Mineral water as a source of dietary calcium: acute effects on parathyroid function and bone resorption in young men¹,²

Josette Guillemant, Huyen-Tran Le, Chantal Accarie, Sophie Tézenas du Montcel, Anne-Marie Delabroise, Maurice J Arnaud, and Serge Guillemant

ABSTRACT

Background: Calcium is a major component of mineralized tissues and is required for normal growth and maintenance of bone. Epidemiologic studies showed that a large percentage of the population fails to meet the currently recommended guidelines for optimal calcium intake.

Objective: The present study was designed to determine whether high-calcium mineral water is an efficient additional source of dietary calcium.

Design: Twelve healthy young men (mean ± SD age: 21.1 ± 1.2 y) ingested in a randomized order either 0.5 L of a mineral water containing 344 mg Ca/L or 0.5 L of a mineral water with a very low concentration of calcium (<10 mg/L) as a control. Blood samples were drawn before and 1, 2, 3, and 4 h after intake of the water. Urine was collected for 2 h before and every 2 h for 4 h after ingestion of the water. Serum concentrations of intact parathyroid hormone (iPTH) and serum concentrations and urinary excretion of type 1 collagen cross-linked C-telopeptide (CTx), were measured.

Results: Serum iPTH was significantly (P < 0.002) lower after ingestion of high-calcium water than after ingestion of the control. There was a significant (P = 0.01) progressive decrease in urinary CTx after ingestion of the high-calcium water, whereas after ingestion of low-calcium water the changes were modest and not significant. The fall in serum CTx concentrations was 34.7% 3 h after ingestion of high-calcium water, compared with 17.6% with the control. The decreases in serum CTx concentrations were significantly (P < 0.05) lower 1, 2, 3, and 4 h after ingestion of high-calcium water than after ingestion of the control.

Conclusion: The present study showed that one oral intake of high-calcium water containing 344 mg Ca/L acutely inhibited iPTH secretion and bone resorption. Am J Clin Nutr 2000;71:999–1002.

KEY WORDS  Calcium, parathormone, bone markers, mineral water, young men, France, parathyroid hormone, parathyroid function, bone resorption

INTRODUCTION

A sufficient calcium intake is now considered to be beneficial to bone mass at all stages of life and some recommended dietary allowances (RDAs) for calcium are between 1200 and 1500 mg/d for adolescents, pregnant and nursing women, and people aged ≥65 y (1). To attain the optimal calcium intake it has been suggested that the frequency of consumption of dairy products and calcium-rich vegetables be increased. Other low-energy calcium sources, not mentioned in the National Institutes of Health Consensus Statement (1), are high-calcium mineral waters. The bioavailability of calcium from mineral waters was recently studied and was found to be at least equal to that from milk (2, 3). Oral intake of a calcium load increases serum calcium and lowers intact parathyroid hormone (iPTH) concentrations. Because elevated concentrations of iPTH induce an increase in bone turnover, which is accompanied by increased bone loss, it is pertinent to determine whether an oral calcium load not only decreases iPTH secretion but also inhibits bone resorption. The products of degradation of collagen are potential bone-resorption markers and specific amino acid sequences of nitrogen- or carbon-terminal telopeptides have been used as markers of type 1 collagen breakdown. We decided, therefore, to assess the acute effects of a low (172 mg) dose of calcium contained in 0.5 L of a high-calcium mineral water on parathyroid function and bone resorption. Bone resorption was assessed by measuring serum concentrations and urinary excretion of type 1 collagen cross-linked C-telopeptide (CTx) by using a recently developed assay.

SUBJECTS AND METHODS

Twelve healthy young men (mean ± SD age: 21.1 ± 1.2 y) were studied. None of the men had disorders or was taking medications known to affect calcium metabolism. The subjects were studied during their usual diet (dietary calcium intake of ≈800 mg/d) except on the evening before the assay, on which they avoided all calcium-rich meals. The subjects' vitamin D status, assessed by measuring 25-hydroxyvitamin D, was found to be replete (>800 mg/d) except on the evening before the assay, on which they avoided all calcium-rich meals. The subjects’ vitamin D status, assessed by measuring 25-hydroxyvitamin D, was found to be replete (29 ± 5.8 μg/L). All subjects were medical students and all gave their informed consent to the procedures, which were approved

¹From the Service de Biochimie, Faculté de Médecine Pitié-Salpêtrière, Paris; Ecole Pratique des Hautes Etudes, Nutrition et Pharmacologie Hydrominérale, Paris; the Département de Biostatistiques, Faculté de Médecine Pitié-Salpêtrière, Paris; and the Institut de l’Eau, Vittel, France

²Address reprint requests to S Guillemant, Faculté de Médecine Pitié-Salpêtrière, Boulevard de l’Hôpital 91, F-75634 Paris Cédex 13, France. E-mail: guillema@ccr.jussieu.fr.
by the local ethics committee (Comité Consultatif de Protection des Personnes dans la Recherche Biomédicale). The study protocol was performed between 0700 and 1300. The subjects fasted from the previous evening (2100–2200) until the end of the assay (1300). Blood samples were drawn before and 1, 2, 3, and 4 h after oral intake of the high-calcium mineral water (referred to hereafter as P0, P1 h, P2 h, P3 h, and P4 h). Urine was collected for 2 h before and every 2 h for 4 h after calcium intake (referred to hereafter as U0, U2 h, and U4 h). The test was started by having the subjects ingest 0.5 L of a high-calcium mineral water containing 172 mg elemental Ca. A control test was performed by having the subjects ingest the same volume of a low-calcium mineral water (< 10 mg/L). The 2 tests were performed at least weekly and the order of the tests was randomized.

The composition of the 2 mineral waters was as follows: 1) High-calcium mineral water: calcium, 344.7 mg/L; magnesium, 50.2 mg/L; sodium, 4.6 mg/L; potassium, 2.3 mg/L; sulfate, 767.9 mg/L; bicarbonate, 356 mg/L; chloride, 3.2 mg/L; and phosphate, 0.08 mg/L. 2) Low-calcium mineral water: calcium, 9.9 mg/L; magnesium, 6.1 mg/L; sodium, 9.4 mg/L; potassium, 5.7 mg/L; sulfate, 6.9 mg/L bicarbonate, 65.3 mg/L; and chloride, 8.4 mg/L.

Serum ionized calcium was measured with a specific calcium electrode (ICa2 ionized calcium analyzer; Radiometer, Copenhagen). Serum PTH (1–84) was measured by immunoradiometric assay for iPTH (Nichols Institute, San Juan Capistrano, CA); intraassay variation was < 4% and interassay variation was < 6% in the explored range of concentrations. Urinary calcium was determined by using a fluorimetric method with calcein and an automatic apparatus (Precision Systems Inc, Sudbury, MA). Urinary CTx was measured with an enzyme immunoassay kit (CrossLapsEIA purchased from CIS Bio International, Gif-sur-Yvette, France). This kit measures an amino acid sequence (EKAHDGGR) specific for a part of the C-telopeptide of the α-1 chain of type I collagen, the major form of collagen in organic matrix of bone that therefore can be used in the estimation of bone resorption. The assay has no cross-reaction with urinary free cross-links. The values were corrected by the urinary creatinine concentration measured by a colorimetric method. Intraassay variation is 6% and interassay variation is < 10% in the explored range of concentrations. Serum CTx was measured by using a one-step enzyme immunologic test (Serum CrossLaps; Osteometer Biotech A/S, Denmark, purchased from CIS Bio International). This test recognizes 2 cross-linked amino acid sequences of EKAHD-β-GGR where the aspartic acid residue is β-isomerized. Intraassay variation is < 5% and interassay variation is < 10% in the explored range of concentrations.

Data are expressed as means ± SDs unless otherwise stated. Repeated-measures two-factor analysis of variance with interaction was performed by using the general linear models procedure (4). The main effects were time of sampling and treatment (high-calcium water or low-calcium water), with subjects as a random factor. When the time-by-treatment interaction was significant (P < 0.05), Bonferroni t tests of differences between means were performed at the global level of probability of 0.05.

RESULTS

Calcium metabolism results are shown in Table 1. Ingestion of high-calcium water induced a significant increase in both ionized calcium (repeated-measures two-factor analysis of variance: time, P < 0.003; treatment, P < 0.005; time-by-treatment interaction, P < 0.02) and urinary calcium (time, P < 0.06; treatment, P < 0.001; time-by-treatment interaction, P < 0.004). After the subjects ingested high-calcium water, serum iPTH decreased significantly (time, P < 0.01; treatment, P < 0.002; time-by-treatment interaction, P = 0.0541), was lowest at P1 h (− 34% of basal values), and returned to baseline at P4 h (Figure 1). After

![FIGURE 1](https://academic.oup.com/ajcn/article-abstract/71/4/999/4729209)

**FIGURE 1.** Time course of changes in mean (±SEM) intact parathyroid hormone (iPTH) after ingestion of 0.5 L high-calcium (172 mg Ca) and low-calcium (< 5 mg Ca) mineral water. Serum iPTH was measured before (P0) and 1 h (P1 h), 2 h (P2 h), 3 h (P3 h), and 4 h (P4 h) after ingestion of water. Time and treatment main effects were significant (P < 0.01 and P < 0.002, respectively) by repeated-measures two-factor ANOVA.
ingestion of high-calcium water there was a significant (time,
$P < 0.02$; treatment, $P = 0.01$; time-by-treatment interaction; 
$P < 0.04$) progressive decrease ($-17\%$ at U2 h and $-34\%$ at 
U4 h) in urinary CTx excretions, whereas after ingestion of low-
calcium water the changes were modest (Figure 2). At U4 h the 
urinary excretion of CTx was significantly ($P < 0.05$) less after 
ingestion of high-calcium water than after ingestion of the con-
trol (Bonferroni $t$ tests). A decrease in serum CTx concentrations 
($-28.9\%$ at P2 h, $-34.7\%$ at P3 h, and $-31.8\%$ at P4 h) was evi-
dent after ingestion of high-calcium water (Figure 3). In the con-
trol assay a more modest ($-10.8\%$ at P2 h, $-17.6\%$ at P3 h, and 
$-15.1\%$ at P4 h) decrease was observed. Repeated-measures
two-factor analysis of variance indicated that the effects of both
time and treatment on serum CTx were significant ($P < 0.001$)
and that there was a significant ($P < 0.007$) interaction between
the 2 main effects. The comparison of the 2 series of assays by
Bonferroni $t$ tests showed that at P1 h, P2 h, P3 h, and P4 h the
serum CTx concentrations were significantly ($P < 0.05$) lower
after ingestion of high-calcium water than after ingestion of low-
calcium water (Figure 3). The basal (P0) values in the 2 series of
assays were not significantly different.

FIGURE 2. Urinary excretion of collagen cross-linked C-telopeptide 
(CTx) before and after ingestion of 0.5 L of either high-calcium (172 mg 
Ca) or low-calcium (< 5 mg Ca) mineral water. Urine was collected 
for 2 h before (U0) and every 2 h for 4 h (U2 h, U4 h) after ingestion of 
water. x ± SEM. High-calcium and low-calcium waters were compared 
at each time point by using Bonferroni $t$ tests of differences between 
means. $P < 0.05$.

DISCUSSION

We showed, for the first time, that a low dose (172 mg) of cal-
cium contained in 0.5 L mineral water and given to young men
inhibited iPTH secretion by 34% 1 h after ingestion and induced
a significant decrease in CTx both in urine ($-34\%$ between 2 
and 4 h) and in serum ($-34.7\%$ at 3 h); this decrease was prob-
ably mediated by the reduction in iPTH secretion. Previous stud-
ies showed that calcium supplements administered at various
times suppressed PTH secretion and decreased bone resorption
markers. A significant decrease in PTH and resorption bone
markers was observed after supplements of calcium ingested for
days (5, 6), months (7), and years (8). Some studies showed the
acute effects of the intake of calcium on serum PTH and on bone
resorption in men but with much larger doses (1000 mg) and
showed an inhibition by 29% of urinary deoxypyridinoline
between 4 and 6 h (9) and by 55% of urinary CTx (between 2 and 
4 h) after the ingestion of the calcium load (10). We observed a
significant decrease in serum CTx after ingestion of high- and 
low-calcium mineral water. However, the decrease in serum CTx
was significantly less after ingestion of low-calcium mineral water
than after ingestion of high-calcium mineral water and could be
attributed to a circadian rhythm of bone resorption. No significant
decrease in urinary CTx was found during the 6 h of
the assay with low-calcium water, although circadian variations
in urinary pyridinium cross-links, N-telopeptide, and CTx, have
been reported (11, 12).

The comparison of the effects of 500- and 1500-mg doses of 
calcium had suggested to us that small oral doses of calcium
could be efficient in acutely inhibiting PTH secretion (13). In a
previous study we showed that 250 mg Ca contained in a cal-
rich mineral water significantly inhibited by 45% the serum concentrations of iPTH (14). In the present study a dose of 
calculator as small as 172 mg contained in 0.5 L mineral water
was ingested and had significant effects on both iPTH concen-
trations and bone resorption. We can estimate, according to
Heaney et al’s data (15), that $\approx 40\%$ of the ingested dose should
have been absorbed. Studies of absorbability of calcium in dif-
ferent high-calcium mineral waters showed that after ingestion
of 100 mg Ca, 47.5% was absorbed (2), whereas after ingestion
of 1000 mg Ca 23.8 ± 4.8% was absorbed (3).

Dietary intakes of calcium are generally much less than the 
RDAs; to attain optimal calcium intake, calcium supplements
are frequently prescribed. These supplements are often administered
once daily and the duration of their effects is restricted to a few
hours. If one agrees with the statement, taken from the National
Institutes of Health Consensus Conference on Optimal Calcium
Intake (1), that “absorption of calcium supplements is most effi-

FIGURE 3. Time course of changes in serum mean (±SEM) collagen 
cross-linked C-telopeptide (CTx) after ingestion of 0.5 L of either high-
calcium (172 mg Ca) or low-calcium (< 5 mg Ca) mineral water. Serum
CTx was measured before (P0) and 1 h (P1 h), 2 h (P2 h), 3 h (P3 h), and 
4 h (P4 h) after ingestion of water. High-calcium and low-calcium waters
were compared at each time point by using Bonferroni $t$ tests of differ-
ences between means. * $P < 0.05$. 

FIGURE 1. Intake (1), that “absorption of calcium supplements is most effi-
cient at individual doses of 500 mg or less and when taken between meals," drinking calcium-rich mineral water several times a day could be recommended because it would provide both supplemental calcium and adequate hydration. Furthermore, better absorption of calcium by ingesting fractionate rather than single doses of calcium has been emphasized on both theoretical and practical bases (16, 17).

In conclusion, the results of the present study showed that high-calcium mineral water not only represented an additional dietary source of calcium but also modulated parathyroid function and bone metabolism. Nevertheless, further long-term studies are needed to confirm these short-term results.

REFERENCES