number by theelin to 8.5 per cross section, and cell height was raised from high cuboidal (in control males) to high columnar. No effect of theelin was distinguishable in the Wolffian duct of the female. In neither sex did theelin increase the diameter of the duct. Testosterone did enlarge the width of the female Wolffian duct slightly, and mitoses were increased to 17.1 per cross section. Testosterone produced similarly slight enlargement in the Wolffian duct of the male, but mitoses were only 11.0 per cross section.

The most marked effects of theelin and testosterone injection were seen in the oviduct. Theelin produced an increase of 40% in cross-sectional diameter, and of more than 100% in thickness of the wall. This increase was due to a proportional thickening of the mucosa and the mucous glands situated beneath the mucosa. Testosterone elicited an increase of 57% in the diameter of the oviduct, and 115% in the thickness of its wall. However, the increase in thickness of the wall was due almost entirely to enlargement of mucous glands, the mucosa appearing stretched over the increased surface. Evidence of secretory activity in theelin- and testosterone-treated oviducts, but not in controls, appeared in the form of a large amount of methyl green-staining (mucous?) material in the lumen. No increase in mitotic activity was apparent in any of the treated oviducts.

11061

Effects of Testosterone Propionate on the Female Viviparous Teleost, *Xiphophorus helleri* Heckel.

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Experimental sex-reversal and the study of intersexes in the vertebrates have been of fundamental importance in the analysis of the rôle of the genes and endocrine secretions in embryonic development, particularly in sex-differentiation.

Gallagher, *et al.*,[1] indicated that urine from adult men and women contains both sex hormones and that the ratio of androgen to estrogen is higher in the urine of males than it is in the urine of females. The results obtained by Callow[2] substantiated the work of Gallagher,

1 Gallagher, *et al., J. Clin. Invest.*, 1937, **16**, 695.

2 Callow, *Proc. Royal Society*, 1938, **31**, 841.

et al. Dingemanse, *et al.*,[3] reported findings that are not altogether harmonious with the work just cited but stated that the androgen-estrogen ratio was similar in the urines of the two sexes. This evidence indicates that each sex is hermaphroditic regarding the sex hormones, the difference being a quantitative one.

Since these differences of the sex hormones' content of the respective urines of the sexes are quantitative and not qualitative, and since they seem proportionately not to be very great, the study of the effect of the synthetically prepared male sex hormone, testosterone propionate,* upon the viviparous teleost, *Xiphophorus helleri* Heckel, was initiated to determine if, and to what extent, the relatively greater amount of one sex hormone checks the development and activity of the functioning sex tissue and accessory ducts of the other sex, and stimulates its own sex ducts and tissue to develop, and to learn if such development be caused by the establishment of a relatively greater amount of the male sex hormone in the female.

Essenberg[4] theorized, from histological examinations of 2 cases of natural complete sex-reversal observed in several hundred fish, that about 50% of all females *might* undergo sex-inversion, but did not necessarily *have* to do so. He concluded that any agent or condition which tends to decrease the capacity for female sex hormone secretion beyond a certain limit, became an immediate factor in the possibility of the sex-reversal in the female of the *X. helleri*.

Witschi and Crown[5] added testosterone to the water in the aquaria in which they kept pregnant *X. helleri* and obtained abortion and resorption of the young. In non-pregnant adult females, under the above conditions, all the large eggs underwent resorption. All treated females developed ovaries that resembled testes, but spermatogenesis was not reported, although the treated fish assumed gradually but completely the male secondary sex characteristics. Regnier,[6] using intra-muscular administration of the male sex hormone, reported that the sexual development of approximately one-third of her treated young female *X. helleri* developed male secondary sex characteristics.

This report involved experiments upon 91 virgin females and 7 normal males of the species. All of the subjects were past the un-

[3] Dingemanse *et al.*, *Biochem. J.*, 1937, **31**, 500.

* By the courtesies of Dr. G. Stragnell of the Schering Corporation, and of the Ciba Pharmaceutical Products, Inc.

[4] Essenberg, J. M., *Biol. Bull.*, 1926, **51**, 98.

[5] Witschi, E., and Crown, E. N., *Anat. Rec.*, 1937, **70**, 121.

[6] Regnier, M. T., *Bull. Biol. de la France et de la Belgique*, **79**, 385 pp.

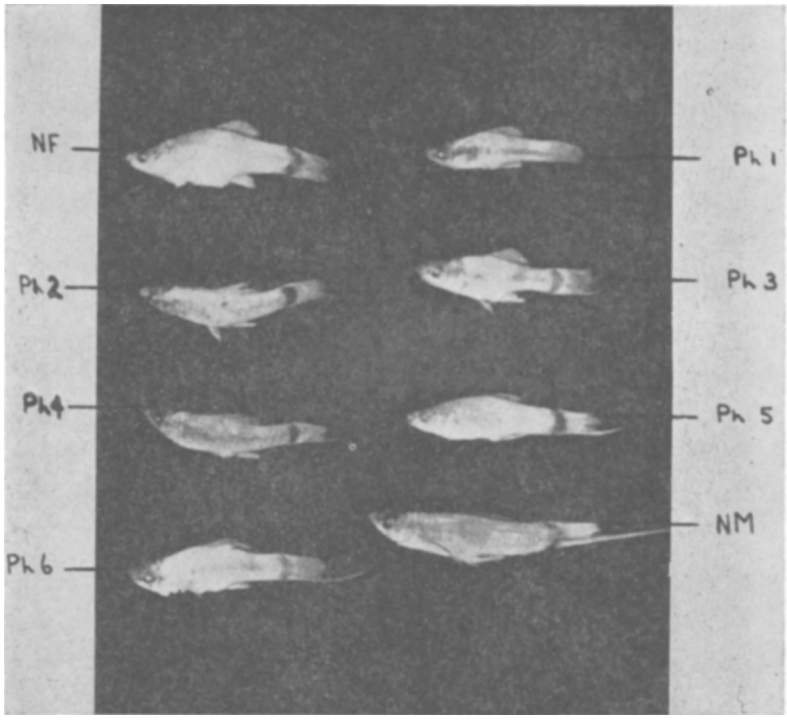

PLATE 1.

Progressive Phases of Development of the Artificially Induced Male Secondary Sex Characteristics in the Treated Females.

NF	Normal Female	NM	Normal Male
Ph 1	Phase 1	Ph 2	Phase 2
Ph 3	Phase 3	Ph 4	Phase 4
Ph 5	Phase 5	Ph 6	Phase 6

differentiated stage, with their sex being definitely established.

The testosterone propionate† was injected retroperitoneally, approximately 2 mm anterior to the anus, ventrally and laterally. Each fish received 0.5 mg testosterone propionate in 0.02 cc sesame oil weekly. The experiments extended over a period of approximately 19 weeks, with the progressive changes shown in Plate 1. The majority of treated fish reached phase 1 at an average time of 10.5 days after the initial injection; subsequent phases were reached on the averages of 21.9 days, 31.1 days, 45.4 days, 57.8 days, and 69.1 days after the initial injections, respectively.

† Withdrawn and used immediately from the original ampules furnished through the courtesies of the Schering Corporation and the Ciba Pharmaceutical Products, Inc.

Controls of several types were used. First, one-sixth of the total number of virgin females were left absolutely untouched. Second, a similar number of virgin female fish were treated only with pure sesame oil Third, normal males of the species were used as the basis of comparison of the extent of morphological and histological transformations expressed in the degree of apparent sex-reversal of the experimentally treated fish.

Not only does the experimental administration of testosterone propionate cause the regular morphological changes (Plate 1) in all

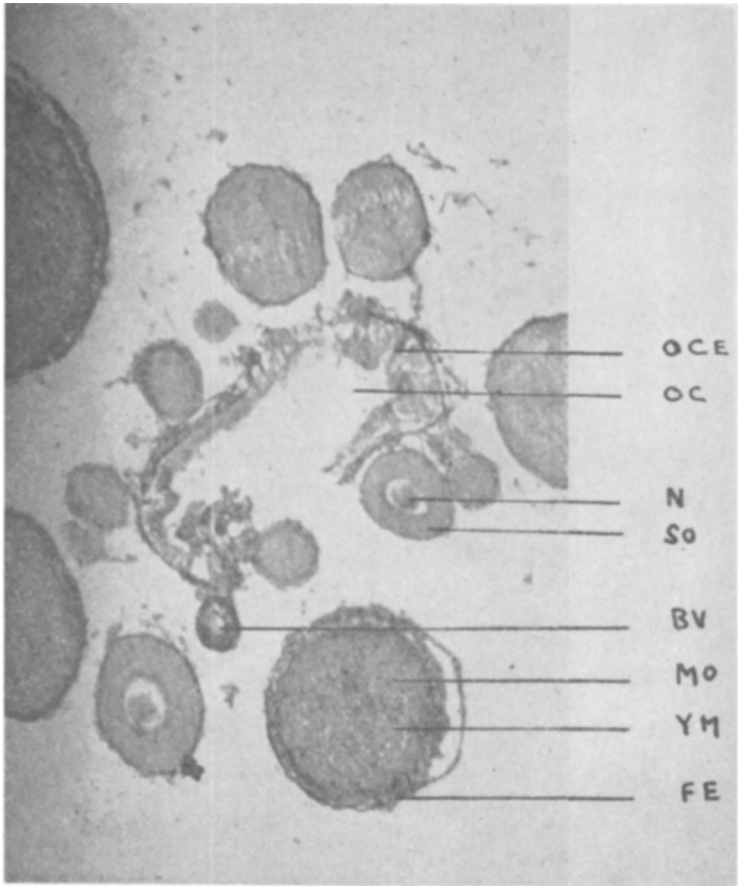

PLATE 2.

Cross-section of treated female gonad: resorption. BV, Blood Vessel. FE, Follicular Epithelium. MO, Medium-sized Oöcyte. N, Nucleus. OC, Ovarian Cavity. OCE, Ovarian Cavity Epithelium. SO, Small-sized Oöcyte. YM, Yolk Material.

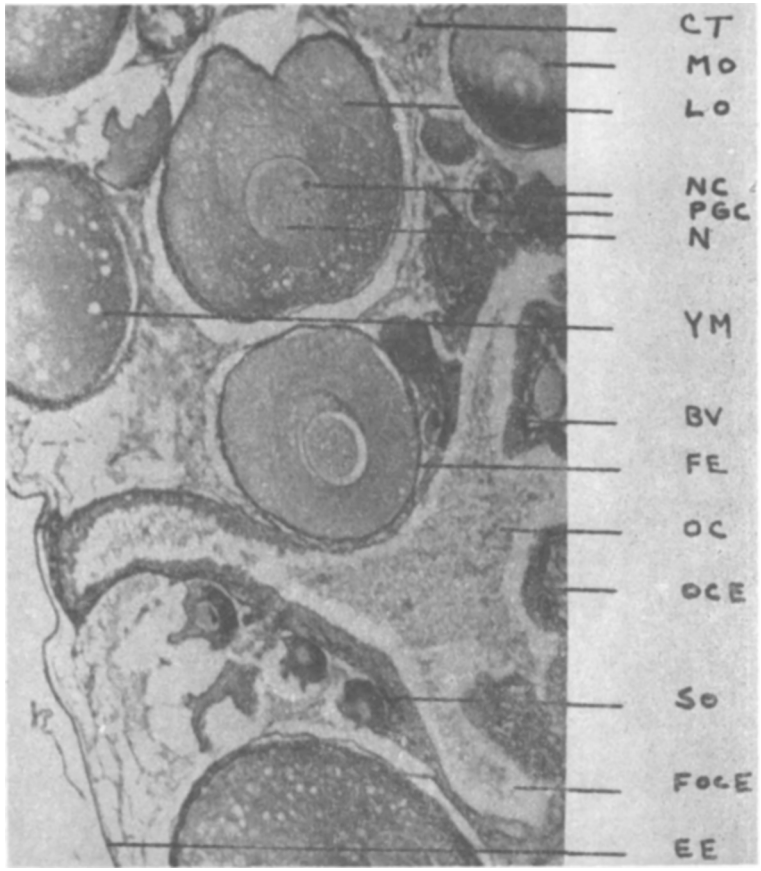

PLATE 3.

Cross-section of normal female gonad. BV, Blood Vessel. CT, Connective Tissue. EE, External Epithelium. FE, Follicular Epithelium. FOCE Folds of Ovarian Cavity Epithelium. LO, Large-sized Oöcyte. MO, Medium-sized Oöcyte. N, Nucleus. NC, Nucleolus. OC, Ovarian Cavity. OCE, Ovarian Cavity Epithelium. PGC, Primordial Germ Cells. SO, Small Oöcyte. YM, Yolk Material.

cases to simulate the male in body color, formation of a caudal sword, and the development of the anal fin into the copulatory organ, the male gonopod, but these changes are accompanied by histological changes in the primary sex organs in approximately 50% of the experimentally treated virgin female fish. Females which respond to such treatment in these experiments are characterized as showing either (a) resorption of the gonad (Plate 2), or (b) some phase of spermatogenesis. Individuals which are completely altered histologically apparently pass from normal female gonadal structure

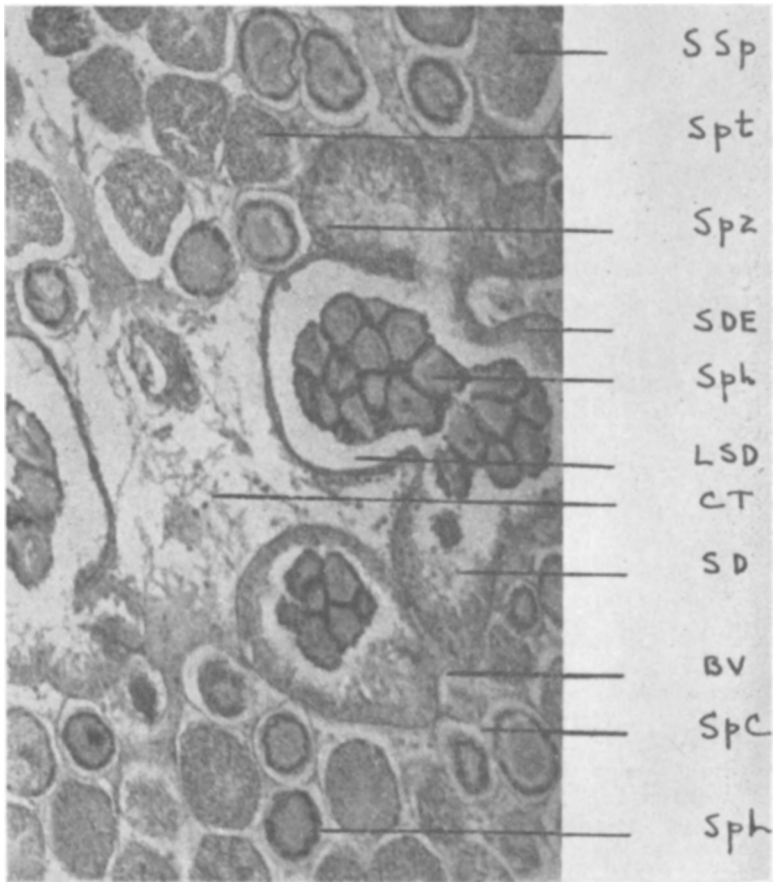

PLATE 4.
Cross-section of treated female gonad: Spermatogenesis, Phase 6. BV, Blood Vessel. CT, Connective Tissue. LSD, Lumen of Sperm Duct. SD, Sperm Duct. SDE, Sperm Duct Epithelium. SpC, Spermatocyst. Sph, Spermatophores. Spt, Spermatids. Spz, Spermatozoa. SSp, Secondary Spermatocytes.

(Plate 3) through progressive resorption stages and finally show histological features (Plate 4) resembling those of the normal male gonad (Plate 5). These results substantiate Essenberg's[4] postulation, previously mentioned. It is thus to be concluded that the administration of the esterified androgen, testosterone propionate, to virgin female *Xiphophorus helleri* Heckel, causes sex-reversal in approximately 50% of the experimentally treated animals. However, it is not to be concluded that the appearance of changes in secondary sex characteristics are to be taken as a certain index of complete sex-reversal in this form.

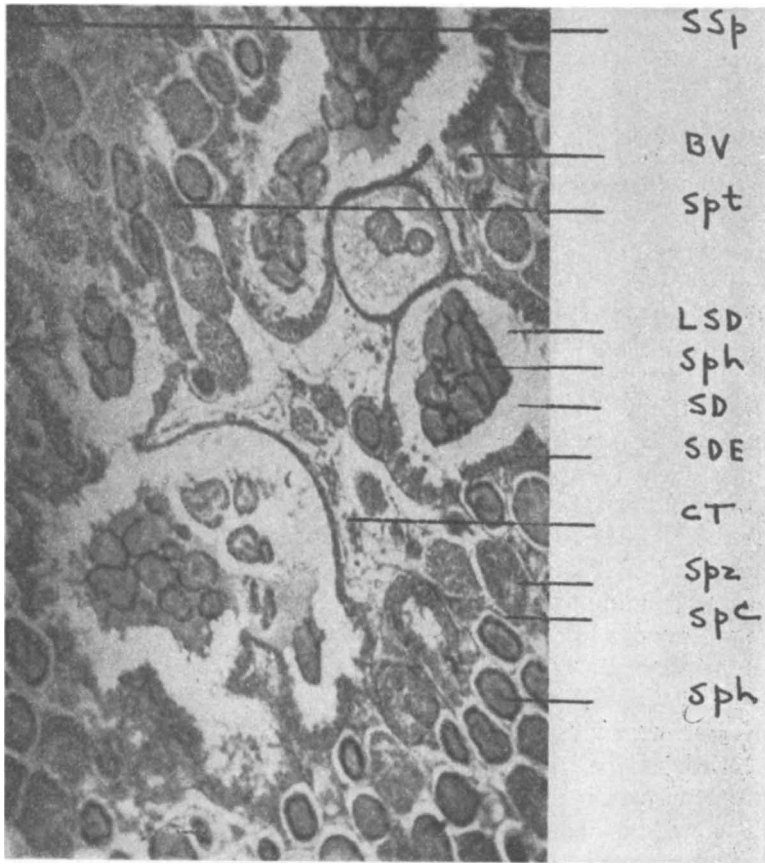

PLATE 5.

Cross-section of Control Male Gonad. BV, Blood Vessel. CT, Connective Tissue. LSD, Lumen of Sperm Duct. SD, Sperm Duct. SDE, Sperm Duct Epithelium. SpC, Spermatocyst. Sph, Spermatophores. Spt, Spermatids. Spz, Spermatozoa. SSp, Secondary Spermatocytes.