American Museum Novitates

PUBLISHED BY THE AMERICAN MUSEUM OF NATURAL HISTORY
CENTRAL PARK WEST AT 79TH STREET, NEW YORK, N. Y. 10024

NUMBER 2407 MARCH 11, 1970

Descriptive Notes on Larvae of Toads of the *debilis* Group, Genus *Bufo*

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Bufo debilis, Bufo retiformis, and *Bufo kelloggi* are three closely related species with a composite range that includes much of the southwestern United States and northern Mexico. Bogert (1962) described the distribution, morphology, and mating calls of these species and presented evidence for their specific rather than subspecific status, as had been advocated for some or all of the forms by earlier authors (Sanders and Smith, 1951; Savage, 1954; Riemer, 1955).

Only two authors have described tadpoles thought to be those of *Bufo debilis,* and the tadpoles of the other two species have been completely unknown. Smith (1934) described larvae from Comanche County, Kansas, and suggested that those were of *debilis.* The identity of the tadpoles was inferred from the known existence of *debilis* in the region and, evidently, also from the fact that the larvae were not readily assignable to another species. Bragg (1955) associated tadpoles and recently transformed *Bufo debilis* at the same pool in Oklahoma, and found that the tadpoles differed in some important respects from those attributed to *debilis* by Smith.

In the course of laboratory experiments on the temperature tolerances of embryos (Zweifel, 1968), I made a number of intraspecific crosses of *Bufo debilis, B. retiformis* and *B. kelloggi.* Relatively few tadpoles were obtained and these proved unusually difficult to raise. Consequently, it

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is not possible to give a thorough description of the larval stages through metamorphosis, but in view of the near lack of information on the larvae of these species it seems desirable to present such information as is available.

I have used the normal table for *Bufo valliceps* developed by Limbaugh and Volpe (1957) as the standard for describing the stage of development of embryos and tadpoles. Colors mentioned are those noted on the living animals. Specimens on which the descriptions are based are catalogued in the herpetological collection of the American Museum of Natural History (each number refers to one or more series of embryos and tadpoles bred in the laboratory). *Bufo debilis insidior:* A.M.N.H. Nos. 81546, 81547, parents from the vicinity of Portal and Douglas, Cochise County, Arizona, and from near Rodeo, Hidalgo County, New Mexico; *Bufo kelloggi:* A.M.N.H. No. 81548, parents from 22.5 miles west of Hermosillo, Sonora, Mexico; *Bufo retiformis:* A.M.N.H. No. 81549, parents from 22.5 miles west of Hermosillo, Sonora, Mexico.

Acknowledgments

This work stems from field and laboratory studies conducted in 1958 and 1960 at the Southwestern Research station of the American Museum of Natural History in Portal, Arizona. Mr. William A. Wimsatt (1958), Mr. F. Harvey Pough (1960) and Mr. Charles J. Cole (1960) assisted me. Mr. Wimsatt and Mr. Pough received support from the Lincoln Ellsworth Fund of the American Museum of Natural History; Mr. Cole was a participant in the Undergraduate Research Program, supported by National Science Foundation Grant EO/3/43-1606. General support came from National Science Foundation Grant G5033. The drawings illustrating the present paper were made by Frances W. Zweifel, who has my sincere thanks.

EGGS

The eggs produced in the laboratory were stripped or dissected directly from the oviducts. Hence, I cannot be certain of the normal condition of the clutch, but I am sure that the eggs would not be laid in the long, continuous strings characteristic of most species of *Bufo,* as species that lay eggs in this fashion (e.g., *Bufo cognatus*) also produce long strings when stripped, and the three species studied produced only short strings or irregular clumps.

The eggs of the three species studied are unusually pale for *Bufo*. Those of *debilis* and *retiformis* are yellow with a band of melanin approximately one-third the distance down from the animal pole with scattered melanin

over the rest of the egg, slightly denser above the aforementioned ring of pigment. The general effect is somewhat like that of a monk's tonsure. The eggs of *kelloggi* have more melanin than those of the other two species, but it is largely restricted to the upper one-third of the ovum (much less of a tonsure effect), and the lower two-thirds are creamy white. In more typical *Bufo* eggs the melanistic pigment is not only more intense but also covers more of the yolk.

"A sample of 25 ova of *B. debilis* averaged 0.98 mm., and another sample of 32 (measured before preservation) averaged 1.15 mm." (Zweifel, 1968, p. 54). Thirty-one ova of *B. kelloggi* averaged 1.17 mm. before preservation. Ova of *B. retiformis* are of essentially the same size, but I have no material in condition for accurate measurement.

GENERAL MORPHOLOGY OF TADPOLES

Tadpoles of the three species are quite similar in general appearance. The body is rounded (not depressed), with the spiracle almost halfway up the left side, and the anal tube opening on the right side of the tail fin. The eyes are situated dorsally, centered about midway between the midline of the body and its outer edge as viewed from directly above. The tail fins are moderately developed, neither so large as in pond-type larvae of other genera, nor so narrow as in stream-type larvae. The greatest distance from the upper edge of the upper tail fin to the lower margin of the lower fin averages 0.20 of the total length in 37 specimens of *B. debilis* (range 0.16–0.26) in stages 27 to 36. The means are similar in the other species: 0.22 (0.18–0.25, n = 12) in *kelloggi;* 0.19 (0.16–0.25, $n = 8$) in *retiformis.*

The length at hatching is approximately 3.1 to 3.4 mm.; six embryos (two *debilis,* three *kelloggi* and one *retiformis*) all fall within this range. None of the tadpoles was raised through metamorphosis, so the maximum size was not determined. The largest *Bufo debilis* larva has a total length of 25.1 mm. (body 9.9 mm., tail 15.2 mm.) in stage 36. I have no specimens of the other species of so large a size or so late a state of development.

Mouth Parts

The tadpoles of all three species have two rows of teeth anterior to the beak and two rows posterior when the mouth parts are fully developed. The second anterior row is broadly broken in the middle. The third and fourth rows are complete and are of virtually the same length, but are shorter than the first row. Papillae are confined to the corners of the mouth, as is typical of *Bufo.* The papillae are few in number, usually

eight to 10 on each side. I can detect no differences between the mouth parts of *debilis* and *kelloggi* (fig. 1). My specimens of *retiformis* have the labial teeth poorly developed, but no differences from the mouth parts of the other two species are evident.

Color and Pattern

Bufo debilis: The newly hatched larva (fig. 2, stage 19) is pale yellowish tan, with only a trace of dark pigment in the form of a few melanophores in the dorsal region of the body. Through stage 25 the tadpole remains essentially yellowish and translucent, although the number of melanic cells increases and they appear over much of the body and also on the tail musculature. Late in stage 25 the first xanthophores appear on the back and on the base of the tail.

In later stages (figs. 3, 4) the melanophores form an even stipple over the body and tail musculature. Patches of golden cells are present on the

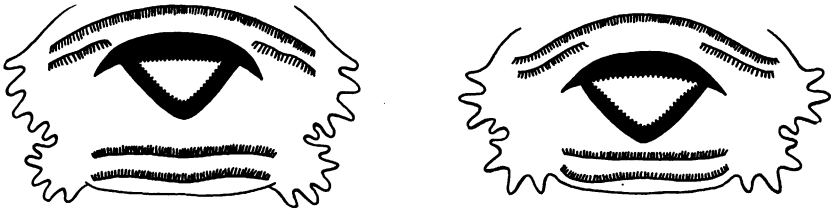

Fig. 1. Mouth parts of *Bufo debilis* (left, stage 31) and *Bufo kelloggi* (right, stage 33).

tail musculature, and such cells are more prominent than melanophores on the dorsal surface of the body. The deep abdominal region is black laterally with golden flecks, but the gular and mid-abdominal regions are without melanic pigment. The first melanophores appear in the dorsal tail fin in stage 28 (fig. 3) and gradually form a general peppering, occasionally with dendritic orientation. Only in the largest tadpoles examined (fig. 4, stage 36) was there melanic pigment in the ventral fin.

In general, these larvae are much paler than is usual in *Bufo*. In contrast to the dense black aspect common to many *Bufo* tadpoles, these are relatively transparent even when well grown.

Bufo kelloggi: The larva is yellow at hatching (stage 19, fig. 5). The first melanic flecks are present on the back at hatching or appear shortly thereafter. Late in stage 24 melanin forms lichen-like patches on the top and sides of the body and on the upper margin of the tail musculature. A few golden cells are mixed with the melanophores. The over-all color

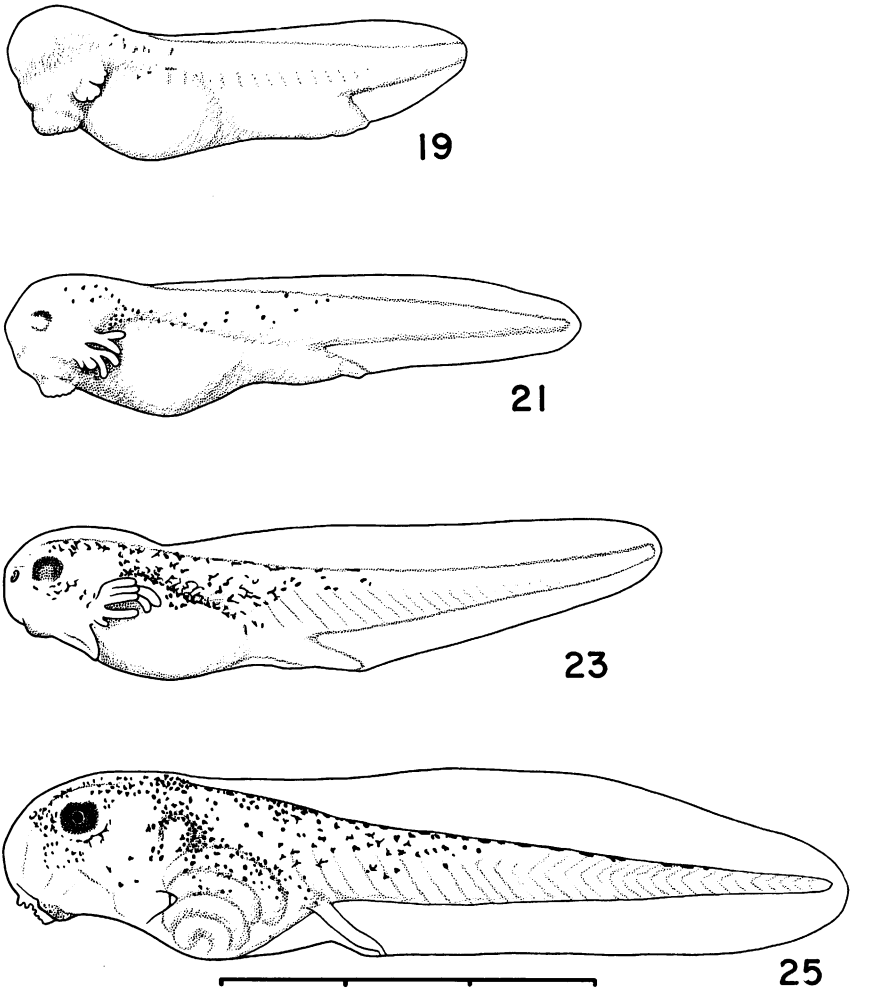

FIG. 2. Early stages of larval development in *Bufo debilis*. Numbers indicate stages of development; scale line represents a length of 3 mm.

of the larva is yellow, but the yolk-filled gut is, of course, brighter than the rest of the body.

The body and the upper parts of the tail musculature develop an even and relatively dense distribution of melanophores in later stages (figs. 6, 7). Golden cells are distributed throughout the melanophore-covered parts of the body but are less abundant on the tail. In the largest individual available (stage 33), there are only a few flecks of melanin in the

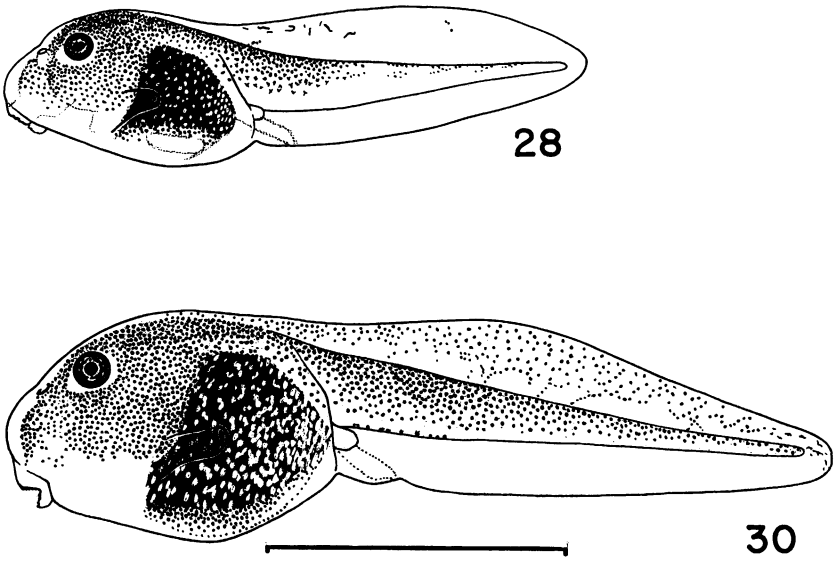

FIG. 3. Larvae of *Bufo debilis*. Numbers indicate stages of development; scale line represents a length of 5 mm.

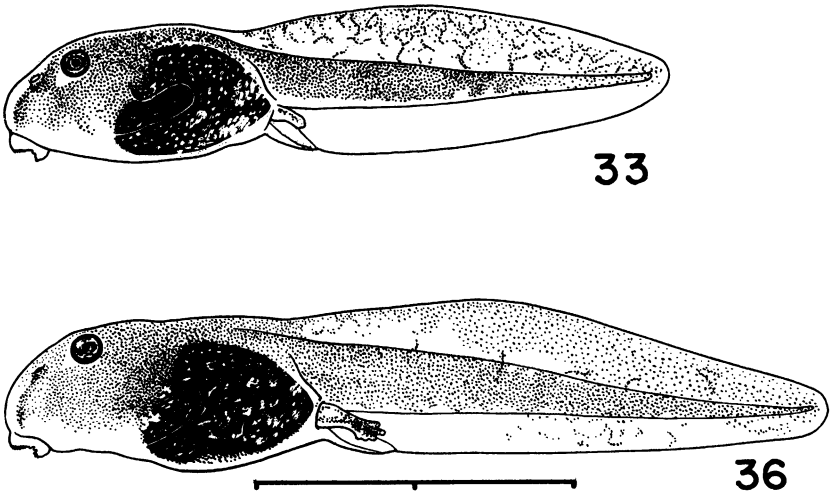

FIG. 4. Larvae of *Bufo debilis*. Numbers indicate stages of development; scale line represents a length of 10 mm.

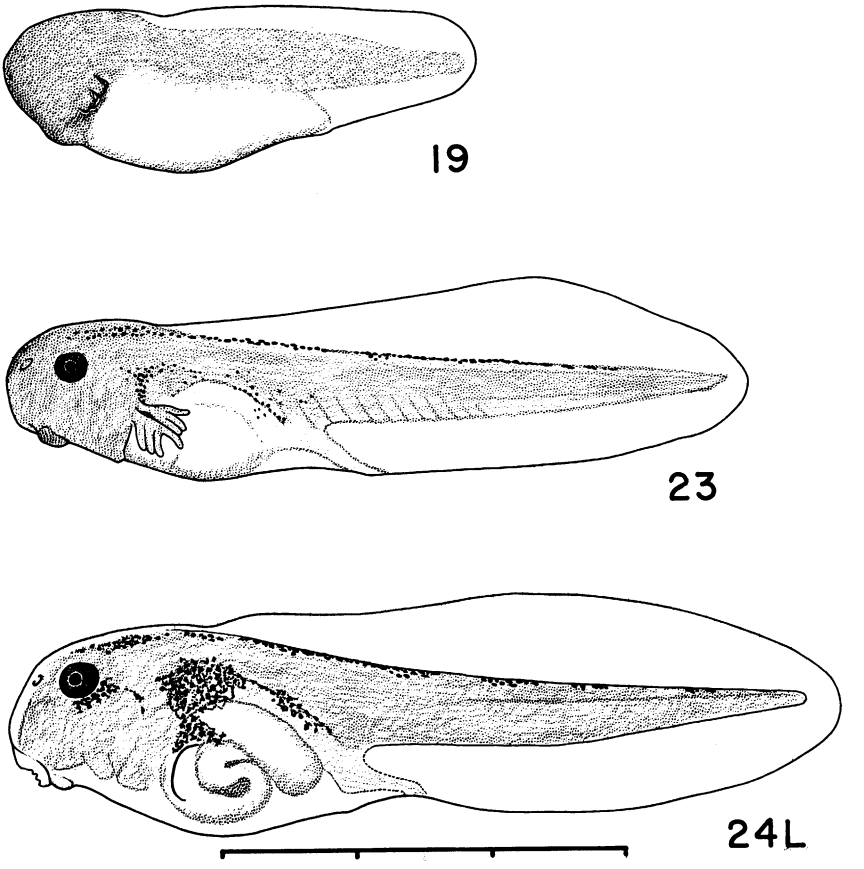

FIG. 5. Early stages of larval development in *Bufo kelloggi*. Numbers indicate stages of development; scale line represents a length of 3 mm.

ventral region of the tail musculature, in contrast to the condition in *debilis* (fig. 4) and in *retiformis* (fig. 9). Dendritic aggregations of melanophores appear in the dorsal tail fin first in stage 28 (fig. 6), and a few melanophores are seen in the ventral fin of later stages. The abdomen has a deep layer of dense melanophores overlaid with a stippling of xanthophores.

Bufo retiformis: Development of pigmentation in this form is much the same as in *B. debilis* and *B. kelloggi*. The hatchling (fig. 8, stage 19) is yellow with some scattered melanic pigment. Melanophores gradually appear over the body and tail musculature, and some golden cells are

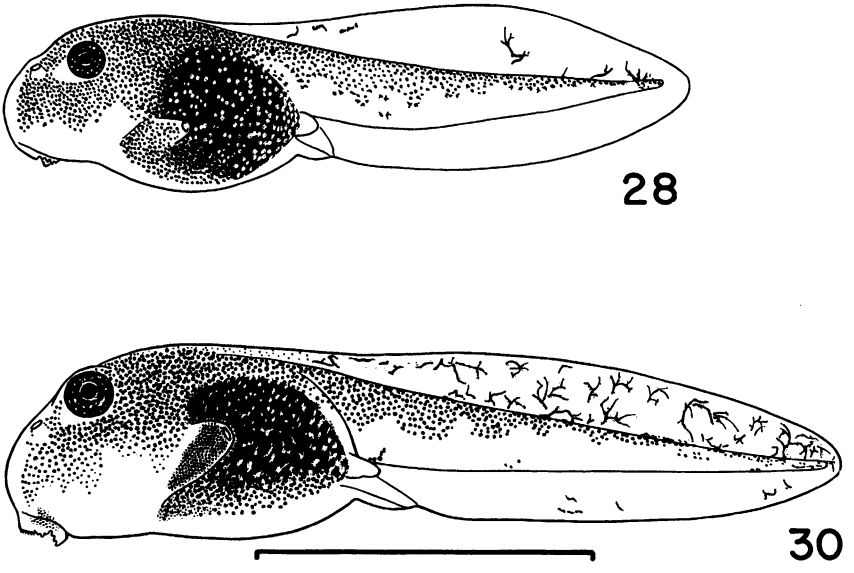

FIG. 6. Larvae of *Bufo kelloggi*. Numbers indicate stages of development; scale line represents a length of 5 mm.

present early in stage 25; but the basic color is pale yellow. The largest tadpoles (stage 28, fig. 9) have a heavy stippling of melanophores on the body (except for a free area where the tail musculature meets the abdomen), and these cells are especially dense on the abdomen. On the tail musculature, melanophores are more abundant on the dorsal half. Golden cells are present everywhere among the melanophores, but are not conspicuous, except on the lateroventral part of the abdomen where they exist in the superficial layers in the absence of melanin. The upper tail fin shows some dendritic accumulations of melanophores, but the lower fin is immaculate.

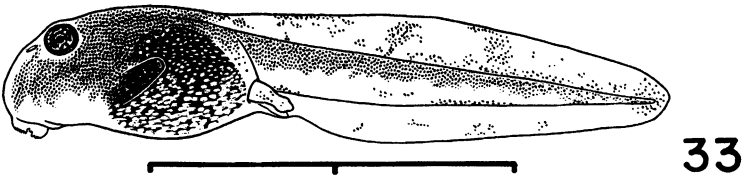

FIG. 7. Larva of *Bufo kelloggi* in stage 33. Scale line represents a length of 10 mm.

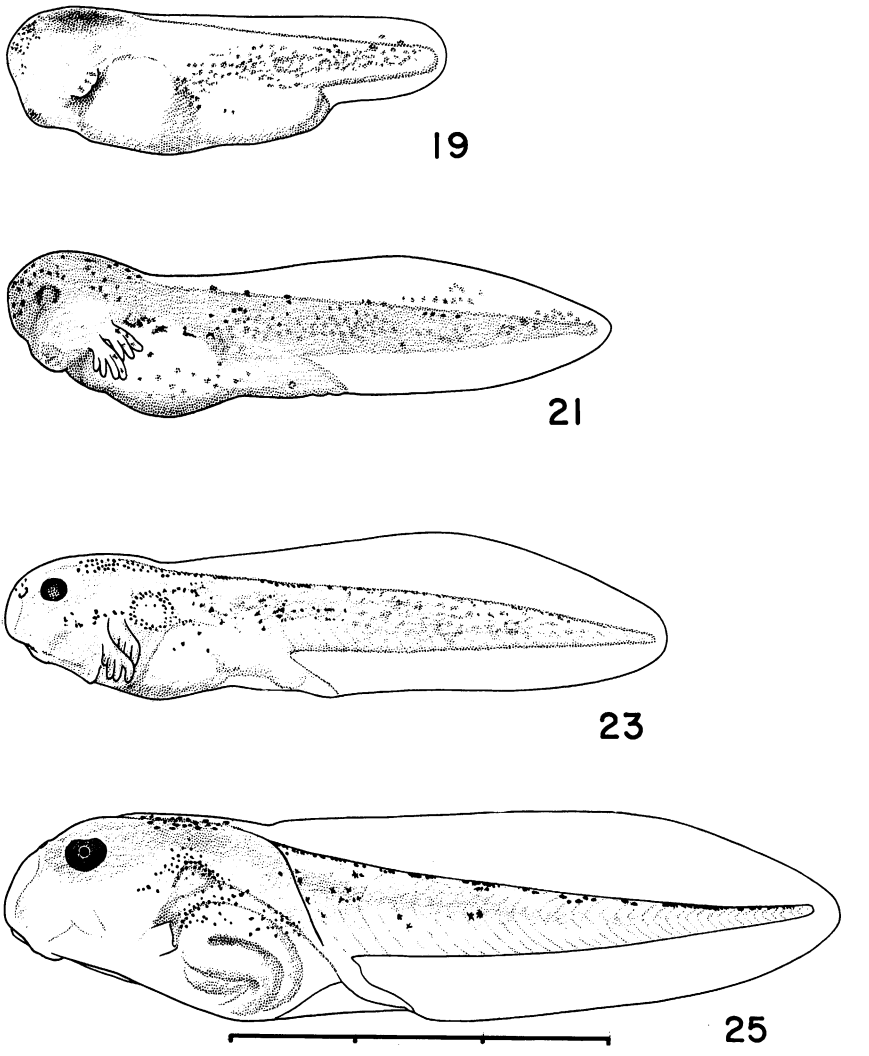

FIG. 8. Early stages of larval development in *Bufo retiformis*. Numbers indicate stages of development; scale line represents a length of 3 mm.

COMPARISONS WITHIN THE *debilis* GROUP

With the limited number of specimens available, the extent of individual and geographic variation cannot be assessed and the significance of apparent differences between species cannot be determined. No differences in the structure of the mouth parts or general morphology are

evident. There is some indication of differences in pigmentation among the three species. In *debilis* the tail musculature tends to be rather uniformly pigmented with melanophores (fig. 4), whereas in my larger specimens of *kelloggi* and *retiformis* the ventral half of the musculature is largely free of this pigment (figs. 7, 9). Limbaugh and Volpe (1957) found the pigmentation of the tail musculature to be of use in diagnosing the larva of *B. valliceps,* and it may prove useful in the *debilis* group. Possibly, if numbers of well-developed living tadpoles of the three species were available for direct comparison, other differences could be found, and the seeming difference in tail pigmentation confirmed, but for the present I

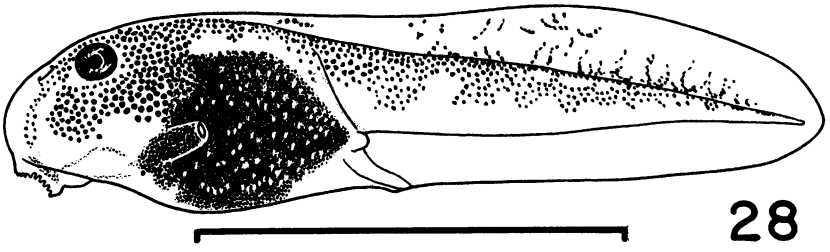

FIG. 9. Larva of *Bufo retiformis* in stage 28. Scale line represents a length of 5 mm.

regard the tadpoles of *debilis* as questionably distinguishable from those of *kelloggi* and *retiformis,* and the last two as indistinguishable from each other.

COMPARISONS WITH OTHER *Bufo*

As a group, the larvae of *Bufo debilis, B. kelloggi,* and *B. retiformis* are unique in the genus *Bufo,* so far as is known, in the possession of only two lower rows of labial teeth. The formula 2/3 describes virtually all other species of *Bufo* throughout the world whose larvae are known. This formula occurs even in a *Bufo* modified for stream dwelling (*B. preussi;* Mertens, 1938, p. 11 and pl. 2), in the related genus *Pedostibes* (Inger, 1966), and in *Ansonia,* with mouth parts greatly modified for life in fast-flowing water (Inger, 1960). The only exception in *Bufo* of which I am aware is *B. exsul* (*B. boreas exsul*) which is reported to have a formula of 1/3 (Wright and Wright, 1949, p. 179). Of course, the larvae of many species of *Bufo* have not yet been described, therefore the 2/2 formula of *debilis* and its relatives may prove not to be unique. There is no doubt, however, that the 2/2 formula is diagnostic of this group among North American *Bufo,* and when used in conjunction with geographic data (see

Bogert, 1962) will serve to identify *debilis* group larvae specifically everywhere except in that part of Sonora in which *B. kelloggi* and *B. retiformis* are sympatric.

The presence of the 2/2 formula in tadpoles of known parentage fully verifies Bragg's (1955) description. His conclusion that the differences between his tadpoles and those referred to *debilis* by Smith (1934), which had the common 2/3 formula, were "most likely due to differences in the environment" is incorrect in the light of present information. The larval mouth parts illustrated by Smith (1934, pl. 12, fig. 8) have the second anterior row of teeth scarcely interrupted, and long posterior rows, as is seen in *B. punctatus.* The habitat Smith described (1934, p. 444) is also appropriate for that species ("in small pools at the rocky bottom of a small, nearly vertical-sided tributary of Schwartz Canyon . . ."), and *B. punctatus* is recorded from the same locality.

Tadpoles of *Bufo* are said to have a median anal opening (Orton, 1952, p. 385), so it is noteworthy that the opening is clearly dextral in members of the *debilis* group.

Another way in which *debilis* group toads differ from other *Bufo* is in the stage at which hatching occurs. Typically, *Bufo* embryos hatch at an earlier stage of development than do those of other anurans. Hatching at late stage 16 or stage 17, before muscular movement is possible, is recorded for many species in different parts of the world. The embryos of *B. debilis, B. kelloggi,* and *B. retiformis* usually hatch in stage 19, although occasional individuals hatch in stage 18 and some remain within the capsules into stage 20, when gill circulation is established. The species of the *debilis* group evidently do not share with many other *Bufo* the habit of producing the eggs in long, continuous strings.

Ferguson and Lowe (1969) studied several features of the biology of the three species I examined, and they concluded that these three species together with *Bufo punctatus* constitute a natural species group. In that the larva of *B. punctatus* has typical *Bufo* mouth parts and hatches at a relatively early stage of development (stage 17; Zweifel, 1968, p. 56), this species is more like *Bufo* of other species groups than it is like *debilis, kelloggi,* or *retiformis.* The anal tube of larval *punctatus* appears to me to be more dextral than median, which lends similarity to *debilis* and its relatives. The habit of producing eggs singly or in short files, rather than in the long strings common to many species of *Bufo,* may ally *B. punctatus* with the three species I studied. It is possible, however, that the manner of deposition in *punctatus* is an adaptation to the stream habitat and does not necessarily reflect relationships (Zweifel, 1968, p. 55). Ferguson and Lowe regard *B. punctatus* to be the most primitive of the four species con-

sidered, and my information is in accord with this viewpoint.

Thus, with respect to some features of embryological development and larval morphology the species *Bufo debilis, B. kelloggi,* and *B. retiformis* differ notably from other *Bufo.* There can be no doubt (nor has there been in the past) that these species are closely related. Attempts to infer relationships to other species of *Bufo* should take these embryological and larval characteristics into consideration.

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