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THE EFFECT OF SODIUM FLUORIDE ON THE METAMORPHOSIS OF TADPOLES*

by

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The supposed effect of fluorine derivatives upon thyroid physiology has elicited wide interest. From purely theoretical points of view, it is worthy of attention that fluorine belongs to the Seventh Periodic Group like its chemical relative, iodine. *Baumann* and his associates[1-3] studied the behavior of the thyroid towards the members of this group by biochemical means. Their results indicated that the thyroid gland possesses an ability to concentrate these elements in a ratio of greater magnitude than the other tissues of the body.

From a practical point of view, the use of fluorine derivatives in industry, and, in lesser degree, its earlier use as a local antiinfective in medicine brought about several studies on the response of various organs to fluorine administration, including that of the thyroid gland. However, the recent suggestions of the fluoridation of drinking water for preventing dental caries have greatly increased the interest in the effect of acute or chronic exposure to fluorine on the metabolism of the thyroid.

The results of animal experiments on the effect of sodium fluoride on the thyroid physiology are highly conflicting. Synergistic[11, 12, 17], inhibitory[6, 9, 13, 16] and neutral[4, 5, 7, 15] effects have been reported. *Ogilvie*[10] described an atrophy of the thyroid after

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toxic doses of sodium fluoride in rats. This divergence is probably in part due to the different doses and species used.

In a recent short communication, *Saadi-Nazim*[14] reported that sodium fluoride in doses of 0.1 g and 0.2 g per liter had some inhibitory effect on the metamorphosis of tadpoles. The metamorphosis in Amphibia is known to be solely induced by thyroid secretion, activated by thyrotropic hormone. The metamorphosis can be considered as a sensitive test of thyroid activity insofar it is disturbed and retarded under the influence of different chemical and physical factors which act on the thyroid (*Lynn & Wachowski*[8]).

The present experiment was carried out in order to investigate whether the addition of sodium fluoride to water interferes with the metamorphosis of tadpoles using doses existing in drinking water or proposed for addition to municipal water supplies.

MATERIAL AND METHODS

The *Rana temporaria* larvae were collected from a pool on May 21, 1960. They were all about the same size, length approx. 10 mm and had no macroscopically visible hindlegs. They were subdivided into four groups, sixty animals in each. Each group was placed in a separate 2 litre aquarium. The tap water for the aquaria was filtered through a carbon filter. The groups were kept under identical conditions with regard to space, food, light and temperature. Each group was given daily an abundant and equal portion of ground meat and dark bread which was eaten readily.

One group served as controls. To the aquaria of the three other groups, sodium fluoride, *pro analysi,* was added in doses of 1, 2 and 10 mg/litre, respectively. The experiment was started on May 25, 1960, and terminated on July 25, 1960, at which time only a few tadpoles from every group had failed to undergo metamorphosis.

The animals were frequently examined under a magnifying glass. As the forelegs emerged from under the branchial arch, the tadpoles were picked up from the aquarium and killed with chloroform. Ten tadpoles from each group were histologically

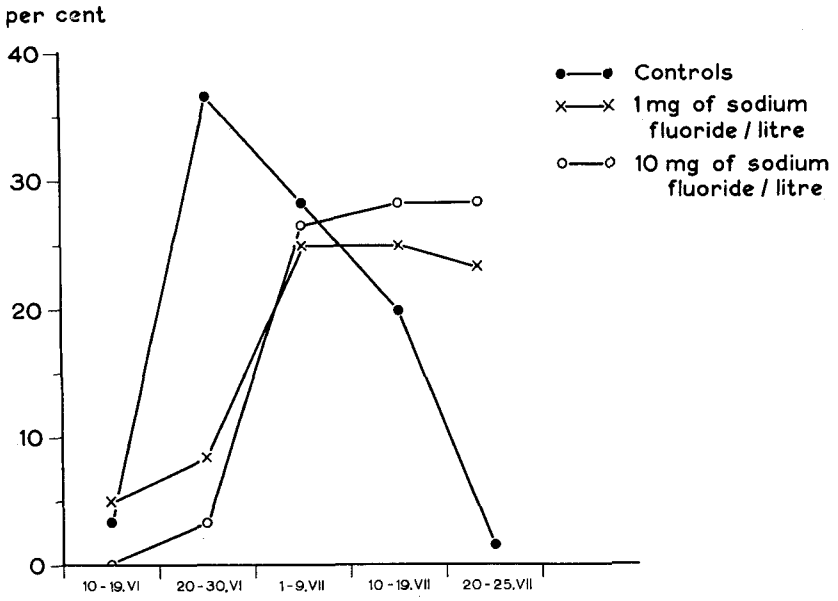

Fig. 1. The times of metamorphosis.

examined. After fixation in *Bouin's* fluid they were embedded in paraffin and sectioned serially. The sections were stained with *Mallory*'s azan method.

OBSERVATIONS AND DISCUSSION

Four to six animals from each group died before the metamorphosis. At the termination of the experiment 5 tadpoles from the control group and 4—6 from the other groups failed to undergo a metamorphosis.

The times of metamorphosis are given in Fig. 1. For the sake of clarity, the curve representing the group which received sodium fluoride in the dose of 2 mg/litre is omitted. This curve was almost parallel with the curve of the group which was given 1 mg/litre.

As is clear from Fig. 1, the metamorphosis of the sodium fluoride groups was delayed as compared with that of the controls. Nearly 40 per cent of the control tadpoles underwent a

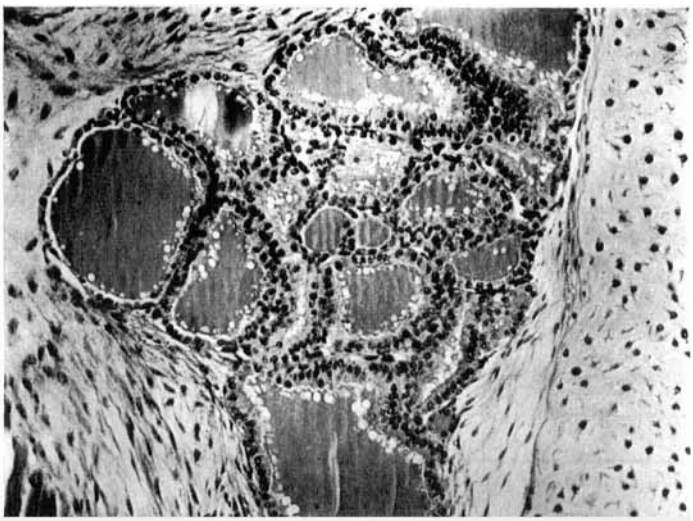

Fig. 3. The thyroid of a tadpole from the 10 mg of sodium fluoride/litre group. 350 ×.

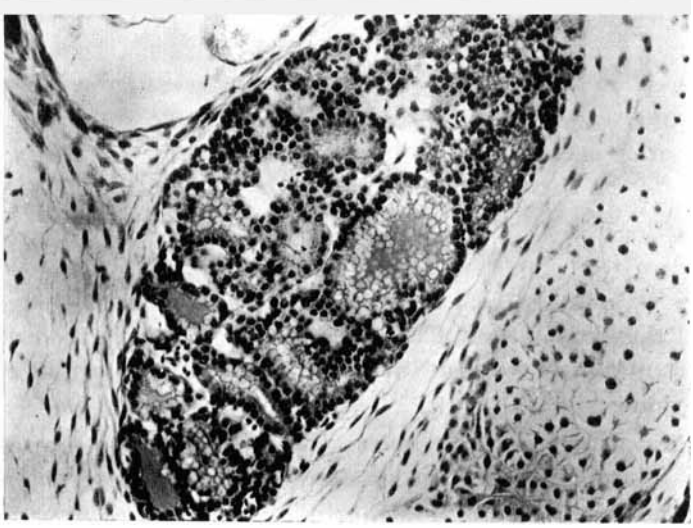

Fig. 2. The thyroid of a control tadpole. 350 ×.

metamorphosis during the period June 20—30, whereas the tadpoles of the sodium fluoride groups metamorphosed quite evenly during the three following periods July, 1—9, July 10—19 and July 20—25 at a rate of 25—30 per cent.

Histological examination revealed that the thyroids of the sodium fluoride-treated tadpoles showed a more flattened epithelium and a greater amount of colloid than those of the control group, *i.e.* in the three experimental groups the thyroid glands were less active as far as the histological picture is concerned (Figs. 2 and 3).

The results of the present experiment indicate that sodium fluoride in the doses used seems to delay the metamorphosis of tadpoles and produce decreased activity of the thyroid gland. According to the common concept amphibian metamorphosis is controlled by the thyroid hormone, the secretion of which, again, is ruled by thyrotropic hormone secreted by the anterior lobe of the pituitary. The delay or failure of the metamorphosis is usually explained to be consequent on deficiency of thyroid secretion, insensitivity of tissues to the thyroid influence, or impairment of the thyrotropic function of the pituitary. The way sodium fluoride influences the complicated metamorphic events remains to be explained.

The tadpole-test is rather sensitive and reflects readily even slight disturbances in the pituitary-thyroid system. Although some known antithyroid agents, like thiourea derivatives, show a strong inhibitory effect on amphibian metamorphosis, too, it is unjustified to draw any definite conclusions on potential antithyroid properties of various substances in mammals based on observations made on amphibian material only.

SUMMARY

The effect of sodium fluoride on *Rana temporaria* tadpoles was investigated by adding it in the doses of 1, 2 and 10 mg/litre to the aquaria where the tadpoles were kept. In all three groups the time of metamorphosis was delayed as compared with that of the controls. Histological examination of the thyroid glands revealed that in the three experimental groups the thyroids were less active than those of the controls.

RÉSUMÉ

ACTION DU FLUORURE DE SODIUM SUR LA MÉTAMORPHOSE DES TÊTARDS

Le fluorure de sodium, ajouté dans les aquariums à raison de 1, 2 et 10mg/litre ralentit la métamorphose des têtards de grenouille *(Rana temporaria)*. L'examen histologique des glandes thyroïdes montre une activité plus faible que celle des animaux de contrôle.

ZUSAMMENFASSUNG

DER EFFEKT VON NATRIUMFLUORID AUF DIE METAMORPHOSE DER KAULQUAPPEN

Natriumfluorid in Konzentrationen von 1, 2 und 10 mg/L verzögert die Metamorphose von *Rana temporaria* Larven. Die Schilddrüsen dieser Tiere zeigen histologisch verminderte Aktivität.

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