STUDIES OF CALCIUM AND PHOSPHORUS METABOLISM

IV. The Effect of the Parathyroid Hormone[1]

By FULLER ALBRIGHT,[2] WALTER BAUER,[3] MARION ROPES and JOSEPH C. AUB

(*From the Medical Clinic of the Massachusetts General Hospital, Boston*)

In our studies of calcium and phosphorus metabolism considerable data have accumulated on the physiological effect of parathormone (Collip's preparation) on normal and pathological subjects. We are presenting in this paper such of these data as seem to us of value in explaining its action.

As explained in a previous paper (1) of this series, we employ a diet inadequate in calcium when studying the effect of various factors on the endogenous calcium metabolism. In a control series of thirteen "normal" male individuals with an average age of 41 years, an average weight of 62 kgm., and an average calcium intake of 0.33 gram per three-day period, there was an average output in the urine of 0.19 gram, in the feces of 0.60 gram, making a total average output of 0.79 gram, and an average negative calcium balance per three-day period of 0.46 gram (1). In the observations about to be described we have studied the quantitative effect of parathormone injection on this negative calcium balance and have as controls both the series summarized above and periods without medication on each individual subject. The methods employed in the preparation of the diet, the collection of the excreta, and the chemical analysis of the material have been described in a previous paper (2).

Case I. This case is presented first because it is in every way characteristic and because it presents the minimum number of

---

complicating factors. The subject was a girl of 16 years whose sole abnormality was otosclerosis of serveral years duration. We have no reason to suppose that otosclerosis affects the action of parathormone, although it may have some quantitative effect on the negative calcium balance while the subject is on a low calcium diet (3). She was studied for twelve three-day periods on a low calcium diet. There was a three-day period on low calcium diet before the investigation was started, and this practice was adhered to in the other investigations reported in this paper. She received parathormone injections from the third to the eighth periods inclusive. In chart Ia and Ib are given the data of the calcium, phosphorus, and nitrogen balances, and of the blood serum calcium and phosphorus.

From the data the following observations of the effect of parathormone upon a subject receiving a low calcium diet are noted:

(i) During parathormone administration there is an increase in the negative calcium balance (in this case 165 per cent).

(ii) Following cessation of parathormone administration, the negative calcium balance is less than in the control periods, suggesting a compensatory mechanism.

(iii) The calcium excretion in the feces is unaffected by parathormone administration.

(iv) During parathormone administration the serum calcium rises. The increased urinary calcium excretion is roughly parallel to this rise.

(v) Both the rise in the urinary calcium on administration of parathormone and the fall in the urinary calcium on cessation of parathormone tend to be gradual. (The rise is unusually abrupt in this case.)

THE PHOSPHORUS METABOLISM

The calcium losses from the body are at the expense of the calcium deposited in the bones. This calcium is largely deposited as tertiary calcium phosphate $(Ca:P::1.93:1)$, but also partly as calcium carbonate, so that the ratio of calcium to phosphorus in bone is approximately $2.23:1$. Therefore, it is of interest to determine whether phosphorus excretion during parathormone injection corresponds to the increased calcium excretion. If such were the case and all other fac-

tors were constant, the ratio of the negative calcium balance to the negative phosphorus balance should be 1.93:1 or 2.23:1, depending on whether parathormone affects only the calcium in calcium phosphate or whether it affects both the calcium in calcium phosphate and the calcium in calcium carbonate. As the difference between these two factors is less than the limit of error of our data, we have arbitrarily chosen the Ca:P factor as 2.23:1. Before a theoretical phosphorus balance thus obtained can be compared with the actual phosphorus balance, a correction has to be made for the phosphorus involved in the deposition or liberation of protein (N:P = 17.4). Therefore, the phosphorus equivalent of the nitrogen balance and the phosphorus equivalent of the calcium balance have been calculated. The sum of these equals the "theoretical phosphorus balance," it being assumed that the phosphorus in muscle, body fluids, etc., would tend to remain in equilibrium. When we later have occasion to use the expression "theoretical phosphorus excretion" we mean the phosphorus intake plus the "theoretical phosphorus balance." It should be noted that since the phosphorus intake is relatively high and since there must be considerable error in its estimation, the determined phosphorus balance is apt to vary from the theoretical by a certain fixed amount. Since the diet is constant throughout the experiment, this error will be constant. In comparing the "theoretical phosphorus balance" with the actual phosphorus balance, the relative changes from period to period are therefore more significant than the actual difference in the two balances in any one period. In order to study these balances, we have constructed chart Ic. Here the actual and theoretical phosphorus balances have been recorded.

From an inspection of the phosphorus data (chart Ib), we have the additional observations:

(vi) The urinary phosphorus is increased by parathormone administration without an effect upon the nitrogen balance.

(vii) The fecal phosphorus is little if at all affected by parathormone injection.

(viii) The serum phosphorus is decreased during parathormone administration.

(ix) As the serum phosphorus falls during parathormone administration, the serum calcium rises.
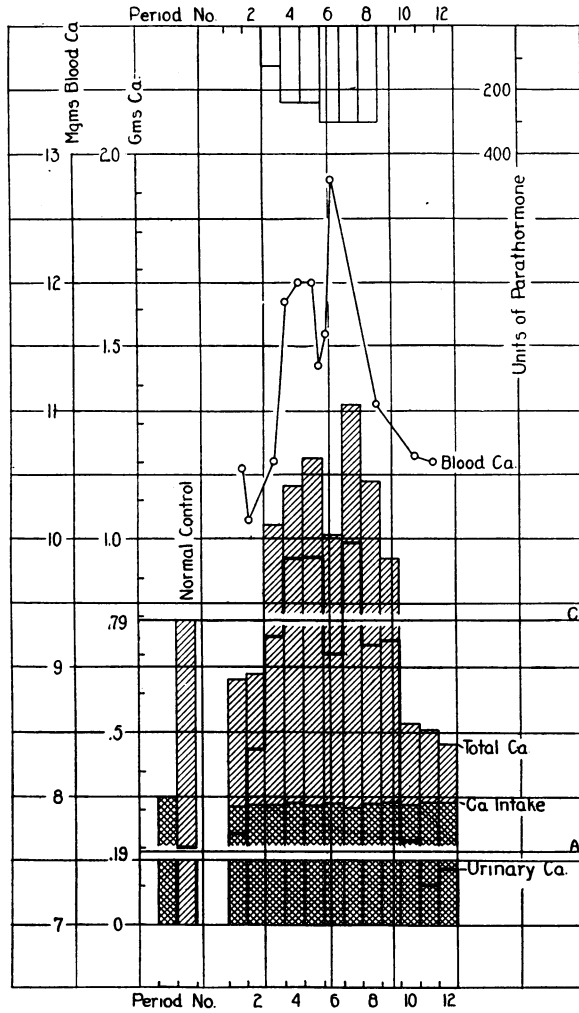
CHART Ia. CALCIUM METABOLISM IN CASE I

Line C represents average total calcium excretion of control series on a low calcium diet. Line A represents average urinary calcium excretion of control series on a low calcium diet (1). These lines have the same significance in later charts.

(x) The urinary phosphorus excretion tends to rise abruptly, often to its highest point during the first period of parathormone administration, and to fall abruptly to the pre-parathormone level in
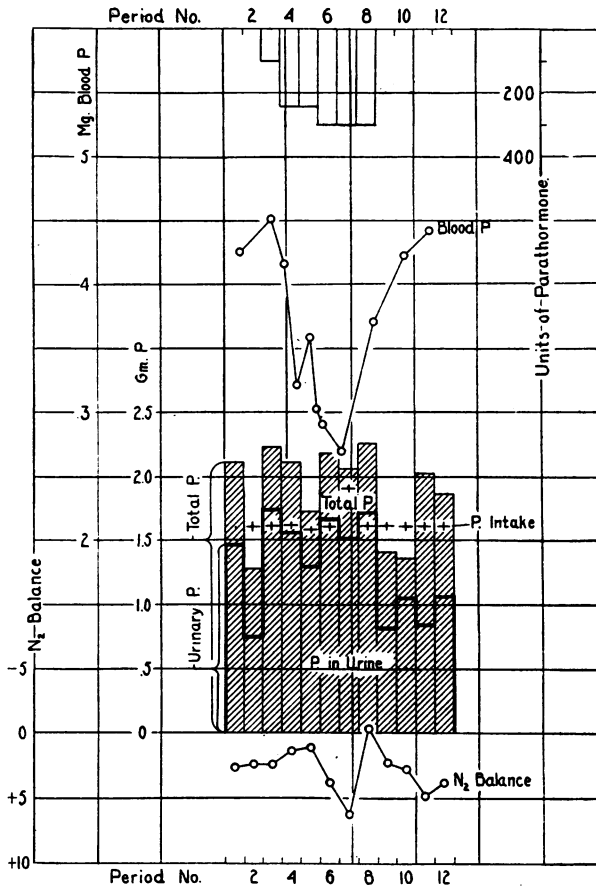

CHART Ib. PHOSPHORUS METABOLISM IN CASE I

the first period following cessation of parathormone.   This is in contrast with the urinary calcium excretion which shows a latent period.

A careful examination of chart Ic shows a relationship between the theoretical and calculated phosphorus balances.   This seems hit or miss on first inspection but will be found constant in all later experi-

ments, namely, that there is *a phosphorus excretion in excess of the theo-
retical amount during the first period or periods of parathormone admin-
istration, and a phosphorus excretion less than the theoretical amount
during the first period or periods following cessation of parathormone.*
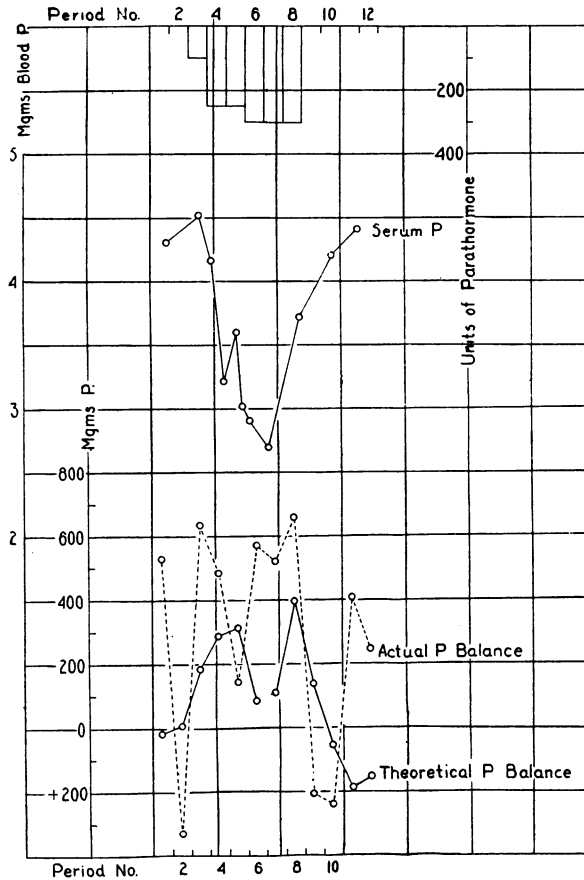

CHART Ic. "THEORETICAL" AND ACTUAL PHOSPHORUS BALANCES IN CASE I

One can predict variations in the urinary calcium excretion for any
given period from chart Ic by the relationship of the theoretical phos-
phorus balance to the calculated phosphorus balance of the preceding
period. Thus periods 3 and 6, where the actual phosphorus excre-

tions rise much more than the theoretical phosphorus excretions, are followed by a rise in the urinary calcium in periods 4 and 7; periods 5 and 9, where the actual phosphorus excretions fall much more than the theoretical phosphorus excretions, are followed by a fall in the urinary calcium in periods 6 and 10.

It will now be necessary to consider the significance of an actual phosphorus excretion in excess of a theoretical phosphorus excretion. One of two explanations seems likely. Possibly when calcium phosphate is mobilized from the bones the phosphorus is excreted more rapidly than the calcium. Evidence against this explanation is found in case VI of this paper. Or possibly it may be due to an excretion of the phosphorus in solution in body fluids, blood, etc. If such is the case one would expect the serum phosphorus to vary with the discrepancy between the actual and theoretical phosphorus excretions. In chart Ic, it will be noted that this appears true. We have then these further observations:

(xi) During the first period of parathormone administration more phosphorus is excreted than can be explained by the calcium and nitrogen balances (theoretical phosphorus excretion).

(xii) During the first period following cessation of parathormone administration, less phosphorus is excreted than can be explained by the calcium and nitrogen balances.

(xiii) During parathormone administration any tendency for the actual phosphorus excretion to rise above the theoretical phosphorus excretion is followed by a tendency for the urinary calcium excretion to rise after a latent period of about 3 days (1 period). The converse is likewise true.

(xiv) A rise in the actual phosphorus excretion above the theoretical phosphorus excretion is apparently partly at the expense of the phosphorus in the body fluids and results in a fall of the serum phosphorus.

From case I we have made certain observations. These are confirmed by the other cases and new observations are added.

Case II. This case is very similar to case I. The subject was a boy of 16 who had symptoms of otosclerosis for about four years. He was studied for thirteen three-day periods on a low calcium diet, then sent home for three months on a normal diet, and finally further studied on a low calcium diet for fifteen more three-day periods. The
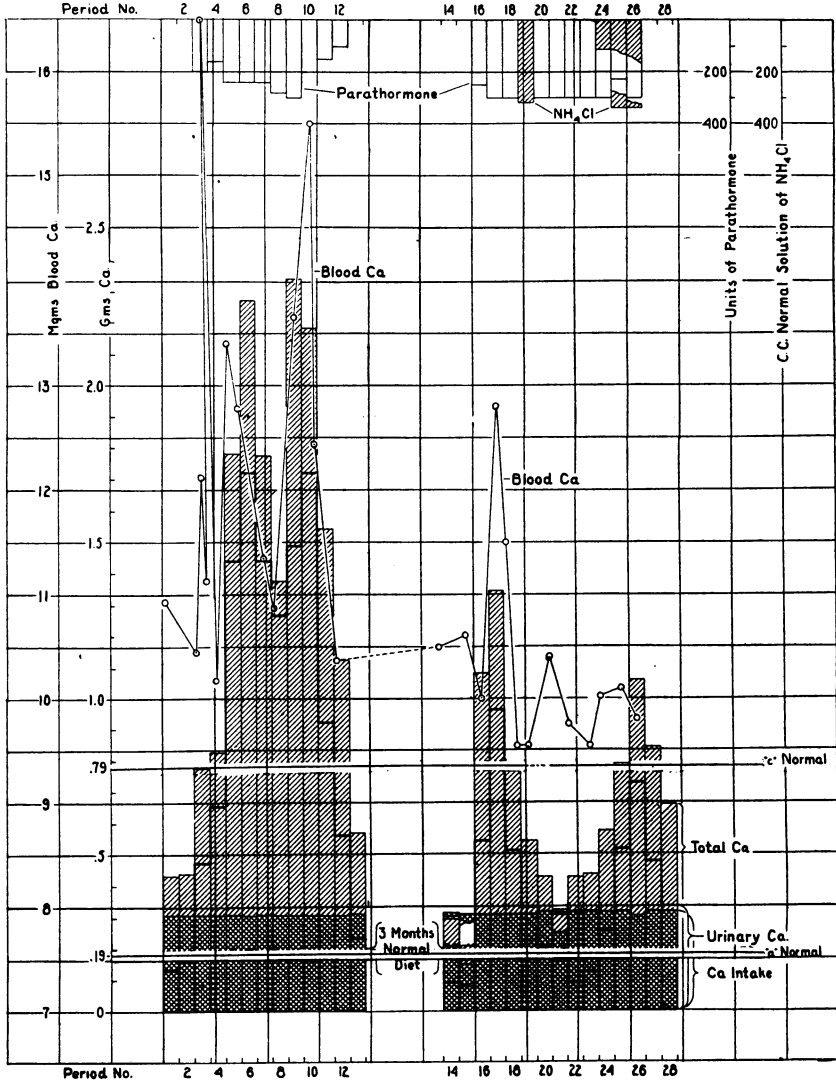
CHART IIa. CALCIUM METABOLISM IN CASE II

medication and the calcium, phosphorus and nitrogen balances are recorded in charts IIa, IIb and IIc.

The calcium data confirm the observations made on case I. The calcium rise (periods 3, 4, and 5) is more step-like than in case I. The negative calcium balance during the control periods (1, 2, 14,
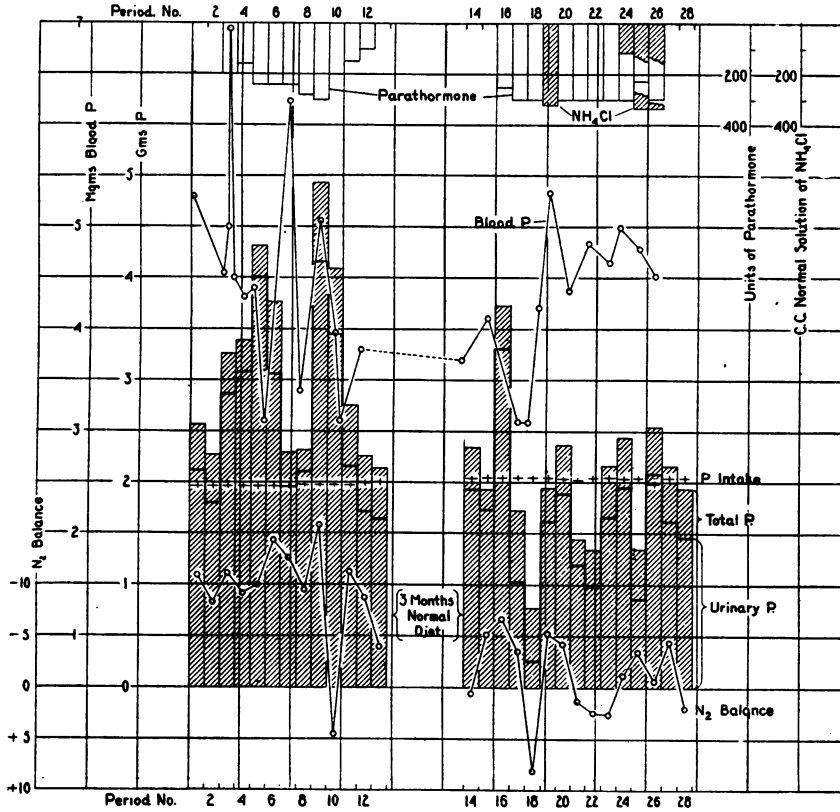
CHART IIb. PHOSPHORUS METABOLISM IN CASE II

and 15) is very low, and actually becomes positive in periods 14 and 15. Whether this is due to the fact that the patient was growing or to the fact that he had otosclerosis has not been determined and does not affect the present discussion. The very high serum calcium in period 3 represents a determination taken in the afternoon and so is not com-
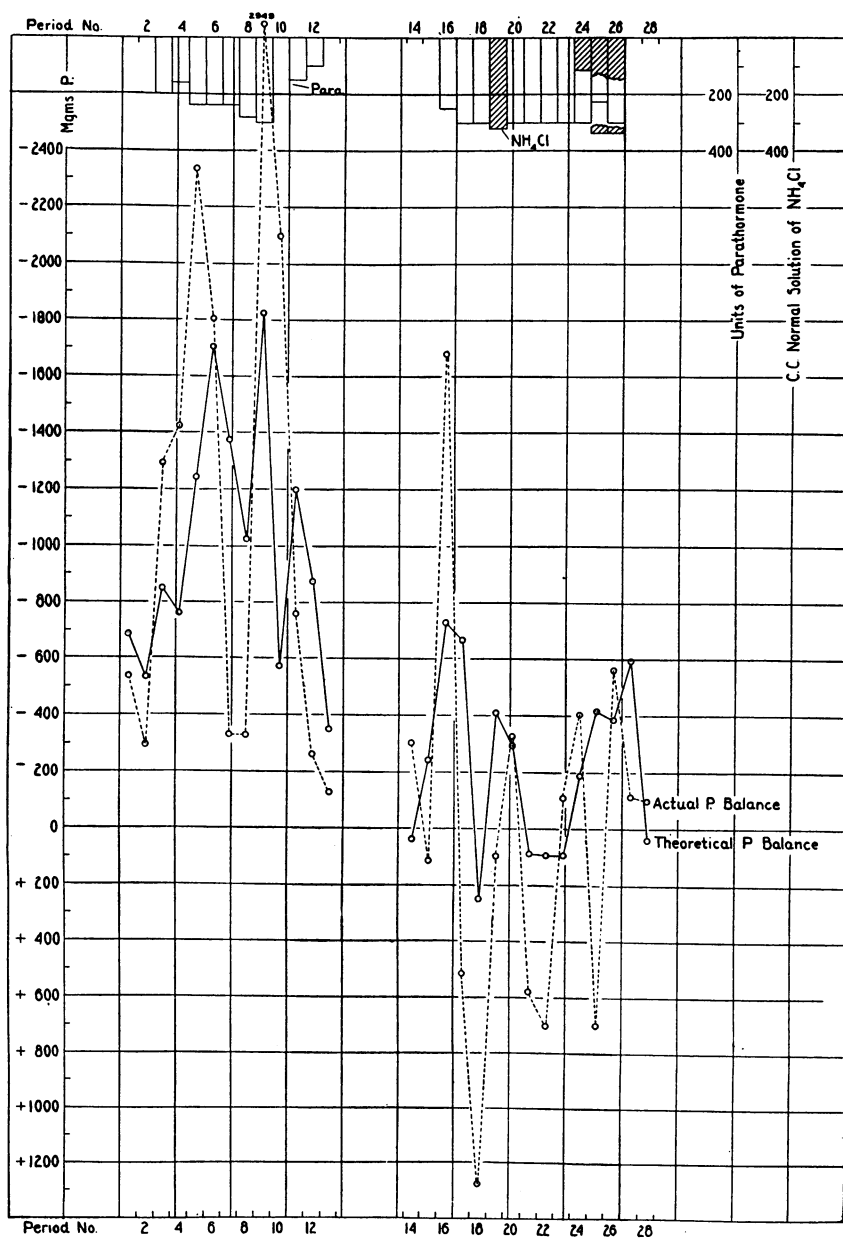
CHART IIc. "THEORETICAL" AND ACTUAL PHOSPHORUS BALANCES IN CASE II

parable with the other calcium determinations which represent morning values.   This is important because the serum calcium level after a dose of parathormone varies over a period of hours, so that in a study like this the injections and the blood examinations should be made at a fixed time each day.   It should be noted that the calcium response to parathormone was much greater in this than in the previous case.   In the second half of the experiment, the response to parathormone, after a satisfactory start in periods 16 and 17, completely wears off in periods 18 to 23.   This is apparently not due to a lack of available calcium as there is a good response to ammonium chloride in periods 24 to 26.   The calcium response to ammonium chloride, like the calcium response to parathormone, is almost entirely urinary.

The phosphorus data (charts IIb and IIc) also corroborate the observations on case I.   Parathormone affected the phosphorus excretion more abruptly than it did the calcium.   The initial rise in the first periods of medication and the rapid return to normal on cessation of the drug is more precipitous in the phosphorus figures.   This is brought out more clearly when comparing the actual phosphorus output with the theoretical figure.   In the first response to parathormone the actual phosphorus excretions exceed the theoretical expectations, and are less than the theoretical expectations when the parathormone response disappears.   Phosphorus excretions above the theoretical appear to be associated with a falling blood phosphorus and to be accompanied in the following period by a rising calcium excretion. During the second admission the same principles prevail, namely, the actual phosphorus excretion exceeds the theoretical phosphorus excretion during the first period of parathormone administration and is accompanied by a falling serum phosphorus; then as the parathormone becomes ineffective the actual phosphorus excretion falls below the theoretical phosphorus excretion and the serum phosphorus rises to an unusually high level.   The effect of the ammonium chloride on the phosphorus excretion is rather inconclusive.   The tendency seems to be for the urinary phosphorus to rise, which was to be expected (4) (11) (see also cases III and VI below).

The additional observations to be noted from case II are:

(xv) The effect of parathormone injection on the urinary calcium

excretion and the blood serum calcium may wear out completely. This has been corroborated by our studies of tetany (5).

(xvi) This impotency of parathormone is apparently not due to an exhaustion of the available supply of calcium, because the calcium excretion can still be increased in a normal manner by ammonium chloride ingestion.

(xvii) The first evidence of a cessation of the effectiveness of parathormone is a fall in the actual phosphorus excretion below the theoretical phosphorus excretion.

(xviii) With ammonium chloride ingestion there is a rise in urinary calcium and phosphorus excretion.

Case III.   In case III, as in case II, we have further opportunity of studying what happens as parathormone injections lose their physiological effect.   The subject was a colored boy of 14 who was suffering from an ossifying hematoma (myositis ossificans) of the right thigh, which had developed as a result of trauma three weeks before admission.   In order to see whether parathormone could mobilize calcium from regions other than the bones, we gave the patient a low calcium diet with parathormone injections.   No metabolism studies were made until the patient had been on this regime for 16 days, though there was obviously a fair response to parathormone as judged from the fact that the blood serum calcium rose to a maximum of 13 mgm. The patient was then studied for 20 three-day periods while still on a low calcium diet.   He was sent home for four months on a normal diet, and finally was studied again for 9 three-day periods on a low calcium diet.   The medication and the calcium, phosphorus, and nitrogen data are given in charts IIIa, IIIb, and IIIc.   During period 2 the patient had an attack of tonsillitis but his temperature remained elevated only 24 hours.   He developed generalized urticaria during period 25, which cleared up promptly when parathormone was discontinued.   All of these efforts to eliminate body calcium produced no change in the density of the ossifying hematoma as demonstrated by x-ray.

An inspection of chart IIIa shows that in spite of increasing doses the effect of parathormone on the serum calcium and urinary calcium excretion was gradually lost.   It is to be noted especially that the calcium excretion falls not only to what it would be without parathor-

mone but even to a much lower level, so that finally a positive balance is obtained in the latter half of the observation.

The second part of this observation follows a rest period of four months. During this time he grew very markedly, and this may be
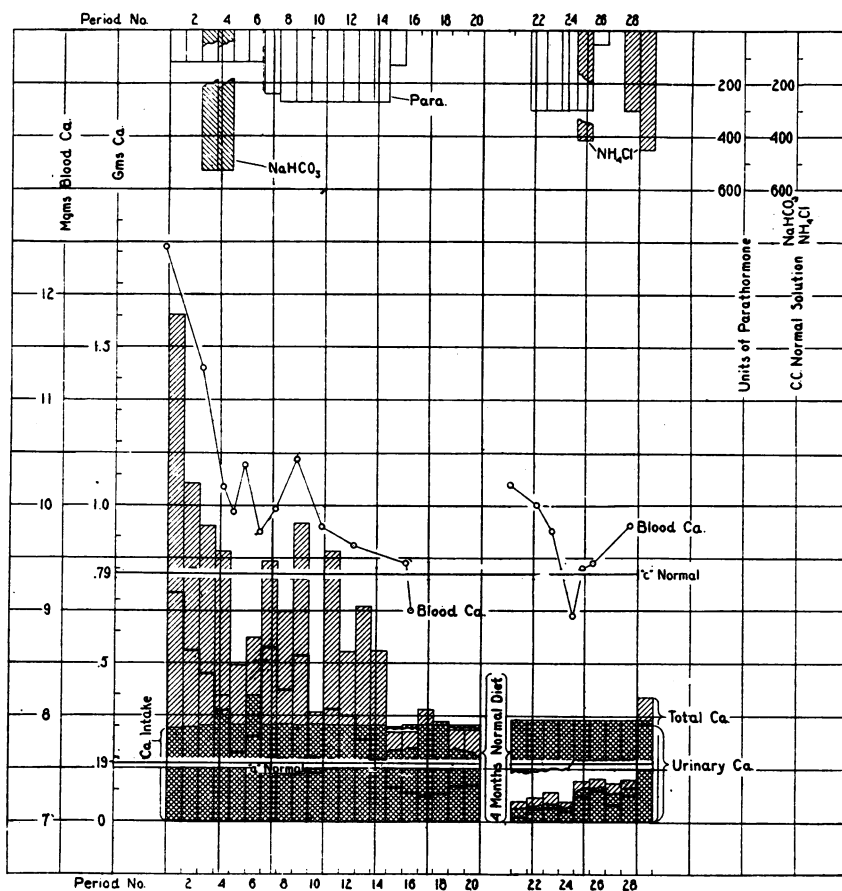

CHART IIIa. CALCIUM METABOLISM IN CASE III

the cause for his having the lowest calcium excretion we have observed in normal individuals on this test diet. In this second observation large doses of parathormone again fail to affect the calcium excretion, and the blood calcium even falls, although ammonium chloride still

proved effective.   Thus the observation is corroborated that mobiliza-
tion of calcium can still be accomplished though parathormone proves
ineffective.

The effect of parathormone on the phosphorus metabolism is
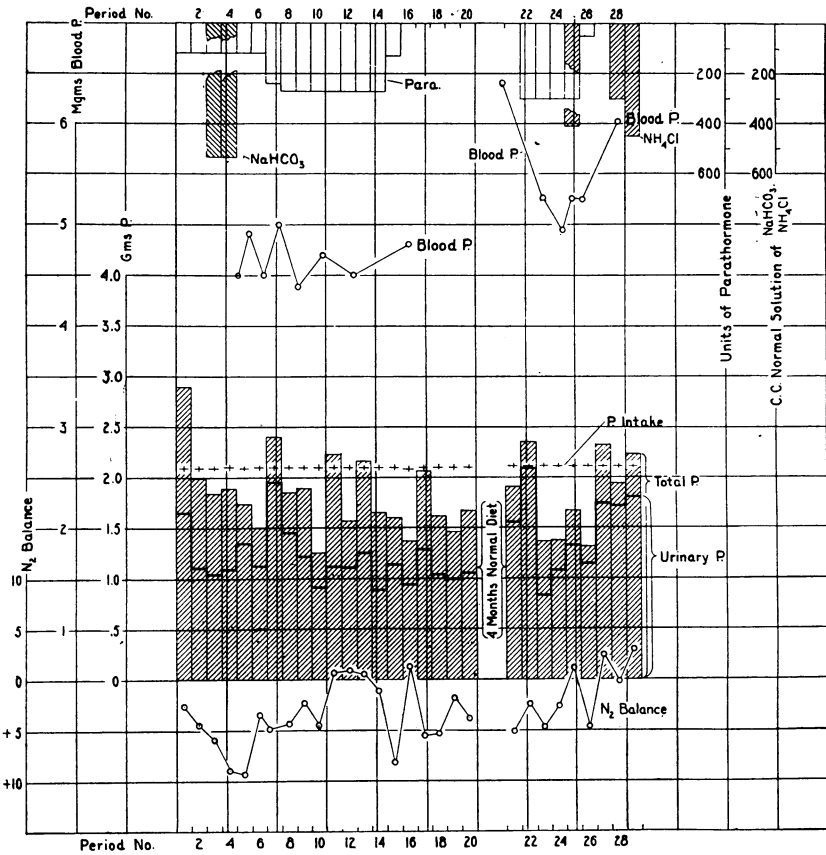similar in kind to that seen in cases I and II.   On his second admis-

CHART IIIb. PHOSPHORUS METABOLISM IN CASE III

sion, however, it is of interest to note the high initial serum phosphorus
level, which may be due to the season of the year (7) and to the fact
that he was growing (7).   As parathormone is administered (period
22), in spite of no effect on the calcium metabolism, there is some rise

in phosphorus excretion, an actual phosphorus excretion in excess of the theoretical phosphorus excretion (chart IIIc), and a corresponding fall of the blood phosphorus.   Thus far, if we judge from the phosphorus data, the parathormone effect has been classical.   But the
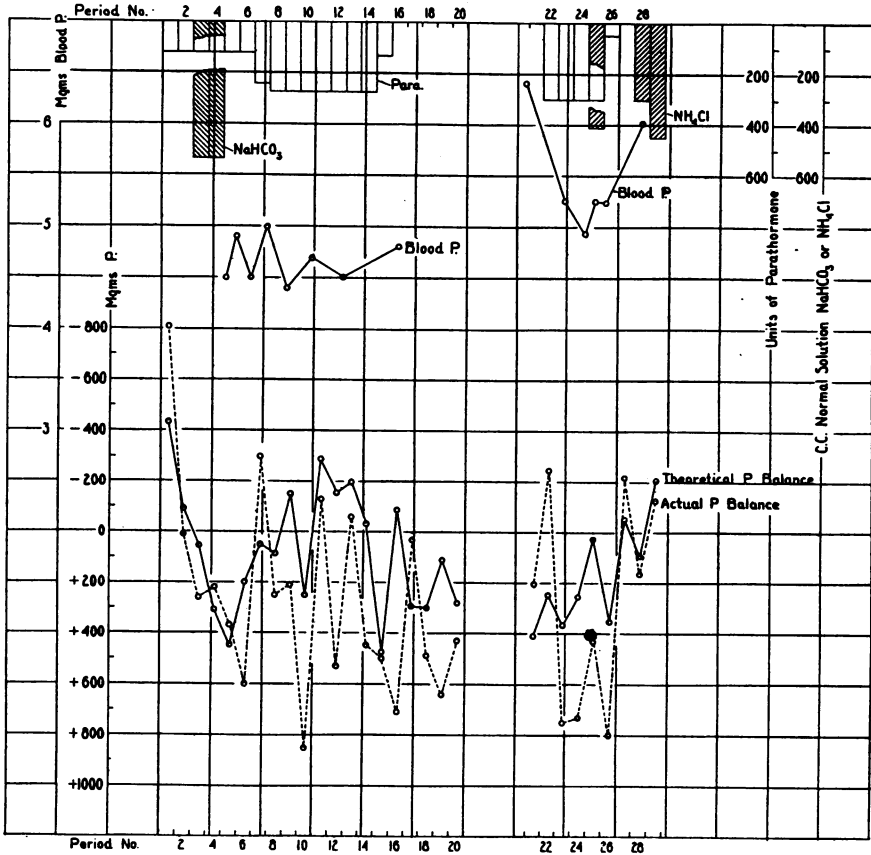

CHART IIIc. "THEORETICAL" AND ACTUAL PHOSPHORUS BALANCES IN CASE III

increase in the actual phosphorus excretion over the theoretical phosphorus excretion in period 22 is not followed by a rise in the calcium excretion.   The increased phosphorus excretion lasts only for one period and then falls just as in period 17 of case II (chart IIb).   It is of interest that the data of Robinson, Huffman and Burt (25) on the

effect of parathyroid extract on normal calves show one case in which a classical phosphorus response was obtained without any concomitant calcium response. As parathormone is discontinued and ammonium chloride is given, the serum phosphorus again rises to the pre-parathormone level (chart IIIb).

From case III we learn:

(xix) In a person who has had parathormone and who has become "immune" to it, the negative calcium balance on a low calcium diet is much reduced or even becomes positive (see also case II).

(xx) The phosphorus response to parathormone administration in a person who fails to show any calcium response is shortlived, but qualitatively normal in this case.

(xxi) Sodium bicarbonate administered to this individual did not effect or decrease the effectiveness of parathormone.

(xxii) Parathormone administration has not been found helpful in decalcifying an ossifying hematoma. (See also case VIII.)

Case IV. The subject of this observation was an Italian laborer of 55 who was suffering from chronic lead poisoning. The effect of parathormone on his lead excretion has been reported as case I by Hunter and Aub (8). We have selected 18 three-day periods on a low calcium diet to report here in more detail. The medication, calcium balance, and blood plasma calcium data are given in chart IV. The nitrogen and phosphorus balances were not determined.

It will be seen that the plasma calcium and the calcium excretion (especially fecal) fall off markedly during the 8 periods (24 days) following parathormone administration. There is then an elevation of the calcium excretion and of the low plasma calcium following ammonium chloride ingestion as shown in the last three periods. The increased calcium excretion appeared largely in the feces.

This experiment confirms observation (ii), that after there has been a marked loss of body calcium produced by parathormone, the negative calcium balance on a low calcium diet is reduced. It also gives us these additional observations:

(xxiii) During calcium starvation over long periods of time, especially if there has been an additional loss of calcium due to periods of parathormone administration, the blood calcium tends to fall (see also case III—periods 21-29 on chart IIIa).

(xxiv) A low blood calcium level due to calcium starvation is raised by ammonium chloride (see also case III).
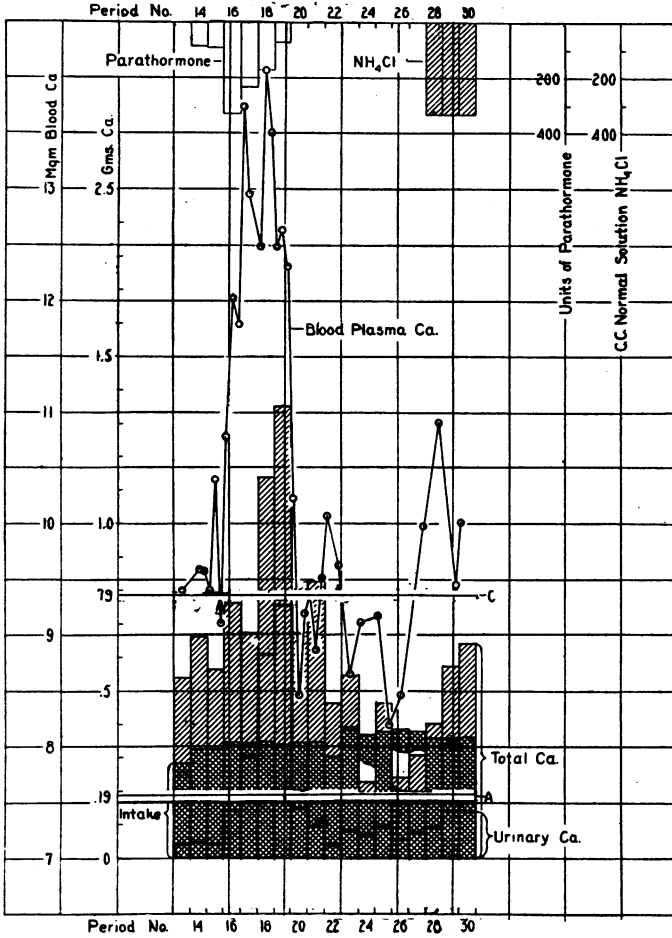

CHART IV. CALCIUM METABOLISM IN CASE IV
The blood values in this case are on blood plasma

Case V. The subject of this investigation was a painter, aged 60, who was suffering from chronic lead poisoning with wrist drop. The results of the studies on lead and calcium excretions during periods

1–36 have been reported by Hunter and Aub (8) as their case IV. We have selected certain of these periods (4–14, 21–23, and 24–36)[4] to report in greater detail and are reporting 19 additional periods on the same patient (5'–24'). The data are shown in charts Va and Vb. It will be noted that periods 4–6 differ from all other periods thus far discussed in that the low calcium in the diet is increased by the addition of calcium lactate.

The most striking part of these data (chart Va) is the extremely high plasma calcium during periods 6, 7, and 8 produced by a dosage of parathormone which was relatively small when compared with that used in previous cases. Furthermore, at these high levels, the urinary calcium excretion does not rise in proportion to the plasma calcium, but remains approximately constant when the plasma calcium rises above 14–15 mgm. (cf. 20 mgm. in period 7). The tendency for the plasma calcium to fall below normal following cessation of parathormone administration and to rise again with ammonium chloride administration is well shown and is confirmatory of observation (xvi) (v. supra). Turning to periods 5'–24' one notes that sodium thiosulphate had no effect on either the calcium or the phosphorus metabolism. This does not concern the present subject but is discussed in a paper dealing with the effect of sodium thiosulphate on lead excretion (9). The response to parathormone in periods 10' and 11' is classical. The phosphorus response in the urine is one full period ahead of the calcium response and greater than can be explained by the calcium-phosphorus ratio of bone. The slightly elevated blood serum phosphorus value on the second day of period 11', corresponding to the calcium value of 14.6, is in disagreement with the observation made in cases I–IV, where it was seen that the serum phosphorus falls when the serum calcium rises. This type of reaction is apt to occur when the calcium value has risen over 14 mgm. and is associated with a sudden diminution in phosphorus excretion. We believe this is entirely a secondary phenomenon dependent on the high serum calcium. It also accounts for certain sudden falls in the actual phosphorus excretion seen in most of the C charts which do not occur when the calcium is kept at a lower level (cf. case VII below). The sodium

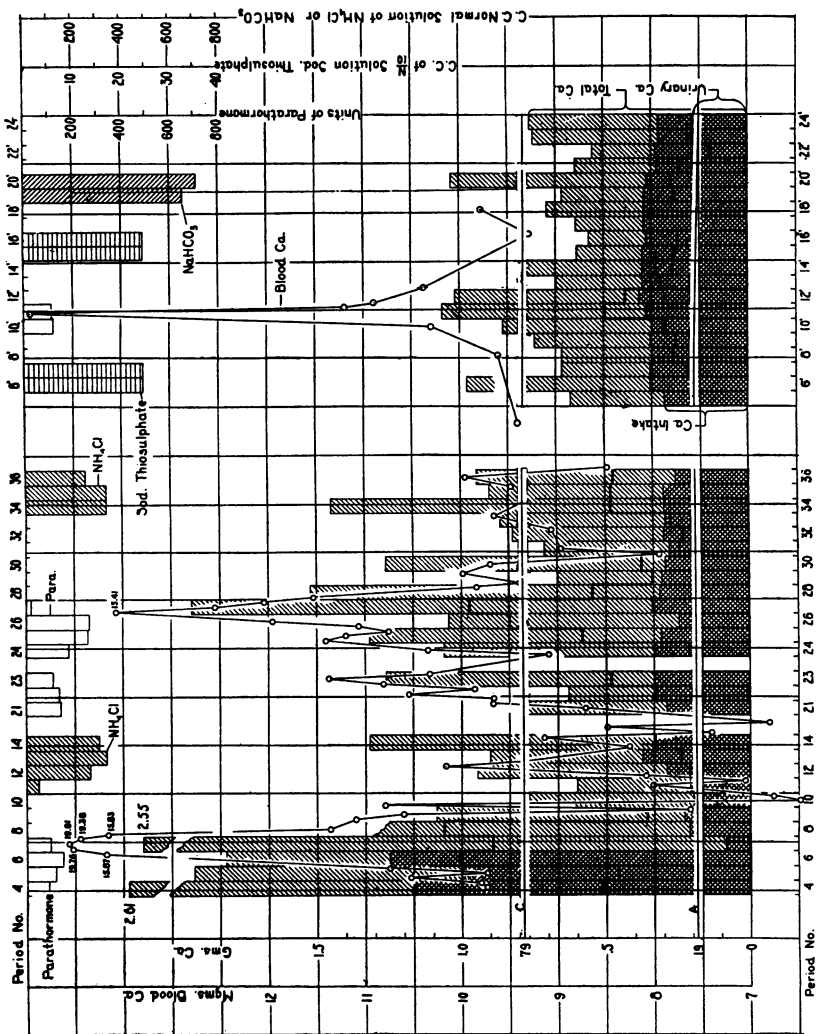[4] The patient left the hospital for one week between periods 23 and 24.

CHART Va. CALCIUM METABOLISM IN CASE V

The blood values during the first admission are on blood plasma and during the second admission on blood serum.

bicarbonate given in periods 19' and 20' increased both the urinary and fecal calcium and phosphorus, but especially the fecal phosphorus.
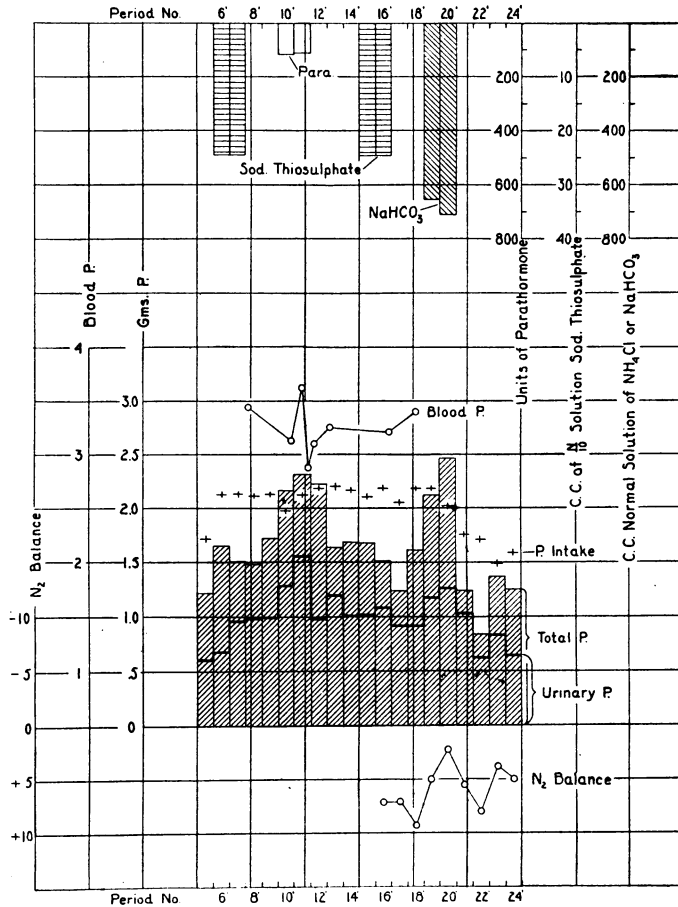

CHART Vb. PHOSPHORUS METABOLISM DURING SECOND ADMISSION ON CASE V

(xxv) Parathormone has more effect on the blood calcium level of this patient than on that of any other in our series. Its effect, therefore, may vary in different individuals.

(xxvi) When parathormone is administered to a patient on a rela-

tively high calcium diet, the serum calcium rises more than if the patient were on a low calcium diet.

(xxvii) When the serum calcium rises above 14–15 mgm., there is not a corresponding increase in the urinary calcium excretion.

(xxviii) When the serum calcium rises above 14–15 mgm., the urinary phosphorus excretion falls and the serum phosphorus rises.

(xxix) Sodium bicarbonate when administered alone causes a slight increase in the calcium and phosphorus excretions in the urine and especially in the feces (single observation—see also conclusion (xxi)). We do not differentiate here between excretion by and lack of absorption from the gastro-intestinal tract.

(xxx) Sodium thiosulphate given intravenously is without effect on the calcium and phosphorus excretions in this case.

Case VI. The subject for this experiment was a college man of 23 who had been afflicted with otosclerosis for ten years. He was studied for 14 periods of 3 days each, and he received parathormone injections for the long consecutive period of 36 days. The data of this experiment are tabulated in charts VIa, VIb and VIc.

The phosphorus response to parathormone is unusually gradual but it still appears to be one period ahead of the calcium response. The excess of the actual phosphorus excretion over the theoretical phosphorus excretion is large (periods 5 and 6—chart VIc) and the response in the urinary calcium is correspondingly large. The tremendous increase in the urinary calcium excretion in periods 10 and 11, when ammonium chloride is given in conjunction with parathormone, is most striking. The total is probably beyond the sum of the increased excretions which would have been obtained had the drugs been administered separately. There was no marked change in the serum calcium as a result of the ammonium chloride. But what is perhaps the most important observation from a theoretical point of view is that the increases in the calcium and phosphorus excretions are coincident (chart VIc). The excretion of the extra calcium does not extend into period 12. Thus when calcium phosphate is pulled from the bones by means of ammonium chloride, the calcium and the phosphorus are excreted in the same period. This is suggestive evidence that the excess phosphorus excretion above the theoretical, which is found at the beginning of parathormone administration, is due to an
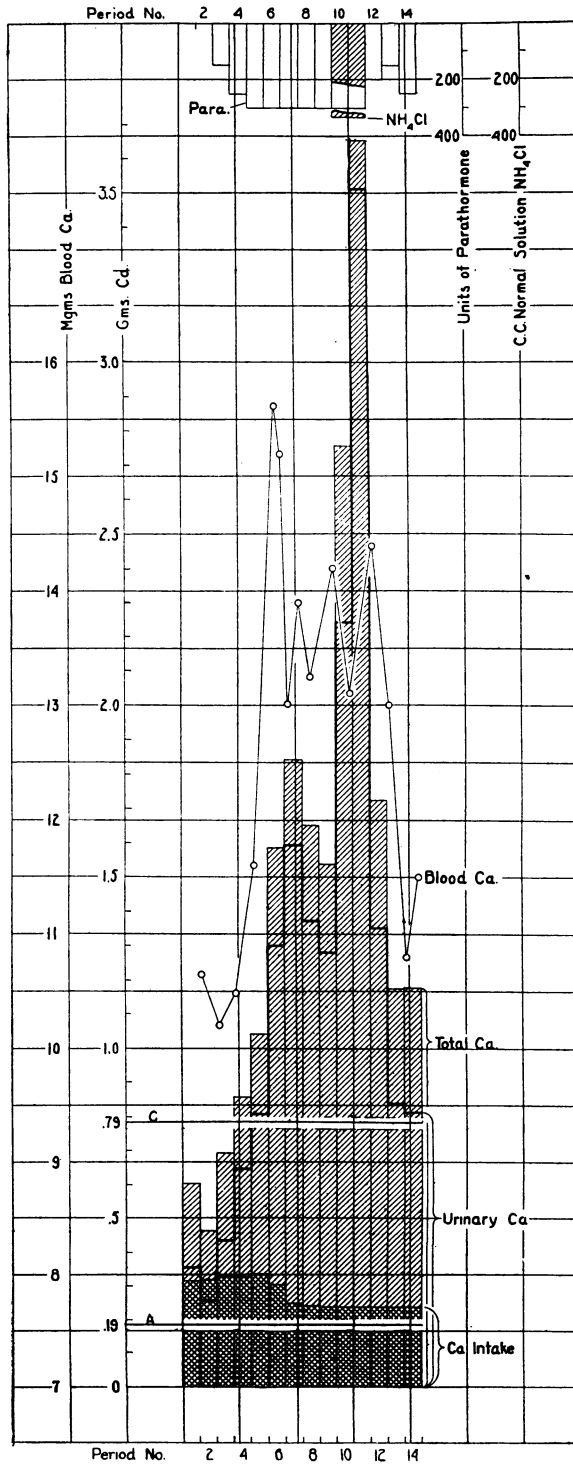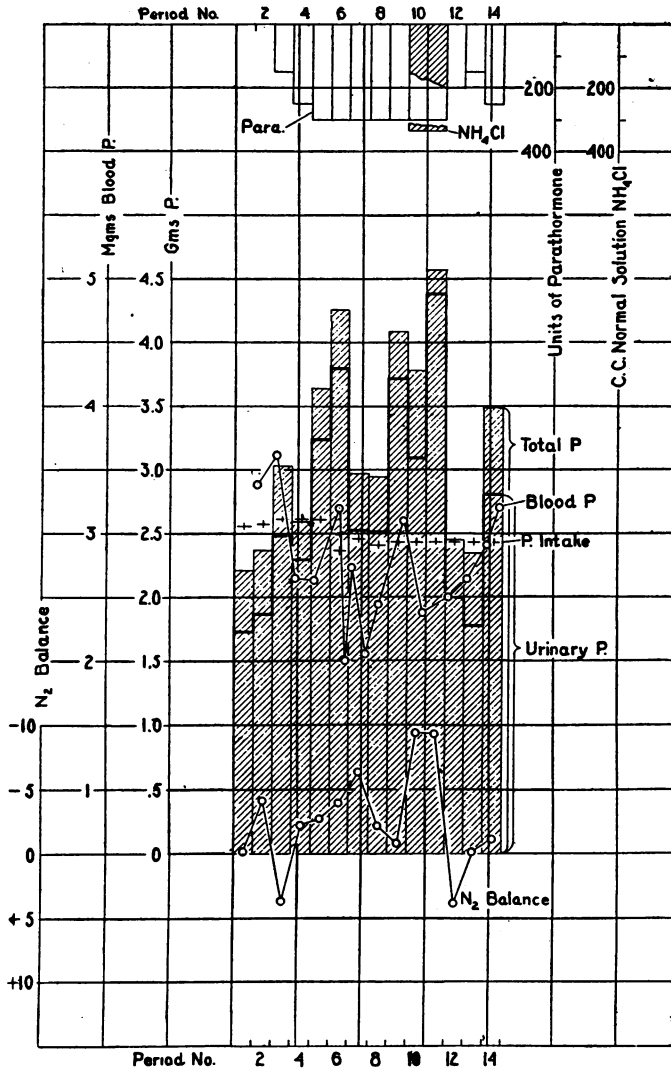
CHART VIa. CALCIUM METABOLISM IN CASE VI

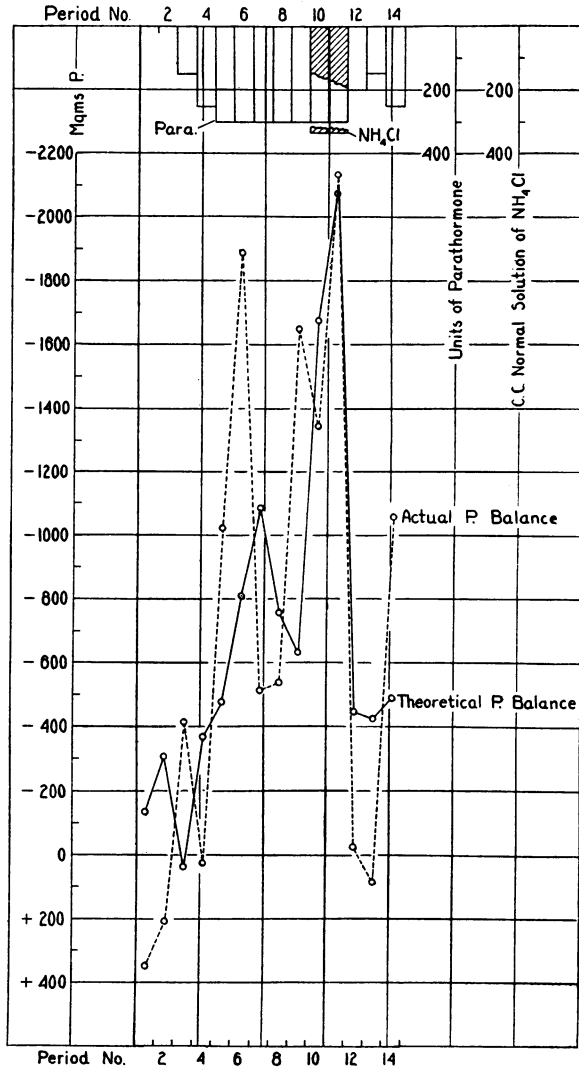CHART VIb. PHOSPHORUS METABOLISM IN CASE VI

CHART VIc. "THEORETICAL" AND ACTUAL PHOSPHORUS BALANCES IN CASE VI

excretion of the phosphorus of body fluids rather than to an easier excretion of phosphorus.   Finally it should be noted that the patient is still responding to parathormone at the end of 36 days of parathormone injection.   The total negative calcium balance during the entire experiment was 17 grams.   The extent of this loss is more obvious when one recalls that a normal fetus at term contains about 30 grams of calcium.[5]   The patient also had an actual phosphorus excretion of 9.926 grams above his intake, which compares favorably with a calculated theoretical figure of 9.639 grams.

From case VI we derive the following new observations:

(xxxi) The effect on calcium excretion of parathormone and ammonium chloride administered simultaneously is probably more than the sum of their individual effects.

(xxxii) Ammonium chloride, administered to a patient who is already receiving parathormone, markedly increases the calcium excretion without any marked effect on the serum calcium.

(xxxiii) The calcium and phosphorus of tertiary calcium phosphate are excreted simultaneously when mobilized from the bones by ammonium chloride ingestion.

(xxxiv) One patient after losing 17 grams of calcium from the body in 42 days still shows a good response to parathormone.

Case VII.   The subject of this investigation was a married woman of 24 who had suffered from otosclerosis for 5 years.   The metabolic data are tabulated in charts VIIa, VIIb, and VIIc.

The low excretion of calcium during the control periods (see also cases I, II, and VI) will be discussed in another paper dealing with the calcium excretion in otosclerosis (3).

This case shows very well what has been suggested by the previous cases, namely that the phosphorus excretion tends to reach its highest limits during the first few periods of parathormone administration. This suggests again that the initial rise in phosphorus excretion is due to an excretion of the phosphorus in body fluids.   The phosphorus excretion then tends to fall to a level corresponding to the phosphorus derived from bones—that is, the "theoretical phosphorus excretion." Following cessation of parathormone the actual phosphorus excretion

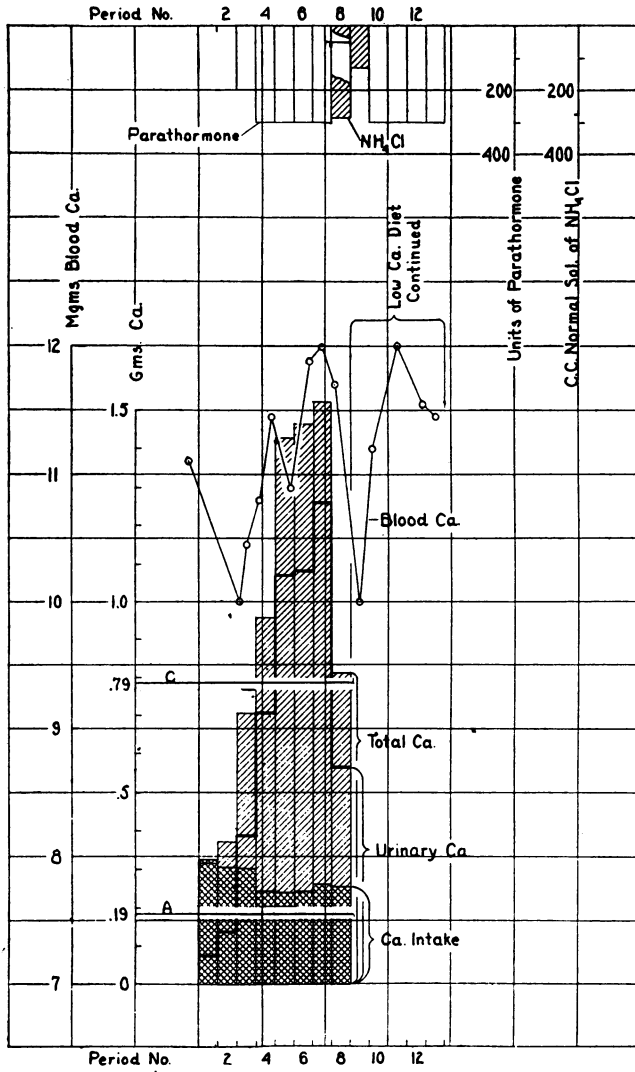[5] Michel quoted by Hamilton (6).

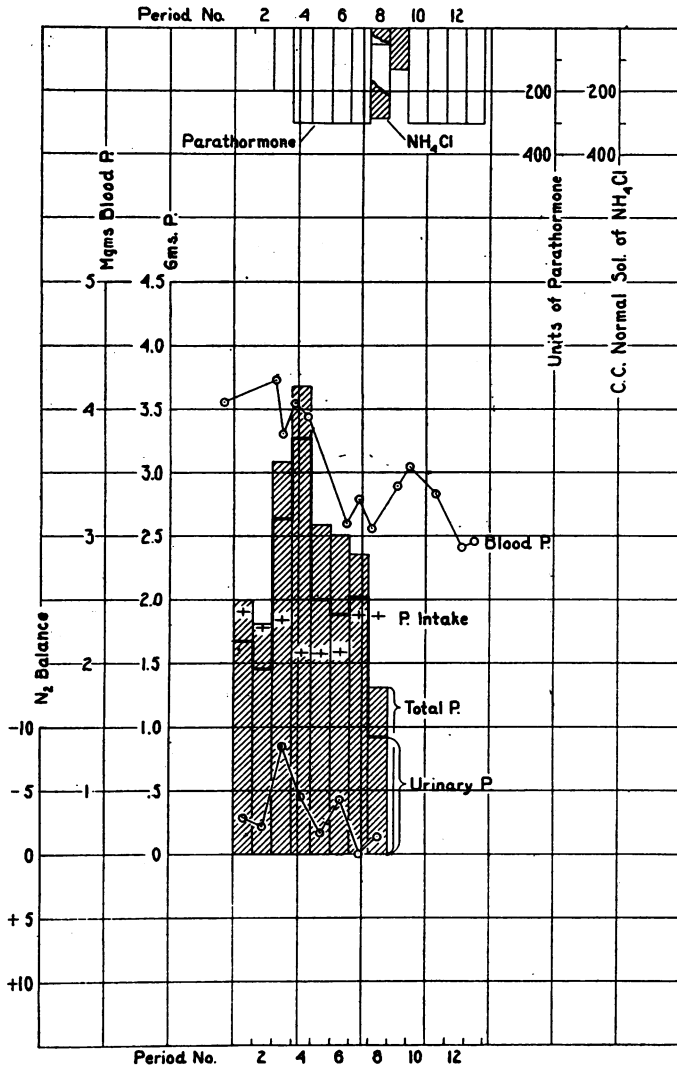CHART VIIa. CALCIUM METABOLISM IN CASE VII

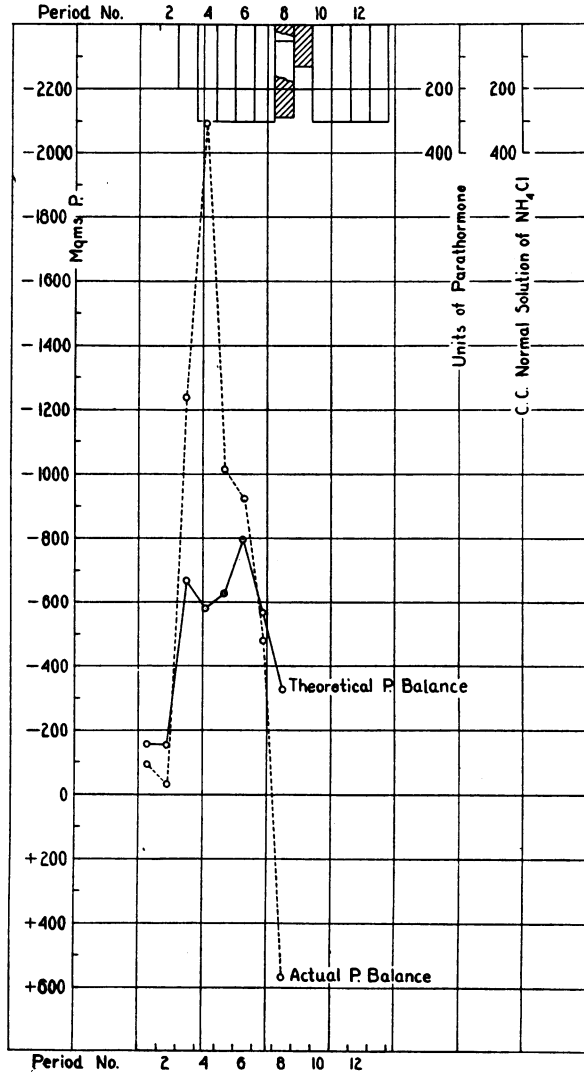CHART VIIb. PHOSPHORUS METABOLISM IN CASE VII

CHART VIIc. "THEORETICAL" AND ACTUAL PHOSPHORUS BALANCES IN
CASE VII

falls below the theoretical phosphorus excretion to about the same degree as it rises above this level at the beginning of parathormone administration.

The new observation to record from this experiment is:

(xxxv) During parathormone administration the phosphorus excretion tends to reach its highest level in the first periods.

Case VIII.   The previous experiments have been confined almost entirely to the study of calcium and phosphorus metabolism during parathormone administration and to a few isolated observations during the ingestion of acids and alkalies.   In view of the conception of bone calcium as a reserve supply of fixed base, elaborated by Gamble (10) (11), it seemed wise to study at least on one patient, the total acid-base changes in the urine, in order to determine whether the increased mobilization of calcium phosphate with parathormone administration is associated with the formation of some acid radicle, as well as to investigate the effects of this drug on inorganic salts other than calcium and phosphorus.

The patient was a laborer of 45 who, like case III, was receiving parathormone in the hope that it would decalcify a large ossified hematoma of three months' duration.   No definite diminished density by x-ray could be demonstrated in the lesion following the parathormone therapy here described.   This result is similar to that in case III.   The patient ate exactly the same diet during the 42 consecutive days of the investigation.   He was also on a constant fluid intake.   The determinations included calcium, phosphorus, and total base analyses on the urine and feces; sulphate, chloride, and ammonia analyses on the urine; and total base, calcium, and phosphorus analyses on the diet.   We also determined what we have called "the titratable acidity minus CO_2."   It was obtained by adding a known amount of $N/10$ hydrochloric acid to the urine, aerating until all the CO_2 was driven off, and then titrating back to a pH of 7.35.   The value was obtained by subtracting the amount of acid used from the amount of alkali used in the titration.   The details of the other methods employed have been discussed in a previous paper (2).   The data for experiment VIII are given in charts VIIIa, VIIIb, VIIIc, VIIId, VIIIe, and VIIIf.
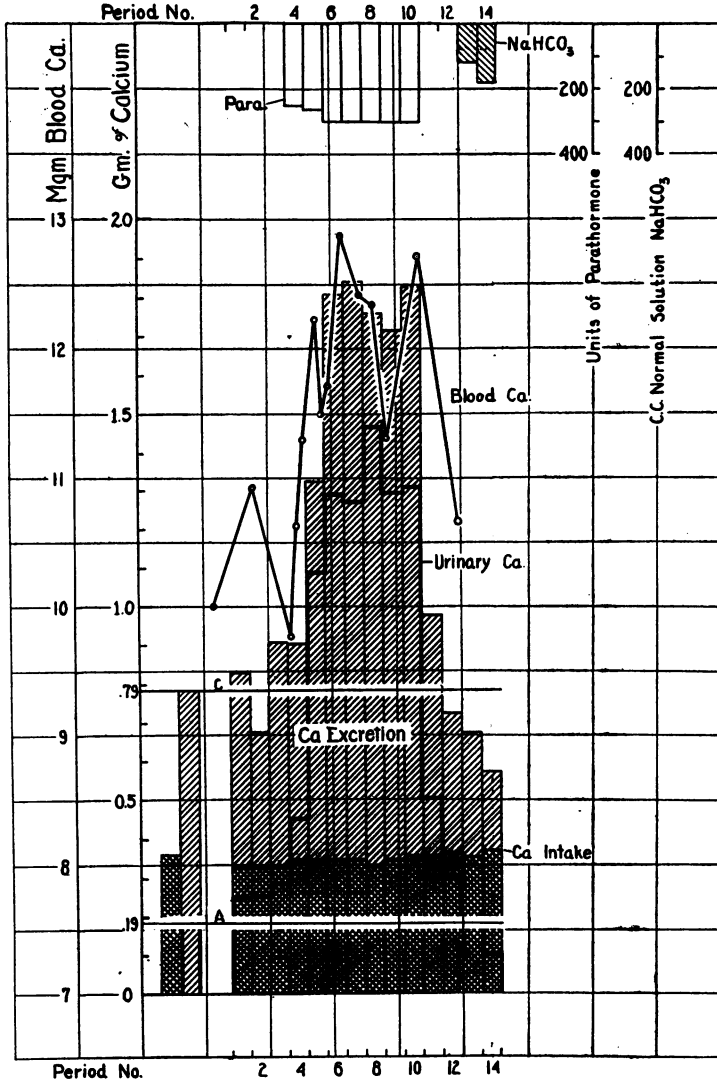
CHART VIIIa. CALCIUM METABOLISM IN CASE VIII

Charts VIIIa, VIIIb, and VIIIc correspond to previous charts except that in charts VIIIb and c the actual phosphorus balance was obtained by using the determined phosphorus intake rather than the estimated phosphorus intake. The determined phosphorus intake
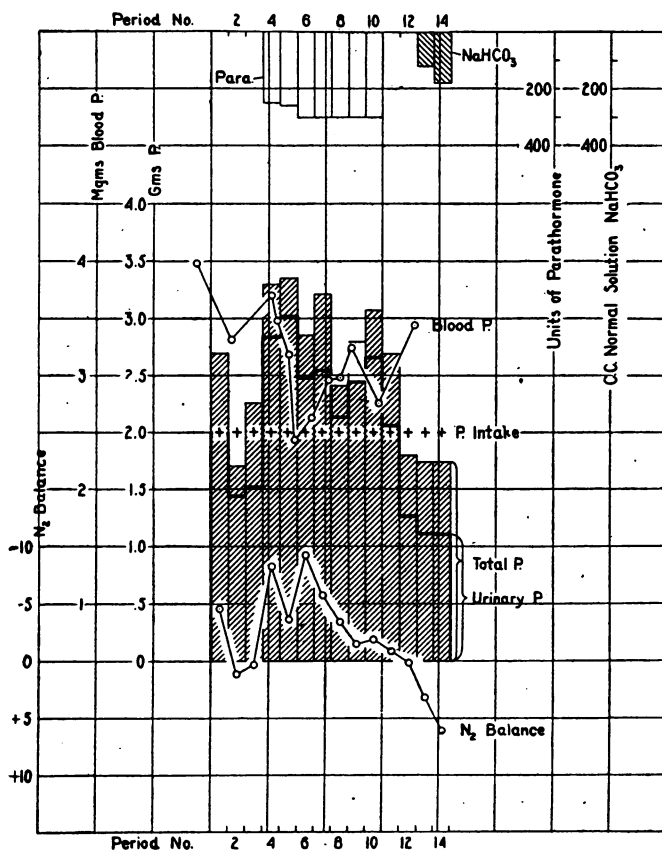

CHART VIIIb. PHOSPHORUS METABOLISM IN CASE VIII

per three-day period was 300 mgm. lower than the estimated phosphorus intake. This gives an indication of the possible error in the estimation of the phosphorus intake in the previous C-charts, and re-emphasizes the fact that the relative changes in the actual and theoretical phosphorus balances from period to period are more significant

than the absolute differences in any one period.   Charts VIIIa, VIIIb, and VIIIc offer further evidence in favor of previous observations but add no new observations.   The sodium bicarbonate seems
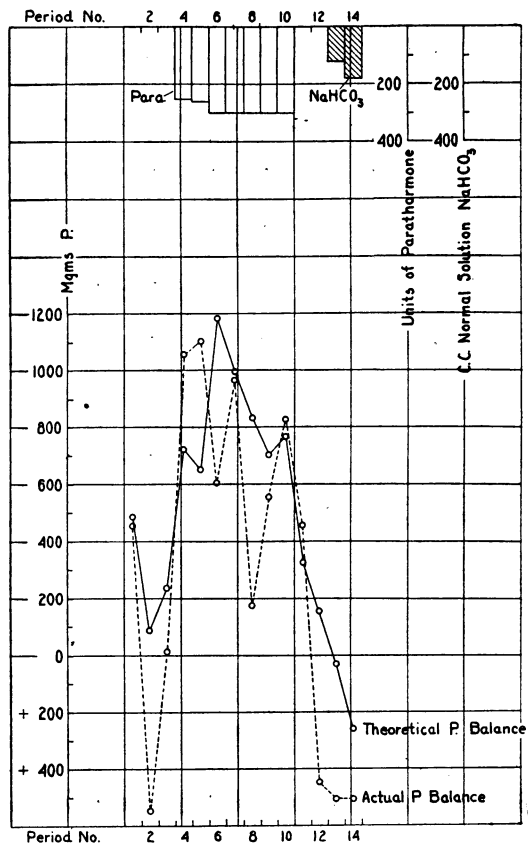

CHART VIIIc. "THEORETICAL" AND ACTUAL PHOSPHORUS BALANCES IN CASE VIII

to have had very little effect on the calcium and phosphorus metabolisms (periods 13–17).

In chart VIIId the effect of parathormone therapy upon the total base balance is recorded.   The experiment checks very well in that the total base ingested during the entire experiment is almost identical

with the total base excreted.   With parathormone one notes a rise in
the urinary excretion of total base, especially periods 3, 4, 8, 9, 10.
In the first period following cessation of parathormone injections
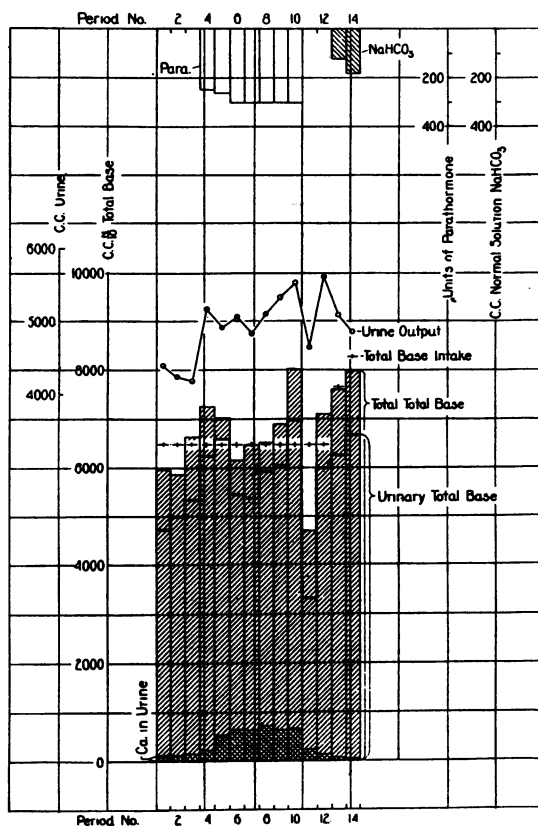
CHART VIIId. TOTAL BASE METABOLISM IN CASE VIII

All values for base are in cubic centimeter of N/10. This chart also shows the
urinary output on a scale chosen such that 1000 cc. of urine is equivalent to 1500
cc. of N/10 base (cf. 1000 cc. of body fluid is equivalent to about 1600 cc. N/10
base).

there was an extreme drop in the total base excretion which suggests
that in this period there was a compensatory retention of total base.
But before we can say that this represents an independent action of

parathormone on total base excretion, certain calculations are neces-
sary.   It seems fair to assume that the theoretical negative total base
balance should equal the total base value of the calcium liberated from
the bones plus the total base in the water held by destroyed muscle
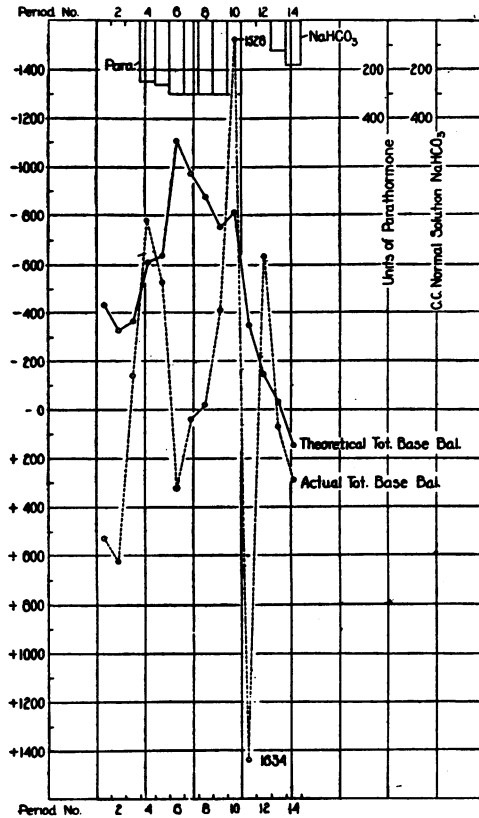
CHART VIIIe. "THEORETICAL" AND ACTUAL TOTAL BASE BALANCE IN
CASE VIII

(10).   Theoretically, any excess excretion of total base beyond this
would be at the expense of the total base in the body fluids.   We have
calculated such a theoretical total base balance and in chart VIIIe
this is plotted against the actual total base balance.   Here, again, as
in the corresponding phosphorus charts, it must be pointed out that
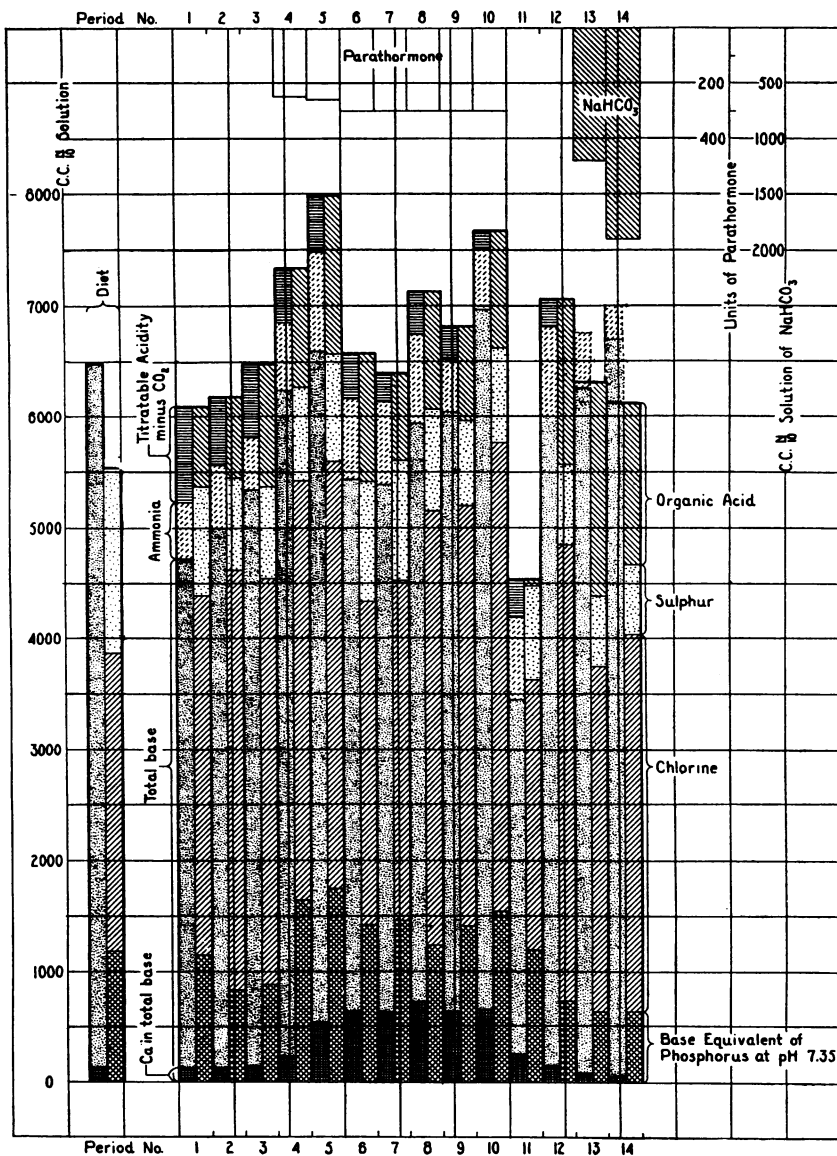
CHART VIIIf. ACID-BASE VALUES OF ANIONS AND CATIONS IN URINE IN CASE VIII

Acid-base properties of diet are represented at left. The chlorine and sulphur of the diet have been estimated from tables and so are only approximate. The total base, calcium and phosphorus of the diet were actually determined.

any small percentage error in determining the total base intake makes a large error in the actual base balance, so that the curve representing this may be incorrect by a uniform amount in all periods.   In calculating the amount of muscle water liberated or stored from the nitrogen balance the following formula has been used:

$$\text{N} \times 29.5 \times 0.76 = \text{muscle water (10)}$$

The first factor provides an estimate of the protoplasm destroyed and the second indicates the corresponding water content.   The total base content of muscle water has been taken to be 180 cc. N/10 per 100 cc. (10).

On examination of chart VIIIe, one notes that the curve of the actual total base balance tends to be below the curve of the theoretical total base balance (448 cc. N/10 by actual determination) due probably to an error in determining the total base intake.   When charts VIIId and VIIIc are compared, it will be noted that the deviations of the actual from the theoretical excretions coincide fairly well.   This leads one to believe that when phosphorus is excreted from the body fluids as a result of parathormone, there is also an excretion of total base held in the body fluids.

Finally chart VIIIf has been made.   Here the cations of the urine have been balanced against the anions in order to see whether any other anion or cation is affected by parathormone.   The principles employed in balancing the electrolytes of the urine are those used by Gamble (10) (11), (12), except that the columns representing the cations and those representing the anions are both shorter by the CO_2 value of the urine than the corresponding ones of this author.   The organic acid value is taken as the difference in the heights of the columns thus established.   In periods 13 and 14 the values for the "titratable acidity minus CO_2" are negative due to the alkaline urine which resulted from sodium bicarbonate ingestion (12).   The values for the intakes are shown on the left of chart VIIIf.   Calcium, total base, and phosphorus were actually determined in the intake.   Sulphur and chlorine were estimated from tables.   The chlorine value in the intake is obviously too low, but this is not surprising as the chlorine content of foods varies considerably.

The deductions to be drawn from chart VIIIf are mostly negative

ones.   The sulphur excretion rises in periods 4, 5, 6, and 7, but so does the negative nitrogen balance.   The chlorine varies considerably and tends to follow the total base.   This is especially obvious in the fluctuation in periods 9, 10, 11, and 12.   Greenwald (13) found a decreased excretion of chloride in the first two days following para-thyroidectomy in dogs.   The urinary ammonia rose slightly and the titratable acidity fell slightly when parathormone was administered. The organic acid values, which of course contain all the errors of the experiment, show no constant changes.

The chief observation made in this case is a negative one, namely that:

(xxxvi) The principle action of parathormone is confined to the calcium and phosphorus metabolisms.

Other observations have not been fully proven but nevertheless are suggested.   These are:

(xxxvii) During parathormone administration, total base, water and chlorine tend to be excreted from the body fluids coincidentally with the excretion of phosphorus from the body fluids and tend to be retained in the body fluids during the periods of phosphorus retention.

(xxxviii) The sulphur, ammonia and titratable acidity of the urine are little affected by parathormone.

<div align="center">DISCUSSION</div>

These observations show quite clearly the metabolic effects of para-thormone on the organism.   It is an active preparation which has an unequal effect in different individuals.   In several of our cases, re-ported here and in other papers, these effects soon wore off, and there-after even large doses had apparently no obvious results.   This is an important factor in its prolonged use for medication.   The most obvious effect of parathormone, as previously reported by others, is in raising the blood calcium level and thereby increasing the urinary calcium excretion.   At first it does this very effectively, but, while it is the only known method for raising appreciably the blood calcium, its effect on the calcium excretion is not as great as that found in thyro-toxicosis (14).   It is unfortunate that the effect of parathormone on both calcium and phosphorus metabolism is gradually lost.   This

immunity to the drug has been found to remain at least a year in a normal individual and in a patient with tetany.

There is evidence among these data that the primary effect of parathormone is on phosphorus excretion rather than on calcium. Thus, when parathormone is administered, the first metabolic changes are a rise in phosphorus excretion and a fall in serum phosphorus; when parathormone is discontinued, the converse is true. At both times the changes in the rate of calcium excretion and in the height of the serum calcium level are more sluggish and lag behind about one period (3 days) (observation (v)). These time relationships are brought out by a study of the "theoretical" and "actual" phosphorus excretions (cf. C-charts). The actual phosphorus excretion is in excess of the calculated at the beginning of parathormone administration, and below, following cessation of parathormone. This implies that the extra phosphorus is drawn from the body fluids. It is also interesting that in cases II and III the wearing out of the parathormone effect was manifested first in the phosphorus metabolism. In the second admission of case III (period 21), although parathormone produced no effect on the calcium metabolism, there was a definite but poorly sustained effect on the serum phosphorus and on the phosphorus excretion. This strongly supports the hypothesis that the primary effect of parathormone is on phosphorus.

The above discussion refers only to the primary effect of parathormone on phosphorus metabolism. It was emphasized first by Collip that when a certain critical level of serum calcium has been reached following parathormone injection, the blood phosphorus rises abruptly. This is probably due to an alteration in kidney function resulting from the high serum calcium, for the non-protein nitrogen in the blood also rises. Thus a late effect of parathormone on phosphorus metabolism is brought about, which is exactly opposite to the primary effect.

There is evidence that the relatively greater excretion of phosphorus over calcium following administration of parathormone cannot be attributed to its more ready excretion through the kidney. The situation is not like the sulphur from protein metabolism which is excreted more rapidly than the nitrogen; for in case VI, when a large amount of $Ca_3(PO_4)_2$ was withdrawn from the bones by ammonium chloride, there was no lag in the excretion of the calcium behind that

of the phosphorus. Furthermore, when thyroid was given to normal controls (14), the increased excretion of calcium and phosphorus ran fairly parallel.

There are several other observations which lend support to the possibility that the primary action of parathormone is on phosphorus. Greenwald (13), who has always emphasized the phosphorus changes, has shown that there is a marked retention of phosphorus in the first two days after parathyroidectomy. The well known deleterious action of a meat diet on parathyroid tetany (15) (16) may be due to its high phosphorus content (17). Collip (18) showed that following parathyroidectomy in the rabbit the serum phosphorus tended to rise very high without there being much change in the serum calcium, and that parathormone, if administered directly following operation, would prevent this rise. In conclusion he stated that, "these observations tend to emphasize the importance of phosphorus in relation to the pathogenesis of tetany." Binger (19) decreased the serum calcium levels in dogs by intravenous injection of phosphates and produced tetany. Salvesen, Hastings and McIntosh (20) likewise produced tetany in dogs by feeding huge amounts of phosphates. This experiment is the exact antithesis to the report of Palmer and Eckles (21) wherein hypercalcemia, hypophosphatemia and osteomalacia in cows resulted from low phosphorus diets. These last two experiments are closely analogous to the situation in hypoparathyroidism and hyperparathyroidism respectively. In the first instance, instead of a decreased phosphorus output, there is an increased phosphorus intake, and in the second, instead of an increased phosphorus output there is a decreased phosphorus intake. The end results are similar. Therefore, the fact that conditions similar to hypoparathyroidism and hyperparathyroidism can be brought about by influencing, through diets, the phosphorus balance, is strong evidence that it is primarily through changes in phosphorus metabolism that the states of hypo- and hyper-parathyroidism are reached.

The observation in case VIII (observation (xxxvii)), that there is a tendency to an excretion of base, water, and chloride from the body fluids along with the loss of phosphorus and calcium, is supported by the recent use of parathormone as a diuretic in nephrosis. Its re-

ported good effects (22) (23) (24) in this disease may be due to an influence on the excretion of base from the tissue fluids.

It is obvious that one effect of parathormone on the calcium-phosphorus metabolism is entirely different from that of acid formers like ammonium chloride or that of the thyroid hormone. Parathormone definitely elevates the blood level of calcium, while the others, which may increase the calcium excretion more than parathormone, do not significantly raise the blood calcium level, unless it is low as in tetany (5).

CONCLUSIONS

The following may be stated as the main conclusions to be derived from this study of the effect of parathormone on patients maintained on a constant diet containing an inadequate amount of calcium.

1. Parathormone administration *gradually* increases the urinary calcium excretion without affecting the fecal excretion. Following cessation of parathormone administration, the urinary calcium excretion *gradually* falls to a level below that found before the administration.

2. The effect on calcium excretion of parathormone and ammonium chloride administered simultaneously is probably more than the sum of their individual effects.

3. The calcium level in the blood is markedly but *gradually* elevated by parathormone injections. The extent of this elevation varies in different individuals, and is more marked when the patient is on a high calcium diet.

4. Parathormone administration *abruptly* increases the urinary phosphorus excretion without affecting the fecal excretion. Following cessation of parathormone administration, the urinary phosphorus excretion *rapidly* falls to a level below that found before the administration. These changes are more rapid than those produced in the calcium metabolism, and greater than can be explained by a theoretical calculation of the phosphorus liberated with calcium and nitrogen.

5. The phosphorus level in the blood is primarily lowered by parathormone. If, however, the serum calcium rises above a critical level of about 14 to 15 mgm., then the urinary phosphorus excretion falls and the blood phosphorus rises.

6. These observations on the effect of parathormone suggest that an increased phosphorus excretion is the primary effect. The excreted phosphorus is partly derived from the body fluids.

7. The effect of parathormone in some cases gradually wears off and the first evidence of this is found in a decreased phosphorus elimination. That this lack of response to parathormone is not due to an exhaustion of calcium reserves, is shown by the fact that ammonium chloride ingestion can still produce an increased elimination in such cases.

8. Nitrogen excretion is not affected by parathormone.

9. Parathormone has not helped to decalcify two ossifying hematomas.

10. The effect of parathormone on other electrolytes is discussed, but no conclusions are formulated.

BIBLIOGRAPHY

1. Bauer, W., Albright, F., and Aub, J. C., J. Clin. Invest., 1929, vii, 75. Studies of Calcium and Phosphorus Metabolism. II. The Calcium Excretion of Normal Individuals on a Low Calcium Diet.
2. Bauer, W. and Aub, J. C., J. Am. Diet. Assoc., 1927, iii, 106. Studies of Inorganic Salt Metabolism. I. The Ward Routine and Methods.
3. Aub, J. C., Albright, F., Bauer, W., Salter, W., and Crockett, F., Studies in Calcium and Phosphorus Metabolism. The Calcium Metabolism in Otosclerosis with Observations on the Therapeutic Value of Decalcifying Agents. (To be published.)
4. Fitz, R., Alsberg, C. L., and Henderson, L. J., Am. J. Physiol., 1907, xviii, 113. Concerning the Excretion of Phosphoric Acid During Experimental Acidosis in Rabbits.
5. Bauer, W., Albright, F., Rossmeisl, E., and Aub, J. C., Studies of Calcium and Phosphorus Metabolism. V. The Calcium and Phosphorus Metabolism in Tetany with Observations on the Therapeutic Action of Thyroid and Ammonium Chloride Medication. (To be published.)
6. Hamilton, Bengt, Acta Pediatrica, 1922, ii, 1. The Calcium and Phosphorus Metabolism of Prematurely Born Infants.
7. Hess, A. F., and Lundagen, M. A., J. Am. Med. Assoc., 1922, lxxix, 2210. A Seasonal Tide of Blood Phosphates in Infants.
8. Hunter, D., and Aub, J. C., Quart. J. Med., 1927, xx, 123. Lead Studies. XV. The Effect of the Parathyroid Hormone on the Excretion of Lead and of Calcium in Patients Suffering from Lead Poisoning.

9. Ropes, M., and Aub, J. C., Lead Studies. The Effect of Sodium Thiosulphate on Lead, Calcium, and Phosphorus Excretion. (To be published.)

10. Gamble, J. L., Ross, G. S., and Tisdall, F. F., J. Biol. Chem., 1923, lvii, 633. Metabolism of Fixed Base During Fasting.

11. Gamble, J. L., and Ross, G. S., Am. J. Dis. Child., 1923, xxv, 470. Studies of Tetany. II. Effect of Ingestion of Hydrochloric Acid Producing Substances on the Acid-base Metabolism of an Infant and the Probable Manner of their Action in the Treatment of Tetany.

12. Gamble, J. L., J. Biol. Chem., 1922, li, 295. Carbonic Acid and Bicarbonate in Urine.

13. Greenwald, I., and Gross, J., J. Biol. Chem., 1926, lxviii, 325. The Effect of Long Continued Administration of Parathyroid Extract upon the Excretion of Phosphorus and Calcium. Greenwald, I., and Gross, J., J. Biol. Chem., 1925, lxvi, 185. The Effect of Thyroparathyroidectomy in Dogs Upon the Excretion of Calcium, Phosphorus, and Magnesium. Greenwald, I., Am. J. Physiol., 1911, xxviii, 103. The Effect of Parathyroidectomy Upon Metabolism. I. Introduction.

14. Aub, J. C., Bauer, W., Heath, C., and Ropes, M., J. Clin. Invest., 1929, vii, 97. Studies of Calcium and Phosphorus Metabolism. III. The Effect of the Thyroid Hormone and Thyroid Diseases.

15. Salvesen, H. A., Acta med. Scandinav., 1923, Supplement 6. Studies on the Physiology of the Parathyroids.

16. Dragstedt, L. R., and Peacock, S. C., Am. J. Physiol., 1923, lxiv, 424. Studies on the Pathogenesis of Tetany. I. The Control and Cure of Parathyroid Tetany by Diet.

17. Greenwald, I., J. Biol. Chem., 1926, lxvii, 1. The Effect of the Administration of Calcium Salts and of Sodium Phosphate Upon the Calcium and Phosphorus Metabolism of Thyro-parathyroidectomized Dogs with a Consideration of the Nature of the Calcium Compounds of Blood and their Relation to the Pathogenesis of Tetany.

18. Collip, J. B., Medicine, 1926, v, 1. The Parathyroid Glands.

19. Binger, Carl, J. Pharm. and Exper. Therap., 1917, x, 105. Toxicity of Phosphates in Relation to Blood Calcium and Tetany.

20. Salvesen, H. A., Hastings, A. B., and McIntosh, J. F., J. Biol. Chem., 1924, lx, 311. Blood Changes and Clinical Symptoms Following Oral Administration of Phosphates.

21. Palmer, L. S., and Eckles, C. H., Proc. Soc. Exper. Biol. and Med., 1927, xxiv, 307. Effect of Phosphorus Deficient Rations on Blood Composition in Cattle.

22. Davidson, J. R., Canad. Med. Assoc. J., 1925, xv, 803. A Case of Adolescent Myxedema Accompanied by Nephrosis and by Tetany of Parathyroid Origin Treated with Thyroid and Collip's Parathyroid Extract.

23. Mason, E. H., Canad. Med. Assoc. J., 1926, xvi, 538.   A Case of Chronic Nephritis Treated with Collip's Parathyroid Extract.
24. McCann, W. S., J. Am. Med. Assoc., 1928, xc, 249.   Diuretic Action of Parathyroid Extract—Collip, in Certain Edematous Patients.
25. Robinson, C. S., Huffman, C. F., and Burt, K. L., J. Biol. Chem., 1927, lxxviii, 477.   The Effect of Parathyroid Extract on Normal Calves.