

Gonadotropic Hormones of the Hypophysis of the Turkey.*

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Relatively little work has been done in the past on the hypophysis of birds. Benoit¹ obtained evidence for the presence of gonadotropic hormones in hypophyses of sexually active ducks but not in those of inactive ones. Similarly Riddle and Schooley² found no effects from implanted hypophyses of pigeons less than 1.8 months old, while those of mature pigeons gave definite indication of the presence of some gonad stimulating principle. Since birds develop no true corpora lutea and differ also otherwise in their physiology of reproduction markedly from mammals, it seemed desirable to make a more complete, qualitative and quantitative analysis of their gonadotropic hormones.

Through the courtesy of Mr. A. C. Gingerich, we were permitted to collect about 6000 hypophyses at the packing plant of the Maplecrest Turkey Farms at Wellman, Iowa. The glands were taken from birds about 6 months old, the hens weighing around 10 and the gobblers around 18 pounds. The fresh glands weigh about 13 mg. and 25 mg. respectively. However, the male glands lose more weight in the process of acetone-drying. One day's collection yielded 6.978 gm. dry powder from 1655 female glands and 9.900 gm. from 2047 male glands.

The dry powder taken up in distilled water was assayed in 4 different tests. Table I gives the results obtained from injections into immature 21 day old rats. The largest dose (100 mg.) produces large ovaries which usually contain about as many corpora lutea as mature follicles. (However, in several cases the corpora were more numerous while in one case they were absent.) In the second group (50 mg.) the increase in ovarian weight is almost entirely due to the presence of many stimulated and mature follicles. A few incomplete corpora lutea are found in 3 cases. The third group (25 mg.) has slightly increased ovaries that contain only few mature or distinctly enlarged follicles. However, sections through the vagina prove that enough estrogenic hormones were released to produce

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¹ Benoit, J., *C. Rend. Soc. Biol. Strasbourg*, 1935, **118**, 672.

² Riddle, O., and Schooley, J. P., *PROC. SOC. EXP. BIOL. AND MED.*, 1935, **32**, 1610.

full cornification in the vaginal epithelia. The lowest dose (10 mg.) produces no visible morphological change in the ovary though it stimulates the endocrine apparatus, as can be seen from the development of the vaginal epithelium. The vaginal stage was in every

TABLE I.

Assay of acetone-dried turkey hypophyses on 21-day rats. The total amount given in the first column was administered in 6 injections during 3 days. The recipients were killed 24 hours after the last injection (except the animals showing vaginal stage 4, which were killed one day later). F, mature follicles; f, enlarged immature follicles; L, corpora lutea; l, luteinizing follicles.

	Hypophysis injected mg.	Wt. of fresh ovaries, mg.	Aver.	Ovarian histology	Vaginal stage
Male	100	19.3	25.1	FL	3
"	"	30.9	37.3	F-	3
Female	"	28.5		33.1	FL
"	"	37.7	48.2	FL	3
Mixed	"	37.4		36.1	FL
"	"	36.1	71.2	F	3
"	"	71.2		48.2	FL
Male	50	28.8	27.7	F	3
"	"	27.6		27.7	Fl
"	"	32.9	23.6	Fl	3
"	"	21.8		18.1	F
Female	"	13.5	18.1	F	3
"	"	25.0		18.1	F
"	"	15.5	26.3	F	3
"	"	18.6		26.3	F
Mixed	"	24.6	28.0	Fl	4
"	"	28.0		26.3	F
Male	25	14.2	14.8	f	3
"	"	14.8		14.8	f
"	"	15.4	17.2	f	3
Female	"	15.0		14.8	f
"	"	13.5	21.9	f	3
"	"	16.1		21.9	f
Mixed	"	20.4	21.9	f	3
"	"	31.1		21.9	f
"	"	14.2	11.5	f	3-4
"	"	14.2		11.5	f
Male	10	12.5	12.7	-	1-2
"	"	10.6		12.7	-
Female	"	14.3	12.1	-	1-2
"	"	11.2		12.1	-
Mixed	"	10.5	12.2	-	1-2
"	"	13.0		12.2	-
"	"	13.0	13.3	-	1
"	"	13.0		12.2	-
Controls		12.4	13.3	-	0
"	"	14.9		-	0
"	"	12.9		-	0
"	"	15.7		-	0
"	"	13.1		-	0
"	"	11.8		-	0
"	"	13.9		-	0
"	"	12.2		-	0

case ascertained by cross sections. The vagina remained closed even in rats that had received the highest dose. Ovulation was never observed. All corpora lutea that were investigated contained eggs, often fragmented in 2 or more pieces. All 4 dosages cause also enlargement of the uteri. At the lowest dose it is already quite definite; at 25 and 50 mg. the uteri are often widely distended while the highest dose produces greatly thickened uterine walls.

TABLE II.

Assay of acetone-dried turkey hypophysis on 28-day rats. The total amount given in the first column was administered in 6 injections during 3 days. The rats were killed 24 hours after the last injection.

Hypophysis injected mg.		Wt. of fresh ovaries, mg.	Aver.	Ovarian histology	Vaginal stage
Mixed	50	58.0	60.9	FL	4
"	"	57.9		FL	4
"	"	66.8		FL	4

TABLE III.

Assay of acetone-dried turkey hypophysis on hypophysectomized rats operated 2 weeks prior to beginning of injections which were given over 5 days in 10 equal amounts.

	Hypophysis injected, mg.	Wt. of fresh ovaries, mg.	Ovarian histology	Vaginal stage	Wt. in gm. at hypophysectomy	Wt. in gm. at autopsy
Male	100	39.8	FL	3	101.0	96.3
"	"	22.6				

In a second test the same powder was assayed on 28-day-old rats. While the ovaries of controls have not enlarged during the fourth week, they have become much more responsive; injections of 50 mg. give nearly 3 times as heavy ovaries as in the 21-day rat. The vaginae open and show stage 4 at the end of the fourth day. Some follicles ovulate, though most of them luteinize while they still enclose the egg.

It is obviously not easy to say just what amount of powder should be considered as having the potency of one rat unit. Since it is customary to use as a test animal the 21-day-old rat in which a distinctly recognizable reaction in ovary, uterus and vagina is produced by 25 mg. (Table I), we may say that this dosage contains one (total) rat unit. It represents the contents of about 6 turkey pituitaries. In view of the work of Benoit and Riddle, one has to consider the possibility that the hypophyses of our birds, which were killed at the very beginning of their first reproductive season, were not yet at the peak of productivity.

The third test was added to make sure that the luteinizing principle was contained in the hypophyseal powder. Two hypophysectomized rats were injected with 50 mg. of turkey pituitary powder in 2 daily injections extending over 5 days. Hypophysectomy had preceded the injections by 2 weeks. Throughout this period the animals remained in dioestrus. On the fourth day of injections, the vaginal smear changed to 3 and remained so until the sixth day, when the animals were killed. At this time the ovaries contained numerous mature follicles, some of them ovulated, and all showed stages of progressive luteinization. The uteri are greatly enlarged. This test removes any possible doubt concerning the actual presence of both the follicle-stimulating and the luteinizing principle in the bird hypophyses. It is of no small interest to note that the gonadotropic hormones are of nearly the same nature in birds and in mammals though the reactions which they control are quite different in the 2 taxonomic classes.

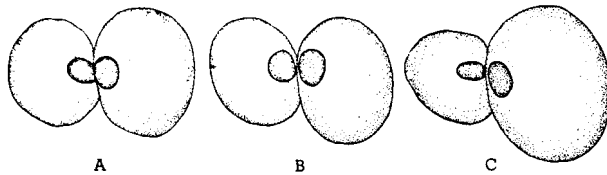


FIG. 1.

Camera lucida drawings of testes of 3 sparrows stimulated by injection of acetone dried, powdered turkey hypophyses, and (inside outlines) of 3 control sparrows which were injected with serum of a pregnant woman, without effect; $\times 2$. Total amounts injected: *A*, 85 mg. turkey hypophysis (15 days), resp. 4 cc. serum p. w. (16 days). *B*, 90 mg. turkey hypophysis (15 days), resp. 4 cc. serum p. w. (16 days). *C*, 130 mg. turkey hypophysis (12 days), resp. 8 cc. serum p. w. (16 days).

In a fourth test the effect of the turkey hypophyses on adult sparrows in the state of sexual quiescence was studied. This condition is characterized in the male by small and inactive testes, thin and only slightly convoluted vasa deferentia and light brown bills, in the female by smallness of the ovaries and oviducts.³ Six sparrows, 3 males and 3 females, were injected with various dosages of powdered hypophyses over periods of 11 to 15 days. The total amounts (85 to 130 mg.) are equivalent to 3.5-5 rat units. The extensive effects on gonads and secondary sex characters are partly represented in Figs. 1 and 2. The left testes of the males are enlarged at least 60 to 120 times. Spermatogenesis is in full progress; the seminal tubules contain masses of mature spermatozoa. The vasa deferentia are slightly convoluted and enlarged. The bills have turned

³ Witschi, E., *Wilson Bull.*, 1935, **47**, 177.

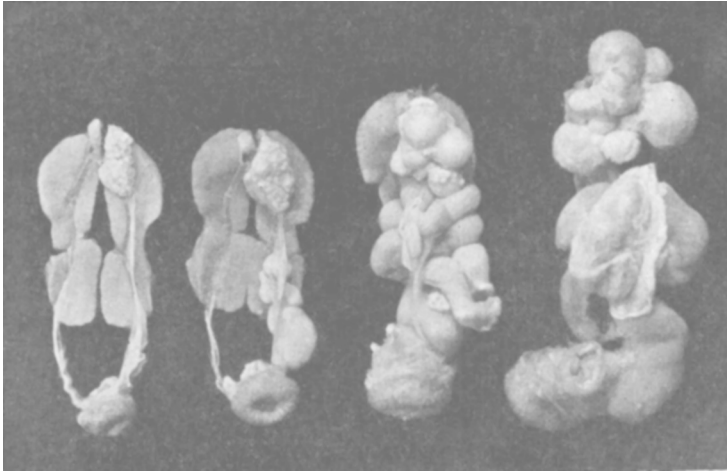


FIG. 2.

Urogenital apparatus of 4 female sparrows, phot. $\times 1.5$. The first one illustrates the quiescent condition, ovary and oviduct covering a minor part of the left kidney; it can serve as control, since the injection of 8 cc. serum p. w. (16 days) has not caused any change. The second and the third received 90 mg. of turkey hypophysis (in 15 days); ovaries and oviducts greatly enlarged. The fourth one received 110 mg. of turkey hypophysis (in 11 days); ovary and oviduct, enlarged to full breeding size, cover the kidneys entirely.

dark blue (stage 2, Witschi and Woods⁴). The corresponding changes in ovaries and oviducts of the females appear clearly in Fig. 2. Control animals were injected with 4-8 cc. of serum of a pregnant woman. This serum contained about 100 rat units of gonadotropic principle per cc. It had no stimulating influence on either gonads or secondary sex characters (Figs. 1 and 2).

From our experiments we can conclude that powdered turkey hypophysis is in quality very similar to corresponding products from cattle, sheep or rats. If equal weights of these powders are compared, the turkey hypophyses appear more potent than bull but less potent than sheep or rat hypophyses. Male and female hypophyses of the turkey show no marked sexual differences in our tests.

⁴ Witschi, E., and Woods, R. P., *J. Exp. Zool.*, 1936, **73**, 445.