The differential diagnosis of hypercalcemia may be difficult. A readily available uncomplicated test that distinguishes between hypercalcemia of parathyroid and nonparathyroid origin would be most useful.

Plasma calcium concentration is a major factor in regulating the rate of parathormone (PTH) secretion. There is an inverse linear relationship between serum calcium and PTH levels when PTH secretion is under normal homeostatic control [1]. The system functions as if there were a calciostat that normally operates around a set point of 10 mg/100 ml [2]. When the serum calcium concentration decreases below 10 mg/100 ml, the rate of PTH secretion increases, and when the serum calcium is elevated above 10 mg/100 ml, PTH secretion is suppressed. Conversely, in patients with hyperparathyroidism due to adenoma or hyperplasia, this normal negative-feedback homeostatic control is lost and PTH levels are increased without respect to serum calcium levels, although complete autonomy is seldom achieved. Thus, patients with hypercalcemia due to hyperparathyroidism have elevated levels of PTH, whereas patients with hypercalcemia due to other causes have decreased or negligible PTH plasma levels. Since the levels of PTH are different in these two groups of patients, perhaps some of the metabolic effect of PTH could be used to distinguish between these two groups.

In addition to raising the serum calcium concentration, PTH has other functions. Its action on the proximal renal tubule to decrease the tubular reabsorption of phosphate has been well documented. This produces a variable decrease in serum phosphate levels, but the reliability of this decrease as a means of diagnosing hyperparathyroidism has frequently been questioned [3-5]. Nordin [6] and Hillman, Au, and Barter [7] have reported that PTH also decreases the reabsorption of bicarbonate from the proximal renal tubule, leading to increased reabsorption of chloride and producing a mild hyperchloremic renal tubular acidosis. Wills and McGowan [8] have reported that the hyperchloremia occurring in hyperparathyroidism is useful in the differential diagnosis of hypercalcemia, but other investigators have questioned this [9,10].

To determine the diagnostic usefulness of these metabolic effects of PTH upon phosphate and bicarbonate excretion, we measured and compared serum chloride and phosphate levels in two groups of patients. One group had hypercalcemia due to hyperparathyroidism, and the other group had hypercalcemia due to a variety of other causes. Some of these patients were included in a previous report by Palmer, Nelson, and Bacchus [11].

Clinical Material

Chloride and phosphate levels were measured and chloride/phosphate ratios determined in eighty-four patients from the medical and surgical services at Riverside General Hospital and Loma Linda University Hospital. Thirty-four patients had surgically proved hyperparathyroidism, thirty-two with adenoma and two with hyperplasia. In fifty patients, hypercalcemia was due to a variety of other conditions. (Table I.)

Serum calcium was measured with the SMA 12/60 Autoanalyzer (normal, 8.5 to 10.5 mg/100 ml). Phosphate was also measured with the SMA 12/60 Autoanalyzer (normal, 2.4 to 5.0 mg/100 ml), and serum chloride was measured with the SMA 6/60 or Autoanalyzer 2 (normal, 95 to 106 mEq/l). Serum phosphate, calcium,
and chloride levels were measured in the same specimen from each patient. Calcium and phosphate levels were expressed in milligrams per 100 milliliters and chloride levels in milliequivalents per liter. The ratio of chloride to phosphate was calculated for each patient in the following manner:

\[
\text{Chloride (mEq/L)} / \text{Phosphate (mg/100 ml)} = \text{Chloride/Phosphate Ratio}
\]

All these patients had normal renal function and none were receiving medications known to influence these parameters.

**Results**

The serum calcium levels in these two groups of hypercalcemic patients were almost identical. (Figure 1.) In patients with hyperparathyroidism, the range was 10.6 to 14.4 mg/100 ml with a mean of 12.1 mg/100 ml. In the group without hyperparathyroidism, the range was 10.6 to 16.4 mg/100 ml with a mean of 12.5 mg/100 ml (p = 0.70).

Conversely, there were significant differences in the serum chloride and phosphate levels between the two groups of patients. The serum chloride levels in patients with hyperparathyroidism ranged from 99 to 120 mEq/L with a mean of 106.7 mEq/L, which was consistently higher than the range of 51 to 112 mEq/L and a mean of 95.6 mEq/L in patients with hypercalcemia due to other causes. (Figure 2.) The serum chloride levels were above 102 mEq/L in 85 per cent of patients with hyperparathyroidism and below 102 mEq/L in 86 per cent of other hypercalcemic patients. These differences were significant at the 0.1 per cent level (p = 0.001). (Table II.)

The phosphate levels in the hyperparathyroid group were 1.2 to 3.4 mg/100 ml with a mean of 2.61 mg/100 ml, which was significantly lower than levels in the nonparathyroid group in which the corresponding values were 2.8 to 6.8 mg/100 ml with a mean of 4.35 mg/100 ml. (Figure 3.) The serum phosphate levels were 3 mg/100 ml or below in 82 per cent of the patients with hyperparathyroidism and above 3 mg/100 ml in 98 per cent of the others. These differences were also highly statistically significant (p = 0.001). (Table II.)

The differences in calculated chloride/phosphate ratios were of even greater significance. The chloride/phosphate ratio in patients with hyper-
TABLE II  Comparison of Accuracy of Differential Diagnosis of Hypercalcemia by Chloride and Phosphate Levels and the Chloride/Phosphate Ratio

<table>
<thead>
<tr>
<th>Diagnostic Test</th>
<th>Hyperparathyroid Patients</th>
<th>Other Hypercalcemic Patients</th>
<th>Per cent Correctly Differentiated (Accuracy)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Number</td>
<td>Per cent</td>
<td>Number</td>
</tr>
<tr>
<td>Chloride &gt; 102 mEq/L</td>
<td>29</td>
<td>85</td>
<td>7</td>
</tr>
<tr>
<td>Chloride &lt; 102 mEq/L</td>
<td>5</td>
<td>15</td>
<td>43</td>
</tr>
<tr>
<td>Phosphate &lt; 3 mg/100 ml</td>
<td>20</td>
<td>62</td>
<td>1</td>
</tr>
<tr>
<td>Phosphate &gt; 3 mg/100 ml</td>
<td>6</td>
<td>18</td>
<td>49</td>
</tr>
<tr>
<td>Cl/PO₄ ratio &gt; 33</td>
<td>32</td>
<td>94</td>
<td>2</td>
</tr>
<tr>
<td>Cl/PO₄ ratio &lt; 33</td>
<td>2</td>
<td>6</td>
<td>48</td>
</tr>
</tbody>
</table>

Figure 3. Serum phosphate levels were below 3 mg/100 ml in 82 per cent of hyperparathyroid patients and above 3 mg/100 ml in 98 per cent of other patients (p < 0.001).

Figure 4. The calculated chloride/phosphate ratio was above 33 in 94 per cent of patients with hyperparathyroidism and below 33 in 96 per cent of other hypercalcemic patients (p < 0.001).

Figure 5. Response in serum calcium levels and calculated chloride/phosphate ratios in fifteen hyperparathyroid patients treated by parathyroidectomy. There was a significant decrease in both values in all cases.

Many authors have questioned the value of serum phosphate and chloride measurements in the differential diagnosis of hypercalcemia, because chloride and phosphate levels are frequently lower in patients with hyperparathyroidism than in those with other causes of hypercalcemia.
TABLE III  Summary of Laboratory Characteristics of Hypercalcemic Patients*

<table>
<thead>
<tr>
<th>Laboratory Data</th>
<th>Normal Value</th>
<th>Hyperparathyroid Patients</th>
<th>Other Hypercalcemic Patients</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum calcium</td>
<td>8.5–10.3 mg/100 ml</td>
<td>&gt;10.3 mg/100 ml</td>
<td>&gt;10.3 mg/100 ml</td>
</tr>
<tr>
<td>Serum chloride</td>
<td>95–105 mEq/L</td>
<td>&gt;102 mEq/L</td>
<td>&lt;102 mEq/L</td>
</tr>
<tr>
<td>Serum phosphate</td>
<td>2.1–5 mg/100 ml</td>
<td>&lt;3.0 mg/100 ml</td>
<td>&gt;3.0 mg/100 ml</td>
</tr>
<tr>
<td>Cl/PO₄ ratio</td>
<td>. . .</td>
<td>&gt;33</td>
<td>&lt;33</td>
</tr>
</tbody>
</table>

*In more than 80 per cent of the eighty-four patients in this series.

in the “normal range” in both hyperparathyroid and in other hypercalcemic patients [9,10]. Although results vary from series to series, approximately 40 to 60 per cent of patients with hyperparathyroidism have serum phosphate levels above 2.4 mg/100 ml. In the series reported by Pyrah, Hodgkinson, and Anderson [3], only 54 per cent of hyperparathyroid patients had serum phosphate levels below 2.4 mg/100 ml, but the remainder were all in the low normal range. In our series, only 35 per cent of hyperparathyroid patients had phosphate levels below 2.4 mg/100 ml, but 82 per cent had phosphate levels below 3 mg/100 ml. In the other group of hypercalcemic patients, 98 per cent had phosphate levels above 3 mg/100 ml.

A similar situation exists with respect to chloride. Mallette et al [4] found serum chloride levels above 107 mEq/L in only 39 per cent of hyperparathyroid patients, but Wills and McGowan [8] reported a series of sixty-one hypercalcemic patients in which chloride levels were above 102 mEq/L in 97 per cent of patients with hyperparathyroidism and below 102 mEq/L in 96 per cent of other hypercalcemic patients. In the hyperparathyroid patients in our series, chloride levels were distinctly abnormal (above 105 mEq/L) in only 38 per cent of patients, but 85 per cent of these patients had chloride levels above 102 mEq/L, and 86 per cent of other hypercalcemic patients had chloride levels below 102 mEq/L. (Table II.)

Since PTH tends to decrease serum phosphate and increase serum chloride levels and the absence of PTH produces opposite changes in chloride and phosphate levels, we have explored the possibility that a simple chloride/phosphate ratio might be more useful diagnostically.

The chloride/phosphate ratio was higher than 33 in 94 per cent of patients with hyperparathyroidism and lower than 33 in 96 per cent of other hypercalcemic patients (p < 0.001). Thus, although both serum chloride and phosphate levels were useful in differentiating between these two groups of patients, the chloride/phosphate ratio was even more reliable in distinguishing between these patients, with an overall accuracy of 95 per cent. (Table II.) The characteristic laboratory findings and their significance in these two groups of hypercalcemic patients are summarized in Table III.

These changes in chloride and phosphate levels are not applicable in normocalcemic patients and, therefore, cannot be used for the diagnosis of normocalcemic hyperparathyroidism. Another possible error could occur in a patient with ectopic PTH-like secretion, usually from a nonendocrine malignant tumor. On the basis of these tests, such a patient would probably appear to have hyperparathyroidism, which would be true—but from a nonparathyroid source.

The measurement of serum chloride and phosphate levels and the calculation of the chloride/phosphate ratio provide a group of readily available screening tests for the evaluation of patients with hypercalcemia. Although these measurements may not provide a definitive diagnosis, they aid in the selection of more specific diagnostic tests, such as parathormone immunoassay, prednisone suppression test, and protein electrophoresis, for definitive diagnosis.

Summary

The serum chloride and phosphate levels were measured and the chloride/phosphate ratios calculated in a group of eighty-four hypercalcemic patients. Although patients with hyperparathyroidism frequently had phosphate levels in the low normal range (< 3 mg/100 ml) and chloride levels in the high normal range (> 102 mEq/L), they were nevertheless significantly different from the groups of patients with nonparathyroid hypercalcemia in whom phosphate levels were usually higher (> 3 mg/100 ml) and chloride levels usually lower (< 102 mEq/L). The chloride/phosphate ratio was higher than 33 in 94 per cent of hyperparathyroid patients and lower than 33 in 96 per cent of other hypercalcemic patients. Thus, the measurements of serum phosphate and chloride levels and the calculation of the chloride/phosphate ratios were useful diagnostic screening tests that discriminated between patients with hypercalcemia of parathyroid and nonparathyroid origin with an accuracy of 95 per cent.
References


Discussion

H. Earl Gordon (Los Angeles, CA): These studies clearly show that the chloride/phosphate ratio can be an effective discriminant between the hypercalcemia of primary hyperparathyroidism and hypercalcemia due to other causes; however, it is important to consider this ratio in its proper perspective. As advocated by the authors, it is of primary value as a screening test. I would seriously question its potential in those patients who present the greatest diagnostic challenge. I would like to ask the authors if they have had an opportunity to evaluate the accuracy of the test in those patients with borderline elevations in serum calcium levels.

A second diagnostic dilemma occurs in a patient with suspected hyperparathyroidism who has concomitant mild to moderate renal failure with an elevated creatinine level, in the range of 2.0 to 4.0. Such a patient excretes phosphate poorly, and I would therefore predict that the chloride/phosphate ratio would not fall in the diagnostic range for patients with hyperparathyroidism. Although the authors' series did not include any patients with abnormal renal function, I would be interested in their comments on patients in this category.

A third diagnostic problem is identification of those patients with an ectopic source of a parathyroid-like hormone from a nonendocrine malignant tumor. As the authors have implied, the chloride/phosphate ratio probably would not be useful in such patients since the aberrations in electrolyte metabolism are frequently similar to those in patients with primary hyperparathyroidism. Adding to the complexity of the problem is the accumulating evidence that patients with primary hyperparathyroidism have a significantly increased risk of malignant disease in another organ. In a recent report from Memorial Hospital in New York, twelve of fifty consecutive patients with primary hyperparathyroidism had active coexistent cancer. Thus, the presence of malignant disease and hypercalcemia does not necessarily mean that one is related to the other but may well indicate a combination of cancer and primary hyperparathyroidism.

Despite these limitations, the chloride/phosphate ratio should prove valuable in the evaluation of hypercalcemic patients. However, its validity depends on the absence of any complicating aspect of the illness that might independently alter the phosphate and chloride levels.

Orlo H. Clark (San Francisco, CA): Doctor Leon Goldman looked forward to discussing this paper but has recently been ill. He did say, however, that the chloride/phosphate ratio is a useful test.

The authors stated that they determined the serum chloride and phosphate concentrations in the same sample of blood. However, I am unsure whether they selected the sample with the highest chloride and lowest phosphate concentration or used mean values when more than one serum determination was available. In a review of the records of fifteen patients who had determination of preoperative serum chloride and phosphate values and who underwent successful operation for primary hyperparathyroidism at the Veterans Administration Hospital in San Francisco during the past eighteen months, we found that twelve of fifteen (80 per cent) had a chloride/phosphate ratio greater than 33 when mean serum concentrations of chloride and phosphate were used. When the highest chloride and lowest phosphate levels were used, all but one patient (93 per cent) had an elevated ratio. However, when the lowest chloride and highest phosphate concentrations were selected, only seven of fifteen (47 per cent) had an elevated ratio.

I also wonder why the hyperchloremia associated with hyperparathyroidism does not decrease after parathyroidectomy, since it is attributed to a parathormone-induced bicarbonate loss and also to hemoconcentration. In seven of eight patients who underwent curative parathyroid operation during the past eighteen months and had postoperative serum chloride determinations, the chloride level decreased appreciably postoperatively in only one patient. The chloride/phosphate ratio did decrease after operation in seven of eight patients, but this was due to an increase in serum phosphate concentration rather than a decrease in serum chloride concentration. In fact, in 50 per cent of these patients the ratio remained higher than 33 postoperatively.

I have several questions for the authors: (1) How were the chloride and phosphate values selected and was this done before or after operation? (2) Do you think phosphate deprivation might improve the results of the ratio, especially in normocalcemic hyperparathyroid patients?
Why is there no decrease in the serum chloride level soon after successful parathyroidectomy if hyperchloremia is due to the mechanisms just described?

George R. Mason (Baltimore, MD): I would like to urge reconsideration of some of these data. I note that in this group major diagnoses have been made on the basis of results from the SMA 12/60 Autoanalyzer or some other screening process. Most of us use recurrent elevation of calcium or recurrent depression of phosphorus in combination as a basis for a presumptive diagnosis of hyperparathyroidism. There is another group with carcinoma or other disease such as sarcoid that would lead to the diagnosis of hypercalcemia.

It is of interest, as Doctor Gordon pointed out, to eliminate these from the group and determine the percentage of success in the questionable group. We now have a study series of about seventy-five patients that includes those with hypercalcemia and hypophosphatemia, fairly clearly of hyperparathyroid origin, and in this group we have 100 per cent diagnostic accuracy. It is the other group that bothers us, those with some renal dysfunction that our endocrinologists have labeled idiopathic hypercalcinuria. There have been six such patients and our diagnostic accuracy is about 50 per cent. Frankly, I am rather surprised it is that high. I would urge you to look at your data and eliminate those with a fairly clear diagnosis, barring concomitant disease, and determine your rate of diagnostic success with this test.

Leonard Rosoff (Los Angeles, CA): As I understand it, Doctor Reeves did not say that they were using the chloride/phosphate ratio as a specific diagnostic test for hyperparathyroidism, but rather to differentiate hypercalcemia due to malignant disease from hypercalcemia due to hyperparathyroidism. When we first learned of these findings by the group at the Loma Linda Medical School, we reviewed some of the laboratory studies obtained in our series of patients who underwent operation for hyperparathyroidism (now approaching 400) and found that the chloride/phosphate ratio was higher than 30 in most instances and closely matched the authors' findings in the patients with hypercalcemia due to parathyroid disease. We do not have data available at the present time regarding the chloride/phosphate ratio in the patients with hypercalcemia due to malignant disease.

At the Los Angeles County-USC Medical Center, some of the patients with hypercalcemia associated with malignant disease have had extremely low or undetectable levels of serum parathormone in the bioassays done in our research laboratories. This finding may prove to be of value in determining whether a patient with hypercalcemia has hyperparathyroidism or an occult tumor. However, further observations on a variety of malignant conditions associated with hypercalcemia are needed and are being obtained currently. Another problem in differential diagnosis is the coexistence of a parathyroid adenoma and malignant disease of another organ system.

Regardless of the term "ectopic hyperparathyroidism" as a designation for hypercalcemia of malignant rather than parathyroid origin, I believe this is a misnomer. Ectopic implies displacement or abnormal location of a structure anatomically; therefore, as a surgeon, I prefer to use the word "ectopic" for a parathyroid tumor that is located in an abnormal anatomic position rather than for hypercalcemia resulting from nonparathyroid malignant disease.

William F. Pollock (Santa Monica, CA): I would like to ask Doctor Longerbeam, since uremia and failing kidney function were not listed as medical problems in this group of hypercalcemic patients, whether renal function in all of these patients was essentially within normal limits, and whether the serum protein levels and other values were essentially normal pre- and postoperatively.

In the nomogram showing the postoperative reduction in the chloride/phosphate ratio, it appeared that a substantial number of the patients had a ratio in excess of 33 postoperatively. Doctor Clark has already documented this at the University of California, but I wonder whether a mean has been calculated for the postoperative ratio in the patients who have been reported on today.

Jerrold K. Longerbeam (closing): I shall attempt to answer the many questions asked, but not necessarily in the order in which they were raised and without attempting to identify the discussant in each case.

First, the presence or significance of hyperchloremia in hyperparathyroidism has not been accepted by all investigators and its mechanism is still being debated. We believe it is real and results from the action of parathormone on the distal tubule causing bicarbonate excretion and increased chloride reabsorption to maintain electrical neutrality in body fluids. In this series of patients, the chloride and phosphate determinations were made in a single serum specimen, and these levels were used to calculate the chloride/phosphate ratio. It has been suggested that a mean of three chloride and phosphate determinations would be more accurate in calculating chloride/phosphate ratios, but we have not done this.

The chloride and phosphate levels and the chloride/phosphate ratio have been particularly useful in those patients with borderline calcium elevations, since in this group of patients the chloride/phosphate ratio was always above 33 in those subsequently found to have hyperparathyroidism. We, like Doctor Clark, have observed a rapid increase in phosphate levels but a much slower decrease in chloride levels post parathyroidectomy; therefore, determination of postoperative chloride/phosphate ratios is best delayed for at least one month after operation.

Finally, as emphasized by Doctor Gordon, this test is not applicable in any patient with a significant degree of renal failure, because renal failure frequently causes the serum phosphate level to be abnormally elevated. All patients in this series had normal renal function, as defined by normal blood urea nitrogen, creatinine, and creatinine clearance values.