Classification of Idiopathic Hypercalciuric Patients by Isotopic Calcium Absorption: A Comparison with Oral Calcium Tolerance Test

Roberto Pacifici, Paolo Filipponi, Carla Mannarelli, Giuseppe Vespasiani, Massimo Porena, Leone Fedeli, Valerio Morucci, and Louis V. Avioli

The Institutes of Clinica Medica I, Clinica Urologica, Medicina Nucleare and Medicina del Lavoro Dell' Universita' Di Perugia, Italy and the Division of Bone and Mineral Metabolism, Washington University School of Medicine, St. Louis, Missouri, USA

Summary. To test the accuracy of calcium tolerance test in estimating calcium absorption, we have measured the radioactive calcium absorption (expressed as Fx) in 27 patients with IH and renal calcium stones. The results of this test were compared with those of a standard oral calcium tolerance test. Although only seven of nine AH patients displayed normal fasting calcium excretion, they all displayed Fx values above normal and a normal parathyroid activity. Conversely, only 5 of our 18 RH patients demonstrated a hyperabsorption of radioactive calcium and an elevation in iPTH and cAMP above normal limits, yet all of them showed an increased calciuric response to an oral calcium challenge. Calcium absorption was inversely related to iPTH (r = -0.82; P < 0.001) and cAMP (r = -0.64; P < 0.05) in AH, but directly proportional to these parameters (r = 0.62; P < 0.001 and r = 0.46; P < 0.05, respectively) in RH patients.

In view of these results, two ratios, iPTH/Fx and cAMP/Fx were used to discriminate between the two groups of patients. Both ratios were over normal limits in all RH patients and within normal range in all but one AH patient. Furthermore, no overlap was found between the two groups. Conversely, we were unable to completely separate AH from RH subjects on the basis of the oral calcium tolerance test, since in both groups the fasting and the absolute (or percentage) changes in urinary calcium, cAMP and blood iPTH levels following oral calcium loading, overlapped in each instance. The result of this study indicates that two indices, iPTH/Fx and cAMP/Fx, may prove particularly useful in differentiating AH from RH patients. Furthermore, since only a subgroup of patients with an abnormal calciuric response to an oral calcium load manifest an increase in calcium absorption, it is concluded that the calcium tolerance test overestimates calcium absorption in IH.

Key words: Hypercalciuria — Calcium absorption — Calcium load.

When first described [1], idiopathic hypercalciuria (IH) was classified as at least two separate entities: absorptive hypercalciuria (AH), and renal hypercalciuria (RH). This classification was based on the observation that some patients with IH exhibited excessive calcium excretion because of calcium hyperabsorption by the intestines, although others presented with hypercalciuria and impaired calcium reabsorption by the renal tubules [2]. More recently, the calcium tolerance test has been proposed and widely used as a standard procedure for distinguishing AH from RH [3–5], assuming that normal calcium excretion during fasting, increased calciuric response to calcium load, and normal or suppressed parathyroid activity are discriminating features of AH; whereas, increased fasting and postload calcium excretion and high parathyroid activity are markers of RH [3, 5, 6, 7–9]. Nevertheless, elevated urinary calcium excretion in the fasting state has been reported in some cases of AH [10–12], and parathyroid activity has been found normal even in patients with RH and compensatory hyperabsorption of calcium [8, 13]. Although a better resolution can be achieved using a fasting
urinary calcium-cAMP discriminant score [12], we are of the opinion that the oral calcium tolerance test does not always reflect the intestinal absorption of calcium and that most of the controversies [3, 8, 14–17] could be resolved if one relates parathyroid activity to a more sensitive and precise index of intestinal calcium absorption.

To substantiate our opinion, we studied a group of patients with hypercalciuria and calcium nephrolithiasis by measuring radioactive intestinal calcium absorption and fasting parathyroid activity. Finally, we compared the results of these studies with those of a standard oral calcium load test.

Materials and Methods

Twenty male and seven female patients with recurrent calcium nephrolithiasis and IH were studied. Patients were defined to have IH consistent with the following parameters: a serum calcium of 8.5–10.5 mg/dl; calcium excretion > 200 mg/day on a daily intake of 400 mg of calcium, > 300 mg/day on a random diet, and/or > 0.2 mg/100 ml G.F.R. after a 1000 mg oral calcium load. Subjects with hyperuricemia and/or hyperuricosuria, recurrent urinary tract infection, obstructive disease, urinary tract malformation or diabetes were excluded from the study. None of the patients received therapy for at least 10 days before entering the study.

Routine laboratory tests (SMA-6 and SMA-12) were within normal limits, mean (mg/dl ± SE) levels of phosphate and magnesium were 3.3 ± 0.3 and 1.8 ± 0.4 respectively.

A basal study was performed by obtaining 24-hour urine collections and midpoint (12 hour) blood samples while patients were on a random diet, (calcium intake was retrospectively estimated by dietary recall analysis as 600–800 mg/day). Intestinal absorption of radioactive calcium was measured by the Gennari technique [18] of the technique of Nordin et al. [19] while patients were still on the random diet, and expressed as Fx. Briefly, calcium absorption was estimated by administering 5 μCi of 45Ca in 100 mg calcium carrier as calcium gluconate after a 12-hour fast. Plasma samples were obtained 15, 30, 60, 90, 120, 180, 240, and 300 minutes thereafter. The fraction of the administered dose per liter was multiplied by 15% of body weight to estimate the fraction of the dose circulating in the extracellular fluid at each time. The total circulating fraction of the dose (Fx) was calculated by multiplying the area bounded by the dose-time curve and a constant that expresses the fraction of the dose removed from the plasma. The reproducibility of this method was ±14%. After 7 days of a restricted calcium intake (400 mg/day), the standard oral calcium-load test [5] was performed; urine samples were collected after a 12-hour overnight fast, between 8 AM and 10 AM, and between 10 AM and 2 PM. Blood samples were taken at 8 AM and 10 AM; at 10 AM patients ingested 250 ml of whole milk supplemented with calcium gluconate to a total calcium content of 1000 mg. A third blood sample was drawn at 2 PM. Patients were allowed to drink 1500 ml distilled water in order to maintain an adequate urine flow. Blood samples, obtained without stasis with patients in the supine position for at least 30 minutes were assayed for creatinine, calcium, magnesium, phosphate, and immunoreactive parathyroid hormone (iPTH). Urine samples were analyzed for creatinine, calcium, magnesium, phosphate and cAMP. Results were normalized to the glomerular filtration rate (GFR).

Analyses

Serum and urinary calcium and magnesium were determined by atomic absorption spectrophotometry; serum and urinary creatinine and phosphate by standard spectrophotometric techniques, and serum; PTH by a specific double antibody radioimmunoassay (Immunonuclear Corporation, Stillwater, Minn.). The antisera was directed against the 48–68 region of human PTH (PTH-mm) and obtained from chickens. The h-PTH44–68 standard was calibrated against the WHO preparation 79/500 and the results expressed in nmol/l (coefficients of variation of this assay were 6% within and 10% between assays and the sensitivity, 10 nmol/l). In order to minimize the intraassay variability, all samples were measured in the same assay. Urine was assayed for cAMP by the competitive protein binding technique, using the Becton Dickinson Immunodiagnostics Kit (Orangeburg, NY). Sensitivity of iPTH and urinary cAMP assays in discriminating between normal and increased parathyroid activity was evaluated in four hypercalcemic patients with surgically proved hyperparathyroidism. All four had iPTH and cAMP above normal limits prior to surgery; both these parameters returned within normal range following successful neck exploration.

Statistics

Statistical evaluation of results was performed by Student’s t test (both paired and unpaired as appropriate) and by linear regression analysis.

Results

Nine patients in this series (33%) (Fig. 1a; circles) were characterized by hyperabsorption of radioactive calcium (Fx: mean ± SE: 0.36 ± 0.03) and normal parathyroid activity. (iPTH: mean ± SE: 37.0 ± 2.7 pmol/l; cAMP, mean ± SE: 2.89 ± 0.26 nmol/100 ml G.F.R). As shown in Figure 1b, five patients (18.5%) (open triangles) had increased calcium absorption (Fx: mean ± SE: 0.30 ± 0.01) and increased parathyroid activity (iPTH, mean ± SE: 166.0 ± 50.3 pmol/l; cAMP, mean ± SE: 7.5 ± 0.6 nmol/100 ml G.F.R.). The remaining 13 subjects (48.1%) (closed triangles) had normal radiocalcium absorption (Fx, mean ± SE: 0.20 ± 0.01) and normal parathyroid function (iPTH, mean ± SE: 59.6 ± 3.0 pmol/l; cAMP, mean ± SE: 5.5 ± 0.2 nmol/100 ml G.F.R.). Serum calcium values (mg/dl ± S.E.) for each of these groups were 9.7 ± 0.2, 9.2 ± 0.2 and 9.5 ± 0.1, respectively.

Those patients with calcium hyperabsorption and normal parathyroid activity were considered to