ORIGINAL ARTICLE

New geographic record of Peters’s Trumpet-eared Bat *Phoniscus jagorii* (Peters, 1866) from India

Sreehari Raman¹,²,³,*, Akhil Padmarajan⁴, Liju Thomas⁵, Arya Sidharthan⁵, Alice C. Hughes¹,²,⁶,*

¹ Center for Integrative Conservation, Xishuangbanna Tropical Botanical Garden, Chinese Academy of Sciences, Menglun, Mengla, Yunnan Province, 666303, PR China.
² University of Chinese Academy of Sciences, Beijing 100049, PR China.
³ Department of Wildlife Sciences, College of Forestry, Kerala Agricultural University, KAU P.O. (680656), Vellanikkara, Thrissur, Kerala, India.
⁴ Rani Bhavan, Muttappalam, Perumugzh P.O., Chirayinkeezhu, 695305, Thiruvananthapuram, Kerala, India.
⁵ School of Ocean Science and Technology, Kerala University of Fisheries and Ocean Studies (KUFOS), Kochi, Kerala, India.
⁶ Southeast Asia Biodiversity Research Institute, Chinese Academy of Sciences, Menglun, Mengla, Yunnan, 666303, PR China.
*Corresponding author: ach_conservation2@hotmail.com


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ABSTRACT

Peters’s Trumpet-eared Bat (*Phoniscus jagorii*) was recently recorded from Sri Lanka (2,100 km west of the nearest known range), which extends its distributional range significantly. We recorded *P. jagorii* in peninsular India, providing the first confirmed record from India and the second from South Asia. Phylogenetic analysis using cytochrome oxidase I (COI) gene shows similarities between the Indian and Southeast Asian specimens. Comprehensive bat surveys carried out in south India showed that the *P. jagorii* is more elusive than other bat species in this region and suggests further studies are needed to enable long-term conservation of the species.

INTRODUCTION

The genus *Phoniscus* includes *Phoniscus atrox*, *P. papuensis*, *P. jagorii* and *P. aerosa* (Hill 1965, Corbet & Hill 1992, Simmons & Cirranello 2020). *Phoniscus* was previously classified within the genus *Kerivoula* owing to their morphological similarity and long woolly fur (Gray 1842, Miller 1905). Hill (1965) separated *Phoniscus* from *Kerivoula* due to differences in the shape of the tragus, fur and dental morphology. The group is largely distributed in Southeast Asia (SEA), except *P. aerosa* which is known from South Africa and *P. papuensis* from Papua and the eastern coast of Australia.

*Phoniscus jagorii* has characteristic four-banded hairs with dark grey base followed by buff band, dark brown, and golden tips along with shiny yellow hairs across the body, forearm and fingers (Hill 1965, Corbet & Hill 1992). The species inhabits lowland rainforest, as well as dry dipterocarp and semi-evergreen forests (Oo et al. 2019 – Fig. 1). Later, a road-killed specimen of this species was collected from Sri Lanka (7.724550°N and 81.212600°E) on 9th August 2015; around 2100 km west of the nearest known range across the Andaman Sea and Bay of Bengal (Edirisinghe et al. 2018).

Here we present a new record of *P. jagorii* from South Asia, the first record from India and the second record outside SEA. We discuss the morphology, vocalisation and the possible colonisation routes that assisted in the movement of the taxon between South Asia and SEA.

MATERIALS AND METHODS

Study area

In the southern Western Ghats (WG), several field surveys were conducted at Nelliampathy Hills of Nenmara forest division between July 2017 and June 2018. The study area is a reserve forest on the western slopes of the WG (10°25’–10°49’N, 76°26’–76°54’E) with a total extent of 348.86sq.km. The elevation ranges from 40 m to 1633 m, and majority of the landscape is covered by tropical wet
evergreen, semi-evergreen, moist deciduous, grasslands, shola forests, and plantations of cardamom, coffee, tea and rubber (Ramachandran & Suganthasakthivel 2010). Recently a comprehensive bat survey was conducted by Wordley et al. 2014, 2015, 2017 in Valparai, which is ~50km from Nelliyampathy Hills and documented 17 bat species. Apart from this study, no detailed bat surveys had previously been undertaken in this region.

Bat sampling and echolocation call recordings

Sampling was carried out at varying elevations using two harp traps of 1.5 m width, 2.2 m height, 7.5 cm between each of four frames, 2.5 cm between vertical monofilament fishing lines for an overall trapping effort of 864 trap hours (6 days x 6 stations x 2 traps x 12 hours) between July 2017 and June 2018. Identification followed available keys (Francis 2008) using standard morphometric measurements and other morphological characters. Echolocation calls were recorded after releasing bats inside a mosquito net made of nylon mesh (dimension of 2 m length × 2 m breadth × 2 m height) using a full spectrum M500-384 ultrasound detector (Pettersson Elektronik AB, frequency range 1–384kHz). Thirty pulses with the highest signal to noise ratio were selected from the recording and nine call parameters such as start frequency (Fstart), end frequency (Fend), call duration (D), Inter-pulse interval (IPI), frequency of maximum energy (FmaxE), Bandwidth (BW) and duty cycle (DC) were measured using BatSound (ver. 3.31, Pettersson Electronik AB). We calculated mean values and standard deviations of all the nine call parameters. After collecting acoustic data, the individual was euthanised following the standard protocol (Sikes et al. 2011), and the specimen was deposited in the wet collections of the Department of Wildlife Sciences, College of Forestry, Kerala Agricultural University.

DNA sequencing and analysis

DNA was extracted from thigh muscle tissues using the DNeasy Blood and Tissue kit (Qiagen, Hilden, Germany. Catalog No. 69504) following the manufacturer’s protocol. The polymerase chain reaction was performed to amplify the mitochondrial cytochrome oxidase 1 (CO1) gene using the primer pair VF1 and VR1 (Ivanova et al. 2006). COI was used due to the availability of comparable material from other parts of SEA (Francis et al. 2010). PCR amplification was done with a reaction profile of 95°C for 180s; 35 cycles of 94°C for 60s, 55°C for 60s, and 72°C for 60s and finally 72°C for 300s, and the PCR products were outsourced for sequencing. The sequence generated as part of the present study was deposited in GenBank (Accession Number: MN255825). Publicly available COI sequences of *P. jagorii* (Francis et al. 2010 –HM541207 to HM541216 and HM541206) and *Kerivoula lenis* (KY034131) were downloaded from NCBI (http://www.ncbi.nlm.nih.gov), and *P. atrox* (ABBID027, ABBID067) from BOLD (http://www.boldsystems.org) database. *Kerivoula lenis* (Chiroptera: Vespertilionidae) was used as an outgroup. The integrity of the sequences was checked using BLAST (Altschul et al. 1990). Sequences were aligned using Clustal Omega (https://
RESULTS

Morphology

On 18 July 2017, a single adult female *P. jagorii* was trapped in a harp trap at 2000 h set across a dirt-road passing through an evergreen forest patch at Nelliyampathy hills (10°31’57”N, 76°40’8”E) at an elevation of 679 m (Fig. 1). The body mass of the collected adult female was found to be 7.5 g and standard measurements (in mm) are given in Table 1A. Body hairs appear to have characteristic four colour bands starting from the dark brown or blackish-brown base, followed by buff, then brown, and finally golden or whitish-yellow tips.

Echolocation calls

*P. jagorii* emits low DC broadband frequency modulated (FM) calls with an FmaxE of 113.79kHz (Fig. 2). Various echolocation call parameters (mean, standard deviation and range) are given in Table 1B.

Table 1 - A: External measurements (in mm) of *Phoniscus jagorii* recently collected from Nelliyampathy Hills, southern WG, peninsular India. B: Echolocation call parameters and its Mean ± SD (minimum–maximum). Fstart, Fend and FmaxE are in kHz. D and IPI are in ms. DC is calculated by (D/IPI)x100.

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<tr>
<th>A. External measurements</th>
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<tr>
<td><strong>Variables</strong></td>
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<td>Forearm length</td>
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<td>Thumb</td>
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<td>Fifth metacarpal</td>
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<td>Second phalanx of the fifth metacarpal</td>
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<td>Wingspan (tip-tip)</td>
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| B. Echolocation call parameters |
|----------------|----------------|
| **Variables** | **Values** |
| Start frequency (Fstart) | 140.88 ± 4.14 (134.25–146.34) |
| End frequency (Fend) | 80.75 ± 2.79 (78.0–86.08) |
| Frequency of maximum energy (FmaxE) | 113.79 ± 4.86 (105.34–120.40) |
| Band width (BW) | 60.14 ± 4.49 (53.76–65.29) |
| Duration (D) | 1.39 ± 0.15 (1.22–1.62) |
| Inter-pulse interval (IPI) | 15.62 ± 2.13 (11.62–18.07) |
| Duty cycle (DC) | 9.02 ± 1.60 (7.67–11.76) |
DISCUSSION

The recent discovery of *P. jagorii* in the evergreen forests of WG confirms the presence of this species in peninsular India. The present record is approximately 600 km from the nearest known location in Sri Lanka and 2850 km from the closest populations in mainland SEA (Myanmar) (Oo et al. 2019). The size and external morphology of the individual collected from WG and the Sri Lankan specimen falls within the size range of SEA population (Francis 2008). *P. jagorii* is known to be a low-flyer (Thong et al. 2006), and distributed in primary to moderately disturbed semi-evergreen forest where it has previously been recorded in SEA (Oo et al. 2019). Despite 72 trapping nights in Nenmara forest division, we were only able to record a single individual suggesting this species, is a rare species in this region.

Echolocation calls

The low duty cycle echolocation calls of *P. jagorii* are similar to the calls produced by Kerivouline and Murininae (Kingston et al. 1999, Schmieder et al. 2010, 2012) and are successful in tracking arthropods in the dense rainforest understorey (Schnitzler et al. 2003, Schmieder et al. 2010). Echolocation calls can vary geographically within a species (Aspetsberger et al. 2003, Yoshino et al. 2006, Armstrong & Coles 2007, Gillam & McCracken 2007, Hughes et al. 2010, Wordley et al. 2014, Zhang et al. 2018) and the extent of call variability over a wider geographical area can help to identify cryptic species and evolutionarily significant units (ESUs) that can contribute in conservation planning (Crandall et al. 2000, Davidson-Watts et al. 2006, Bickford et al. 2007, Frankham 2010). However, the

**Phylogenetic analysis**

Maximum Likelihood tree constructed using MEGA 7 shows that the sequence from India lies between the specimens from Laos (Fig. 3), which shows the affinity of the Indian population of *P. jagorii* with SEA. The phylogenetic reconstruction shows 1–14 nucleotide difference between the sequences.
frequency ($F_{\text{maxE}}$) emitted by the captured individual was within the frequency limit 94.4–124.1kHz that has been previously recorded from its known distribution range (Kingston et al. 1999, Thong et al. 2006, Schmieder et al. 2010, 2012).

Phylogenetic relationship and possible colonisation passages

The affinity of peninsular Indian species with those from the eastern Himalayan or Malayan region has been observed since Hora’s (1949) Satpura hypothesis. The similarity was also validated by various biogeographic studies on different taxa (Karanth 2003, Puri et al. 2016, Garg & Biju 2019). Westward colonisation of Malayan fauna began when the Indian plate joined mainland Eurasia during mid-Eocene, around 45 million years ago (Chapman & Reiss 1992), and the dispersal rate peaked during mid-Miocene (Klaus et al. 2016). The Eastern Himalaya is often considered as a ‘gateway’ that bridges Indian subcontinent with the rest of Asia (Mani 1974). Due to continued changing climatic conditions and tectonic movements, the dispersal rate started decreasing from 14 Mya (Patnaik 2011, Klaus et al. 2016). Few possible passages have been suggested to explain the dispersal of species from the eastern Himalayas to the Western Ghats (Fig.1). ‘Satpura hypothesis’ (Hora 1949) suggested a specific colonization route through the wet forests of Satpura Hill ranges in central India for species to disperse between the two mountain ranges. Areas that lie to the north of the Satpura range and the ‘Brij area’ may have acted as corridors (Dilger 1952). The Satpura hypothesis has been supported by observations from various freshwater fish species (Hora 1944, Silas 1952, Daniels 2001, Negi et al. 2017), birds (Ali 1949, Srinivasan & Prashanth 2006, Wagh et al. 2011); and certain plants (Jain et al. 2000, Kutapetty et al. 2014, Sen et al. 2019). However, the hypothesis has been debated due to the relationship with species in peninsular India with Shivalik and western Himalayan species suggesting a possible colonisation route through the Aravalli Hill ranges--‘Medlicott-Blanford theory’ (Medlicott & Blanford 1879, Daniels 2001). Another proposed colonisation route is between Eastern Himalaya and WG through the Eastern Ghats (Abdulali 1949, Mani 1974), and this view is supported by the distribution of 13 bird species (for example, Coprimalus atripennis, Alcedo meninting, Merops leschenaultia – Srinivasan & Prashanth 2006). However, in the absence of comprehensive bat surveys from the Aravallis, Satpura Hills, and the Eastern Ghats, it is hard to tell which of the above routes is most likely. *P. jagorii* might have colonised Sri Lanka from peninsular India when the sea level dropped as a result of global glaciation and drying during the Eocene-Oligocene boundary, i.e., about 34 Mya (Salamy & Zachos 1999, Liu et al. 2009). This may have facilitated the movement of taxa between peninsular India and Sri Lanka (Jacob 1949, Bossuyt et al. 2004, Biswas & Pawar 2006, Meegaskumbura et al. 2019). We, therefore, recommend that intensive surveys be undertaken in potential habitats in peninsular India, followed by comprehensive genetic studies to elucidate the colonisation patterns of bats in India.

CONCLUSION

The recent discovery of *P. jagorii* in peninsular India shows that there is still much to learn about the species and others in this region. Phylogenetic analysis shows no significant differentiation across its distributional range, indicating the similarities between South Asian population of *P. jagorii* with that of SEA. We recommend further inventories across the region for a better understanding of the species range, and therefore provide baseline information for the long-term conservation of the elusive species.

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