Effect of cellulose phosphate on calcium and magnesium homeostasis: studies in normal subjects and patients with latent hypoparathyroidism

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(Received 1 October 1974)

Summary

1. The bivalent cation-binding agent, cellulose phosphate, was given for 6 days to four normal subjects and six patients with latent hypoparathyroidism (diagnosed by impaired response to EDTA infusion), all of whom were on a moderately low calcium diet.

2. In normal subjects, there was a prompt and sustained fall in urinary calcium with no change in plasma calcium, indicating increased tubular reabsorption. Plasma and urinary magnesium fell, without increase in tubular reabsorption. The urinary total hydroxyproline increased and Tm,p/glomerular filtration rate fell after 2 days; these changes were transient and were consistent with a transient increase in parathyroid hormone secretion.

3. In the hypoparathyroid patients, urinary calcium fell more slowly and a fall in plasma calcium occurred in several subjects, the extent and duration of which corresponded with parathyroid status determined by EDTA infusion. Urinary conservation of calcium was impaired but plasma and urinary magnesium fell as in normal subjects. Urinary total hydroxyproline did not change and Tm,p/glomerular filtration rate fell more slowly than in the normal subjects.

4. The relative contributions of increased tubular reabsorption and reduced filtered load to calcium conservation in response to calcium depletion depend on the prevailing level of parathyroid function; the former is more important when parathyroid function is normal, the latter when parathyroid function is impaired.

5. In the detection of reduced parathyroid reserve, the assessment based on the plasma calcium response to cellulose phosphate agrees closely with the assessment based on the degree of recovery from EDTA-induced hypocalcaemia.

Key words: calcium, cellulose phosphate, hypocalcaemia, hypoparathyroidism, magnesium.

Introduction

It is well known that normal subjects are able to reduce urinary calcium excretion in response to dietary calcium restriction (Bauer, Albright & Aub, 1929; McIntosh & Seraglia, 1963), but the timing and magnitude of this homeostatic adjustment are not clearly defined. Also, the relative importance of reduced filtered load (MacFadyen, Nordin, Smith, Wayne & Rae, 1965) and increased tubular reabsorption (Phang, Berman, Finerman, Neer, Rosenberg & Hahn, 1969) of calcium is unknown. A small fall in plasma calcium has been detected (MacFadyen et al., 1965), but it is uncertain whether this accounts adequately for the observed changes in urinary calcium.

The role of the parathyroid glands in mediating the response to dietary calcium deprivation is unclear. Kinetic studies with radiocalcium (Phang et al., 1969) show changes which are consistent with increased parathyroid hormone secretion but documentation of this is still lacking. A fall in plasma calcium in response to sodium phytate (the sodium
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TABLE 1. Details of experimental subjects

Means of two control values are given, and means of each group ± SEM. (a) and (b), the same patient studied on two different occasions. Normal ranges: plasma Ca: 2.4–2.7 mmol/l; plasma P: 0.9–1.5 mmol/l; urine Ca: 3.0–9.0 mmol/24 h; urine total hydroxyproline: 130–350 μmol/24 h (adult); Tm,p/GFR: 0.8–1.4 mmol/l. N.S. = not significant.

<table>
<thead>
<tr>
<th>No.</th>
<th>Age (years)</th>
<th>Sex</th>
<th>Plasma Ca (mmol/l)</th>
<th>Plasma P</th>
<th>Urine Ca (mmol/24 h)</th>
<th>Urine total hydroxyproline (μmol/24 h)</th>
<th>Tm,p/GFR (mmol/l)</th>
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</thead>
<tbody>
<tr>
<td>Patients</td>
<td></td>
<td></td>
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<td></td>
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<td></td>
</tr>
<tr>
<td>1 (a)</td>
<td>44</td>
<td>F</td>
<td>2.38</td>
<td>1.02</td>
<td>2.32</td>
<td>193</td>
<td>0.89</td>
</tr>
<tr>
<td>2</td>
<td>41</td>
<td>F</td>
<td>2.39</td>
<td>1.24</td>
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<td>2.40</td>
<td>1.31</td>
<td>3.08</td>
<td>334</td>
<td>1.65</td>
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<tr>
<td>4</td>
<td>47</td>
<td>F</td>
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<td>1.24</td>
<td>4.62</td>
<td>263</td>
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<tr>
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<td>56</td>
<td>F</td>
<td>2.46</td>
<td>1.55</td>
<td>3.82</td>
<td>174</td>
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<td>6</td>
<td>39</td>
<td>F</td>
<td>2.42</td>
<td>1.16</td>
<td>2.80</td>
<td>355</td>
<td>1.01</td>
</tr>
<tr>
<td>1 (b)</td>
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<td>F</td>
<td>2.46</td>
<td>1.09</td>
<td>5.94</td>
<td>253</td>
<td>1.27</td>
</tr>
<tr>
<td>Mean ± SEM</td>
<td></td>
<td></td>
<td>2.41 ± 0.02</td>
<td>1.21 ± 0.08</td>
<td>3.45 ± 0.55</td>
<td>250 ± 28</td>
<td>1.08 ± 0.11</td>
</tr>
<tr>
<td>Control subjects</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>23</td>
<td>F</td>
<td>2.41</td>
<td>1.19</td>
<td>3.45</td>
<td>157</td>
<td>0.92</td>
</tr>
<tr>
<td>8</td>
<td>24</td>
<td>M</td>
<td>2.51</td>
<td>1.13</td>
<td>9.06</td>
<td>341</td>
<td>0.94</td>
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<tr>
<td>9</td>
<td>22</td>
<td>M</td>
<td>2.59</td>
<td>1.18</td>
<td>6.25</td>
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<td>10</td>
<td>37</td>
<td>M</td>
<td>2.48</td>
<td>1.26</td>
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<td>231</td>
<td>1.14</td>
</tr>
<tr>
<td>Mean ± SEM</td>
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<td></td>
<td>2.50 ± 0.04</td>
<td>1.19 ± 0.03</td>
<td>6.10 ± 1.15</td>
<td>311 ± 78</td>
<td>0.98 ± 0.06</td>
</tr>
</tbody>
</table>

P < 0.05 N.S. (P > 0.8) P < 0.05 N.S. (P > 0.3) N.S. (P > 0.5)

salt of inositol hexaphosphoric acid), which reduces calcium absorption, was interpreted as evidence of impaired parathyroid function by Smith, Davis & Fourman (1960), and by Davis, Fourman & Smith (1961). Sodium phytate forms an insoluble complex with calcium and magnesium but, in spite of this, much of the phytate phosphorus is hydrolysed and absorbed, so that the effects of sodium phytate are dependent on phosphate loading as well as on calcium and magnesium deprivation (Parfitt, Higgins, Nassim, Collins & Hilb, 1964).

Cellulose phosphate is an ion-exchange substance with a particular affinity for bivalent cations (Parfitt et al., 1964; Dent, Harper & Parfitt, 1964). In contrast to sodium phytate, only a small fraction of the cellulose phosphate is hydrolysed in the gut so that the effects of administration are due primarily to calcium and magnesium deprivation. In the present study, cellulose phosphate has been given to normal subjects and to patients with latent (normocalcaemic) hypoparathyroidism, diagnosed by impaired recovery from EDTA-induced hypocalcaemia (Parfitt, 1969a).

The purpose was threefold. First, to characterize the normal response to simultaneous calcium and magnesium deprivation in more detail, secondly to examine the effect of impaired parathyroid function on urinary calcium conservation, and, thirdly, to compare cellulose phosphate administration and EDTA(1) infusion as tests of parathyroid function.

Methods

Four normal subjects and six patients with normocalcaemic hypoparathyroidism were studied. The patients were cases nos. 11–13 and nos. 18–20 of a previous report (Parfitt, 1969a). Pertinent details are given in Table 1. The patients were selected on the basis of four criteria: (1) the occurrence of overt tetany after thyroid surgery performed more than 2 years previously; (2) normal plasma calcium without treatment at the time of study; (3) a recent assessment of parathyroid function by EDTA infusion (Parfitt, 1969a); (4) willingness to participate in the study, the purpose of which was explained to each patient. One patient (no. 1) was studied again after an interval of 2 years.

The EDTA infusions were performed and interpreted as previously described (Parfitt, 1969a). The responses were classified as: grade 0 (plasma calcium ≥95% of preinfusion values at 10 and 22 h after end of infusion; area under curve of recovery at

(1) Abbreviations: EDTA, ethylenediamine tetra-acetate; GFR, glomerular filtration rate.
Table 2. Response to EDTA infusion in patients with normocalcaemic hypoparathyroidism and in two control subjects

Unchelated calcium is expressed as percentage of the preinfusion value, at 10 h and 22 h after the end of infusion. Area (\%) = area under curve of recovery as fraction of area corresponding to instantaneous recovery. Numbering of subjects as in Table 1.

<table>
<thead>
<tr>
<th>Patient no.</th>
<th>Calcium (%)</th>
<th>Area (%)</th>
<th>Grade EDTA</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>10 h</td>
<td>22 h</td>
<td>10 h</td>
</tr>
<tr>
<td>1 (a)</td>
<td>86</td>
<td>89</td>
<td>14.7</td>
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<tr>
<td>2</td>
<td>83</td>
<td>89</td>
<td>26.4</td>
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<td>3</td>
<td>83</td>
<td>88</td>
<td>34.3</td>
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<td>94</td>
<td>97</td>
<td>26.7</td>
</tr>
<tr>
<td>5</td>
<td>92</td>
<td>97</td>
<td>44.2</td>
</tr>
<tr>
<td>6</td>
<td>98</td>
<td>100</td>
<td>46.7</td>
</tr>
<tr>
<td>Control subjects</td>
<td></td>
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<td></td>
</tr>
<tr>
<td>9</td>
<td>99</td>
<td>102</td>
<td>59.5</td>
</tr>
<tr>
<td>10</td>
<td>100</td>
<td>102</td>
<td>63.6</td>
</tr>
</tbody>
</table>

Changes in the renal tubular reabsorption of calcium were assessed by comparing the relation between urinary calcium (expressed as \(\mu\)mol/100 ml creatinine clearance) and plasma calcium with the ranges established for this relation in different states of parathyroid function by means of calcium infusion (Peacock, Robertson & Nordin, 1969). Changes in the renal tubular reabsorption of phosphate were assessed by estimating \(T_{\text{m,p}}/GFR\) from a nomogram (Bijvoet & Van der Sluys Veer, 1972). This quantity is less dependent on changes in plasma phosphate than other indices of phosphate reabsorption. The normal range is 0.8–1.4 mmol/l.

Results

Comparison of the hypoparathyroid patients as a group with the controls (Fig. 1)

Calcium. There was no change in plasma calcium in the normal subjects, either individually or collectively. In the hypoparathyroid patients the mean control value was lower (Table 1) and there was a significant fall with cellulose phosphate (0.1 ± 0.04 mmol/l, \(P < 0.01\) by paired \(t\)-test). Urinary calcium excretion fell promptly in both groups, by 75% in the control subjects and by 69% in the hypoparathyroid patients. Analysis of variance showed no significant difference between the mean values on days 3–8 in the control subjects, but a significantly larger value on day 3 in the patients, indicating that the lowest value was attained on the first day of the experimental regime in the control subjects but not until the second day in the hypoparathyroid patients.

Magnesium. The plasma magnesium fell steadily in both groups, with no significant difference in the levels reached. Urinary magnesium excretion fell in parallel with calcium excretion, by 76% in the control subjects and by 77% in the hypoparathyroid patients. As with calcium, maximal conservation was achieved on day 3 in the control subjects and day 4 in the patients.

Inorganic phosphate. The plasma inorganic phosphate fell transiently on day 5 in the control subjects but did not change in the patients. Urinary inorganic phosphate excretion increased by about 8 mmol/24 h in both groups, the percentage change being greater in the hypoparathyroid patients.

Phosphate reabsorption. There was a significant
Fig. 1. Effect of cellulose phosphate and low calcium diet on four normal subjects and six patients with normocalcaemic hypoparathyroidism. The vertical bars indicate SEM. Note the change in scale between (a) and (b) for the urinary constituents. THP = total hydroxyproline. $T_{m,p}/\text{GFR}$ = maximum tubular reabsorption of phosphate per unit of glomerular filtration rate, estimated from a nomogram.

fall in $T_{m,p}/\text{GFR}$ ($P < 0.05$ by paired $t$-test) in both groups. This occurred earlier in the normal subjects than in the patients.

Total hydroxyproline. There was an increase of 35% in urinary total hydroxyproline excretion on day 4 in the normal subjects. This difference was not statistically significant because the peak increment occurred on different days in each individual, but the peak value on day 4, 5 or 6 in each subject exceeded the 95% confidence limits derived from the control values. No significant change in urinary total hydroxyproline occurred in the hypoparathyroid patients, either individually or collectively.

Sodium. There was a modest but significant increase in sodium excretion in both groups, no doubt due to the sodium contained in the cellulose phosphate.
Cellulose phosphate and hypoparathyroidism

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Grade

1

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Time (days)

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2. Changes in plasma and urinary calcium in patients with normocalcaemic hypoparathyroidism classified according to parathyroid status determined by EDTA infusion. O—O, Results in the normal control subjects; •—•, results in the hypoparathyroid patients. Vertical bars indicate SEM.

TABLE 3. Mean changes in plasma calcium concentration during cellulose phosphate administration

Deviation (mean±SEM) in plasma calcium (mmol/l) from mean values of control measurements (Table 1) during cellulose phosphate administration is shown. P values refer to significance of difference from normal subjects in the magnitude of these deviations.

<table>
<thead>
<tr>
<th>Parathyroid status</th>
<th>2 days</th>
<th>4 days</th>
<th>6 days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal</td>
<td>+0·02±0·02</td>
<td>−0·00±0·02</td>
<td>+0·01±0·02</td>
</tr>
<tr>
<td>Grade 0</td>
<td>+0·03±0·04 (P&gt;0·7)</td>
<td>+0·05±0·04 (P&gt;0·2)</td>
<td>+0·04±0·02 (P&gt;0·2)</td>
</tr>
<tr>
<td>Grade 1</td>
<td>−0·17±0·04 (P&lt;0·02)</td>
<td>−0·04±0·04 (P&gt;0·3)</td>
<td>+0·04±0·01 (P&gt;0·3)</td>
</tr>
<tr>
<td>Grade 2</td>
<td>−0·24±0·09 (P&lt;0·05)</td>
<td>−0·30±0·04 (P&lt;0·005)</td>
<td>−0·30±0·03 (P&lt;0·001)</td>
</tr>
</tbody>
</table>

Subdivision of the hypoparathyroid group according to the EDTA response (Fig. 2, Table 3)

In three patients with a grade 2 response to the EDTA infusion, there was a substantial fall in plasma calcium (0·24 mmol/l) which continued throughout the period of study; in one patient with idiopathic epilepsy (case 2) a grand mal seizure occurred at the end of the period of hypocalcaemia. In two patients with a grade 1 response there was an initial fall of moderate magnitude (0·17 mmol/l) by the end of day 4, which recovered to the pretreatment value by the end of day 8. Finally, in one patient with a grade 0 response and in case 1, in whom it was not possible to repeat the EDTA infusion at the time of the second cellulose phosphate study, there was no significant change in plasma calcium, as in the normal subjects.

Changes in other variables in the three subgroups. There was no significant difference between the three subgroups for plasma and urinary phosphate, Tm,p/GFR, plasma and urinary magnesium, urinary
Changes in tubular reabsorption of calcium in the three subgroups. In Fig. 3, the relationship between $C_{aE}$ (urinary calcium in $\mu$mol/100 ml creatinine clearance) and total plasma calcium is shown. For both normal subjects and those with grade 0 normocalcaemic hypoparathyroidism, urinary calcium conservation occurred with no change in plasma calcium. In grade 1 calcium conservation was less effective, and in grade 2 calcium conservation was achieved only at the expense of a substantial fall in plasma calcium. Calcium reabsorption did not increase at all, the points following closely the theoretical curve (Fig. 3).

Discussion

Normal subjects are evidently able to reduce urinary calcium promptly in response to calcium deprivation. As previously found by MacFadyen et al. (1965), the maximum fall occurred on the first day with little change thereafter. By contrast, maximum sodium conservation on a low sodium diet is not attained for
Calcium are restricted with a diet which is completely electrolyte free, urinary calcium falls in a stepwise manner, like sodium (Vertes, 1960). This immediate short-term response in urinary calcium must be distinguished from long-term adaptive changes whereby urinary calcium may continue to fall slowly for many months (Malm, 1958), since the mechanisms of these two types of calcium conservation may be different.

In previous studies the extent of fall in urinary calcium was partly dependent on the degree of dietary restriction (Table 4), but comparably low excretion of urinary calcium was achieved only in the study of Bauer et al. (1929). Their experimental diets were moderately low in sodium (40–70 mmol/24 h) and since dietary sodium restriction may reduce urinary calcium (Kleeman, Bohannan, Bernstein, Ling & Maxwell, 1964), the low sodium intake may have contributed to the unusually low urinary calcium that was found. In the present study, both sodium intake and excretion were increased. The major difference is that the regimen of moderate dietary calcium restriction with cellulose phosphate also reduces the absorption of magnesium and it is possible that simultaneous magnesium depletion and hypomagnesaemia may increase the efficiency of urinary calcium conservation.

From what is known of parathyroid physiology, it would be expected that dietary calcium restriction, which leads to a small but significant fall in plasma calcium (MacFadyen et al., 1965), would lead to parathyroid stimulation (Phang et al., 1969). In the present study, the increase in urinary total hydroxyproline, and increase in tubular reabsorption of calcium, both of which occurred in the normal subjects but not in those with normocalcaemic hypoparathyroidism, suggest that secretion of parathyroid hormone was increased. This may have occurred without a change in plasma calcium because of the hypomagnesaemia, which stimulates secretion of parathyroid hormone directly (Buckle, Care, Cooper & Gittelman, 1968). Pak, Delea & Bartter (1974) found normal parathyroid hormone levels in patients with idiopathic hypercalciuria who had been on cellulose phosphate for 1 year or longer, but no earlier measurements have been reported. The decrease in Tm,p/GFR which occurred in both groups may be unrelated to parathyroid function since a similar fall occurs in completely hypoparathyroid patients on vitamin D in response to increased dietary phosphorus (A. M. Parfitt, unpublished work).

The conclusion that secretion of parathyroid hormone probably increased in the normal subjects is strengthened by more detailed consideration of the changes in the patients with normocalcaemic hypoparathyroidism. The data in Fig. 2 and Fig. 3 indicate that the separate contributions of reduced filtered load and increased tubular reabsorption to renal calcium conservation is determined by the prevailing level of parathyroid function. Of the four groups, the fall in urinary calcium was wholly dependent on increased tubular reabsorption of calcium in either normal subjects or hypoparathyroid patients, the fall in urinary magnesium being consistent with the fall in plasma magnesium in every case. Evidently urinary conservation of magnesium, unlike calcium, is unaffected by a moderate deficiency of parathyroid hormone.

The close correspondence between the two different methods of assessing parathyroid function is of interest and lends support to the interpretations of impaired response to EDTA infusion advanced previously (Parfitt, 1969a). The results suggest that in the recognition of impaired parathyroid reserve, either test may be employed. EDTA infusion gives an answer more quickly, but requires admission to hospital. The potential hazards of EDTA infusion, although real, are easy to avoid with careful control of the rate of infusion, and the only serious complication of either test (precipitation of a grand mal seizure) occurred with cellulose phosphate in a patient who had previously undergone an EDTA infusion without incident. The correspondence between the two tests was much better than that found by Davis, Fourman & Smith (1961) using sodium phytate instead of cellulose phosphate. This is partly because of the increased accuracy of the EDTA infusion, when interpretation is based on changes at two different times rather than at only one time, and partly because of the additional complication of phosphate loading, which is less with cellulose.
phosphate than with phytate. Based on previous balance data (Parfitt et al., 1964; Dent et al., 1964), about 21% of the phosphate in cellulose phosphate is absorbed compared with 46% for sodium phytate and 81% for sodium phosphate. From the magnitude of the hypocalciuric effect of the latter (Parfitt et al., 1964) it can be estimated that the fall in urinary calcium attributable to the effect of increased phosphorus absorption was only 0.5–1.0 mmol/24 h, much less than that observed in any group.

There was no evidence that the fall in plasma calcium in those with grade 1 and 2 hypoparathyroidism was related to the amount of phosphorus absorbed (in contrast to hypocalcaemia induced by sodium phytate), and magnesium depletion does not lower the plasma total calcium in normal subjects for several weeks (Shils, 1969). Consequently a reduction in the supply of available calcium, together with inability to augment tubular reabsorption of calcium, were probably the main factors leading to hypocalcaemia in the patients with impaired parathyroid function.

Acknowledgments

I thank Dr W. Douglas, Dr. B. Hirschfeld, Dr D. Hobson and Dr I. McKelvie for referring patients; J. Wilson, C. Cornish, T. French, S. McCullum and G. Chalk for giving (at different times) technical assistance; Reeve Angel and Co. for a gift of cellulose phosphate; and the University of Queensland and the National Health and Medical Research Council of Australia for financial support.

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