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Syndrome of Amelogenesis Imperfecta, Nephrocalcinosis, Impaired Renal Concentration, and Possible Abnormality of Calcium Metabolism

Mark Lubinsky, Carol Angle, P. Wayne Marsh, and C.J. Witkop, Jr.

Childrens Memorial Hospital, Omaha, Nebraska (M.L.), Department of Pediatrics, University of Nebraska Medical Center, Omaha, NE (C.A., P.W.M.), and University of Minnesota, Minneapolis, Minnesota (C.J.W.)

We describe a brother and sister with amelogenesis imperfecta, nephrocalcinosis and impaired renal concentrating ability. This is the second sibship reported, further substantiating autosomal recessive inheritance of this condition. There is lack of enamel, lifelong nocturnal enuresis, progressive punctate nephrocalcinosis, and decreased calcium and phosphate excretion over 24 hours and after an acute load. Increased serum osteocalcin and decreased urine δ-carboxyglutamic acid suggest involvement of vitamin K-dependent calcium binding proteins, although this may represent a secondary finding. No other evidence of abnormal calcium metabolism was found. Renal function is stable in the early teens, but the previously reported patients went on to renal failure.

Key words: Amelogenesis imperfecta, autosomal recessive inheritance, calcium binding proteins (vitamin K-dependent), concentration defect, enuresis, δ-carboxyglutamic acid, hypocalciuria, hypophosphaturia, nephrocalcinosis, osteocalcin, polyuria

INTRODUCTION

Hypoplastic amelogenesis imperfecta can occur per se or in a variety of ectodermal syndromes and metabolic disorders [Witkop and Sauk, 1976; Catena et al, 1970; Edward and Nord, 1974; Chosack et al, 1979; Frank and Bolender, 1962; Wennström, 1963; Bergman et al, 1964; Witkop et al, 1975; Nikiforouk and Fraser, 1979; Köhlschütter et al, 1974; Jorgenson and Warson, 1973; Koshiba et al, 1978].

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Address reprint requests to Dr. Mark Lubinsky, Clinical Genetics Center, Childrens Memorial Hospital, 8301 Dodge Street, Omaha, Nebraska 68114.

In 1972, MacGibbon reported a brother and sister with absent enamel, nephrocalcinosis, and apparently normal calcium metabolism. We have studied a second sibship, confirming the existence of this syndrome. Abnormalities of calcium-binding proteins suggest a pathogenetic basis for this unusual disorder, but may be secondary disturbances.

CLINICAL REPORTS

A brother (P.M.) and sister (S.M.), ages 11 and 9 years respectively, presented with enuresis and dental problems.

Both had achieved some daytime bladder control by 2 years, but nocturnal enurersis continued. Intermittent urinary tract infections began at 5 years and continued for several years. Later urines have been sterile. Enuresis and polyuria continue, worse in the brother.

Their lower incisors erupted at 5 to 6 months and they had a full set of primary teeth by 2½ years. The mother described these as yellow-white in color and gradually darkening with age, but did not consider them unusual. There were no complaints of unusual sensitivity to foods or thermal changes. Permanent maxillary and mandibular central incisors erupted by 7 to 7½ years of age. These were yellow-brown, did not contact each other proximally, and appeared to have short crowns. The parents became concerned when the remaining permanent teeth failed to erupt at the expected time and the incisors showed marked wear. Their dentists told them that the teeth lacked enamel completely.

Aside from one broken arm in the girl, there had been no fractures. Mental and motor development and growth parameters have all been normal.

Family History

The father is Czechoslovakian, and the mother of Swedish ancestry; they are not consanguineous. They were 23 and 21 years old at the time of the birth of the boy. Both pregnancies were unremarkable, and there are no other children. The father and a maternal aunt had delayed nocturnal bladder control, but there is no other known urinary tract problem in the family. Neither parent has defective enamel on examination.

On examination, vital signs, blood pressure, height, weight, and head circumferences were all normal. There were no orthopedic deformities. Dentition was abnormal (see below) and the mandibles small.

Bone age and density, skeletal and soft tissue surveys, and slit-lamp examination were normal in both.

Oral Examination

P.M.: At 12 years only the canines and incisors had erupted (Fig. 1). There was no clinically detectable enamel on any tooth. The color of the crowns varied from yellow-brown in protected areas to brown in exposed areas where extrinsic staining or dental caries had occurred. The crowns of the teeth did not meet at the proximal contact points. The crown surfaces were slightly rough, could be penetrated by a sharp explorer with firm pressure, and had the general consistency of dentin. Areas of attrition were present on the incisal and occlusal surfaces of most teeth.

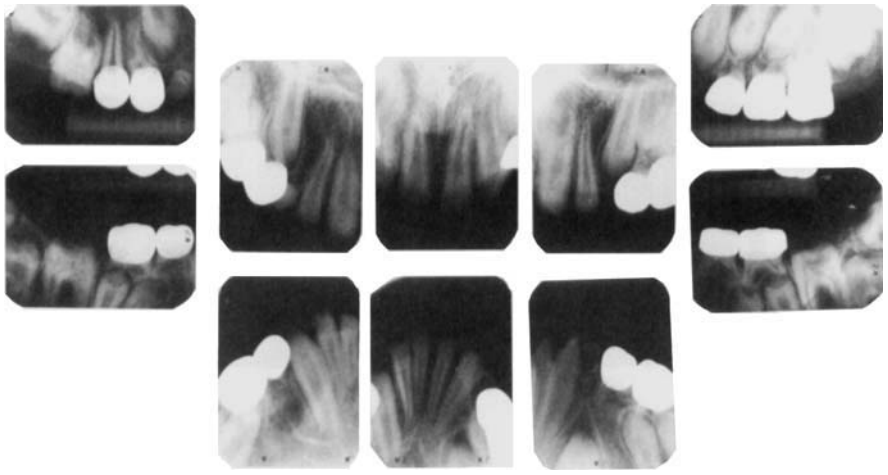

Fig. 1. Radiographs of P.M. at 12 years show a full complement of permanent teeth but with marked delay in eruption of the permanent premolar and molar teeth. The left permanent first molar is the only tooth in occlusion other than the incisors.

Roentgenograms showed that all permanent canines, premolars and molars were present but unerupted. No enamel layer could be discerned on erupted or unerupted teeth, and the latter had large follicles surrounding the crowns. The unerupted right mandibular first molar crown had a rough outline suggesting that a portion of the occlusal surface had been resorbed within the alveolus. All erupted teeth and many unerupted permanent teeth, particularly canines and first molars, showed multiple radiopaque, often dagger-shaped bodies in the pulp chambers (Fig. 2, arrow) which probably represented pulpal calcifications. The lateral incisors showed slight dilaceration of the roots.

S.M.: Roentgenograms at 10 years showed development of all permanent teeth, except that a follicle for the maxillary right third molar was not detected. The maxillary incisors and the mandibular central incisors were the only permanent erupted teeth (Fig. 3). Marked attrition was present on the incisal surfaces. Clinically, the crowns of the teeth resembled her brother's.

Radiographically, enamel was absent on erupted and unerupted teeth (Fig. 4), and large follicles were present over the crowns of unerupted teeth. A rough appearance of the occlusal aspect of the first mandibular molars suggested that these crowns were undergoing resorption within the alveolus. Radiopaque masses were present in the pulp chambers of erupted incisor teeth and in the unerupted first permanent molar and canine teeth. The maxillary incisors and unerupted mandibular first permanent molars showed slight dilaceration of the roots.

Histologic Preparation

An exfoliated crown of the primary maxillary right canine tooth of patient S.M. was examined. No enamel was detected on gross or microradiographic examination. Thick (120 μm) and thin (70 μm) sagittal ground sections were prepared by embedding the teeth in dental stone and sectioning on a Motter lathe microtome. Some

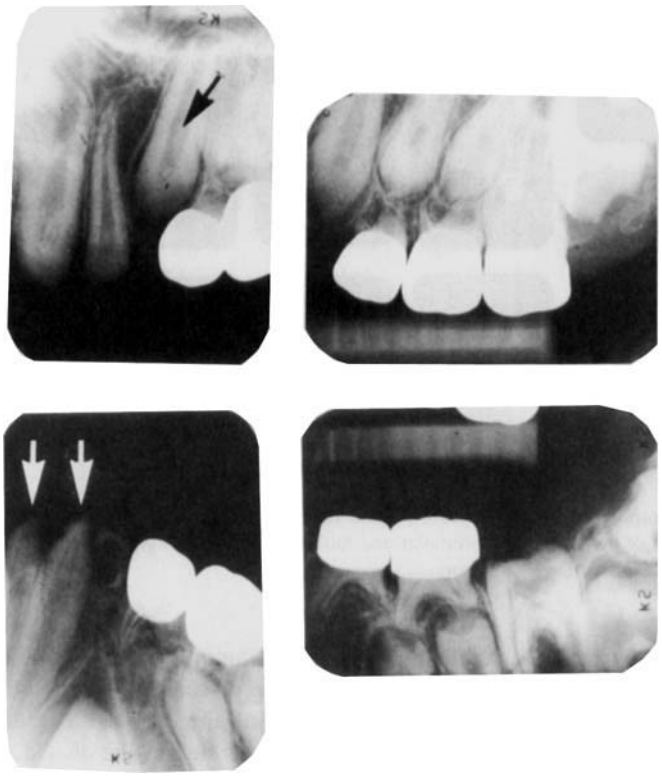

Fig. 2. Radiographs of P.M. at 12 years show no detectable enamel on erupted incisors or unerupted teeth. Arrows indicate radiopaque masses in pulp chambers of erupted and unerupted teeth which are dagger-shaped in the anterior teeth.

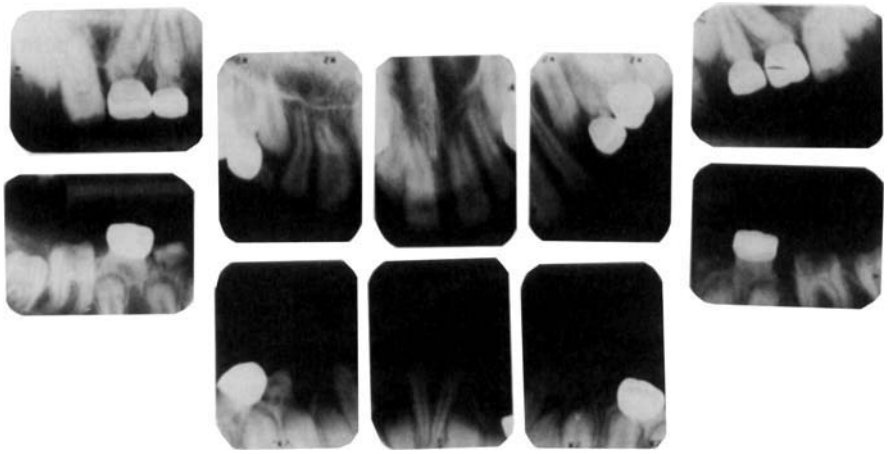

Fig. 3. Radiographs of S.M. at 10 years show marked delay in eruption of permanent teeth. The only permanent teeth in occlusion are the maxillary incisors, the maxillary first molars and mandibular central incisors. Note the rough mottled appearance of the unerupted crowns of the mandibular first molars suggesting that they have been partially resorbed.

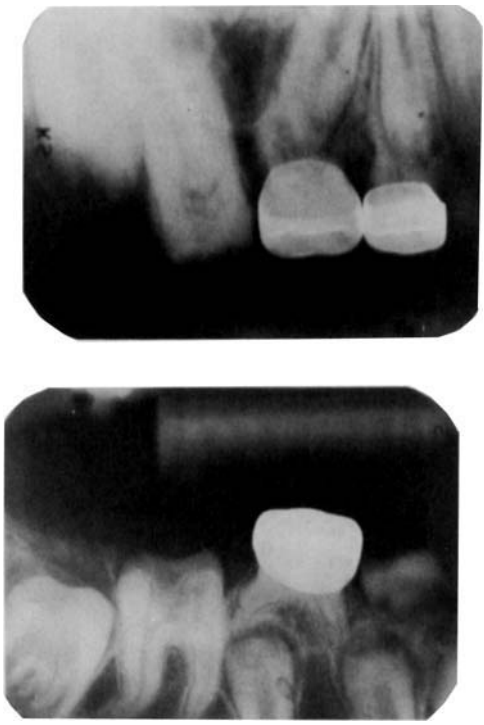

Fig. 4. Radiographs of S.M. at 10 years show no enamel on the primary first molar or unerupted permanent teeth. Calcifications are present in the pulp chambers of the first permanent molars and an area of resorption appears to involve the lower molar crown.

sections were lapped using a series of #3M lapping papers to a final thickness of 50 μm. Unstained and lightly stained (Masson trichrome) undecalcified sections were examined and photographed using a Zeiss photoscope. Examination under low power ($\times$10) showed no normal enamel (Fig. 5). The dentin was normal except for isolated areas of stained and sclerotic tubules and a calcified mass of tubular secondary dentin with vascular inclusions attached to the incisal aspect of the roof of the pulpal dentin and extending into the pulp chamber (Fig. 5, arrow). Areas of interglobular calcification were not observed in the dentin. Inspection of the dentin surface normally in contact with enamel showed dentinal tubules extending to a flat unscalloped surface with no evidence of an enamel layer (Fig. 6). The root surface was covered by acellular cementum (Fig. 7). The only evidence of calcified material on the crown occurred incisally to the cemental junction, where in the tilted thick specimen (Fig. 7), a few small islands of amorphous globular calcified material were found. These were from 0.5 to 2.0 μm in thickness and resembled globules of cementum.

Renal Evaluation

P.M.: At 5 years, IVP showed grade I reflux into the right ureter; this was not found subsequently. There were minimal, barely visible fine calcifications in the medullary areas which became progressively coarser and denser in roentgenograms at 8, 11, and 14 years. No distortion of the calyces has been evident.

Fig. 5. Thick ground section of the exfoliated primary right maxillary canine tooth from S.M. showed no evidence of enamel, and a large, dagger-shaped mass of secondary dentin with vascular inclusions occupying the incisal aspect of the pulp chamber (arrow). (Masson trichrome stain, magnification ×10).

SM.: At 6 years, IVP showed faint medullary calcinosis. This was more apparent at 9 years, but less severe than in her brother and with minimal progression in the interval. No calyceal distortion developed.

Laboratory Studies

Serum electrolytes, pH, bicarbonate, parathormone, calcitonin, 20(OH) vitamin D, Ca, PO_4, and alkaline phosphatase, as well as urine amino acids and oxalates were normal. Four years of renal function tests from 9 years in the girl and from 11 years in the boy are summarized in Table I. Both had impaired concentrating ability. The brother had a slightly reduced creatinine clearance. The excretion of titrable acid in response to oral NH_4Cl was decreased in both and urine ammonia excretion was subnormal in the brother. Both children consistently excreted low amounts of urinary Ca and PO_4 in 24 hours. The urine calcium:creatinine ratio (U_{Ca}/U_{cr}) was low in both and there was less than the normal 2–3 fold increase after 1.0 g of oral calcium [Sutton and Walker, 1980; Pak et al, 1975]. A test dose of furosemide (40 mg) in the boy raised Ca excretion to 129 mg per 24 hour, but the Ca:cr ratio of 0.81 was still subnormal. Because of borderline impairment of urinary acidification, a daily oral supplement of 4.4 g potassium citrate monohydrate, 4.0 g sodium citrate dihydrate

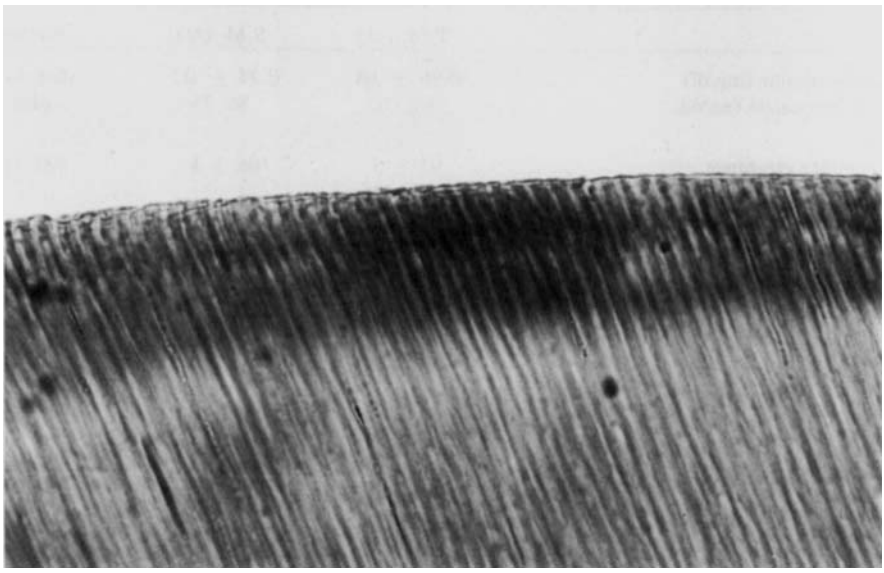

Fig. 6. Unstained thin ground section shows dentinal tubules coursing to the surface with no evidence of enamel or a calcified material covering dentin. This appearance was present over all crown surfaces examined except as shown in Figure 7. (Magnification ×400).

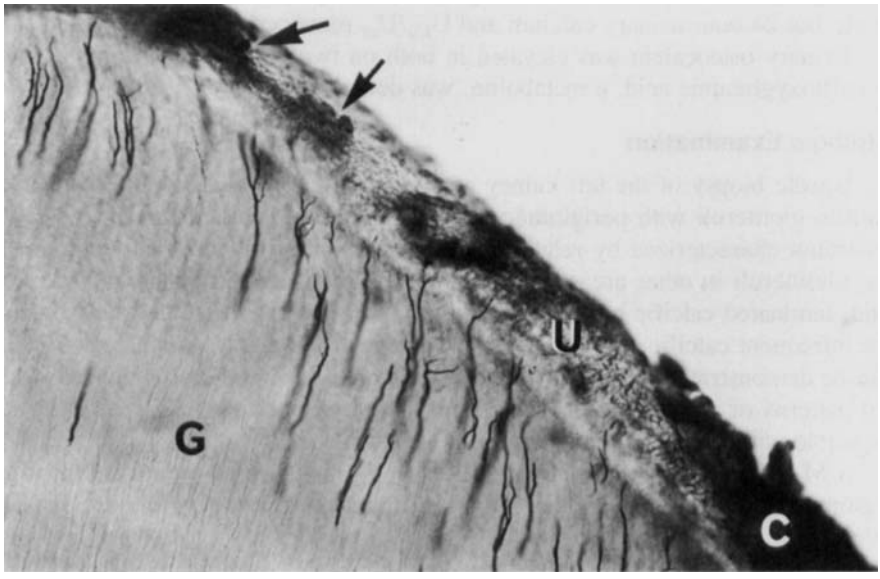

Fig. 7. Thick ground section (G) tilted to show unground (U) crown surface at the cemental (C) junction. Small islands of globular material (arrows) resemble cementum rather than enamel. (Masson trichrome stain, magnification ×320).

Table I. Results of Selected Laboratory Tests

Test	P.M. (♂)	S.M. (♀)	Normal
Serum creatinine (mg/dl)	0.96 ± .04	0.74 ± .02	(0.4–1.2)
Serum osteocalcin (ng/ml)	68; 202	86; 75	(40)
Urine			
Creatinine clearance (mg/min/1.73 M^2)	91 ± 5	106 ± 4	(90–110)
Maximal concentration (mosm/L urine/plasma)	496/286	660/293	(800)
Maximum specific gravity[a]	1.012	1.018	
pH (routine)	5.0–8.0	5.0–8.0	
(NH_4 g/min/1.73 M^2)	48	75	(46–100)
Total acidity (g/min/M^2/1.73 M^2)[b]	33	25	(33–71)
Calcium (mg/24 hr)	33 ± 10	25 ± 6	(200)
Phos (mg/24 hr)	764 ± 65	591 ± 40	(900–1300)
U_{Ca}/U_{cr}[c]			
Fasting	.028 ± .009	.046 ± .005	(.053 ± .007)
Ca Load	.037 ± .008	.061 ± .017	(.147 ± .014)
Gamma carboxyglutamic acid mc moles/gm	15	20	33–35
Oxalate mg/24 h	4	6	(40)
Citrate mg/24 h control	26–66	39–224	(140–940)
on oral citrate	405–721	695–832	

[a]Following overnight water deprivation.
[b]After loading dose of NH_4Cl 150 meq/m^2.
[c]Mean of 12 tests by the method of Pak: 2 hour fasting and 4 hour post load urine collections.

and 2.7 g citric acid monohydrate was given for one year. Citrate excretion increased in both, but 24 hour urinary calcium and U_{Ca}/U_{cr} remained unchanged.

Urinary osteocalcin was elevated in both on two occasions. Urinary excretion of δ-carboxyglutamic acid, a metabolite, was decreased.

Histologic Examination

Needle biopsy of the left kidney at 11 years showed focal areas of hyalinized, shrunken glomeruli with periglomerular fibrosis within areas of tubular loss and an interstitium characterized by relatively large numbers of mononuclear inflammatory cells. Glomeruli in other areas were normal. There was rare tubular dilatation. One round, laminated calcific body was noted within a hyalinized glomerular tuft. There were infrequent calcific deposits in the tubular interstitium. No immunofluorescence could be demonstrated. On electron microscopy 14 of 28 glomeruli were sclerosed in focal patterns of tubular atrophy with intense chronic inflammatory cell infiltration, compatible with chronic pyelonephritis or ischemia.

S.M.: Needle biopsy of the left kidney at 9 years showed mild focal scarring of the glomeruli and interstitial inflammation similar to, but less severe than, that of her brother.

DISCUSSION

The enamel defect in this condition most closely resembles that of autosomal recessive hypoplastic (enamel agenesis) type of amelogenesis imperfecta. However,

in that condition a laminated, structureless calcified material resembling layers of agate covers the surface of the dentin. A similar material may have been present on the teeth of these patients at one time, but the lack of radiographic and clinical evidence of enamel suggests otherwise. One patient who may represent an example of the agenesis type of amelogenesis imperfecta was reported by Bergman et al [1964], and had polycystic kidneys and cataracts; none of the other patients that definitely represent this type of amelogenesis imperfecta [Witkop and Sauk, 1976; Catena et al, 1970; Edward and Nord, 1974; Chosack et al, 1979], or are possible examples [Frank and Bolender, 1962; Wennström, 1963; Bergman et al, 1964] were known to have had kidney disease. As a general rule, hypoplastic enamel defects secondary to defects in calcium metabolism result when hypocalcemia occurs during odontogenesis. Interglobular calcification of the dentin develops during periods of hypophosphatemia [Nikiforouk and Fraser, 1979]. The enamel defects seen in hypocalcemic conditions such as vitamin D-dependent rickets and pseudohypoparathyroidism present a pitted clinical appearance and do not have complete absence of enamel as in our patients [Witkop and Sauk, 1976].

Hypoplasia of enamel is also associated with a wide variety of ectodermal syndromes [Witkop et al, 1975]. However, some detectable amounts of enamel are present even in severe ectodermal disturbances such as the amelocerebro-hypohidrotic syndrome of Köhlschutter [1974] and the dystrophic forms of epidermolysis bullosa [Witkop and Sauk, 1976].

Failure of eruption of teeth and/or resorption of unerupted teeth within the alveolus is a manifestation often seen in the hypoplastic types of amelogenesis imperfecta [Witkop and Sauk, 1976] and in syndromes associated with hypoplasia of enamel such as the trichodentoosseous syndrome [Jorgenson and Warson, 1973], the ameloonychohypohidrotic syndrome [Witkop et al, 1975], and the trichoonychodental syndrome [Koshiba et al, 1978].

In the present condition, the inner enamel organ epithelial cells apparently induce odontoblastic differentiation of ectomesenchymal cells, but then fail to respond to induction from dentin to form enamel.

The nephrocalcinosis is also atypical for its early onset and the absence of typical predisposing factors. Since there was progression despite consistently low urinary calcium excretion, calcium absorption is probably not defective. Therefore, this syndrome appears to be in the unusual category of nephrocalcinosis with hypocalcinuria.

The above findings document a syndrome of amelogenesis imperfecta, progressive nephrocalcinosis, and decreased renal concentrating ability in our patients. This represents a single entity, rather than a chance concurrence, since similar findings were reported in another sibship [MacGibbon, 1972]. This is presumably an autosomal recessive condition since both sexes are affected and there were no findings in the parents.

Pathogenesis is unknown. The nephrocalcinosis and decreased urinary calcium and phosphate excretion suggest a possible defect in calcium metabolism. In this respect, the chronic concentrating problems, antedating other findings and in excess of other abnormalities of tubular dysfunction, are of interest. Chronic hypercalcemia can cause a diabetes insipidus, indicating that calcium metabolism can be related to concentrating ability [Gill and Bartter, 1961]. However, studies of serum levels, parathyroid function, and vitamin D metabolism were all normal.

The abnormalities of vitamin K-dependent calcium binding proteins are suggestive, although too little is known of their function to make a definitive statement. These abnormalities are associated with ectopic calcifications, and the proteins are found in relatively high concentrations in kidney and tooth dentin [Gallop et al, 1980]. Serum osteocalcin, one of the major species of these proteins, was elevated in both of our patients. Urinary δ-carboxyglutamic acid, a component and derivative, was paradoxically reduced, and paralleled the low urine calcium.

The locations of these proteins and their relationship to calcium metabolism provide a tempting molecular linkage for the multiple anomalies in our patients. However, the protein findings may be secondary, rather than causative, or perhaps even artifactual. No other possible cause for the nephrocalcinosis was apparent. Despite problems with tubular function, including acidification, there was no evidence for a systemic acidosis.

A major concern for children with this disorder is prognosis. While renal function seems stable at age 14 and 16, the progressive renal failure and death of MacGibbon's patients [1972] is ominous. We were unable to affect excretion with calcium chelators in our patients and are uncertain about treatment prospects. Renal transplantation may become necessary, although the response of a new kidney is unknown. We would be interested to hear of additional patients with this syndrome and to learn of their course and results of treatment.

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