

Notes on the genus *Amerianna* Strand, 1928 (Gastropoda, Planorbidae) in India

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Abstract

A previous confirmed report of the planorbid genus *Amerianna* from India was limited to the description of a new species, *Bulinus indicus*, from Pune, Maharashtra. Later, this species was recognised as *Amerianna carinata*. This study provides the first molecular data of *Amerianna* obtained from India, as well as the first confirmed report of its presence in southern India. Based on a BLAST query of a cytochrome oxidase subunit I sequence (*cox1*), the Indian material (Kochi, Kerala) showed 99.2% similarity with an *Amerianna* sp. from New Guinea, thus suggesting conspecificity. Owing to the tentative taxonomic status of the genus, extensive revision is required in order for the species status of the study material to be resolved.

Keywords: *Bulinus indicus*, Edappally Canal, Freshwater, Kerala, Kochi, *Planorbella scalaris*

Introduction

Members of the Planorbid genus *Amerianna* Strand, 1928 are known from northern regions of the Australian mainland, New Guinea, Moluccas and Philippines, and have been introduced to Java, Thailand and Nigeria (Ponder *et al.*, 2020). Previous reports of this genus in India are limited to a description of the species *Bulinus indicus* by Subba Rao *et al.* (1994) from Pune, Maharashtra, which was later synonymised as *Amerianna carinata* (H. Adams, 1861) by Brown (1997). However, the number of species in the genus and their distribution remain uncertain, requiring comprehensive revision (Ponder *et al.*, 2020). This study confirms the occurrence of a hitherto undetermined *Amerianna* sp. in southern India, and provides first molecular genus data obtained from Indian material.

Material and Methods

During a diversity survey in September 2020, ten live specimens of *Amerianna* were collected from the Erroor region (9°58'56.99"N 76°19'38.13"E) of Edappally canal, Kochi, Kerala (Figure 1). The specimens were collected and fixed in 4% buffered formalin. In the laboratory,

samples were sorted and identified to genus level. Specimens intended for molecular study were preserved in 70% ethanol. Basic water quality parameters (water temperature, turbidity, salinity and pH) were recorded using a portable multi-parameter waterproof meter (Make: HANNA, Model: HI98194).

Molecular Analysis

Genomic DNA was extracted from foot tissue using a DNA easy Blood and Tissue extraction kit (Qiagen). The mitochondrial cytochrome c oxidase subunit I (*cox1*) marker was amplified using the universal primers LCO1490 and HCO2198 (Folmer *et al.*, 1994). The cycling conditions were set as per the previous report (Jayachandran *et al.*, 2019). DNA sequencing was performed with an ABI PRISM Big Dye Terminator v3.1 cycle sequencing kit in an AB 3730 DNA analyzer (Life Technologies). The sequences obtained were compiled, aligned and analysed using BioEdit v7.2.5. (Hall, 1999). A sequence similarity query was conducted with the GenBank database, using the Standard Nucleotide Basic Local Alignment Search Tool [BLAST] (<https://blast.ncbi.nlm.nih.gov/Blast.cgi>). A phylogenetic tree was constructed based on the maximum likelihood method using MEGA 10.2.2 with

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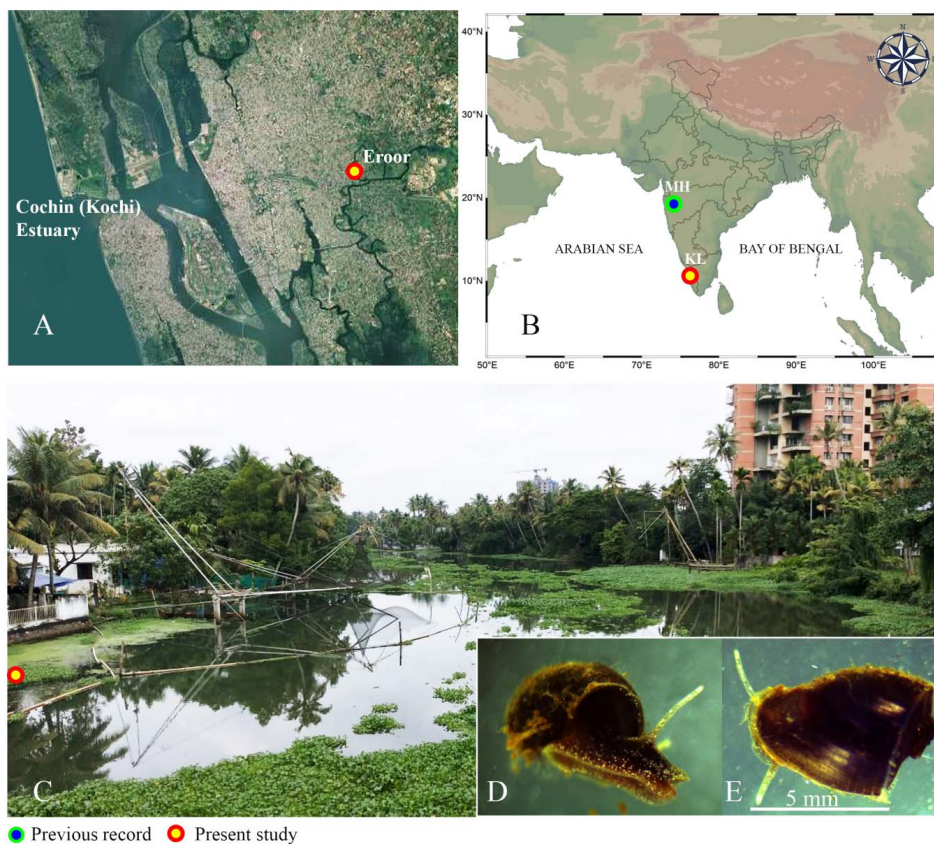


Figure 1. (A) present study location in Edapally canal of Kochi, Kerala, India; (B) Map showing the location of previous and present record of species of genus *Amerianna* from India (MH: Maharashtra & KL: Kerala); (C) habitat of *Amerianna* sp. collected from Edapally canal of Kochi, Kerala, India; (D and E) Habitus of *Amerianna* sp. from the study area.

bootstrap values set for 1000 replicates (Tamura & Nei, 1993; Kumar *et al.*, 2018). The partial *cox1* sequences were submitted to GenBank (accession numbers MW386166, MW386169). The voucher specimens were deposited in the museum collections of the Department of Marine Biology, Microbiology and Biochemistry, CUSAT, Kochi, India (MBM/SBN/JCPR/31-34/2020).

Results

Identification and description of the shell.

Remarks

Shell broadly sinistral, with truncate spire; last whorl strongly keeled, with rounded shoulders and raised stepped spire; aperture length approaching shell length, angular posteriorly, narrowly curve danteriorly, with slight columellar fold. Shell surface smooth, with spiral

rows of periostracal hairs. Average dimensions of voucher shells were: length 7 ± 0.9 mm, width 4.9 ± 0.5 mm, aperture length 6.4 ± 0.6 mm, aperture width 2.8 ± 0.4 mm, and spire length 1.1 ± 0.3 mm. Empty shells has a semi-transparent golden yellowish colour, while the body whorl of live specimens appears to be dark brown (Figure 2A–D) (Iredale 1943; Walker 1988). Based on the above-mentioned features as well as molecular data (see Molecular Results), the material studied herein is assigned to *Amerianna* (Figure 3A–B).

Molecular Results

Four specimens were identified by sequencing the *cox1* gene. The nucleotide BLAST results of all sequences showed a 99.2% similarity with '*Amerianna* sp. 1' isolates from New Guinean material (MT883681). A phylogenetic

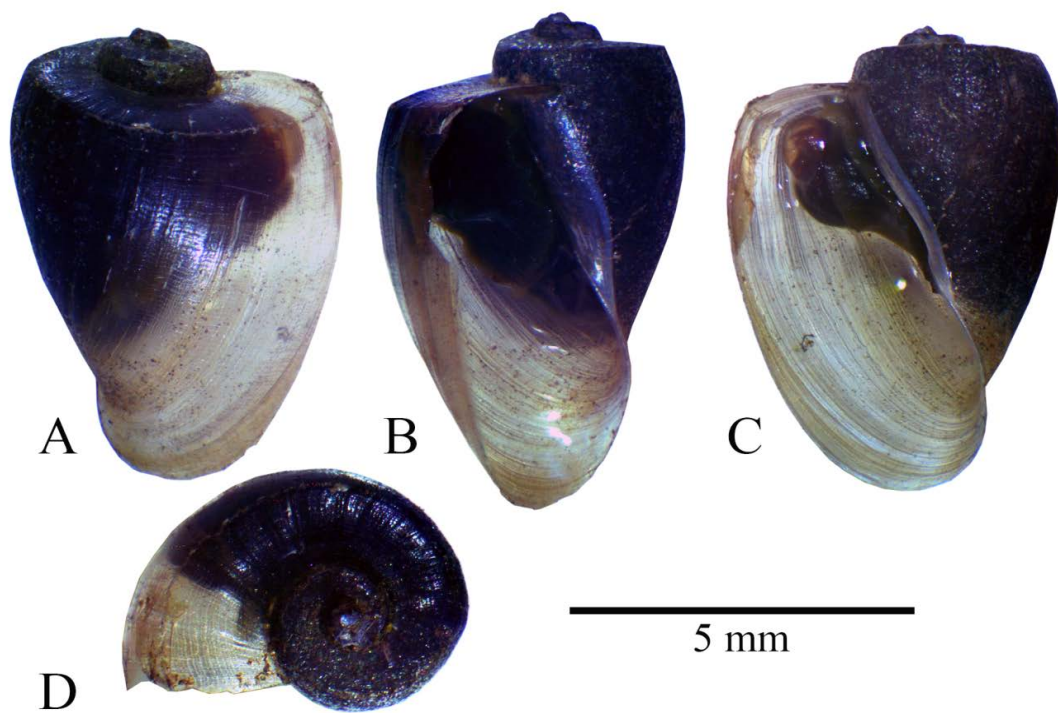


Figure 2. A–D. Shell of *Amerianna* sp. found in the Edappally canal, Kochi, Kerala (freshly preserved specimen).

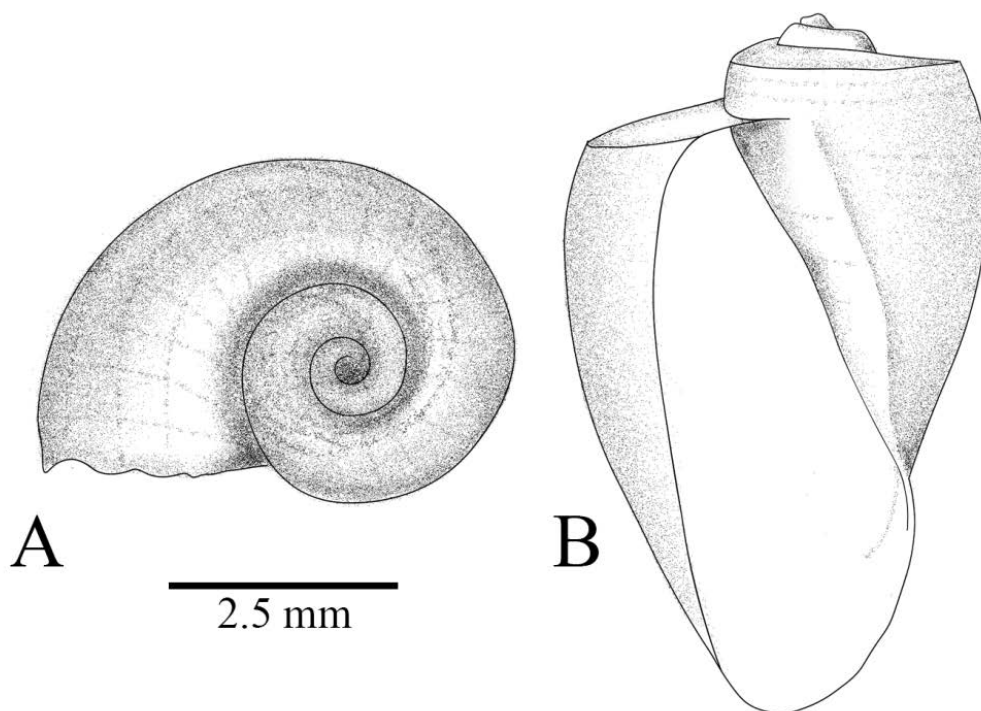


Figure 3. A–B. Illustration of the shell of *Amerianna* sp. found in the Edappally canal, Kochi, Kerala.

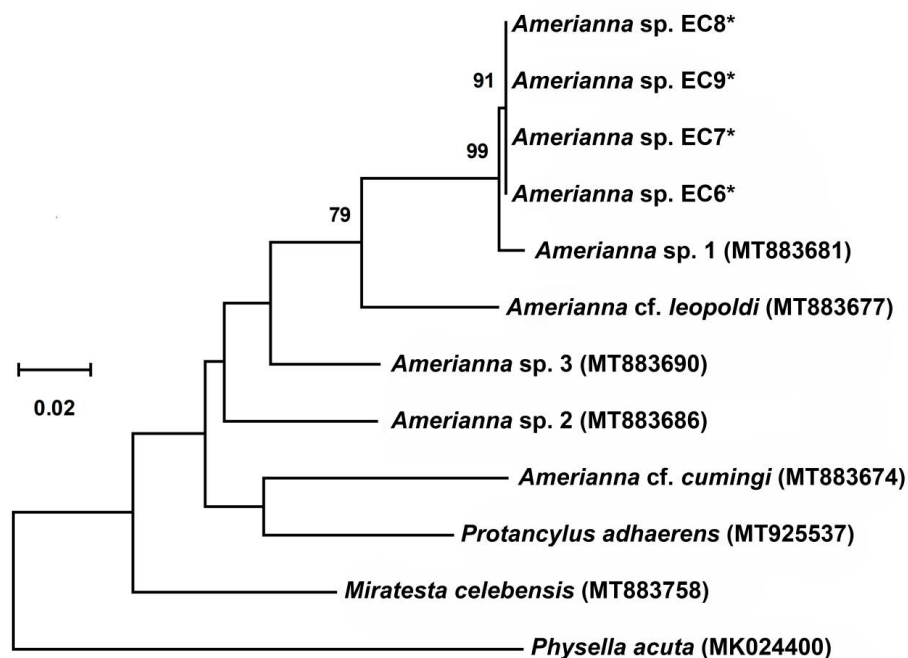


Figure 4. Maximum Likelihood tree based on the *cox1* gene showing the phylogenetic relationship between *Amerianna* sp. 1 from New Guinea with *Physella acuta* as outgroup. Bootstrap results from 1000 replicates with >70% support are shown on the branches. Sequences of *Amerianna* sp. obtained from the Edappally canal, Kochi, Kerala, India in the present study are given with the strain numbers EC6–EC9*.

tree (Figure 4) was constructed based on *cox1* sequences using the Maximum Likelihood method.

Field Observations

In the present study, *Amerianna* sp.1 co-occurred with gastropod species belonging to Physidae, *Physella acuta* (Draparnaud, 1805), Bulinidae, *Indoplanorbis exustus* (Deshayes, 1833), and Lymnaeidae, *Recesina luteola* (Lamarck, 1822). All these snails were attached to the freshwater plant species *Ceratophyllum demersum* (Linnaeus, 1753). The density of the *Amerianna* sp. was recorded as 10 ± 2 individuals on 40 cm plant shoot. The recorded depth ranges from 0.5 to 1 m. Other parameters were: water temperature $28.79 \pm 0.06^\circ\text{C}$; turbidity 3.15 ± 0.07 NTU; salinity 0.105 ± 0.01 PSU; pH 6.36 ± 0.02 , and dissolved oxygen 2.71 ± 0.49 mg/L.

Discussion

The genus *Amerianna* is known from the northern regions of the Australian mainland, New Guinea, Moluccas and

the Philippines. It was reported for the first time out of its native range in Java in 1951 by Butotin aquariums and ponds of the Bogor Botanic Gardens. Its spread in Java was confirmed in the following years by Van Benthem Jutting (1956). *Amerianna carinata* was subsequently reported from Thailand by Brandt (1974), Nigeria by Brown (1983), and the Lesser Antilles by Pointier (1996). Little is known about its ecology and invasive capability. However, this species thus far appears to have limited its introduction to particular habitats such as artificial ponds, botanical gardens, and small ditches along river banks.

In India, this genus has been recorded from fresh water habitats of Pune, Maharashtra state, as *Bulinus indicus*. Recently, Ramitha and Vasandakumar (2015) reported *Bulinus indicus* from freshwater habitats of the Malabar region in Kerala. However, there are no photographs or drawings available for comparison. Similarly, Mukhopadhyay *et al.* (2017) published an article regarding the introduction of *Planorbella scalaris* (Jay, 1839) (figures 1, 2, p. 517) in India, but photographs from the publication indicates a far closer affinity to *Amerianna*. The present study confirms the presence

of *Amerianna* in southern India and presents with first molecular data from India. Based on a comparison of the *cox1* gene, the Indian material (Kochi, Kerala) appears closely matched with *Amerianna* sp. 1 from West Papua (see Gauffre-Autelin *et al.*, 2021). However, we are unable to assess the morphology of the New Guinean material, since the authors did not provide figure or incorporate any morphological information. At the same time, Van Benthem Jutting (1956) figured an introduced species from Indonesia (Java) (figure 1, 2) referred to as *Amerianna carinata*. However, the recent study by Gauffre-Autelin *et al.* (2021) on biogeographic patterns of fresh water planorbids in the Indo-Australian Archipelago shows that *Amerianna* systematics is more complex than previously thought. Pending a thorough taxonomic revision of the genus, species assignments are therefore fraught with uncertainty and must be approached as tentative only.

Generally, members of the genus are found in shallow lentic and lotic water bodies, and are commonly inadvertently introduced to fresh water habitats outside of their natural range. Their air-breathing capability may facilitate this dispersal and adaptability to new habitats, even in eutrophicated water bodies. In fresh water systems around the globe, the aquarium trade is considered a major vector for the introduction of nearly 150 invasive animal species from 50 families (Chang *et al.*, 2009). Furthermore, the increasing demand for exotic ornamental species results in an ever-growing human-mediated transport of exotic species, and it is therefore likely these figures will grow. In the present study, the undetermined *Amerianna* sp. has so far only been recorded

from the Edappally canal of Kochi (Figure 1). However, the species may exhibit high potential for further spread in southern India. Therefore, a comprehensive study on the molecular phylogenetics and evolution of this genus is required in order to enhance our understanding of their distribution, invasiveness, and impact on the native resources, to facilitate effective management of the fresh waters systems of Kerala. Hopefully, the sequence material and morphological information presented in this study should provide a baseline with which further records can be compared. This may serve to establish whether there is, to date, only one or more *Amerianna* species present in India.

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