Hypophyseal Nucleic Acids: Castration and Thiouracil[1] (34347)

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Pituitary gland mass is influenced by age and hormonal imbalances but little is known about potentially associated changes in nucleic acids. Kraicer and Cheng (1) found that castration and hypothyroidism increased cell numbers (DNA) and cell size (p/DNA) in the anterior hypophysis of adult male rats maintained under constant light. However similar information on the immature rat under light–dark conditions and subjected to castration, thiouracil feeding or both has not been reported.

Materials and Methods. Immature male Long-Evans rats from our colony were maintained on a 12-hr light, 12-hr dark cycle. The animals were fed a semipurified diet containing 20% casein (2), with or without thiouracil (0.1%) starting at 25 days of age, and were killed by decapitation at either 50 or 65 days of age. Addition of 0.1% thiouracil to the diet fed does not influence food intake (3). Castration was performed at 23 days of age to permit a 2-day recovery prior to starting the experiment. Pituitaries (anterior and posterior lobes) were removed immediately following necropsy and homogenized in 2 ml of distilled water. Single pituitary homogenates were then frozen and later analyzed for protein (4) and nucleic acids (5). Significant differences between experimental and control rats of the same age were estimated by the Student's t test at the 95% confidence level.

Results. Pituitary gland mass was significantly greater in rats gonadectomized at 23 days of age and examined 27 days later when compared to intact controls (Table I). Increase in mass was accompanied by an increase in total protein and cell size (protein/DNA) but total DNA and RNA did not change. Thiouracil feeding (0.1%) was without effect in intact rats, but the drug significantly enhanced the influence of castration on pituitary weight and protein content.

When rats were sacrificed at 65 days of age further pituitary growth was noted and total protein increased in normal rats. The gonadectomized rat exhibited a greater increase in pituitary mass and total protein, but contrary to the 50-day-old rats a cell size increase did not occur but an increase in cell number (DNA) was noted. Thiouracil alone had no significant effect in the intact rat, but again enhanced the effect of castration on hypophyseal mass and total protein.

Discussion. Hypophyseal hypertrophy and/or hyperplasia is an eventual consequence of castration or thyroidectomy but the feeding of antithyroid drugs has provided varied results (6). Castration is followed by an increase in hypophyseal and serum FSH and LH (7, 8) suggesting a modified protein synthesis and indeed the rate of incorporation of alanine (9) but not of leucine is increased (10). Castration of the immature male rat caused significant hypophyseal growth as shown by protein deposition. Initially cell size increased (protein/DNA) but the subsequent gland growth was associated with an increase in cell number. In adult rats, (1) castration was followed by an increase in cell size and cell number which may reflect the difference in anabolic potential between the immature and mature rat.

It has been assumed that polyploidy does not occur in the pituitary, or occurs in only a small percentage of hypophyseal cells (11–13). Our data support this view in that no increase in total DNA occurred in the control animals between 50 and 65 days of age, but marked changes did occur in castrated rats.

[1] Supported by USPHS Grant AM 00462.

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TABLE I. Influence of Thiouracil and Castration on Hypophyseal Protein and Nucleic Acids.[a]

	Hypophyseal wt. (mg)	Total protein (µg)	Total DNA (µg)	Protein/DNA	Total RNA (µg)	RNA/DNA
Rats necropsied at 50 days of age						
Controls (12)[b]	5.9 ± 0.2	742.2 ± 30.8	26.8 ± 2.9	30.7 ± 3.0	68.4 ± 3.6	2.9 ± 0.4
Thiouracil (12)	6.3 ± 0.2	757.0 ± 28.6	30.6 ± 3.0	26.4 ± 2.0	75.1 ± 6.8	2.7 ± 0.3
Castrated (10)	7.7[c] ± 0.4	922.9[c] ± 53.4	23.3 ± 2.6	43.4[c] ± 4.9	83.0 ± 7.3	4.1 ± 0.6
Castrated + Thiouracil (12)	9.0[d] ± 0.3	1148.9[d] ± 88.7	22.2 ± 3.0	59.0[c] ± 6.5	86.0[c] ± 3.8	4.7[c] ± 0.7
Rats necropsied at 65 days of age						
Controls (12)	7.0 ± 0.3	860.6 ± 36.8	26.2 ± 2.5	36.6 ± 4.6	72.7 ± 3.8	3.1 ± 0.4
Thiouracil (12)	7.5 ± 0.2	923.8 ± 54.8	29.7 ± 2.8	33.8 ± 3.5	66.7 ± 2.5	2.5 ± 0.2
Castrated (12)	8.8[c] ± 0.3	1095.0[c] ± 35.7	36.4[c] ± 2.9	31.2 ± 1.5	71.8 ± 2.6	2.1[c] ± 0.1
Castrated + Thiouracil (11)	10.4[d] ± 0.5	1265.1[d] ± 49.8	44.6[c] ± 2.8	29.0 ± 1.3	84.5 ± 6.2	1.9[c] ± 0.1

[a] Values are means ± standard error of the mean.
[b] Number of animals used for determinations, one hypophysis/determination.
[c] Significantly different from control at 95% confidence level.
[d] Significantly different from castrated at 95% confidence level.

Antithyroid drug administration to female rats caused a decrease in hypophyseal TSH and an increase in serum TSH but the pituitary gland mass decreased (6). However, synthesis and release of other pituitary hormones was reduced (14, 15). Thus protein anabolism and catabolism for the gland as a whole may be unchanged despite evidence for increased amino acid incorporation into pituitary tissue (9, 10, 16) and increased incorporation of uridine into RNA following thyroidectomy (11). Indeed our results failed to reveal an effect of thiouracil on pituitary gland protein or nucleic acid content. However, the adult rat pituitary did exhibit an increase in nucleic acids in response to propylthiouracil (1). Clearly the 0.1% level of thiouracil fed to immature male rats produced only a partial block of thyroid hormone production and may contribute to the failure to simulate the results in adult male rats; dosages have not been investigated. Age at treatment may also be a factor since 0.5% thiouracil fed to immature female rats for 30 days also failed to increase pituitary weight (17).

Hypophyseal weight response to ovarian ablation was augmented by concomitant thiouracil (17) and cytological studies indicate a cellular response to both endocrine modifications. Contopoulos *et al.* (18) found that castration plus thyroidectomy led to increased plasma levels of FSH, LH and TSH. Thiouracil feeding to the castrated immature male augmented the effect of castration alone on pituitary weight and total protein but cell size and cell numbers were not increased. Thiouracil and castration appear to complement each other by influencing different mechanisms. Thus thiouracil increases oxygen consumption in the gland but castration does not (19). Thymidine incorporation into DNA was increased by castration but not by thyroidectomy (20) whereas the opposite responses were noted when uridine incorporation into RNA was examined (11).

Summary. Increased hypophyseal mass which follows castration in immature male rats, was due to cellular hypertrophy at 50 days of age and hyperplasia at 65 days of age. Thiouracil (0.1%) had no significant

effect on hypophyseal protein, RNA, or DNA in the immature rat fed the drug for 40 days. However thiouracil enhanced some of the effects of castration on the pituitary.

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Received June 12, 1969. P.S.E.B.M., 1969, Vol. 132.