

Suppressive Effect of *Spirometra erinacei* Plerocercoids on Thyroid Function in Golden Hamsters

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Abstract

Plerocercoids of the tapeworm, *Spirometra erinacei*, produce a growth hormone (GH)-like factor that stimulates growth of normal and hormonally deficient mice but does not stimulate growth of golden hamsters. Plerocercoid infection (30 scoleces per hamster) markedly suppressed the serum levels of thyroxin (T_4) (48%) and triiodothyronine (T_3) (40%) and increased the ^{125}I - T_3 uptake (20%) in golden hamsters. These results suggest that plerocercoid infection causes a reduction in T_4 -binding globulin (TBG) and the free T_4 index. Furthermore, the plerocercoid-infected golden hamster exhibited a significant decrease in the cell height of the acinar epithelium of the thyroid gland, indicating an inactive thyroid function and suppressed thyroid-stimulating hormone (TSH) secretion. This hypothyroid state induced by plerocercoid infection may be associated with a GH-like factor, which is recognized as a GH by hosts and stimulates hypothalamic production of somatostatin and, in turn, suppresses the levels of the TBG and endogenous T_4 and T_3 .

Key words: Plerocercoid, *Spirometra erinacei*, Growth Factor, Thyroid Gland

Introduction

Plerocercoids of the tapeworm, *Spirometra erinacei*, produce a growth hormone (GH)-like factor, which stimulates body growth of normal mice (Hirai *et al.*, 1978; Shiwaku and Hirai, 1982), chinese hamsters (Hirai *et al.*, 1983a) and hypothyroid (Hirai *et al.*, 1983b) or dwarf mice (Shiwaku *et al.*, 1983), and displaced [^{125}I]-human GH from its receptor on hepatic membranes prepared from a pregnant rabbit (Hirai *et al.*, 1986). The presence of plerocercoids in normal and dwarf mice results in a significant increase in the circulating levels of somatomedins (Shiwaku *et al.*, 1986) and an enhancement of the liver and skeletal muscle weights (Shiwaku and Hirai, 1982) but these effects are not seen in golden hamsters (Hirai *et al.*, 1983a). Furthermore, plerocercoid infection decreases the concentration of serum

glucose in golden hamsters but increases that in mice (Hirai *et al.*, 1983a). The reasons for these differences in the biological effects of plerocercoid infection between mice and golden hamsters are yet unclear.

The plerocercoid-infected golden hamster, in which there is no promotion of body growth, shows a marked decrease in the serum levels of thyroxin (T_4) (Tsuboi and Hirai, 1986) similar to infected mice (Hirai *et al.*, 1983b) which exhibited promoted body growth. Therefore, it is suggested that golden hamsters are suitable experimental animals for observing the direct effects of the GH-like factor, designated the plerocercoid growth factor (PGF), on thyroid functions. The present study was performed to investigate the effects of PGF on thyroid function in golden hamsters as a continuing series of experiments on the physiological characteristics of PGF.

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Materials and Methods

Reagents.

The radioimmunoassay kits used for the

measurements of the serum levels of T_4 and triiodothyronine (T_3) and the ^{125}I - T_3 uptake were purchased from Dainabot Co. (T_4 -Ria Kit, T_3 -Ria Kit II, Tokyo, Japan) and Daiichi Radioisotope Labs. Ltd. (Spac T_3 Uptake Kit, Tokyo, Japan), respectively.

Plerocercoids and animals.

Plerocercoids of *Spirometra erinacei* were collected from two species of snake, *Rhabdophis tigrinus* and *Elaphe quadrivirgata*, captured in the southern part of Ehime Prefecture, Japan. These larvae were maintained in the subcutaneous tissue of golden hamsters until they were used.

Male Syrian-golden hamsters, 8 weeks old, were purchased from the Inoue Jikkendoubutsu Center Co. (Kumamoto, Japan). The animals in the experimental group received 30 scoleces of plerocercoids in 0.4 ml of physiological saline solution containing 1000 U/ml Penicillin-G and 0.5 mg/ml Streptomycin, under the skin of the back. The animals in the control group received a similar injection of saline solution containing antibiotics only. All the animals were kept in plastic cages on 12 hr light-dark cycles at 24°C and were given laboratory chow and water *ad libitum*.

Measurements of thyroid hormones.

On the 14th day after infection, the golden hamsters of each respective group were weighed and sacrificed by decapitation after being starved for 24 hr. The serum from each respective animal was obtained from collected blood by centrifugation at 4°C for 10 min at 3000 rpm and used for measurements of thyroid hormones by radioimmunoassay.

As an estimate of free T_4 index, the T_3 uptake index was multiplied by the serum T_4 levels as reported by the Committee on Nomenclature of the American Thyroid Association (Solomon *et al.*, 1976).

Measurement of the cell height of acinar epithelium in the thyroid gland.

The thyroid glands of two animals each from the control and experimental group were perfused with 2% glutalaldehyde in 0.1 M sodium phosphate buffer (pH 7.4) and then immediately before they were removed. The removed thyroid glands were fixed in 2% glutalaldehyde in the same buffer for 2 hr at 4°C. Following buffer rinse, the specimens were post-fixed in 1% OsO_4 in the same buffer for 2 hr at 4°C. The fixed thyroid glands were dehydrated in a graded series of ethanol and embedded in Epon 812. The embedded specimen was cut in 0.5 μm thickness and stained with toluidin blue O. In order to estimate thyroid function the height of one hundred cells of some acini in the respective thyroid gland was measured under a light microscope (Ingbar and Woeber, 1981).

Statistical differences between the control and experimental group were assessed by the unpaired Student's *t*-test.

Results

Table 1 shows various parameters of the thyroid function in the control and experimental groups on day 14 after infection. The serum levels of T_4 and T_3 and the free T_4 index in the experimental animals were significantly lower than those in the control animals. On

Table 1 Effect of plerocercoid infection on various parameters of thyroid function in golden hamsters

	No. of animals	T_4 ($\mu\text{g}/\text{dl}$)	T_3 (ng/ml)	^{125}I - T_3 uptake index	Free T_4 index	T_3/T_4 (ng/ μg)
Control	5	3.71 \pm 0.23	0.465 \pm 0.055	2.09 \pm 0.05	7.77 \pm 0.56	12.6 \pm 1.9
Experimental	5	1.92 \pm 0.17 \ddagger	0.281 \pm 0.050 \dagger	2.50 \pm 0.12 \ddagger	4.79 \pm 0.41 \ddagger	14.7 \pm 2.4

Values are means \pm standard deviations. The statistical significance of differences between the means for the control and the experimental group was estimated by Student's *t*-test. $\dagger P < 0.01$, $\ddagger P < 0.001$ compared with controls.

the other hand, the $^{125}\text{I-T}_3$ uptake index was significantly increased in the experimental group as compared with the control group. There was, however, no difference in the T_3/T_4 ratio between the control and the experimental groups.

In order to investigate the cause of the decrease in the serum levels of T_4 and T_3 , the cell height of the acinar epithelium in the thyroid gland was measured in the control and experimental animals, to determine whether the thyroid function was active or not (Ingbar and Woeber, 1981). Plerocercoid-infected animal

caused a significant decrease in the cell height of the acinar epithelium of the thyroid gland as compared with the control animals (Fig. 1) but did not exhibit other pathological changes (Fig. 2-5).

Discussion

PGF produced by plerocercoids of *S. erinacei* stimulates body growth of mice by inducing somatomedins (Shiwaku *et al.*, 1986) but does not stimulate body growth of golden hamsters (Hirai *et al.*, 1983a). Infection of these larvae, however, lowers the serum levels of T_4 in both of these animals, as shown in our previous reports (Hirai *et al.*, 1983b; Tsuboi and Hirai, 1986).

In this study, plerocercoid infection lowered the serum levels of T_4 and T_3 , and the free T_4 index while it raised the $^{125}\text{I-T}_3$ uptake, indicating a suppressive effect on the biosynthesis of thyroid hormones in golden hamsters which were indispensable for the growth of hosts. In addition, the fact that plerocercoid infection does not alter the T_3/T_4 ratio indicates that there is no effect on the T_3 neogenesis from T_4 in the peripheral tissue. The decrease in the cell height of acinar epithelium of thyroid gland in the plerocercoid-infected golden hamster shows histologically that plerocercoids suppress the biosynthesis of T_4 and do not stimulate the thyroid-stimulating hormone (TSH) secretion in adenohypophysis. Furthermore, the decrease in the serum T_4 levels and free T_4 index and the increase in the $^{125}\text{I-T}_3$ uptake caused by plerocercoid infection suggest a reduction in the T_4 -binding globulin (TBG), as shown in a previous report (Daughaday, 1981).

Our previous studies showed that PGF produced by plerocercoids of *S. erinacei* was a GH-like substance (Hirai *et al.*, 1986), and caused an increase in the concentration of serum free fatty acid (Hirai *et al.*, 1983a) and hypertriglyceridaemia with suppression of the lipoprotein lipase activity in golden hamsters (Tsuboi and Hirai, 1986) as well as physiological actions of GH (Murase *et al.*, 1980; Murase

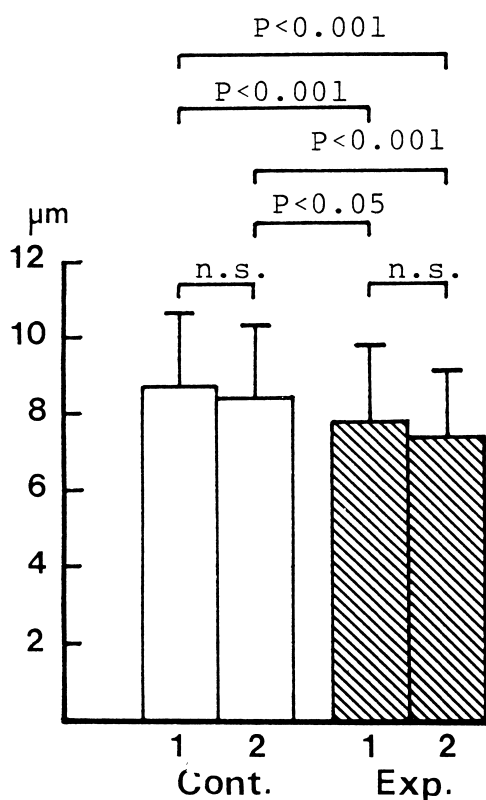
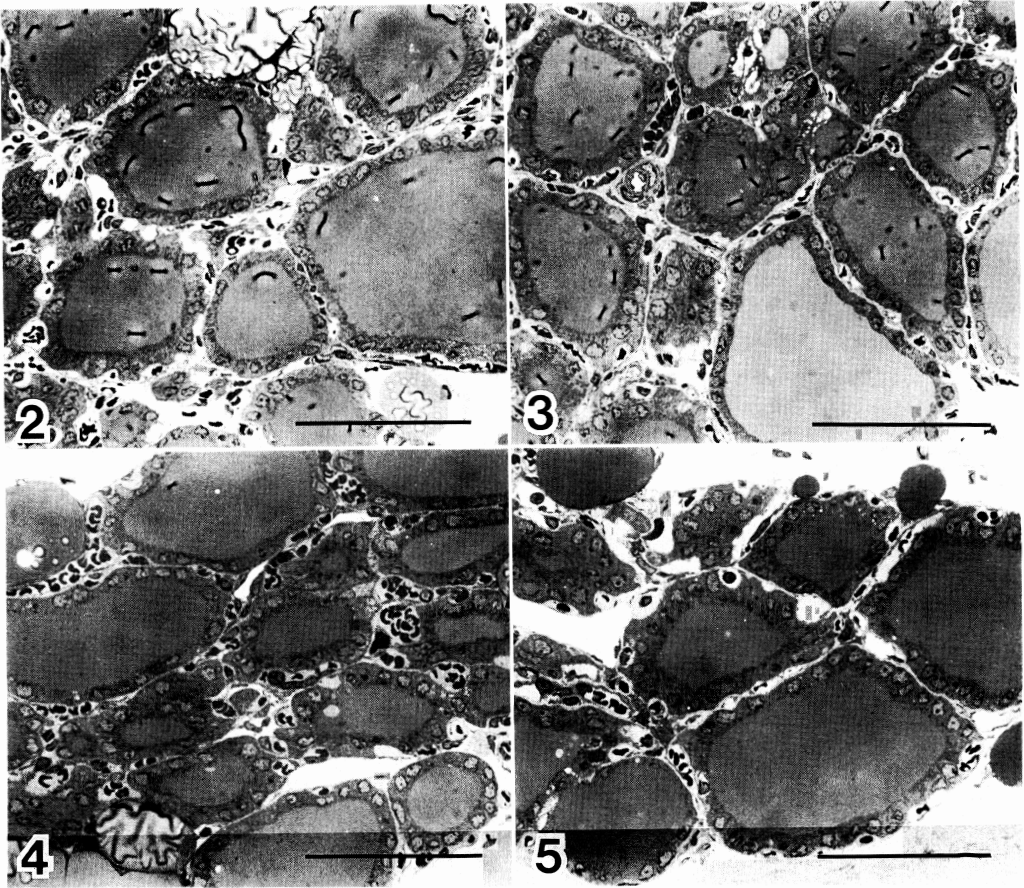


Fig. 1 Effect of plerocercoid infection on the cell height of the acinar epithelium in the thyroid gland.

Values are means \pm standard deviations. The statistical significance of differences between the respective mean of the cell height for the control (Cont.) and the experimental (Exp.) animal was estimated by Student's *t*-test.

n.s. : not significant



Morphological aspect of the acinus of thyroid in the control and experimental animals.

Figs. 2 and 3: Control group

Figs. 4 and 5: Experimental group

Scale bar = 100 μ m

et al., 1981). On the other hand, there are some reports that an excessive secretion of GH in a patient led to decrease in the concentration of TBG (Daughaday, 1981) and somatostatin, which is secreted by the hypothalamus due to excess of GH, suppresses the activity of TSH (Ahren *et al.*, 1978). In addition, PGF produced by plerocercoids of *S. mansonioides* suppresses the circulating amount of GH as well as serum T_4 levels in hosts (Phares, 1982).

In light of these reports, the results reported here indicate that PGF produced by plerocercoids of *S. erinacei*, which mimics the physiological actions of GH, is recognized as a GH and stimulates hypothalamic production of soma-

tostatin and, in turn, suppresses the endogenous T_4 and T_3 . However, PGF seems to be effective in maintaining normal growth of plerocercoid-infected golden hamsters in spite of a possible retardation of growth caused by a significant reduction in the serum T_4 and T_3 levels.

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