular infant formula soon after hospital discharge and then an early wean from formula. He was seen by a nutritionist in the pediatric endocrinology clinic to ensure adequate macronutrient and micronutrient intake in the context of his restricted diet.

Azole antifungal medications, which inhibit 1α-hydroxylase and thus decrease production of 1,25-dihydroxyvitamin D, have been shown to be effective in treating hypercalcemia associated with *CYP24A1* variants in small studies[31,42-44]; however, the use of these medications is limited because of the associated hepatic toxic effects. An alternative catabolic pathway for both 25-hydroxyvitamin D and 1,25-dihydroxyvitamin D that is mediated by CYP3A4, an inducible cytochrome P450, has been identified (Fig. 3).[45,46] It is important to note that this alternative pathway can be induced by medications including rifampin and many antiepileptic drugs, which provides a molecular explanation for the low blood levels of 25-hydroxyvitamin D and the drug-induced osteomalacia that has been observed in patients taking these medications.[47] Proof of the importance of this pathway in human physiology has been described in two patients with low levels of circulating 25-hydroxyvitamin D and severe rickets. These patients had resistance to standard therapeutic doses of vitamin D and were found to have activating variants in *CYP3A4*.[48] These observations prompted the off-label use of rifampin for the treatment of hypercalcemia and hypercalciuria in two patients with 24-hydroxylase deficiency,[27] which resulted in improvements in calcium levels in both blood and urine. An ongoing clinical trial sponsored by the National Institutes of Health (ClinicalTrials.gov number, NCT03301038) is further investigating this approach. The patient's family elected to enroll him in this trial.

After completion of the clinical trial, the patient continued to receive rifampin therapy. He eats an unrestricted diet, and his blood calcium level has stayed in the normal range. Random urine samples have shown that the ratio of calcium to creatinine has been intermittently elevated; however, the most recent ratio, calculated at 31 months of age, was normal (0.02; reference value, <0.2). His growth, which was also evaluated at 31 months of age, had rebounded; his weight was at the 10th percentile and length was at the 22nd percentile for age. Development of language and motor skills is appropriate for his age. The nephrocalcinosis is persistent and stable.

FINAL DIAGNOSIS

24-Hydroxylase deficiency due to a homozygous *CYP24A1* variant.

This case was presented at the 2023 Harvard Medical School postgraduate course "Primary Care Pediatrics," directed by Drs. Peter T. Greenspan, Benjamin A. Nelson, John Patrick T. Co, Janice A. Lowe, and Ronni L. Goldsmith.

Disclosure forms provided by the authors are available with the full text of this article at NEJM.org.

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