reached until 15 to 18 minutes later. Between heatings the temperature was allowed to drop gradually to a point sufficiently low for further treatment. The return to normal temperature required from an hour to an hour and a half. In myxomatous animals, the diseased condition was so advanced after 6 days that treatment was discontinued and death occurred from 2 to 3 days later. In animals infected with fibroma, daily treatments over a period of 2 weeks did not retard the progress of the disease. Again fever therapy had no effect upon the ultimate development of these 2 virus diseases in the animals.

While our report is, therefore, of a negative nature, it is important perhaps to point out that, under the conditions of our experiments, and with the 2 viruses mentioned, fever therapy is apparently without value. In the cases of these 2 viruses, therefore, we have contrast with certain bacterial infections (such as *Neisseria gonorrhœæ*) where fever therapy has been found efficacious. It would seem that at least some viruses are more resistant to induced high temperatures than are some bacterial forms.

9252 P

Effect of Estrogenic Substances in *Lebistes reticulatus* (Guppy).

PHILIP BERKOWITZ. (Introduced by Harry A. Charipper.)

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The adult male *Lebistes reticulatus* possesses, as its secondary sex characters, a gonopod (intromittent organ which is the anal fin in the female, and in young fish of both sexes) and various color patterns. The adult female possesses no such gonopod or colorations, but is much larger than the male. The average length of the male (from tip of snout to base of caudal fin) is approximately 18 mm.; that for the adult female is about 30 mm. (Goodrich, *et al.*,[1] confirmed by present observations). These differentiating secondary sex characters are not present at birth, at which time both male and female are identical in appearance. At about 35 days after birth there is a divergence in size of the 2 sexes, from which time the females normally become larger than the males. The gonopod of the male makes

[1] Goodrich, H. B., Dee, J. E., Flynn, C. M., and Mercer, R. N., *Biol. Bull.*, 1934, **67**, 83.

its first appearance at approximately 40 days, and the colorations from 50-60 days after birth.

In view of the scarcity of information concerning hormonic influences on these and related processes in the fish, an attempt has been begun, under the direction of Dr. Robert Gaunt,* to analyze such influences in the guppy. The first phase of this study, concerned with the effect of estrogenic substances, is here reported.

Sixty fish have been treated with Progynon tablets.† These tablets contained 45 R. U. of estrogenic substance, active by mouth in mammals, the exact chemical nature of which was not known. When crumbled into the aquaria (one tablet per 2-gallon tank, 3 times a week) the fish ate the tablets freely. The fish were fed on this hormone from birth for periods of one to 5 months. No male secondary sex characters appeared during the period of treatment although the fish had reached the size and age at which sexual differentiation should have been completed. The size and shape of these fish approached that of the female sex; the coloring and condition of the anal fin were entirely those of the female.

Fourteen fish were treated for 2 months and then placed in tanks with fresh water. Eight assumed male secondary sex characters 40 days after discontinuance of the hormone, *i. e.*, a period closely corresponding to the time necessary for normal males to attain sexual differentiation from birth. The absence of male secondary sex characters during treatment is not due to slow growth resulting from unfavorable conditions of treatment, for all treated males outgrew their controls and went well above the normal size for males. The average length of treated males was 26 mm. as compared to 18 mm. for controls. When hormone was discontinued treated males reverted to normal as regards the gonopod and the color conditions, but they did not revert to normal male size.

Of 5 fish treated for 4-5 months, all showed external sex characters of the female type. Four were killed; 3 possessed ovaries, and 2 also had young. The fourth had a gonad definitely not normal for either sex.

Examination of this gonad and the testes of other males treated for shorter periods showed a distinct effect of hormone treatment. There was a considerable suppression of spermatogenesis, and a relative increase in the number of young germ cells. In the latter

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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,[1] using *Bufo marinus* males, demonstrated that hypophyseal removal causes degeneration and hypophyseal implantation causes hypertrophy of the testes. O. M. Wolfe[2] implanted pituitary glands subcutaneously to induce amplexus in *Rana pipiens* and Rugh[3] 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[4] 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

[1] Houssay, B. A., and Lascano-Gonzalez, J. M., *Compt. rend. Soc. biol.*, 1929, **101**, 938.

[2] Wolfe, O. M., PROC. SOC. EXP. BIOL. AND MED., 1929, **26**, 692.

[3] Rugh, R., *Biol. Bull.*, 1935, **68**, 74.

[4] Rugh, R., *Biol. Bull.*, 1934, **66**, 22.