

THE EFFECT OF AUREOMYCIN ON TADPOLE DEVELOPMENT AS INFLUENCED BY TEMPERATURE¹

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ABSTRACT

Control and aureomycin-treated tadpoles of *Rana plancyi* were reared at different temperatures of 19 and 30 C respectively. At 19 C progress of metamorphosis and growth of total length of the treated tadpoles were inhibited while at 30 C tadpole development was not affected by aureomycin treatment. Observation on body weight of the froglets also showed that there was practically no difference between control and experimental groups at 30 C. The inhibiting effect of aureomycin at 19 C was considered to be due to its antibiotic activity and the indifferent effect at 30 C. was attributed to the loss of antibiotic potency after heat degradation.

Aureomycin is labile in aqueous solution and its stability is a function of temperature and pH (1-4). The heat- and alkali-degraded aureomycin lose their antibiotic potency (5-8). Hsü *et al.* reported that the alkalinized aureomycin promoted while the untreated aureomycin inhibited tadpole growth (9). It appears then that the inhibiting effect of aureomycin was closely related to its antibacterial activity. The present experiment was designed to test the effect of aureomycin which was decomposed by warm temperature on tadpole development.

MATERIALS AND METHODS

Fertilized eggs were obtained by induced breeding of the pituitary-injected frogs (10) of *Rana plancyi*. At stage 0 (11) when the hindlimb buds had not

yet been discernible, 30 tadpoles of the same size were reared in an aquarium containing 2,000 ml of tap water and another 30 with additional 5 ppm crystalline chlortetracycline HCl*. The 2 aquaria were kept in a water bath of 30 C maintained by heaters and thermostat. Two similar groups of 30 each at stage 0 were reared at 19.3 ± 0.5 C.

For the tadpoles at 30 C, the metamorphic stage and total length were examined biweekly during the first 6 weeks and weekly during the last 4 weeks while those of the tadpoles reared at 19 C were recorded biweekly throughout the experimental period of 20 weeks.

Tadpoles in the 2 series of experiments were fed par-boiled spinach *ad libitum* and the rearing media were changed 3 times a week.

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RESULTS

Solutions containing 5 ppm of aureomycin in tap water were of a light lemon color for at least 48 hours when kept at 19 C. However, at 30 C the same antibiotic solution changed into light brown color at the end of 12 hours, probably resulting from degradation of aureomycin.

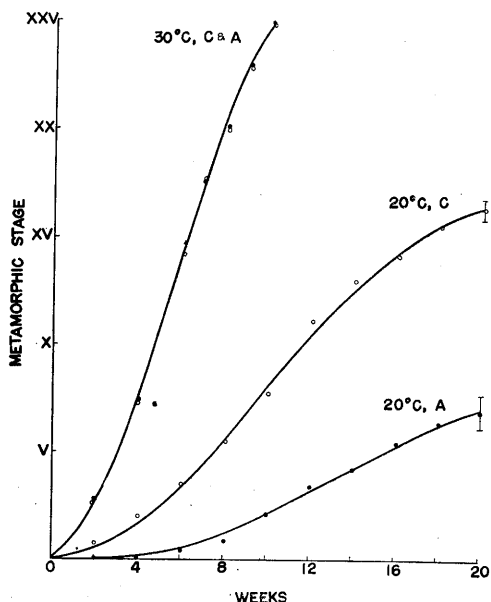


Fig. 1. Progress of metamorphosis of control (circles) and aureomycin-treated (dots) tadpoles under different temperatures. Vertical bars stand for standard errors.

Figure 1 indicates clearly that at 19 C aureomycin treatment inhibited tadpole metamorphosis with a difference of 9.8 ± 1.09 stages between control and experimental tadpoles at the end of the experiment; while at 30 C the 2 groups of tadpoles did not show any difference, both reaching stage XXV at the 10th week.

The development of total length in the 2 series of tadpoles as represented by growth curves in Figure 2 showed the same phenomenon that aureomycin exerted its inhibiting effect at 19 C but not at 30 C. At the latter temperature tadpoles had a shorter life history and

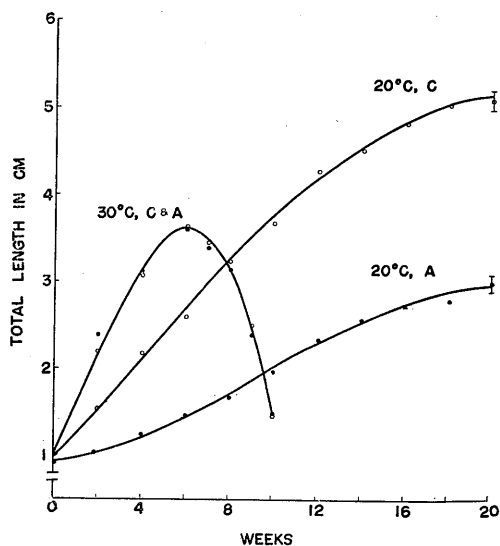


Fig. 2. Growth of total length of control (circles) and aureomycin-treated (dots) tadpoles under different temperatures. Vertical bars stand for standard errors.

their tails were beginning to regress at the 6th week while those reared at 19 C were still growing in length as illustrated in the growth curves.

A comparison of the final body weight at stage XXV (froglets) between the control and the experimental also indicated the lack of aureomycin effect at 30 C (344 ± 11 mg vs 345 ± 13 mg with a difference of 1 ± 17 mg).

DISCUSSION

The bacteriostatic action of aureomycin on micro-organisms is generally considered to be due to the interference with protein synthesis (12-15). This consideration may be applied to higher organisms too and the antibiotic nature of aureomycin could be extended to all living forms providing aureomycin is kept intact to maintain its antibacterial activity.

In this experiment, since degradation due to temperature was minimal at 19 C (7), it may be assumed that it was the antibiotic nature of the intact aureomycin which hampered tadpole metamorphosis and growth as compared with the

controls. The result confirms previous reports from this laboratory and elsewhere on *Rana catesbeiana*, *Rana limnocharis* (16), thyroidectomized tadpoles (17) and *Rana temporaria* (18).

Contrariwise, at 30 C aureomycin loses its antibacterial activity by heat degradation (7, 8) which explains why the antibiotic could no longer adversely affect the tadpoles under the present experimental conditions. However, this effect was different from that after alkali-degradation in which aureomycin was [shown to promote growth (9), probably due to different degradation products.

In regard to organisms other than tadpoles, aureomycin was shown to retard cell proliferation in tissue culture (19) and in protozoa (20) and to inhibit growth of guppies (21) and young rats (22). It was also reported that achromycin, another antibiotic of the same family to which aureomycin belongs, caused inhibition of the development of larval sand dollars (23), of chick embryos (24), of the teeth of rats (25) and of infants and new born rats (26). All these effects are considered to be due to the antibiotic nature of the tetracyclines which were preserved intact on parenteral administration, under low temperature or with large dosage, if the first 2 conditions were not present.

In temperate zones, 32 C is considered the upper limit of temperature range for tadpole development and this warmth could evoke sex reversal in tadpoles of *Rana sylvatica* (27). However, in the subtropical island of Taiwan the tadpoles such as those of indigenous *Rana plancyi*, *Rana narina* and *Rana limnocharis*, as well as the introduced species of *Rana catesbeiana* develop normally in the hot summer days when the outdoor temperature can be as high as 35 C. Hence 30 C is well tolerated by tadpoles in Taiwan.

Figures 1 and 2 also show the effect of temperature on normal development

of tadpoles. At 30 C it took 10 weeks for the tadpoles to complete metamorphosis up to stage XXV while at 19 C the same species of tadpoles developed to stage XVI only after 20 weeks. The result demonstrates very well the general biological principle that the relation between temperature and rate of biological process is characterized by the temperature coefficient $Q_{10}=2-3$ (28).

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