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EFFECT OF EXPERIMENTALLY PRODUCED HYPERTHYROIDISM UPON THE REPRODUCTIVE AND ASSOCIATED ORGANS OF THE MALE RAT

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I. Introduction

The existence of a relationship between the thyroid gland and the development and functioning of the reproductive organs has been the subject of considerable research for the past several decades. Experimental findings have, however, until recently been too disharmonious for any definite conclusions to be drawn.

The problem has been attacked by ablation of the thyroid gland and by feeding of thyroid substances. Ablation is said by most investigators to have an injurious effect upon the reproductive organs. Tatum '13 reported gonad degeneration in male and female rabbits thyroidectomized early in life. Kunde, Carlson and Proud '29 reported similar results in female rabbits, and found in addition that the ovary could be restored to its normal condition by thyroid feeding. Hammett '26 found no specific relation between thyroid activity and the growth of the reproductive organs prior to puberty.

The experiments involving thyroid feeding have shown widely varied results. Gudernatsch '15 showed that rats fed rather large doses of fresh thyroid tissue exhibited pronounced loss in weight, diarrhea and muscular emaciation and, in most cases, finally died. Administration of doses which kept the animals in apparently good health resulted either in failure to breed, or if the thyroid-fed males were bred to normal females, the young died at birth

or remained stunted in their growth. Courrier '21, however, reported neither a stimulating nor a deleterious effect upon the reproductive organs following thyroid feeding. The dose used by him was probably quite small since his animals gained in weight. Belawenetz '28 noted that thyroid feeding resulted in a testicular degeneration and a loss of potency in male rats. Döderlein '29 found that with a slight dose the males lost interest in normal females although the structure of the testis remained unchanged. DaCosta and Carlson '33 reported a depression of sexual maturation and a pronounced lowering of the relative weight of the testes of male rats as a result of thyroid feeding. Weichert and Boyd '33 noted the induction of typical pseudopregnancy in thyroid-fed female rats.

The lack of harmony in the results of the experiments cited above is due partially to the variance in dosage, the form in which the glandular substance is administered and the method of administration.

II. Experimental

In the following experiments Parke Davis' desiccated sheep thyroid gland in powder form was used. The dosage varied within the limits of .125 - .5 gm. daily, depending upon the age of the recipient. In order to insure consumption of a definite amount of the thyroid material, the powder was weighed and mixed with enough water to

make a thin paste which was fed to the rat by means of a medicine dropper. The animals, with few exceptions, within a few days learned to take the mixture quite readily.

As a preliminary step to the experiments reported below, 35 rats, 3 - 6 months of age were fed thyroid gland in doses varying from .25 - .5 gm. daily and their tissues prepared for histological study. Guided by the results obtained, the following series of experiments was then planned and performed.

Experiment 1. Effect of thyroid feeding on the weights of the reproductive organs and accessory glands.

Seven litters comprising 35 rats and varying in age from 3 weeks to 10 weeks were used. Nineteen were fed .125 - .5 gm. thyroid powder per day. In 3 - 5 weeks old litters the dose at the beginning of the experiment was .125 gm. but this was increased to .25 gm. if the experiment was continued until the animals were 7 weeks of age or older. Litters 7 weeks old or older received .25 gm. or .5 gm. thyroid gland throughout the experiment. At the end of the experiment, the animals were sacrificed and their reproductive organs and the adrenal and pituitary glands were weighed. The percentage of body weight of these organs was calculated and the averages of these percentages per litter have been tabulated in table 1.

Table 1.

Percentage of body weight of reproductive organs and adrenal and pituitary glands of thyroid-fed and control rats.

Litter Number		Number of anim.	Age, weeks	Age at autopsy, Weeks	Duration of exp. "	Daily dose Grams	Average percentage of body weight				
							Testes	Prostate Glands	Seminal Vesicles	Adrenal Glands	Pituitary Gland
I	T*	3	3	9	6	.125	1.199	.037	.076	.030	.0029**
	C	3		9			1.139	.057	.083	.017	.0028**
II	T	2	5	14	9	.125-.25	1.187	.100	.171	.024	.0049
	C	2		14			1.020	.144	.256	.015	.0037
III	T	4	7	10	3	.25	1.262	.138	.079	.018	.0029**
	C	3		10			1.179	.211	.101	.013	.0024**
IV	T	2	8	11	3	.25	1.428	.152	.095	.020	.0036**
	C	2		11			1.272	.215	.102	.013	.0026**
V	T	2	8	14	6	.5	.933	.116	.232	.015	.0034
	C	2		14			.896	.140	.270	.011	.0032
VI	T	2	10	14	4	.25		.122	.178	.022	.0037
	C	2		14				.125	.226	.014	.0033
VII	T	4	10	17	7	.25	1.159	.102	.226	.025	.0040
	C	2		17			.967	.122	.189	.017	.0032
VIII Castrates	T	4	15	18	3	.25		.012	.048	.051	.0028**
	C	4		18				.012	.048	.012	.0028**

* T - Thyroid-fed; C - Control
** Anterior lobe only.

In the case of the testis and adrenal and pituitary glands, a slight increase in the relative weights of these organs is seen in the thyroid-fed animals of each litter when compared to their normal control litter-mates. The prostate glands and the seminal vesicles are relatively smaller in the experimental animals than in the controls. The differences between the percentage weights of the organs of the thyroid-fed and control animals is slight, but, with one exception (the seminal vesicle in litter 7) is consistant.

Experiment 2. Effect of thyroid feeding upon the histological appearance of the reproductive organs and associated glands.

The testis and seminal vesicle and the prostate, thyroid, pituitary, adrenal and mammary glands of the animals used in experiment 1 were fixed in Bouin's picro-formol solution, stained in either Delafield's or Ehrlich's haematoxylyn and studied.

Histologically testis, seminal vesicle and adrenal and prostate glands present the same picture in the experimental as in the control animals (Fig. 1 to 6). The cells of the prostate glands of the thyroid-fed animals show plainly the clear secretory area midway between the lumen and the nucleus of the cell indicative of a normal prostate cell (Moore, Price and Gallagher '30). The seminal vesicles of the experimental animals exhibit the "haloed" secretory

granules described in the rat by Moore, Hughes and Gallagher '30 to be evidence of a functionally secreting cell of that organ.

The thyroid gland of the normal animals shows a predominance of follicles with a high cuboidal epithelium and a small colloid content. Some follicles are present with a low epithelium and an abundance of colloid. The thyroid glands of the treated animals show only the latter type of follicle.

In addition to the cell types normally seen in the anterior pituitary gland, that of the thyroid-fed animal also shows the castration cell described by Addison '17. This type of anterior pituitary gland cell is not so abundant in the thyroid-fed animals as it is in the pituitary gland of a castrated animal, but it may be seen in appreciable numbers (Fig. 7,9,11).

The mammary glands of the experimental and control animals are quite different. The normal male mammary gland consists of masses of tissue with illy defined lumina. The cells are large, with abundant cytoplasm usually showing some vacuolization (Fig.8). The glands of the thyroid-fed animals, on the other hand, show very few of such masses of tissue. The cells apparently have become rearranged about well defined lumina of ducts. The amount of cytoplasm in the cells is reduced as evidenced by the more crowded condition of the nuclei (Fig.12).

The mammary glands of normal castrates and thyroid-fed castrates were also examined. The castrate glands resemble normal mammary glands (Fig.10). Those of the thyroid-fed castrates rather closely parallel in appearance the mammary glands of the thyroid-fed animals as described above.

A possible explanation for the histological findings in these experiments will be considered later.

Experiment 3. Survival of spermatozoa in the isolated epididymides of thyroid-fed animals.

Six litters containing 28 mature rats were used. In each animal the right testis was removed and the epididymis fastened to the base of the scrotal sac by a thread. Fifteen of the rats were fed .25 gm. thyroid daily for 30 days and the rest were kept as controls. Some of the epididymides become infected at the sites of the ligatures, but in the healthy animals it was found that the thyroid feeding had no effect upon the time of survival of the spermatozoa in the isolated epididymis. The spermatozoa of the experimental animals were found in several cases to have survived as long as those of their control litter-mates. Survival was noted in both types of animals for the duration of the experiment, 30 days.

Experiment 4. Effect of thyroid feeding on fertility.

Two litters, designated as A and B for convenience, containing 4 and 6 rats respectively were used.

Litter A consisted of 4 male rats 5 weeks of age. Two of these were fed thyroid powder for 9 weeks. The beginning dosage of .125 gm. was increased to .25 gm. when the rats were 7 weeks old. At the end of the seventh week of thyroid feeding, these animals and their litter-mate controls were put into separate cages, each with a normal female in dioestrus. Within 3 days spermatozoa were found in the vaginas of all the females. Pregnancy followed in each case and 4 healthy litters, ranging in number from 8 to 10 individuals were born and reared.

Litter B consisted of 6 male rats 10 weeks of age. Four were fed .25 gm. thyroid powder daily for 7 weeks. At the end of the fourth week of thyroid feeding all the animals in the litter were placed in separate cages, each with a normal female in dioestrus, and within 3 days all the females were found to have spermatozoa in the vagina. Each female gave birth to normal young which were reared to maturity.

From these results it seems safe to conclude that thyroid feeding in amounts indicated above, begun before or after puberty and continued for 4 to 7 weeks, does not have any effect upon the mating reactions or fertility.

Experiment 5. Castrate male rats fed desiccated thyroid gland.

Eight litter mate males were castrated at 15 weeks of age. Four were fed .25 gm. thyroid powder for 3 weeks,

beginning the day after castration. Four were kept as controls. At the end of 3 weeks all the animals were sacrificed and their accessory reproductive organs and adrenal and pituitary glands were weighed. The results are tabulated in table 1, litter VIII.

As may be seen from the table, in the case of each organ with the exception of the seminal vesicles, the average percentage of the body weight was the same for both the thyroid-fed and the control animals. The hyperthyroid condition seemed to have no effect upon the weights of the organs in the absence of the testes.

Experiment 6. Effect of thyroid feeding upon the potency of the anterior pituitary gland in stimulating precocious sexual maturity in immature female rats.

Two litters, 7 and 8 weeks old, containing 7 and 4 animals respectively were used. In the first litter 4 animals were fed thyroid powder; in the second litter, 2 animals were similarly treated. The dosage of .25 gm. daily was continued for 3 weeks. At the end of this time, the animals were decapitated and their anterior pituitary glands implanted into the hind leg muscles of female rats belonging to 2 litters 3 and 4 weeks old. The result obtained as indicated in table 2 was a greater increase in the relative weights of the ovaries of the animals receiving implants from thyroid-fed donors than was shown by the animals receiving implants from normal donors, with one exception.

Table 2.

Percentage of body weight of ovaries of immature female rats receiving implants from thyroid-fed and control males.

Recipients from thyroid-fed donors	Recipients from control donors
.026	.024
.025	.031
.047	.032
.037 .049	.027
.041 .040	.033

III. Discussion

The result of the first experiment, i.e., slight decrease in the weights of the prostate glands and seminal vesicles, suggested that the testicular hormone was present in a smaller concentration than normally, in other words, there seemed to be a slight castration effect. The sperm motility tests were performed to determine whether the concentration of the testicular hormone in the thyroid-fed animals was sufficient to enable the spermatozoa in the isolated epididymides to retain their motility for as long a period as the spermatozoa in the isolated epididymides of the normal animals (Benoit '26). The results of experiment 2 showed the concentration to be adequate. Experiment 3 demonstrated that the spermatozoa not only retain their motility but, contrary to the findings of Gudernatsch '15, they also retain their fertilizing power to the fullest extent.

The indication of a slight castration effect as a result of thyroid feeding is substantiated by experiment 6 in which the anterior pituitary glands of the thyroid-fed animals proved more potent than those of the normal animals in stimulating precocious sexual maturity in immature females. Van Horn '31 has shown that a hyperthyroid condition in female rats increases the gonad stimulating power of the hypophysis 19 - 63%. Evans and Simpson '30 have also found an increased potency of the anterior pituitary

gland of hyperthyroid female rats. Schockaert '31 noted that after thyroidectomy in males, the response of the secondary sex organs to the injection of anterior pituitary was increased. This effect upon the pituitary gland has been explained (Van Horn '31) by a change in the concentration of the gonadal hormones with a resultant increase or decrease in the concentration of the gonadotropic pituitary hormone. Moore and Price '32, using male and female rats, injected oestrin and testicular hormone alone, incombination, and with the gonadotropic hypophyseal hormone or hormones. They have demonstrated a pituitary-gonad interrelationship. The gonads function only when stimulated by the anterior pituitary gland. But the pituitary gland is controlled to some extent by the gonadal secretions for when these are present in effective amounts, the hypophyseal activity is lowered. The converse of this statement, i.e., when the amount of gonadal hormone is lowered, the activity of the anterior pituitary gland is increased, has been demonstrated by Engle '29 and Evans and Simpson '29. Weichert and Boyd '34 have suggested that a hyperthyroid condition may cause a lowering of the theelin content of the body and thus indirectly affect the pituitary gland as evidenced by the responses of the ovary and the mammary gland in the female rat.

If in the experiments described in this paper, there has been an elimination of testicular hormone, as a result of hyperthyroidism, it has not apparently been rapid enough

nor overwhelming enough to produce the typical castrate picture in the accessory reproductive organs, i.e., the prostate gland and the seminal vesicle, since much less hormone than is normally present is adequate for maintaining the normal histological structure of these organs. The decrease in the weights of the organs may be explained by their secreting less actively than normally.

The presence of castration cells in the anterior pituitary gland but the absence of the castration type of cells in the prostate glands and seminal vesicles of the thyroid-fed animals may be explained in another way. It has been suggested by several investigators cited below that there may be two hormones present in the testis, one of which maintains the secondary sex organs, and the other of which acts as a check upon the anterior pituitary gland. Martins and Rocha '30 have implanted testis tissue into castrates and prevented the appearance of castration cells in the anterior pituitary gland without preventing changes in the accessory reproductive organs. Castrated male rats injected with purified bull testes extract have been found to possess normal accessories, yet the hypophysis showed castration cells (Nelson, unpublished - from Allen, Sex and the Internal Secretions). The effects of thyroid feeding may then be ascribed to an elimination of testis hormone or hormones possibly due to the heightened rate of metabolism, a consequent decrease in the secretory action

of the prostate glands and seminal vesicles, and an increase in potency of the anterior pituitary gland.

At present I am unable to formulate an adequate explanation for the changes occurring in the mammary gland following thyroid feeding. The change cannot be due to elimination of testicular hormone since similar changes are not found following castration. It is possible that there may be a stimulation of the galactogogue principle of the anterior pituitary gland described by Corner '30, Turner and Gardner '31 and Riddle, Bates and Dykshorn '32. However, no positive statement can be made as to how this principle is affecting the male gland. Further experiments are being planned to make an extensive study of these mammary gland changes.

IV. Summary and Conclusions

1. Seventy-five male rats ranging from 3 - 15 weeks in age were fed thyroid powder in doses of .125 gm., .25 gm. or .5 gm. daily for 3 to 9 weeks.

2 The weights of the testes, seminal vesicles and prostate, adrenal and pituitary glands were determined in 35 animals. In the 19 thyroid-fed animals, the weights of the testes, adrenals and pituitary glands were found to be relatively larger than the weights of the corresponding organs in the control animals. The weights of the prostate glands and seminal vesicles of the experimental animals were found to be relatively lower than those of the control animals.

3. The thyroid-fed animals bred normally and fathered healthy litters, after 4 - 7 weeks of thyroid feeding regardless of whether the feeding was begun before or after puberty.

4. The histological appearance of the testes, seminal vesicles and prostate glands of the experimental animals is normal.

5. The anterior pituitary glands of the thyroid-fed animals show an appreciable number of castration cells.

6. The hypophyses of the experimental animals are more potent in stimulating precocious sexual maturity in immature females.

7. The results are believed to be due to a more rapid than normal elimination of the testicular hormone or hormones resulting in a lessened activity of the seminal vesicles and prostate glands and a heightened activity of the anterior pituitary gland.

9. A change occurs in the structure of the mammary gland of the thyroid-fed rats but no explanation is offered for this phenomenon.

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