Report

Mineral Metabolism of Aging Female Rats Fed Various Commercially Available Calcium Supplements or Yogurt

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The utilization of calcium from commercially available calcium supplements and yogurt and the effects of these calcium supplements on the utilization of other minerals were evaluated. Moderate and high levels (4 and 8 mg Ca/g diet) of calcium from four different sources of dietary calcium (yogurt, calcium phosphate dibasic, calcium magnesium chelate, and oyster shells) were fed to retired female breeder rats. Rats absorbed calcium equally efficiently from all four sources but ingestion of calcium phosphate dibasic tended to cause abnormal accumulation of calcium in kidneys. Ingestion of the calcium magnesium chelate improved calcium retention in bone but depressed the digestibility of the total diet. The elevation of dietary calcium did not affect tissue calcium levels orecal β-glucuronidase activity but depressed the apparent absorption of phosphorus, increased kidney phosphorus levels, decreased tibia iron levels, and decreased the digestibility of the total diet.

KEY WORDS: calcium; supplements; nephrocalcinosis; magnesium.

INTRODUCTION

The Consensus Development Panel on Osteoporosis (1) suggested that one strategy for preventing or at least slowing the development of osteoporosis was the ingestion of 1000 to 1500 mg calcium daily. Other investigators have suggested that the consumption of additional calcium was protective against colon cancer (2,3). One result of this advice was that sales of calcium supplements increased five-fold between 1980 and 1985 (4).

However, few data are available on the relative utilization of calcium from supplements. Most investigators have examined the utilization of calcium from supplements or foods consumed in a single meal (5–7) but women are advised to consume supplements daily for a number of years. Moreover, the effects of chronic use of calcium supplementation on the utilization of other minerals have been studied primarily with young male rats (8).

Thus the objectives of this study were (i) to compare the utilization of calcium from a dairy product (yogurt) and three commercially available calcium supplements in mature female rats, (ii) to compare the effects of moderate and high calcium intakes on the utilization of calcium, and (iii) to examine the effects of the calcium source and level on the utilization of other essential minerals (i.e., magnesium, iron, phosphorus, zinc, and copper) on gut function and flora.

MATERIALS AND METHODS

Forty-eight retired female breeder rats were fed one of eight dietary treatments for 33 days. The dietary treatments differed in their levels of calcium (4 and 8 mg Ca/g diet) and in their sources of calcium. Diets 4 yogurt and 8 yogurt contained 4 and 8 mg Ca/g diet, respectively, from freeze-dried yogurt (The Dannon Company, White Plains, N.Y.); Diets 4 oyster shell and 8 oyster shell contained 4 and 8 mg Ca/g diet, respectively, from oyster shells (OsCal 500, Marion Laboratories, Inc., Kansas City, Mo.); Diets 4 Ca Mg chelate and 8 Ca Mg chelate contained 4 and 8 mg Ca/g diet, respectively, from a yeast chelated calcium and magnesium preparation (Schiff Bio-Food Products Manufacturing, Moonachie, N.J.); and Diets 4 CAHPO₄ and 8 CAHPO₄ contained 4 and 8 mg Ca/g diet, respectively, from calcium phosphate dibasic (Fisher Scientific, Fair Lawn, N.J.).

The semipurified diets fed conformed to the guidelines of the American Institute of Nutrition (9) and were similar in formulation to those fed to weanling rats in previous studies (8). The sole source of calcium in each diet was either yogurt or one of the three supplements because all calcium was eliminated from the basic AIN-76 mineral mixture used to prepare the diets. Diets were formulated to provide 18% protein from either yogurt and/or casein and to provide 9.5% disaccharide from yogurt or sucrose except for Diet 8 yogurt, which contained approximately 19% disaccharide and 23% protein because of the protein and disaccharide content of yogurt. All diets also contained 5.0% cellulose (Teklad Test Diets, Madison, Wis.), 5% corn oil (Best Foods, Englewood Cliffs, N.J.), 3.5% AIN-76 mineral mixture without calcium, 1.0% AIN-76 vitamin mixture (Teklad Test Diets), 0.3% dl-methionine, 0.2% choline bitartrate, and cornstarch to achieve 100%. The analyzed composition of the diets is shown in Table 1.

Retired female breeder Sprague–Dawley rats (Harlan Sprague–Dawley, Indianapolis, Ind.) were housed individu-
ally in stainless-steel wire-bottomed cages. The facilities met the standards of the American Association for Accreditation of Laboratory Animal Care.

Deionized water was offered ad libitum. Food consumption was recorded daily and animals were pair-fed to maintain feed intake at similar levels (11.0 ± 0.1 g feed/day). Rats were weighed twice a week.

Sample Collection and Analyses

Fecal samples were collected on days 8–9 and on days 31–32 of the study. Percentage dry matter digestibility was determined by the following equation: (weight of food intake – weight of dry feces) × 100 / weight food intake. Percentage apparent absorption of minerals was determined by the following equation: (intake of mineral – fecal loss of mineral) × 100 / intake of mineral.

Rats were anesthetized and killed by exsanguination on day 34 of the study after being fasted overnight. Kidneys, livers, and tibias were removed and frozen in acid-washed plastic containers. Fecal pellets were expressed from each rat’s colon; β-glucuronidase activity using phenolphthalein glucuronide (Sigma Chemical Company, St. Louis, Mo.) and protein content of diluted fecal pellets were determined (10,11).

Diets, tissues, and fecal samples were analyzed for calcium, magnesium, iron, zinc, and copper by atomic absorption spectroscopy and for phosphorus content by a colorimetric procedure (12,13). Livers were analyzed for iron and copper only; diets were also analyzed for lactose (14). Bovine liver standards (SRM No. 1577a) or milk standards (SRM No. 1549) obtained from the National Bureau of Standards were analyzed with several batches of experimental samples. Liver standards (N = 24) were determined to contain 600 ± 6 (mean ± SE) µg Mg/g (certified value, 600 µg Mg/g), 123 ± 1 µg Zn/g (certified value, 123 µg Zn/g), 177 ± 4 µg Fe/g (certified value, 194 µg Fe/g), 158 ± 1 µg Cu/g (certified value, 158 µg Cu/g), and 1.13 ± 0.01% P (certified value, 1.11%). Milk standards (N = 14) were determined to contain 1.25 ± 0.01% Ca (certified value, 1.3% Ca), 0.121 ± 0.001% Mg (certified value, 0.120%), and 49.1 ± 1.3 µg Zn/g (certified value, 46.1 µg Zn/g).

The effects of dietary treatments were evaluated by analysis of variance (15). Effects of levels and sources of calcium and their interactions were determined.

RESULTS AND DISCUSSION

The dietary treatments did not affect the body or organ weights of rats. The final average weight of rats was 264 ± 4 g.

Table II. Extent of Utilization of Calcium from Various Supplements as Judged by Tissue Calcium Levels and Apparent Absorption of Calcium

<table>
<thead>
<tr>
<th>Diet</th>
<th>Kidney Ca (µmol/g)</th>
<th>Tibia Ca (mmol/g)</th>
<th>Apparent absorption of Ca (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Days 8 &amp; 9</td>
<td>Days 31 &amp; 32</td>
<td></td>
</tr>
<tr>
<td>4 yogurt</td>
<td>1.40 ± 0.07a</td>
<td>4.62 ± 0.10</td>
<td>24 ± 9a</td>
</tr>
<tr>
<td>8 yoghurt</td>
<td>3.68 ± 1.45</td>
<td>4.75 ± 0.05</td>
<td>11 ± 10</td>
</tr>
<tr>
<td>4 CaHPO₄</td>
<td>7.96 ± 2.80</td>
<td>4.85 ± 0.05</td>
<td>33 ± 5</td>
</tr>
<tr>
<td>8 CaHPO₄</td>
<td>7.48 ± 4.22</td>
<td>4.90 ± 0.10</td>
<td>22 ± 5</td>
</tr>
<tr>
<td>4 Ca Mg chelate</td>
<td>1.80 ± 0.32</td>
<td>5.02 ± 0.10</td>
<td>20 ± 8</td>
</tr>
<tr>
<td>8 Ca Mg chelate</td>
<td>3.82 ± 2.05</td>
<td>5.05 ± 0.08</td>
<td>27 ± 7</td>
</tr>
<tr>
<td>4 oyster shell</td>
<td>2.95 ± 1.50</td>
<td>4.78 ± 0.08</td>
<td>20 ± 8</td>
</tr>
<tr>
<td>8 oyster shell</td>
<td>2.30 ± 0.85</td>
<td>4.80 ± 0.05</td>
<td>19 ± 7</td>
</tr>
</tbody>
</table>

Statistical effect of:
- Level of Ca      | NS                 | NS                | NS                            |
- Source of Ca     | 0.05               | 0.005             | NS                            |
- Interaction of   | NS                 | NS                | NS                            |
  level & source   | NS                 | NS                | NS                            |

a Mean ± SE (N = 6). Data expressed as the basis of wet weights of tissues.  

b Differences expressed as significant (P < 0.05 or P < 0.005) or not significant (NS).