# The Influence of the Thyroid Gland on the Survival of Rats and Mice Bearing Transplanted Lymphoid Leukemia*

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Relatively little attention has been given to the hormones of the thyroid-pituitary axis in leukemogenesis, despite evidence that they may influence the activity of lymphoid tissues (5, 10). It has been reported that spontaneous lymphoma-formation in mice is not affected by the administration of thiourea (25) or by thiouracil or thyroxine except in association with changes in body weight (7). The administration of thyroid hormone has likewise been observed to have no effect on the rate of induction of lymphoid tumors by irradiation in C57BL mice (10).

In the present experiments the effects of altered thyroid activity on the survival of mice and rats bearing transplanted lymphoid leukemia were studied. While this investigation was in progress, it was reported by Grad *et al.* (7) that weekly injections of thyroxine did not influence the survival of AKR mice with transplanted lymphatic leukemia.

## MATERIALS AND METHODS

*Rats.*—Inbred male and female rats of a Wistar subline were divided into two experimental groups at 6 weeks of age; one group was given propylthiouracil in the drinking water (0.025 per cent), and the other received tap water. The animals were caged individually, weighed regularly, and pair-fed to maintain uniform body weight throughout both groups. Approximately 9 weeks after beginning the administration of propylthiouracil, all rats were given injections intraperitoneally of 0.1 ml. of a suspension of isologous lymphoma cells in physiological saline. On the day after inoculation and every 3 days thereafter until death one-half of the rats in each group received a subcutaneous injection of thyroxine (crystals, Squibb; 100 μg. dissolved in 5 drops of 4 per cent NaOH and diluted with distilled water to 20 ml.), 1.0 mg. in each of the first two injections, and 0.5 mg/dose subsequently (Table 1).

*Mice.*—Young adult male AKR mice were thyroidectomized by the subcutaneous or intraperitoneal injection of 200–230 μc. of iodine-131, a dose observed to destroy the thyroid gland in mice maintained on an ordinary laboratory diet containing moderate amounts of iodine (4). Fourteen days later the subcutaneous administration of saline, thyroid-stimulating hormone (TSH), or thyroid hormone was begun, the mice being inoculated with 0.1 ml. once every other day for four injections and once every 3d day thereafter until death. Thyroxine (crystals, Squibb) was prepared, as before, in a concentration of 1 mg/ml and TSH (Thytropar, Armour) in a concentration of 10 USP units/ml in physiological saline. The thyroxine solution and the saline for the controls were made slightly alkaline. One or 2 days after the third injection of the hormone or vehicle, 0.1 ml. of a suspension of isologous lymphoma particles in physiological saline was injected intraperitoneally. The suspension consisted of finely divided fragments of tissue from the thymus, spleen, and mesenteric lymph nodes of leukemic AKR mice. All animals were autopsied, and sections of the thyroid gland and other tissues were studied histologically. The amount of food offered the mice was regulated in the first three experiments to maintain a uniform dietary intake in all treatment groups.

TABLE 1

EFFECT OF PROPYLTHIOURACIL AND THYROXINE ON SURVIVAL OF RATS BEARING TRANSPLANTED LYMPHOMA

| | Treatment | | | |
|---|---|---|---|---|
| Exper. no. | Control | Propyl-thiouracil | Propyl-thiouracil and thyroxine | Thyroxine |
| | Mean survival time (days) | | | |
| I | 18.0 (6)* | 25.5 (6) | 21.4 (5) | 15.8 (6) |
| II | 16.8 (15) | 20.6 (8) | | |
| III | 14.1 (6) | | | 12.3 (6) |
| Av. | 16.3 (27) | 23.0 (14) | 21.4 (5) | 14.0 (12) |

* No. of rats indicated in parentheses.

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## RESULTS

Every animal given inoculations of lymphoma cells died of transplanted leukemia. The mean survival time of euthyroid rats was 15–18 days (Table 1) and of euthyroid mice, 11 days (Table 2). Hypothyroidism prolonged the mean survival 25–40 per cent (4–7 days) in rats ($P = 0.05$) and 17 per cent (2 days) in mice ($P = 0.01$), as shown in Charts 1 and 2. This was associated with delay in the onset of lymphadenopathy preceding death and with reduction in the extent of leukemic infiltrations observed post mortem. When hypothyroid animals were given thyroxine, these effects were reduced or abolished (Tables 1, 2). In euthyroid rats the administration of thyroxine hastened death from leukemia (Table 1; Chart 1); but in euthyroid mice TSH had no effect on survival. In radiothyroidectomized mice the injection of TSH prolonged survival more than radiothyroidectomy alone ($P < 0.03$). By reducing the food intake of all animals to the amount consumed by the hypothyroid animals, the appetite of which was significantly diminished, the body weight was kept essentially uniform in all treatment groups until the terminal stages of the disease (Chart 1).

TABLE 2

EFFECT OF THYROID STATUS ON SURVIVAL OF AKR MICE BEARING TRANSPLANTED LYMPHOID LEUKEMIA

| Exper. no. | Treatment: Control | Thyroidectomy | Thyroidectomy and thyroxine | Thyroidectomy and TSH | TSH | Thyroidectomy and thyroxine and TSH |
|---|---|---|---|---|---|---|
| | Mean survival time (days) | | | | | |
| I | 10.2 | 11.7 | 11.9 | | 13.5 | 11.6 |
| II | 13.3 | 15.5 | 10.5 | | 9.8 | 10.6 |
| III | 11.4 | 14.5 | 12.3 | 15.6 | | 12.4 |
| IV | 10.4 | 10.5 | 9.1 | | | |
| Av. | 11.3 | 13.3 | 10.9 | 15.6 | 11.6 | 11.5 |
| Total no. mice* | 39 | 49 | 44 | 10 | 19 | 27 |

* In each experiment there were nine to fifteen mice per treatment group.

## DISCUSSION

The retardation of the growth of transplanted lymphomas in hypothyroid hosts and the acceleration of their growth in hyperthyroid animals are consistent with earlier observations on the response of lymphoid tissues to thyroid hormone. Marine *et al.* (13) emphasized that lymphoid hyperplasia occurs frequently in patients with Graves' disease. Others (3, 8, 12, 17) have noted in experimental animals involution of lymphoid tissues after thyroidectomy and lymphoid hyperplasia after the administration of thyroxine or TSH.

It has also been observed that lymphosarcomas in rats concentrate $I^{131}$-labeled diiodotyrosine (21) and that TSH is inactivated by rabbit thymus at a rate far exceeding that of all other tissues except the thyroid (16). Similarly, Sonenberg (24) has found a selective absorption of $S^{35}$-labeled TSH by the thymus and lymph nodes second only to that of the thyroid gland.

The influence of nutritional factors was controlled by regulating the caloric intake of the lymphoma-bearing animals, since underfeeding has been shown to inhibit the spontaneous development of lymphoid leukemia in AK mice (18) and to extend the survival of mice bearing transmitted leukemia (6). It is evident, in view of the uniformity of body weight among the various treatment groups, that the differences in survival and lymphoma growth resulted from factors other than nutritional variation. It is not likely that the observed effects stemmed from secondary changes in adrenocortical function, because hypothyroidism is known to result in adrenal atrophy (15), which by itself enhances the growth of lymphoid tissues (5, 9).

The administration of thyroxine to leukemic rats significantly reduced their survival, in contrast with the observations of Grad *et al.* (7) on lymphoma-bearing mice. This difference may be attributable to the more intensive thyroxine dosage employed in the present investigation; however, in agreement with Grad's results, TSH had no significant effects on the survival of euthyroid mice in the present study. Hence, the possibility of a species difference in responsiveness to thyroxine or TSH should be explored further. It is also conceivable, however, that the greater responsiveness of the rat resulted from a lower degree of virulence, or autonomy, of the rat neoplasm employed. The rat lymphosarcoma used in these experiments arose spontaneously in a WR female, had been transmitted through fifteen transplant generations at the beginning of the investigation, and uniformly killed isologous recipients on intra-

peritoneal injection in 12–25 days. The mouse lymphosarcoma, which originated spontaneously in an AKR breeder and had been transmitted through eight transplant generations, killed slightly more rapidly, isologous recipients dying uniformly on intraperitoneal inoculation after a mean interval of only 11 days. An even more dramatic response to induced hormonal changes might conceivably be exhibited by lymphomas arising *de novo*, which are presumably of lesser autonomy than transplanted lymphomas, if the responsiveness of the lymphoma is inversely related to its degree of autonomy.

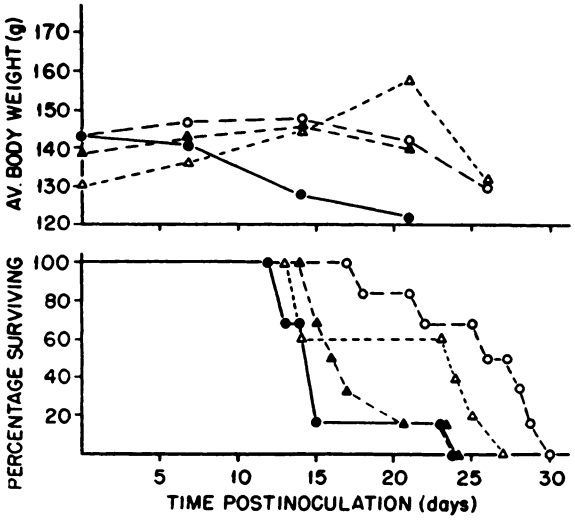


CHART 1.—Body weight and survival of propylthiouracil- and thyroxine-treated rats bearing a transplanted lymphoid leukemia (data of Experiment 1, Table 2). ▲, controls; ●, thyroxine; △, propylthiouracil + thyroxine; ○, propylthiouracil.

The effect of thyroidectomy on the growth of neoplasms other than lymphomas has been studied in mice, rats, and rabbits. The early literature was reviewed by Levine and Kugel (11), who observed acceleration of the growth of Tumor-180 in mice. Others have observed thyroidectomy to cause regression (14) or resistance to induction (22), or to have no effect (2, 23) on various neoplasms. Although the activity of the thyroid has not been observed to affect the spontaneous development and induction of lymphomas in mice (10), in none of the previous investigations has the influence of nutritional factors and of endocrinological changes secondary to altered thyroid function been adequately controlled. The significance of these experiments is therefore uncertain.

The reduced responsiveness of the thyroid to goitrogenic agents in animals bearing transplanted neoplasms (1) and the detection of an iodide-trapping substance in the blood of tumor-bearing rats (20, 21) suggest that the metabolism of the thyroid gland is itself disturbed in the tumor-bearing host. In view of the results of the present study, the role of the thyroid-pituitary axis in neoplastic growth deserves further attention.

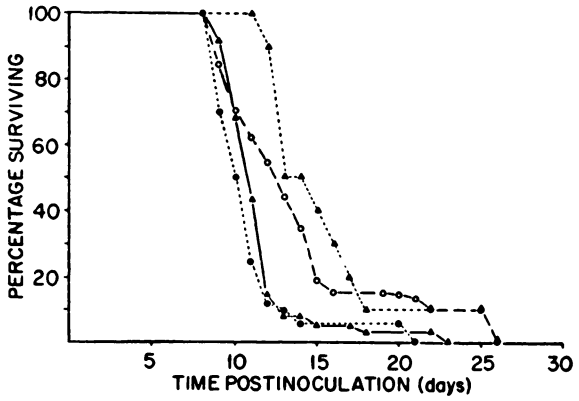


CHART 2.—Survival time of AKR mice bearing transplanted lymphoid leukemia as affected by thyroidectomy, TSH, and thyroxine. ▲, controls; △, thyroidectomy + TSH; ○, thyroidectomy: ●, thyroidectomy + thyroxine.

## SUMMARY

1. The mean survival of rats and mice bearing transplanted lymphomas was significantly prolonged by the administration of propylthiouracil or by thyroidectomy; this effect was inhibited by the administration of thyroxine.

2. In rats rendered hypothyroid by propylthiouracil, the lymphomatous infiltrations were less extensive than in the euthyroid controls.

3. The administration of thyroxine to euthyroid hosts hastened their death from transplanted leukemia.

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