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EFFECTS OF CHRONIC HYPERTHYROIDISM ON THE LIFESPAN OF THE RAT

HIROSHI OOKA and TADASHI SHINKAI

Department of Biology, Tokyo Metropolitan Institute of Gerontology, 35-2 Sakaecho, Itabashi-ku, Tokyo 173 (Japan)

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SUMMARY

The life durations of hypo- and hyperthyroid Wistar rats were measured under clean conventional conditions. The amount of exogenous T_4 (thyroxine), which is sufficient to elevate T_4 levels in the blood, decreased with age. The rats which were made hypothyroid by the neonatal T_4 treatment had a longer lifespan than control. The lifespans of hyperthyroid rats, to which T_4 solutions were given as drinking water during either the first or the second half of the life period, were shorter than control. The life-shortening effect of T_4 was not detected when T_4 was administered to already aged animals. These results indicate that the effect of T_4 administration is not due to the direct promotion of the diseases which cause the death, but to the acceleration of aging during the young or middle-aged period.

Key words: Rat; Hyperthyroidism; Lifespan

INTRODUCTION

The accelerative effect of thyroid hormones on the rate of aging has been suggested by various works [1–3]. Our previous work has indicated that the lifespan of rats which were made hypothyroid by neonatal thyroxine (T_4) treatment is significantly longer than that of normal rats [4]. In the present study, cohorts of rats were made hyperthyroid by continuous administration of T_4 in drinking water, and the lifespan and other biological data were compared with those of control rats. The lifespan of neonatal T_4-treated rats was also measured again in this study for comparison, because of the considerable variation in the mean lifespan among cohorts of normal rats [5,6].

MATERIALS AND METHODS

Wistar SPF rats bred in the Tokyo Metropolitan Institute of Gerontology were used. Rats were fed *ad libitum* with Charles River CRF1 chow under clean conventional condi-

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tions during the experiment. Blood samples were collected under light anaesthesia with ether. Effect of this ether anaesthesia on the level of blood T_4 was checked by comparison with the samples from decapitated rats in a preliminary experiment, and ascertained that there is no significant difference. Most blood samples were collected by heart puncture. Some of them in successive sampling from the same rat were carried out from the tail vein, which also showed no significant difference in the T_4 level from the heart blood of the same rat in the preliminary experiment. Heparinized plasma, after the blood corpuscles were centrifuged down, was stored in −20°C. Radioimmunoassay for the estimation of thyroid hormones was carried out with Amerlex T_4 and T_3 (Amersham) RIA kits with the correction according to the results of a preliminary test using T_4- and T_3-free rat plasma. T_4 administration, to make the rats hyperthyroid, was done with T_4 solution in 0.002 N NaOH prepared 2 times per week. The T_4 solution, administered ad libitum, was used as drinking water. To make the rats hypothyroid, the pups were injected intraperitoneally with 1.5 μg T_4/g body weight on days 2, 4, 6, 8, and 10 of newborn life. T_4 levels of these rats are significantly lower than normal rats throughout the lifespan [4]. Statistical analysis was done by Student's *t*-test.

RESULTS

Figure 1 shows age changes in the level of T_4 in the blood of normal male Wistar rats and the elevation of T_4 concentration after exogenous T_4 administration through drinking water. Blood T_4 decreased gradually with age. In middle aged (8 months old) and senescent (26 months old) rats, T_4 levels increased after drinking 10 mg T_4/l of the

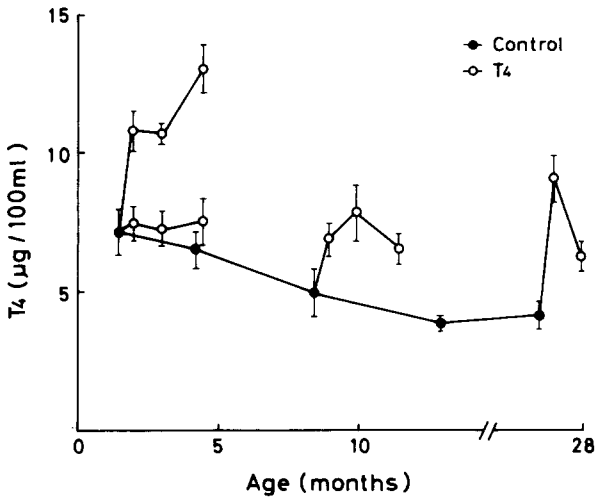

Fig. 1. Concentrations of T_4 in the blood of normal and T_4-administered male rats at various age. The experimental rats were given 10 mg T_4/l solution as drinking water, except for a group of young rats which were given 20 mg T_4/l solution (upper circles).

solution. However, changes in the T_4 level were not observed in young (2 months old) rats after drinking the T_4 solution of this concentration. When the concentration of T_4 increased to 20 mg/l, a remarkable elevation of the T_4 level was induced. A similar result was obtained when T_3 was measured in the blood of the same rats used in T_4 estimation (Fig. 2). The result that the increase in the T_4 level of older animals was larger than that of young rats might reflect the fact that the catabolic rate of T_4 in young rats was faster than that of old ones. From this result, we selected the dose of T_4 in drinking water for the lifespan experiment. We gave 20 mg T_4/l to the rats from 1 to 6 months of age, and 10 mg T_4/l to the rats older than 6 months.

Figure 3 shows the time course of the changes in the levels of T_4 and T_3 in the blood of the rats continuously given 10 mg T_4/l as drinking water after 8 months of age. Levels of both hormones were higher in the T_4-treated group than in control, but they gradually decreased after a maximum value, and T_3 in T_4-treated rats was almost the same level as control after T_4 treatment for 13 months. Some of the T_4-treated rats were given normal water instead of T_4 solution after the 8- and 12-month experimental period. The levels of both T_4 and T_3 of those rats returned to normal within 1 month after the cessation of T_4 administration.

Figure 4 shows the survival curves of normal, neonatal T_4-treated (hypothyroid), and long-term T_4-administered (hyperthyroid) male rats. The rats in the hyperthyroid group were given 10 mg T_4/l as drinking water after 12 months of age throughout the rest of their lifespan. The lifespan of hypothyroid rats is the longest of the three, and the hyperthyroid rats have shorter lifespan than normal rats, although the duration of the hyperthyroid state is approximately the last half of the whole life period in this experiment.

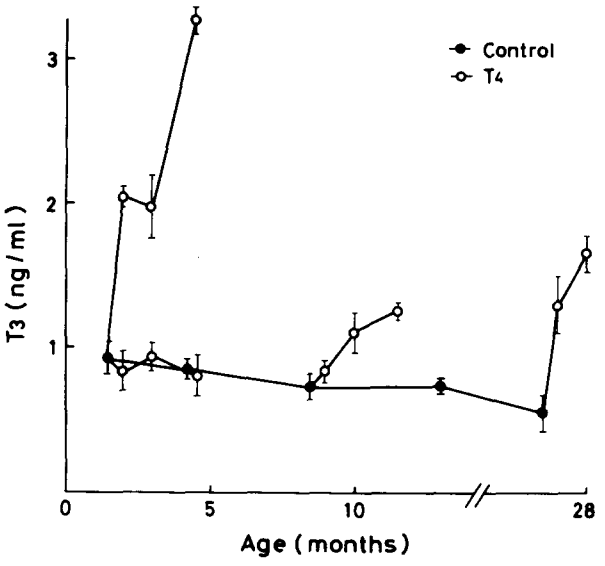

Fig. 2. Concentrations of T_3 in the same blood samples as shown in Fig. 1.

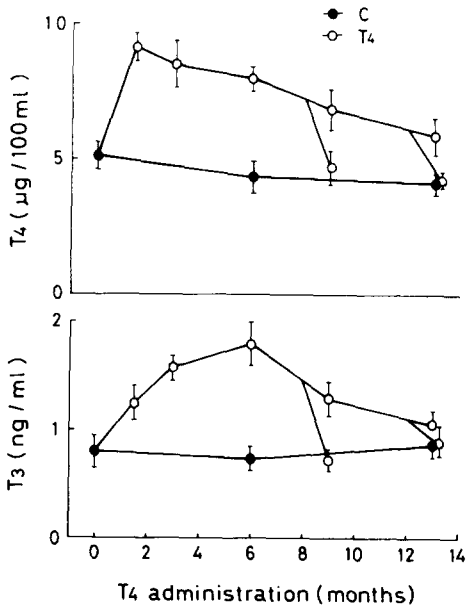

Fig. 3. Age-changes in concentrations of T_4 and T_3 in the blood of normal rats and T_4-administered rats which were given 10 mg T_4/l solution for 13 months. A part of the rats in the T_4 group was given water instead of T_4 solution after 8 and 12 months (lower circles at 9 and 13 months).

Figure 5 shows the result of the second experiment on the lifespan. Control rats were given 0.002 N NaOH solution as drinking water from 1 to 22 months of age. Experimental rats were divided into 2 groups. In one group they were given T_4 solution from 1 to 22 months, and in the other group they were given T_4 solution from 1 to 12 months and NaOH solution between 12 and 22 months. The rats of all groups were given acidic water, which is used routinely to prevent infection in our animal station, after 22 months old, to eliminate the direct effect of hyperthyroidism to the death rate of the rat. The

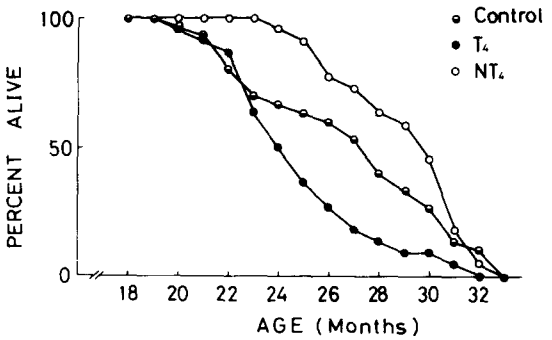

Fig. 4. The survival curves of control, continuously T_4-treated (T_4), and neonatally T_4-treated (NT_4) male rats. The rats in T_4 group were given 10 mg T_4/l solution after 12 months old. The numbers of rats were 30 (control), 22 (T_4) and 22 (NT_4).

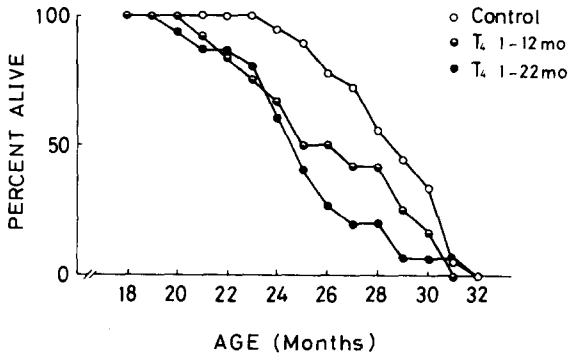

Fig. 5. The survival curves of control and continuously T_4-treated male rats. Control rats were given NaOH solution from 1 to 22 months of age. Some of the rats in the T_4 group were given T_4 solution from 1 to 12 months, and NaOH from 12 to 22 months. The rest of the rats in the T_4 group were given T_4 from 1 to 22 months. The concentration of T_4 solution was 20 mg/l from 1 to 6 months, and 10 mg/l after 6 months. The numbers of rats were 18 (control), 12 (T_4 1–12 months) and 15 (T_4 1–22 months).

lifespans of experimental groups were significantly shorter than that of the control group without significant difference, however, between the rats given T_4 throughout the whole lifespan and those given T_4 during the first half of the life period. This result is possibly due to the fact that T_4 and T_3 levels showed no remarkable difference after continuous administration for more than 13 months (Fig. 3). It is concluded that the life-shortening effect of thyroid hormones shown in this study is not due to the direct positive effect of the hormones to the initiation or promotion of diseases which are a direct cause of death, but due to the acceleration of aging during the whole period of the experiment, because the lifespan of the rats was significantly shorter than control in the second experiment, even when the administration of T_4 was discontinued after 12 months of

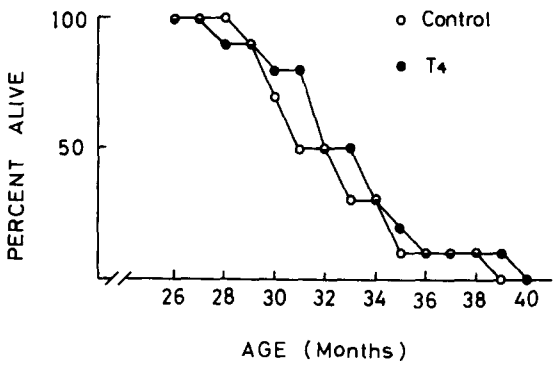

Fig. 6. The survival curves of control and continuously T_4-treated female rats. The rats in the experimental group were given T_4 solution after 26 months of age, throughout the rest of the life period. The numbers of the rats were 10 in both groups.

TABLE I

FOOD CONSUMPTION (g/DAY/RAT) OF CONTROL AND T_4-ADMINISTERED (FOR 1 MONTH) RATS OF VARIOUS AGES

	3 months	*10 months*	*26 months*
Control	19.4 ± 2.1[a]	15.7 ± 2.0	10.6 ± 0.8
T_4 (10 mg/l)	18.1 ± 0.9	14.6 ± 1.8	9.2 ± 1.6
T_4 (20 mg/l)	24.6 ± 1.3	–	–

[a]Measurements were carried out on the food consumed by three rats from each cage. Standard deviations were calculated among cages.

age, earlier than the period of actual death by more than 6 months. This conclusion is supported by the result shown in Fig. 6. The lifespan of female rats which were given T_4 when they were already senescent (after 26 months) was not shorter than that of controls.

Table I shows the food consumption of the control and T_4-administered rats. Significant increase in food intake by T_4 administration was observed only in the very early age period.

Table II shows the body weights of the rats in the second experiments on lifespan. Body weights of the rats in the T_4-administered group were smaller throughout the period of administration. The rats to which T_4 was given for 12 months recovered their normal body weight within a month after cessation of T_4 administration.

DISCUSSION

In laboratory rats and mice, a trend towards increasing longevity has been documented for a number of inbred strains [5]. The mean lifespan of cohorts of Wistar male rats in our laboratory has demonstrated the same trends during the past 8 years (from 24.5 months to 27 months), and part of the cause of this increase is ascribed to a change

TABLE II

BODY WEIGHTS OF CONTROL AND CONTINUOUSLY T_4-ADMINISTERED RATS AT VARIOUS AGE (g)

	3 months	*13 months*
Control	286 ± 3.2	489 ± 23.6
T_4	249 ± 17.4*	459 ± 18.3*
		486 ± 31.5[a,**]

[a]Administration of T_4 was discontinued after 12 months.
*$P < 0.01$, **$P > 0.1$ (control *vs.* T_4).

of diet (Funabashi F2 → Charles River CRF1). The present study indicates that the hypothyroidism induced by neonatal T_4 treatment prolongs the lifespan of the rat under the recent improved conditions which permit long lifespan to the normal rats, and confirms the previous result which has already been reported in a study on the rats of shorter lifespan [4].

The present study also showed that the hyperthyroidism accelerates the aging process, since T_4 administration shortens the lifespan of the rat without the direct action to the cause of death. In 1928, Robertson [1] reported that mice fed desiccated thyroid throughout their life had shorter lifespans than control mice. Everitt [7] showed that middle-aged rats treated with thyroxine had a reduced lifespan, although the difference was not statistically significant. In the present study, the accurate measurements of concentrations of thyroid hormones in the blood by radioimmunoassay enabled us to induce moderate hyperthyroidism which was enough to reveal the acceleration of aging without any drastic harmful effects.

It has frequently been reported that food restriction prolongs the lifespan of rats [8,9]. In the present experiment, body weights of short-lived hyperthyroid rats are smaller than controls whereas long-lived hypothyroid rats are also smaller than controls [4]. Therefore, changes in body weight can not be the cause of the aging-accelerating effect of thyroid hormones. The amount of food does not change by the administration of T_4 at the dose used in this study except at a very early age. On the other hand, the life-shortening effect of T_4 is observed in the second half of the life period as much as in the first half. Therefore reduction of food intake can not mediate the effect of hyperthyroidism. Everitt [10] has shown that the accelerative effect of T_4 on aging of the rat, observed by the solubility of collagen and urinary protein excretion, is diminished by food restriction. One of the possible explanations is that thyroid hormones accelerate aging by promotion of an intrinsic process which is disturbed exogenously by food restriction.

It has been reported that the lifespan correlates with metabolic rate both among various species of mammals [11] and among various strains of the mouse [12]. Since blood levels of thyroid hormones are correlated with metabolic rate [13], mechanism of aging-acceleration effect of thyroid hormones may include the elevation of O_2 uptake. On the other hand, the data in some reports do not support the concept that nutritional restriction slows the rate of aging by decreasing the rate of energy metabolism [9,14]. The stimulation of protein and RNA synthesis by thyroid hormones [15,16] can also be responsible for their action to promote aging.

REFERENCES

1 T.B. Robertson, The influence of thyroid alone and of thyroid administrated together with nucleic acids upon the growth and longevity of the white mouse. *Aust. J. Exp. Biol. Med. Sci.*, *5* (1928) 69–74.

2 H.D. Johnson, L.D. Kinter and H.H. Kibler, Effect of 48°F (8.9°C) and 83°F (28°C) on longevity and pathology of male rats. *J. Gerontol.*, *18* (1963) 29–36.

3 W.D. Denckla, Role of the pituitary and thyroid glands in the decline of minimal O_2 consumption with age. *J. Clin. Invest., 53* (1974) 572–581.
4 H. Ooka, S. Fujita and E. Yoshimoto, Pituitary-thyroid activity and longevity in neonatally thyroxine-treated rats. *Mech. Ageing Dev., 22* (1983) 113–120.
5 C.A. Curcio, N.A. McNelly and J.W. Hinds, Variation in longevity of rats: Evidence for a systematic increase in lifespan over time. *Exp. Aging Res., 10* (1984) 137–140.
6 C.F. Hollander, H.A. Solleveld, C. Zurcher, A.L. Nooteboom and M.J.V. Zwieten, Biological and clinical consequences of longitudinal studies in rodents: Their possibilities and limitations. An overview. *Mech. Ageing Dev., 28* (1984) 249–260.
7 A.V. Everitt, The effect of prolonged thyroxine treatment on the aging male rat. *Gerontologia, 3* (1959) 37–54.
8 C.M. McCay, M.F. Crowell and L.A. Maynard, The effect of retarded growth upon the length of life span and upon the ultimate body size. *J. Nutr., 10* (1935) 63–79.
9 E.J. Masoro, B.P. Yu and H.A. Bertrand, Action of food restriction in delaying the aging process. *Proc. Natl. Acad. Sci. USA, 79* (1982) 4239–4241.
10 A.V. Everitt, The thyroid gland, metabolic rate and aging. In A.V. Everitt and J.A. Burgess (eds.), *Hypothalamus, Pituitary and Aging*, Charls C. Thomas, Springfield, 1976, pp. 511–528.
11 G.A. Sacher, Evaluation of the entropy and information terms governing mammalian longevity. *Interdiscip. Top. Gerontol., 9* (1976) 69–83.
12 G.A. Sacher and P.H. Duffy, Genetic relation of life span to metabolic rate for inbred mouse strains and their hybrids. *Fed. Proc., 38* (1979) 184–188.
13 S.B. Barker and H.M. Klitgaard, Metabolism of tissues excised from thyroxine-injected rats. *Am. J. Physiol., 170* (1952) 81–86.
14 S. Leto, G.C. Kokkonen and C.H. Barrows, Dietary protein, life-span, and physiological varieties in female mice. *J. Gerontol., 31* (1976) 149–154.
15 L. Sokoloff, P.A. Roberts, M.M. Januska and J.E. Kline, Mechanisms of stimulation of protein synthesis by thyroid hormones in vitro. *Proc. Natl. Acad. Sci. USA, 60* (1968) 652–659.
16 J.R. Tata and C.C. Widnell, Ribonucleic acid synthesis during the early action of thyroid hormones. *Biochem. J., 98* (1968) 604–620.