

A new distribution record of *Polypedates taeniatus* (Amphibia : Anura : Rhacophoridae) with notes on its Breeding Call and Post-Embryonic developmental stages

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Abstract

The Terai Reed frog, *Polypedates taeniatus* (Boulenger, 1906) was described on the basis of two specimens collected from Purneah, Bihar by W. Partridge. IUCN categorized the species as Least Concern due to its wide distribution all along the Himalayan foothills of Nepal and India at elevations below 500 meters a.s.l, supposedly large population and unlikeliness of declining fast enough to be considered in a threatened category. Although their breeding call was syllabilized (Ray, 1991) it was never recorded and analyzed. Nothing was known about the developmental stages of this Rhacophorid anuran which was known to inhabit tall grasslands. While conducting casual field-work in Behala Aerodrome area of south Kolkata, West Bengal, we observed the presence of the species calling from *Typha angustifolia* reeds. Three foam-nests were also collected and brought to the laboratory for study. The tadpoles were studied and photographed to determine their life stages (Gosner stages). It's each post-embryonic stage was measured later for their body lengths and tail lengths. Collection of three adult frogs (Registration No: ZSI A 11457 - A 11459) constitutes the first record of the species from the State of West Bengal, India and is more than 600 km from the foothills of the Himalayas. The breeding call of the species was recorded and analyzed. One call was analyzed; the call width was 4.103 seconds with 46 pulses. The pulse rate was 11.21 pulses per second. The fundamental frequency was found to be 1156 Hertz and the peak frequency was 2832 Hertz.

Keywords: Amphibia, Breeding Call, Distribution, Developmental Stages, Frog, Tadpoles

Introduction

The Rhacophorid frog, *Polypedates taeniatus* was described by Boulenger (1906) on the basis of two specimens collected from Purneah, Bihar by W. Partridge. One of the syntypes lodged in the British Museum of Natural History was considered as the lectotype. The other syntype (Reg No. 15715) lodged at the Zoological Survey of India, Kolkata was designated as the paralectotype by Ray (1991) who also collected the species from Dudhwa NP, Uttar Pradesh in 1986. Thereafter Anders et al (1998) reported it from Royal Chitwan NP and Koshi Tappu Wildlife Reserve, Nepal. Ahmed and Dutta (2000) found a specimen from Orang NP, Assam. Subsequently, Hegde et al (2009) collected six specimens from Bijnor, Meerut and Moradabad districts of western Uttar Pradesh

in 2008. Das et al., (2012) also found the species in Katarniaghat Wildlife Sanctuary, Uttar Pradesh. IUCN categorized the species as Least Concern due to its wide distribution all along the Himalayan foothills of Nepal and India at elevations below 500 meters a.s.l, supposedly large population and unlikeliness of declining fast enough to be considered in a threatened category. Although their breeding call was syllabilized (Ray, 1991) it was not recorded and analyzed. Nothing was known about the developmental stages of this anuran which was known to inhabit tall grasslands.

Study Area

While conducting casual field-work in Behala Aerodrome area of south Kolkata, West Bengal, we observed the

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species calling from *Typha angustifolia* reeds. This small aerodrome (22°50.544 N, 88°29.661 E, altitude: 20 m) with an area almost 3 sq km is used for flying small training aircrafts for trainee pilots and has a single runway of about 1 km with adjoining marshy areas covered with tall *Typha* grasslands, which gets inundated during the monsoon with rain-water standing up to a depth of up to 1 m.

Methodology

A survey was undertaken in the study area between 18:00-20:00 hr and three adult males were collected on 09.09.2012. These were compared with the paratype (ZSI 15715) to confirm the identity of the species. Their breeding calls were recorded with a SONY ICD P520 digital sound recorder and analyzed at Indian Institute of Science, Bengaluru. *Polypedates taeniatus* was found to hide at the base of the tall *Typha angustifolia* reeds and breed on them by constructing hanging foam-nests of size 6 X 6 cm. Three foam-nests were also collected and brought to the laboratory and kept in three different enamel trays with pond water up to a depth of 5 cm. Tadpoles from all the foam-nests hatched out two days later. The tadpoles were fed daily with boiled spinach as done in case of earlier studies on *Polypedates maculatus* in our laboratory and the water in the trays were changed regularly. 3-4 tadpoles were preserved every day at 14:00 hr and studied and photographed under Leica DFC 500 camera attached stereo-binocular microscope to determine their life stages (Gosner stages). Tadpoles of each post-embryonic stage was measured later for their body length and tail lengths.

Results

Survey: Collection of three adult frogs (Registration No: ZSIC A 11457 - A 11459) constitutes the first record of the species from the State of West Bengal, India. Snout-vent lengths of the collected frogs varied from 41.37-42.46 mm (mean 42.07 mm, n = 3).

Call Analysis: One breeding call of the species was recorded and analyzed. The call width was found to be 4.103 seconds with 46 pulses. The pulse rate was 11.21 pulses per second. The fundamental frequency was found to be 1156 Hertz and the peak frequency was 2832 Hertz.

Developmental Stages: From the foam-nests collected from the study area 218, 207 and 232 tadpoles (mean 219, n = 3) hatched out after two days and began swimming in the pond water put in the enamel trays. The post-embryonic developmental stages from Gosner Stage 20 (Gill Circulation Stage) to Gosner Stage 46 (Metamorphosed Froglet) were observed and photographed under camera attached stereo-binocular microscope. These developmental stages are described below along with their measurements (body and tail lengths and age in days).

Stage 20 (Gill Circulation Stage): (Body length 2.55mm, Tail length 3.44mm, age 1 day)

The tadpoles had distinct minute-sized gills and their own yolk sac from which they derived nourishment.

Stage 21 (Free-swimming larval Stage): (Body length 2.98 mm, Tail length 4.12 mm, age 2 days)

Tail straight, tail fin is dusky. Mouth opens as a deep triangular pit at the stomodeum. The sides of the body are heavily pigmented and two rows of pigments pass from above the head over the dorsal side of body to the dorsal side of the tail.

Stage 22 (Tail fin circulation Stage): (Body length 3.16 mm, Tail length 4.31 mm, age 4 days)

Tail fin becomes transparent. Circulation begins in the tail fin below the front part of the dorsal fin. Mouth becomes wide. Fringes develop on the upper and lower labium but no papillae.

Stage 23 (Opercular fold Stage): (Body length 4.02 mm, Tail length 7.37 mm, age 6 days)

Opercular fold covers base of gills. Lips and denticles begin to differentiate. Papillae and faint labial ridges develop on upper and lower labial fringes. Cloaca not yet opens.

Stage 24 (Opercular fold closed on right side): (Body length 4.39 mm, Tail length 7.56 mm, age 7 days)

Opercular fold closes on right side. Supra and infra rostront develop from labial jaws. A faint row of keratodont teeth start starts to develop.

Stage 25 (Opercular fold closed on left side): (Body length 4.73 mm, Tail length 7.75 mm, age 8 days)

Operculum closes and gills disappear totally. Spiracle formed. Cloaca opens and tadpole starts feeding. Upper supra-angular keratodont teeth fully formed. Tail is lightly pigmented.

Stage 26 (Hind limb bud Stage): (Body length 5.42 mm, Tail length 9.33 mm, age 9 days)

Hind limb bud appears inside a groove between the ventral wall and base of tail. Length of tail bud is less than half its diameter. Pigmentation spreads to base of dorsal fin. A faint row of lower supra-angular keratodont teeth starts to develop.

Stage 27 (Length of limb bud half its diameter): (Body length 5.82 mm, Tail length 11.47 mm, age 11 days)

Length of hind limb bud is equal to half its diameter. Pigmentation on the tail fin spreads considerably. Lower supra-angular keratodont teeth fully formed.

Stage 28 (Length of limb bud equal to its diameter): (Body length 6.04 mm, Tail length 11.88 mm, age 12 days)

Length of hind limb bud is equal to its diameter. End of limb bud becomes slightly conical. Pigmentation spreads all along the tail.

Stage 29 (Length of limb bud equal to one and a half times its diameter): (Body length 6.64 mm, Tail length 12.45 mm, age 14 days)

Length of hind limb bud becomes equal to one and a half times its diameter. End of limb bud becomes conical. First and second row of keratodont teeth is complete but third row is incomplete.

Stage 30 (Length of limb bud is equal to twice its diameter): (Body length 6.88 mm, Tail length 13.32 mm, age 15 days)

Length of hind limb bud is greater than twice its diameter. Last half of conical limb bud is slightly becomes bent. First row of keratodont teeth complete but second and third row incomplete.

Stage 31 (Foot Paddle Stage): (Body length 7.57 mm, Tail length 14.61 mm, age 17 days)

End of the limb bud becomes flattened laterally to form a foot paddle with no inter-digital indentations. Bend of knee bend becomes prominent.

Stage 32 (First Indentation Stage): (Body length 7.87 mm, Tail length 14.64 mm, age 18 days)

The margin of the foot paddle becomes slightly indented on the ventral side which marks the prominences of the future 4th and 5th toes.

Stage 33 (Second Indentation Stage): (Body length 8.15 mm, Tail length 15.24 mm, age 19 days)

Margin of the foot paddle becomes slightly indented on the ventral side start formation of the 4th toe, forming the 3rd, 4th and 5th toes.

Stage 34 (Third Indentation Stage): (Body length 8.33 mm, Tail length 15.44 mm, age 21 days)

Margin of the foot paddle becomes indented on the ventral side behind the 3rd toe, forming the 2nd, 3rd, 4th and 5th toes.

Stage 35 (Fourth Indentation Stage): (Body length 8.82 mm, Tail length 15.48 mm., age 22 days)

Margin of the foot paddle is indented behind the 2nd toe forming the 1st toe. All the five toes are now fully formed.

Stage 36 (Margin of 5th toe web directed towards tip of 2nd toe): (Body length 9.29 mm, Tail length 15.64 mm, age 23 days)

Margin of the 5th toe web is directed towards the tip of the 2nd toe.

Stage 37 (Margin of 5th toe web directed towards tip of 1st toe): (Body length 10.35 mm, Tail length 15.83 mm, age 25 days)

Margin of the 5th toe web is directed towards the tip of the 1st toe. Toes grow longer.

Stage 38 (Margin of 5th toe web directed towards prehallux): (Body length 11.29 mm, Tail length 16.24 mm, age 26 days)

Margin of the 5th toe web is directed towards the prehallux. Inner metatarsal tubercle start appearing.

Stage 39 (Appearance of subarticular tubercles on toes): (Body length 11.76 mm, Tail length 16.47 mm, age 28 days)

Subarticular tubercles appear on the ventral surface of the toes. The inner metatarsal tubercle is still small.

Stage 40 (Toe pads complete): (Body length 12.15 mm, Tail length 16.87 mm, age 30 days)

Distal end of toes thicken. Subarticular tubercles become prominent. The cloacal tail piece is still prolonged.

Stage 41 (Cloacal tail piece reduced): (Body length 11.93 mm, Tail length 9.22 mm, age 32 days)

The cloacal tail piece suddenly reduces considerably. The forelimbs become visible under the skin. Larval mouthparts begin to break down. Keratodonts start shedding but oral papillae on labial fringe remains intact.

Stage 42 (Both forelimbs emerge): (Body length 11.68 mm, Tail length 7.09 mm, age 34 days)

Both forelimbs emerge, the right forelimb first followed by the left. Absorption of labial fringe begins. Mouth starts widening. The cloacal tail piece reduces further.

Stage 43 (Angle of mouth between the eye and nostril): (Body length 11.56 mm, Tail length 4.96 mm, age 35 days)

The widening angle of the mouth reaches midway between nostril and front margin of eye. Jaws and tongue are formed. The dorsal and ventral fins in the tail reduce and the tail is reduced considerably.

Stage 44 (Angle of mouth reached the middle of eye): (Body length 11.42 mm., Tail length 3.26 mm, age 36 days)

The widening angle of the mouth reaches the middle of the eye. Dorsal and ventral fins disappear in the tail. Tail absorbed further.

Stage 45 (Angle of mouth reached posterior margin of the eye): (Body length 11.28 mm, Tail length 2.04 mm, age 37 days)

The widening angle of the mouth reaches the hind margin of the eye. The tail is now reduced to a small stub.

Stage 46 (Metamorphosed Froglet): (Body length 11.22 mm, Tail length 1.26 mm, age 37 days)

The triangular tail stub disappears completely. Tail resorbed, metamorphosis is complete. The metamorphosed froglets climb up the wall of the rearing tray.

Thus, it took 37 days from egg stage to metamorphosed froglet stage, the tadpoles had tail for 30 days and on the 32nd day the tail started reducing. The tadpoles metamorphosed and climbed out of water on the 35th day. The tadpoles with a snout-vent length of about 11 mm climbed out of the trays after 37 days of development on 17.10.2012.

Discussion

The Terai Reed Frog (*Polypedates taeniatus*) was known from the terai grassland areas at the base of the Himalayas from Uttar Pradesh through Bihar to Assam. However, the species was never reported more than 100 km from the Himalayan foothills. The discovery of this species from an aerodrome inside Kolkata city is not only the first record of the species from the State of West Bengal but also the first report from about 600 km away from the Himalayan terai regions. However, wherever the species was known it was always seen in tall grasslands which gets inundated during the monsoon allowing the frogs to breed and construct their hanging foam-nests. The presence of remnant of tall Typha grasslands in this aerodrome area may have allowed the species to survive in this site for the last few centuries. How far this species have withstood habitat changes in an urban area is not known but apparently the presence of the remnant of tall grasslands similar to terai areas seems to have allowed the species to survive there.

Although the breeding call of the species was syllabilized by Ray, 1991 it was not recorded and analyzed. The recording of a single call was done for the first time in this study by digital sound recorder and subsequently analyzed. However, as analysis of a single call may be pre-mature interpretation of the call of the species, more acoustic studies on the calls of the species should be done.

Nothing was known about the developmental stages and life history of this species although its foam-nesting

behavior was noted earlier by Ray (1991). Although we could not successfully study the embryonic stages inside the collected foam-nests, yet we could manage to develop the hatched out tadpoles to froglet stage and study each and every post-embryonic stage from Gosner Stage 20 to Gosner Stage 46 and photograph most of the stages under microscope. Gosner staging of tadpoles is the standard procedure used throughout the world nowadays for study on developmental stages of anuran embryos and larvae and the proper identification of the developing stages as devised by K.L. Gosner (1960).

The observations showed that the head and body length of the tadpoles increased steadily from Gosner Stage 20 (2.55 mm) to Gosner Stage 40 (12.15 mm) in the first 30 days and then decreased slightly to 11.22 mm at complete metamorphosis (Gosner Stage 46) over the next 7 days. Similarly the tail length also increased steadily from Gosner Stage 20 (3.44 mm) to Gosner Stage 40 (16.87 mm) in the first 30 days and then suddenly decreased rapidly to 9.22 mm (Gosner Stage 41) in the next two days when the cloacal tail piece reduced sharply and then

further reduced to 1.26 mm at complete metamorphosis (Gosner Stage 46).

Thus, this species under laboratory conditions took 37 days to develop from egg stage to metamorphosed froglet stage. The tadpoles had tail for 30 days and on the 32nd day tail started reducing. The tadpoles metamorphosed and climbed out of water on the 35th day. The froglets with a snout-vent length of about 11 mm climbed out of the trays and completed their life cycle.

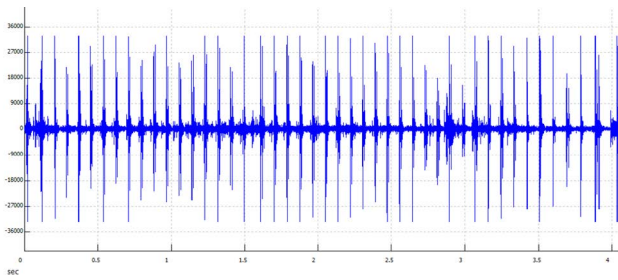
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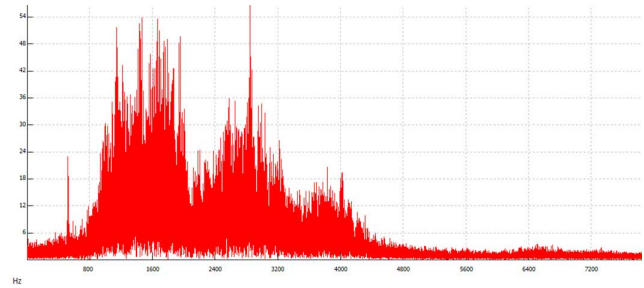
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Plate 1



1. Sonogram of call of *Polypedates taeniatus*.



2. Oscillogram of call of *Polypedates taeniatus*.



3. Adult of *Polypedates taeniatus*.



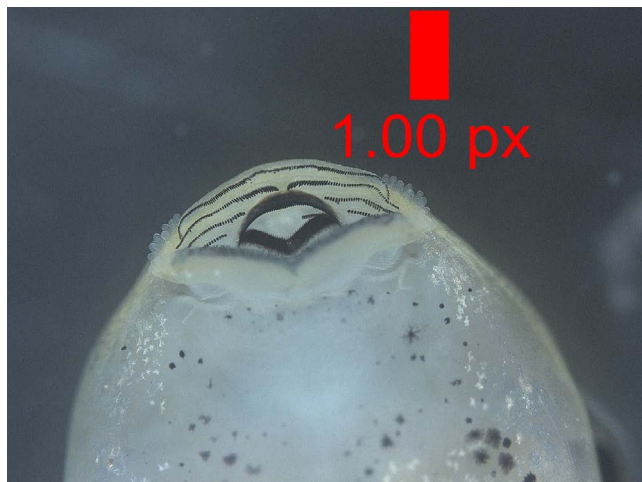
4. Adult on grass.



5. Foam-nest among grass.



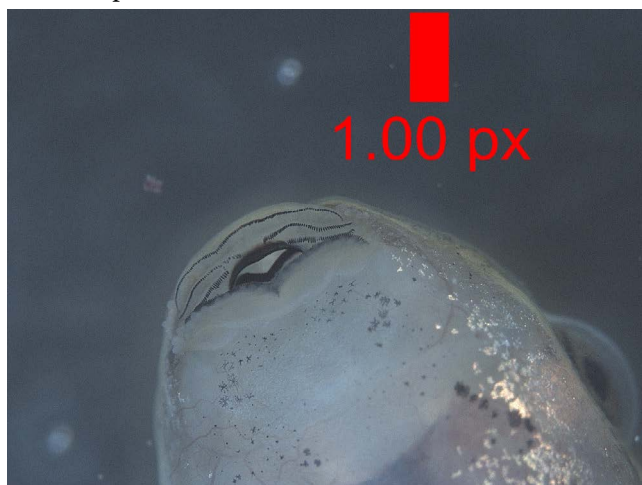
6. Stage 23 (free swimming larval stage) (40.4 X).



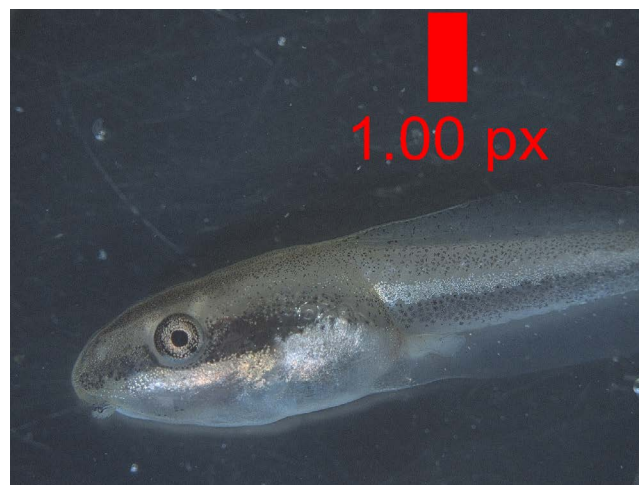
7. Stage 29 (1st & 2nd keratodont row complete 3rd incomplete) (28.8 X).



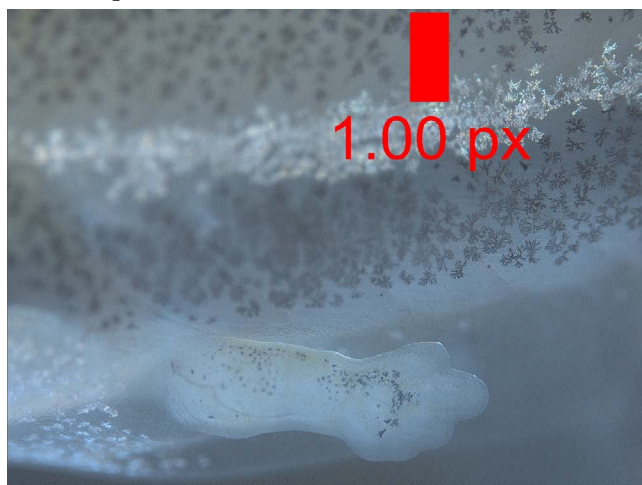
8. Stage 29 (limb bud one and half its diameter) (11.1 X).



9. Stage 30 (1st keratodont row complete, 2nd & 3rd incomplete) (29.0 X).



10. Stage 31 (limb bud twice its diameter) (9.7 X).

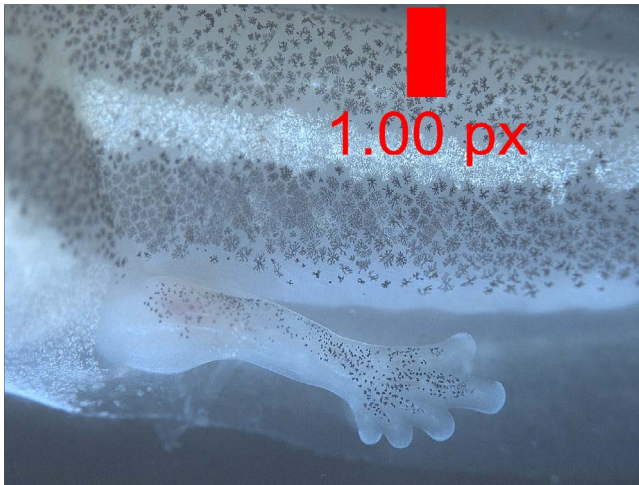


11. Stage 33 (Foot paddle Stage) (7.4 X).

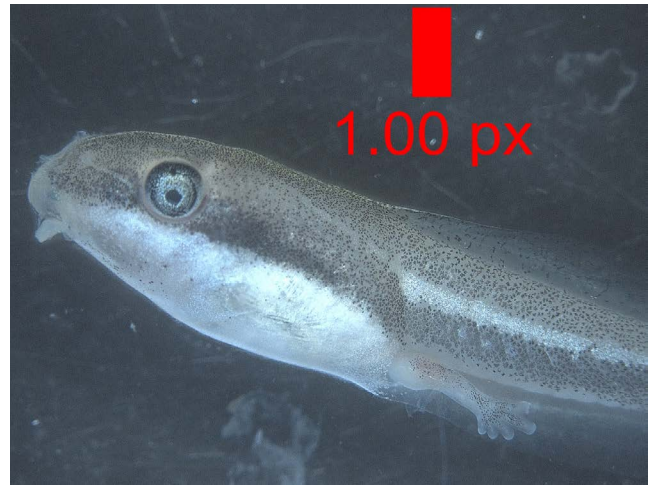


12. Stage 34 (tadpole at foot paddle stage) (10.3 X).

Plate 2



13. Stage 35 (Fourth indentation stage on toes) (24.3 X).



14. Stage 35 (tadpole with fourth indentation on toes) (9.2 X).



15. Stage 38 (all toes formed) (10.3 X).



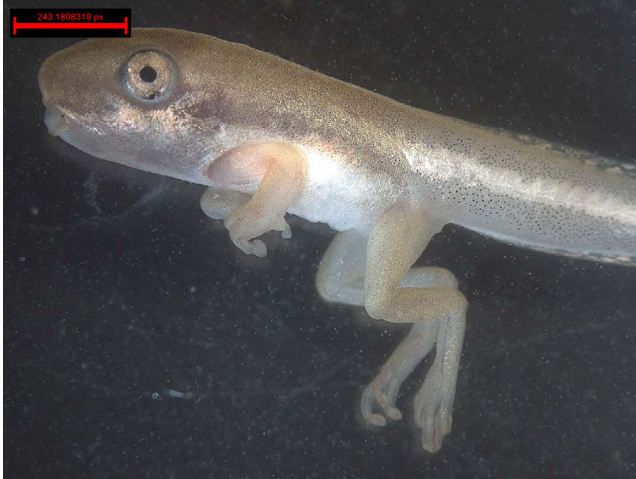
16. Stage 38 (tadpole with all toes formed) (4.91 X).



17. Stage 40 (Toe pads complete) (7.6 X).



18. Stage 41 (Keratodonts shed oral papillae on labial fringe) (28.9 X).



19. Stage 42 (Both forelimbs emerge) (5.28 X).



20. Stage 42 (Mouth formed) (13.9 X).



21. Stage 43 (Angle of mouth between nostril & eye).



22. Stage 45 (Angle of mouth to posterior margin of eye).



23. Stage 45 (Tail is a triangular stub).