Uptake of ^{131}I by the Thyroid Gland during Metamorphosis in *Xenopus laevis*[1)]

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(*With 2 Text-figures*)

Morphological studies of the amphibian thyroid gland during metamorphosis have been well investigated by many workers as reviewed by Allen (1938), Etkin (1955) and Saxén *et al.* (1957 b). The gland in the premetamorphic stage is small and histologically inactive, but during prometamorphosis it grows rapidly both in absolute and relative size. The follicular epithelium of the thyroid gland increases in height, reaching a maximum at the beginning of the metamorphic climax. In *Xenopus* remarkable evacuation of colloid occurs at this stage (Saxén *et al.*, 1957 a, b). Subsequently, the height of the epithelium decreases with the absorption of the tail.

With the availability of radioiodine the quantitative study of the activity of the thyroid gland has been reinvestigated by several workers (Saxén *et al.*, 1957 a; Hunt and Dent, 1957; Kaye, 1961). Their results have indicated that the uptake of ^{131}I by the thyroid gland increases in the prometamorphic stage up to the eruption of forelimbs and then decreases in metamorphic climax. Therefore, it has been considered that the thyroid gland stores the hormone during the prometamorphic stage and releases it in metamorphic climax. Actually, the thyroxine requirement in the tissue has been shown to increase during metamorphic climax (Kollros, 1961; Henriques, 1962, 1964).

The present paper deals with the time curves of the ^{131}I uptake by the thyroid gland in *Xenopus* larvae at different developmental stages.

Material and Method: The larvae of the South African clawed toad, *Xenopus laevis* Daudin were obtained by induced ovulation with an injection of 200 IU of chorionic gonadotropin (Teikoku-zoki Co.) in the laboratory. They were reared in a large pond at 20°–25°C, and fed with alfalfa powder and dry milk. For the experiment of iodine uptake the larvae were kept in a radioactive iodide solution (200 μC, carrier-free ^{131}I per liter of water) at 24°±1°C. During the experiments the animals were not fed. They were fixed

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in Bouin's solution after 24, 48 and 72 hours of immersion, respectively, and their thyroid glands were carefully removed by using a watchmaker's forceps under a dissecting microscope. The radioactive iodine contained in the gland was measured with a well-type scintillation counter. All the mean values for the iodine content of the gland represented at least 6 animals in each developmental stage according to Nieuwkoop and Faber (1956). Since the animals varied considerably in size, the iodine content of the gland was expressed per unit of fresh body weight. Furthermore, the thyroid gland count was divided by the count of 0.1ml of immersion solution in order to compare the uptake of ^{131}I by the thyroid gland in each day.

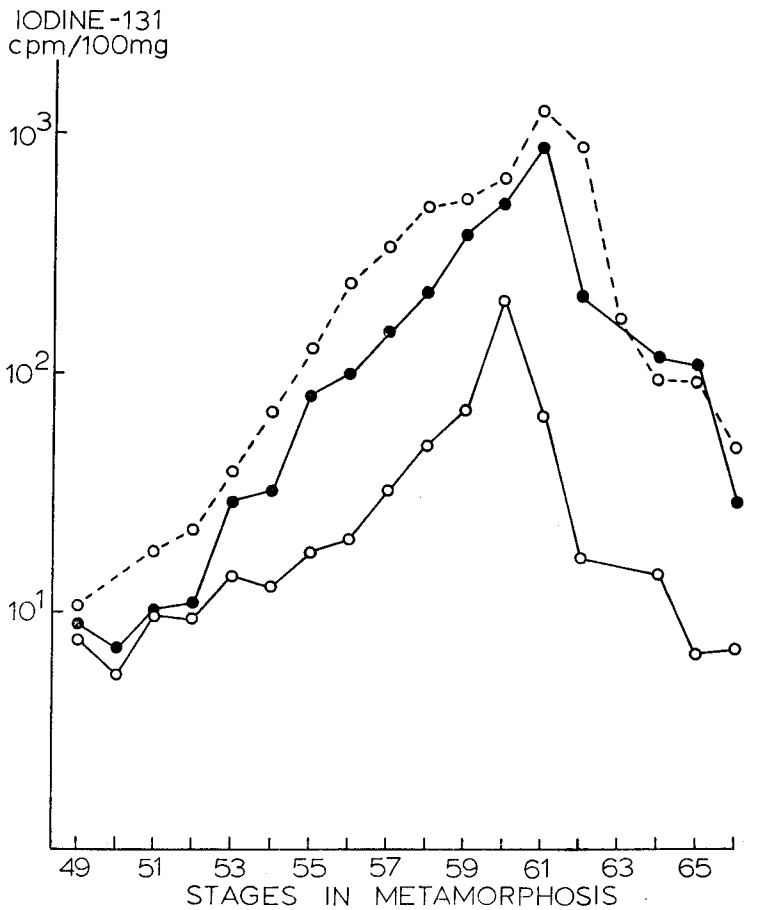

Fig. 1. The uptake of ^{131}I throughout the metamorphosis after 24, 48 and 72 hours of treatment. For explanation of figure, see text.

Results

Figure 1 shows the ^{131}I uptake by the thyroid gland in each stage during the metamorphosis. The uptakes after 24, 48 and 72 hours of immersion in radioactive iodide solution are represented by a solid line connecting open circles, a solid line connecting solid cricles and a borken line connecting open circles, respectively. It was clearly shown that the ^{131}I uptake was very low in level during the premetamorphic stage (49–53) and then increased considerably in the prometamorphic stage (54–58) regardless of the time of immersion. In every developmental stage the uptakes after 48 or 72 hours of immersion became higher than that after 24 hours of immersion. The maximum uptake of ^{131}I was observed at stage 60 after 24 hours of treatment, whereas in the tadpoles treated for 48 or 72 hours the uptakes reached the maximum at stage 61.

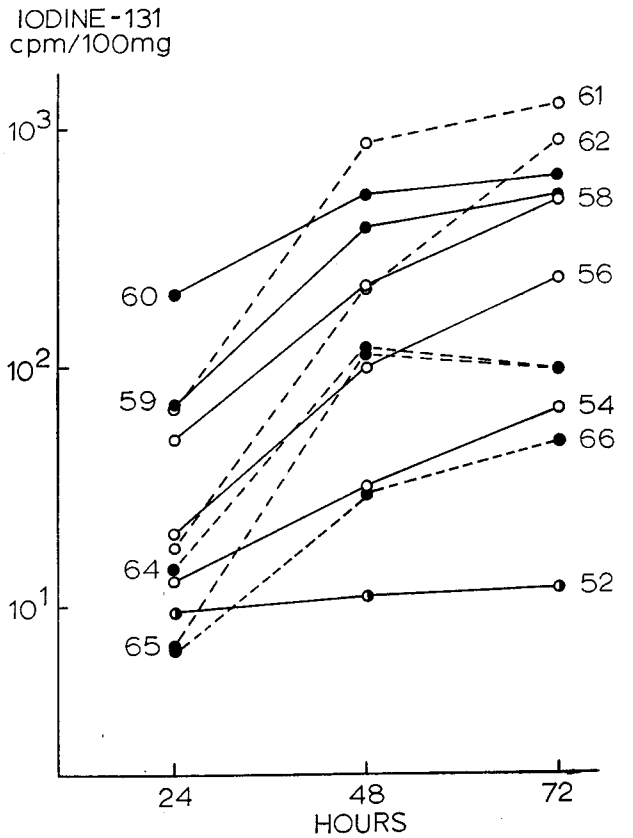

Fig. 2. Time curves of ^{131}I uptake after treatment in each metamorphic stage. For explanation of figure, see text.

The time curves of the ^{131}I uptake in each developmental stage after treatment are shown in Figure 2, in which the numbers indicate the developmental stage of the larvae. The mode of the ^{131}I uptake by the thyroid during metamorphosis can be divided into the following five types. In the first type, observed from stage 49 to 53, the maximum uptake was reached in 24 hours and the constant level followed (indicated by solid lines connecting semisolid circles). In the second type the uptake gradually increased up to 72 hours. This type was observed during the prometamorphosis from stage 54 to 58 (indicated by solid lines connecting open circles). At the beginning of the metamorphic climax (stages 59 and 60), the uptake increased rapidly for 24 hours and a relatively slow increase followed. This characterizes the third type (indicated by solid lines connecting solid circles). The fourth type was seen in the middle of the metamorphic climax at stages 61 and 62. The uptake at these stages was relatively low at 24 hours of immersion, but after 72 hours of immersion it reached the highest level (indicated by broken lines connecting open circles). The fifth type was seen near the end of the metamorphosis (stages 64 and 65), when the uptake by the thyroid gland again decreased to the level at the prometamorphic stage (indicated by broken lines connecting solid circles).

Discussion

The real rate of iodine uptake by the thyroid gland in each developmental stage may be indicated by the initial velocity of the ^{131}I uptake rather than by the maximum uptake. The initial velocity, which is represented by the uptake in the 24-hour immersion in this study, means the affinity of iodine to the thyroid gland, whereas the maximum uptake may indicate the capacity of iodine in the thyroid gland.

From the study of the time curves of the ^{131}I uptake it was demonstrated that the rate of iodine uptake increased during the prometamorphic stage and reached the maximum at stage 60 (Fig. 2). Saxén *et al.* (1957 a, b) and Kaye (1961) have also observed the increase of iodine uptake up to the beginning of the metamorphic climax in *Xenopus laevis* and in *Rana pipiens*. According to Saxén *et al.* (1957 a, b) the intensive hormone synthesis occurs in the thyroid gland during the prometamorphic stage but its release from the gland does not increase, and the synthesis of the thyroid hormone at this stage proceeds without the stimulation by TSH from hypophysis. However, hypophysectomized tadpoles only advance to stage VII in *Rana pipiens* (Kollros and McMurray, 1956) or stage XIII in *R. japonica* and *R. ornativentris* (Hanaoka, 1966). These tadpoles required a certain concentration of thyroxine to complete the prometamorphic stage. Moreover, the uptake of ^{131}I in hypophysectomized tadpoles of *R. japonica* and *R. ornativentris* was remarkably less than that of intact tadpoles in the prometamorphic stage (Hanaoka, unpublished data). These results indicate that the thyroid gland of these animals in the prometamorphic stage secretes a small amount of hormone under the influence of TSH from hypophysis. It may be concluded from available

data both the synthesis and the release of hormones gradually increase in both the thyroid gland and the hypophysis during the prometamorphic stage.

Interesting time curves of the ^{131}I uptake by the thyroid were seen at the middle of the metamorphic climax (stages 61 and 62). After 24 hours of immersion the uptake of ^{131}I at these stages was lower than that at stage 59 or 60, showing a decrease of iodine uptake rate at these stages. After 48 or 72 hours of immersion, however, the uptake reached higher levels as compared with that at stage 59 or 60, showing the increase of capacity in the gland for iodine uptake. This increase may be caused by the release and loss of iodine already present in the gland. Therefore, the gland at these stages seems to be in the most active phase of hormone release throughout the metamorphosis.

During the metamorphic climax the ^{131}I uptake again decreased. However, the uptake of ^{131}I is only a part of the thyroidal function, and its degree does not always indicate the degree of release of the thyroid hormone. Although the secretion of thyroid hormone has never been established in the tadpoles, protein-bound iodine (PBI) in the serum of the tadpole may be considered as indicating the releaseing activity of thyroid hormone. Saxén *et al.* (1957 a, b) have reported that the content of PBI rises during the metamorphic climax up to the end of the metamorphosis. Actually, the thyroxine requirement in tissue increases up to the end of the metamorphic climax in *Xenopus* (Henriques, 1962, 1964). Allen (1938) and Etkin (1936) have interpreted that the thyroid at the metamorphic climax intensively secretes the hormone, although the decrease in hormone synthesis occurs. Saxén *et al.* (1957 a, b) were in agreement with these investigators. It is generally accepted that the increase in thyroid hormone secretion is the earliest detectable response to the treatment by the thyroid stimulating hormone (TSH).

The TSH is found to cause the thyroid to release its hormone within 30 minutes in dogs (Ackerman and Arons, 1958) and in rats (Rosenberg *et al.*, 1960; Taurog *et al.*, 1964). On the other hand, the effect of TSH on the thyroid's iodine uptake is remarkably slow, the latent period being at least 8 hours in man (Stanly and Astwood, 1949) and in rats (Halmi *et al.*, 1953, 1960; Taurog *et al.*, 1958). These facts suggest that the TSH released at the metamorphic climax first induces hormone release in the thyroid and then the iodine uptake at later stages. The increases of the ^{131}I uptake after the end of metamorphosis was observed in the thyroid gland of *Xenopus* (Saxén *et al.*, 1957 b).

Summary

The time curve of the ^{131}I uptake by the thyroid gland in each metamorphic stage was determined in the present experiment. The rate of the ^{131}I uptake was slow in the premetamorphic stage (49–53) and increased in the prometamorphic stage (54–58), reaching the maximum at stage 60. These facts may indicate that the synthesis of thyroid hormone increases gradually in the prometamorphic stage and reaches the maximum at stage 60.

In the middle of the metamorphic climax (stages 61 and 62) initial velocity became decreased, although the uptake after 72-hour immersion reached the highest level. It is surmised that the releasing function of the thyroid was very active at these stages.

Near the end of metamorphosis (stages 64 and 65) the synthetic activity of the thyroid gland seems to become inactive.

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