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Effect of Thyroid Hormone on Growth of Thyrotrophin-Secreting Pituitary Tumors.* (20400)

EVELYN L. GADSDEN, AND JACOB FURTH.

From Biology Division, Oak Ridge National Laboratory, Oak Ridge, Tenn.

Tumors of the pituitary develop in almost every mouse following destruction of the thyroid by I^{131}(1,2). These tumors secrete large quantities of thyroid-stimulating hormone (TSH)(3). The mechanism of the development of these tumors is not known with certainty. Lack of TH (thyroid hormone) provides the major stimulus for proliferation of TSH secreting cells, but whether or not ionizing radiation, incidental to radiothyroidectomy, is an essential contributing factor is under investigation(4,5). These induced tumors are readily transplantable on radiothyroidectomized but not on normal hosts(2). However, in the course of subpassages the tumors may gain autonomy and grow even better in normal than in radiothyroidectomized animals(3). Induction of the primary tumors can be prevented by administration of TH(6,7). If lack of TH is the stimulus for the growth of these tumor cells, can the growth of tumor grafts be prevented by TH? Will TH also affect the growth of autonomous tumors? Will thiouracil, an antithyroid compound which blocks the synthesis of TH, promote the growth of dependent tumors? The 3 sets of experiments now to be reported were designed to answer these questions.

Materials and methods. C57 black mice of both sexes were used. Transplantations of the I^{131}-induced pituitary tumors were made by cutting the tumor into fine particles in physiological saline and injecting it into the thigh muscle. Thyroid hormone was administered (a) by feeding desiccated thyroid in food or (b) by intramuscular injections or subcutaneous grafts of enlarged thyroids obtained from mice bearing autonomous grafted pituitary tumors. In one experiment (Table I) thyroids of tumor-bearing mice fed thiouracil for a long period of time were used. The desiccated thyroid† was mixed with ground Purina Chow, ½ g thyroid per 1000 g food. Each animal was fed approximately 3 g of this food mixture per day (1½ mg thyroid). Thyroids to be grafted were either cut into fine particles in physiological saline and injected in the muscle of the thigh or were grafted subcutaneously. Thyroid feeding was usually begun on the day of tumor grafting; the time relation is shown in the tables. Thiouracil‡ was administered as a 0.1% solution in drinking water given *ad libitum.* Thiouracil treatment was begun prior to tumor grafting in most experiments and at the time of tumor grafting in 2 experiments as indicated in Table III.

Results. Effect of TH on dependent tumors.

* Work performed under contract for the U. S. Atomic Energy Commission.

† "Thyroid Strong" obtained through the courtesy of Parke-Davis.

‡ Obtained through the courtesy of the American Cyanamid Co.

TABLE I. Effect of Thyroid Hormone on Growth of Dependent Pituitary Tumors.

Tumor strain	TH given: Time, days*	TH given: Mode	Radiothyroidectomized hosts: TH treated: No. inj. / No. takes	Radiothyroidectomized hosts: TH treated: Latency, days†	Radiothyroidectomized hosts: Not treated: No. inj. / No. takes	Radiothyroidectomized hosts: Not treated: Latency, days	Normal hosts: Not treated: No. inj. / No. takes	Normal hosts: Not treated: Latency, days
Strains remained dependent:								
101	0	Food	7/0	—	10/10	137 (124–158)	4/0	—
101	—3	Graft	7/0	—	10/10	134 (123–167)	10/0	—
124	+1	Food	10/0	—	7/7	78 (63–99)	11/0	—
124	0	Graft	7/2	137 (132–141)	8/8	116 (109–121)	10/0	—
77	—4	"	10/0	—	6/6	181 (143–205)	6/0	—
Strains turned autonomous:								
3-D	+4	Food	3/3	157 (148–161)	9/9	75 (65–90)	11/11	99 (83–133)
3-D	0	Graft‡	5/5	83 (75–96)	4/4	59 (54–75)	5/5	211 (159–236)
163	0	"	2/0	—	9/9	81 (56–125)	9/2	139 (139)
19	—8	"	8/6	132 (68–208)	7/7	80 (68–110)	8/0	—

* In relation to pituitary tumor graft.
† Means; range is in parentheses.
‡ Hyperplastic thyroid was obtained from thiouracil-treated mice.

TABLE II. Effect of Thyroid Hormone on Growth of Autonomous Pituitary Tumors.

TH given: Time, days*	TH given: Mode	Radiothyroidectomized hosts: TH treated: No. inj. / No. takes	Radiothyroidectomized hosts: TH treated: Latency, days†	Radiothyroidectomized hosts: Not treated: No. inj. / No. takes	Radiothyroidectomized hosts: Not treated: Latency, days	Normal hosts: TH treated: No. inj. / No. takes	Normal hosts: TH treated: Latency, days	Normal hosts: Not treated: No. inj. / No. takes	Normal hosts: Not treated: Latency, days
0	Food	3/3	76 (66–96)	3/3	117 (117)	5/4	66 (66)	5/5	47 (40–54)
0	"	5/5	46 (29–81)	5/5	46 (40–60)	10/10	39 (29–40)	11/11	27 (16–29)
+1	"	—	—	10/10	33 (21–51)	10/10	19 (16–21)	10/10	13 (11–16)
0	Graft	—	—	5/5	111 (74–177)	4/4	49 (42–63)	7/7	53 (42–74)
0	"	5/5	30 (30)	5/5	32 (30–40)	5/5	47 (47)	5/5	30 (30)
—6	"	3/3	30 (30)	7/6	75 (70–80)	3/3	30 (30)	10/10	32 (30–58)
0	"	—	—	10/10	77 (55–118)	5/5	54 (46–90)	5/5	46 (46)
–13	"	3/3	84 (70–97)	4/4	96 (86–107)	—	—	6/6	58 (53–70)

*† For explanations, see Table I.

The 9 experiments can be presented in 2 groups: (a) In 5 experiments 3 transplantable tumor strains were used which thus far did not turn autonomous. Grafts of these failed to grow in the 41 normal mice used in these studies, but grew in all but one of the 40 radiothyroidectomized mice. Thirty-nine radiothyroidectomized mice grafted with such tumors were also treated with TH and of these only 2 developed tumors. Thus TH was effective in preventing tumor growth. In the 2 animals in which the tumors grew the thyroid grafts could not be identified at autopsy; thus the tumor growth could be explained by unsuccessful thyroid graftings. (b) In 4 experiments strains of pituitary tumors were used which had a tendency to become autonomous. Eighteen of 33 normal mice were receptive to grafted tumors of these strains which appeared after a long latency period. The tumors grew after a shorter latency period in all of 29 grafted radiothyroidectomized mice. The administration of TH to radiothyroidectomized mice reduced the number of successful tumor grafts and increased their latency period (Table I).

Inhibitory effect of TH on growth of established pituitary tumors. In experiments performed on a small scale TH retarded the growth of already established tumors but did not cause their complete regression.

Two preparations containing TH were used. (a) thyroxine (Roche-Organon) was given 6 times weekly in doses of 0.5 μg in 0.05 cc of saline and (b) thyroprotein (Parke-Davis) was given 6 times weekly in doses of 0.125 mg in 0.1 ml of distilled water.

In one experiment thyroxine was adminis-

TABLE III. Effect of Thiouracil on Growth of Pituitary Tumors.

Tumor strain	Thiouracil treatment, days	Hosts: Radiothyroidectomized No. inj.	Radiothyroidectomized No. takes	Radiothyroidectomized Latency, days	Thiouracil-treated No. inj.	Thiouracil-treated No. takes	Thiouracil-treated Latency, days	Normal No. inj.	Normal No. takes	Normal Latency, days
Strains remained dependent:										
82	—16	5/5	125 (105–161)		5/0	—		4/0	—	
101	—126	9/9	134 (121–157)		4/0	—		10/0	—	
Strains turned autonomous:										
3-D	0	8/8	75 (61–103)		5/5	104 (94–136)		—	—	
3-D	0	6/6	73 (65–75)		4/4	233 (197–267)		5/5	246 (230–267)	
3-D	0	6/6	85 (74–100)		5/5	174 (165–188)		4/4	226 (215–236)	
3-D	—93	8/8	67 (53–90)		6/6	175 (165–186)		8/8	222 (207–251)	
3-D	—24	5/5	56 (42–60)		6/6	101 (84–137)		6/6	173 (163–178)	
3-D	—89	5/5	70 (62–85)		8/8	79 (72–85)		5/4	113 (113)	
163	—117	9/9	39 (32–59)		9/9	121 (107–172)		10/7	268 (210–282)	
3A	—127	4/4	84 (71–95)		4/4	59 (46–71)		5/5	51 (46–56)	

For explanations, see Table I.

tered for 8 weeks to 4 animals each having tumors ranging from 1 to 3 cm across. Control animals with tumors of comparable sizes were observed without treatment. In 3 of the treated animals the tumors regressed, though not completely; the size of the tumor of the fourth animal remained nearly unchanged. The control animals died with very large, progressively growing tumors.

In another experiment thyroprotein was administered to 2 mice for 4 months. The treatment began when the tumors measured about 0.5 x 2 cm. These tumors greatly diminished in size, measuring at death 5 x 8 mm. The tumor of a control mouse grew progressively, and the animal died with a large tumor.

Another 3 mice were injected with thyroprotein when their tumors measured about 2 cm across. Tumors in 2 of these mice failed to progress during 15 weeks of treatment. The 2 controls died with large tumors. In another experiment the tumors of 2 mice remained about unchanged in size after 14 weeks of thyroprotein injections and these animals lived much longer than the controls which died with large tumors.

Although the total number of animals in these experiments is small, the results are highly significant since several strains of tumors were used in these studies and progressive growth occurred invariably in hundreds of untreated mice with grafted tumors.

Effect of TH on autonomous tumors. In all 8 experiments listed in Table II the autonomous strain 3-A was used. These autonomous tumors grew in all but one of the 150 mice grafted, and it is possible that the animal in which the tumor failed to grow did not receive viable cells. Table II indicates that while TH treatment failed to prevent tumor growth in any of the animals, it hastened tumor growth somewhat in most radiothyroidectomized mice. In normal hosts the results were variable. In some experiments the latency period was somewhat longer; in others, somewhat shorter, indicating that the mild TH treatment did not significantly alter the growth rate of the tumors. The thyroids of mice bearing autonomous pituitary tumors were greatly enlarged and the amount of TH secreted by them was probably greater than that administered. What the continued administration of very large quantities of TH would do remains to be studied.

A correlation of the size of thyroid grafts with tumor growth was not made. When the grafts were identified and studied microscopically, they exhibited the same changes as those seen in the thyroids of the hosts, namely, hyperplasia, reduction of colloid content, intracellular colloid deposits and formation of adenomas.

Effect of thiouracil on the growth of pituitary tumors. The 10 experiments performed can be conveniently presented in 3 groups: (a) In 2 experiments in which tumor strains

were used which remained dependent in the course of a large series of transplantation studies (not detailed in this report), the grafts succeeded in all of 14 radiothyroidectomized mice and in none of 14 normal mice. Thiouracil treatment did not render the 9 animals studied susceptible to these tumors. (b) In 7 experiments strains of pituitary tumors were used which tended to become autonomous. Transplantations succeeded in all of 47 radiothyroidectomized mice and failed in 4 of the 38 normal control mice. In the radiothyroidectomized animals the average latency period varied between 39 and 85 days; in normal mice between 113 and 268 days. The tumor grafts succeeded in all of 43 thiouracil treated mice. The latency period was in every experiment shorter than in normal hosts and much longer than in radiothyroidectomized hosts (Table III). (c) In one experiment an autonomous pituitary tumor strain was used. The grafts succeeded in all animals and appeared first in normal, next in thiouracil-treated, and last in radiothyroidectomized hosts. These experiments indicate that blocking TH synthesis by thiouracil does not accomplish the results obtained by massive destruction of the thyroid with radioiodine. In an earlier study the effect of thiouracil treatment on tumor growth was compared with that of depression of the thyroid by graded doses of radioiodine, and it was estimated that the thiouracil effect was equivalent to that of a small dose of I^{131} ($\sim$25 μc) which caused but slight depression of the thyroid.

Thiouracil-treated animals had greatly enlarged thyroids which were studded with adenomas, as described by others.

Summary and conclusions. 1. The administration of thyroid hormone effectively prevents the growth of dependent pituitary tumors induced in and grafted on radiothyroidectomized mice. Autonomous pituitary tumors grow faster in normal than in radiothyroidectomized hosts, and, in the latter, administration of TH hastens the growth of the tumor. Sustained thiouracil treatment does not render normal mice susceptible to dependent pituitary tumors but hastens somewhat the growth of such pituitary tumors which tend to become autonomous. 2. These experiments support the view that lack of TH is the major growth stimulant of pituitary growths in radiothyroidectomized mice and that sustained lack of this hormone is essential for the proliferation of these tumor cells. In the course of sustained proliferation a new (autonomous) type of tumor cell arises which is not only uninhibited but is often stimulated by TH, the physiological depressant of normal pituitary cells producing thyrotrophin.

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