THE EFFECT OF EXPERIMENTAL REDUCTION OF KIDNEY SUBSTANCE UPON THE PARATHYROID GLANDS AND SKELETAL TISSUE

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PLATES 51 TO 54

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There are many established facts pointing to a relationship between kidneys and parathyroids.

1. MacCallum (1), Bergstrand (2), Hubbard and Wentworth (3), Pollack and Siegal (4), Magnus and Scott (5), have demonstrated the occurrence of parathyroid hyperplasia in individual cases of chronic nephritis. Pappenheimer and Wilens (6) have shown that such enlargement occurs regularly, and is proportional to the degree of renal damage.

2. Renal lesions occur in approximately 50 per cent of the cases of hyperparathyroidism with osteitis fibrosa (Albright, Baird, Cope and Bloomberg (7)). These are associated with formation of calculi (Type I) or with a diffuse calcinosis (Type II). The calcium content of the kidneys may be greatly elevated by injection of parathormone in suitable doses (Molinari-Tosatti (8), Olsen (9), Spingarn (10)).

3. In the few cases of renal rickets in which the parathyroids have been studied, they have been found to be enlarged (Langmead and Orr (11), Smyth and Goldman (12), Schelling and Remsen (13)). The last authors, using the technique of Hamilton and Schwartz (14), were able to demonstrate an increased amount of parathyroid hormone in the blood during life.

4. Indirect evidence of a relationship between parathyroid and renal function is seen in the facts that (a) injection of parathormone is followed almost immediately by an increased urinary excretion of
phosphate (Greenwald and Gross (15), Albright and Ellsworth (16), Albright, Bauer, Roper and Aub (17)); (b) that this does not occur in nephritics in whom the excreting power of the kidneys is impaired (Goadby and Stacey (18)); (c) that after complete ablation of kidneys in dogs, injection of parathormone fails to cause a significant rise of calcium in the blood above that of untreated nephrectomized controls. The mobilization of the calcium store of the body by parathyroid hormone thus depends on kidney function (Tweedy, Templeton, McJunkin (19)); (d) that there is increase in the phosphatase activity of the kidney after parathyroidectomy, and decrease after parathormone injections (Pisa (20)). We shall not attempt to discuss the theoretical implications of these several observations. They have been cited to show that there is considerable evidence of an interrelation between parathyroids and kidneys, and need of further experimentation on the subject.

The concrete questions which we shall attempt to answer in this paper are these:

1. Does experimental reduction of kidney tissue lead to significant hyperplasia of the parathyroid glands?
2. If so, is this increased functional activity reflected in alterations of the osseous tissue, and is it possible to produce experimentally in rats a picture analogous to renal rickets in man?

EXPERIMENTAL

In the first series of experiments, healthy white rats weighing from 150 to 250 gm. were used. They were kept on a mixed diet. One kidney was removed through a lumbar incision, and a considerable part of the opposite kidney was destroyed by thermocautery in two subsequent operations. All operations were performed under ether anesthesia.

The animals were allowed to live for varying periods of time; when they were killed or died spontaneously, a litter mate unoperated control was killed for comparison.

The volume of the parathyroids was determined in the following way. The glands, with attached thyroid tissue, were fixed in Zenker's fluid, and serially

2 A commercial chicken mash, containing cereals, alfalfa, meal, meat scraps, skimmed milk, cod liver meal, sardine oil, etc. Rats on this diet show excellent growth and fertility.
3 All operations were performed under ether anesthesia.
sectioned at 10μ. Their volume was calculated by multiplying the combined areas of the sections, as obtained with the planimeter from drawings projected at known magnification, by the thickness of the section. The volume was calculated for 100 gm. of weight. Since many of the partially nephrectomized rats lost weight before death, the calculations were based upon the normal weight at time of death, as estimated from Donaldson's tables, and not upon the actual weight.

In addition to the parathyroids, sections were prepared from kidneys and bones, the latter decalcified by prolonged fixation in Mueller's fluid.

After a preliminary period of apparent health, the majority of the rats after the second cauterization showed a stationary or declining weight curve, drowsiness, roughness of the hair and loss of appetite.

The operated rats fall into two groups according to the time of survival. Those allowed to live for a period of 113 to 124 days after the second cauterization showed much more intense lesions of the remaining kidney tissue than did those in the group killed or dying before 46 days. The data are summarized in Table I.

TABLE I

<table>
<thead>
<tr>
<th>Average Combined Volume of Parathyroids per 100 Gm. of Rat</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group A—Controls (9) .................................. 0.1441 0.0130 0.0551</td>
</tr>
<tr>
<td>Group B*—Early nephritics (5) .......................... 0.1679 0.0066 0.0197</td>
</tr>
<tr>
<td>Group C*—Late nephritics (5) .......................... 0.4117 0.0553 0.1659</td>
</tr>
</tbody>
</table>

* Calculations of B and C are based on estimated normal weights at time of death.

Comparing the combined volume of the parathyroids per 100 gm. of rat in the operated and control animals, we find, in spite of the small numbers of animals, a decided increase in parathyroid volume in the partially nephrectomized rats as compared with the controls. In the group which survived a longer period and showed more intense renal lesions, the difference is statistically valid; in the earlier group it is suggestive but not conclusive, although in each rat the volume of the gland exceeded the mean of the control series.

One may reasonably conclude—and this will be supported by subsequent experiment—that the reduction of functional renal tissue has led to a decided increase in the size of the parathyroids.

The question arises as to whether this increase is to be ascribed to enlargement of the cells, or to their multiplication. Measurements
of the nuclei in two diameters indicated that the nuclei in the glands of operated rats were larger than those of the controls. Thus the mean diameters of 250 nuclei in the 5 nephritic rats were 8.06 x 6.2 µ as against 6.8 x 4.4 µ in normal nuclei. It could also be demonstrated that the increase in nuclear size was attended by an increase in cytoplasmic volume. Mitotic figures were rarely found. It would seem that the increase in the total bulk of the gland may be accounted for in part, at least, by the increased volume of both nucleus and cytoplasm.

The pathologic changes which develop in the remaining kidney substance in the later group C, are both diffuse and severe, simulating an advanced stage of glomerulonephritis (Fig. 1). At least 90 per cent of the glomeruli are greatly enlarged, bloodless, the capillary loops distended with hyaline and granular material, the capsular space obliterated by adhesions (Fig. 2). Often there is a crescentic proliferation of epithelial cells. In many tufts tissue changes progress to complete obliteration.

The majority of the tubules are greatly dilated, and their lumina filled with dense hyaline coagulum. The epithelial cells are flattened, so that in some areas the tissue resembles thyroid. There is irregular interstitial fibrosis with moderate lymphoid infiltration of the stroma. In a few of the larger arteries and in some of the arterioles there is fibrinoid or hyaline material in the subendothelial tissues.

With minor variations in intensity, the same picture is found in all 5 animals of the later group. Although the pathogenesis of the lesions is not clear, it is of interest that the cauterization leads to the gradual development of a diffuse nephritis with glomerular lesions comparable to those of advanced human glomerulonephritis. Such diffuse changes were not seen in the kidneys of group B killed within 46 days of the last cauterization.

The second series of experiments is essentially a repetition of the first, save that many of the rats were operated upon at an earlier age, and kept under observation for longer periods. Excluding those that died prematurely or of incidental causes, there remained 16 operated, and 12 control rats. The additional data afforded by these experiments are summarized in Table II.

These figures amply confirm those of the earlier experiments. In spite of their lower body weight, there is an approximately 100 per cent increase in the mean parathyroid volume of the operated rats, over
that found in their litter mate controls. Calculated on the basis of 
maximal weight, and expressed as volume of parathyroid per 100 gm. 
of rat, the difference is still more striking. The severity of the renal 
lesions was graded without knowledge of the parathyroid measure-
ments. It is seen that the degree of hypertrophy is closely correlated 
with the intensity of the kidney lesions. In one instance in which 
the nephritic changes were pronounced, the parathyroids were 5.6

TABLE II

<table>
<thead>
<tr>
<th>No.</th>
<th>Normal controls</th>
<th>Nephritic rats</th>
<th>Severity of kidney lesions</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Absolute volume</td>
<td>Volume per 100 gm. (maximum weight)</td>
<td>Absolute volume</td>
</tr>
<tr>
<td></td>
<td>c.mm.</td>
<td>c.mm.</td>
<td>c.mm.</td>
</tr>
<tr>
<td>1</td>
<td>0.536</td>
<td>0.158</td>
<td>0.675</td>
</tr>
<tr>
<td>2</td>
<td>0.235</td>
<td>0.066</td>
<td>0.462</td>
</tr>
<tr>
<td>3</td>
<td>0.418</td>
<td>0.167</td>
<td>1.31</td>
</tr>
<tr>
<td>4</td>
<td>0.307</td>
<td>0.135</td>
<td>0.433</td>
</tr>
<tr>
<td>5</td>
<td>0.403</td>
<td>0.175</td>
<td>0.755</td>
</tr>
<tr>
<td>6</td>
<td>0.540</td>
<td>0.135</td>
<td>0.931</td>
</tr>
<tr>
<td>7</td>
<td>0.518</td>
<td>0.173</td>
<td>0.686</td>
</tr>
<tr>
<td>8</td>
<td>0.180</td>
<td>0.090</td>
<td>0.709</td>
</tr>
<tr>
<td>9</td>
<td>0.541</td>
<td>0.183</td>
<td>1.52</td>
</tr>
<tr>
<td>10</td>
<td>0.432</td>
<td>0.141</td>
<td>0.398</td>
</tr>
<tr>
<td>11</td>
<td>0.590</td>
<td>0.147</td>
<td>0.945</td>
</tr>
<tr>
<td>12</td>
<td>0.400</td>
<td>0.167</td>
<td>0.528</td>
</tr>
<tr>
<td>13</td>
<td></td>
<td></td>
<td>0.362</td>
</tr>
<tr>
<td>14</td>
<td></td>
<td></td>
<td>0.800</td>
</tr>
<tr>
<td>15</td>
<td></td>
<td></td>
<td>2.421</td>
</tr>
<tr>
<td>16</td>
<td></td>
<td></td>
<td>0.700</td>
</tr>
</tbody>
</table>

Mean . . . . 0.425 0.145 0.852 0.343

times as large as those of the control litter mate of the same sex, age 
and weight. In every instance, regardless of the severity of the renal 
lesions, both absolute and relative volume in the operated rats exceeded 
the mean volume of the controls.

The question may be raised at this point as to a possible sex differ-
ence in parathyroid volume. In human beings, Pappenheimer and 
Wilens (6) found that the mean weight of the female glands during the
period of active sex life was 22 per cent in excess of the male glands. In the small series of 10 male and 11 female unoperated controls, the mean volume per 100 gm. of rat was 0.1610 c.mm. in the females as against 0.1348 c.mm. in the males—a preponderance of about 20 per cent. The significance of this sex difference and its possible relation to certain phases of the reproductive cycle is being studied by Peters and Andersen, and will be reported upon elsewhere. The point of interest in connection with the present problem is that the enlargement due to reduction of kidney substance greatly transcends that attributable to sex difference.

It was expected that the parathyroid hypertrophy might bring about skeletal changes in the direction of osteitis fibrosa. In the second group of 16 rats surviving from 148 to 306 days after the second operation, only 2 showed histological changes in the bones, and these were of a trifling character. The epiphyses were normal for the age. In rat B7 the perforating vessels of the cortex in femur and tibia were surrounded by fibrous tissue in which the fibers were impregnated with calcium. There was increased osteoclastic resorption. Rat H7 showed areas of marrow fibrosis and a few osteoclasts in the rib (Fig. 3). In the other cases in spite of the renal insufficiency and parathyroid enlargement, no evidence of excessive bone resorption was found.

In view of these negative results further experiments were performed which differed (a) in that the rats were operated upon at an earlier age, (b) the operative technique was modified in the hope of effecting a still greater reduction in functional kidney tissue, (c) the calcium intake was reduced to various levels.

The operation was a modification of that devised by Chanutin and Ferris (21) for the study of renal insufficiency in rats. The kidney exposed through a lumbar incision was delivered through the wound, and both poles snipped off with sharp scissors. The cut surfaces were then seared with a hot knife, no ligature being used. The remaining kidney tissue, reduced to about ½ or ⅓ of the original volume, was replaced and peritoneum and skin closed with sutures. A few days later the opposite kidney was removed *in toto*. The wounds invariably healed without infection.

The operations were performed when the rats were 10 to 15 days old. After weaning, they were placed on the following low calcium diet, with or without a supplement of CaCO₃.
No antirachitic was added, and direct sunlight was excluded. The unoperated controls on this low calcium diet showed a fairly satisfactory but not entirely normal growth curve, whereas the partially nephrectomized animals lagged behind and in most instances were stunted in their development; after an initial period of growth the weight curve became stationary or declined. The dwarfed appearance of the operated animals in comparison to their litter mate controls is well shown in Fig. 4.

As was anticipated from the work of Luce (22), the lowered calcium content of the diet in itself led to a noteworthy increase in the volume of the parathyroids. Thus the mean volume per 100 gm. in 14 unoperated controls on low calcium diet killed at various ages was 0.305 c.mm., as compared with 0.144 and 0.145 in the two series given the stock laboratory diet with adequate calcium.

The hyperplasia of the parathyroid in the operated rats, however, was very much greater than in the controls. In spite of the retarded growth, the mean absolute volume in 8 operated rats was almost double that of the controls (0.657 c.mm. as compared with 0.376 c.mm.); the volume per 100 gm. of rat (based on maximal weight) was 0.615 c.mm. as compared with 0.305 c.mm.

Most interesting were the changes produced in the bones. In the unoperated controls, the lesions were those characteristic of a low calcium diet in the presence of an adequate amount of phosphate and conformed to the description of Pappenheimer, McCann and Zucker (23), and did not show the more extreme lesions described by Shipley, Park, McCollum and Simmonds (24) on their low calcium diet. The bones cut with less than normal resistance, but showed no marked deformity and little swelling at the epiphyses. Microscopically there
was a moderate uniform increase in the width of the zone of preparatory calcification and matrical deposition of calcium was somewhat defective. The trabeculae of the spongiosa were orderly and parallel in alignment, but rather thin, as was the cortex. Both trabeculae and cortex were surrounded by an osteoid border slightly in excess of the normal width. Osteogenesis was active, and there was no osteoclastic resorption or marrow fibrosis (Fig. 6).

The bones of the partially nephrectomized rats presented a striking contrast. In the gross, there was extreme rachitic deformity of the thorax with beading and angulation at the chondrocostal junctions, leading to narrowing of the thoracic cavity and extensive atelectasis of the lungs. The wrists and long bones showed typical epiphyseal swelling, and very characteristic rachitic cupping in the x-rays (Fig. 7a).

The histological picture differed in no respect from that of a florid low phosphorus rickets. There was great widening and irregularity of the cartilage, with almost complete failure of calcium deposition in the matrix. There was an extraordinary excess of calcium-free osteoid in the metaphyseal region and about the cortex (Figs. 8, 5).

Two representative experiments may be presented in tabular form. (Table III and Table IV.)

From these illustrative experiments several facts emerge. While the low calcium diet in itself brings about retardation of growth, the chronic renal insufficiency induced by experimental reduction of kidney tissue leads to more extreme stunting, in some instances (rat AE4) to dwarfism, comparable to that which accompanies renal rickets in human beings. In these cases, the skeletal lesions are extreme and the histological changes are those of florid rickets, with almost complete failure of calcium deposition in cartilage and osteoid.

While the low calcium diet in itself leads to a decided increase in the volume of the parathyroids, partial nephrectomy brings about a striking additional enlargement of the glands.

In the following experiment the low calcium diet was supplemented with 250 mg. per cent of CaCO₃ (Table V).

Rat AG4, operated upon at a very early age, died spontaneously after 29 days; it was greatly dwarfed, weighing only 36 gm. at death, as compared with 94 gm. in the litter mate control. The kidney was reduced to a thin walled hydronephrotic sac. Microscopically, the pelvis and tubules were greatly dilated, lined
### TABLE III

**Litter AD, Born Nov. 3, 1935, Low Calcium Diet**

<table>
<thead>
<tr>
<th>No.</th>
<th>Operation</th>
<th>Total age</th>
<th>Time after 2nd operation</th>
<th>Final Weight</th>
<th>Final Length</th>
<th>PO₄ Absolute</th>
<th>Per 100 gm. (maxim.)</th>
<th>Per 100 gm. (final)</th>
<th>Kidney lesions</th>
<th>X-ray</th>
<th>Length of Femur</th>
<th>Length of Tibia</th>
<th>Rachitic lesions</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Control</td>
<td>79 days</td>
<td>122 days</td>
<td>115 mm.</td>
<td>15.3 mm.</td>
<td>0.649</td>
<td>0.532</td>
<td>0.532</td>
<td>±</td>
<td></td>
<td>25 mm.</td>
<td>31 mm.</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>Nov. 13, 1935, partial L</td>
<td>98 days</td>
<td>75 days</td>
<td>140 mm.</td>
<td>16.0 mm.</td>
<td>0.844</td>
<td>0.562</td>
<td>0.603</td>
<td>+</td>
<td>+</td>
<td>28 mm.</td>
<td>36 mm.</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Nov. 13, partial L</td>
<td>98 days</td>
<td>96 days</td>
<td>130 mm.</td>
<td>17.5 mm.</td>
<td>1.288</td>
<td>0.810</td>
<td>0.993</td>
<td>+</td>
<td>+</td>
<td>26 mm.</td>
<td>31 mm.</td>
<td>+</td>
</tr>
<tr>
<td>4</td>
<td>Dec. 4, complete R</td>
<td>119 days</td>
<td>75 days</td>
<td>140 mm.</td>
<td>16.0 mm.</td>
<td>0.492</td>
<td>0.262</td>
<td>0.262</td>
<td>±</td>
<td></td>
<td>30 mm.</td>
<td>35 mm.</td>
<td></td>
</tr>
</tbody>
</table>

### TABLE IV

**Litter AE, Born Nov. 30, 1935, Low Calcium Diet**

<table>
<thead>
<tr>
<th>No.</th>
<th>Operation</th>
<th>Total age</th>
<th>Time after 2nd operation</th>
<th>Final Weight</th>
<th>Final Length</th>
<th>Serum Ca</th>
<th>Volume of parathyroid</th>
<th>Kidney lesions</th>
<th>Bones</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>Dec. 9, 1935, partial L</td>
<td>69 days</td>
<td>59 days</td>
<td>76 mm.</td>
<td>14.8 mm.</td>
<td>0.530</td>
<td>0.696</td>
<td>0.696</td>
<td>+</td>
</tr>
<tr>
<td>4</td>
<td>Dec. 9, partial R</td>
<td>69 days</td>
<td>59 days</td>
<td>46.5 mm.</td>
<td>12.8 mm.</td>
<td>?</td>
<td>?</td>
<td>?</td>
<td>+</td>
</tr>
<tr>
<td>5</td>
<td>Dec. 19, complete L</td>
<td>69 days</td>
<td>59 days</td>
<td>89 mm.</td>
<td>16.5 mm.</td>
<td>0.544</td>
<td>0.570</td>
<td>0.589</td>
<td>+</td>
</tr>
<tr>
<td>7</td>
<td>Control</td>
<td>69 days</td>
<td>105 days</td>
<td>17.5 mm.</td>
<td>5.9 mm.</td>
<td>0.362</td>
<td>0.331</td>
<td>0.342</td>
<td>+</td>
</tr>
<tr>
<td>8</td>
<td>Control</td>
<td>69 days</td>
<td>98 days</td>
<td>17.5 mm.</td>
<td></td>
<td>0.310</td>
<td>0.305</td>
<td>0.316</td>
<td>+</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>No.</th>
<th>Operation</th>
<th>Total age</th>
<th>Time after 2nd operation</th>
<th>Final Weight</th>
<th>Final Length</th>
<th>Stock Diet</th>
</tr>
</thead>
<tbody>
<tr>
<td>9</td>
<td>Control</td>
<td>69 days</td>
<td>124 days</td>
<td>20</td>
<td>10.6</td>
<td>0.236</td>
</tr>
<tr>
<td>10</td>
<td>Control</td>
<td>69 days</td>
<td>130 days</td>
<td>19.5</td>
<td>10.6</td>
<td>0.161</td>
</tr>
</tbody>
</table>
### TABLE V

*Litter AG, Born Feb. 10, 1936, Low Calcium Diet + 250 Mg. Per Cent CaCO₃*

<table>
<thead>
<tr>
<th>No.</th>
<th>Operation</th>
<th>Total age</th>
<th>Time after 2nd operation</th>
<th>Final</th>
<th>Nonprotein nitrogen</th>
<th>Ca</th>
<th>Volume of parathyroid</th>
<th>Kidney lesions</th>
<th>Bones</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>days</td>
<td>days</td>
<td>gm.</td>
<td>mm.</td>
<td>mg. per cent</td>
<td>Absolute</td>
<td>Per 100 gm. (maximal)</td>
<td>Per 100 gm. (final)</td>
</tr>
<tr>
<td>4</td>
<td>Feb. 15, 1936, partial L</td>
<td>38</td>
<td>29</td>
<td>36</td>
<td>11</td>
<td>0.3512</td>
<td>0.903</td>
<td>0.974</td>
<td>++++</td>
</tr>
<tr>
<td></td>
<td>Feb. 19, complete R</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>7</td>
<td>Control</td>
<td>39</td>
<td></td>
<td>94</td>
<td>15</td>
<td>0.118</td>
<td>0.1256</td>
<td>0.1256</td>
<td>-</td>
</tr>
<tr>
<td>5</td>
<td>Feb. 19, partial R</td>
<td>104</td>
<td>91</td>
<td>218</td>
<td>19.5</td>
<td>0.6587</td>
<td>0.3021</td>
<td>0.3021</td>
<td>++</td>
</tr>
<tr>
<td></td>
<td>Feb. 23, complete L</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
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<td>Control</td>
<td>104</td>
<td></td>
<td>248</td>
<td>20</td>
<td>0.4889</td>
<td>0.1971</td>
<td>0.1971</td>
<td>-</td>
</tr>
<tr>
<td>9</td>
<td>Control</td>
<td>104</td>
<td></td>
<td>260</td>
<td>19.5</td>
<td>0.4304</td>
<td>0.1655</td>
<td>0.1655</td>
<td>-</td>
</tr>
</tbody>
</table>
with flattened cells. Casts were numerous. The glomeruli were small, the
capsular spaces distended with albuminous precipitate, but there were no adhesions
or hyaline changes. A few calcified casts were found in the collecting tubules.
Quite marked changes were found in femur, tibia and ribs. The lesions may be
described as combining the features of a mild rickets with those of osteitis fibrosa.
The cartilage showed a few blunt imperfectly calcified prolongations towards the
metaphysis, but in general hardly exceeded the normal width. The spongiosa was
composed of irregularly disposed, thin trabeculae, separated by patches of fibrous
narrow with numerous osteoclasts (Fig. 9). The cortex in the region of the
metaphysis was thinned, and in places completely defective, being replaced by
fibrous tissue with many giant cells. Further along the shaft, the marrow resumed
its normal appearance, but the osteoid margin in some places was distinctly
increased in width. The control rat, killed on the same day, had normal bones,
the added CaCO₃ being sufficient to prevent obvious histologic lesions.

Other experiments in which lesser or greater amounts of CaCO₃
were added to the low calcium diet, have not given additional inform-

**Effect of Parathormone Injections upon the Bones of Partially
Nephrectomised Rats**

A possibility, which we have not seen discussed, is that the organism
is rendered more sensitive to the effect of parathyroid hormone when
there is a state of renal insufficiency. One of the factors leading in this
direction may be the accompanying acidosis. Indeed, it has been
well shown by Olsen (9) that the activity of paroidin, as measured by
the urinary excretion of calcium after injection, is greatly enhanced
when the animals are placed on an acid forming diet (NH₄Cl, meat).

That the bone lesions resulting from repeated injections of para-
thalmone in young rats are exaggerated, when the renal function is
experimentally reduced, is indicated by the following experiment.

Litter AH, Mar. 20, 1936. Partial nephrectomy (R), at age of 8 days, Mar. 23
left kidney removed. Beginning Mar. 25 rats 2 and 4 of the litter and unoperated
control 7 were given daily two intraperitoneal injections of parathyroid extract
(Lilly), in all 60 to 90 units. The two operated rats died 7 days after the second
operation, and the control was killed on the same day.

A histological study of ribs, tibia and femora showed distinctly
more severe changes in the injected nephrectomized rats (cf. Fig. 10).
Both in ribs and long bones, the submetaphyseal region is almost
replaced by fibrous tissue, containing only a few fragmented bone trabeculae. The cortex is reduced to a thin ribbon and entirely lost over considerable areas. Osteoclasts are extremely numerous in this region, but fibrosis of the marrow and osteoclasts are seen along the entire course of the rib and along the shafts of the tibia and fibula.

In contrast to these extreme lesions, the unoperated injected control shows only minor changes. The rib (Fig. 11) is normal save for a few osteoclasts and slight thinning of the cortex just below the cartilage. Tibia and femur show minimal lesions.

It would seem from this and other similar experiments that demineralization of the bones in young rats following parathyroid extract administration is intensified by the reduction of renal function. To determine whether this is caused by acidosis, alterations in calcium-phosphorus metabolism, diminished calcium intake or other unknown factors will require much further investigation.

DISCUSSION

It has been shown that the parathyroid glands react to a loss of functional renal tissue by an increase in volume, roughly proportional to the severity of the kidney damage. The immediate stimulus to this enlargement of the gland, which one may assume to signify increased functional activity, has not been determined. The suggestion of Schelling (25) and others that the enlargement in chronic nephritis is probably the "result of a functional demand on the glandules to rid the body of retained phosphates" is not wholly satisfactory. There is no direct evidence that the hyperphosphatemia per se is the chemical incitant to hyperplasia, and we are confronted with the fact that such hyperplasia occurs with equal constancy in low phosphorus rickets. Many of the reported cases of renal rickets have shown plasma phosphate values within the normal limits (Ellis and Evans, (26)). In the case reported by Elsom, Wood and Ravdin (27) of hyperparathyroidism with renal insufficiency, removal of an adenoma was followed by a permanent return of the serum calcium and PO₄ values to normal. In this patient, phosphate retention was never observed and could not have supplied the primary stimulus to parathyroid hyperplasia.

4 Schelling (25), page 278.
It seems idle to enter into a theoretical discussion of this point, since the precise mode of action of the parathyroid secretion is still obscure.

In reviewing the literature of renal rickets, there is a recurring suggestion that the hyperactive parathyroids play a part in the demineralization of the skeleton, bringing about the rachitic or osteofibrotic lesions which characterize the disease. Indeed, in certain cases occurring in adolescence or early adult life, in which there has been found a diffuse hypertrophy of all the parathyroids, the differentiation between primary and secondary hyperparathyroidism may be difficult or impossible.

Our experiments thus far have given no clear cut information as to the part played by the parathyroids in the production of the skeletal lesions. On a diet containing adequate calcium and phosphorus the parathyroid hyperplasia induced by partial nephrectomy was only exceptionally accompanied by slight osteofibrotic changes in the bones and these were never of an intensity comparable to those observed in "primary" hyperparathyroidism or in renal rickets.

On the other hand, when rats with experimental renal insufficiency were placed on a low calcium diet, the resulting skeletal lesions were greatly intensified. On a very low calcium intake, they took on the character of florid rickets indistinguishable from that produced by the usual low phosphorus rachitogenic diets. With a moderate addition of calcium, the picture was rather that of an osteitis fibrosa.

The crucial experiment, which has not yet been performed, will be to study the effect of experimental renal insufficiency in the absence of the parathyroids. In this way we should secure information as to the rôle played by these glands in the production of the skeletal lesions.

One of the purposes of this study was to reproduce, if possible, in rats the clinical picture of renal rickets or renal dwarfism in man. The outstanding features of this disease are stunted development, sometimes with delay or failure in the acquisition of the secondary sex characters; bone deformities, often genu valgum, with x-ray changes, sometimes suggesting osteitis fibrosa, sometimes florid rickets; chronic renal insufficiency, ending in marked nitrogen retention and uremia. Polydipsia and polyuria have been frequently noted. The blood calcium may be normal or slightly reduced, the inorganic phosphate, in the later stages, tends to be elevated.
The renal changes at autopsy are usually described as chronic interstitial nephritis, but recently attention has been drawn to the frequency of accompanying hydronephrosis and dilatation of the ureters, sometimes with vesical hypertrophy (Ellis and Evans (26), Roberts (28)). In some cases anatomic explanation for the renal obstruction has been found (phimosis, valve-like folds in the urethra (Ellis, (29)). In other cases (Ellis and Evans (26)) no organic stricture has been discovered, and a disturbance in sphincteric control has been assumed.

The histologic alterations in the bones have been studied in a number of instances. The descriptions vary from case to case, and there appears to be every gradation between a florid rickets, indistinguishable from ordinary rickets, to a picture identical with osteitis fibrosa in the adult (11, 24, 30–39). It would be impossible from the histologic changes alone to establish the diagnosis of renal rickets, as distinct from that due to vitamin D deficiency, on the one hand, or to primary hyperparathyroidism on the other.

The same variability has been found in the skeletal changes which follow experimental reduction of renal tissue in young rats. On a very low calcium intake they have resembled the lesions of florid rickets; with a moderate deficiency of calcium, changes are in the direction of osteitis fibrosa.

The stunting of growth and development in our rats is comparable to that noted in human cases of renal rickets. Our data on the blood calcium PO₄ and nonprotein nitrogen are not sufficiently numerous to report in detail, but so far as they go, agree with those reported in human cases of renal rickets, i.e., hyperphosphatemia and high nonprotein nitrogen in the late stages of the experimental disease.

We believe therefore that we have induced a condition essentially like that which accompanies advanced chronic renal disease in childhood. This experimental renal rickets should be useful in working out the various unknown factors concerned in the human disease.

CONCLUSIONS

Reduction of renal tissue in young rats regularly leads to a marked increase in the volume of the parathyroid glands.

If partially nephrectomized rats are maintained on a low calcium diet, growth is stunted, and skeletal lesions are produced, of far greater
severity than can be ascribed to the dietary calcium deficiency alone. The picture closely resembles that found in cases of renal rickets in children.

I am indebted to Dr. Paul Swenson for taking and interpreting radiographs of the bones. Dr. Thomas Todd kindly assisted in some of the experiments.

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EXPLANATION OF PLATES

PLATE 51

FIG. 1. Rat AA2. Removal of right kidney. Cauterization of left. Survived second operation 37 days. Low power showing chronic nephritis, dilatation of tubules, casts, glomerular adhesions, interstitial fibrosis.

FIG. 2. High power, showing glomerular lesions.


PLATE 52

FIG. 4. Litter AE. Low calcium diet. Rats 1 and 2, complete nephrectomy, right, partial left; rat 3, unilateral nephrectomy. Rats 4 and 5, unoperated controls. Rat 6, control on stock diet.


PLATE 53

FIG. 6. Rat AC1. Unoperated control on low calcium diet. Slight increase in width of proliferating cartilage zone.

FIG. 7. Litter AC. Low calcium diet. X-ray of knee joint in operated (a) and control (b) rats.

FIG. 8. Rat AC1. Partial nephrectomy. Low calcium diet. Rib, showing very marked rachitic changes.

PLATE 54


FIG. 10. Rat AH2. Low calcium diet + 150 mg. per cent added CaCO₃. Partial nephrectomy. 80 units of parathyroid extract (Lilly) intraperitoneally in 4 day periods. Rib showing thinning and destruction of cortex and trabeculae, fibrosis of marrow and numerous osteoclasts.

FIG. 11. Rat AH7. Unoperated control to rat AH2. Received similar dose of parathyroid extract. Practically normal rib.
(Pappenheimer: Kidney substance and parathyroid glands)
(Pappenheimer: Kidney substance and parathyroid glands)
(Pappenheimer: Kidney substance and parathyroid glands)