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## Oestrogenic Treatment of Hypophysectomized Male Mice.*

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With increasing numbers of investigations on the effect of oestrogenic treatment of hypophysectomized animals 2 schools of thought seem to have arisen. Individual studies on hypophysectomized rats and guinea pigs[1, 2, 3] seemed to show that in no case were the mammary glands able to respond to relatively large doses of female hormone. Other investigators[4-7] report that they could obtain normal mammary development even in the absence of the pituitary. One is at a loss to correlate these conflicting results, but it is hoped that this and future studies will help clarify the situation.

Hypophysectomy of the mouse is carried out in much the same fashion as in the rat or guinea pig. The hardiness of some strains of mice allows them to stand the operation very well, and postoperative recovery offers no significant difference from that described for the rat. In the male, the gonads and accessories begin regression at once, and approach a static condition about a month after operation. Regardless of when oestrogenic injections are started in hypophysectomized mice, they do not tolerate this treatment well, and a majority succumb within 48 hours. For this reason the number of animals herein described is not large, but analysis of the data is considered significant.

A total of 21 hypophysectomized mice were treated with various types of oestrogenic hormone. Of these animals 10 were later

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

2 Gomez, E. T., and Turner, C. W., PROC. SOC. EXP. BIOL. AND MED., 1936, **34**, 320.

3 Reece, R. P., Turner, C. W., and Hill, R. T., PROC. SOC. EXP. BIOL. AND MED., 1936, **34**, 204.

4 Nelson, W. O., PROC. SOC. EXP. BIOL. AND MED., 1935, **33**, 222.

5 Ruinen, F. H., *Acta Brevia Neerl.*, 1932, **2**, 161.

6 Freud, J., and De Jongh, S. E., *Acta Brevia Neerl.*, 1935, **5**, 47.

7 Asdell, S. A., and Seidenstein, H. R., PROC. SOC. EXP. BIOL. AND MED., 1935, **32**, 931.

found to be completely hypophysectomized. In most of the animals the base of the skull was serially sectioned, and in some the remaining fragments were calculated on 3-dimension volumetric measurements.

Recently Hill and Gardner[8] have shown that small remaining fragments of anterior lobe will not maintain the gonads in either male or female hypophysectomized mice. Unpublished data (Hill and Gardner) show that in hypophysectomized male mice bearing ovarian grafts, the addition of a pituitary graft will allow growth of the mammary glands. The results now under discussion confirm these observations. Two animals, 4 and 44, retained 2 and 8% of the pituitary respectively, the fragments containing all 3 parts of the original gland. In the remaining incompletely hypophysectomized animals volume measurements were not made. The testes of these same 2 animals were very much regressed, as shown by tubule diameter and the condition of the accessory sex glands. It will also be noted that the adrenal cortex had undergone much involution as indicated by its reduced thickness. Gardner[9] has shown that female hormone will cause the replacement of the bony pubic symphysis by a ligament. In 3 of the cases reported in this paper, the same reaction had taken place, although the pituitary was completely absent.

Absolutely no mammary response was obtained by the treatment in those animals which were completely hypophysectomized. However, if even so small an amount as 2% of the pituitary remained intact, part of which was intermediate and posterior lobes, the mammary response was very good. The response obtained was essentially the same as that found in intact animals. Therefore in mice we may consider that some part of the anterior pituitary is essential to mammary response by theelin treatment. Our experience leads to the suggestion that in some types of experiments, especially those in testing oestrogenic hormone response on mammary glands, it is absolutely essential to know, by stained serial sections, just how much of the pituitary may remain intact. Small remaining fragments cannot be seen in fresh material, even with the aid of a low power dissecting microscope. It is certain that in dealing with anterior pituitary physiology, one must consider the fragment as well as the whole gland.

*Summary.* In completely hypophysectomized male mice, treated with female hormone, the mammary gland rudiments do not respond.

[8] Hill, R. T., and Gardner, W. U., PROC. SOC. EXP. BIOL. AND MED., 1936, **34**, 78.

[9] Gardner, W. U., PROC. SOC. EXP. BIOL. AND MED., 1935, **33**, 104.

TABLE I.

| No. | Days injected | I.U. weekly | I.U. daily | I.U. Total | Pituitary Left % | Mammary response | Tubule diam. (microns) | Acces-sories | Thickness adrenal cortex (microns) | Symphysis |
|---|---|---|---|---|---|---|---|---|---|---|
| 4 | 114 | 150 O.B. | | 2400 | 2 | Good | 90-100 | Cast. | | |
| 16 | 8 | 150 " | | 150 | 0 | None | | | | |
| 41 | 11 | 250 " | | 500 | 0 | " | 90-100 | " | 140-150 | Loose |
| 42 | 28 | 250 " | | 1000 | 0 | " | 70-80 | " | 140-150 | |
| 43 | 26 | 250 " | | 2000 | 0 | " | | " | | |
| 44 | 38 | 500 " | | 3000 | 8 | Good | 85-90 | " | | " |
| 45 | 28 | 250 " | | 1000 | 0 | None | 70-80 | " | 145-155 | " |
| 2 | 30 | | 50 P.B. | 1500 | 0 | " | | | | |
| 7 | 20 | | 50 T.O. | 1000 | 0 | " | | | | |
| 8 | 20 | | 50 " | 1000 | 0 | " | | | | |
| 13 | 20 | | 50 O.B. | 1000 | 0 | " | | | | |
| 14 | 25 | | 50 " | 1250 | 0 | " | | | | |
| 1 | 20 | | 50 P.B. | 1000 | unmeasured fragments | Good | | | | |
| 3 | 60 | | 50 " | 3000 | " | " | | | | |
| 5 | 20 | | 50 " | 1000 | " | " | | | | |
| 6 | 20 | | 50 " | 1000 | " | " | | | | |
| 9 | 20 | | 50 " | 1000 | " | " | | | | |
| 10 | 20 | | 50 T.A. | 1000 | " | " | | | | |
| 11 | 20 | | 50 " | 1000 | " | " | | | | |
| 12 | 20 | | 50 " | 1000 | " | " | | | | |
| 15 | 25 | | 50 O.B. | 1250 | " | " | | | | |
| | 25 | 150 " | | 525 | All | " | 190-210 | Normal | 240-260 | Loose 3-6 weeks |

P.B. = Progynon-B.† T.O. = Theelin in oil.† T.A. = Theelin (aqueous).† O.B. = Oestroform-B.

The gonads, accessories and adrenal cortex undergo regression regardless of the female hormone present. The resorption of the pubic symphysis does not depend on the presence of anterior lobe material. A small fragment of prehypophysis enables the administered female hormone to induce growth of the mammary gland. Such a small pituitary fragment permits regression of the gonads, accessories and adrenal cortex. Caution must be urged in use of the term "physiologically hypophysectomized" animals.†

## 9204 P

## Effects of Saponin and Digitonin on Lipase and Phosphatase Action.

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In the course of the investigation of the effects of various snake venoms on lipolytic action controls were run to determine whether any of the observed effects might be due to the decreased surface tension caused by the venoms. Two of the agents so employed were "purified" saponin, Merck, and Hoffman-LaRoche digitonin. The following communication is a report on the rather interesting observation that the former markedly inhibits the activity of pancreatic lipase but inhibits the lipase activity of blood only slightly, while digitonin in very low concentrations increases the lipolytic action of pancreatic lipase and has little or no effect on blood lipase action. That the activity in each case seems to be due to some factor apart from the effect on surface tension is indicated by the fact that both of these reagents are very powerful surface tension depressants. The inability of both to show the same marked effects on blood lipase seems to indicate that they are inactivated or removed by some constituent in blood serum. It is likely that the serum cholesterol combines with the saponin or the digitonin to form a very slightly soluble digitonide, thus removing the inhibiting or stimulating factor.

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