

**Figure 4.** Removal of lead and cadmium from (a) *N. jatamansi* extract and (b) *V. vinifera* extract using a packed bed reactor containing beads of *C. cladosporioides* # 2.

there were no alterations in pH, UV-visible spectra and TDS and also no changes in number, size and position of spots appearing in thin layer chromatograms, it could be said that medicinal properties of the extracts remained unchanged after biosorption.

The results of column experiments clearly indicate that the toxic metals from herbal extracts could be brought down to below permissible limits. Large-scale trials are however required to be performed to test the commercial applicability of the process.

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**ACKNOWLEDGEMENTS.** We thank the Director, Agharkar Research Institute for help and facilities provided. A.V.P. thanks Hi-Tech Bio Laboratories, Pune for providing research fellowship.

Received 11 October 2000; revised accepted 10 January 2001

## First record of the Ranid frog *Paa annandalii* (Boulenger 1920) from north-eastern region (Arunachal Pradesh) of India with a note on its larval stages

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**Arunachal Pradesh has remained underexplored for its amphibian faunal wealth. *Paa annandalii* adults and tadpoles were collected from a snowfed stream in Tawang, Tawang district of Arunachal Pradesh. In India this frog was first recorded by Annandale<sup>1</sup> from Darjeeling district in West Bengal. This species was included in the genus *Rana*. Dubois<sup>5</sup> later included this species in the genus *Paa*. Detailed taxonomic description of the adults and tadpoles, and food habits of the tadpoles are presented in this paper.**

ARUNACHAL Pradesh is a part of the Eastern Himalayan region, a hotspot of biodiversity. Report of amphibian fauna of this region is very fragmentary<sup>1–4</sup>. The present collection of *Paa annandalii* (Boulenger, 1920) is from the north-eastern region of India. This frog was originally known as *Rana annandalii* and was collected by Annandale<sup>1</sup> from Sureil, Darjeeling district (1800 m amsl), West Bengal. Later this species was included in the subgenus *Paa*<sup>5</sup> of the genus *Rana*. Subsequently the species has been included in the genus *Paa* and in *Paa liebegii*

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group<sup>6,7</sup>. The present collection of adults (three) is from the bank of a stream and the tadpoles are from the same stream in Tawang, Tawang district of Arunachal Pradesh, India.

Tawang is situated at an altitude of 4031 m amsl and is located at 27°35'25"N latitude and 91°45'E longitude. The still water body formed by melting snow was inhabited by tadpoles of *P. annandalii*. Description of the adult and tadpoles was compared with Boulenger<sup>8</sup>.

The colour of the dorsal surface is olive green in living condition. On preservation the colour changes to black (Figure 1). A dark crossbar exists between the eyes. Limbs have numerous dark crossbars. Throat and breast are spotted with brown spots.

The dorsal surface is smooth with scattered minute granules and small warts (Figure 2). A strong glandular fold extends from eye to the shoulder. Glandular longitudinal ridges are prominent on the dorsal side. Ventral surface is smooth.

Males are without external vocal sacs. Forelimbs are very strong. A patch of large black spines is present on each side of the breast and on the dorsal surface of the first and second finger and on the metacarpal tubercles. Vomerine teeth are found in short oblique series close together. Head is broader than long and much depressed. Snout is rounded, scarcely projecting beyond the mouth and as long as the eye (antero-posterior axis). Canthus rostralis is obtuse; loreal region is concave. Nostrils are equidistant from the eye and from the end of the snout. Internarial distance is greater than the interorbital width. Tympanum is hidden.

Fingertip is rounded, the first and second fingers are equal, the third finger is a little longer than the snout. Sub articular tubercles are small and moderately prominent. Hind limbs are long. Tibio tarsal articulation reaches nostril. Heels strongly overlap when the limbs are folded at right angles to the body. Tibia is three and a half times as long as broad. Tibia length is a little shorter than the forelimb. The toes have tips swollen into small discs

which are fully webbed, and two phalanges of the fourth toe are narrowly bordered by the web. Outer metatarsals are separated nearly to the base. Sub-articular tubercles are prominent. Tarsal fold is absent. Inner metatarsal tubercle is narrow and prominent. Outer metatarsal tubercle is absent. Morphometric measurements of the three frogs are presented in Table 1.

For taxonomic description of tadpoles, Altig<sup>9</sup> was consulted. Staging of tadpoles was done with the help of Gosner<sup>10</sup>. Collected tadpoles belonged to late stages. Early stages were not found. Morphometric measurements of all the stages found have been incorporated in Table 2. Detailed description of stage 38 tadpole is as follows.

The body of the tadpole is black. The tail has brown irregular spots (Figure 3), is obtusely pointed and about twice as long as the body. The head is round in shape and the snout is blunt. Eyes are dorsally placed and round in

**Table 1.** Morphometric measurements of body parts of adult *Paa annandalii*

	Male (mm)	Male (mm)	Female (mm)
Snout vent length	50	51	50
Length of head	15	16	15
Width of head	17	18	17
Length of snout	7	8	7
Length of eye (antero-posterior axis)	7	8	7
Interorbital width	4	4	4
Internarial width	5	5	5
Length of forelimb	31	32	31
Relative length of fingers	1 = 2 < 4 < 3		
Length of hind limb	90	93	91
Length of tibia	29	30	30
Width of tibia	9	9	9
Length of foot	30	31	30
Relative length of toes	1 < 2 < 3 < 5 < 4		
TTA reaches	Nostril		



**Figure 1.** Secondary sexual characters of the male frog during breeding season.



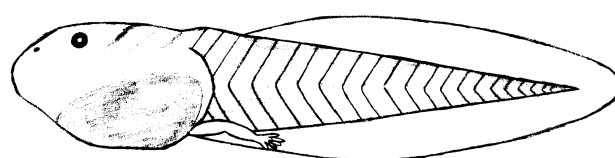
**Figure 2.** Dorsal surface of *Paa annandalii*. a, Dorsal surface with minute granules and small warts; b, Glandular fold extending from eye to shoulder; c, Prominent longitudinal ridges on the dorsal surface; d, Round snout scarcely projecting beyond the mouth; e, Concave loreal region.

**Table 2.** Morphometric features of *Paa annandalii* tadpoles

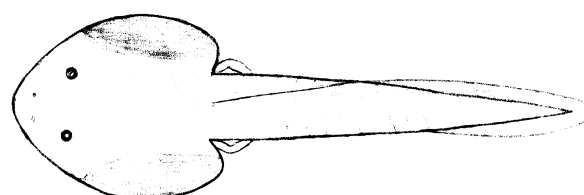
Description (mm)	1	2	3	4	5	6	7	8	9	10	11	12	13
Total length	25.2	44.8	44.8	46.6	46.2	53.8	51.8	53.9	61.3	65.8	61.1	64.6	84.1
Body length	10.2	15.9	15.1	16.0	15.1	18.9	18.6	18.0	15.1	23.1	21.7	22.6	25.0
Body width	6.8	9.5	9.2	10.2	10.1	13.0	12.1	12.1	17.5	17.5	17.0	17.9	17.7
Body height	5.2	7.9	7.8	8.1	8.0	10.5	10.1	10.0	12.9	13.4	11.5	12.9	13.2
Head width	5.7	8.3	7.5	9.1	8.8	10.2	10.9	10.6	13.8	13.9	12.5	13.1	14.9
Head height	4.1	5.6	5.4	5.8	6.2	7.8	6.6	7.3	9.5	8.9	7.8	8.8	8.1
Interocular space	3.5	5.0	5.3	5.0	5.0	5.8	5.9	5.0	6.4	6.4	6.2	6.4	7.1
Internarial space	2.2	3.2	3.4	3.3	3.2	4.2	4.1	4.7	4.5	4.1	4.2	4.2	4.1
Diameter of eye	0.9	1.9	1.9	1.9	1.9	2.0	2.0	2.0	2.7	2.5	2.7	2.5	2.7
Narial eye distance	1.7	2.0	2.4	2.0	2.0	2.4	2.5	2.8	3.2	2.8	3.0	2.9	3.2
Mouth width	3.5	4.5	4.9	4.5	5.1	6.0	6.0	6.2	7.8	7.6	7.3	7.3	8.1
Snout spiracle distance	7.1	10.1	10.0	10.4	10.2	13.3	12.7	13.4	16.3	15.6	13.7	15.8	16.5
Snout nare distance	2.8	2.7	3.4	2.6	2.7	3.7	3.6	3.7	4.3	4.1	3.8	4.2	4.2
Snout eye distance	3.9	5.5	5.3	5.5	5.5	6.6	6.5	6.8	7.2	7.0	6.3	7.1	7.4
Spiracle eye distance	3.4	5.1	4.1	5.1	5.1	6.2	5.3	6.4	7.1	7.5	6.5	7.5	8.1
Spiracle nare distance	5.2	7.6	7.1	7.9	7.6	10.6	9.6	11.5	12.3	12.5	11.1	12.3	14.0
Tail length	15.0	28.9	29.7	30.6	31.1	34.9	37.5	38.0	46.2	42.7	39.4	42.0	59.1
Tail height	6.0	9.9	8.5	9.8	9.5	12.9	12.6	12.8	16.0	16.2	15.1	16.8	16.8
Diameter of tail muscle	3.5	6.2	5.5	6.0	5.9	8.0	6.9	7.8	10.6	9.5	10.2	8.9	11.6
Length of hind limb	0.4	1.8	1.8	2.0	2.4	2.6	6.1	8.2	11.0	13.7	17.8	24.5	32.1
Gosner stage	26	29	30	31	32	33	35	36	37	38	39	40	41



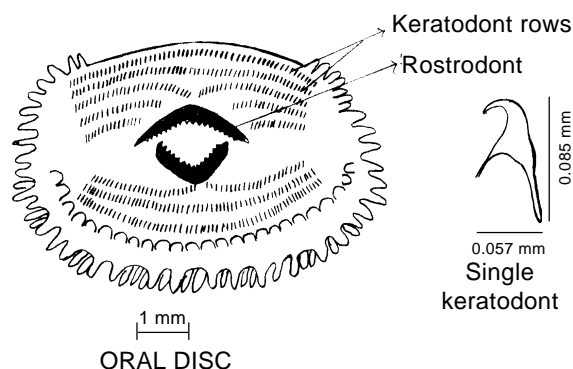
**Figure 3.** Various stages of the tadpoles of *Paa annandalii*.



LATERAL VIEW



DORSAL VIEW



**Figure 4.** Structural details of *Paa annandalii* tadpole.

shape. The interocular distance is comparatively larger. The nares are oval in shape and with a rim. Internarial distance is large. Nares are placed nearer to the eyes than to the snout. Spiracle is sinistral. Muscle blocks on the dorsal side are very distinct. The vent is median. Intestinal coils are not very distinct from outside. There are four and a half coils.

The tail and the tail fins are well developed. The height and thickness of the tail musculature is maximum near the vent. The muscle blocks are also distinct. The dorsal and ventral fins are of equal size and the tail tip is blunt. Fins are irregularly pigmented.

Oral disc is well developed. The rostral gap is large and distinct, while the mental gap is not seen. The supra and infra rostrodonts are distinctly serrated. Keratodont rows are distinct. In the upper labium, the first and second rows are uninterrupted, while the third, fourth and fifth rows are interrupted by the median gap. In the lower labium

the first row is interrupted by a median gap, while the other two rows are uninterrupted. The dental formula is (2 : 3 + 3/1 + 1 : 2). Oral papillae are distinct and well

developed. A single keratodont measures 0.085 mm in length and 0.057 mm in breadth. Measurements of the tadpoles are presented in Table 2 and the structural details of the tadpoles in Figure 4.

Tadpoles are mainly surface feeders. Keratinized mouth parts help the tadpoles in utilizing the periphyton. Gut contents of tadpoles belonging to early stages were analysed to know the food items. Phytoplanktons belonging to the following genera were recorded from the gut contents. *Oscillatoria*, *Phormidium*, *Anabaena*, *Gloeo-trichia* (Myxophyceae), *Achnanthes*, *Synendra*, *Tabellaria*, *Navicula*, *Melosira*, *Surirella*, *Nititzschia*, *Cymbella* (Bacillariophyceae), *Ulothrix*, *Chaetophora*, *Spirogyra*, *Zygnema*, *Desmidium*, *Volvox*, *Scenedesmus* (Chlorophyceae). Zooplanktons (*Philodina*, *Monostyla*, *Cyclops*) were encountered only in two cases. In one case a nematode was found. Occurrence of nematode in gut contents was earlier reported by Sahu<sup>11</sup> in the gut of *Rana alticola* tadpoles. Food items were identified with the help of Edmondson<sup>12</sup>.

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ACKNOWLEDGEMENTS. We thank Dr S. K. Dutta, Department of Zoology, Utkal University, Bhubaneswar for confirmation of identification of the frog. Financial support by the G.B. Pant Institute of Himalayan Environment and Development, Kosi-Katarmal, Almora, is gratefully acknowledged.

Received 13 October 2000; revised accepted 29 December 2000

## A deep-sea bacterium with unique nitrifying property

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**Sediment cores collected from the Central Indian Basin (10°1′–10°2′S, 75°59′–76°2′E) harboured a high population of nitrifying bacteria which ranged from 10<sup>5</sup> to 10<sup>7</sup> cfu per gram dry weight. This high density could contribute to the high nitrate concentration observed in these sediments. We report for the first time the unusual property of having both the phases of nitrification, i.e. NH<sub>4</sub><sup>+</sup> → NO<sub>2</sub><sup>-</sup> (Phase I) and NO<sub>2</sub><sup>-</sup> → NO<sub>3</sub><sup>-</sup> (Phase II) under normal and high pressure conditions in an autotrophic nitrifying bacterium isolated from the deep sediments cores at 5000 m depth (15–20 cm below the sea floor).**

THE Central Indian Basin (CIB) is characterized by unusually high concentration of nitrate in the pore water of sediments, which are highly siliceous. The nitrate concentration generally ranged from 31.25 to 50 µg at NI<sup>-1</sup> and occasionally reached 76 µg at NI<sup>-1</sup>, unlike the Arabian Sea where it ranged from 4.99 to 35.82 µg at NI<sup>-1</sup>. Moreover, nitrate levels in the CIB sediments showed no decline with increasing depth<sup>1,2</sup>. Nagendernath and

Mudholkar<sup>2</sup> hypothesized that this high concentration and constant value of nitrate in the sediments may be due to the intense nitrification process. This process has long been known to be restricted to aerobic or micro-aerophilic zone of water<sup>3</sup> and shallow sediments<sup>4</sup> and not to deep-sea sediment cores. The nitrification process is often deduced from the chemical profiles in water or from activity measurements<sup>5</sup>. Though Carlucci and Strickland<sup>6</sup> reported more than three decades ago that nitrifiers are widely distributed in the marine environment, the number of nitrifiers mediating this process has rarely been determined. Nitrification is generally carried out by known nitrifiers which either oxidize ammonia to nitrite (Phase I) or nitrite to nitrate (Phase II)<sup>7</sup>. The present work reports an observation of a deep-sea nitrifying bacterium capable of performing both the phases of nitrification.

Serially diluted samples from the sediment cores in the CIB (10°1′–10°2′S, 75°59′–76°2′E) were plated onto a mineral medium which is essentially a modified Winogradsky medium<sup>8</sup> with pure agar (Himedia, Mumbai) as gelling agent. The medium was substituted with ammonium sulphate at 2 mM (final concentration) or sodium nitrite at 0.5 mM (final concentration) as energy source and incubated for a period of 45 days at <10°C. The colonies were enumerated and expressed as colony forming units (cfu) per gram dry weight sediment. Representatives from morphologically different colonies were isolated and checked for the purity and nitrifying ability (both phases) in liquid Winogradsky's mineral medium. Among the 101 isolates one (CIB12) was found capable of performing both the phases of nitrification and henceforth selected for further studies.

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