THE LARVA OF THE OAK TOAD, *BUFO QUERCICUS* HOLBROOK

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One aspect of systematic studies on anuran species is the establishment of criteria by which larvae can be unequivocally identified, particularly when samples are collected from areas in which two or more species coexist. In southeastern Louisiana and Mississippi, the genus *Bufo* is represented by four species, whose ranges partially or entirely overlap (fig. 1). Precise knowledge of the early development of each species should facilitate accurate diagnoses of larvae in mixed collections, even in samples derived from localities in which interspecific hybridization occurs (Volpe, 1956).

Tadpoles of *B. terrestris* and *B. fowleri* are distinguished primarily on the basis of the relative distribution of melanophores in the tail musculature (Gosner and Black, 1958). Larvae of the Gulf Coast toad, *B. valliceps,* differ mainly from those of the Southern and Fowler's toads in labial tooth row proportions and pigmentation pattern of the tail (Limbaugh and Volpe, 1957). The tadpole of the Oak toad, *B. quercicus,* has been reported to have a distinctive tail pattern. The pattern of "six or seven black saddles on the musculature, and with heavily marked upper tail crest" (Wright and Wright, 1949, p. 199) should serve to differentiate larvae of *B. quercicus* from those of other species of toads. However, such a supposition proves untenable.

The investigation reported here was undertaken to amplify the description of the larvae of *B. quercicus,* particularly with the view of establishing characters useful in identification.

Embryonic Development

The opportunity to study the early development of the Oak toad was afforded when several adults were collected, on May 27, 1958, from a rain pool in pine-oak woodlands 1.4 miles northwest of the junction of State Highways 604 and 43, near Pearlington, Hancock County, Mississippi, 2.6 miles east of the Louisiana border (fig. 1). Amplexing pairs were placed in a gravel-bottomed terrarium containing a small amount of pond water. Eggs were obtained from one mated pair. The eggs were laid in small groups, each consisting of a short strand or "bar" of three to six eggs. The entire complement deposited by the female was estimated at 500 eggs. The mean diameter of the vitellus was 1.09 mm (range of 21 eggs : 0.98 - 1.13 mm). Each egg was enclosed in a single layer of hyaline jelly; the diameter of this gelatinous envelope averaged 1.25 mm (range of 20 measurements : 1.20 - 1.34 mm). A thin layer of jelly of loose consistency, comprising the jelly tube, united the individual eggs. The jelly tube was drawn out and pinched off between individual bars of eggs (fig. 2).

Strands of fertilized eggs were transferred to finger bowls (350 ml capacity), each containing 200 ml of pond water. The development of the embryos was observed at approximately three-hour intervals. Stages in the embryological development were similar to those of *valliceps,* as illustrated by Limbaugh and Volpe (1957). As is characteristic of toads, the embryos hatched before they were motile (tail bud stage 17). Embryonic development was extremely rapid, in comparison with other species of toads known to the authors. At room temperature (27.5° - 29.5° C), the embryos hatched within 24 hours after fertilization. The embryos were noticeably smaller than those of *valliceps.* At stage 20, the onset of gill circulation, the average total length (snout to tip of tail) was 3.2 mm; at stage 25, the last of the prefeeding stages, 5.5 mm. Embryos of *valliceps* at equivalent stages measure 3.7 and 7.3 mm, respectively.

The mouthparts differentiated during the closing stages of the embryonic period. Cornified frameworks of the beaks and rudiments of the labial ridges were recognizable

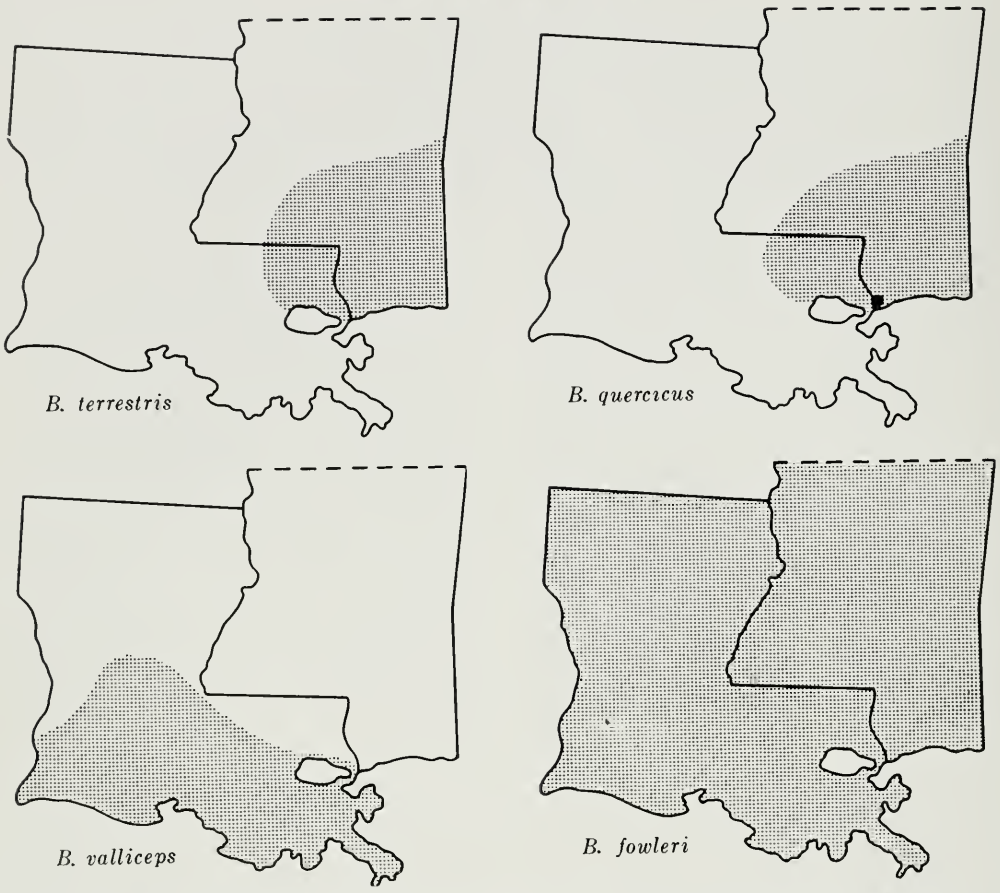

Figure 1. Approximate ranges of four species of toads in Louisiana and Mississippi. The black dot signifies the source of origin of the *B. quercicus* adults utilized in the investigation. (*Note:* The status of *B. fowleri* west of the Mississippi River is questionable.)

at embryonic stage 23. Teeth were evident at stage 24, arising in the first upper labial ridge or row. The inner edges of the horny beaks were pigmented and serrated. At stage 25, the lateral papillary fringes were emarginate and contained small papillae. Teeth were conspicuous on all labial ridges (two upper and three lower); however, no row possessed a full complement of teeth.

During the period between the origin and resorption of the external gills (stages 20-25), melanophores and xanthophores arose in the dorsolateral surfaces of the tail musculature. At stage 25, the last embryonic stage, the tail musculature was sharply bicolor; the dorsal half was darkened by melanophores, the ventral half was immaculate. Clusters of xanthophores occurred at irregular intervals along the dorsal edge of the tail musculature.

Larval Development

At the onset of feeding activity (stage 26, defined in Limbaugh and Volpe, 1957), the larvae were transferred to 12 X 8 X 2 inch enamel pans containing pond water to a depth of one inch. Each enamel pan, housing approximately 10 larvae, was kept at room temperature (27.5° - 29.5° C). The larvae were fed slightly boiled letture until the closing stages of metamorphosis.

The larvae were examined at approximately 24-hour intervals. Records were kept of the stages of development, distribution of chromatophores, and linear dimensions (snout to tail tip) of the larvae. Photographs were taken of living larvae (nar-

cotized in "M.S. 222"[1]) to reveal the distribution of formalin-soluble yellow chromatophores (xanthophores). The following discussion of larval characteristics emphasizes the structure of the oral disc, the size in relation to stage, and the pigmentation pattern of the tail.

Mouthparts.—The configuration of the oral disc is illustrated in figure 2. The labial tooth formula is 2/3. The first tooth row in the upper labium is continuous; the second is divided medially. The median space of the second upper tooth row is narrow, comprising 0.15 - 0.20 times the length of either lateral segment of teeth. The first

[1] Trade name for tricaine methanesulfonate, prepared by the Sandoz Chemical Works, New York, New York.

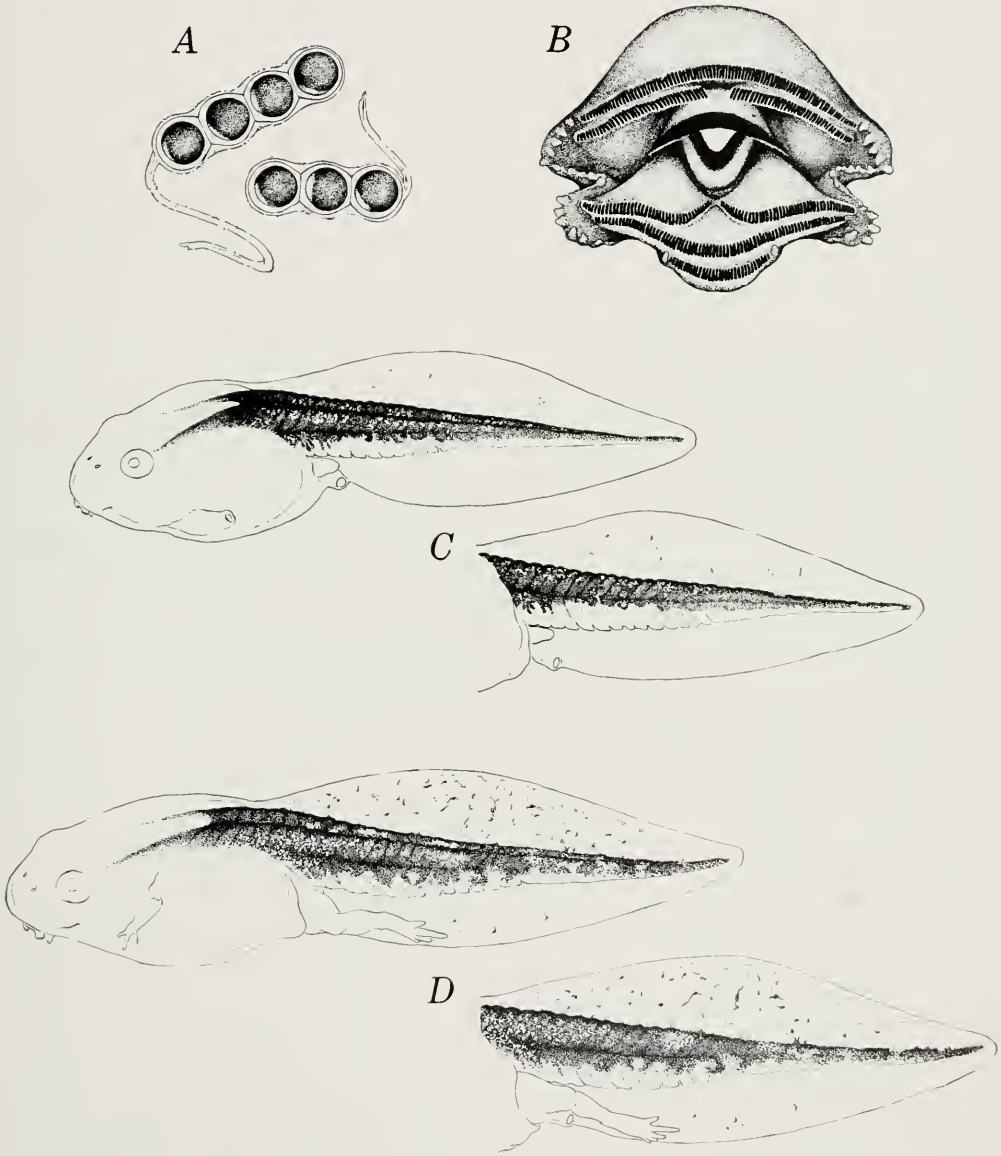

Figure 2. "Bars" of eggs (A); oral disc (B); oblique and lateral views of formalin-preserved larvae at stages 30 (C) and 41 (D). (Illustrated by Mrs. Carolyn Thorne Volpe.)

lower tooth row is interrupted medially by a short space[2] and rises to a point in the midline in the direction of the broadly V-shaped lower beak. The second lower row corresponds in outline to the first but is slightly shorter. The third lower tooth row is approximately half the length of the second lower row. The oral disc is emarginate. Marginal papillae are present along the sides of the upper and lower labia.

The relative lengths of the three lower tooth rows constitute the best criterion for distinguishing the oral discs of *quercicus* and *valliceps*. In the latter species, the three lower tooth rows are of nearly equal chord length (Limbaugh and Volpe, 1957; Gosner and Black, 1958). In mouthparts of larval *fowleri* and *terrestris*, the lateral papillae of the lower labium extend ventrad to slightly below the outer edge of the short third lower row of teeth (see fig. 10 in Volpe, 1956). The lower labial papillary fringe of *quercicus* extends to the outer edge of the second lower tooth row (fig. 2). In our sample, the third lower row is usually flanked on each side by a single, isolated papilla. We do not know whether this feature is generally characteristic of *quercicus* larvae, or is peculiar to the particular group of larvae studied.

Linear dimensions.—The size-stage relationships during larval development are graphically represented in figure 3. The growth curve of *quercicus* is an approximation based on small samples of two or three larvae at each stage. Obviously, greater numbers would result in a refinement of the curve. The present data, however, are of value in revealing the relatively small size of the tadpole of *quercicus*. The larvae of *terrestris, fowleri,* and *valliceps* are conspicuously larger. Gosner and Black (1958) found that the size-staging curves of the latter three species are nearly equivalent. Data presented in figure 3, based on large samples of laboratory-reared larvae of *terrestris, fowleri,* and *valliceps* from southeastern Louisiana, substantiate their findings.

Tadpoles of *quercicus* reached a maximum length of 19.4 mm at stage 41. The body lengths (snout to vent) of ten toads preserved at transformation averaged 8.2 mm (range: 7.2 - 8.9 mm). Transformation sizes recorded by Wright (1932) for the Oak toad are between 7 and 8 mm.

Pigmentation pattern of the tail.—Two types of chromatophores, the black (melanophore) and the yellow (xanthophore),[3] impart a characteristic pattern to the tail. The distribution of xanthophores is shown in photographs of living larvae at stages 30, 37, 39, and 41 (fig. 4). The appearance of the tails of two of the larvae (stages 30 and 41) after formalin preservation is illustrated in figure 2. The striking contrast before and after preservation reflects the fact that the pigment of xanthophores is soluble in formalin. It is evident that larval descriptions based solely on preserved specimens are not only incomplete but may be misleading.

The dorsal portion of the tail musculature is covered with melanophores; the greater part of the ventral half is devoid of melanophores. The dividing line between the melanophore-darkened dorsal region and the immaculate ventral area may be referred to as the "melanophore border". This border tends to be more ventrad in advanced larvae (*cf.*, stages 30 and 41 in fig. 4). The dorsal tail fin in our sample can not be designated as "heavily marked" (Wright and Wright, 1949). Rather, the dorsal fin is sparsely flecked with melanophores, the number varying with the stage of development. In general, the intensity of mottling increases progressively during development (fig. 4).

The greatest concentration of xanthopores occurs along the dorsal edge of the tail musculature and along the melanophore border. In a formalin-preserved specimen viewed dorsally or obliquely (fig. 2), a series of light areas may be seen in the dorsal edge of the musculature. These light areas represent the sites of the formalin-soluble xanthophores. In like manner, the melanophore

[2] The majority of larvae examined possessed a divided first lower tooth row. A median break in this row occurs (or is considered) as a variant condition in other species of toads.

[3] The term "xanthophore" is used with reservation. At least two types of chromatophores, xanthophores and leucophores, comprise the "light spots" of the tadpole. The predominant type appears to be the xanthophore (=lipophore), containing a yellow pigment. Others fit the description of an iridescent leucophore (=iridophore, guanophore), appearing silvery with reflected light. Additional studies are needed.

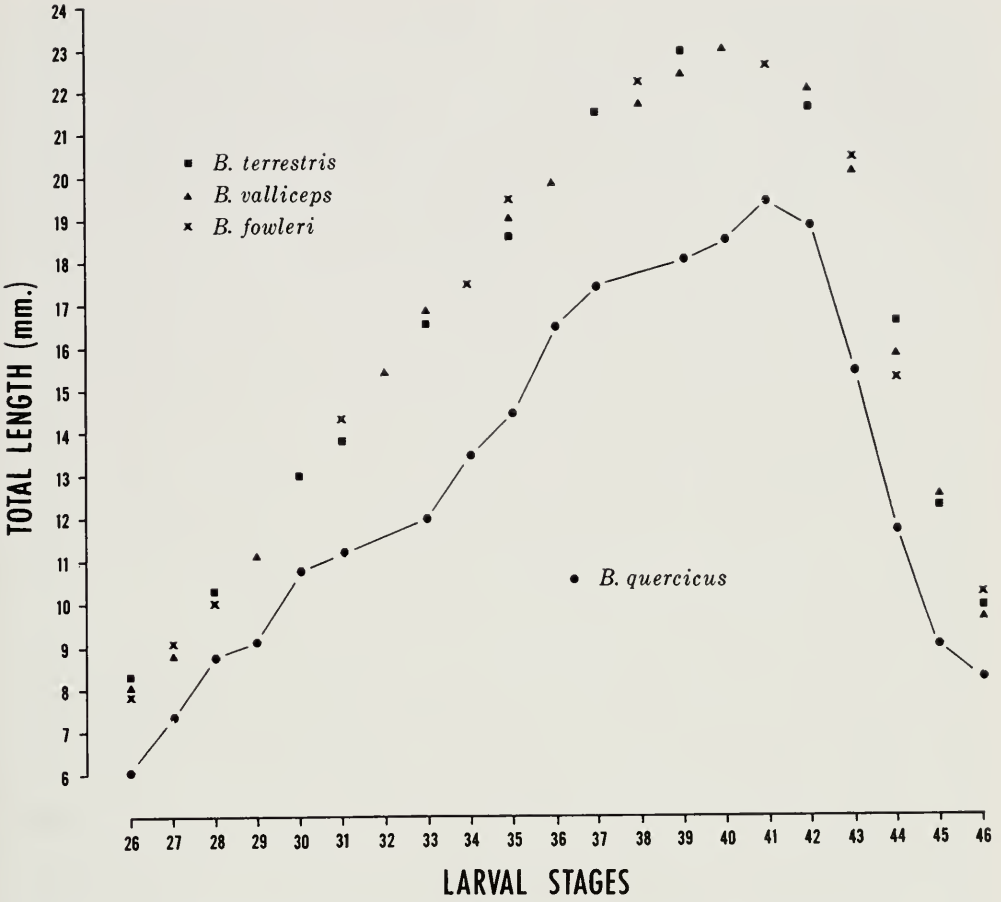

Figure 3. Average dimensions in millimeters of the total length of tadpoles of four species of toads during development. Stages of larval development are defined and illustrated in Limbaugh and Volpe (1957).

border in formalin-preserved specimens is irregular in outline, the indentations signifying those areas occupied by clusters of xanthophores. The scalloped appearance of the melanophore border may conceivably create an overall pattern effect of "six or seven black saddles on the musculature", as envisioned by Wright and Wright (1949).

Comparison of Tail Patterns

Xanthophores are prevalent also in the tail of the larva of *valliceps;* larvae of *terrestris* and *fowleri* contain relatively insignificant amounts of yellow chromatophores (fig. 5). Although the disposition of xanthophores in *quercicus* may be somewhat comparable, particularly along the dorsal edge of the musculature, to that of *valliceps,* larvae of the latter species can be distinguished readily on the basis of the amount and distribution of melanophores. In sharp contrast to *quercicus,* melanophores are distributed throughout the tail musculature in *valliceps* and form an elaborate, reticulate pattern in the dorsal tail fin.

Formalin-preserved specimens of *terrestris* and *fowleri* resemble superficially the tadpole of *quercicus,* especially when comparisons are made utilizing larvae of *terrestris* that possess unspotted dorsal fins (*cf., A* in fig. 5 with *C* in fig. 2). In each of these three species, the melanophore-darkened dorsal region contrasts sharply with the ventral area lacking melanophores. However, a close examination of the dorsal edge of the musculature of larvae of *quercicus*

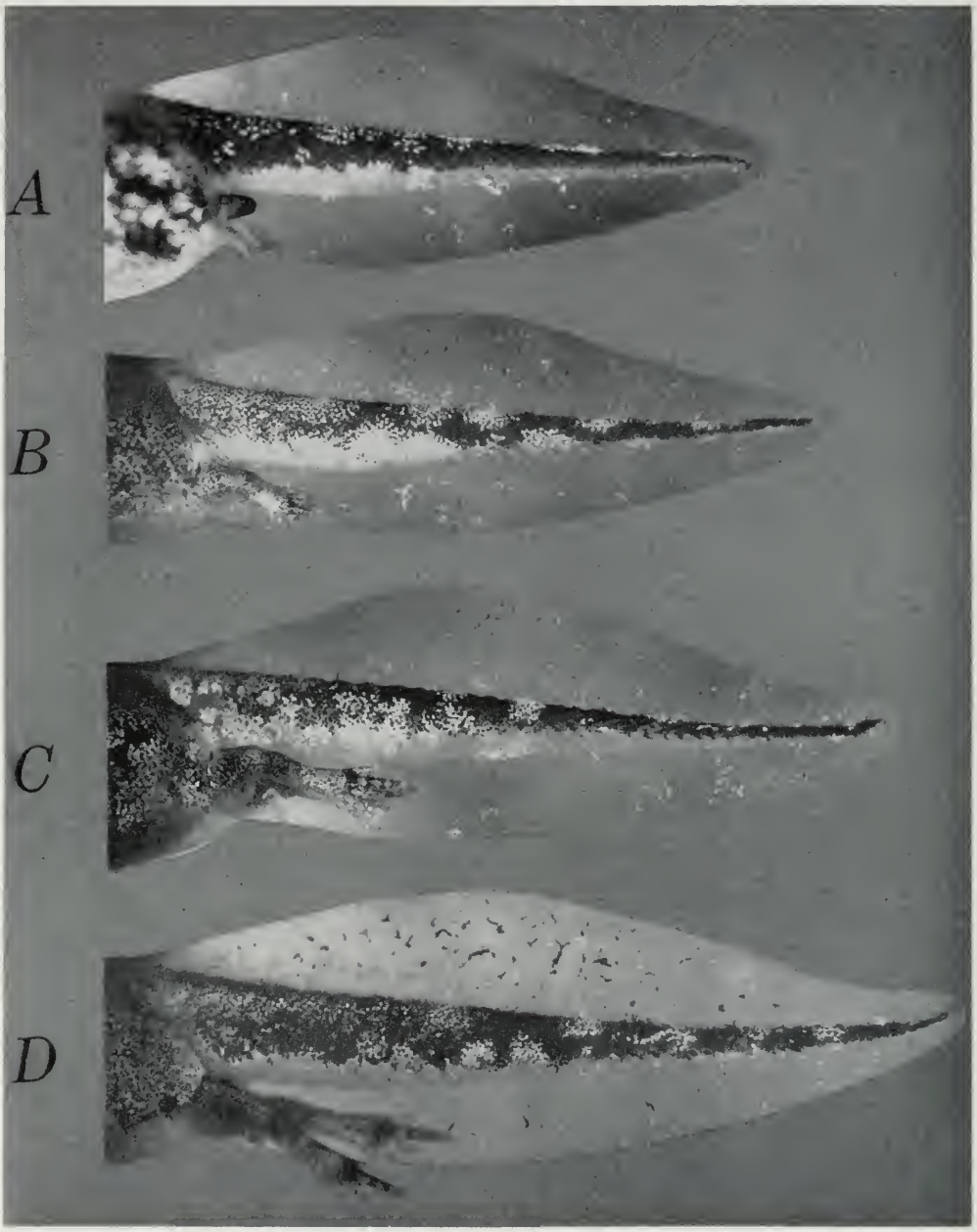

Figure 4. Lateral views of the tails of living larvae of *Bufo quercicus* at stages 30 (A), 37 (B), 39 (C), and 41 (D).

reveals light areas or patches representing the sites of formalin-soluble xanthophores, whereas the same region in larvae of *terrestris* or *fowleri* is covered uniformly with melanophores. Furthermore, the melanophore border tends to be more regular in outline in *terrestris* and *fowleri* than in *quercicus*.

Summary

The only previous description of the tadpole of the Oak toad, *Bufo quercicus*,

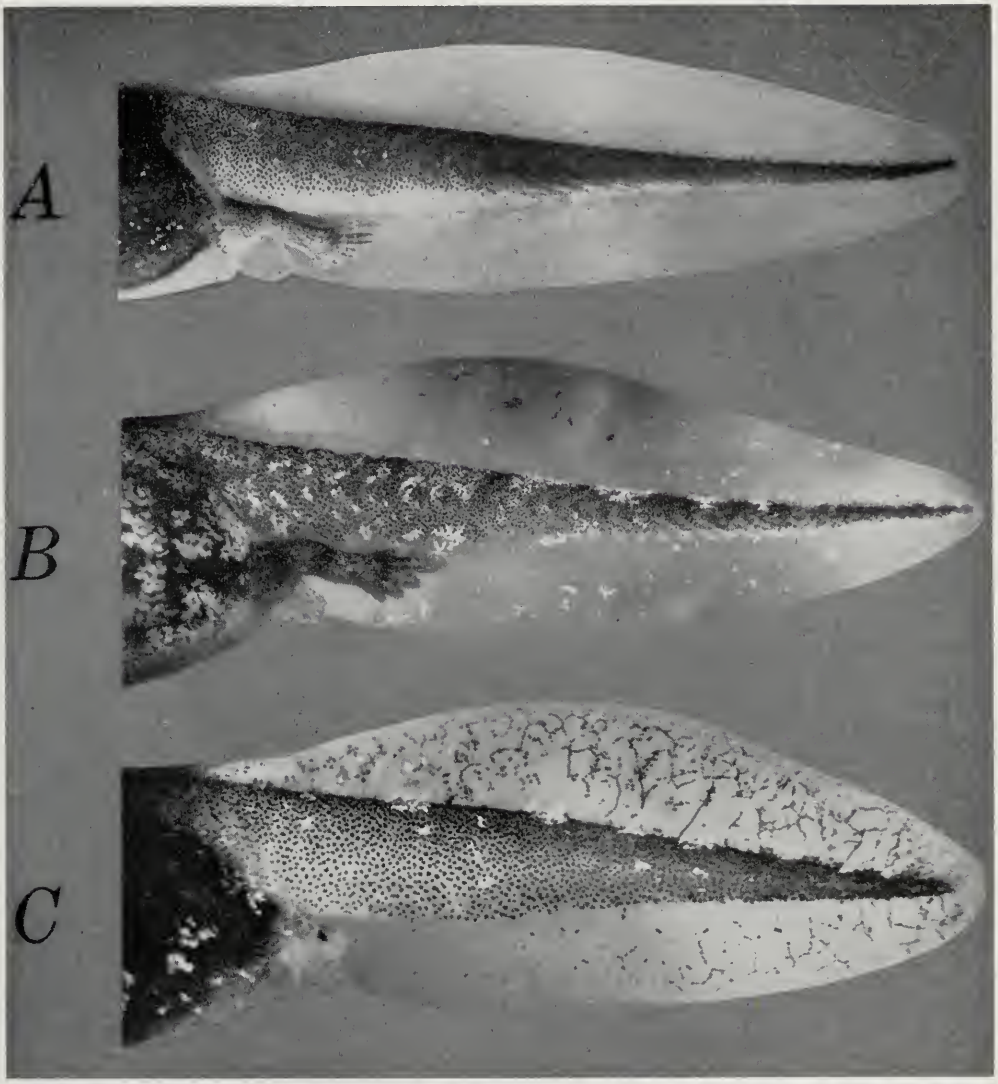

Figure 5. Lateral views of the tails of living larvae of *Bufo terrestris* (A), *B. quercicus* (B), and *B. valliceps* (C).

stemmed from studies of specimens collected from the Okefinokee swamp area in Georgia (Wright, 1932). A detailed description of laboratory-reared larvae derived from a single mated pair collected near Pearlington, Mississippi has been presented here. Emphasis has been placed on those characters that serve to differentiate these tadpoles of *B. quercicus* from those of *B. valliceps, B. terrestris,* and *B. fowleri.*

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