

regard, an immature condition was suggested. Sperm ducts were apparently stimulated to increased growth by the hormone. The spermatogenic suppression might have been due either to a direct effect of estrogens on the male gonad or to an inhibition of hypophyseal activity. The factors involved are being studied further.

The secondary sex characters of adult males could not be modified by estrogenic hormone treatment beginning at maturity and continued for periods up to 4 months.

*Conclusions.* Estrogenic hormone feeding, started at birth, caused the suppression of male secondary sex characters and a marked suppression of spermatogenesis in the male guppy.

### 9253 P

#### Release of Spermatozoa by Anterior Pituitary Treatment of the Male Frog, *Rana pipiens*.

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Houssay and Lascano-Gonzalez,<sup>1</sup> using *Bufo marinus* males, demonstrated that hypophyseal removal causes degeneration and hypophyseal implantation causes hypertrophy of the testes. O. M. Wolfe<sup>2</sup> implanted pituitary glands subcutaneously to induce amplexus in *Rana pipiens* and Rugh<sup>3</sup> showed that extracts of mammalian anterior pituitary (antuitrin-S or whole sheep gland) would induce amplexus in toads but not in frogs.

The standard technique for securing developing frog's eggs<sup>4</sup> does not require pituitary treatment of males. Functional sperm are available at all times during hibernation simply by cutting up the testes of mature, hibernating frogs in spring water. However, anterior pituitary injection of hibernating male frogs not only induces amplexus (with ovulating females of the same species, only) but releases from the Sertoli cells all mature spermatozoa. This can be demonstrated by subjecting hibernating males from which single testes have been removed to anterior pituitary treatment. Amplexus

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<sup>1</sup> Houssay, B. A., and Lascano-Gonzalez, J. M., *Compt. rend. Soc. biol.*, 1929, **101**, 938.

<sup>2</sup> Wolfe, O. M., *PROC. SOC. EXP. BIOL. AND MED.*, 1929, **26**, 692.

<sup>3</sup> Rugh, R., *Biol. Bull.*, 1935, **68**, 74.

<sup>4</sup> Rugh, R., *Biol. Bull.*, 1934, **66**, 22.

will be achieved in about 16 hours and the remaining testis may then be removed and studied. In the control testis (Fig. 1) the mature sperm may be seen clustered in groups around their Sertoli cells while the testis from the same frog after sexual stimulation by

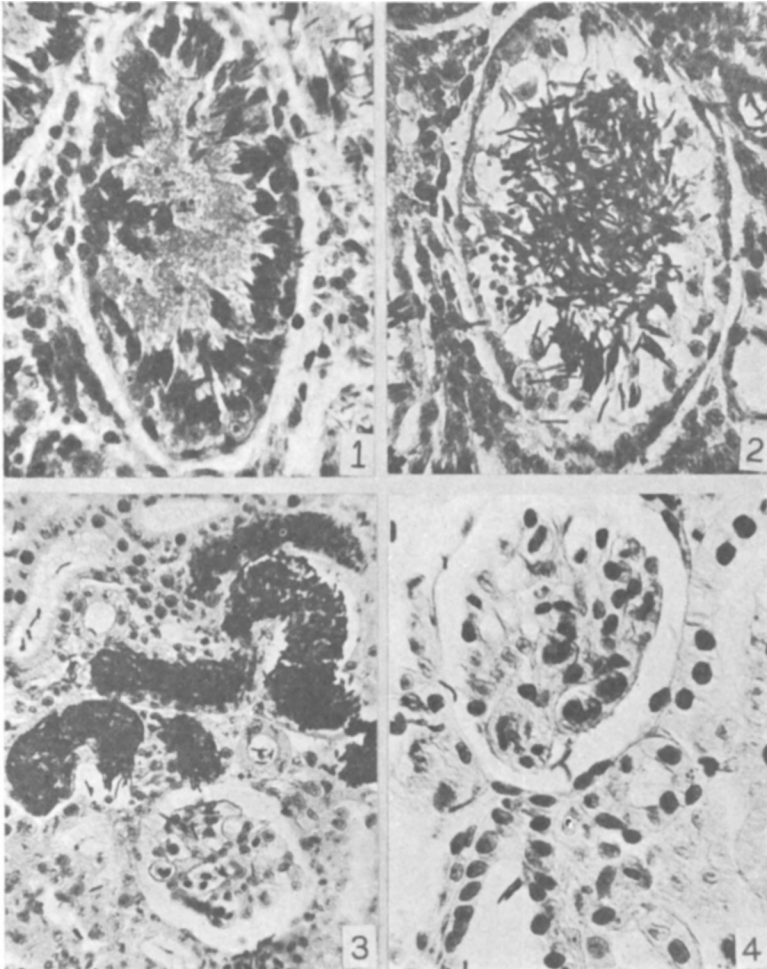


FIG. 1.

Seminiferous tubule from the right testis of an hibernating frog.

FIG. 2.

Seminiferous tubule from the left testis of the same frog (Fig. 1) 16 hours after the frog was injected with anterior pituitary hormone.

FIG. 3.

Section of kidney of frog 16 hours after anterior pituitary injection showing tubules filled with spermatozoa.

FIG. 4.

Highly magnified section of kidney showing Bowman's capsule and connecting uriniferous tubule, both containing spermatozoa.

anterior pituitary injection shows all of the mature sperm liberated into the lumen of the seminiferous tubule (Fig. 2). This effect can be achieved only by anterior pituitary treatment, the controls having received other frog organs and having been subjected to temperature changes from 4°C. to 28°C. The reaction is comparable to the follicle changes induced in females by anterior pituitary treatment<sup>5</sup> except that in the female the egg is released into the body cavity.

In sections of the kidney adjacent to the stimulated testis, spermatozoa may be seen in the uriniferous tubules, Bowman's capsule, and ureter (Figs. 3 and 4). The exact path of these spermatozoa, through the kidney, is being worked out for *Rana pipiens*, *Rana catesbiana*, *Bufo fowleri*, and *Hyla crucifer*.

Studies are at present being made to determine the source of the male gonad hormone, and the seasonal differences in maturation exhibited by *Rana pipiens* and *Rana catesbiana*. In the one there is a single breeding period, with a single expulsion of spermatozoa. In the other there seems to be an extended breeding period, at a different time of the year, with staggered maturation of spermatozoa.

## 9254

### Water Intake and the Blood Sugar Level.

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In our studies which have involved blood sugar determinations on rats, considerable effort has been expended to secure uniformity both in the animal material and in the routine care of the colony. But in spite of all this care to secure uniformity we have found at times that the normal blood sugar level showed considerable variation.<sup>1</sup> Recently, upon our return from the summer vacation, the routine normal control series of sugar determinations was made. At the same time, determinations were made upon a group of rats used in the study of prolonged high carbohydrate feeding.<sup>2</sup> These animals represented the 4th, 5th and 6th generations which had been kept continuously on the high carbohydrate diet. It was noticed that the sugar levels in both groups were not only high but far above

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<sup>5</sup> Rugh, R., *J. Exp. Zool.*, 1935, **71**, 163.

<sup>1</sup> Hrubetz, M. C., *J. Biol. Chem.*, 1934, **107**, 731.

<sup>2</sup> Hrubetz, M. C., *J. Lab. Clin. Med.*, 1936, **21**, 1142.