

## 11998

## Relation of Nutrition to Mammary Growth After Estradiol Administration to Hypophysectomized Rats.

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To explain the failure of estrogens to stimulate mammary growth in hypophysectomized animals, as first observed by Lyons and Pencharz,<sup>1</sup> Turner<sup>2</sup> has introduced the hypothesis that the effects of the estrogens and progesterone on the mammary gland are not direct but are due to respectively stimulating the secretion of a duct-growth and an alveolar factor by the anterior lobe of the pituitary gland. On the other hand Lyons, *et al.*,<sup>3</sup> and Speert<sup>4</sup> have shown that if estrogens are administered by inunction over one mammary gland, this gland will show greater development than that on the opposite side. They conclude, therefore, that estrogens must have a direct action on the mammary gland.

The experiments of Astwood, Geschickter and Rausch<sup>5</sup> raised the question of the importance of food intake on mammary development. They restricted intact rats to a food intake comparable to that of hypophysectomized littermates and found the same refractoriness of the mammae to estrogen stimulation.

The hypothesis that the failure of estrogens to act directly on the mammary glands of hypophysectomized animals was due to their state of chronic under-nutrition was given further support by the work of Nathanson, Shaw and Franseen.<sup>6</sup> These workers reported that a growth extract which had no direct effect on the mammary glands enabled estradiol to cause proliferation when given

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<sup>1</sup> Lyons, W. R., and Pencharz, R. I., *PROC. SOC. EXP. BIOL. AND MED.*, 1936, **33**, 589.

<sup>2</sup> Lewis, A. A., and Turner, C. W., *Mo. Agr. Exp. Sta. Res. Bul.* 310, 1939.

<sup>3</sup> Lyons, W. R., and Sako, Y., *PROC. SOC. EXP. BIOL. AND MED.*, 1940, **44**, 398.

<sup>4</sup> Speert, H., *Science*, 1940, **92**, 461.

<sup>5</sup> Astwood, E. B., Geschickter, C. F., and Rausch, E. O., *Am. J. Anat.*, 1937, **61**, 373.

<sup>6</sup> Nathanson, I. T., Shaw, D. T., and Franseen, C. C., *PROC. SOC. EXP. BIOL. AND MED.*, 1939, **42**, 652.

in conjunction with it. The effect of the estradiol seemed to be roughly proportionate to the gain in weight produced by the pituitary preparation. This experiment, however, did not distinguish between the influence of the extract on nutrition (food intake and absorption) and its influence on the internal metabolism. In the following experiment we have attempted to distinguish these two effects by artificially maintaining the food intake of the hypophysectomized rats at such a level that weight was gained instead of lost. With the nutrition adequately maintained in this manner we have studied the response of the mammary gland to estradiol benzoate.

Three litters of 3 young adult virgin female rats each were selected for the experiment. One rat of each litter was kept as a control and the others were hypophysectomized. All animals were then fed a balanced mixed diet by stomach tube in such quantity that body weight was approximately maintained.<sup>7</sup> Five milligrams of desiccated thyroid was given daily mixed in the diet. The rats received sodium chloride-sodium carbonate solution to drink, to which was added 10 cc of Upjohn's cortical extract per 100 cc solution. Estradiol benzoate, 1000 IU in oil,<sup>†</sup> was injected every other day for 28 days. Vaginal smears were run daily. All animals showed estrus smears. At the end of this time gross observations showed that the nipples of the control rats were considerably enlarged while those of the hypophysectomized rats were as small as at the beginning. Both hypophysectomized and normal rats had gained weight during the experimental period, the former an average of 19 g (6-38 g) from an average original weight of 242 g and the latter an average of 27 g (21-33 g) from an average original weight of 204 g.

The animals were then killed, skinned, and the skins preserved in Bouin's solution. The individual mammary glands were dissected free with the nipple attached. They were then stained with hemalum and mounted according to the technic of Lewis.<sup>8</sup> All the control rats showed considerable lobular development with wide branching ducts. On the other hand, the glands of the hypophysectomized rats were so small as to be difficult to dissect. They showed a few twigs and branches, but not significant development of either ducts or secretory alveoli. A typical specimen of a gland from each group is shown in Fig. 1.

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<sup>7</sup> Reinecke, R. M., Ball, H. A., and Samuels, L. T., *PROC. SOC. EXP. BIOL. AND MED.*, 1939, **41**, 44.

<sup>†</sup> The estradiol benzoate was kindly furnished by the Schering Corporation, Bloomfield, N.J.

<sup>8</sup> Lewis, A. A., personal communication.

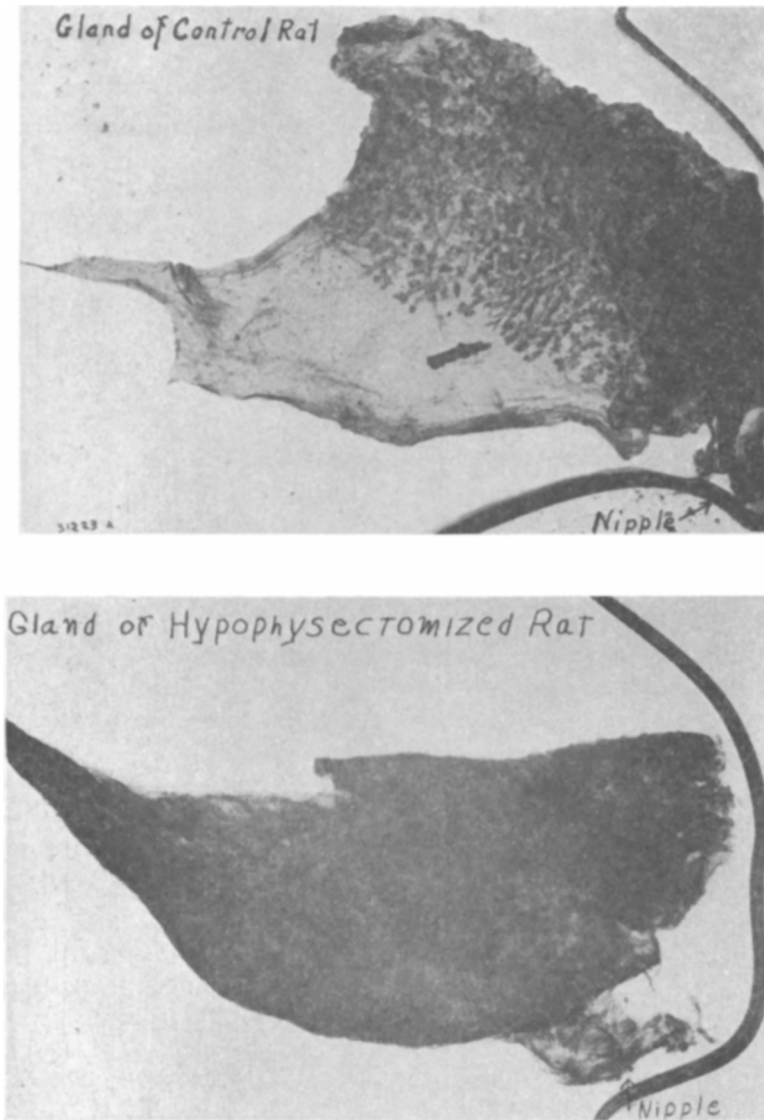


FIG. 1.

Typical Mammary Glands of Hypophysectomized and Control Rats Treated with Estradiol Benzoate.

In both slides the entire fat pad above and including a nipple has been mounted. The pad of the hypophysectomized rat was not destined as much as the control, and the vascular system can therefore be seen. There is obviously no duct system entering the nipple of the hypophysectomized animal.

It seems, therefore, that while the mammary gland may react directly to the stimulation of estradiol, it is essential that there be

present at the same time an active pituitary gland. The supplying of adequate amounts of caloric material is insufficient of itself to enable the mammary gland to respond to estradiol. Either the change in internal metabolism brought about by the growth fraction is essential to mammary development or some specific mammogenic factor is needed.

On the basis of the work of Nathanson, *et al.*,<sup>6</sup> the pituitary factor will apparently only affect the mammae in the presence of estrogens. Since Turner's extracts have not been demonstrated to be estrogen-free, it may be that their effectiveness in hypophysectomized rats was due to the combined presence of small amounts of such substances and some pituitary factor as yet not clearly distinguished. This apparently was present in both Nathanson's and Turner's extracts.

The experiments of Astwood, *et al.*,<sup>5</sup> can best be explained by assuming that the pituitary gland is sensitive to undernutrition. This was first shown by Moore and Samuels<sup>9</sup> in connection with the gonadotropic factor. If this assumption is correct Astwood's rats failed to show mammary development because of lack of the pituitary factor.

*Summary.* Well nourished hypophysectomized rats did not show mammary development when stimulated by large doses of estradiol benzoate for a period of 28 days. When this result is correlated with those of other workers it appears that both estrogens and some pituitary factor must act directly on the mammary gland to produce normal development.

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### Efficiency of Electrical Energy Production by Surviving Frog Skin, Measured by Iodine Coulometer.\*

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Surviving frog skin is the seat of an electrical potential which has been related to factors affecting its metabolic activity.<sup>1, 2</sup> To deter-

<sup>9</sup> Moore, C. R., and Samuels, L. T., *Am. J. Physiol.*, 1931, **96**, 278.

\* Based on data submitted in partial fulfillment of the requirements for the degree of Doctor of Philosophy at the University of Texas, Austin, Texas.

<sup>1</sup> Lund, E. J., *J. Exp. Zool.*, 1926, **44**, 383; 1928, **51**, 291, 327; 1931, **60**, 249.

<sup>2</sup> Francis, W. L., *Nature*, 1933, **131**, 805.