

reappearing with a slight prolongation of the diestrus interval. The ovaries of the treated animals are loaded with corpora lutea and weigh 4 or 5 times as much as those of the controls.

Kelly³ has reported that the injection of pregnancy urine into pregnant guinea pigs invariably produces abortion. The administration of our extracts to rats greatly prolongs the gestation period.⁴ This is true when the injections are started at any time between the fourteenth and nineteenth days of pregnancy. At autopsy the fetuses are usually dead and partially resorbed but fully developed and often of greater than normal size. The ovaries are much larger and more luteinized than those of control pregnant rats.

Since the luteinization is so pronounced it would seem reasonable to attribute the delay of parturition to this phenomenon. To test this idea we have administered the extracts to ovariectomized pregnant rats but in no case was the gestation period prolonged. Apparently, the presence of the ovary is necessary to produce this effect and since large quantities of theelin have no influence on the pregnancy, it seems probable that the corpora lutea are responsible for the retention of the fetuses.

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Chromatophore Reaction to an Oestrogenic Hormone.

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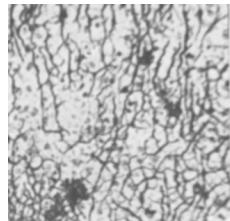
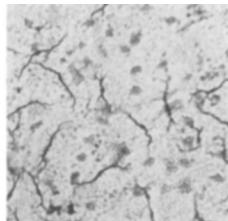
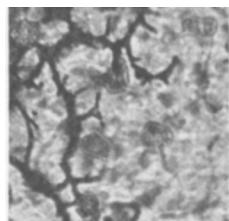
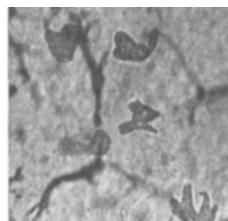
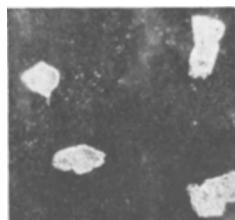
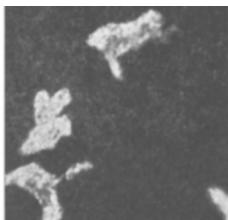
Some 3000 tadpoles (*Rana pipiens*) were treated during 5 months with the oestrogenic hormone theelin obtained from the Biochemical Laboratory of Doctor Doisy. Several series of experiments were carried on simultaneously in which the tadpoles were exposed to different concentrations of theelin over varying periods of time. In other experiments 100 tadpoles were injected with the hormone.

In one series 250 tadpoles just emerged from the gelatin coating were placed in a 1000 cc. aqueous solution of theelin 0.0000003125 mg./cc. for 60 minutes. On successive days the exposure time was increased by 60 minutes for a period of 10 days. This time increment was kept constant during the rest of the experiment but the concentration of the hormone was doubled every 10 days until a

³ Kelly, L. G., *Anat. Rec.*, 1931, **48**, 50 (supplement).

⁴ Levin, L., Katzman, P. A., and Doisy, E. A., in press.

concentration of 0.0000025 mg./cc. was attained. The experiment was completed on the 40th day. Higher concentrations of the hormone effected a marked change during the last 10 days of the ex-

FIG. 1. $\times 1.5$ FIG. 2. $\times 72$ FIG. 3. $\times 72$ FIG. 4. $\times 154$ FIG. 5. $\times 154$ FIG. 6. $\times 450$ FIG. 7. $\times 450$

periment. Microscopic examination of fresh epidermis mounts revealed a recession of pigment centrally toward the cell body in the epidermal melanophores and a pseudopodial expansion of the leucophores.

Some 65 tadpoles (total length 26 mm.) were injected intraperitoneally with 0.05 cc. theelin 0.00035 mg./cc. on 3 successive days by means of a glass needle (0.1 mm. diameter) attached to a Chambers microinjection apparatus. Color change (silvering) was first observed on the 6th day after the first injection. Tadpoles A and C (Fig. 1) show the color change following the injection. Tadpole B (Fig. 1) is a control. This is a photograph of the living animals quieted in iced water. Fig. 2 shows a mounted section of epidermis from the mid-tail region of tadpole B. The epidermal melanophores are multi-branched with frequent anastomosis. Fig. 3 shows a similar section from tadpole A. A recession of pigment centrally toward the cell body is seen in the epidermal melanophores. Practically no anastomosis nor marked branching is evident. Fig. 4 shows the normal contour of the leucophores in the control tadpole. Fig. 5 shows the pseudopodial expansion of the leucophores in the experimental animal. Figs. 6 and 7 are microphotographs taken by dark-field illumination of the leucophores from control and experimental animals respectively. The normal contour of the leucophores in the control animal and the pseudopodial contour of the leucophores of the experimental animal are evident. All of the photographs are made from untouched negatives.

Conclusions. The oestrogenic hormone apparently has an effect upon the pigmentary system of the tadpole causing albinism. The chromatophore reaction involves a centripetal movement of pigment in the epidermal melanophores and a pseudopodial expansion of the leucophores. The velocity of the reaction is directly related to the concentration of the hormone and the duration of exposure. Intraperitoneal injection causes a more rapid response. It is generally admitted that the pigmentary system of amphibia is controlled by the secretions of the pituitary. Extracts from the pituitary gland cause melanism. The oestrogenic hormone causes albinism. It is probable that in the control of the melanophores and the leucophores there exists a physiological antagonism between the secretions of the pituitary and the oestrogenic hormone. A further histological study of the effect of the injected theelin upon the amphibian pituitary will be presented in later studies.