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GROWTH OF THYROIDLESS TADPOLES

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ABSTRACT

The growth rates of thyroidless and control tadpoles of *Rana catesbeiana* and *Rana plancyi* were compared respectively and the comparison was also made on thyroidless tadpoles between the 2 species. The following results were thus obtained. Before the onset of metamorphic climax, controls and experimentals grew with a similar rate; after that the thyroidless tadpoles outgrew the controls in body weight and total length. Under the same laboratory conditions, thyroidless tadpoles of *Rana plancyi* became senile earlier than those of *Rana catesbeiana* because of a shorter period of metamorphosis acquired by the former. The relationship of growth of body weight and total length in thyroidless tadpoles followed the equation of Huxley, $y=bx^k$, thus simulating the growth of fishes. The ratio of tail length over body length in thyroidless tadpoles increased at first and then remained constant while the body form of controls was unchanged as the tadpoles grew old.

Thyroidectomy in frog tadpoles was first performed by Allen in 1916 (1). The operation consisted of the extirpation of thyroid anlage at very early embryonic stage when the function of the gland was far from beginning. However, unlike the thyroidectomized mammalians, the thyroidless tadpoles live normally; albeit they do not metamorphose. In our laboratory some thyroidectomized tadpoles kept on growing up to the age of four and half years and still remained at the hindlimb bud stage. It seems that permanent dissociation of differentiation and growth is possible in tadpoles. It should be interesting to observe, under this condition, the growth of these thyroidless tadpoles in terms of species difference, aging process and change of body form which were seldomly touched by the earlier workers such as Allen (2) and Hoskins (3).

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MATERIALS AND METHODS

Larvae of *Rana catesbeiana* raised from eggs of induced breeding were surgically thyroidectomized at the gill circulation stage. Two months after operation, 50 tadpoles were reared in an aquarium (32 × 26 × 12 cm) containing 4 liters of dechlorinated tap water. Fifty normal tadpoles in the same alloted aqueous space served as the control group. Whenever an unsuccessfully thyroidectomized tadpole with growth of hindlimb beyond stage

IV of Taylor and Kollros (4) was found, it was removed together with a control at the same time. Thus a final number of 30 thyroidless and of 30 normal tadpoles in each group was maintained.

In order to give the growing tadpoles enough space, the aquaria water of the 2 groups of tadpoles was increased gradually up to 8 liters at the end of 4 months. Thereafter, the tadpoles were separated in equal number into 2 aquaria with the same amount of water at every 4-month interval until a final space of 3 or 4 tadpoles per aquarium with 8 liters of water was reached.

The 2 groups of tadpoles were fed with thawed leaves of water-convolvulus *ad libitum.* The aquaria water was changed thrice weekly. Room temperature was controlled at 20 ± 1 C. Ten hours of incandescent illumination was given daily.

The body weight of tadpoles was measured with a torsion balance to the hundredth of a gram and the stage of development was examined monthly. Total length, tail length, body length and body width were measured to the nearest mm from the photographs of the tadpoles taken once a month. The control tadpoles took 18 months to complete metamorphosis while the thyroidless ones were observed for 25 months.

Pari passu with the observation on the growth of *Rana catesbeiana* tadpoles, that of another species, *Rana plancyi,* was carried out. This is a local species with a much shorter tadpole life of approximately 4 months under laboratory conditions. The growth of 30 normal and 30 thyroidless tadpoles of this species was observed for 36 weeks beginning 6 weeks after the operation. The aquaria space was increased gradually from 80 ml to 400 ml per tadpole. All measurements were taken biweekly. Other conditions were the same as for those of *Rana catesbeiana.*

RESULTS AND DISCUSSION

I. *Outgrowth of thyroidless tadpoles over the controls*

The growth curves in terms of body

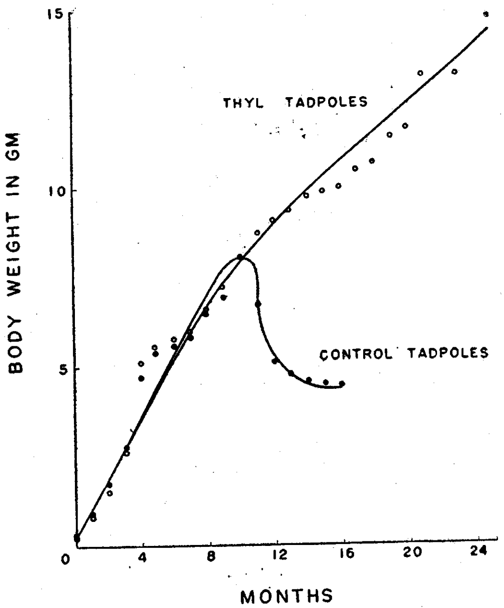

Fig. 1. Growth curves of body weight of *Rana catesbeiana* tadpoles.

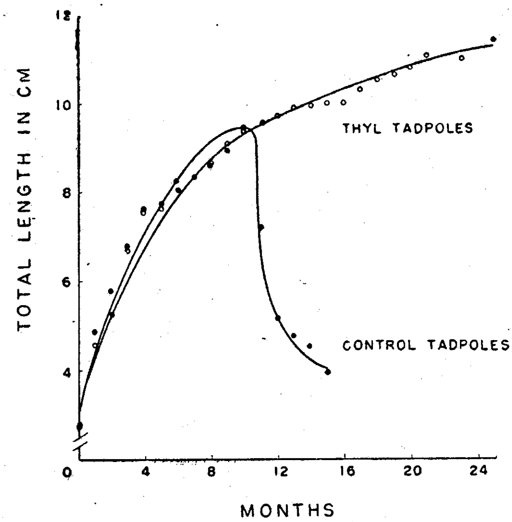

Fig. 2. Growth curves of total length of *Rana catesbeian* tadpoles.

weight and total length of *Rana catesbeiana* and *Rana plancyi* are shown in *Figs. 1-4* respectively. The trend of growth for the 2 species was similar. Control and thyroidless tadpoles increased their body weight and total

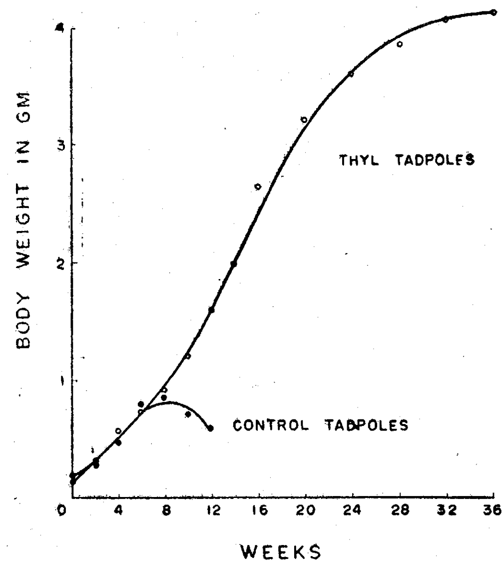

Fig. 3. Growth curves of body weight of *Rana plancyi* tadpoles.

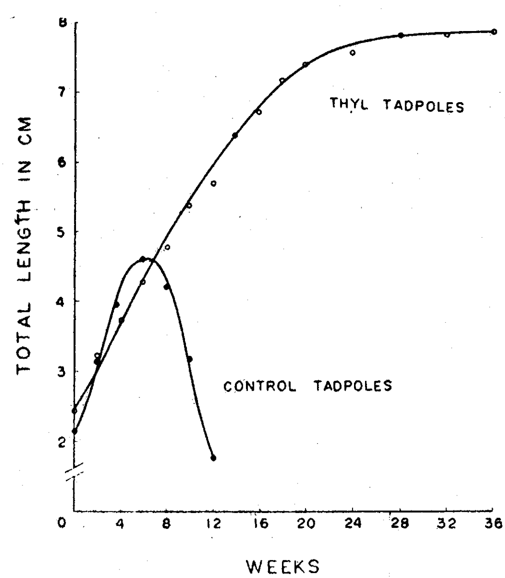

Fig. 4. Growth curves of total length of *Rana plancyi* tadpoles.

length at approximately the same rate until the climax of metamorphosis set in. Thereafter, the growth rate of controls declined while that of the thyroidless tadpoles kept on growing at a slower pacer until nearly a stand still was reached. The age of the thyroidless tadpoles of *Rana plancyi* to reach this limit, according to *Figs. 3* and *4,* was 42 weeks; approximately 2.3 folds of 18 weeks required to complete metamorphosis. The completion of metamorphosis for *Rana catesbeiana* required 18 months and therefore the terminal phase of growth was estimated at 42 months (180 weeks) after fertilization under the present conditions.

Since decrease in growth rate is one of the expression of aging, the terminal growth phase of thyroidless tadpoles of *Rana plancyi* at the age of 42 weeks might be considered senile effect. The phenomenon of aging was in accord with that of *Rana catesbeiana* in a previous report (5).

The aging difference for the terminal phase of growth in thyroidless tadpoles of the 2 species under the same environmental conditions might result from the unequal rates of metamorphosis and growth. The biological principle that an inverse relationship exists between the degree of differentiation and the rate of growth holds that the more highly an organism is differentiated the slower it grows. The high energy requirement of growth in the tadpoles of *Rana catesbeiana* reduces their speed of differentiation, thus prolonging the period of metamorphosis. On the other hand, tadpoles of *Rana plancyi* shows a faster rate of metamorphosis and thus a lower rate of growth. According to Voitkevich this specific characteristics in amphibia results from the dominance of acidophils and deppression of basophils or *vice versa* in the pituitary (6). When metamorphosis was precipitated by thyroidectomy, acidophils on account of either greater number or higher activity might augment the growth rates, leading thus to the outgrowth of thyroidless tadpoles of *Rana catesbeiana* when compared to those of *Rana plancyi.*

II. *Relationship between growth of body weight and of total length in thyroidless tadpoles*

When body weight of the thyroidless tadpoles of *Rana plancyi* was plotted against the total length on a double logarithmic grid, the result was a straight line as shown in *Fig. 5* which bears a resemblance to the equation of Huxley, $y=bx^k$, where x and y be the magnitudes of 2 correlative parameters; b and k are constant (7). K is an important value derived from the slope of the line and represents the regression of body weight (y) on total length (x). It is of interest to note that the comparable data of *Rana catesbeiana* fell within the same straight line, meaning that k values of both species were identical. In the present case k approximated 2.8 which was within the range of the exponential of fish growth, 2.5 to 4.0 (8). This finding suggested a very close resemblance between growth of thyroidless tadpoles and fishes in respect to the correlation of body weight to total length.

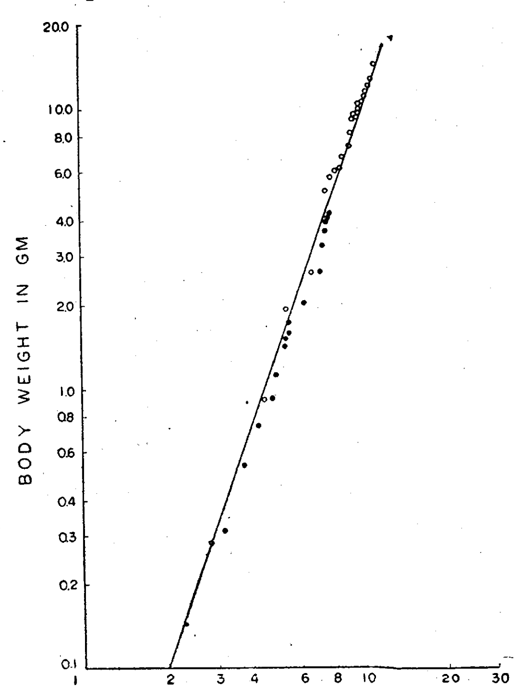

Fig. 5. Correlation between body weight and total length of thyroidless tadpoles in double logarithmic grid; o, *Rana catesbeiana;* •, *Rana plancyi.*

III. *Growth of the form of tadpoles*

In order to compare the shape between control and thyroidless tadpoles during growth and aging, curves in *Figs. 6* and *7* were constructed. *Fig. 6* shows the change of ratio between tail length and body length of *Rana plancyi* as the tadpoles grew. For controls it was only natural that the decreasing ratios brought the curve to zero as metamorphosis was completed after tail absoption. On the contrary, the same ratios for thyroidless tadpoles maintained a slowly upward trend indicating outgrowth of tail over body length and then the ratio was kept constant. *Fig. 7* indicates the change of body form. In controls, the body became elongated during metamorphosis while that of thyroidless tadpoles was kept approximately the same shape through-

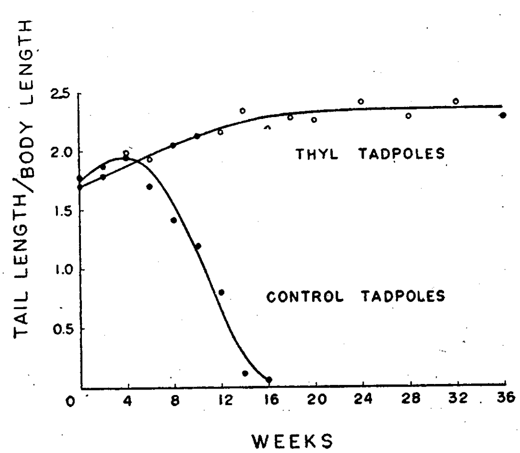

Fig. 6. Change of the ratio of tail length over body length during growth of *Rana plancyi* tadpoles.

out the observation period. The difference lay in the fact that the alimentary tract of controls underwent a process of histologic reorganization, resulting in reduction of intestinal length during metamorphosis while that of thyroidless tadpoles did not.

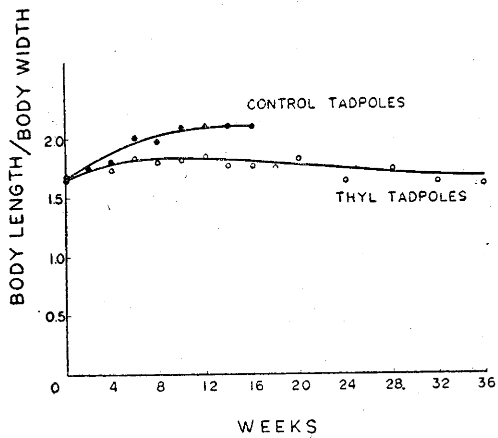

Fig. 7. Change of the ratio of body length over body width during growth of *Rana plancyi* tadpoles.

IV. *Discrepancies between the present data and those of earlier workers*

Hoskin and Hoskins (3) held that when controls developed to their maximal total length, the thyroidless tadpoles had gained very much in size over the controls. However, the statement had no statistical support and the laboratory conditions were not defined. The growth curves in the present study showed that before the development of maximal total length controls and experimentals progressed with the same rate and that it was only after the metamorphic climax that the experimentals outgrew the controls.

Allen (2) and Hoskin and Hoskins (3) reported that after thyroidectomy sex differentiation went on normally as testes became fully mature and ovaries were large and filled with oocytes. However, in the present study all thyroidless tadpoles of *Rana catesbeiana* were female after gonadal examination and all 60 thyroidless tadpoles of the same species in another study of ours (9) were also exclusively females. The discrepancy might be due to the different sex races employed in the 2 studies.

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