brain tissues, they do not exhibit prolactin and ACTH releasing ability possessed by the brain tissues. It is not known whether these brain tissues acted directly on the anterior hypophysis or indirectly through other centers. Preliminary assays of bovine hypothalamic and cerebral cortical tissues‡ show that these may also induce mammary secretion in rats.

Summary. Rat tissue containing the hypothalamus, cerebral tissue, kidney or liver were injected subcutaneously in saline suspension into rats for 5 days, after they had previously been injected with estradiol for 10 days to develop their mammary glands. Saline alone was injected into control rats. The tissue containing the hypothalamus initiated lactation in 12 out of 15 rats; cerebrum in 4 out of 15 rats; and liver, kidney or saline in none out of 5 rats each. Tissue containing the hypothalamus was injected intradermally over crop sacs of 5 pigeons for 5 days and failed to induce a response, indicating that the active principle is not prolactin. It is suggested that rat hypothalamus and possibly cerebral tissue produce release of prolactin and probably ACTH from the anterior pituitary in amounts sufficient to initiate lactation.

‡ Actone-dried preparations of these tissues were kindly supplied by Dr. C. E. Graham, Wilson Labs., Chicago.

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Pituitary ACTH Levels During Adrenal Involution Following Thiouracil.* (25496)

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Adrenal atrophy following administration of goitrogen thiouracil has been amply demonstrated(1,2,3,4) but the causal mechanism has not been fully elucidated. Atrophy of the adrenal cortex after thiouracil and inability to obtain adrenal hypertrophy following exposure to cold in thiouracil fed rats led Zarrow and Money(5) to conclude that adrenal involution was due either to decreased ACTH production by the pituitary or decreased sensitivity of the adrenal to ACTH. Subsequent studies by Zarrow and Zarrow(6) showing that the atrophic adrenal gland of thiouracil treated rat retains its sensitivity to ACTH were interpreted to indicate that adrenal involution in thiouracil fed rats is caused by decreased ACTH secretion. In the present experiment levels of pituitary ACTH were measured in rats showing adrenal involution induced by feeding thiouracil.

Materials and methods. Normal male albino rats, descendants from Sprague-Dawley strain weighing 150 ± 10 g were separated into 2 groups. Ten rats were kept as untreated controls and were fed Rockland rat

TABLE I. Mean Body and Fresh Organ Weights of Male Rats Fed a Diet Containing 0.3% Thiouracil for 3 Weeks.

Group	Body wt, g	Fresh organ wt, mg/100 g body wt		
		Thyroid	Adrenals	Pituitary
Thiouracil	196 ± 6*	44.7 ±2.8	13.1 ± .2	4.6 ±.3
Controls	246 ±17	7.2 ± .7	18.3 ±1.2	2.8 ±.3

* Stand. error of mean. All differences significant at P .05.

* The author gratefully acknowledges technical assistance of William Watson and Joan Michalski.

TABLE II. Mean Pituitary Weights and ACTH Content of Male Rats Fed a Diet Containing 0.3% Thiouracil for 3 Weeks.

	Pituitary wt				Pituitary ACTH		
	mg		mg/100 g body wt		U.S.P. milliunits		
Group	Fresh	Dry*	Fresh	Dry	Per mg fresh	Per mg dry	Per pituitary
Thiouracil	9	1.6	4.6	.8	10.5	59	94
Controls	7	1.4	2.8	.6	39.4	187	262

* Acetone-dried.

diet and tap water *ad lib*. Six other rats were fed Rockland rat diet containing 0.3% thiouracil (Schwarz Labs) and tap water *ad lib*. All animals were kept in quarters with controlled temperature (78 ± 2°F) and humidity (60 ± 5%). At the end of 3 week experimental period, animals were weighed and sacrificed with ether. Immediately thereafter, their pituitary glands were removed, weighed on torsion balance, and quickly placed in cold acetone. Thyroids and adrenals were also weighed at autopsy. After 24 hours in acetone pituitary glands were air dried at room temperature, reweighed and pulverized. For ACTH assay, the pituitary powder from thiouracil or control rats was suspended in 0.9% saline and assayed by intravenous U.S. Pharmacopeia (USP) ACTH assay against USP Corticotropin Reference Standard.

Results. Mean body and fresh organ weights after 3 week experimental period are shown in Table I. Pituitary weights and ACTH contents are summarized in Table II.

Thiouracil rats showed an increase in fresh thyroid and pituitary weights over untreated controls, but no significant difference in acetone-dried pituitary weights was evident. This indicates that the increase in fresh pituitary weight of thiouracil rats was largely due to higher water and/or fat content.

Body weights, adrenal weights and pituitary ACTH levels were lower in the thiouracil rats. Pituitary ACTH content of thiouracil rats was one-third or less than that of controls whether expressed in terms of units/mg of fresh weight, dry weight, or total units/pituitary gland.

Discussion. Adrenal-thyroid interaction was first suggested by Hoskins(7) when he reported adrenal hypertrophy after treatment with thyroid extract. Since then, adrenal atrophy following thyroidectomy(4,8) or treatment with thiouracil(1-4,9) has been shown in various species, but the mechanism of interaction has remained obscure.

When the hypothyroid state is induced by surgical thyroidectomy or thiouracil (a form of chemical thyroidectomy), the following hypotheses may be considered as far as adrenocortical atrophy and hypofunction(9,10): 1) A metabolic effect on adrenals due to lack of thyroxine, 2) A decrease in ACTH secretion by direct effect or by shift in pituitary production from ACTH to TSH. Since adrenal atrophy does occur following surgical thyroidectomy in absence of thiouracil and the condition can be corrected by small simultaneous doses of thyroxine(4), a direct toxic effect by goitrogen is unlikely.

The present findings of decreased pituitary ACTH levels following thiouracil strongly support the hypothesis first suggested by Zarrow and Zarrow(6), namely, that adrenal atrophy of the thiouracil rat is caused by decreased ACTH secretion. It may be, of course, that low levels of both thyroxine and ACTH are responsible for the adrenal involution effect, since lack of circulating thyroxine could quite conceivably aggravate the effect of low ACTH secretion.

Conclusions. Feeding rats a diet containing 0.3% thiouracil for 3 weeks brought about adrenal atrophy and thyroid and pituitary enlargement as compared to non-thiouracil controls. Pituitary ACTH content of thiouracil rats was less than ⅓ that in controls. The data support the hypothesis that adrenal atrophy following thiouracil is caused by lowered ACTH titers.

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Effect of N-(1-Methyl-2,3,di-p-chlorophenylpropyl)-Maleamic Acid (Benzmalecene) on Serum Lipids and Lipoproteins.* (25497)

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Among hypocholesterolemic agents recently made available for clinical trial is N-(1-methyl - 2,3,di-p-chlorophenylpropyl) - maleamic acid, Benzmalecene,† said to inhibit incorporation of 1-C^{14} acetate and 2-C^{14} mevalonic acid into cholesterol by rat liver homogenates, to inhibit development of hypercholesterolemia in chickens fed a diet containing 2% cholesterol and 5% cottonseed oil, to reduce the magnitude of cholesterolemia in rats fed a natural diet to which saturated fat was added, and to reduce serum cholesterol levels in dogs maintained on standard laboratory rations(1). We administered Benzmalecene to one hypercholesterolemic human subject and 2 normocholesterolemic subjects and noted hypocholesterolemic effects in each. Unusual changes in serum lipid pattern were induced.

Methods. Serum total and unesterified cholesterol and phospholipid concentrations and cholesterol and phospholipid content of serum lipoprotein fractions obtained by differential preparative ultracentrifugation were determined by methods we previously reported(2). "Phospholipids" were calculated by multiplying lipid P values by 25. Serum triglycerides were determined by the method of Van Handel and Zilversmit(3). Benzmalecene was administered to the following subjects: (1) PB a 39 year old woman with familial hypercholesterolemia who received 1 g/day for 7 days followed by 1.5 g/day for next 7 days; (2) ABB a 41 year old man with Klinefelter's syndrome who received 1.5 g/day for 10 days; (3) GM a 27 year old man with myotonic dystrophy and hypogonadism who received 1.5 g/day for 12 days. Both pre- and post-treatment control sera were studied. Post-treatment sera were obtained beginning 12 to 21 days after cessation of treatment.

Results. No change in serum transaminase (glutamic-oxaloacetic and glutamic-pyruvic) or van den Bergh levels were noted during the relatively short period of study. (Bromsulphalein retention has been noted in chronic toxicity studies in animals(1)). The compound uniformly produced anorexia or nausea which prompted one subject to refuse further medication after 12 days of treatment. No dietary restrictions were imposed and no changes in weight were noted. Since weight loss did not occur, the changes in serum lipids are not attributable to reduced caloric intake. The influence of anorexia *per se* on serum lipids is unknown.

The effects on serum lipids were similar in all subjects (Table I). The decrease in serum total cholesterol was limited to esterified cholesterol. Analysis of the ultracentrifugally separated lipoprotein fractions of densities <1.063 g/ml (includes "β" and all lower

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