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Description of tadpole stages of the Malabar Tree Toad *Pedostibes tuberculosus* Günther, 1875 (Anura: Bufonidae)

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Pedostibes tuberculosus is a member of the toad family Bufonidae exhibiting a phytotelmatic mode of life (leading an arboreal mode for the most part of its life). This toad was described by Günther in 1875 from southern India with the type locality mentioned as 'Malabar' (now part of coastal Kerala). The species is commonly known as the Malabar Tree Toad and is endemic to the Western Ghats. This toad was not known from any of its natural environs for more than a span of 100 years since its description, till it's rediscovery by S.K. Bhattacharya from the Silent Valley National Park in 1980 (Pillai 1986). There was no information on its natural history and life cycle and the only information available was its range of distribution and advertisement call details (Gururaja & Ramachandra 2006; Dinesh & Radhakrishnan 2008). However, Boxclaer et al. (2009) mentioned a specialized development and an egg clutch size of 250 eggs laid at the edge of streams, wherein the tadpoles develop by bottom feeding.

The species is placed under the 'Endangered' category in the IUCN Red List (Biju et al. 2004).

On 2 July 2009, during our search for the reed associated frogs at Kakkayam Reserve Forest (11.54875^{0}N & 75.923788^{0}E), Kozhikode District, Kerala, we encountered nine motile tiny tadpoles (assignable to Stage 21 of Gosner 1960), inside the culm of a standing, dried, *Ochlandra* plant with some water trapped in the nodal region. All the nine tadpoles were collected and moved to a laboratory for further observation.

Materials and Methods: The tadpoles were kept in a petri dish containing rain water and maintained at room temperature between 22^{0}C and 24^{0}C. The tadpoles were fed with boiled chicken egg yolk. The tadpole stage numbers were assigned as per Gosner (1960). Live tadpole images were captured using a Leica M 205A stereo microscope, with a Leica DFC 500 camera. The tadpoles were observed periodically (but not on a daily basis) for 26 days from the date of collection. From the1st day of collection from the field, significant changes seen on the 7th, 14th, 18th, 20th, 22nd, 24th and the 26th day were studied and recorded.

Observations: On the day of the collection from the field, the tadpoles were at Stage 21 (Image 1) and were creamy white in colour, fragile and motile. The body of the tadpoles was transparent, dorso-laterally compressed, with a narrow head and distinct oral disc, and a sucker median in position (a in Image 1). The eyes

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were dark circular (b in Image 1) and abdomen with a mass of yolk in a sac like structure bulging towards the tail musculature (c in Image 1). Tail muscle axis, tail musculature (d in Image 1) and vent tube, were clearly visible and the tail fin was transparent (e in Image 1).

At Stage 27 (on the 7th day from the date of collection of the tadpoles from field) (Image 2) the tadpoles were active and motile, their body dorsoventrally compressed (c in Image 2). The Dorsum of the body became brownish and the eyes were distinct at the anterior part of the body (b in Image 2). The region behind the eyes was wider and the sucker was in a ventral position (a in Image 2). The ventral side of the body appeared transparent (a in Image 3) and the intestine was spirally coiled (b in Image 3). Distinct limb buds (d in Image 2 & c in Image 3) could be seen at the junction of the body and a tail fin was present.

At Stage 41 (on the 14th day from the date of collection of the tadpoles from the field) (Image 4), the body of the tadpoles appeared dorsolaterally compressed, brownish and with enlarged eyes (b in Image 4). The width and length of the tail were reduced (e in Image 4), the mouth region appeared arched (rounded) (a in Image 4) and the skin over the forelimb transparent (c in Image 4), the hind limbs had emerged out of the body (d in Image 4).

At Stage 42 (on the 18th day from the date of collection of the tadpoles from the field) (Image 5), the body colour turned brownish; the eyes grew large, round and black in colour (b in Image 5); the oral disc got dorso-laterally placed and the mouth region gained a circular shape (a in Image 5). The forelimbs were within the skin sac (c in Image 5), and the hind limbs nearly completely grown (d in Image 5). The tail appeared rounded and tapered (e in Image 5).

At Stage 43 (on the 20th day from the date of collection of the tadpoles from the field) (Image 6), the body of the tadpoles appeared brownish dorsally with two lateral silvery yellow stripes. The eyes were rounded and clear (b in Image 6) with a formation of snout and canthus rostralis (a in Image 6). The forelimbs and hind limbs were completely formed (c & d in Image 6) with a clear demarcation of fingers and toes. The tail became more tapered and appeared to be sinking (e in Image 6).

At Stage 44 (on the 22nd day from the date of collection of the tadpoles from the field) (Image 7), the body colour of the tadpole turned brownish with the yellow silvery lateral band extended on to the head region; the snout and canthus rostralis became distinct (a in Image 7) and the region of the orbit flat; the hind and fore limbs were completely developed (b & c in Image 7), with the tail further diminished. The tadpoles started moving to the

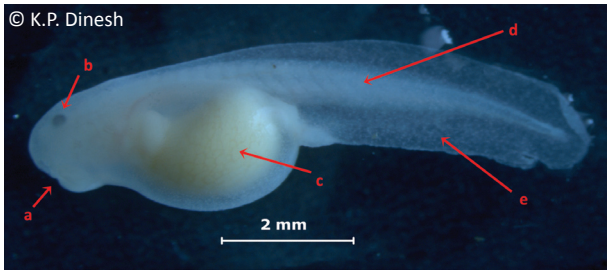

Image 1. Tadpole Stage 21 of *P. tuberculosus.*

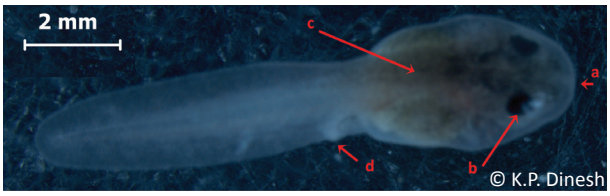

Image 2. Tadpole Stage 27 of *P. tuberculosus* - dorsal view.

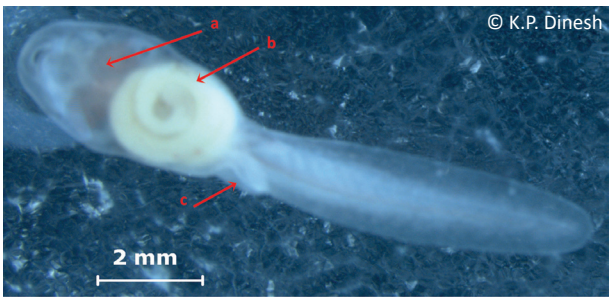

Image 3. Tadpole Stage 27 of *P. tuberculosus* - ventral view

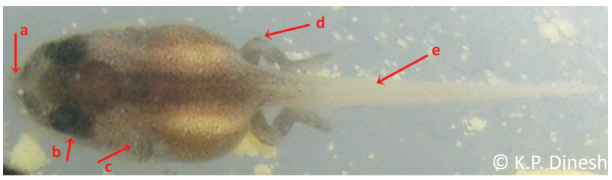

Image 4. Tadpole Stage 41 of *P. tuberculosus* - dorsal view.

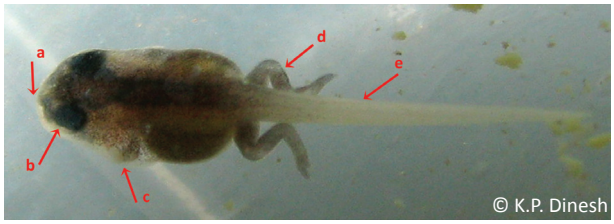

Image 5. Tadpole Stage 42 of *P. tuberculosus* - dorsal view.

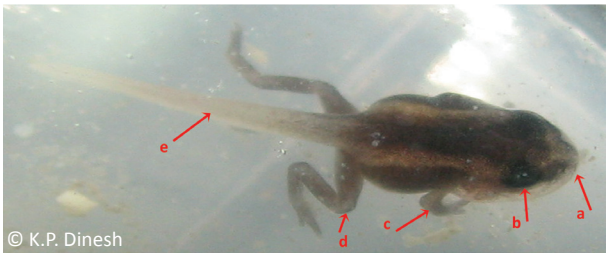

Image 6. Tadpole Stage 43 of *P. tuberculosus* - dorsal view.

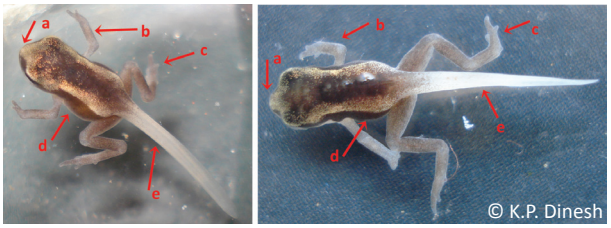

Image 7. Tadpole Stage 44 of *P. tuberculosus* - dorsal view.

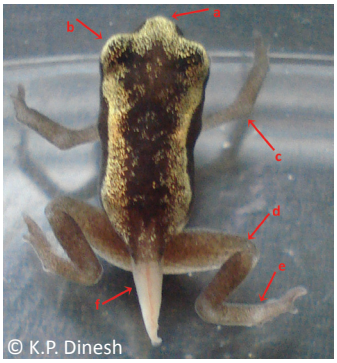

Image 8. Tadpole Stage 45 of *P. tuberculosus* - dorsal view.

periphery of the water without much use of the tail.

At Stage 45 (on the 24th day from the date of collection of the tadpoles from the field) (Image 8), the edges of the dorsum of the tadpoles appeared silvery-yellow in colour (a & b in Image 8), the lateral sides appearing blackish-brown; the snout was obtuse in shape and the orbit region elevated. The fore and hind limbs were operational (c & d in Image 8), and the tail size much diminished (f in Image 8) resembling a young one of *P. tuberculosus*, climbing out of the water.

At Stage 46 (on the 26th day from the date of collection of the tadpoles from the field), the tail of the tadpoles was completely absorbed into the body and the imagos resembled the adults of the species and were seen hopping around.

Discussion: The IUCN Red List (http://www.iucnredlist.org/details/16470/0) categorises *P. tuberculosus* as 'Endangered', in spite of the fact that the species is known to have a confirmed distribution range from Cotegoa (14.978^{0}N & 74.148^{0}E) to Kalakkad (8.5305^{0}N & 77.456^{0}E) (Dinesh & Radhakrishnan in 2008). During many of our field explorations in the Western Ghats, we have seen this species locally abundant (seasonal) in certain pockets of evergreen forests. We suggest that a re-assessment study based on authentic field work of this tree toad is required to gather more information on the natural history of this species.

During our visits to the collection site from 2009 to 2012, we came across young ones of *P. tuberculosus* at various stages inside the *Ochlandrae* culm and the adults making advertisement calls on various occasions. However, we could not get amplecting pairs so as to observe the entire egg laying procedure. Therefore, in our present observation we report only the transformation of tadpoles from Stage 21 to independent young ones (imago), taking 26 days to complete the process.

The present report forms the first record of some of the tadpole stages for *P. tuberculosus*. In our general observations, we note that this species breeds next to shallow streams of forests where the males assemble next to streams and call in chorus during the monsoon showers of June. Our current observation of the collection of eggs from the dried reed confirms its phytotelmatic mode of life and breeding. Our observations are contrary to Boxclaer et al. (2009) report of about 250 eggs in a clutch at the edge of streams and bottom feeding tadpoles pertaining to this tree toad.

Our observations warrant further detailed investigations to understand amplexus and how an amplected pair lays eggs inside the culm.

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