10539

Inducing Land Stage of *Triturus viridescens* to Assume Water Habitat by Pituitary Implantations.

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The studies of Gage[1] and Pope[2] have shown the life cycle of *Triturus viridescens* to consist of 3 phases. (1) an aquatic larval stage of about 3 months' duration; (2) an immature, red-colored land stage, lasting from 2½ to 4 years and, finally, (3) an aquatic adult stage. While there are some variations in this typical life cycle, apparently depending upon the locality,[3, 4] the newt in the Blue Ridge Mountains of extreme Western North Carolina shows exactly the life cycle described by Gage and Pope, with a possible extension of the second stage into a fifth year.

The life cycle of Triturus is very interesting because of the 2 complete changes in habitat that take place. First, at 3 to 6 months of age, the metamorphosis from the aquatic larval to the terrestrial condition with the accompanying migration to land, and second, in the fourth to fifth year, the migration back to water as the animal approaches or reaches sexual maturity with consequent readaptation to a water habitat.

Metamorphosis among the Amphibia has been shown to be essentially an effect of the endocrine glands, particularly of the thyroid and indirectly the pituitary.[5] In our investigations on Triturus we have been interested, not in particular morphological changes, but in the relationship that might possibly exist between the endocrine glands and the phenomenon of *migration* from land to water and *vice versa.* Do the pituitary and thyroid glands, in addition to bringing about certain morphological changes, provide the "drive" which induces the change in habitat following metamorphosis? In seeking the answer to this question we sought first to determine the relation of the pituitary, if any, to the return of the land stage of Triturus to water. Our method was to implant whole pituitaries from adult water-stage newts intramuscularly into immature land stages (red efts) of various sizes and ages. The implanted efts were then

1 Gage, S. H., *Am. Nat.*, 1891, **25**, 1084.
2 Pope, P. H., *Ann. Carnegie Mus.*, 1924, **15**, 305.
3 Noble, G. K., *Am. Mus. Novitates*, 1926, No. 228.
4 Noble, G. K., *Am. Mus. Novitates*, 1929, No. 348.
5 Allen, B. M., *Biol. Rev.*, 1938, **13**, 1.

placed in observation containers of such a nature that either a land or a water habitat could be elected. Controls, either unimplanted or implanted with bits of tongue, liver or brain tissue, were placed in the containers along with the experimental animals and all were observed over a period of 6 weeks.

Thirty-two red efts of sizes ranging from 46 to 90 mm in overall length and of ages ranging from 1 to 4 years were successfully implanted. The dosage varied from 1 to 5 adult pituitaries for each eft and was given over a period of 2 to 6 days. A slightly greater number of male than female pituitaries were used for implantation purposes. Of the 32 animals implanted, regardless of their size or age, 31 voluntarily assumed a water habitat in from 2 to 6 days (46 to 144 hours). Only one animal, 77 mm in length, failed to enter water and in this case autopsy revealed faulty implantation.

Usually on the third to fourth day following the initial implant, the efts were observed in the process of shedding their skins. These molts in most cases occurred before the animals went to water. After about a week in water the experimental animals began to darken perceptibly dorsally and become lighter ventrally. As time passed the color tended more and more towards that of the adult. In certain animals that were observed as long as 6 weeks, the formation of a tail keel was noted. In all cases the skin assumed the smooth and moist condition of the adult.

None of the control animals entered water in spite of repeated, forcible attempts to make them do so. To test the completeness of their adaptation to a land habitat a number of normal efts of differing age and size were placed in a deep battery jar of pond water in such a way that they could bring their external nares to the surface but could not crawl from the water. Not only did these animals constantly struggle to get out but the majority of them died within 1 to 7 days. The few survivors when allowed the choice of remaining in water or going to land, immediately returned to land.

To determine the particular lobe of the pituitary in which the water drive factor is produced, 6 efts were implanted with anterior lobes and 6 others with posterior lobes. The 2 lobes were carefully separated by dissection after the whole gland had been removed from the donor. The 6 anterior lobe-implanted efts showed exactly the same sequence of changes as did those implanted with whole pituitaries. The posterior lobe-implanted animals behaved exactly as unimplanted efts.

Microscopic study of the thyroids and gonads of the pituitary-

implanted efts revealed that the thyroids had been stimulated, but there were no changes in the gonads as compared with the controls. The microscopic changes in the skin, particularly in the lateral line organs, were definitely towards the adult condition.

Although Adams[6] and Dawson[7] were able to induce efts to acquire the skin color and texture of the adult, the former by injecting phyone (an anterior lobe extract), the latter by intraperitoneal implants of frog anterior pituitaries, neither mention the assumption of a water habitat following such treatment.

Since the thyroids are stimulated by pituitary implants and since the thyroids have been shown to be necessary for normal metamorphosis among the Amphibia, we are conducting experiments at present to determine whether the effect of the pituitary in inducing the water drive is direct or through the intermediation of the thyroids. Though the experiments are by no means complete, pituitary implants have induced the water drive in 8 out of 8 completely thyroidectomized efts. We are testing also the effects of pituitary implants in completely gonadectomized animals.

These experiments demonstrate that the pituitary gland of adult *Triturus viridescens* produces a substance which, when properly administered, will induce the water drive in the red land stage of this species. The threshold dosage of pituitary substance necessary to bring about the water drive is low and the effect is reasonably quickly produced. The sequence of changes following implantation are, first, molting with the consequent assumption of a smooth, moist skin, second, the assumption of a water habitat and finally, a color change towards the adult condition.

[6] Adams, A. Elizabeth, *Anat. Rec.*, 1932, **52** (Supp.), 46.

[7] Dawson, A. B., *J. Exp. Zool.*, 1936, **74**, 221.