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**Front cover:** *Zhangixalus smaragdinus* from Dimapur tiger reserve, Mizoram, India. Photographed by Samuel Lalronunga.

**Back cover:** *Calotes irawadi* from Mizoram, India. Photographed by Lal Muansanga.

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# A long-term monitoring programme to understand the impact of climate change on terrestrial herpetofauna of India.

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**ABSTRACT. Anthropogenic climate change is likely to have wide ranging impacts on all aspects of life on earth. Among the many challenges posed by climate change, of particular importance to biologists is its impact on biodiversity and its consequences for ecosystems. South Asia harbours a rich diversity of amphibians and reptiles, which are likely to be impacted by changes in temperature and precipitation patterns. Any effort to mitigate the negative impact of climate change on herpetofauna in the region requires critical ecological and biological data, such as species distributions, population dynamics, phenology, thermal biology etc. Measures have to be put in place to monitor the status of species in key biodiversity areas so that detection of climate change impacts can help frame management interventions. The Long-term Ecological Observatories programme on herpetofauna is an initiative to create such a monitoring programme in different parts of India. In this paper, we provide a background to this project and outline an overview and some expected outcomes.**

**KEYWORDS. Climate change, South Asia, Herpetofauna, Ecology, Conservation**

## Introduction

Anthropogenic global climate change can no longer be ignored by environmental, ecological, and conservation programmes. Set in motion by the industrial revolution and the growth of the fossil fuel industry, massive amounts of greenhouse gases are still released into the atmosphere. In the last century alone, average global temperatures increased by more than 1.07 °C (NASA 2021, NOAA 2021). The global mean sea level has increased by 0.2 m, with an accelerating trend in the last few decades (Chen et al. 2017). Climate zones over land have generally shifted towards the poles, and warm seasons are on average two days longer every decade since the 1950s. Extreme weather events including heat waves and heavy precipitation have also increased in the same period (Masson-Delmotte et al. 2021). When global climate change interacts with another large-scale crisis, the rapid loss of biodiversity that is now identified as the sixth mass extinction, the consequences are many (Thomas et al. 2004, Wake and Vredenburg 2008, Bellard et al. 2012, Araújo et al. 2013, Ceballos et al. 2015, Urban 2015, Urban et al. 2016, Pecl et al. 2017).

To avoid extinction due to climate change, species may respond in three ways: (a) by moving across space based on changing environmental conditions, (b) by temporally changing states or events, such as life history traits or breeding periods and/or (c) adaptively adjusting physiological and evolutionary responses (Bellard et al. 2012). The first of these responses can lead to changes in spatial distribution patterns, range sizes, and range limits of species. For example, northward range shifts in many temperate taxa (Parmesan 2006, Duan et al. 2016), and upward elevation changes in distribution in tropical species due to the strong temperature gradient that exists in many tropical mountain ranges (Colwell et al. 2008, Bickford et al. 2010, Menéndez et al. 2014, Kusrini et al. 2017). The second type of response, temporal changes in states or events, can lead to changes in activity patterns, phenology, dispersal, migration, etc. Many species rely on environmental cues such as rainfall to time their life-history attributes, such as metamorphosis, breeding, number of offspring produced etc. Adaptive changes in temporal activity patterns, either seasonal or daily, will be necessary for many species. The third type of response, termed 'self' by Bellard et al (2012) includes physiological and genetic changes that can lead to changes in the biology of the species, such as thermal responses and strategies, or preference of food or microhabitat (Williams et al. 2008). For example, within a population, selection for a particular allele that influences higher thermal tolerance may happen, leading to rapid microevolution. Failing to respond adaptively along any of these three pathways may lead a species to reduction in populations, ultimately leading to either population or species extinction (Bellard et al. 2012, Thurman et al. 2020). The speed with which current climate change is occurring makes it unlikely that many species will evolve adaptations in time. Additionally, our knowledge of the biology and ecology of majority of species is so incomplete that we cannot predict with confidence the number of affected species and the consequences for ecosystems.

In this paper, we provide a broad introduction to the impact of climate change on herpetofauna globally, and review knowledge of impacts on these taxa in South Asia. We then introduce the Long-Term Ecological Observatories programme, and outline our rationale and approach to establishing long-term monitoring for herpetofauna in India. We conclude by encouraging other researchers to take up similar efforts so that we collectively fill the gaps in our knowledge and work to conserve the herpetofaunal diversity of this ancient landscape.

## Herpetofauna and climate change

There are nearly 20,000 species of extant amphibians and reptiles, with more species added every year (Frost 2022, Uetz et al. 2022). Unfortunately, herpetofauna are also among the most threatened group of vertebrates, with at least 41% percent of amphibians and 21% of reptiles considered threatened (Green 1997, Gibbons et al. 2000, Beebee and Griffiths 2005, McCallum 2007, Inger et al. 2009, IUCN 2021). Globally, estimates suggest a minimum extinction rate of nearly 7% for amphibians over the next hundred years (McCallum 2007, Alroy 2015). This is a result of most of the common reasons attributed to biodiversity loss in general: habitat loss and degradation, unsustainable use, infectious diseases, etc (Lips 1998, Daszak et al. 1999, Gibbons et al. 2000, Stuart et al. 2004, McCallum 2007, Nunes et al. 2019). While most of the above have caused local or regional losses of herpetofaunal diversity, global climate change is a large-scale threat that also interacts with all the others, and can have far-reaching effects on the future of herpetofaunal diversity (Thomas et al. 2004, Lopez-Alcaide and Macip-Rios 2011).

Abiotic environmental factors, such as temperature and precipitation, have an enormous influence on the biology of amphibians and reptiles (Vitt and Caldwell 2009). Compared to endotherms, changes in the predicted distribution of ectotherms, such as amphibians and reptiles, more closely track the changes in temperature across the globe (Aragón et al. 2010). Thermal biology constrains herpetofauna to limited environmental conditions, though species can respond with behavioural strategies to maintain optimal or preferred thermal conditions (Angilletta 2009). With increasing temperatures, many herpetofauna are expected to face restricted activity windows and considerable physiological costs (Clusella-Trullas and Chown 2011). For tropical herpetofauna, this is an even greater problem as thermal tolerance ranges are already narrow and many species already function at the upper part of their preferred thermal ranges (Huey et al. 2012, Griffis-Kyle et al. 2018). Changes in intensity and timing of precipitation can affect many amphibians that synchronize their breeding activities with specific weather conditions (Blaustein et al. 2010). Changes in temperature and precipitation patterns can alter vegetation type, structure, composition, availability of both food and shelter, and predator-prey/parasite-host dynamics for many species (Parmesan 2006, Blaustein et al. 2010, Bastille-Rousseau et al. 2018). There is some evidence that increase in temperature can reduce the resistance of amphibians to the fungal pathogen *Batrachochytrium dendrobatidis* (also called Bd fungus) which is currently considered one of the leading causes of amphibian extinctions (Raffel et al. 2013). The response of herpetofauna to such changes could be along any of the previously mentioned pathways: spatial changes (e.g., range reductions, shifts, and changes in species abundance), temporal changes (e.g., changes in daily or seasonal activity patterns and breeding phenology), and physiological or genetic changes (e.g., changes in preferred active body temperatures) (Blaustein et al. 2010, Griffis-Kyle et al. 2018).

## South Asian herpetofauna and the threat of climate change

The tropical regions of the world have the highest diversity of herpetofauna, with a large majority of all known species occurring in the Neotropics, West Africa, Southeast Asia, and South Asia. These also include some of the most populous regions in the world. India, the second most populous country in the world, hosts four of the world's biodiversity hotspots: The Western Ghats – Sri Lanka hotspot, the Himalayas, the Indo-Burma hotspot (including parts of Northeast India and the Andaman Islands), and the Sundaland hotspot (including the Nicobar Islands) (Mittermeier et al. 2011). These regions are also amphibian and reptile biodiversity hotspots with more species discovered on a regular basis (e.g. Agarwal et al. 2014, Vijayakumar et al. 2014, Pal et al. 2021, Garg et al. 2021). These hotspots host a significant number of highly threatened, yet largely understudied herpetofauna. For example, more than 35% of amphibian species from the Western Ghats are considered Critically Endangered or Endangered, while a further 45% are Data Deficient or were not assessed for their conservation status (IUCN 2021). The latest assessment by IUCN suggests that among the 107 globally threatened Indian species of reptiles, 12 species are affected due to climate change and severe weather-related causes such as climate change mediated habitat shifting and alterations, droughts, temperature extremes, storms, flooding, and other impacts (IUCN 2021). Since a majority of species from this region lack data on population size, distribution, and other aspects of biology, these assessments are arrived at based largely on expert opinion on commonness or rarity of species, its current distribution, and observed as well as perceived threats. Data based assessments of vulnerability or adaptability to changing climate is not available for of most species.

With some of the highest human densities in the world coexisting with rich biota in South Asia, it is essential to consider these aspects when planning biodiversity conservation. Even conservative estimates suggest an increase of 2 °C in both mean and maximum temperatures throughout most of south Asia in the next fifty years or so (Iturbide et al. 2021, Masson-Delmotte et al. 2021). Days when maximum temperature rises above 35 °C could see an increase to 18 – 48 days, with more hot days in the southern and western regions (Masson-Delmotte et al. 2021). The south-west monsoon, which provides most of the rainfall to a large part of South Asia, is set to increase in the amount of rainfall, but with changes in intensity and timings (Katzenberger et al. 2021). The Himalayas are highly susceptible to adverse effects of global climate change, as the rate of increase in average temperature in the Himalayas is three times the global average (Xu et al. 2009) with a 0.06 °C increase per year (Shrestha et al. 2012). One should expect such changes to create significant fluctuations in local weather patterns also, with direct and indirect impacts on biodiversity throughout the region. However, a review of studies on the impact of climate change on herpetofauna across the world from 2005 to 2015 found no quantitative studies from this entire region (Winter et al. 2016). Instead, more than 70% of all studies pertained to North America and Europe. Addressing this major disparity in the distribution of diversity and information should be a priority in efforts to put the brakes on the biodiversity crisis.

## Current knowledge of climate change effects on Indian herpetofauna

Generally, there is a dearth of long-term ecological studies on amphibians and reptiles (other than Chelonians and Crocodilians) in south Asia. This is not to discount short-term studies on aspects of natural history and ecology, which are foundational. Among reptiles, extreme temperatures may affect sex ratios due to Temperature mediated Sex Determination (TSD) (Pezaro et al. 2017). Higher incubation temperatures result in producing more females in turtles and more males in crocodilians, both of which can affect long-term population survival (Lang and Andrews 1994, Valenzuela et al. 2019). Large-scale studies, such as those spanning the range of a species, or a landscape, or studies that span multiple generations are generally missing for most amphibians and squamate reptiles. Tackling the problem of climate change impact on biodiversity requires baseline information on distribution and population status, which is lacking for majority of Indian species. This information has to be combined with long-term ecological monitoring to detect changes and identify thresholds at which management intervention might be necessary.

Since the publication of the Winter et al. (2016) paper, there have been a few studies examining the potential impact of climate change on herpetofauna in this region (Srinivasulu and Srinivasulu 2016, Subba et al. 2018, Srinivasulu et al. 2021). Range shifts to higher elevations by low elevation herpetofauna have been reported for several species in the Himalayas (Acharya and Chettri 2012). This study also recorded changes in breeding seasonality of two amphibian species. Four species of Himalayan amphibians are expected to reduce their geographic distributions, indicating increased extinction risk (Subba et al. 2018). Habitat suitability models in conjunction with future climate models predict several endemic herpetofauna of southern India to shrink or shift their geographic ranges in response to increasing temperatures, with up to 73.48% reduction in climatically suitable habitat for some species by the year 2050 (Srinivasulu and Srinivasulu 2016, Srinivasulu et al. 2021). It should be noted that these studies analysed a single pathway of potential impact and response – potential changes in future climate suitability using current occurrence data – and cover less than 5% of all the species found in this region. Only one study (Subba et al. 2018) examined potential species migration scenarios, the likelihood of which can vary according to the nature of available habitats and other anthropogenic pressures. Species may respond to climate change and associated impacts through other response pathways. Therefore, depending on the specific scenarios, the chances of extinction may be lower or higher than that indicated by these studies. Therein lays a large space for studies, both local and regional, to fill information gaps, monitor status, and create better quantitative predictions of species responses to current and future threats.

## The Long-Term Ecological Observatories programme in India

Long-term studies targeting amphibians and reptiles are rare in general. Some of the more well-known, organized programmes that covered large areas include the ‘North American Amphibian Monitoring Program’ (NAAMP) (https://www.usgs.gov/centers/eesc/science/north-american-amphibian-monitoring-program), ‘Amphibian Research and Monitoring Initiative’ (ARMI) (https://armi.usgs.gov), and the United Kingdom’s National Amphibian and Reptile Monitoring Programme (https://monitoring.arc-trust.org). There are a few other somewhat long-term studies that focused on a species or a small region, such as the 11-year mark-recapture study of boreal toads (*Bufo boreas*) in Colorado, and the 31-year study of

Coachella Fringe-toed Lizards (*Uma inornata*) (Scherer et al. 2005, Fisher et al. 2020). However, almost all such efforts are in North America and Europe. To support effective climate change mitigation and biodiversity conservation, South Asia needs far more effort in large-scale and long-term projects.

The urgent need to understand climate change impacts was the main driver behind the launch of the 'Indian Long-Term Ecological Observatories' programme (https://lteo.iisc.ac.in/research/herpertofauna/) by India's Ministry of Environment, Forest and Climate Change (MoEF&CC), which envisioned an extensive network of biodiversity monitoring programmes spread across India (ILTEO, 2015). Under this umbrella project, there are multiple themes and groups working towards the common goal of understanding the effect of global climate change on India's biodiversity. For the first phase of the project, six regions representative of the biogeographic diversity of India were selected for establishment of long-term ecological observatories. These are North-west Arid Zone (NWAZ), Western Himalayas (WH), Eastern Himalayas (EH), Central Indian Forests (CI), Western Ghats (WG), and Andaman Islands (ANI). The herpetofauna group is a multi-institutional partnership (Fig. 1)

## Establishing a Long term herpetofaunal monitoring programme in India

The LTEO herpetofauna programme has three major objectives. The first is the establishment of monitoring sites and standardization of protocols. The LTEO monitoring programme will use spatial environmental gradients to understand the effects of environmental and climatic variables on distribution, abundance, thermal adaptation, and phenology of herpetofauna in six regions. In three regions, viz. WH, EH, and WG, monitoring will be along elevation gradients. Both WH and EH are part of the Himalayan Mountain range, and are characterized by extreme elevation gradients, though for the pur-

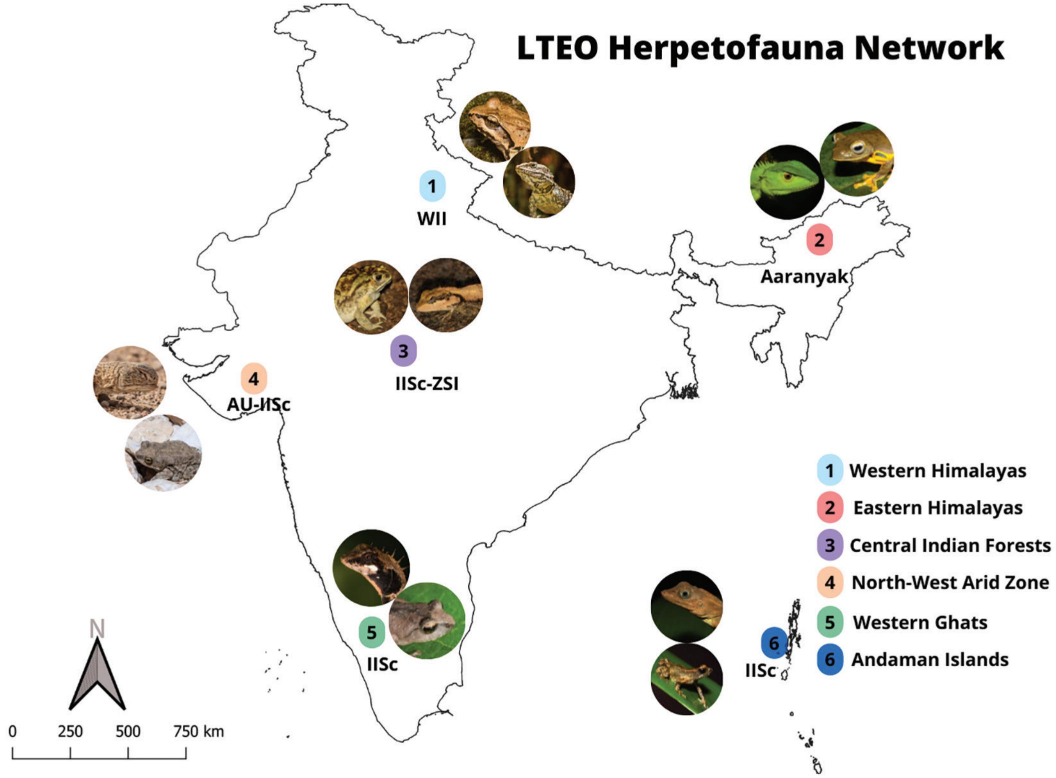

**Figure 1.** The six sites selected for the Long-Term Ecological Observatories for herpetofaunal monitoring in India. These sites cover both rainfall (sites 3, 4, & 5) and temperature gradients (sites 1, 2, & 5) along which representative species of amphibians and reptiles will be monitored. Institutional abbreviations are: WII – Wildlife Institute of India, Dehra Dun, Aaranyak – Aaranyak, Guwahati, ZSI – Zoological Survey of India, Kolkata, AU – Ahmedabad University, Ahmedabad, IISc – Indian Institute of Science, Bengaluru.

pose of herpetofaunal monitoring, sampling will be largely restricted to elevations below 3500 m above sea level (asl). The WG is a mountain chain running along the southwest coast of India. The region chosen for the LTEO sites fall in the Nilgiri-Wayanad region, having an elevation gradient between 500 to 2500 m asl.

In two other regions, viz., NWAZ and CIF, the monitoring will be along a rainfall gradient, in a west-southwest to northeast direction. This includes arid zones in the western part of India in the state of Gujarat and Rajasthan, with average annual rainfall ranging from 185 mm in the arid regions in the west to 2000 mm in the CIF. The ANI is a chain of islands in the Bay of Bengal. ANI has high average annual rainfall (over 3000 mm) and relatively warm, relatively stable temperature (20–35 °C) throughout the year. Since there are no major climatic gradients in ANI, monitoring will focus on long-term ecological changes in a few selected sites. We selected these sites based on representative vegetation for the region, level of anthropogenic disturbance, and ease of access for long-term, repeated sampling.

The second, and long-term, objective of the monitoring programme is to predict and detect changes in herpetofaunal communities, such as range shifts and changes in relative abundance, in response to climate change.  There is limited information available on commonness and rarity of species in the regions under consideration, as well as on efficiency of various sampling methods. Therefore, we plan to start by using a community or guild-based approach to sampling.  The short-term objective is therefore the collection of preliminary data and standardization of sampling/monitoring methods across the regions. We will collect data on occurrence of species, abundance, species richness, and a number of environmental covariates at each of the monitoring sites.

The primary requirement for detecting range shift along elevation gradient is mapping the upper and lower range limits as well as a measure of abundance. Often, a lowering of abundance at the range boundary is the first sign of range shifts. Therefore, early detection of range shifts will require detecting changes in abundance as well. The sampling strategy adopted should therefore be able to detect changes in occurrence as well as abundance over several years. Considering the logistic and analytical difficulties in estimating abundance of amphibians and reptiles, using site occupancy is the best choice for detecting changes over several years (MacKenzie et al. 2002, Mackenzie 2005, Muths et al. 2012, Adams et al. 2013, Grant et al. 2016). In the absence of initial estimates of occupancy and detection probabilities, we cannot derive objective estimates of the number of sites and surveys necessary for the most efficient survey design (Mackenzie and Royle 2005). Therefore, the goal of preliminary surveys in the first year will be to document species occurrence and relative abundance in the study sites through repeated sampling. We will use these results to refine the study design further in terms of focal taxa, number of sites, and sampling occasions.

In five of the six regions, we have identified transects along elevation and rainfall gradients, divided into minimum three strata (low, medium, and high). The survey locations within strata will be of dimensions 2 km x 2 km, consisting of 100 m × 100 m grids at each stratum. The general survey method used will be Visual Encounter Surveys (VES) in combination with acoustic surveys (Crump and Scott Jr 1994). VES yields the highest number of species and individuals for a given survey effort, while creating the least amount of disturbance to the habitat (Doan 2003). This makes the method repeatable during the same season and year after year. These surveys will provide occurrence, count, and species richness information as well as other biological parameters.

The third major objective of the programme is to understand species adaptations to changing climate. This will include changes in breeding biology, phenology, and thermal adaptations. These will be in the form of species-specific studies in each region, along the environmental gradient. After some preliminary data is collected, we plan to select a subset of species (where feasible) for population monitoring using a mark-recapture framework (Sutherland 2006, Patel and Das 2020). Repeated surveys will also help us collect information of breeding phenology for many species of amphibians. Based on this information, we will select a subset of species representative of the spatio-temporal and ecological space occupied by all species in the region, for long-term monitoring of population and breeding phenology. For understanding thermal adaptations, the primary requirement is capturing the thermal heterogeneity by sampling along the natural environmental gradient (e.g., rainfall or temperature) (Weatherhead et al. 2012, Díaz de la Vega-Pérez et al. 2019). We will collect Operative Temperature data for focal species by use copper models with thermo-loggers along the gradients, capturing as much of the thermal heterogeneity as possible (Angilletta 2009). To obtain body temperatures of animals and surface temperatures of substrates, we will use thermal images of lizards (Goller et al. 2014). LTEO weather station data will provide ambient temperature data.

At the end of preliminary sampling, we expect to have site-specific species lists, estimates of species occupancy along with detection probabilities for at least a subset of species present, and another measure of commonness/rarity of species (abundance or relative abundance) in all regions. During this period, we also expect to generate baseline data on thermal adaptations and breeding phenology of a subset of species. Based on these, we hope to refine long-term monitoring strategy. The six regions selected here are representative of the climatic variation across India. By identifying species, or species groups (based on biology and ecology) that are most vulnerable to changes in climatic factors in these regions, or traits that are common for species across these regions, we hope to extend the findings from these sites to other areas across South Asia. The LTEO programme is an ambitious project seeking to establish a first-of-its-kind ecological monitoring for herpetofauna across India. While the challenges are many, the successful implementation of the project can have immense impact on the conservation of species in this region.

## Conclusion

The current climate change crisis is perhaps the most drastic global climatic event that biodiversity has faced since the end of the Pleistocene ice age. Even with a significant reduction in future carbon emissions, the legacy of last two centuries of emissions is likely to have a lingering effect on earth's climate and push global temperatures up for several more decades. Therefore, there is a pressing need to devise mitigation strategies for the detrimental effects of global climate change. For biodiversity, this begins with monitoring changes, predicting species responses, and identifying thresholds for management intervention. The LTEO programme will help us predict and monitor the effects of climate change on herpetofaunal species so that detrimental changes are detected in time for appropriate adaptive management interventions. For example, enhancement and restoration of breeding sites may be necessary for some amphibian species under altered rainfall regimes, while habitat enhancement/manipulations might be necessary for survival of many amphibians and reptiles in hotter environments (Shoo et al. 2011) However, there are still large regions of the country, such as the Eastern Ghats and the coastal plains, and many species that cannot be covered through the current LTEO programme. It is our hope that more researchers will initiate long-term studies on herpetofaunal species across the country, especially in areas not covered by the LTEO programme, so that we can collectively build the knowledge-base necessary to combat the effects of global climate change.

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# First record of Anderson's Bubble-nest Frog *Raorchestes andersoni* (Ahl, 1927) (Amphibian: Rhacophoridae) from the Himalayan Kingdom of Bhutan.

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**ABSTRACT. We report the discovery of the Anderson's Bubble-nest Frog, Raorchestes andersoni, from Central Bhutan's Zhemgang district, in the Himalayan Kingdom of Bhutan. The frog was found in moist dense forests with lots of green bushes. This record will help Bhutan understand its species diversity and help in prioritizing conservation actions for this species and other batrachian fauna.**

**KEYWORDS. Conservation, distribution, habitat, location, morphometrics, species description**

## Introduction

Family Rhacophoridae (Hoffman, 1932), the closest relative of the true frogs Ranidae, Rafinesque, 1814, is so far represented by as many as 440 species belonging to 21 genera worldwide (Frost 2020), of which the genus *Raorchestes* has kept changing over the last few years based on proposals by various batrachologists conducting molecular as well as morphometric research (Hou et al. 2017). However, the accepted name *Raorchestes andersoni* (Ahl, 1927) is considered valid for now (Hou et al. 2017; Chen et al. 2020) for Anderson's Bubble-nest Frog, which has several nomenclatures like, *Ixalus tuberculatus* Anderson 1879, *Rhacophorus andersoni* Ahl 1927, *Rhacophorus (Philautus) andersoni*, Ahl 1931, *Philautus tuberculatus*, Bourret 1942, *Philautus andersonii*, Bourret 1942, *Rhacophorus (Philautus) andersoni*, Bourret 1942, *Philautus andersoni*, Bourret 1942, *Philautus (Philautus) tuberculatus* Bossuyt & Dubois 2001, *Aquixalus tuberculatus*, Fei, Hu, Ye & Huang 2009, *Theloderma andersoni*, Li, Che, Murphy, Zhao, Zhao, Rao & Zhang 2009, *Liuixalus tuberculatus*, Fei, Ye & Jiang 2012, *Theloderma (Theloderma) andersoni*, Poyarkov, Orlov, Moiseeva, Pawangkhanant, Ruangsuwan, Vassilieva, Galoyan, Nguyen & Gogoleva 2015, *Theloderma tuberculatus*, Hou, Yu, Chen, Liao, Zhang, Chen, Li & Orlov 2017 and *Raorchestes andersoni* Chen, Prendini, Wu, Zhang, Suwannapoom, Chen, Jin, Lemmon, Lemmon, Stuart, Raxworthy, Murphy, Yuan & Che 2020.

Other than the mention of habitat by Anderson (1878) as "on level marshy flats on the banks of the Nampoung in the centre of the Kakhyen Hills Myanmar", which was later explored by Hou et al. (2017) and confirmed to be in fact in China's Yingjiang County, Yunnan Province, there are no detailed studies on any other aspects of the species. Thus, it is pertinent that we report the presence of the species in Bhutan, towards studying this lesser-known frog.

The anuran fauna of the Kingdom of Bhutan is represented by ca. 79 species (Wangyal & Gurung 2017; Tenzin & Wangyal 2019; Wangyal et al. 2020; Wangyal et al. 2021) and the family Rhacophoridae, which is represented by 21 genera gets another species, *Raorchestes andersoni* from Bhutan in this research. Wangyal & Das (2014) mentioned the presence of Anderson's Bubble-nest Frog from Bhutan based on the information obtained through verbal interviews with local people living in Central Bhutan. Through a crude method of data collection where they showed the photos of the frogs to the interviewees and when they identified the type of frogs they have seen around in the region, they listed the species as present. They used habitat clues to predict the presence of *Theloderma* or *Raorchestes* species, and went on to ask people if they have seen the species at any point in time. Thus, they did not have enough information to verifiably say that the species existed. However, with this report, the occurrence of *Raorchestes andersoni* in Bhutan can be confirmed.

## Material and Methods

A visual encounter survey (VES) (Heyer et al. 1994) was conducted in the last week of August 2020 to look at the biodiversity of selected forest types of Shingkhar Gewog (sub-district). The frog was located on a curvilinear trail that led inside the forest vicinity of Zangling village. The second author noted geo-coordinates, elevation, plant species of the forest patch (Figure 1) near Zangling Village, in Shingkhar Gewog (sub-district), Zhemgang District, Central Bhutan (Figure 2).

The specimen was caught, photographed, measured and released back to the same place. Another round of VES did not yield any results till the end of August, as the vicinity is pretty cold. Ohler et al. (2018) was used to identify the species, and the speciment was also verified as *Raorchestes andersoni* by Annemarie Ohler through personal communication.

Habitat data, especially those of plant species surrounding the habitat, were collected, and plant species were identified using the book Flora of Bhutan (Barneby 1988).

## Results

**Species description.** Snout to vent length = 20.5 mm, length of hind limb = 35 mm, length of foot = 12 mm, and width of gape (Opened mouth) / head width = 7.5 mm. Snout — short (rather broad), canthus rostralis (CR) — feeble and rounded. Nostril little below the CR, small but with swollen margin. Eyes — large with finely tubercular eyelids. Tympanum about one-fourth the dimension of eye. Finger disc dilated and larger than those on the toes. Feebly webbed toes, reaching only up to the first phalanx. Small, scattered, isolated tubercles on the dorsum and sides of the body and belly. Inner part of the upper third of the thighs finely granular. General color of the upper parts is uniform dark olive with all white small tubercles (Figure 3a, 3b). The ventral color is brown with white granulated tubercles, but the belly has a white patch with white granulated tubercles too (Figure 3c). The groin has large black irregular spot. Morphological comparisons that distinguish *Raorchestes andersoni* from other species of *Raorchestes* in the region that include southeast Asia, Himalaya and northeastern India are provided in Table 1, which has been adapted from Wu et al. (2021) with minor changes.

**Location.** Zangling village, Shingkhar Gewog (sub-District), Zhemgang District. Geo-coordinates, 27.159722°N, 90.884748°E, 2006.300 m asl, obtained using a GPS with WGS 84 datum. Observed and caught on 25th August 2020 at 14:34:19 hrs (GMT+06:00). The species was found 730.9 km (to 4 SF*) away from the type Locality (24.445457°N, 97.535572°E, 1408 m asl), on the bank of the Nampoung River, China's Yingjiang County, Yunnan Province. The distance was obtained by using Movable Type Scripts (Veness 2002) found at URL: https://www.movable-type.co.uk/scripts/latlong.html. (Figure 5).

**Habitat.** There are no specific habitat or reproduction ecology studies on the species. However, it is known to occupy tropical forest edges and marshes (Döring 2020). In our study, the area had thick bushes (see Figure 1) and the species of trees found in the habitat were, *Symplocos sumuntia, S. ramossisima, Quercus ategoriz, Elaeocarpus sikkimensis, Acer campbellii, Eurya acuminata, Viburnum erubescens* and *Measa rugosa*. The shrubs on which the

**Figure 1.** The second author, while searching for orchids, bumped into a frog (Photo: Sherab Jamtsho).

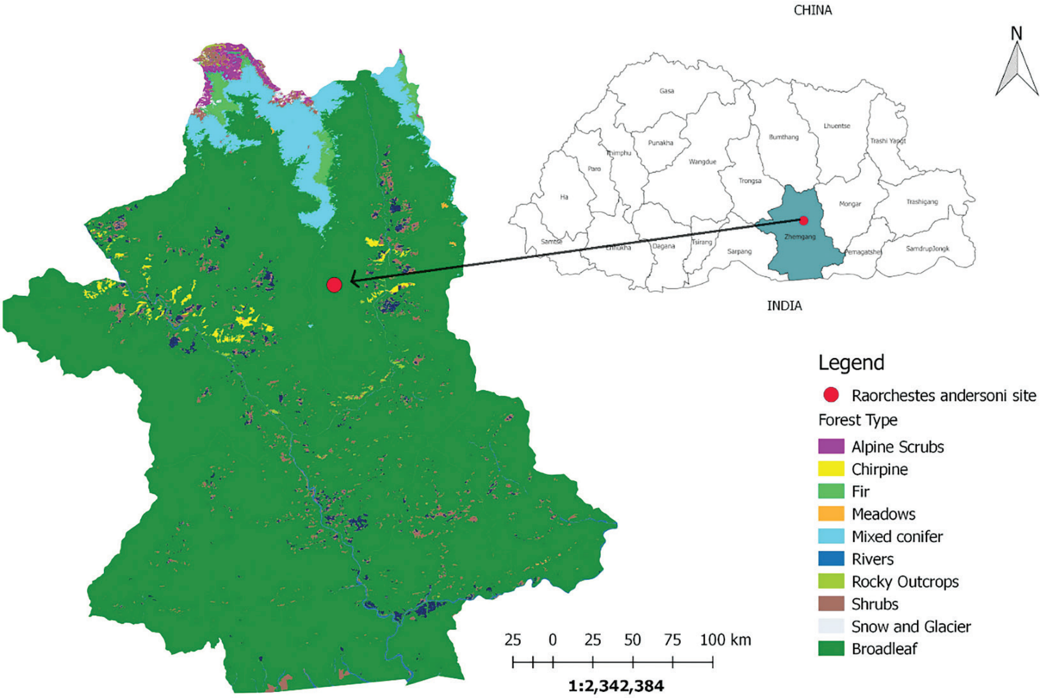

**Figure 2.** Map showing the exact spot where the *Raorchestes andersoni* specimen was found.

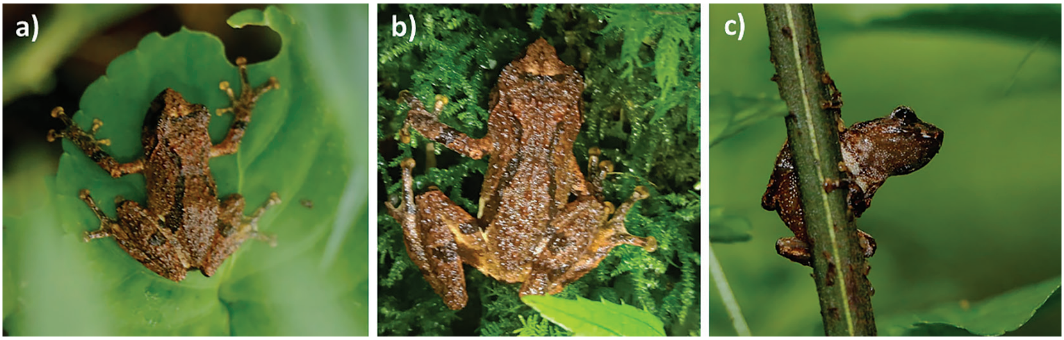

**Figure 3.** a) Dorsal view of *Raorchestes andersoni* in the field (Photo: Sherab Jamtsho).; b) Close-up view of the dorsum of *Raorchestes andersoni* (Photo: Sherab Jamtsho).; c) Ventral view of *Raorchestes andersoni* from the habitat (Photo: Sherab Jamtsho).

**Figure 4.** Distant view of the habitat from where *Raorchestes andersoni* was found (Photo: Sherab Jamtsho). In the background is Zhangling village.

**Figure 5.** The distance calculation between the type locality and the current location of *Raorchestes andersoni.*

frog hopped before being caught were *Dichora febrifuga*, *Melissa axillaries*, *Impatiens pseudo-lavigata*, *Elatostema sessile*, *E. himalayana, Piper suipigua, Thladianthia cordifolia, Urtica dioca, Viola atego, Rubus calycinus, Sarcopyramis* sp., *Clematis* sp. And plenty of *Diplazium* spp.(ferns). Our preliminary observation shows that the species likes thick forests with abundant bushes that hold a lot of moisture.

**Weather and climate.** The highest daily maximum temperatures recorded at Zhemgang when the frog was first spotted in May 2020 was 24.5 °C, while the minimum was 9 °C which averages to 17.5 °C. the highest daily rainfall recorded at Zhemgang in May 2020 was 101.6 mm, with the total rainfall for the month being 374.9 mm (https://www.nchm.gov.bt/).

**Conservation.** In the Red List of the International Union for Conservation of Nature (IUCN), the species is categorised as Least Concern (Van Dijk et al. 2004), while the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) does not list it. Unfortunately, the species does not receive any protection as no amphibians are listed either for protection or use in Bhutan. However, the habitats are well protected because of which animals are deemed safe. The photo at the study site (Figure 4) show that this species was discovered in a moist pristine forest protected by the Kingdom as a

**Table 1.** Morphological characters that distinguish *R. andersoni* from other *Raorchestes* species in the region (Adopted/adapted from Wu et al. 2021)

| Character | *R. dulongensis* | *R. menglaensis* | *R. longchuanesis* | *R. cangyuanensis* | *R. parvulus* | *R. gryllus* |
|---|---|---|---|---|---|---|
| **SVL of male (mm)** | 15.0–19.0 | 16 | 17.8–21.2 | 16.1–20.0 | | 25.0–27.0 |
| **HDL/HDW** | HDL>HDW | HDL almost equal with HDW | HDL almost equal with HDW | HDW>HDL | HDW>HDL | HDW>HDL |
| **Tympanum** | Distinct | Indistinct | Distinct | Distinct | Distinct | Indistinct |
| **Nuptial pad** | Absent | Present | Present | Present | | |
| **Vocal sac** | External single subgular vocal sac | Internal single subgular vocal sac | External single subgular vocal sac | External single subgular vocal sac | | A large subgular vocal sac |
| **Finger web** | Absent | Absent | Absent | Absent | Absent | A rudiment of a web between the two outers |
| **Toe web** | Rudimentary webbing | Rudimentary webbing or 1/4 webbing | 1/4 webbing | Rudimentary webbing | Webbing present, medium | A little more than half webbing |
| **Inner metacarpal tubercle** | Present | Present | Present | Present | Present | |
| **Outer metacarpal tubercle** | Present | Present | Present | Present | Present | |
| **Inner metatarsal tubercle** | Present | Present | Present | Present | Present | Present |
| **Outer metatarsal tubercle** | Absent | Present | Absent | Absent | Absent | Present |
| **Relative toe lengths** | I<II<V<III<IV | The third and fifth toes almost equal in length, or fifth toe slightly longer | The third and fifth toes almost equal in length | I<II<V<III<IV | I<II<III<V<IV | |
| **Range** | Gongshan, Yunnan, China | Mengla, Yunnan, China | Gaoligong Shan and Hengduanshan mountains, Yunnan, China; Lai Chau, Vietnam | Cangyuan, Yunnan, China | Indochina Peninsula and peninsular Malaysia | South Annam and Tonkin, Vietnam; Sepian, Boloven Highlands, Champasak Province, Laos |

Table 1. (continued)

| Character | *R. rezakhani* | *R. andersonii* | *R. annandalii* | *R. manipurensis* | *R. sahai* | *R. shillongensis* |
|---|---|---|---|---|---|---|
| **SVL of male (mm)** | 18.9–20.9 | 24 | 16 | 25 | 25.0–26.0 | 10.0-20.0 |
| **HDL/HDW** | HDW>HDL | HDW>HDL | HDW>HDL | HDW>HDL | HDW>HDL | HDW>HDL |
| **Tympanum** | Indistinct | Distinct | Distinct | Distinct | Distinct | Distinct |
| **Nuptial pad** | Absent | | | | | |
| **Vocal sac** | Vocal sac single, subgular, translucent | Internal single subgular vocal sac | A large subgular vocal sac | | | Males with large vocal sac |
| **Finger web** | Absent | Absent | Absent | Rudimentary webbing | Absent | Absent |
| **Toe web** | Webbing moderate, formula: I2-2+II1¾- 2+III1½-3IV2¾-2-V | Rudimentary webbing or 1/3 webbing | Webbed at the base | Almost 2/3rd webbed | Nearly half-webbed | A very indistinct rudiment of web present between the fourth and fifth |
| **Inner metacarpal tubercle** | Absent | Present | Present | | | |
| **Outer metacarpal tubercle** | Absent | Present | Present | | | |
| **Inner metatarsal tubercle** | Absent | Present | Absent | Present | | Indistinct |
| **Outer metatarsal tubercle** | Absent | Absent | Absent | Absent | | Present |
| **Relative toe lengths** | I<II<V<III<IV | I<II<IV | I<II<III<V<IV | | | I≤II<V≤III<IV |
| **Range** | Northeastern Bangladesh | Northeast India, North Myanmar, Southeast Tibet and Yunnan, China | Himalayas and northeastern India | Churachandpur district, Manipur, northeastern India | Eastern Himalayas | Khasi Hills in the region of Shillong, Meghalaya, northeastern India |

biological corridor (which has a special conservation status) that connects two protected areas of Bhutan. Also, the country with a majority of Buddhism followers considers the killing of any animals a sin and thus, the animals can be considered to be safe in Bhutan.

**Distribution.** The species is so far not known from other areas of Bhutan other than the current spot. Outside Bhutan, the species is known to occur in the northeast Indian states of Arunachal Pradesh (Sarkar & Ray 2006), Assam, Meghalaya, Mizoram and Nagaland (Sen & Mathew 2009). The species is also reported from northern Myanmar and south-eastern Tibet (Yingjiang County), Yunnan, China.

Since the species is lacking information on its ecology, nothing much can be discussed until more research is conducted. However, on the list for the country, it is the fifth formal species of *Raorchestes,* the others being *R. annandalii*, *R. longchuanensis, R. menglaensis* and *R. shillongensis* (Wangyal et al. 2020). However, with the ever-changing taxonomy due to the influence of molecular studies, the species gets placed in a new genus and specific epithet from time to time.

Amphibians are not the conservation priority of the Kingdom of Bhutan, but the people do not harm animals of any taxon unless it is a real threat. The majority of Buddhist followers do not think it is correct to kill any animal. Thus, the animals (in any group or taxa) are safer in Bhutan than in other countries. Although protected by the societal concept, the problem is the lack of data to conserve and monitor this species. Thus, research is important for this group of animals.

This is the first record of *Raorchestes andersoni* which adds value to the biodiversity of the country. Studies on this species must be initiated to understand its systematic status and ecology, to conserve it.

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# On the distribution of *Myriopholis macrorhyncha* (Jan, 1860) (Squamata, Serpentes, Leptotyphlopidae) from India, with comments on the occurrence of members of Family Leptotyphlopidae from India.

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**ABSTRACT. We confirm the presence of *Myriopholis macrorhyncha* (Jan, 1860) from India, based on reptile surveys conducted in north Gujarat and Saurashtra regions of Gujarat state. We also discuss the historical records of the family Leptotyphlopidae from the country. The study also provides new information on the morphology and microhabitat of the species.**

**KEYWORDS. Thread snakes, species distribution, Scolecophidia, morphology, taxonomy**

## Introduction

Scolecophidians are small, burrowing, worm-like snakes with rudimentary vision. Most species of this group have greatly reduced eyes and less flexible head scalation, are pinkish or brownish in colouration, usually unicoloured or bicoloured tubular-shaped body with subequal, smooth scales, and are frequently mistaken for earthworms by laymen. All scolecophidians actively feed on small social insects (ants and termites) and their larvae (Cundall & Greene 2000). They include the world's smallest snakes and rarely exceed 30 cm in length (Hedges 2008). Although largely neglected in vertebrate research, being basal to other serpents, knowledge on their biogeographical history is crucial for evaluating hypotheses on the origin of serpents (Vidal et al. 2010). The superfamily Typhlopoidea (Scolecophidia) contains some 463 species, which is further divided into five families: Anomalepididae (21 species), Gerrhopilidae (23 species), Typhlopidae (275 species), Xenotyphlopidae (2 species) and Leptotyphlopidae (142 species) (Uetz et al. 2022). However, a recent phylogenetic study proposed a division of Scolecophidians into two superfamilies: Typhlopoidea (blind snakes) and Leptotyphlopoidea (thread snakes). It excluded the family Anomalepididae from Scolecophidia as they were phylogenetically more closely related to 'typical snakes' (Miralles et al. 2018).

The family Leptotyphlopidae Stejneger, 1892, is among the most poorly known groups of all terrestrial vertebrates from the standpoint of systematics and ecology (Adalsteinsson et al. 2009). Members of this group are among the thinnest and smallest snakes, and they are commonly known as 'thread snakes.' Leptotyphlopids have a West Gondwanan distribution, occurring primarily in Asia, Africa and the Neotropics (South America, Middle America, and the West Indies). They occupy a wide variety of habitats and elevations, occurring in deserts (Branch 1998; Broadley & Wallach 2007), forests (Broadley & Wallach 1997), wetlands, savannahs (Broadley & Broadley 1999; Broadley & Wallach 2007), and transformed habitats (Thomas et al. 1985), from below sea level to 3250 meters (Thomas et al. 1985; Zug 1977). Some leptotyphlopids occur on islands that were never connected to mainland areas, hinting at a trans-marine dispersal mode (Adalsteinsson et al. 2009). Adalsteinsson et al. (2009) further classified the family into two sub-families: Epictinae (distribution- new and old world, relatively short-tailed: contains 10 genera, and 89 species) and Leptotyphlopinae (distribution-old world relatively long-tailed; four genera, and 53 species) (Uetz et al. 2022).

*Myriopholis* is a diverse genus with 24 recognized species, distributed throughout Africa (north and south of the Sahara Desert), the Arabian Peninsula and Socotra Island, and southwest Asia (Turkey, Iran, Pakistan, and northwest India) (Adalsteinsson et al. 2009). *Myriopholis macrorhyncha* was originally described by Jan in 1860 as *Stenosoma macrorhynchum* from 'Sennaar' (now in Sudan) (Jan 1860). This is the most widely distributed leptotyphlopid (Adalsteinsson et al. 2009). It is reported from the African to the Arabian Peninsula, Middle East and southwest Asia (Uetz et al. 2022). However, the exact distribution of this species needs a reassessment, since several records of this species have recently been attributed to other species as a result of taxonomic reassessments of certain populations, leading to description of new species as well as revalidation of synonymized taxa (Venchi & Sindaco 2006).

The published literature indicates that *Myriopholis macrorhyncha* was reported from Sind in British India (now in Pakistan) (Boulenger 1890a, 1893; Gharpurey 1937; Smith 1943). Further, Minton (1966), Khan (2006), Baig et al. (2008) and Masroor (2012) added new information on the species and new distribution records from Pakistan. The species was included in the list of Indian snakes by some (Hahn 1978; Das 1994), but not all (Deoras 1965; Whitaker 1978; Murthy 1985; Whitaker & Captain 2004; Aengals et al. 2018). Similarly, the species was included in the list of snake fauna of Rajasthan by some (Patel et al. 2021), but not all (Bhatnagar et al. 2013). Thus, occurrence of this leptotyphlopid in India has been a question of debate. Here, we confirm the occurrence of *Myriopholis macrorhyncha* in India based on new material collected from Gujarat. Further, we discuss the occurrence of other members of family Leptotyphlopidae from India.

## Materials and methods

The present study is based on three specimens collected during our ongoing survey to document the herpetofauna of Gujarat, with permissions issued by the Forest Department of Gujarat state (permit numbers WZP/5585/22/C/590-92/3-8-1990, B/WPS/8/9388-92/2013-14 and WPS/T4/682/B/2931/2016-17) and in accordance with the Indian Wild Life (Protection) Act 1972. The specimens were euthanized with halothane, fixed in 4% formaldehyde, and later transferred to 70% ethanol and deposited in the collections of the Bombay Natural History Society (BNHS), Mumbai (BNHS 3661, BNHS 3662 and BNHS 3663). The pholidosis and morphometric data of all the specimens are given in Table 1. The study also includes two live specimens (field numbers: RVT 01 and RVT 02) that were photographed, examined, and released at the capture site in a few days.

Mensural, meristic and qualitative data were recorded by following Broadley & Wallach (2007). All pholidotic features were examined using a stereo microscope (Omano OM2360-BL). Morphological measurements like snout-vent lengths (SVL), tail lengths (TaL) and total lengths (TL), were measured to the nearest millimetre using a non-elastic string and a ruler. The number of scales around the body were counted at mid-body and the scales around the tail were counted near the cloaca. The total number of dorsal scales between the rostral shield and the terminal spine were counted as the total middorsal scales. Subcaudal counts reported here do not include the terminal scute. Description style follows Broadley & Wallach (2007) with some modifications (Patel et al. 2019a).

*Collection abbreviations*: BNHS, Bombay Natural History Society, Mumbai, India; NHMUK, Natural History Museum, London, United Kingdom; MCZ, Museum of Comparative Zoology, Massachusetts, United States.

## Results

*Taxonomic accounts*

Order: Squamata
Family: Leptotyphlopidae
Genus: *Myriopholis* Hedges, Adalsteinsson & Branch, 2009
*Myriopholis macrorhyncha* (Jan, 1860)
(Figure 1, 2, 3; Table 1)
*Stenosoma macrorhynchum* Jan, 1860
*Glauconia macrorhynchus* Boulenger, 1890a, 1893
*Leptotyphlops macrorhynchus* Smith, 1943
*Myriopholis macrorhyncha* Adalsteinsson et al. 2009; Patel et al. 2021

**Common name.** Long-nosed Worm Snake

*Material examined:* BNHS 3661, unsexed adult, from Zat-Satsan (24.5155°N 72.3613°E; ca. 209 m a.s.l.), Banaskantha district, Gujarat, India, collected by Raju Vyas, Harshil Patel and Vaibhav Naik on 29 September 2021; BNHS 3662 (Fig. 1), unsexed adult, from near Bhimrana (22.3610°N 68.9649°E; ca. 6 m a.s.l.), Devbhoomi Dwarka district, Gujarat, India, collected by Vivek Sharma on 14 October 2016; BNHS 3663, unsexed adult, from Gadhechi (21.7580°N 72.1122°E; ca. 28 m a.s.l.), Bhavanagar city, Bhavanagar district, Gujarat, India, collected by Raju Vyas and Deepak Makawana in August 1982; RVT 01, unsexed adult, from near Bhanvad (21.9499°N 69.7641°E; ca. 48 m a.s.l.), Devbhoomi Dwarka district, Gujarat, India, examined by Raju Vyas on 21 March 2018; RVT 02, unsexed adult, from near Rajula (21.0083°N 71.4903°E; ca. 11 m a.s.l.), Amreli district, Gujarat, India, examined by Raju Vyas in October 2019.

*Morphology and description:* Body long, thin and cylindrical, with head and neck slightly broader than the body, short tail tapers to a small terminal spine. Body covered with smooth, imbricate and subequal scales in 14 rows, which reduce to 10 rows near the cloaca and on the tail; precloacal shield semilunate; total mid-dorsals 374–438 (315–404 fide. Broadley & Wallach 2007); subcaudals 45–55 (26–43 fide. Broadley & Wallach 2007). Snout hooked in lateral view with a distinct beak, rostral wide, much wider than nasals, not reaching level of eyes; nasal divided into a large upper supranasal and a small lower infranasal; upper lip covered by infranasal, anterior supralabial, ocular, and posterior supralabial; anterior supralabial small, as wide as infranasal; ocular large, with moderate rounded black eyespot centrally placed in the upper half; posterior supralabial moderate. Nostril midway between rostral and anterior supralabial along nasal suture. Frontal and supraoculars subequal, postfrontal subpentagonal or semicircular; supraoculars anteriorly wedged between supranasal and ocular, posteriorly wedged between frontal and postfrontal, both smaller than interparietal and interoccipital; parietals transverse, slightly larger than the occipitals, in contact with posterior supralabials; single temporal; mental absent, four infralabials (Figure 3).

**Colouration.** Dorsal ground colour of live individual pinkish; a blotch in a shade of lighter pink on the neck and nape, a similar band is sometimes present in the anterior half of the body; underside light pink to white and forms an uneven zig-zag border with upper body colouration on flanks.

**Distribution.** The present study records this species from Banaskantha district in north Gujarat and Bhavnagar, Devbhoomi Dwarka and Amreli districts in the Saurashtra region (Figure 4).

**Habit, Habitat and Natural History.** All individuals were found during the daytime or late evening, under rocks or decaying logs while conducting surveys in semiarid regions (Figure 5). The animals were found under rocks and logs that were of a moderate size, not deeply embedded in the ground, and which bore cavities beneath, where these snakes sheltered. Soil was made of a mixture of dry clay, sand, and decaying organic material. Other reptiles reported from same habitats were *Echis carinatus* (Schneider, 1801), *Psammophis leithii* Günther 1869, *Eryx johnii* (Russell, 1801), *Acanthodactylus cantoris* Günther 1864, *Ophisops* sp., *Hemidactylus* sp. and *Indotyphlops* sp. The specimen BNHS 3662 was found in saline scrubland, hardly 600 m away from the coastline. All the localities were in scrublands, either in the coastal regions or in arid regions, except for one (BNHS 3663) found in a home garden.

**Table 1.** Scale counts and measurements (mm) of specimens of *Myriopholis macrorhyncha* from Gujarat, India.

| **Specimen** | **BNHS 3661** | **BNHS 3662** | **BNHS 3663** | **RVT 01** | **RVT 02** |
|---|---|---|---|---|---|
| **Location** | Zat-Satsan, Banaskantha | Bhimrana, Devbhoomi Dwarka | Desai Nagar, Gadhechi, Bhavanagar | Bhanvad, Devbhoomi Dwarka | Rajula, Amreli |
| **Habitat** | Scrub forest | Coastal scrubland | Kitchen Garden | Scrubland | Scrubland |
| **TL** | 142 | 140 | 180 | 155 | 160 |
| **SVL** | 130 | 129 | 162 | 138 | - |
| **TaL** | 12 | 11 | 18 | 17 | - |
| **Scales around the body** | 14 | 14 | 14 | 14 | 14 |
| **Scales around the tail** | 10 | 10 | 10 | - | - |
| **Mid-dorsals** | 438 | 413 | 410 | 374 | 421 |
| **Subcaudal** | 55 | - | 53 | 45 | - |

**TL: total lengths, SVL: snout-vent lengths and TaL: tail lengths.*

**Figure 1.** *Myriopholis macrorhyncha* (BNHS 3662) from Devbhoomi Dwarka, Gujarat (A) dorsal view; (B) Head dorso-lateral and (C) Head dorsal (Photos by Vivek Sharma).

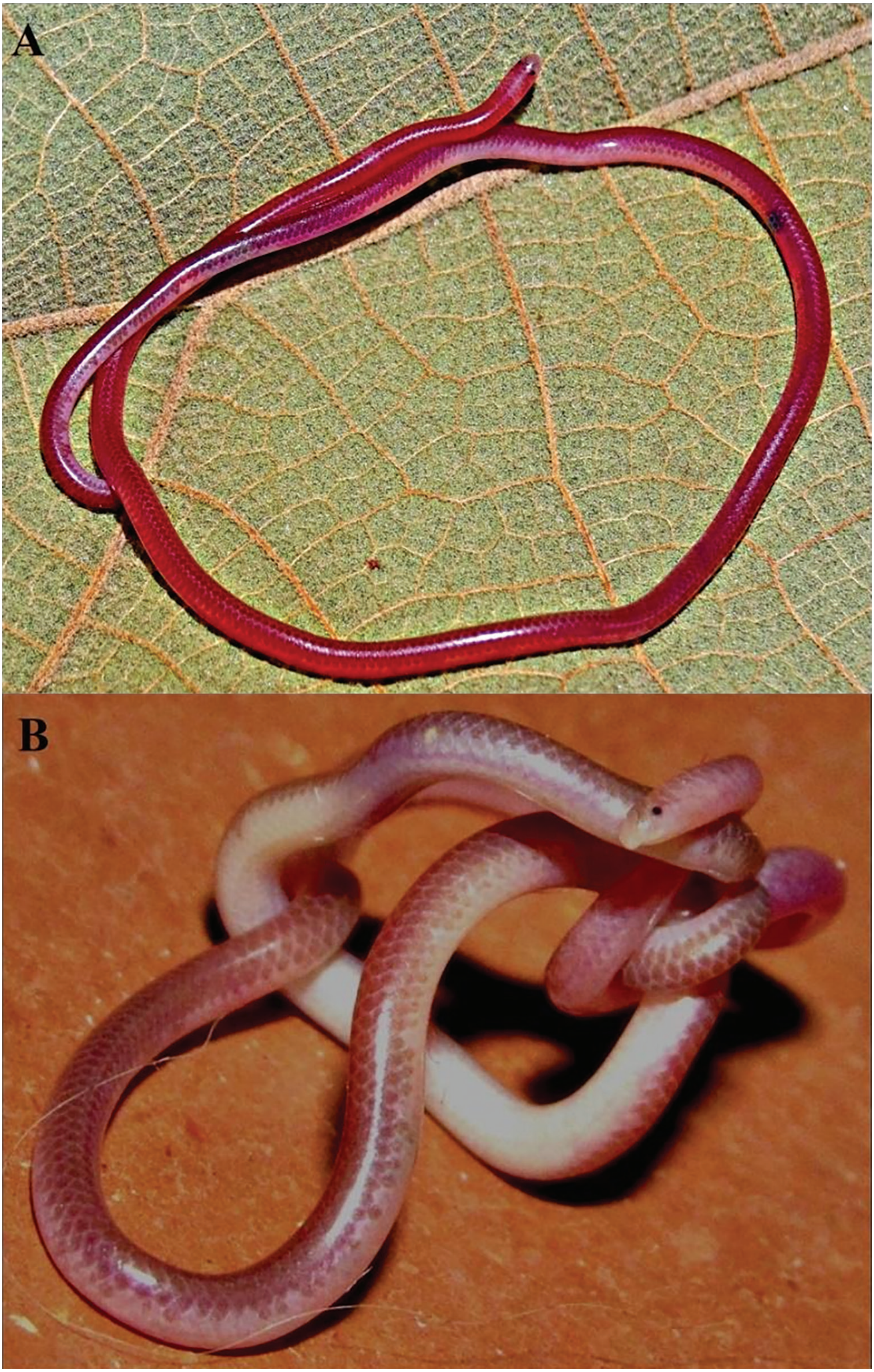

**Figure 2.** Uncollected vouchers of *Myriopholis macrorhyncha* from Gujarat (A) RVT 01 from Bhanvad, Devbhoomi Dwarka, Gujarat; (B) RVT 02 from Rajula, Amreli, Gujarat (Photos by Raju Vyas).

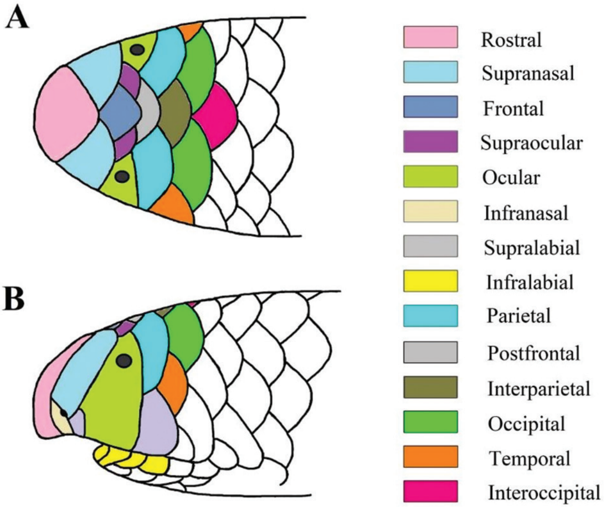

**Figure 3.** Schematic representation of head scalation of *Myriopholis macrorhyncha* (BNHS 3663) (A) Head dorsal and (B) Head lateral.

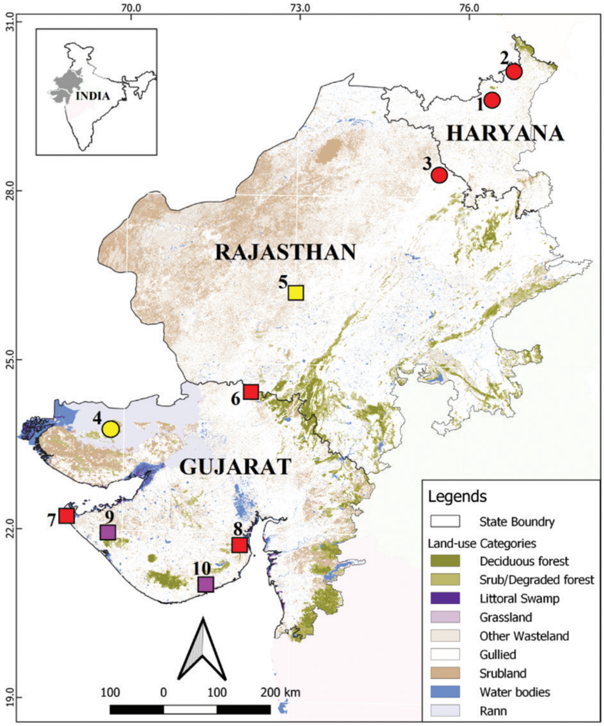

**Figure 4.** Map of Western India showing locations from where *Myriopholis blanfordi* (circles) and *Myriopholis macrorhyncha* (squares) were reported in India (red fill: locations from where museum vouchers were originated; purple fill: locations from where photo vouchers were originated; yellow fill: locations reported in literature without details of voucher specimens, need further verification); see Appendix 1 for details of locations.

## Discussion

Two members of the family Leptotyphlopidae, namely, *Myriopholis blanfordi* (Boulenger, 1890) and *M. macrorhyncha* are reported in literature (both pre-independent and post-independent India) as inhabiting India (Gray 1845; Günther 1869; Boulenger 1890a, 1890b, 1893; Smith 1943; Sharma 2007).

During the pre-independence period in India, *M. blanfordi* was described from Sind (now in Pakistan) and subsequently reported from Multan (now in Punjab, Pakistan) and from Ambala (now in Haryana, India); whereas *M. macrorhyncha* was recorded from Karachi (now in Sind, Pakistan) (Boulenger 1890a; Smith 1943). Later, both the species were included in the list of serpents of India or arid regions of the country (Hahn 1980; Das 1994; Patel et al. 2021). Later, *M. blanfordi* was reported from a few localities in Rajasthan (Sundersingh 1960; Murthy et al. 1993) and one location (Kavada, Kutch) in Gujarat (Sharma 2007); and *M. macrorhyncha* was reported from Jodhpur, Rajasthan (Sharma 2007). However, no voucher specimens or morphological details for the post-independence reports were provided. Records of *M. blanfordi* from India is accepted by few researchers (Whitaker & Captain 2004; Aengals et al. 2018). Thus, the present communication is noteworthy, given that this is the first authentic report of *M. macrorhyncha* from India.

The present study shows that *M. macrorhyncha* is widely distributed in Gujarat State, and could also be occurring in the western Indian state of Rajasthan (Fig. 4). However, surveys thus far had not reported this species from Gujarat (Acharya 1949; Kapadia 1951; Sharma 1982, 2000; Vyas, 1993, 1998, 2000, 2007; Gayen 1999; Patel & Vyas 2019). With these new records of *M. macrorhyncha*, which is the first authentic record of Family Leptotyphlopidae from the state, the total number of snake species in Gujarat is now 58 (Pa-

**Figure 5.** Habitat of *Myriopholis macrorhyncha* (A) at Zat-Satsan, Banaskantha, Gujarat and (B) near Bhimrana, Denbhoomi Dwarka, Gujarat [Photos by Raju Vyas (A) and Harshil Patel (B)].

tel & Vyas 2019; Patel et al. 2019a & 2019b). The sole record of *M. blanfordi* from Khavada (in Kutch), Gujarat by Sharma (2007) needs further confirmation (see Patel & Vyas 2019). Records of members of the family Leptotyphlopidae from India are presented in Appendix 1 and Figure 4. In addition to these reports, we received images of three thread snakes from the

Saurashtra region (one each from Bhavnagar, Beyt Dwarka and Veraval). The images were sent by naturalists and snake rescuers, and appear to be the members of the genus *Myriopholis*. However, lack of specimens hindered species identification.

*M. macrorhyncha*, being a wide-ranging species spanning from African to Arabian Peninsula, Middle East and southwest Asia (Uetz et al. 2022), could be a species complex comprising multiple lineages. The present records are the eastern most population of the species. The eastern population of *M. macrorhyncha* was described as *Glauconia hamulirostris* Nikolsky, 1907, with its type locality in Iran, which is usually treated as a subjective synonym by most researchers (Smith 1943; Broadley & Wallach 2007; Adalsteinsson et al. 2009; Sharma 2007). However, Hahn (1980) and Wallach et al. (2014) considered it as a valid taxon. Molecular and morphological studies are required to clarify the status of *Glauconia hamulirostris*. Until then, it should be treated as a synonym of *M. macrorhyncha*. A thorough revision and reassessment involving detailed morphological comparisons, aided with molecular data from samples throughout the range of *M. macrorhyncha*, is needed to resolve this species complex.

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**Appendix 1.** Records of members of family Leptotyphlopidae from India, in correspondence with Figure 4.

| | Species | Name of Locality | Geo-coordinates | Voucher No. | Source |
|---|---|---|---|---|---|
| 1 | *Myriopholis blanfordi* | from the Plains 70 miles SW of Ambala[1] (in Haryana, India) | 29.6278°N; 76.1392°E | MCZ:Herp:R-3749 | Barbour 1908 |
| 2 | *Myriopholis blanfordi* | Umballah N India[2] (Ambala, Haryana, India) | 30.3609°N; 76.7978°E | MCZ:Herp:R-3217 | Barbour 1908 |
| 3 | *Myriopholis blanfordi* | Umballah N India[2] (Ambala, Haryana, India) | 30.3609°N; 76.7978°E | MCZ:Herp:R-3218 | Barbour 1908 |
| 4 | *Myriopholis blanfordi* | Umballah N India[2] (Ambala, Haryana, India) | 30.3609°N; 76.7978°E | MCZ:Herp:R-175103 | Barbour 1908 |
| 5 | *Myriopholis blanfordi* | Pilani, Rajasthan[3] | 28.3813°N; 75.6139°E | NHMUK:1957.1.9.7 | |
| 6 | *Myriopholis blanfordi* | Khavada, Kutch, Gujarat[4] | 23.8439°N; 69.7340°E | - | Sharma 2007 |
| 7 | *Myriopholis macrorhyncha* | Jodhpur, Rajasthan[5] | 26.2515°N; 72.9934°E | - | Sharma 2007 |
| 8 | *Myriopholis macrorhyncha* | Zat-Satsan, Banaskantha, Gujarat[6] | 24.5155°N; 72.3613°E | BNHS 3661 | Present Study |
| 9 | *Myriopholis macrorhyncha* | Bhimrana, Devbhoomi Dwarka, Gujarat[7] | 22.3610°N; 68.9649°E | BNHS 3662 | Present Study |
| 10 | *Myriopholis macrorhyncha* | Ghadechi, Bhavnagar, Gujarat[8] | 21.7580°N; 72.1122°E | BNHS 3663 | Present Study |
| 11 | *Myriopholis macrorhyncha* | Near Bhanvad, Devbhoomi Dwarka , Gujarat[9] | 21.9499°N; 69.7641°E | RVT 01 | Present Study |
| 12 | *Myriopholis macrorhyncha* | Near Rajula, Amreli, Gujarat[10] | 21.0083°N; 71.4903°E | RVT 02 | Present Study |

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# Uchang-sa: A report on frog consumption around Dampa Tiger Reserve, Mizoram, northeast India

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**ABSTRACT. The global amphibian population decline has been linked to various factors, among which overexploitation as a food source is one. Amphibians have been a part of the local delicacy in many parts of northeast India, including Mizoram. Using a questionnaire survey involving 260 households, we documented 11 species of anurans, belonging to four families, which were consumed locally at the periphery of Dampa Tiger Reserve. We also present our result in the context of seasonality of collection and livelihood among respondents, and discuss future conservation issues.**

**KEYWORDS. Amphibians, collection, food, Dampa Tiger Reserve, Mizoram**

## Introduction

Various wild animals, both from terrestrial and aquatic systems, form the primary source of meat for hundreds of indigenous, rural and other marginalized communities across the globe (Asprilla-Perea & Díaz-Puente, 2019; Brashares et al. 2011). This consumption, valued at several billion dollars annually, forms a key component of local food security (Nielsen et al. 2018). Among the various wild taxa consumed, amphibians have been identified as a key source of protein (Valencia-Aguilar et al. 2013). The Global Amphibian Assessment (GAA) shows that about 220 species of amphibians are used as food across the globe (Stuart et al. 2008), with species assessed potential functional food sources for humans (Oliviera et al. 2017). Many species of amphibians are facing extinction today, and human exploitation, which includes harvesting frogs for food, has been identified as one of the critical reasons for the decline in frog populations worldwide (Whittaker et al. 2013; Grano 2020).

India is not only a biodiversity rich country with a rapidly developing economy, but it is also home to thousands of indigenous and tribal communities. Closeness to nature and lack of livelihood opportunities have resulted in a way of life that is heavily dependent on natural resources for many of these communities. Northeast India is rich in biodiversity and is home to around 145 tribal communities; most practise shifting cultivation and are dependent on natural resources for sustenance. Wild meat is considered a delicacy across the region and many taxa are also used in traditional medicines (Aiyadurai et al. 2010; Jugli et al. 2010; Verma et al. 2014; Borah & Prasad 2017; Chanu 2017; Dhakal et al. 2019; Talukdar et al. 2020). Due to the remoteness, rugged terrain, and lack of opportunities, many parts of the region are understudied when it comes to the socio-ecological nexus in the region.

Mizoram is the southernmost state of northeast India, and is part of the Indo-Burma biodiversity hotspot (Mittermeier et al. 2004);

however, its unique biodiversity is understudied (Pawar & Birand 2001). Though the consumption of frog meat is prevalent among the tribes of northeast India, little published literature is available in this regard (Talukdar & Sengupta 2020; Talukdar et al. 2020). Mizoram state has ca. 70 species of amphibians, with representatives from eight families under two orders (Chanda 2007; Lalremsanga et al. 2021 a,b; Sengupta et al. 2010; Lalronunga et al. 2020, 2021; Decemson et al. 2021; Lalmuansanga et al. 2020; Malsawmhriatzuali et al. 2020; Kundu et al. 2020). However, the faunal discovery is far from being exhausted for this area (Pawar & Birand 2001).

A preliminary market survey and anecdotal observations show that consumption of frogs is still rampant in the region, and forms a part of the local cuisine. Knowing which species are preferred and more prone to offtake will enable us to establish conservation priorities. Hence, this study is aimed at identifying the preferred species of frogs by the local communities within the study area.

## Methods

**Study area.** Dampa Tiger Reserve (DTR) is situated in the western part of Mizoram state, bordering Bangladesh in the west (Figure 1). With an area of 500 km$^2$, it is the largest Protected Area of the state. The fringe villages of DTR are inhabited by the Mizo, Bru and Chakma tribes. The study was conducted in four fringe villages, *viz*. Teirei, Damparengpui, Rajivnagar and Lallen of DTR (Figure 1), from January 2021 to April 2021.

**Data collection and analysis.** A semi-structured questionnaire, informal interviews and interactions with local communities were used to gather information relating to the consumption of anurans (see Appendix 1). At least 10% of the population was surveyed randomly in each study village, with each respondent representing one household, and a proportionate representation of gender was attempted (Choudhury et al. 2019). The respondents were asked about the local names of harvested frogs, and time and places of collection. The identity of the frog species was also confirmed by showing the respondent photographs of species recorded in the area from published literature (Ahmed et al. 2009; Lalronunga et al. 2020a; Decemson et al. 2021) and field surveys.

## Results

During the study period, we sampled 40 households in Lallen Village inhabited by the Mizo tribe, 40 households in Teirei village inhabited by the Mizo and Bru tribes, 80 households in Damparengpui village inhabited by the Bru and Mizo tribes, and 100 households in Rajivnagar village inhabited by the Chakma tribe. Our study showed that 72.7% of total respondents consumed frogs. About 55% of respondents from Teirei, 81.3% from Damparengpui, 76.0% from Rajivnagar and 65% from Lallen consumed frogs. The occupation-wise frog consumption among the respondents (or sampled household) is shown in figure 2. A chi-squared test indicated that frog consumption was significantly higher among daily-wage labourers compared to other occupations ($p = 0.0015$).

A total of 11 species belonging to four families were consumed in the study area, as shown in table 2 and figure 4. Frog harvesting was done primarily during the rainy season. This is also the time frogs are most vocal and can be located easily because of their activities.

## Discussion

Out of the 11 reported species of frogs, nine species are found to be consumed by all three tribes; however, *Duttaphrynus melanostictus* was consumed only by the Bru and Chakma tribes, after removing the skin. The harvesting season also coincides with the breeding season of the species in Mizoram, indicating that there is a dedicated effort towards collection during the peak activity period by following their acoustic trail. In our subsequent surveys in the study area, we noted that locally abundant species such as *Ingerana borealis*, *Raorchestes* sp., etc., are not eaten. Few species such as *Leptobrachium* sp and *Theloderma* sp. Are not consumed, even though they are present in the region, due to their apparent rarity. The frog species harvested for food were comparatively large-sized species, attaining a size class > 50 mm SVL. Such size preference could be due to the ease in locating and catching these species. The habitat of preferred species varies from streams to pools and trees, so the observation indicates that the

**Table 1.** Anuran species harvested in the study area, with their local name and seasonality of collection.

| Species | Local Name | Time of collection | Previous records | IUCN status |
|---|---|---|---|---|
| ***Duttaphrynus melanostictus*** | Utawk | March to November | Decemson et al., 2021 | Least concern |
| ***Hoplobatrachus tigerinus*** | Usai/Ukei/Usial | March to September | Decemson et al., 2021 | Least concern |
| ***Hoplobatrachus litoralis*** | Usai/Ukei/Usial | March to september | Decemson et al., 2021;<br>Kundu et al., 2021 | Data deficient |
| ***Euphlyctis kalasgramensis*** | Chungu | March to August | Decemson et al., 2021 | Least concern |
| ***Amolops indoburmanensis*** | Usial/Lengkir uchang | February to August | Decemson et al., 2021;<br>Lalronunga et al., 2020a | Not Assessed |
| ***Pterorana khare*** | Usial thla nei/Uthlawk/ Uhlap | September to December | Decemson et al., 2021 | Vulnerable |
| ***Odorrana chloronota*** | Uhring | February to August | Decemson et al., 2021 | Least concern |
| ***Hydrophylax leptoglossa*** | Changtuk | March to August | Decemson et al., 2021 | Least concern |
| ***Sylvirana lacrima*** | Utereng | September to January | Decemson et al., 2021;<br>Lalronunga et al.,2021 a. | Least concern |
| ***Polypedates teraiensis*** | Changban | March to August | Decemson et al, 2021 | Not assessed |
| ***Zhangixalus smaragdinus*** | Uchhawlhring | March to August | Decemson et al, 2021 | Not assessed |

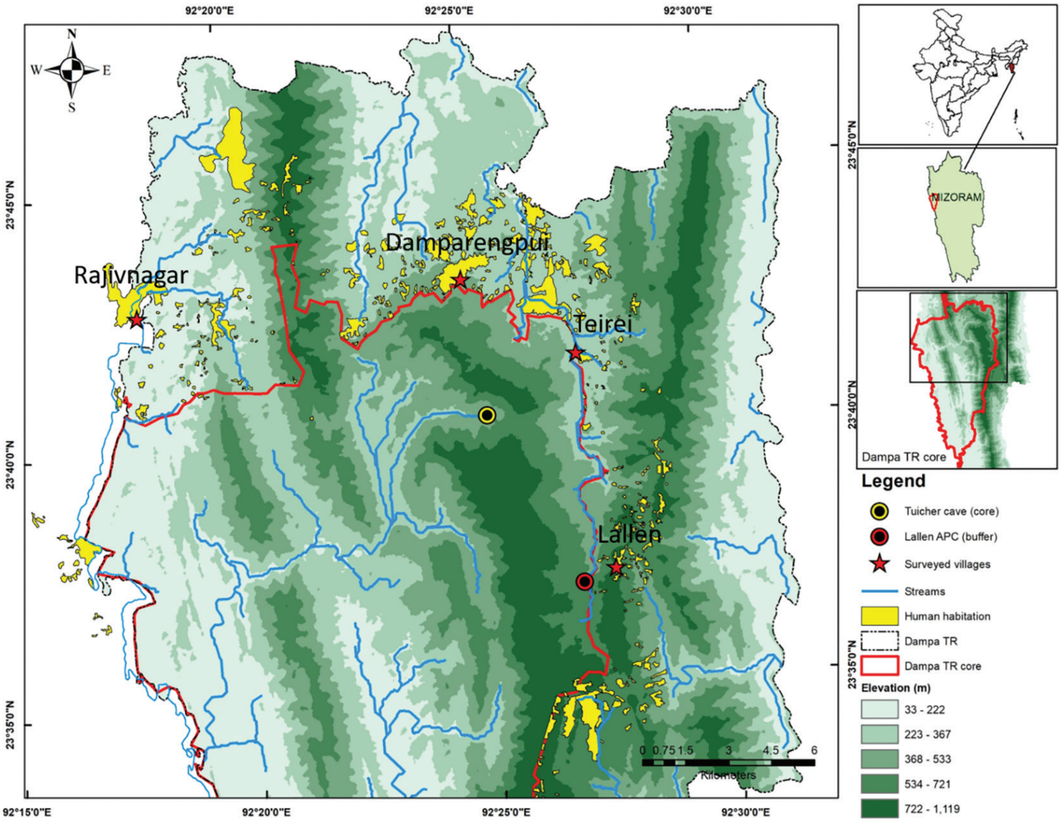

**Figure 1.** Study area showing the location of the study sites with respect to Dampa Tiger reserve.

collection of frogs for consumption may not be habitat specific.

Our results also indicated that the majority of households that collected frogs fell in either of the two livelihood categories *viz.,* daily-wage workers or individuals working in the agriculture sector. Both these occupations are primarily season dependent and could entail more free days, compared to those working in government (regular) jobs or those involved in businesses. These livelihood strategies also give irregular income, and villagers engaged in these might look for free sources of food. Among the Chakma tribes in Rajivnagar village, a small portion of the sampled population (3%) stated that *Duttaphrynus melanostictus* is also used for the treatment of cold and cough. They further added that eating frog meat reduces chances of getting diseases.

The study flags an important conservation issue that frog consumption not only threatens the local anuran biodiversity, but could also pose zoonotic risks for local communities (Funk et al. 2021; Grano 2020). Persistent collection of larger body sized breeding populations might be detrimental for the local populations of various frog species. A detailed study needs to be conducted to understand the population level impacts of such harvesting practices. Except for *Pterorana khare*, the species consumed as a delicacy by the local communities are either Data Deficient or Least Concern as per the IUCN status. Thus, the amount of impact caused in the ecosystem due to the off-take of frogs is yet to be assessed. This would provide a way forward to developing sustainable alternatives for select aquatic fauna that are a crucial part of local culinary practices.

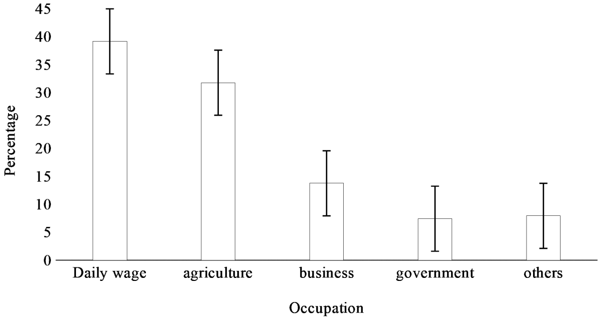

**Figure 2.** Occupation of respondents who harvest frogs for consumption in the study area.

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We thank the Chief Wildlife Warden, Department of Environment, Forest

**Figure 3.** Frogs and snails collected by hunters.

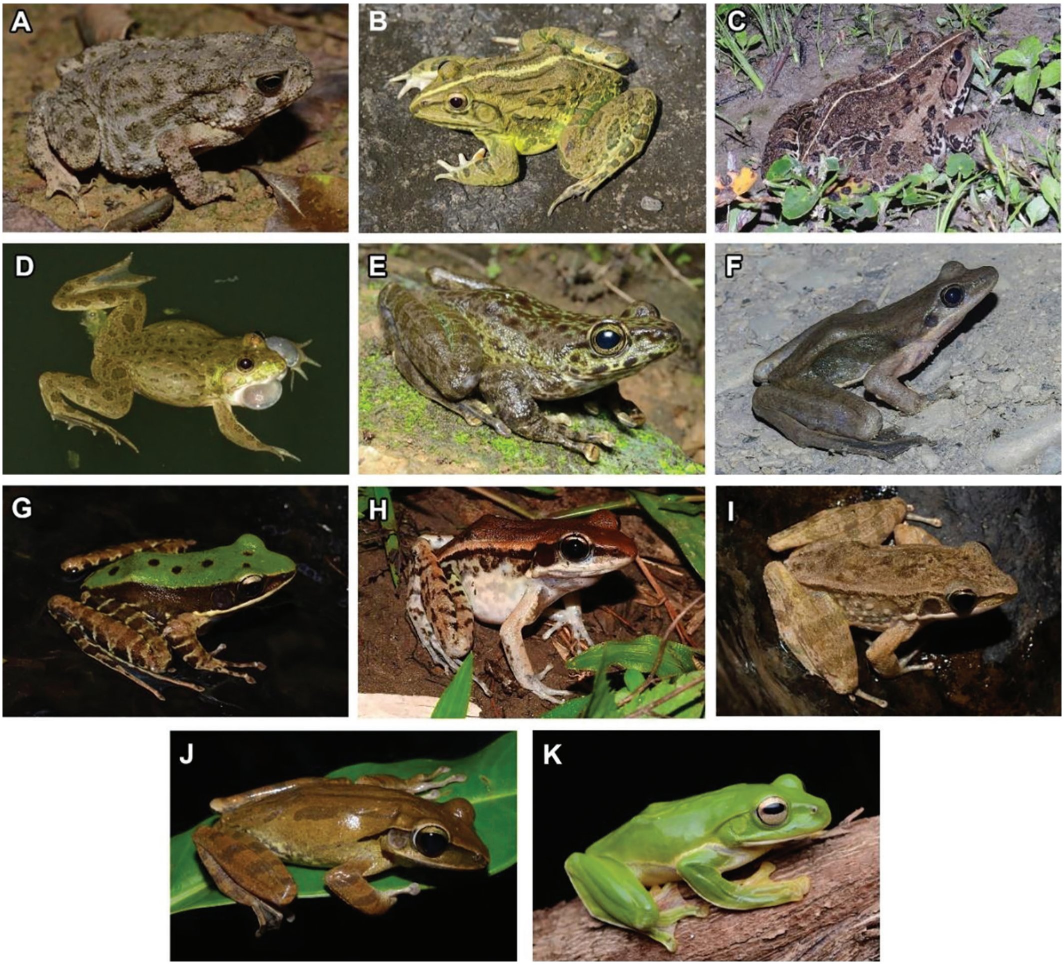

**Figure 4.** Frog species harvested for consumption in the study area. A) *Duttaphrynus melanocstictus* B) *Hoplobatrachus litoralis* C) *Hoplobatrachus litoralis* D) *Euphlyctis kalasgramensis* E) *Amolops indoburmanensis* F) *Pterorana khare* G) *Odorrana chloronota* H) *Hydrophylax leptoglossa* I) *Sylvirana lacrima* J) *Polypedates teraiensis* K) *Zhangixalus smaragdinus*

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*Available online 22/07/2022*

**Appendix 1.** Sample of questions asked during interviews.

| | |
|---|---|
| Basic demography of respondentsÂ | Age |
| | Gender |
| | Educational qualification |
| | Family members |
| Frog collection | Yes or No? |
| | Time of collection? |
| | Species (in local names, confirmed with photographs from published literatures) |
| | Need for conservation? |
| | Medicinal values? |

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# Mortality rates of snakes on the roads of Vazhachal Forest Division, Kerala, Southern-Western Ghats, India.

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**CITATION.** Yadav, P.B.S., Prakash, L. and Karthik, P. (2022). Mortality rates of snakes on the roads of Vazhachal Forest Division, Kerala, Southern-Western Ghats (SWG), India. *Hamadryad : 39, 37-42*.

**ABSTRACT. Vehicular mortalities have a significant impact on wildlife, and pose a substantial threat to biodiversity. Several national highways (NH) bisect important reserve forest patches in India, resulting in severe habitat fragmentation. This study records snake mortalities between from June 2019 to November 2019, on State Highway 21, covering a ca.80km stretch adjoining the Vazhachal forest division. A total of 18 species belonging to five families and 18 genera were recorded. The families are Uropeltidae with one genus (1 sp.), Pythonidae with one genus (1 sp.), Colubridae with 11 genera (11 spp.), Elapidae with 3 genera (3 spp.) and Viperidae with 2 genera (2 spp.). Of these, three are endemic to Western Ghats, one species is near threatened, one species is vulnerable, and one species is listed as endangered. This region is a potential habitat to endemic species, and also home to many faunal species. Hence, a lower speed limit would be advisable to protect wildlife and mitigate future threats.**

**KEYWORDS. snake, highway, mortality, conservation, protected forest**

## Introduction

Road networks and vehicular traffic play a crucial role in wildlife mortality (Plante et al. 2019). Often, the highways pass through the protected forest zones which affect the native fauna's movement and survival (Shepard et al. 2008). Subsequently, the studies from across the globe have been highlighting the adverse impacts of the road networks on wild animals, and the consequent mortality across invertebrates and higher vertebrates (Fahriget al. 1995; Clarke et al. 1998; Mackenzie et al. 2001; Erritzoe et al. 2003).

The Western Ghats harbours about 30% of India's biodiversity, and is one of the 34 biodiversity hotspots of the world (Rodgers & Panwar 1988, Mittermeier et al. 2005). A total of 193 species of reptiles have been documented from the Western Ghats (Srinivasulu et al. 2014). In India, there is a dearth of information on vehicular mortalities of wildlife, and its ecological implications (Sundar 2004). Hence, small-scale investigations have been carried out on the road mortality of organisms ranging from soil invertebrates to amphibians, reptiles, birds, and mammals (Gokula, 1997; Vijayakumar et al. 2001; Boominathan et al. 2008; Bhupathy et al. 2011; Santhoshkumar et al. 2017; Prakash & Karthik 2020). Therefore, the present study attempts to record snake mortality on State highway 21, adjoining the Vazhachal forest division.

## Materials and Methods

**Study area.** The sampling was performed on State Highway 21 (SH 21), covering an 80 km stretch, which passes through Vazhachal forest division (Fig. 1). The Vazhachal forest division spans 414 km$^2$ and includes five forest ranges (Athirapally, Charpa, Kollathirumedu, Sholayar, and Vazhachal), all of which are located on either side of SH 21. The temperature ranges from ca. 23 °C to 31 °C, and the elevation varied from 200 m to 1300 m asl. The heaviest rainfall typically occurs in the months of June and July, and the habitat type is mostly tropical wet evergreen forests, tropical semi-evergreen forests, and tropical moist deciduous forests. The forest division and either side of highway road is rich in floral and faunal diversity (Bachan, 2010)

**Methods.** The road mortality survey sampling was performed between from June 2019 to November 2019, on the road stretch of SH 21 (Fig. 1). The study followed the fortnight sampling method in Boominathan et al. (2008), and the vehicle driving method followed Das et al (2007). A total number of sampling days were 48(N = 48), and each survey covered aca. 50 km stretch. Thus, the sampling was carried out over a ca. 2400 km, and was a two-man effort. Furthermore, the survey was carried out through reserve forests (Athirapally to Malakkapara) and human settlements (Chalakudy to Athirapally). The sampling was restricted to snake-mortalities, and observations were made during mornings (0700–1000 h) and late evenings (1600–1800 h). While sampling, the road carcasses were examined and identified to the species level using available literature (Daniel 2002). Also, the dead carcasses were removed from the road to avoid multiple counts, and photographs were taken for documentation. Apart from that, the survey collected the following variables: family name, species name, observed individuals, venomous or non-venomous, conservation status (IUCN & endemic). The location of each road kill was marked with a GPS and mapped (Fig. 1).The study did not involve collecting any samples for preservation or lab purposes.

**Table 1.** The road-kill observation on the Chalakudy to Valparai State Highway (SH 21) through the Vazhachal Forest Division.

| S. No. | Family | Species Name | # Indi. | Venomous /Non-Venomous | IUCN status |
|---|---|---|---|---|---|
| 1 | Uropeltidae | _Rhinophistravancoricus*_ | 1 | N-V | EN |
| 2 | Phythonidae | _Python molurus_ | 3 | N-V | NT |
| 3 | Colubridae | _Ahaetullaisabellina_ | 2 | M-V | LC |
| 4 |  | _Rhabdophisplumbicolor_ | 4 | V | NA |
| 5 |  | _Amphiesmastolatum_ | 2 | N-V | NA |
| 6 |  | _Oligodonaffinis*_ | 1 | N-V | LC |
| 7 |  | _Ptyas mucosa_ | 2 | N-V | NA |
| 8 |  | _Boiga trigonata_ | 1 | M-V | NA |
| 9 |  | _Coelognathushelenamonticollaris_ | 1 | N-V | NA |
| 10 |  | _Dendrelaphis tristis_ | 5 | N-V | NA |
| 11 |  | _Fowlea piscator_ | 3 | N-V | NA |
| 12 |  | _Hebiusbeddomei*_ | 1 | N-V | LC |
| 13 |  | _Lycodonaulicus_ | 1 | N-V | LC |
| 14 | Elapidae | _Najanaja_ | 2 | V | LC |
| 15 |  | _Bungarus caeruleus_ | 3 | V | NA |
| 16 |  | _Ophiophagus hannah_ | 3 | V | VU |
| 17 | Viperidae | _Hypnalehypnale_ | 1 | V | LC |
| 18 |  | _Daboia russelii_ | 3 | V | NA |

_# N-V denotes non-venomous, V denotes venomous, M-V denotes mildly venomous, * indicates the species endemic to the Western Ghats._

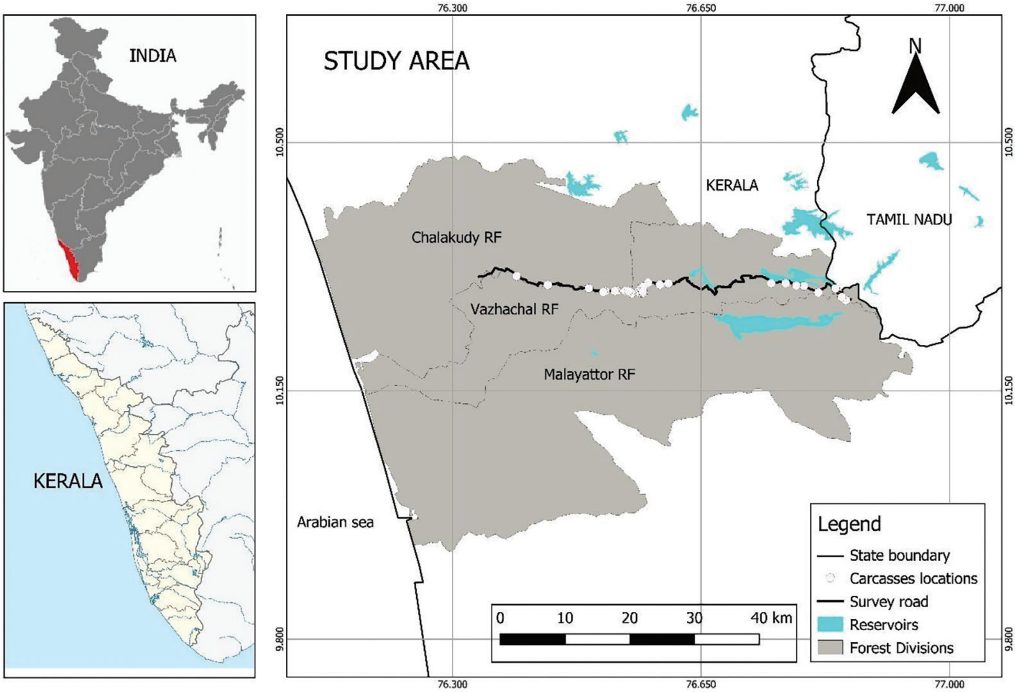

**Figure 1.** Map showing surveyed highway SH 21 road lane with carcass sighting locations.

## Results

Over 48 sampling days, and approximately 2400 km, the study recorded road-kills of 18 species of snakes belonging to 18 genera and five families (Table 1). A total of 39 freshly killed individuals were observed during the sampling hours, of which a few are locally endemic. Of these, six (33.33%) species are venomous; two (11.11%) species are mildly venomous, and ten (55.55%) species are non-venomous (Table 1). However, in comparison to other family members, the family Colubridae is relatively abundant (11 spp. and 23 individuals), followed by families Elapidae (3 spp. and 8 individuals), Viperidae (2 spp. and 4 individuals), Pythonidae (1 sp. and 3 individuals) and Uropeltidae (1 sp. and 1 individual). However, the species *Dendrelaphis tristis* and *Rhabdophis plumbicolor* had the highest carcass rate among the 39 individuals. Furthermore, the snake species *Hebius beddomei, Oligodon affinis* and *Rhinophis travancoricus* are endemic to the Western Ghats. Based on the IUCN Red-list categories (2021), one endangered (*Rhinophis travancoricus*), one near threatened (*Python molurus*) and one vulnerable species (*Ophiophagus hannah*) were also recorded. The species *Rhinophis travancoricus* and *Oligodon affinis* are highly elusive and rare (Srinivasuluet al. 2014). Also, the species *Ophiophagus hannah*islisted in schedule II category (acc. WPA 1972) and appendix II (acc. CITES). Therefore, negligence in enforcing conservation measures is also impacting conservation priority species such as *Ophiophagus hannah*.

## Discussion

Vijayakumar et al. (2001) have observed a total of 49 individuals during a road kill survey, in which most of carcasses fall in the family Colubridae (n = 22). The present study observations corroborate the observations made by Vijayakumar et al. (2001). Snakes play an intrinsic role in nature, particularly in food webs and other ecological aspects (Andrews & Gibbons 2005). This study highlights a few critical factors that threaten animals: (a) people's ignorance about lower vertebrates, (b) feeding wild animals, and (c) food garbage thrown in roadside bushes (Spellerberg 2002). Tourists throw food trash on the roadside, attracting rodents and other small animals. Snakes, while chasing these prey end

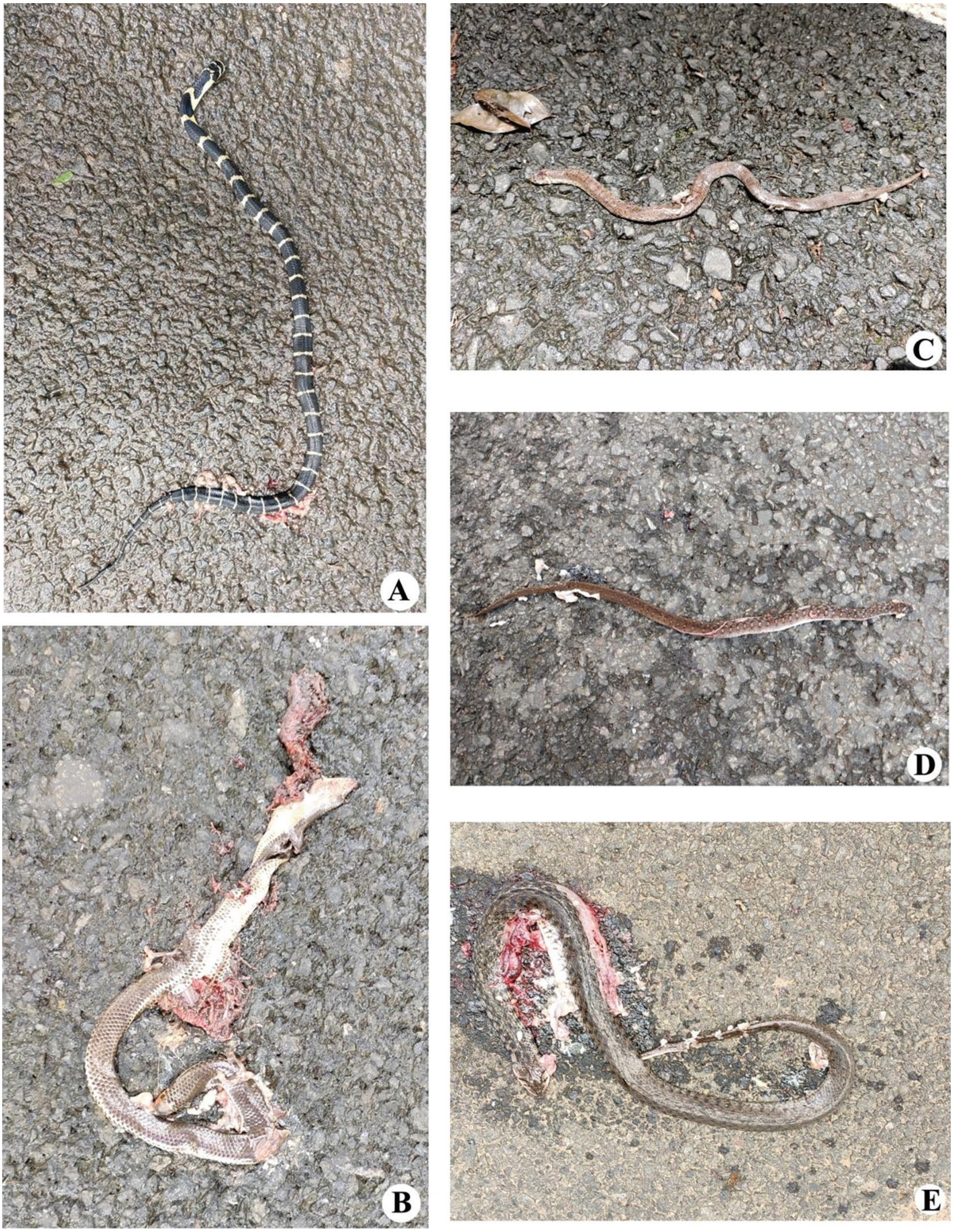

**Figure 2.** A) *Ophiophagus hannah*, B) *Rhinophis travancoricus*, C) *Oligodon affinis*, D) *Hebius beddomei*, E) *Fowlea piscator*.

up as road-kill (Secco et al. 2014). Snakes are also known to be poikilothermic. Therefore, to regulate their body temperatures, the snakes tend to move towards tar roads for heat, and end up getting run over.

## Conclusion

Over one hundred million snakes have been killed in the United States by vehicles, and this is attributed to snakes resting or coiling on the roads for heat (Rosen & Lowe 1994). In India, there are numerous road lanes running through protected zones, which cause a significant effect on wildlife fauna (Baskaran and Boominathan 2010). Emphasising the importance of conservation of lower vertebrates, here is a small attempt to quantify snake mortality on roads. To mitigate the impact of vehicle mortality on wild animals, these road lanes should be monitored prudently, along with strict guidelines to protect wild animals (Karthik et al. 2018). The results indicate that such stretches need strict guidelines on vehicle movements, with vehicle speeds on the highways that passes through the forests being limited to 30 km per hour. Also, speed mounds can be constructed at regular intervals to ensure that speed limits are adhered to (Glista et al. 2009; Seshadri& Ganesh 2011), and signboards indicating wildlife crossing zones can be installed (Samson et al. 2016; Prakash & Karthik 2021).

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# Reproductive behaviour of *Polypedates taeniatus* (Boulenger 1906) (Anura: Rhacophoridae)

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**ABSTRACT. We studied the breeding and nesting behaviour of the endemic terai tree frog, *Polypedates taeniatus* in Uttar Pradesh state of India. We describe amplexus, spawning, unique ground nesting behaviour, and larval developmental stages of the species. Comparative reproductive behaviour of foam-nesting rhacophorids from India is also provided. The present study emphasises the conservation of this obligate grassland species in the Terai Arc Landscape.**

**KEYWORDS. Rhacophoridae, breeding behaviour, grassland, endemic.**

## Introduction

Over 40 different reproductive modes are known among the anurans (Gururaja et al. 2014; Seshadri et al. 2014; Meegaskumbura et al. 2015). The family Rhacophoridae exhibits the most diverse reproductive modes in both aquatic and terrestrial habitats (Bahir et al. 2005; Iskandar et al. 2014; Seshadri et al. 2014; Meegaskumbura et al. 2015; Biju et al. 2016; Khongwir et al. 2016). Terrestrial breeding rhacophorids deposit their eggs in a gelatinous matrix (*Beddomixalus, Feihyla, Gracixalus, Kurixalus, Mercurana, Nasutixalus, Nyctixalus* and *Theloderma*), a foam nest above or near water bodies (*Polypedates*, *Rhacophorus, Chiromantis* and *Chirixalus*), or in a bubble nest (*Rohanixalus*) (Meegaskumbura et al. 2015; Biju et al. 2016; Biju et al. 2020). Other terrestrial breeders (*Philautus*, *Pseudophilautus* and *Raorchestes*) undergo direct development with lecithotrophic larvae except for *Philautus macroscelis* (Hertwig et al. 2012, Meegaskumbura et al. 2015; Biju et al. 2016). Foam nesting rhacophorids construct their nests on a variety of substrates such as under vegetation, on the ground, or on branches or leaves hanging over standing water where free swimming and ectotrophic tadpoles undergo development (Biswas 2000; Deuti 2001; Biju 2009; Hooroo et al. 2017).

Foam nesting rhacophorids of the Himalayan terai region are represented by six species, viz, *Polypedates teraiensis*, *P. maculatus*, *P. taeniatus*, *P. zed, Chirixalus dudhwaensis*, and *Chirixalus simus*. The terai tree frog, *Polypedates taeniatus* was described from Purneah, Bihar, India (Boulenger 1906) and is currently restricted to the grasslands along the Ganga and Brahmaputra rivers in India and Nepal (Fig. 1). Despite several reports of the species from Nepal and India (Ray, 1991; Anders et al. 1998; Ahmed and Dutta 2000; Schleich and Kästle 2002; Hedge et al. 2009; Das et al. 2012; Deuti et al. 2018) very little is known about the ecology and reproductive biology of *Polypedates taeniatus*.

## Material and Methods

**Study area and data collection.** During August 2018, we observed the breeding behaviour of *P. taeniatus* along the River Ganga at Bidurkuti (29.28301º N and 78.10165º E, elevation 208 m a.s.l.) of Bijnor district, Uttar Pradesh. The area is characterized by a large patch of *Typha angustifolia* with temporary water pools created by rain and river flooding. The vegetation of the area is fragmented by a few roads connecting fringe villages.

We observed the breeding activity from 17 August to 22 August 2018 between 18:00 h and 01:00 h. We actively searched for calling males, a pair in amplexus or a female until it engaged in amplexus. All observations were made with red light or a dim torch from about 1 m distance. The duration of amplexus and foam nesting was recorded with a stopwatch. The perch height (distance from the ground to the height at which individuals were roosting or calling) of the individuals and the distance of the nests from the nearest waterbody was measured using a 5 m tape. After spawning, the body size (snout-vent length, SVL) of the amplectant pair and size (length, width and height) of the nest were taken in the natural habitat using a digital calliper (Mitutoyo®) to the nearest 0.1 mm. In addition to nocturnal surveys, we opportunistically searched for foam nests by thoroughly inspecting the ground under the vegetation during the day. We collected nests (n = 2) to determine the clutch size and egg size. Egg extraction from the foam nest follows Coe (1974). Collected nests were placed on the water surface in a glass jar; carefully shaking the nest until all the eggs separated from the foam and settled down at the bottom of the jar. After the complete dispersal of the foam, the fluid and water were poured out and the eggs were counted. The diameter of the eggs was taken within 24 h of foam nest construction using a digital calliper. We photographed and video-recorded the behavioural activities in the field and analysed them in the lab for additional data. We also collected tadpoles from temporary water puddles of the same habitat to study the developmental stages. Collected tadpoles were reared in a plastic tray (50 cm × 30 cm × 8 cm) in the laboratory. We provided rainwater with algae and debris from similar habitats (Chakravarty et al. 2011; Lalremsanga 2017). Tadpoles were preserved at different stages. Developmental stages were identified using Gosner (1960). Collected adults, eggs and tadpoles of *P. taeniatus* were fixed in 4% formalin, washed and finally stored in 70% ethanol. Specimens were housed in the repository of the Wildlife Institute of India, Dehradun. Microscopic observations of tadpoles were made under a stereo microscope (LEICA MZ 125) and measurements were obtained in ImageJ software.

## Abbreviations used

SVL- snout-vent length (for adult individuals), IOS- inter-orbital space (minimum distance between upper eyelids), BL- body length, TL- tail length, MTH- maximum tail height, TMW- tail muscle width, TMH- tail muscle height, LTRF- labial tooth row formula. Tadpole morphometry follows Altig (2007), except for IOS.

## Results

Calling activity and perch height of *Polypedates taeniatus*

Males started calling after 18:30 h. Initially, the males called from the ground and as time progresses, they gradually exposed themselves, calling from the perch above ground (Figure 2). The maximum perch height of the calling male was 126 cm above the ground (mean perch height= 23.24 ± 36.21 cm, n = 118). Calling intensity decreased after 22:00 h. Occasionally, a call could be heard in the early morning. The female perch height observed was 60–110 cm (88.33 ± 25.66 cm, n = 3).

## Sexual dimorphism

During the present study, we found the body size (SVL) of the adult male of *P. taeniatus* to be smaller than that of the female. SVL of the male individuals varies between 35.8–40 mm (37.28 ± 1.48, n = 8) and SVL of the female individuals ranges between 42.4–44.1 (43.46 ± 0.92, n = 3).

## Amplexus and nesting

We encountered six pairs in amplexus between 17 August to 22 August 2018. Our observation on breeding behaviour is based on four amplectant pairs (Table 1). Amplexus and nesting behaviour ensued in the following steps:

1. Females slowly moved on grass blades towards the calling male.
2. The female slowly descended to the ground at a distance of 110–140 cm from the calling male (n = 2).
3. The female carefully entered the grass (n = 2) or came to the same perch (n = 2) where the male is calling. The total distance travelled by the female to reach the mate was 160–610 cm (n = 3) within a

time range of 30–115 min (n = 3) (Table 1).

4. When the female approached very close, the male quickly mounted the female and engaged in axillary amplexus.
5. Amplexus occurred on the ground (n = 2) or above ground up to a height of 70 cm (48.33 ± 18.93 cm, n = 3). As soon as amplexus was initiated, the amplectant male stopped calling.
6. The amplectant pair remained in the same place where the male was calling (n = 2) or if amplexus occurred on grass blades then the amplectant pair descended to the ground after 5–15 min (n = 2) to find a suitable place to construct foam nest. The distance travelled by the amplectant pairs was 13–305 cm (n = 2).
7. After 28–125 min (n = 2) (Table 1) of amplexus, the pair started constructing a foam nest at the same place where the amplectant male was calling. Foam nests were constructed on the ground at the base of *Typha* or under the grass (Figure 4E). Before initiating foam nest construction, the female moved in a semi-circular way with the amplectant male for a few seconds. This is probably to clear the obstacles for foam nesting.
8. Then with the amplectant male, the female stood on the hind limbs grasping the surrounding grasses (Figure 4D).
9. The female initiated the foam nest construction by rubbing her hind limbs below the vent. The female moved her hind limbs 6–8 times (6.64 ± 2.09, n = 11) with an interval of 4–11 sec (6.44 ± 1.93 sec, n = 16). The male also showed similar hind limb movements and pelvic thrusts during spawning. But this behaviour was not as frequent as in the female. Initially, the male shook its hind limbs once, and later increased it to twice, and sometimes thrice. This movement is probably to spread the sperm for effective fertilization of the egg mass, as mentioned by Biswas (2000). This process continued until the construction of the foam nest was completed. The time taken to complete the foam nest was 29–57 min (n = 2) (Table 1).
10. After spawning, the amplectant male slowly moved out of the amplexus and left the place (n = 2).
11. The female stayed on the nest for 3 minutes (n = 1) or left the nest just after the male (n = 1).

In one case (20.08.2018) the amplectant pair dislodged after 25 min of coming to the ground and moved away from each other (Table 1). Within 15 min, the same male grasped another female which was at a distance of 110 cm and a perch height of 35 cm. We followed the pair up to 01:00 h but the pair didn’t start foam nesting (Table 1).

## Nest size and egg clutch

The foam nest was nearly rounded and cream-coloured (Figure 3E). The size of the foam nest was L = 48.64 ± 6.71 mm, W = 48.98 ± 7.51 mm, and H = 39.34 ± 7.21 mm (n = 6). The distance between the foam and the nearest water body was 47–410 cm (224.33 ± 124.9 cm, n = 6). The eggs numbered 220 and 308 in the two collected clutches. The eggs were round, unpigmented, and cream-coloured. The diameter of the egg was 1.35 ± 0.06 mm (n = 27).

## Larval description

The hatchlings of *P. taeniatus* develop in temporary water pools created by rain or overflow from the river (Figure 3). Tadpoles were elliptical and elongated in shape. Tail long, muscular, and laterally compressed with a pointed tip (Figure 5A-C, E, H). Snout rounded in dorsal view and oval in lateral view. Eyes lateral. Nostril lateral, small, closer to the snout. Pineal ocellus not distinct. Single rectangular shaped spiracle ventro-laterally placed on the left side (Figure 6G). Tail fin broad; dorsal fin originated at the junction of body and tail; ventral fin originated from anterior of the vent tube. Vent tube dextral. Oral disc anteroventral. Marginal papilla with a wide gap on anterior labium and small gap at the middle of the posterior labium. Labial tooth row formula (LTRF) observed at Gosner stage 31 was 4(2-4)/3(1) (Figure 6H) and at Gosner stage 27 was 3(2-3)/3(1). Jaw sheaths serrated, upper jaw sheath arch shaped and lower jaw sheath “V” shaped. Marginal papillae, jaw sheath and labial tooth started degenerating at stage 41. Details of tadpole stages (Gosner 1960) of *P. tae-*

*niatus* observed in the present study are given in the Table 2.

### Live colouration of the tadpole

Dorsal body and the muscular part of the tail is dark or light brown (Figure 7). Ventral and lateral parts of the body below the level of the eyes is creamy white, sometimes with dark brown patches. Eyes red with black rounded pupils. Lateral line present on body and tail. A faint mid-dorsal line may be visible. A lateral golden stripe runs up to the tail tip radiating at the middle of the body, broader at the base of tail. Fins of the tail transparent with dark brown irregular markings.

## Discussion

*Polypedates taeniatus* is locally abundant in the study area (Bijnor, Uttar Pradesh), mostly restricted to the fragmented grasslands like *Typha angustifolia*, and *Saccharum spontaneum*. We also observed the species in crop fields, sugarcane cultivations, and marsh lands along the Ganga river, albeit rarely. Earlier, Hegde et al. (2009) reported the species from Ravali forest of Bijnor, Hastinapur forest, and Ram-Ganga river bank. Also, Boruah et al. (2018) recorded the species in the riverine vegetations along the River Ganga between Bijnor and Narora (Ramsar conservation site), Uttar Pradesh.

Earlier documentation of larval development of Indian foam nesting rhacophorids includes *P. teraiensis* (Chakravarty et al. 2011; Tamuly and Dey 2014), *P. maculatus* (Mishra and Dash, 1984; Mohanty-Hejmadi & Dutta 1988; Girish and Saidapur 1998), *P. megacephalus* (Tesia et al. 2017), *P. leucomystax* (Kiyasetuo and Khare 1986; Iangrai 2007), *R. malabaricus* (Sekar 1990), *Z. smaragdinus* (= *maximus*) (Khongwir et al. 2003), and *R. bipunctatus* (Iangrai, 2007). Although, development of *R. rhodopus* and *R. smaragdinus* (= *maximus*) is reported from Thailand (Grosjean and Inthara 2016) and Vietnam (Wildenhues et al. 2010), respectively. Postembryonic development of *P. taeniatus* is described by Deuti et al. (2018). (Table 3). However, Deuti et al. (2018) have not provided information on breeding behaviour of the species. In the present study, we provided information on amplexus, spawning, clutch size, and also live colouration of the tadpole. *Polypedates taeniatus* breeds for a short period between mid-July and end of August. The duration of amplexus (85–154 min), foam nesting (29–57 min) and clutch size (220–308) is nearly similar with that of congeneric species (Table 3). The diameter of the egg is smaller than that of other foam nesting rhacoph-

**Table 1.** Details of the breeding and foam nesting behaviour of *P. taeniatus*. (M-male, F- female, L- length, W- width, H- height, measurements are given in mm).

| Date | Temp. (˚C) / humidity (%) | Perch height | | Dist. Between M and F | Amplexus start time | Foam nesting | | M dislodge time | F leaving time | Nest size (L/W/H) | Dist. from water | Clutch size | SVL | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | F | M | | | Start time | Finished time | | | | | | F | M |
| **19 August 2018** | 29.2/96.0 | 1100 | ground | 2300 | 21:15 h | 23:20 h | 23:49 h | As soon as the nest was completed | 00:04 h | 43/49.5/35.4 | 1800 | eggs eaten by maggots | 44.1 | 36.6 |
| **21 August 2018** | 28.4/96.1 | 950 | ground | 1600 | 20:56 h | 21:24 h | 22:21 h | 22:24 h | 22:25 h | 50.7/43.1/38.8 | 470 | 220 | 42.4 | 35.8 |
| **20 August 2018** | 27.3/98.0 | - | 400 | - | 21:15 h | Dis-lodged | - | - | - | - | - | - | - | - |
| **20 August 2018** | 27.3/98.0 | 600 | 350 | 6100 | 21.55 h | Not com-menced | - | - | - | - | - | - | - | - |

**Table 2.** Morphological measeurements (given in mm) and characteristics of *P. taeniatus* tadpole in different stages (Gosner 1960).

| Gosner Stage | BL | TL | MTH | TMH | TMW | IOS | ED | Characteristics |
|---|---|---|---|---|---|---|---|---|
| **27 (n = 1) (Figure 5A)** | 3.6 | 6 | - | 1.1 | 0.7 | 1.1 | 0.6 | Hind limb bud length greater than half of its diameter. Melanophores spread along lateral side of the body, tail muscle and fins; dense on dorsal side of the head. |
| **28 (n = 1) (Figure 5B)** | 4.3 | 8.4 | 1.85 | 1.2 | 1 | 1.3 | 0.7 | Hind limb bud greater than its diameter. Melanophores denser than previous stage. On fins melanophore forms irregular lines. |
| **31 (n = 3) (Figure 5C-D)** | 5.5 ± 26 | 10.6 ± 0.5 | 3.6 ± 0.1 | 1.6 ± 0.1 | 1.3 ± 0.05 | 1.7 ± 0.06 | 0.9 ± 0.06 | Hind limb bud becomes spatula shaped. Toe development starts. Melanophore pigmentation more dispersed. |
| **33 (n = 1) (Figure 5E-F)** | 6 | 11.8 | 3.7 | 2.1 | 1.5 | 1.9 | 1 | Three toe nubs of 3rd, 4th and 5th toe develops. Melanophore spread on base of the hind limb. |
| **35 (n = 3) (Figure 5G)** | 7.1 ± 0.2 | 14.1 ± 0.3 | 4.7 ± 0.1 | 2.6 ± 0.3 | 1.9 ± 0.06 | 2.2 ± 0.1 | 1.1 ± 0.06 | Hind limb more elongated. 5 toe nubs well defined. |
| **36 (n = 1) (Figure 5H)** | 6.6 | 12.4 | 4 | 2 | 1.7 | 1.9 | 1.1 | 3rd, 4th and 5th toe separated. Melanophores concentrated on distal part. |
| **37 (n = 2) (Figure 5I)** | 7.9 ± 0.4 | 15.7 ± 0.04 | 4.4 ± 0.1 | 2.5 ± 0.1 | 2 ± 0.1 | 2.4 ± 0.2 | 1.3 ±0.01 | Thigh, tibia and tarsus distinct; all toes separated and elongated. Webbing clearly visible. Melanophore pigmentations more prominent on gular and abdomen. |
| **39 (n = 2) (Figure 5J)** | 8.3 ± 0.4 | 16.4 ± 0.5 | 4.8 ± 0.1 | 2.7 ± 0.08 | 2.3 ± 0.1 | 2.4 ± 0.3 | 1.3 ± 0.03 | Inner metatarsal tubercle visible; subarticular tubercles appeared as clear round patch. |
| **40 (n = 2) (Figure 6A)** | 8.4 ± 0.4 | 16.4 ± 0.8 | 4.5 ± 0.7 | 2.6 ± 0.05 | 2.2 ± 0.1 | 2.3 ± 0.1 | 1.4 ± 0.05 | Subarticular tubercle, circum-marginal groove distinct with well-developed toes. Fold of skin present between thighs. Forelimb bulges slightly visible. |
| **41 (n = 2) (Figure 6B)** | 8.3 ± 0.02 | 17.4 ± 0.8 | 4.1 ± 0.2 | 2.5 ± 0.05 | 2.3 ± 0.01 | 2.2 ± 0.01 | 1.4 ± 0.04 | Cloacal tail piece disappeared. Forelimb bulges clearly visible. |
| **42 (n = 3) (Figure 6C-D)** | 8.6 ± 0.3 | 17.3 ± 0.8 | 3 ± 1.1 | 2.5 ± 0.4 | 2.2 ± 0.2 | 2 ± 0.05 | 1.5 ± 0.04 | Forelimb emerges with well-developed fingers; left forelimb emerges first. Granules on thigh and posterior part of the abdomen prominent. |
| **43 (n = 2) (Figure 6E-F)** | 8.9 ± 0.3 | 14 ± 2.8 | 2.2 ± 0.9 | 2 ± 0.05 | 2.3 ± 0.1 | 1.8 ± 0.1 | 1.4 ± 0.2 | Lateral margin of the mouth reaches between nostril and eye. Tail and fins reduced. |

**Table 3.** A comparative chart of foam nesting and breeding behaviour in Rhacophorids of India.

| Species Name | Duration of Amplexus (min) | Duration of Foam Nesting (min) | Size of Nest (L/W/H) (mm) | Substratum | Clutch Size | Diameter of Egg (mm) | Locality | Reference |
|---|---|---|---|---|---|---|---|---|
| ***Chirixalus simus* (Annandale 1915)** | - | - | 52–73/19–36/16–28 | On grass, 6–58 cm above water, 17–89 cm above land | 153–234 | - | West Bengal | Deuti (2001) |
| ***Chirixalus dudhwaensis* (Ray 1992)** | - | - | 30–67/27–55/13–17 | On leaves 0.6–1.8 m above water | 201–231 | - | WII, Uttarakhand | Biswas (2000) |
| ***Rhacophorus bipunctatus* Ahl 1927** | - | - | 60–80 (diameter) | On grass near water, tree branches overhanging water | 45–76 | 3 | Meghalaya | Iangrai (2007) |
| ***Zhangixalus smaragdinus* (= *maximus*) (Blyth 1852)** | - | - | 155–205 (L) | On vegetation 2–3 cm above water, earthen bank, Rock 30 cm above water | 809–2059 | 2 | Cherrapunjee, Meghalaya | Khongwir et al. (2016) |
| ***Rhacophorus malabaricus* Jerdon 1870** | 120–180 | 15–20 | - | Overhanging branches, on ground with leaf litter | 160–192 | 2.62 (n=10) | Western Ghats | Kadadevaru and Kanamadi (2000) |
| ***Rhacophorus lateralis* Boulenger 1883** | 35–50 | 17–29 | 53–80 (L), 28–64 (diameter) | Single leaf | 43–72 | - | Western Ghats | Biju (2009) |
| ***Polypedates leucomystax* (Gravenhorst 1829)** | - | | 60–120 (diameter) | Vegetation along pond or on concrete 5–40 cm above water | 250–530 | 1.5 | Meghalaya | Iangrai (2007) |
| ***Polypedates leucomystax* (Gravenhorst 1829)** | 270 | 60 | 58–63/58–61/65–80 | On Branch 5.6 ft above water | - | - | Meghalaya | Banerjee and Deuti (2006) |
| ***Polypedates maculatus* (Gray 1830)** | 150 | - | - | Concrete above water | 210–448 | - | South India (locality not specified) | Girish and Saidapur (1999) |
| ***Polypedates himalayensis* (Annandale 1912)** | - | - | - | Earthen hole, under moist vegetation | | - | Meghalaya | Hooroo et al., (2017) |
| ***Polypedates megacephalus* Hallowell 1861** | - | - | - | Vegetation and twigs overhanging water | 500–600 | 2.1–2.9 | Arunachal Pradesh | Tesia et al., (2017) |
| ***Polypedates taeniatus* (Bpoulenger 1906)** | 85–154 | 29–57 | 39.6–58.3/37.2–57.3/27.2–44.7 | On ground; under grass or base of Typha | 220–308 | 1.2– 1.4 (N= 27) | Uttar Pradesh | Present Study |

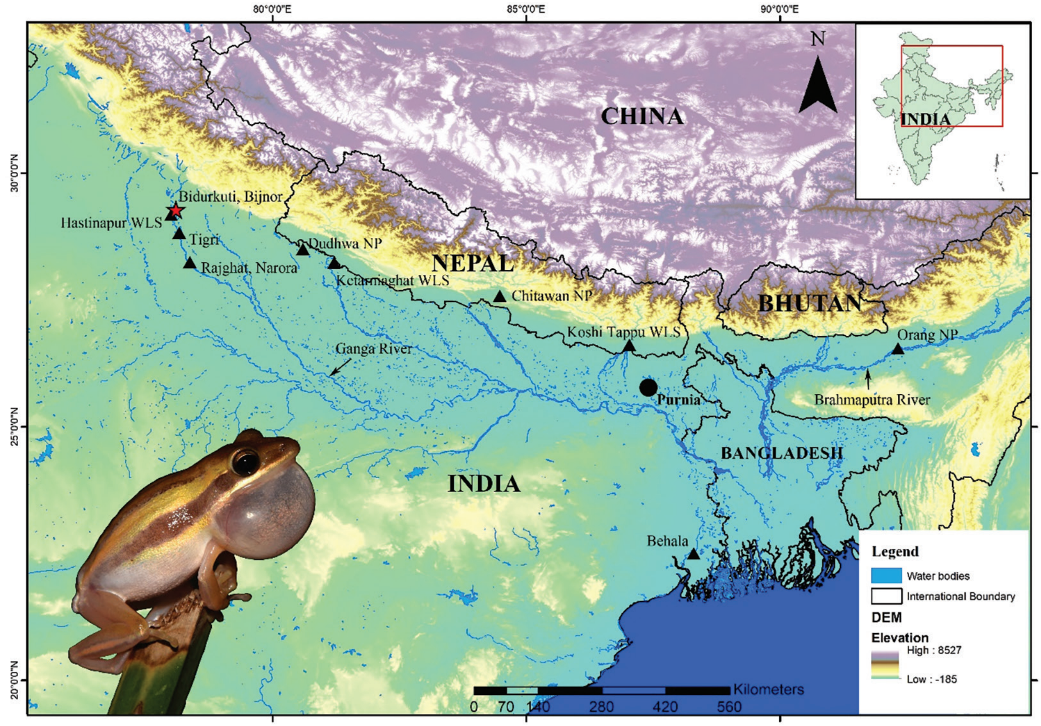

**Figure 1.** Distribution of *P. taeniatus* in India and Nepal. Recorded locality of *P. taeniatus* based on available literature (black triangle), type locality (black circle), study site (red star).

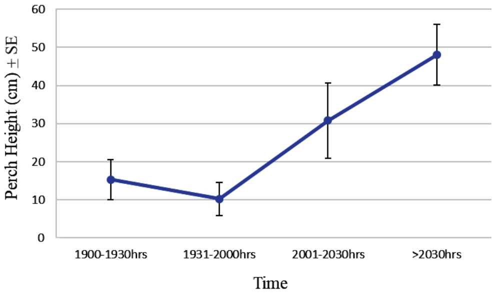

**Figure 2.** Perch height of the male individuals (*P. taeniatus*) with respect to time, N = 145.

**Figure 3.** A typical breeding habitat of *Polypedates taeniatus*. Scale bar = ~ 1 m. and Das 03.tif

orids (Table 3). Size of the foam nest found in the present study was smaller than that observed by Deuti et al. (2018); i.e. 60 mm × 60 mm. Like in other rhacophorid frogs (*C. dudhwaensis, R. malabaricus*, *R. lateralis*, *C simus*, *Z. smaragdinus* (= *maximus*)), (Biswas 2000; Kadadevaru and Kanamadi 2000; Biju 2009; Banerjee 2010, 2014; Khongwir et al. 2016), we did not find any satellite male disturbing the amplectant pair. The reproductive mode of *P. taeniatus* belongs to the type 28 (i.e. foam nest on ground and after flooding, exotrophic tadpoles develop in standing water body) as delineated by Haddad and Prado (2005). During the study period, foam nests of *P. taeniatus* were found at a distance of 47–410 cm from the nearest water puddle. Another congeneric species from Northeast India, *P. himalayensis*, is known to construct the foam nest inside earthen holes or under vegetation, 50–100 cm away from waterbodies (Hooroo et al. 2017). This could be to minimize the predation risk from aquatic predators. Moreover, the possible reasons for avoiding arboreal foam nesting could be: (1) to prevent the desiccation of the egg mass from sunlight as desiccation is the main threat to larval development in tropical

**Figure 4.** Breeding behaviour of *Polypedates taeniatus*. A. calling male on its perch, B. an amplectant pair in arboreal habitat, C. an amplectant pair on the ground, D. foam nesting on the ground, E. a completed nest, F. male and female after leaving the nest.

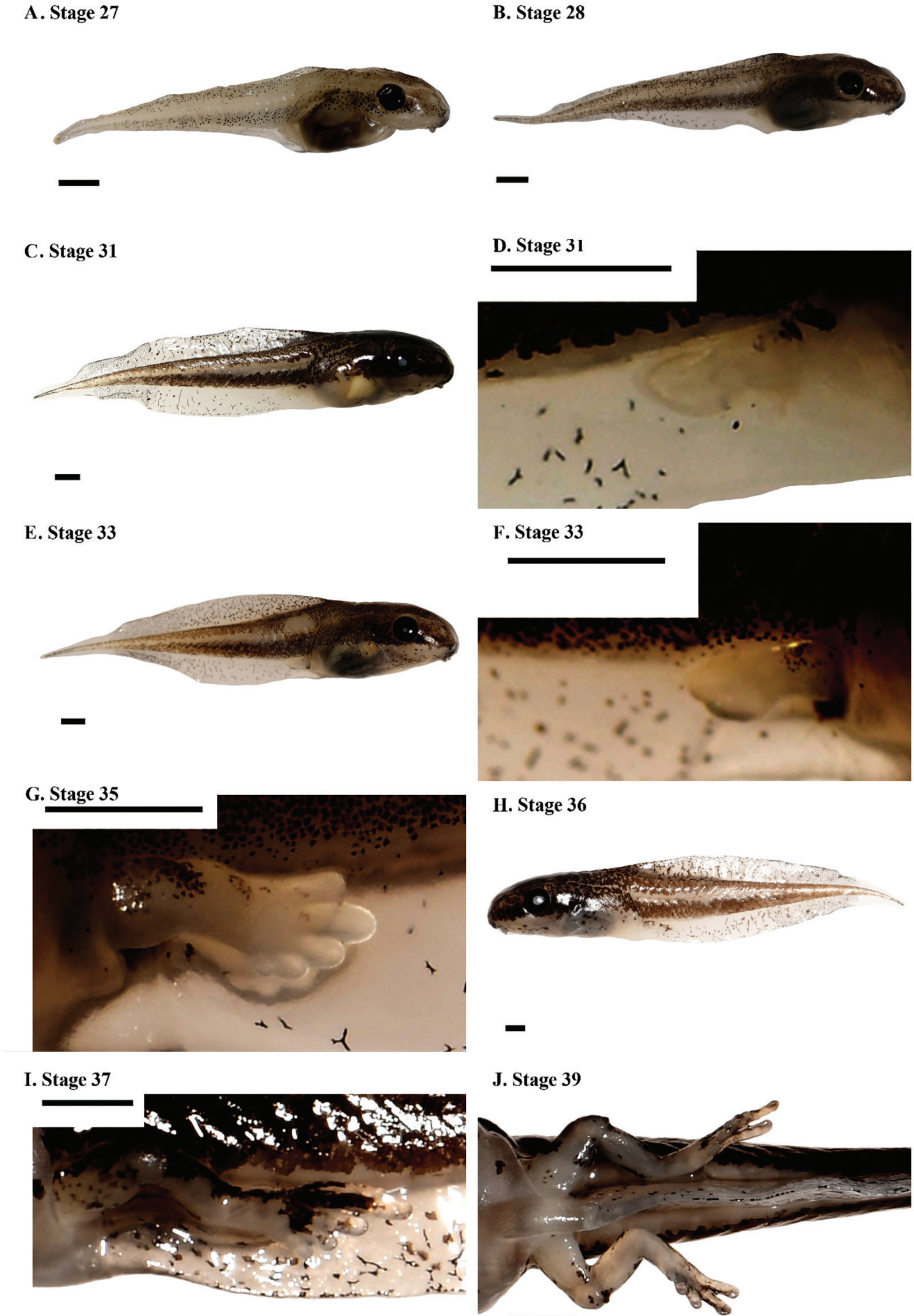

**Figure 5.** Gosner stages of P. taeniatus. A- stage 27, B- stage 28, C-D- stage 31, E-F- stage 33, G- stage 35, H- stage 36, I- stage 37, J- stage 39. (Scale- 1 mm).

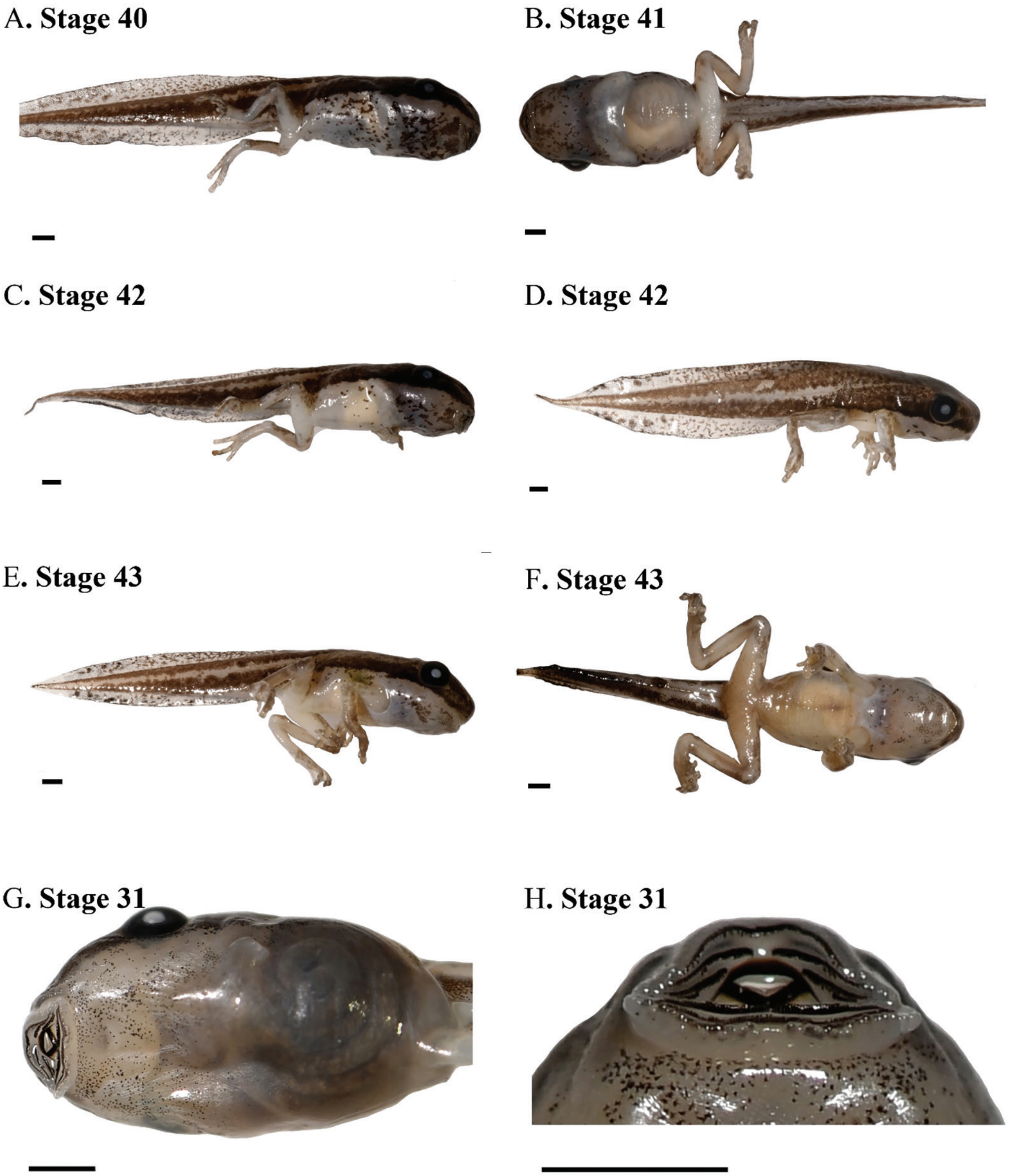

**Figure 6.** Gosner stages of *P. taeniatus*. A- stage 40, B- stage 41, C-D- stage 42, E-F- stage 43, G- stage 31 showing spiracle, H- stage 31 showing LTRF. (Scale- 1 mm).

region (Mohanty-Hejmadi and Dutta 1988), (2) dependency on overflow water of the river rather than rain for larval development.

The external morphology of the tadpole of *P. taeniatus* is similar to that of the congeneric species from India. The LTRF of *P. taeniatus* is similar to that of *P. teraiensis* [4(2-4)/3(1)]. Additionally, there is no interruptions in the first tooth row in the posterior papilla (i.e., 4(2-4)/3) as seen in *P. maculatus* (Mohanty-Hejmadi & Dutta 1988) and *P. himalayensis* (Gogoi and Sengupta 2017). The LTRF of *R. malabaricus, Z. smaragdinus* (= *maximus*) and *P. megacephalus* is 5(2-5)/3(1) (Sekar 1990; Wildenhues et al. 2010; Tesia et al. 2017). The LTRF of *R. rhodopus* is 6(2-6)/3(1) (Grosjean and Inthara 2016). The tadpole of *P. teraiensis* does not have the lateral line which is present in the tadpoles of *P. taeniatus* and *R. rhodopus*.

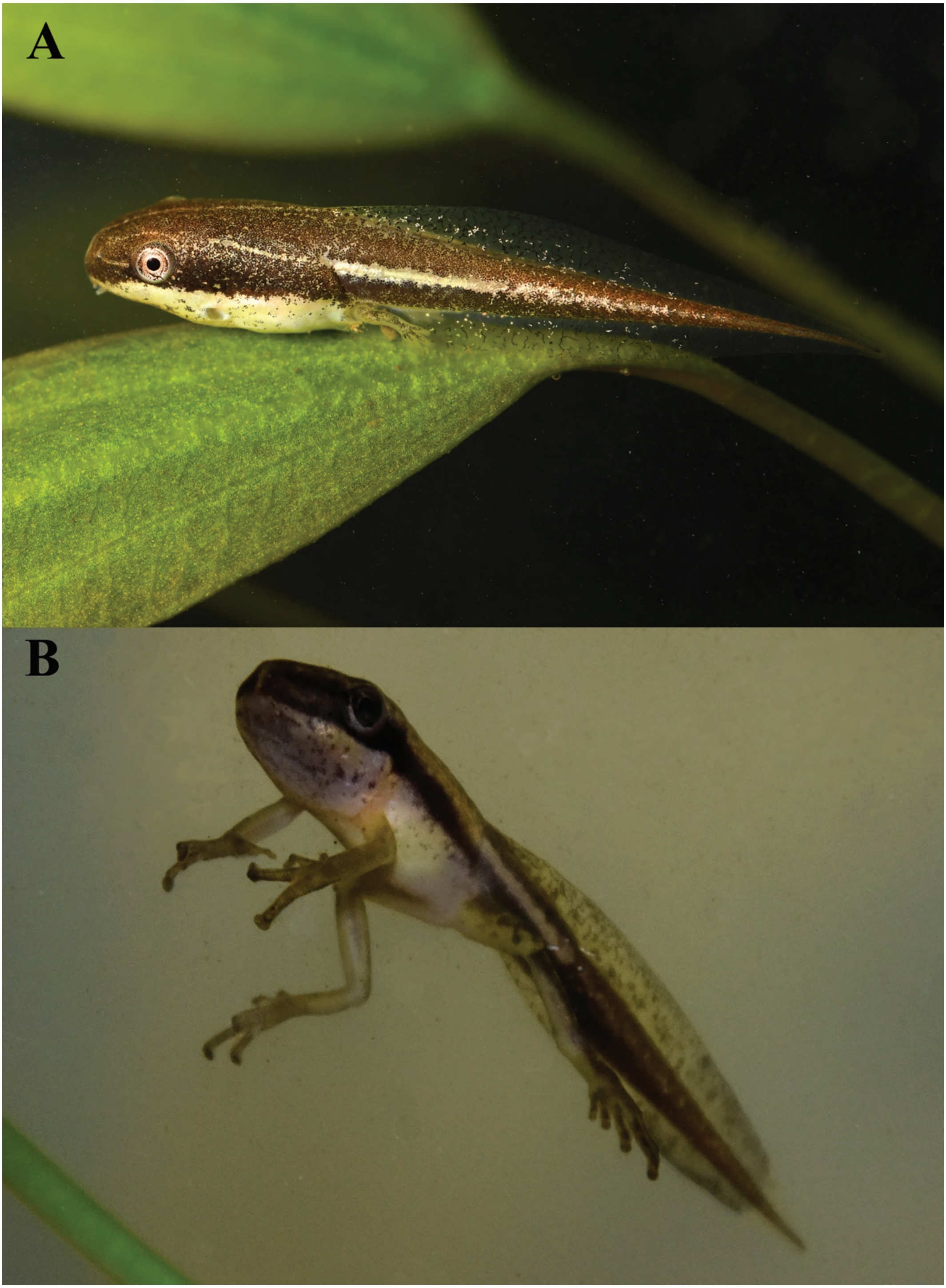

**Figure 7.** Tadpole of *P. taeniatus* in life. A. stage- 37, B. stage- 43.

*Polypedates taeniatus* appears to have the narrowest distribution range at an elevation below 500m a.s.l., inhabiting the fragmented grassland and *Typha* patches of the Gangetic and Brahmaputra plains. Rapid conversion of these natural habitats to agricultural land and settlement may lead to a decline in the populations of this species. Therefore, immediate conservation of the species is vital.

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# The Asian Water Monitor, *Varanus salvator*: An Introduction to its Non-Breeding Ethology.

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**ABSTRACT.– The Asian Water Monitor (*Varanus salvator*) is a highly adaptable species that lives in close proximity to humans. The present study aims to construct an ethogram of *V. salvator* to obtain a complete record of its behaviours in its natural environment. This ethogram is constructed on the basis of all the exclusive behaviours (39) and behavioural categories (8) documented for 43 individuals from two study sites situated on the outskirts of Kolkata, India. The ethogram reveals that *V. salvator* devotes most of its time to foraging (84.63 minutes, 37.58%) and sluggish movements (50.73 minutes, 22.52%). This study also indicates the activity period of the species and its interaction with anthropogenic disturbances, interestingly showing very less temporal overlap with human activity. These results reflect an overall idea of the non-breeding behaviour of *V. salvator* and highlight the survival strategies of such opportunistic species amidst two highly congested areas.**

**KEYWORDS. Focal Animal Sampling, Repertoire, Behavioural Pattern, Ethogram, Co-exist**

## Introduction

An ethological study begins with observing each behaviour of the species exclusively and constructing a repertoire known as an ethogram. An ethogram is a list of exclusive behaviours of the studied animal describing its elementary and functional patterns (Altmann 1974, Lehner 1998). The behaviour of an animal is the link between a species and its environment and plays a crucial role in understanding its evolutionary perspective (Breed and Sanchez 2010). Behavioural understanding deals with systematic field observations merging with prior theoretical knowledge of the species, and quantitative analysis of their ethological pattern. Such *apprehension* is helpful to *understand the* behavioural complexity of species and build conservation measures for them and their habitat (Sutherland 1998).

The Asian water monitor (*Varanus salvator*) has a wide distribution in Tropical Asia and has occupied a large range of habitats (Bennett et al. 2010). *Varanus salvator* is the most widespread species among all varanids (Koch 2007) found in Sri Lanka, northern India, Bangladesh, Burma, Vietnam and Hainan to Peninsular Malaysia and the Indonesian islands (Traeholt 1993; De Lisle 2007). It is one of the largest extant lizards in the world, reaching an average length of 1.5 m (Karunarathna et al. 2017). *Varanus salvator* is known to maintain healthy populations and can very well adapt to human-moulded habitats (Uyeda 2015, 2009; Karunarathna *et al.* 2017; Chatterjee and Bhattaccharya 2014). It is the only monitor that is known to have colonised the marine habitat successfully and is adapted to both freshwater as well as salt water habitats (Traeholt 1993). They are a semi-aquatic and generalist species inhabiting a variety of natural habitats (Gaulke and Horn, 2004). Being a diet generalist provides added ecological plasticity to this species (Karunarathna *et al.* 2017). Ac-

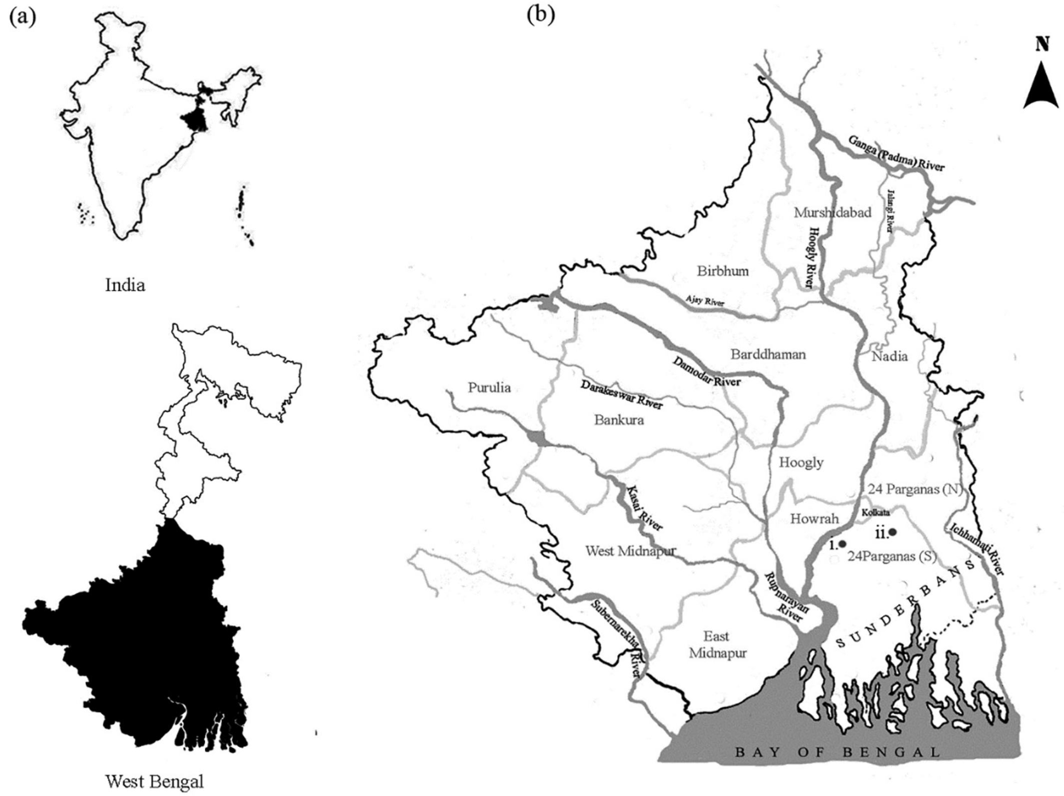


**Figure 1.** Map showing (a) Location of West Bengal in India. (b) Study locations: i. Budgebudge, ii. CKBS

cording to Carl Traeholt (1993) water monitors are territorial and show area-concentrated foraging behaviour (Traeholt 1997; Gaulke *et al.* 1999; Gaulke and Horn 2004).

The water monitor is the third most heavily traded species in the world (Pernetta 2009) and has been subjected to hunting pressures in the international leather trade due to the demand for their skin. Its meat is eaten and the fat is used in traditional medicine (Uyeda 2009; Bennett *et al.* 2010). Despite such threats, the species is not enlisted as a protected species according to international organizations like IUCN and CITES. IUCN listed *V. salvator* in the category of least concern (LC) whereas the Convention on International Trade in Endangered Species (CITES) trade database included them in Appendix II which comprises species that are not presently threatened with extinction but are at risk unless trade is closely controlled. (Bennet *et al.* 2010; CITES 2020). In India they are included in the highest category of protection i.e., Schedule I, according to the Indian Wildlife Protection Act, 1972 (IWPA, 1972).

Several ethological studies exist on different species of varanids throughout the world but until now a limited number of studies have been conducted on the behaviour of *V. salvator* in their wild habitat (Auffenberg 1994; Uyeda 2015; Chatterjee and Bhattaccharya 2014). The present study aims to describe the non-breeding behaviours of *V. salvator* in its natural habitat (both urban and semi-urbanised settings) in southern part of West Bengal, India and to prepare a detailed ethogram for understanding their behavioural patterns which can lead developing explicit conservation strategies for them in future.

## Material and Methods

**Study sites.** A pilot survey was conducted in different locations on the outskirts of Kolkata city, West Bengal, India. The two sites finalized for the in-depth study are rural areas of Budgebudge (22°24’54”N; 88°11’20”E) and Chintamani Kar Bird Sanctuary (CKBS) (22°25’45”N; 88°24’4”E) (Fig. 1). Previous studies (Chatterjee and Bhattacharyya 2014;

Chitra 2015) suggested that both the areas comprise a substantial number of *V. salvator* along with its potential habitat. Budgebudge is a village set up with typical elements such as crop field and settlement areas dotted with small wetlands; whereas CKBS is a small sanctuary (0.07 sq km area) with extremely high tree density situated amidst a congested urban system interspersed with small water bodies like a few ponds and a canal (Wildlife Wing, 2020). These study locations were selected due to their differences in conservation practices. CKBS is a bird sanctuary managed and maintained by the state forest department whereas Budgebudge is a village area with the absence of any formal wildlife protection measures (Wildlife Wing, 2020, Chatterjee and Bhattacharyya, 2014). The geological and climatic conditions are similar in both the places as these are situated on the same side of the lower Gangetic alluvial flood plains (Sirohi, 1989). Mean daily temperature varies around 31 ˚C. Summer temperatures cross 42 ˚C and winter temperatures fall up to 12 ˚C. The atmosphere is moist humid (70.8%) throughout the year except in winter, and annual precipitation is moderate (1641.4 mm) (Indian Meteorological Department 2014).

**Data collection.** This study was conducted during the non-breeding period (May–June 2018) of the species. Initially an *ad libitum* survey was performed in the study locations to identify the potential habitats for the Asian Water Monitor. We used opportunistic encounter followed by the Focal Animal Sampling (FAS) method for the in-depth study in which all occurrences of interactions of an individual were recorded without interfering or disturbing their activities (distance of minimum 150 m from the individual) and each exclusive behaviour was noted down with a detailed description of the posture. The length of each sample period for each focal individual and the amount of time during which the individual is in view was recorded (Lehner 1996; Altmann 1974). The amount of time the animal was in sight and the frequency of each sub-behaviours was noted down. While conducting the FAS in an area from a vantage point it was only possible to observe the focal animal when it was within the viewing range of the observer. Therefore, the activity data was taken for those individuals till they were in sight and recorded till the time they moved out of the viewing range of the observer during each time slot. In each time slot the habitats were scanned thoroughly and if any individual were sighted, the observation was conducted for the individual till it was in sight with each behavioural detail along with duration and frequency of each behaviour being noted down. Once chosen, a focal individual was followed to whatever extent possible (till the focal animal was in view) during each sample periods (Altmann, 1974). Binoculars (Nikon; resolution: 8 x 42) were used for enhancing the visibility of the species and occasionally a camera (Model: Canon 700D, Lens: 55-250 mm) was employed to record their different behavioural activities. Later the direct observation notes were cross checked with the video to standardize the technique. Using this data an ethogram was constructed describing their postures and displays and to understand the patterns of behaviour. A repertoire was made consisting of all the behaviours shown by the monitor while it was in sight. These behaviours were assigned to functional categories based on prior studies on lizards (Tracy Langkilde et al., 2003; Carpenter et al., 1970; Done & Heatwole, 1977; Whittier & Martin, 1992; Torr & Shine, 1994; Whittier, 1994).

Observations were made for 43 individuals (N=43) in different developmental stages (juvenile, sub-adult, adult) that were identified from the SVL (Snout to Vent length) of the individual (Amarasinghe *et al.* 2009; Langkilde *et al.* 2003; Torr and Shine 1994). Thirty eight adults, two sub-adults and three juveniles were observed. Data was collected over a period of seven different time slots (each slot comprises 2 hrs.) in a day starting from 06:00 hrs. in the morning to 20:00 hrs. in the evening. In a single field day, a maximum of three time slots were covered to minimise observer fatigue and bias in data collection. Night sampling was not done due to poor availability of light after 20:00 hrs. (Bandyopadhyay *et al.* 2014). Climate data (temperature, humidity and rainfall) were recorded using HTC thermo-hygrometer for the days (for each time slots) when field surveys were conducted. Total 225 minutes and 14 seconds of on-field sighting data during May–June period was documented covering different times of the day to construct a detailed exclusive ethogram (Langkilde *et al.*

2003; Torr and Shine, 1994; Bandyopadhyay *et al.* 2014). An exclusive ethogram is where each behaviour performed by the species is considered as single behavioural type in the ethogram (Lehner 1998). Data collected from the two survey sites were pooled to perform further analysis and construct the ethogram.

**Data analysis.** Analyses of the number of times each animal performed each behaviour during each time slot yielded quantitative results. The data from each observation period for the two sites were combined. For statistical analysis we have combined behavioural data from all observed animals, treating all instances of a particular behaviour pattern as independent observations (Torr and Shine 1994). This assumption was done due to small sample size for many behaviours and to aid in statistical analysis. The temperature recorded during survey days were grouped into three clusters of $< 30$ $^{0}$C, $\geq 30–40$ $^{0}$C, and $\geq 34–38$ $^{0}$C. The number of individuals sighted during each temperature cluster were then analysed in MS Excel, and a regression analysis was performed. Sightings of focal animals during each time slots were then plotted in a graph showing the number of individuals sighted during each of the surveyed time slots.

### Results

Behaviours were assigned to four functional categories based on prior studies on lizards (Carpenter *et al.* 1970; Greenberg 1977a; Whittier and Martin 1992; Torr and Shine 1994; Whittier 1994; Langkilde *et al.* 2003). Each behaviour were further broken down into exclusive behaviours (Lehner 1996) in the ethograms for time budgeting and to reflect detailed behaviours of *V. salvator*.

### *Ethogram*

A total of 39 exclusive behaviours of *V. salvator* were observed and identified. According to their elements of behaviour, the observed 39 behaviours are clustered into eight behavioural categories. i.e., foraging, feeding, movement, sluggish movements, resting, basking, alert, and interaction. Furthermore, these behaviours were assigned to four functional categories (exploratory, maintenance, escape and social). The detailed repertoire of each exclusive behaviours and their behavioural and functional categories are described in Table 1.

### *Functional category I: Exploratory*

#### <u>Behavioural category I: Foraging</u>

*Exclusive Behaviour I: Swimming*

Exploring for potential prey. Swift locomotion in water bodies for exploration of prey using the two pairs of limbs and the long tail accompanied by tongue flicks and head turns. They forage in the deepest part of the water bodies i.e., in the centre, mostly searching for fish as well as along its banks covered with submerged vegetation in search of small invertebrates and amphibians.

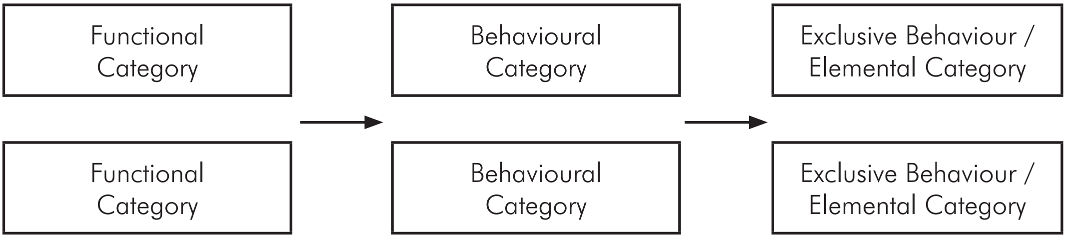
*Exclusive Behaviour II: Crawl*

A lateral undulatory gait, wherein the body flexes sideways while walking. An investigative search for food (Treaholt 1993). A swift forward movement of the body on land with two pairs of limbs and the tail in contact with the surface. Foraging walk is accompanied by constant tongue flicking and looking sideways trying to locate prey using their specialized tongue.

*Exclusive Behaviour III: Tongue flick*

Tongue is specialized to sense the environment and helps in locating prey. Unlike snakes, monitors lack a slit in-between the upper and the lower jaw through which the tongue could make its way out easily, therefore they have to open their mouth to expose their bi-forked tongue. During ‘tongue flicking’ the tongue

**Table 1.**



droops down close to the ground before it is retracted back inside. The rate of tongue flicking is seen to vary from being as rapid as two flicks a second to as slow as one flick per 2 seconds or more. While foraging in water most of the times the tongue touches the water and then goes under the water surface.

*Exclusive Behaviour IV: Head turn in movement*

Swaying its neck and head sideways to have a 180° view while crawling or swimming; on turning its head and neck 90° left or right, the monitor's upper body (from head to neck) looks like alphabet 'C'.

*Exclusive Behaviour V: Motionless head turn at rest*

Turning its head sideways or gazing straight while the body stays still on ground or in water showing very limited movement.

*Exclusive Behaviour VI: Head and neck raise*

At times the monitor stops the foraging walk or swim and fully stretches its neck and turns its head from left or right to have a better view of its surroundings, giving the neck and the head a telescopic appearance.

*Exclusive Behaviour VII: Dip*

Individuals dipped their snout inside water while foraging for food in water, or from the bank. While on the bank they dipped their snout inside the water to search for food.

*Exclusive Behaviour VIII: Sudden stall*

Continuous foraging walk or swim is interrupted by sudden stalling for a few minutes and then the tongue flicking and walk/swim continues.

*Exclusive Behaviour IX: Climbing*

Going up a tree using two pairs of limbs accompanied by the hook shaped nails and the long tail with occasional tongue flicks, in search for food.

*Exclusive Behaviour X: Dive*

Leap into the air such that all four limbs leave the substrate and landing or plunging in water with a heavy splash usually from a tree or some other elevated surface, followed by a quick swim.

*Exclusive Behaviour XI: Underwater Dive*

While diving in the pond, the whole body goes underwater instantly. The animal resurfaces in a while.

### ***Functional category II: Maintenance***

#### **Behavioural category I: Feeding**

*Exclusive Behaviour I: Grab*

Rapid forward movement of the head in order to grasp the food using the jaws. The food is held horizontally in the jaws in order to get grip.

*Exclusive Behaviour II: Jerk*

Shaking the head sideways while grabbing the food in the jaws in order to kill the prey completely.

*Exclusive Behaviour III: Swallow*

Engulfing the entire food (anterior part first) by directing the head upwards followed by stretching and nodding of the neck.

*Exclusive Behaviour IV: Gape*

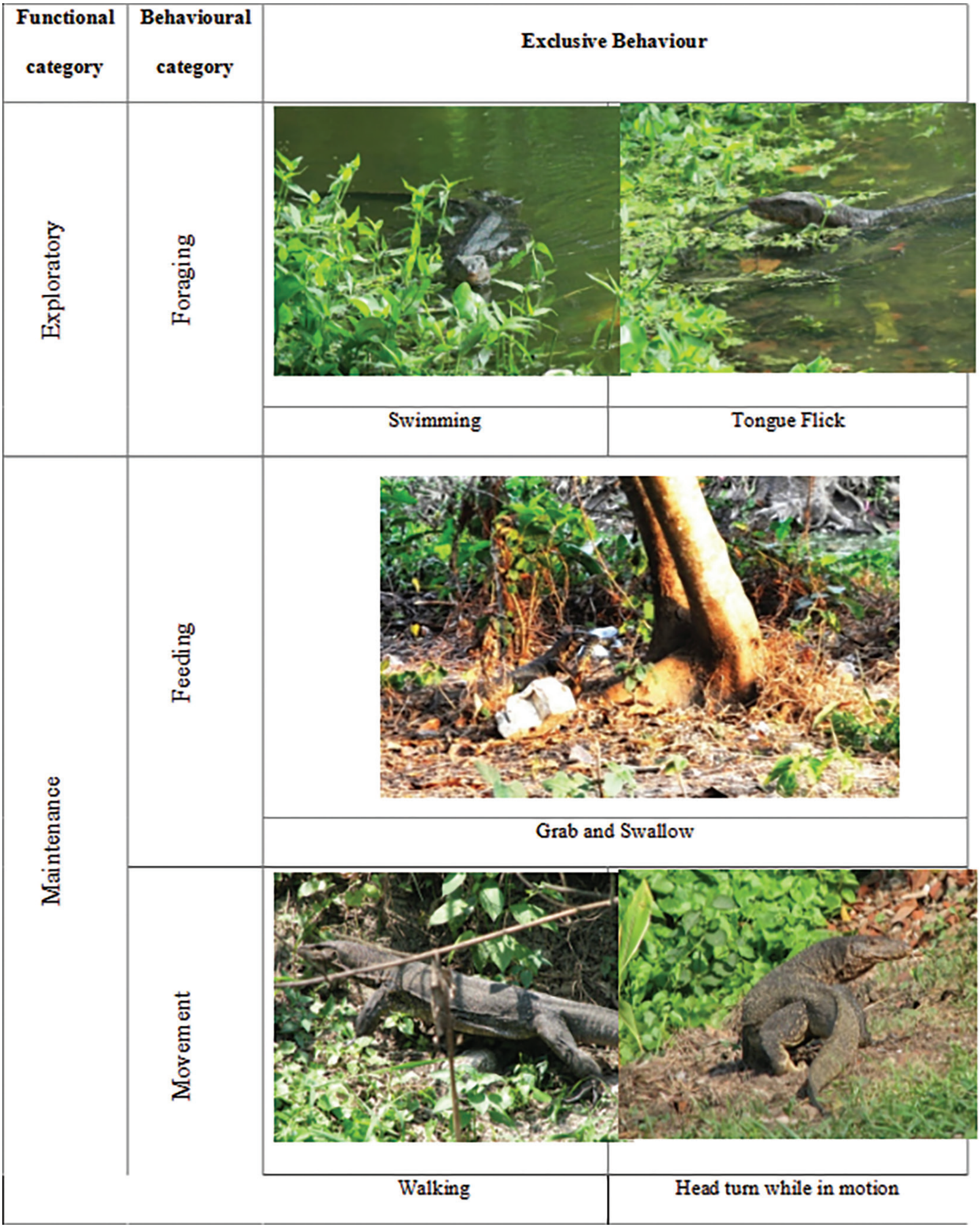
After engulfing food, the monitor let its mouth wide open to take in air. If the food is dry or bigger in size it takes more time to engulf and hence becomes difficult to pass through the throat. This might be the reason behind the high oxygen demand post swallowing the food (Hicks *et al.* 2000), engulfing the food might sometimes be followed by wide opening of the mouth to take in air.

*Exclusive Behaviour V: Wipe*

Wiping or encircling the mouth with tongue after the food is down the throat.

*Exclusive Behaviour VI: Drinking*

The snout is placed in water while the monitor sips water using its tongue.



| Functional category | Behavioural category | Exclusive Behaviour | |
|---|---|---|---|
| Exploratory | Foraging | Swimming | Tongue Flick |
| Maintenance | Feeding | Grab and Swallow | |
| | Movement | Walking | Head turn while in motion |

**Figure 2.** Behavioural patterns of the Asian Water Monitor

### Behavioural category II: Movement

*Exclusive Behaviour I: Walk*

A swift forward movement of the body on land without tongue flicking. The hind limbs land 3–4 cm away from the fore limbs. Walking is a wavelike movement of the body and the body forms both the crest and trough in a single crawl.

*Exclusive Behaviour II: Slow motion*

Unrushed movement or gentle forward movement of the body at a pace far slower than that of the normal walk. Hind limbs lands 4–8 cm away from the fore limbs.

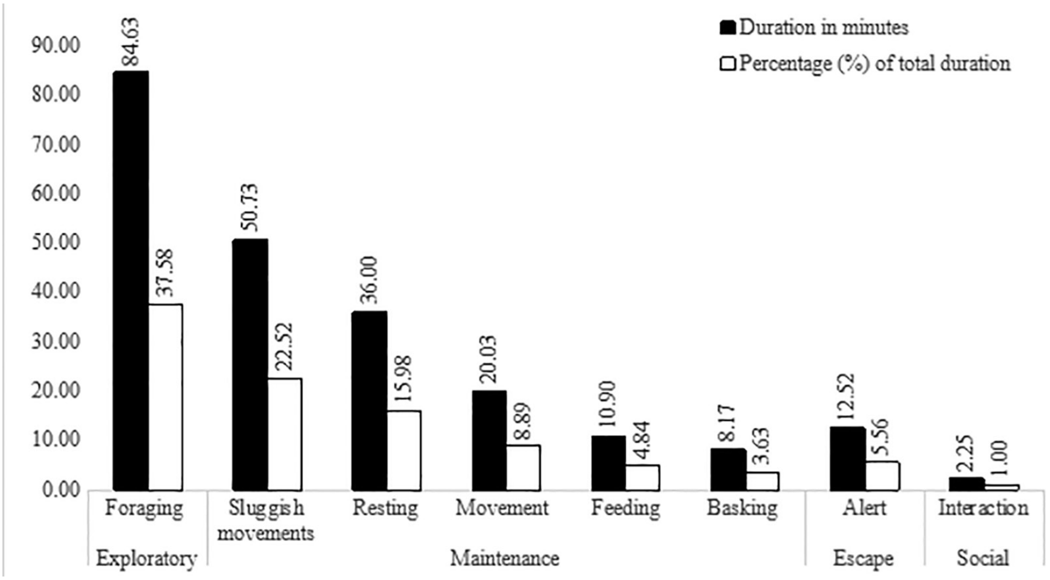


**Figure 3.** Duration of different behavioural activities of *Varanus salvator*

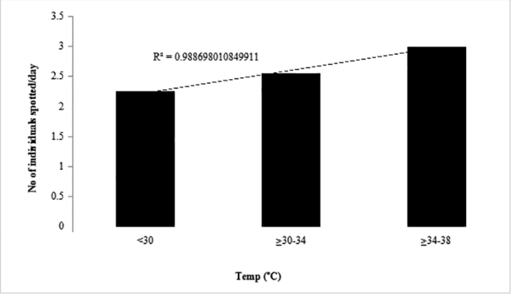


**Figure 4.** Number of individuals observed in different temperature intervals.

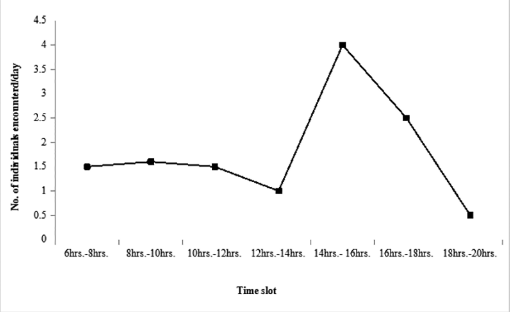


**Figure 5.** Activity of *Varanus salvator* spotted in different periods of a day.

*Exclusive Behaviour III: Run*

While escaping from anthropogenic disturbances or on coming in very close proximity to humans, monitors increase their pace of walking and move fast by raising the body off the ground. While running only certain part of their tail stays in contact with the ground surface.

*Exclusive Behaviour IV: Floating*

It implies swift locomotion in water or swimming which does not involve foraging. The spindle shaped body aids in efficient movement through water while the tail helps in steering or changing direction. While floating, only the head remains above water surface and portions of dorsal part of the body and the tail is visible.

*Exclusive Behaviour V: Head turn while in motion*

Swaying the head right and left to have a view of the surroundings while the monitor is either floating or walking.

*Exclusive Behaviour VI: Leap*

Jump into the air from a tree trunk or some other substrate towards the ground such that all four feet leave the substrate.

## Behavioural category III: Sluggish or restricted movements

The behavioural category sluggish or restricted movements deals with lazy unenergetic behaviours showing hardly any or very little motion and almost no locomotion.

*Exclusive Behaviour I: Stand still*

Stalling or showing no or very little movement for a considerable time. The lower part of the body i.e., from the neck up to the tail is in contact with the surface, while the neck is above the ground (but not fully stretched) and the monitor is gazing in a particular direction for quite some time (may extend up to 15 minutes) in the stand-still position.

*Exclusive Behaviour II: Throat expansion*

Motionless stalling is accompanied by gazing in a particular direction and expanding and contracting the throat at regular intervals; may be followed by a few tongue flicks.

*Exclusive Behaviour III: Yawn*

Throat expansion sometimes followed by a yawn i.e., opening the mouth wide and exposing the region inside the mouth. Individuals show such activity as a sign of being tired and wanting to rest. Mostly seen in the afternoon.

*Exclusive Behaviour IV: Jaw licking*

A yawn is followed by licking the upper and lower jaw using tongue. Just after a yawn the individual wipes its mouth and starts walking. After this the individual is not seen to be very active and either goes for basking or rests.

*Exclusive Behaviour V: Scrape*

Rubbing the mouth to a hard surface (for e.g., tree trunk or any solid substratum). Mostly observed in juveniles.

*Exclusive Behaviour VI: Head raise*

Raising the head lateral to the body to some height.

#### Behavioural category IV: Resting

*Exclusive Behaviour I: Lie flat*

The whole body is kept in contact with the surface while the monitor is lying motionless under the shade with the forelimbs directed backwards tucked close to the body and hind limbs placed sideways. The body looks even more bulky and D shaped. A monitor can rest for hours at a stretch lying in the same position with eyes open (lack of eyelids) while the nictitating membrane covers the eyes at times and opens again like a high-speed shutter of the camera.

*Exclusive Behaviour VII: Watching around*

Looking sideways by turning the head and neck while resting.

#### Behavioural category V: Basking

*Exclusive Behaviour I: Bask flat*

A monitor lying in the open with the dorsoventrally flattened body against the substrate with all the body parts touching the surface and often sunbathing (on the ground or on the trunk of trees) in this position.

*Exclusive Behaviour II: Head and neck stretch*

Basking while stretching the neck and moving the head upwards.

*Exclusive Behaviour III: Head rest*

Head and neck stretch are followed by head rest wherein the monitor resumes lying in a flat position while being under the sun.

*Exclusive Behaviour IV: Repositioning*

While basking the monitor changes its position frequently so that every part of the body is exposed to the sunlight equally.

### Functional category III: Escape

#### Behavioural category I: Alert

*Exclusive Behaviour I: Halt*

On land or in water continuous walk/swim is interrupted by pausing for a few seconds/minutes to observe their surroundings before continuing the walk.

*Exclusive Behaviour II: Flee*

On coming in close proximity to humans (or other disturbances), monitors increase their pace and move fast or change the direction of their crawl by raising the body off the ground and creating heavy thumping sound. Sometime the run is not continuous; it runs a certain distance, pausing to look behind in order to keep an eye on the individual.

### Functional category IV: Social

#### Behavioural category I: Interaction

*Exclusive Behaviour I: Chase away*

Smaller sized individuals of *V. salvator* are chased by the dominant and larger ones.

*Exclusive Behaviour I: Submissive*

A thrust of the body in water when chased by another individual of larger size, followed by the one that submits to the interaction getting displaced from that area. This involves direct one on one interaction.

### *Behavioural pattern*

Quantification of each behavioural category of the Asian Water Monitor reflects that in a day they devote most of their time (76.08% of the total activity duration) to three activities, foraging (84.63 mins, 37.58%), sluggish movements (50.73 mins, 22.52%) and resting (36 mins, 15.98%). Here all activities including resting or sleeping have been considered a behaviour.

Regardless of a massive time spent foraging, very little effort is transformed into feeding (10.9 mins, 4.84%). The monitors are very curious animals and hence remain alert (12.52 mins, 5.56%) mostly due to anthropogenic disturbances such as eye contact or sudden encounter with villagers while bathing in the pond. Basking (8.17 mins, 3.63%) is another common behaviour practiced by *V. salvator*, particularly in the late afternoon for a very limited period of time (Fig. 3). The duration of basking might be less but the sighting frequency is maximum in the afternoon. It is very evident from Fig. 5 that the sighting frequency of the monitor lizards are quite considerable throughout the morning i.e., 6 am–10 am, decreasing very steeply from 12 pm–2 pm and then peaking between 2 pm–4 pm, followed by a gradual drop in activity. It can be inferred from this figure that late afternoon is the best time for foraging since the monitors in the study sites are coexisting with humans. Human activity is low in late afternoon (2pm to 4pm) and increases between 12 pm – 2 pm and in the evening, when human activities like bathing, washing etc. increase. The period between 2 pm–4 pm is the time when the villagers show least activity and this is the time when the monitor lizards are the most active. Therefore, the monitor's activity time does not coincide with human activity time or shows very little overlap. Comparing daily temperature with the activity of monitor lizards (Fig. 4) showed a positive correlation ($r = 0.988$). Though it is not possible to draw any conclusions from this relation, given the limited sampling, the trend of increase in activity with temperature can be studied further. As the study was conducted in the non-breeding season of the animal, no mating behaviours were documented.

## Discussion

Preparing an ethogram is an introductory process for any kind of behavioural study. This study clearly reveals all the behaviours observed during the study tenure (total 39 exclusive behaviours clustered into eight behavioural categories and four functional categories) of *V. salvator* during their non-breeding period. The repertoire (along with the pictures) was an effort to describe every aspect of each behaviour of *V. salvator* observed during the brief study period in its wild habitat, which actually helps to understand their overall activity and resource utilization regime.

Among all the behaviours, the monitor lizards devote maximum activity time to foraging (84.63 minute, 37.58%). Their feeding behaviour reflects that these monitors are active hunters and swallow the prey entirely. Along with fishes, rat, and aquatic invertebrates, *V. salvator* was also found to hunt snakes for food. Sighting frequency followed by different behavioural activities of monitor lizards are maximum in the afternoon in both the study areas. Budgebudge area is a village, densely populated by human where anthropological activity is less in the afternoon as compared to mornings and evenings. Meanwhile, CKBS is situated just beside a broad metallic road where the frequency of cars passing by this area is less in the afternoon. The study also highlights the fact that the activity of *V. salvator* increases with increase in atmospheric temperature.

This ethogram clearly depicts the overall behavioural patterns of the species in its non-breeding period and the reasons behind their survival in a highly congested human habitat. It is to be noted that observations made on field might be altered by the presence of the observer or interactions such as eye contact between the animal and the observer. Also, we note the incomplete nature of the present ethogram since we undoubtedly failed to document all the behaviour exhibited by *V. salvator* during the non-breeding period. Though the study primarily aimed at understand the behavioural differences in the two sites having different characteristics, there were no significant differences observed and it is difficult to draw any conclusion from the limited sample size (observation time of each behaviour) and time. Hence, the behaviour data from both the sites were pooled for consistency in the statistical analysis. With a larger sample size and a greater input of time one can draw concluding results on the differences in behaviour amongst the two sites and also amongst different age groups in *V. salvator*.

Eating snakes (both venomous and non-venomous) in the villages and controlling their population enhances the credibility of monitors in front of the local villagers, because according to the popular belief venomous snake is more harmful to human rather than monitor lizards. This positive existence value helps them co-exist with the humans in these localities. Apart from increasing the activity in the afternoon, it separates its niche from humans and avoids any serious conflicts. This study was conducted under a time constraint, but still gives a wholesome idea about their behaviour during the non-breeding season and the reason behind their sustainability in these highly populated human altered ecosystems.

## Acknowledgement

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### A case of radio tag recovery from *Nanorana vicina* (Stoliczka, 1872) (Anura: Dicroglossidae)

**CITATION.** Nawani, S., Banerjee, K. and Das, A. (2022). A case of radio tag recovery from *Nanorana vicina* (Stoliczka, 1872) (Anura: Dicroglossidae). *Hamadryad: 39, 69–71*.

Radio telemetry is used to study habitat use, movement pattern and migratory behaviour in anurans (Tatarian 2008; Ludwig et al. 2013; Pitt et al. 2017), and has been thoroughly experimented with, by using different techniques of tag attachments (Goldberg et al. 2002; Muths 2003; Watson et al. 2003; Leskover & Sinsch 2005; Berg et al. 2010). One of the constraints in anuran telemetry is the size of the transmitter concerning its battery life, which necessitates recovering the tag before signal loss and exhaustion of battery life. Moreover, habitats of some anurans contain potential snags such as vegetation, coarse woody debris that might get entangled in the waistbands and even the antennae, causing dislodgment of the transmitters (Groff et al. 2015). Studies have reported the recovery of functional radio tags shed by animals (Rathbun & Murphey 1996; Muths 2003; Forester et al. 2006; Sepulveda & Layhee 2015; Cooper 2017; Tabata et al. 2018). However, recovery of nonfunctional radio tags retained by the animals has not been documented thus far.

We used this technique to understand the ecology of *Nanorana vicina* (Stoliczka, 1872), commonly known as Stoliczka's torrent frog, adapted for life in mountain streams in the Western Himalayas. This stream-breeding dicroglossid frog is uniquely adapted to pool sections of the streams, as observed during our survey in Benog Wildlife Sanctuary, Uttarakhand (28° 60´ - 31°28´ N, 70°49´ - 80°60´ E). We radio-tagged and tracked a total of 16 individuals of *N. vicina* over a period of five months (May to September 2019). Altogether, 11 radio transmitters (SOPR- 2070, Wildlife Materials Inc, US) were used in this study, out of which, seven tags were attached to seven different individuals and the rest of the four transmitters were reused on nine

**Figure 1.** A radio-tagged *Nanorana vicina*.

different individuals over a different period. The transmitter has a battery body (Battery life — 134 days) and a whip antenna attached to the device. We used a very fine and stretchable Teflon tube with a nylon thread inside the radio tag through the crafted fine hole present on the tags. Body weight and snout-vent length of the tagged frogs were recorded before and after tagging. Body weight of individuals ranged from 115–190 gm (Average = 150 gm, n = 16). The tags were carefully tied to the groin region of the frogs with a harness weighing 3.5 gm (2.3% of average body weight) maintaining the consideration that it must not exceed 5% of the body weight of the frog (Goldberg et al. 2002). The transmitter frequency range (150.000 to 150.480) of the tags and individual IDs of each tagged frog were noted for further monitoring. The frogs were kept overnight under controlled conditions before they were released at the same area of capture. Throughout the course of our study, we have successfully recovered five nonfunctional tags within the span of five months (June to October) in the year 2019; however, the rest of the six tags remained missing due to the transmitter's signal failure.

On 27 April 2021, we manually recovered two more nonfunctional transmitters with intact radio belts tied to individuals. However, we did not find the remaining four tags. These two recovered tags were attached on 16 July and 8 August 2019. Subsequent examination revealed no potential signs of abrasion or lesion on the recovered frogs even after a long period of attachment to the transmitters. The individuals were seen within a 20 m radius from their initial place of tagging. To our knowledge, this is the first-ever report of such incidence taking place where the tags were recovered after a span of 21 months, even when they were nonfunctional.

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## A note on the larval stage of the Sikkimese caecilian, *Icthyophis sikkimensis* Taylor, 1960

**CITATION.** Mondal, S., Bhattacharya, S. and Deuti, K. (2022). A note on the larval stage of the Sikkimese caecilian, *Icthyophis sikkimensis* Taylor, 1960. *Hamadryad*: *39, 71–75.*

**KEYWORDS.** Caecilian, Developmental stages, Transitory stage, Sensory tentacles, Nuchal collars, Tail fin, Annuli

Among the three modern amphibian orders, Gymnophiona (caecilians) are the least known (Zhang & Wake 2009). Caecilians are readily distinguished from members of Anura (frogs and toads) and Caudata (salamanders and newts) by their elongated, annulate and limbless bodies. Gymnophiona are found in the moist tropical and proximate temperate regions of South-east and South Asia, Africa, the Seychelles, Central and South America (Zhang & Wake 2009). Among the 215 currently recognised extant species under 10 families, the genus *Ichthyophis* is the most speciose, with 50 species (April 10, AmphibiaWeb 2022)

The Sikkimese caecilian, *Ichthyophis sikkimensis* Taylor, 1960, was described on the basis of a holotype and three paratypes from the Indian states of Sikkim and West Bengal. The species is known to occur in Sikkim and Darjeeling regions of West Bengal and the Ilam district of eastern Nepal (Taylor 1960; Sarkar et al. 1992; Pillai & Ravichandran 1999; Anders et al. 2002; Shah & Tiwari 2004; Kamei et al. 2009; Kamei 2017; Frost et al. 2022). *Ichthyophis sikkimensis* is classified as Data Deficient (DD) in the IUCN Red list of Threatened species because of "ongoing uncertainties as to its extent of occurrence, status and ecological requirements" (Ohler et al. 2004).

During a field survey conducted between 15–21 July, 2021 at Kalikhola Latpanchar, Darjeeling district, West Bengal State, a caecilian was found while searching for earthworms under wet stones on sandy-gravel substratum beside a running stream inside a forest at Kalikhola (26° 44′ 34″ N, 88° 34′ 22″ E). The specimen was collected by Md. Nurul Hassan on 19.07.2021 on a rainy day during peak monsoon season and euthanised by injecting 70% ethanol into the heart, then washed with water and preserved for two days in 4% formaldehyde solution. Later it was washed in running tap water and transferred to 70% ethanol for final preservation and deposited in the collections of the Zoological Survey of India as ZSI A 15401. The specimen was identified as a larva of *Ichthyophis* because of the presence of a spiracle and tail fin and also because the tentacle had not erupted. There are no published keys to ichthyophiid larvae, but the specimen is tentatively identified as *I. sikkimensis* (Taylor 1960) because it is a late-stage larva that lacks lateral yellow stripes. Measurements were taken with a Mitutoyo$^{\text{TM}}$ digital calliper to the nearest 0.1 mm under a Leica EZ4 stereo binocular microscope and a Lensel magnanoscope.

The developmental stages of *Icthyophis sikkimensis* is poorly known, with no published description of the larvae. For the first time, we present information on the larval morphology

**Table 1.** Morphometric and meristic data for the larvae of *Ichthyophis sikkimensis* from Darjeeling district, West Bengal. Measurements to the nearest 0.01 mm.

| Characteristics | ZSI A 15401 | ZSI 19049 A | ZSI 19049 B | ZSI 19049 C |
|---|---|---|---|---|
| **Snout to vent length** | 120.0 | 144.0 | 131.0 | 103.0 |
| **Tail length** | 2.3 | 1.1 | 2.2 | Damaged |
| **Head length** | 4.0 | 3.0 | 3.4 | 3.1 |
| **Head width** | 4.2 | 3.9 | 3.5 | 3.4 |
| **Snout length** | 3.1 | 2.1 | 2.4 | 2.3 |
| **Eye to nostril distance** | 2.0 | 1.9 | 2.0 | 2.0 |
| **Eye to corner of mouth** | 1.6 | 1.1 | 1.5 | 1.6 |
| **Nostril to snout tip distance** | 1.2 | 0.4 | 0.7 | 0.6 |
| **Inter-narial distance** | 2.0 | 1.5 | 1.9 | 1.6 |
| **Inter-orbital distance** | 3.1 | 3.2 | 2.3 | 2.3 |
| **1st nuchal groove distance from the tip of the snout** | 7.1 | 6.9 | 8.4 | 6.3 |
| **2nd nuchal groove distance from tip of the snout** | 8.9 | 10.1 | 10.1 | 8.5 |
| **Anterior tip of the vent to the tail tip** | 4.1 | 2.8 | 3.7 | Damaged |
| **Distance between two annuli on the dorsal part of the body** | 0.9 | 0.4 | 0.5 | 0.4 |
| **Length of the cloacal slit** | 1.3 | 1.1 | 1.5 | 1.1 |
| **No. of Annuli** | 287 | 286 | 286 | 275 |
| **Tail structure** | Tail tip more rounded, tail fin less broad, curved dorsally | Tail tip more rounded, tail fin less broad | Tail tip more rounded, tail fin less broad | Tail tip more rounded, tail fin less broad |
| **Gill structure** | Not elongated, small pin-head shaped, not deep opening | Elongated, deep opening | Elongated, deep opening | Elongated |
| **Mechanosensory organs on mandible** | Thick white dots with small central grooves, considerably larger and circumscribed by a dark circular groove | Thick white dots | Thick white dots | Thick white dots |
| **Nasal opening** | Triangular shaped, frontal nasal opening | Circular shaped, frontal nasal opening | Circular shaped, frontal nasal opening | Circular shaped, frontal nasal opening |
| **Glandular papillae** | Present on chin | Absent | Absent | Absent |

of *I. sikkimensis* along with a brief comparison with the juvenile of *Ichthyophis kohtaoensis* (Dünker et al. 2000) and late larval stage and juvenile of *Ichthyophis glutinosus* (Breckenridge et al. 1987). The head of ZSI A 15401 is flattened, snout rounded in front, the mouth has well-developed labial folds typical of ichthyophiid larvae. The small eyes are covered with semi-transparent skin. Breckenridge et al. (1987) reported that the gills of hatchlings of *I. glutinosus* fall off just two days after hatching. ZSI A 15401 lacks any sign of gills, and a single spiracle is visible on each side laterally just behind the head (Figure 1a). Sensory tentacles have not yet erupted. Mechanoreceptive sense organs on the head are visible as minute white spots. Two partially developed collars are visible on the ventral side only (Figure 1b). A short

**Figure 1.** *Ichthyophis sikkimensis* larvae ZSI A 15401 from Kalikhola, Latpanchar, Darjeeling district, West Bengal (26◦ 44′ 34″ N, 88◦ 34′ 22″ E). a. Lateral view of head, b. Ventral view of head, c. Lateral view of the tail fin, d. Ventral view of the tail fin.

laterally compressed tail fin is present (Figure 1c & d) In *I. glutinosus,* the transition from late larval to "juvenile" stage is marked by the gradual disappearance of the tail fin (Breckenridge et al. 1987). The annular folds of the skin are not clearly demarcated, making a count of the folds very difficult, as is typical of ichthyophiid larvae (e.g., Breckenridge et al. 1987). In ZSI A 15401 the number of annuli is approximately 287. Snout-vent length is 120 mm and tail length is 2.3 mm.

After studying the specimen, we have come to the conclusion that ZSI A 15401 is in the stage 38 of Dünker et al. (2000) because mechanosensory organs are present on the mandible as thick white dots with small central grooves, considerably larger and circumscribed by a dark circular groove; glandular papillae present on the chin; triangular shaped, frontal nasal opening; tail tip more rounded, tail fin less broad, curved dorsally. Therefore, we conclude that due to absence of gills (which is regressed forming only a tiny aperture), presence of a reduced tail fin and yet absence of any sensory tentacles, this specimen is in a transitory stage between the late larva and small metamorphosed stage of *Ichthyophis sikkimensis.*

We also examined three previously collected larvae (ZSI 19049A, B, C) from Darjeeling district of West Bengal state, India by S.W. Kemp in 1918 (Pillai & Ravichandran 1999). These larvae are in the stage 37 of Dünker et al (2000) because mechanosensory organs are present on the mandible as thick white dots; gills are stripped off, no external gills and opening to gill chambers are deep and elongated; tail tip more rounded than arrow-shaped, tail fin less broad and no longer delineated.

Although Pillai & Ravichandran (1999) also mentioned ZSI 19334 and ZSI 19335 as larvae of *Icthyophis sikkimensis* collected by F.M. Bailey ("Barley" in Pillai & Ravichandran [1999]) from Gangtok, Sikkim on 23.i.1922, we however found that ZSI 19334 is an adult while ZSI 19335 is probably lost. Further collections and comparative studies of different developmental stages of *I. sikkimensis* will enhance our knowledge about the life history of this interesting caecilian in future.

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# A report on ophiophagy observed in *Naja oxiana* (Eichwald 1831) from Himachal Pradesh, India

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**KEYWORDS.** Elapidae*,* diet, Himalayas*, Elaphe hodgsoni,* Serpentes

The central Asian cobra *Naja oxiana* (Eichwald 1831) is a species of venomous snake in the family Elapidae that ranges from the Transcaspian region of central Asia all the way down to India (Smith 1943; Wall 1911; Wüster & Thorpe 1992; Wallach et al. 2009). The presence of *Naja oxiana* in Himachal Pradesh was recently confirmed by Santra et al. (2019). Despite a seemingly wide distribution, there is a broad gap in our knowledge of the natural history of the species. There are no published records of its dietary composition, except a broad mention that it feeds on amphibians, lizards, and small rodents (Whitaker & Captain 2008). Here we present the first verified observations to our knowledge of ophiophagy in *Naja oxiana.*

The first instance was observed in Jukyani village (32°51’32”N, 76°08’41”E, 1624 m a.s.l.), situated in Chamba district of Himachal Pradesh. On 7 July 2020 at 17:25 h, a *Naja oxiana* was found in a villager’s house during a rescue operation. The snake measuring a total length of ca. 119 cm was observed feeding on

**Figure 1.** *Naja oxiana* returning to the dead *Elaphe hodgsoni.*

**Figure 2.** *Naja oxiana* feeding on *Elaphe hodgsoni.*

an *Elaphe hodgsoni* (Günther 1860). During capture, the snake regurgitated the specimen of *E. hodgsoni,* which measured a total length of ca. 92 cm.

The second instance was observed near Chamara village (32°50’13”N, 76°08’40”E, 1624 m a.s.l.), situated in Chamba district of Himachal Pradesh, 2.37 km away from Jukyani. On 15 August 2020, at 14:21 h, an adult *N. oxiana* was observed feeding on an *E. hodgsoni*. The anterior portion of *N. oxiana* was hidden behind a stone with one third of the prey snake already swallowed. The cobra regurgitated the prey as it was disturbed by a crowd that had gathered and entered a burrow nearby. The snake returned to the dead *E. hodgsoni* after a few minutes, but was disturbed by the crowd again and moved away.

The range of *E. hodgsoni* and *N. oxiana* overlaps in Himachal Pradesh, as well as in Jammu and Kashmir in India. It is likely that *E. hodgsoni* might form a part of the diet of the latter in this range. Further studies are necessary to ascertain if *Naja oxiana* feeds exclusively on serpents, or includes snakes as part of a broader range of prey items. The present report is noteworthy as it adds to our understanding of an otherwise poorly known elapid species.

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# Additional locality reports of *Calliophis castoe* Smith, Ogale, Deepak & Giri, 2012 (Squamata, Elapidae) from the Western Ghats, India

**CITATION.** Praveen, HN., Badiger, R., Zarmekar, R. and Kamble, S. (2022). Additional locality reports of *Calliophis castoe* Smith, Ogale, Deepak & Giri, 2012 (Squamata, Elapidae) from the Western Ghats, India. *Hamadryad*: *39, 76–79.*

**KEYWORDS.** Reptilia, Elapidae, Coral snake, *Calliophis castoe*, Range Extension

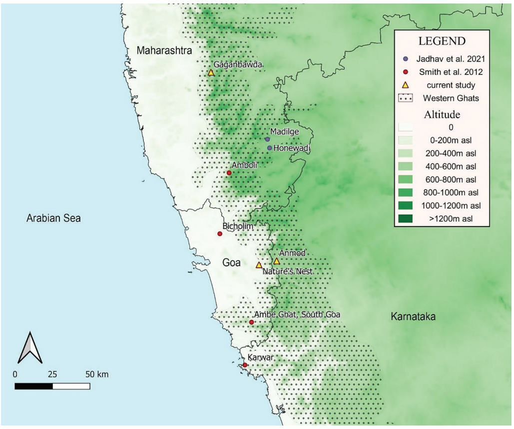

**Figure 1.** Map showing known and new localities of *Calliophis castoe*

The genus *Calliophis* Gray, 1834 currently has 15 recognised species distributed throughout the oriental region, of which five occur in India (Uetz et al. 2021). *Calliophis beddomei* has been previously reported from Shevaroy hills in the Eastern Ghats, Mudumalai and Koppa in the central Western Ghats, and Kallar in the southern Western Ghats (Jins et al. 2014); *C. bibroni* is known to be present along the Western Ghats range with Agumbe hills marking the northern limit, to Agasthiyar hills in Kerala (Deepak et al. 2010); *C. melanurus* has been reported from multiple localities across the Indian peninsula and Sri Lanka (Whitaker & Captain 2004; Shine & Nameer 2012); and *C. nigrescens* distributed across the Western Ghats is sympatric with *C. castoe* in Goa (Smith et al. 2012). Rarely seen, these are some of the least known snakes in India.

Smith et al. (2012) described a new species of coral snake, *Calliophis castoe* from the Western Ghats, based on three specimens from three different localities, namely Amboli, Maharashtra; Amba Ghat, South Goa and Karwar, Karnataka. They also reported the species from Dicholi (Bicholim), North Goa. Subsequently, this species has been reported from Cotigao Wildlife Sanctuary, Goa (Khandekar et al. 2021) and also recently reported from Madilge and Honewadi, Maharashtra (Jadhav et al. 2021). It is an uncommon species with limited information on its natural history, except for a few anecdotal reports suggesting its fossorial habit (Smith et al 2012). We report the species from Kolhapur district (Maharashtra), South Goa district (Goa) and Uttara Kannada district (Karnataka).

On 28th June 2020, SK reported an individual in Sangashi (16.5614° N, 73.8595° E; altitude 630m asl) near Gaganbawada, Kolhapur district, Maharashtra. This new locality is approximately

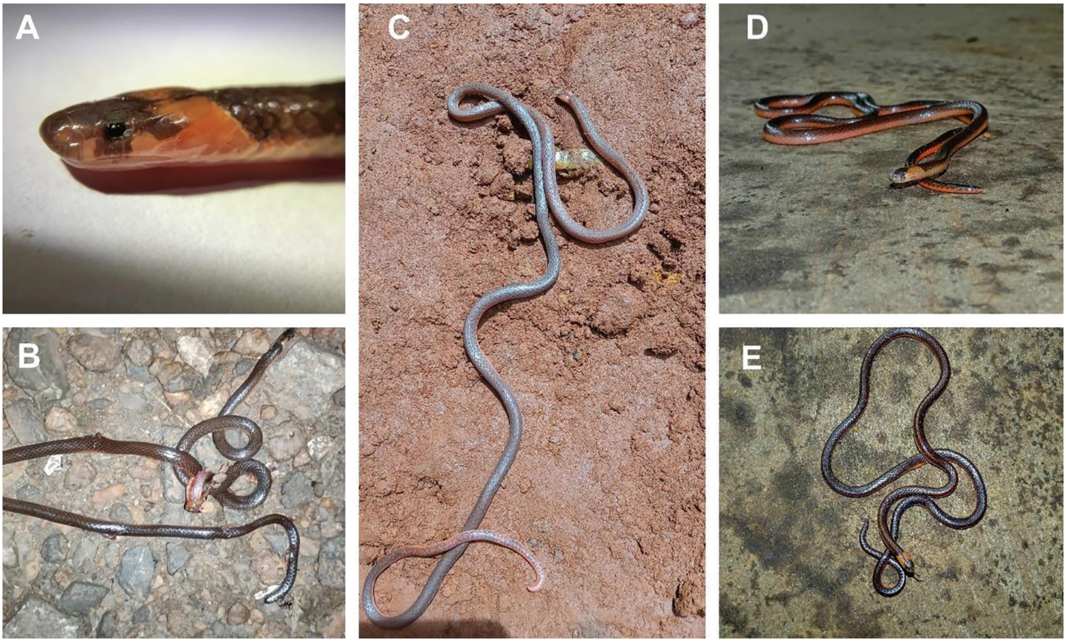

**Figure 2.** A) and B) Road-killed *C. castoe* specimen from Anmod, Karnataka; Picture credit: Ramesh Badiger. C) *C. castoe* specimen from Gaganbawada, Maharashtra Picture credit: Sachin Kamble. D) and E). *C.castoe* specimen from Goa; Picture credit: Ramesh Zarmekar

70 km (straight line distance) north of the type locality of this species, Amboli. It was identified as *Calliophis castoe* based on the unicoloured, unpatterned dorsum, slender body, orange band on the head, and the underside of the tail which was uniformly orange. This individual was ~80 cm in total length. It was seen approximately two feet below the ground in loose soil while digging a pit in the home garden. Typical tail curling behaviour of *Calliophis* was not seen.

The second individual was seen at Nature's Nest, Sacordem, Dharbandora taluka, South Goa district, Goa (15.411256, 74.200198; altitude 60 m asl). The nearest known locality is Bicholim, North Goa, which lies ~31 km aerially from Sacordem. It was examined and identified as *Calliophis castoe* based on the unicoloured, dark dorsum and prominent orange band on the head. It was then photographed and released. From snout to tail-tip, it measured 56 cm, and the specimen did not display any form of aggression or tail curling behaviour when handled.

One road-killed specimen was observed by RB at Anmod (N 15.438602, E 74.309182; altitude 597 m asl), Karnataka on 31 October 2020. This locality is ~45 km aerially from the nearest known locality of occurrence (Ambe Ghat, South Goa). This road-killed specimen was fresh, and seen on the main road passing through the semi-evergreen forest near the forest check post. Morphological data of this specimen was taken before it was left in the forest. We followed Smith (1943) for dorsal row scale count. These dorsal rows were counted two head lengths behind the head, at midbody, and two head lengths before the vent. Dorsal scales were in 13:13:13 rows. We followed Dowling's (1951) method to count ventral scales and they were 242, divided subcaudals 36, supralabials 7, 3 and 4 touching the eye, preocular 1 and post oculars 2 on both sides. It had a total length of 60 cm and colouration was as mentioned in Smith et al. (2012).

The Western Ghats, India, is home to five species of coral snakes of the genus *Calliophis* (Uetz et al. 2021). Among these, *Calliophis nigrescens* is one of the widely distributed species and known from Gujarat, Maharashtra, Goa, Kerala, and Tamil Nadu (Whitaker and Captain 2004). Although a widely distributed species, it is a poorly known species in terms of ecology and natural history. Although considered terrestrial, coral snakes are best at burrowing. Thus, the apparent rarity of these coral snakes would be directly associated with their fossorial nature. *Calliophis castoe* is also a poorly known species. The only information about their habitat and behaviour is based on a single live individual collected from Amba Ghat, Goa (Smith et al. 2012). This individual was seen among the leaf litter close to the road. Our observation based on a live specimen at Gaganbawada, Maharashtra also suggests that this species is burrowing. The observation of a live individual from Sacordem, Goa indicates their nocturnal habits. Our report of a road-killed specimen from Anmod, Karnataka assures the presence of this species in this landscape.

For the effective conservation assessment of a poorly known taxon like *Calliophis,* information about the distribution and natural history is vital. Hence, we take this opportunity to report the occurrence of *Calliophis castoe* from additional localities and a few notes on their habitat and natural history.

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## Evidence towards antipredatory behaviour by Indian Spiny-tailed lizard *Saara hardwickii* (Gray, 1827) towards Indian Red sand boa in Desert National Park, India

**CITATION.** Jangid, A.K., Tripathi, R., and Daniel C.G. (2022). Evidence towards antipredatory behaviour by Indian Spiny-tailed lizard (*Saara hardwickii* GRAY 1827) towards Indian Red sand boa in Desert National Park, India. *Hamadryad: 39, 79-82.*

**KEYWORDS.** Arid, Defensive strategy, Natural history, Tail spination, Thar desert.

The Indian Spiny-tailed lizard *Saara hardwickii* (Gray 1827) is the only herbivorous lizard in the Indian subcontinent. Earlier reported to be widespread, the current known largest population density of this species is confined to the arid tracts of India, which are in Kutch district in Gujarat (Dutta & Jhala 2007), and Jaisalmer district in Rajasthan (Ramesh & Ishwar 2008; Kaur et al. 2020). These lizards have an average snout-to-vent length of 175 mm, with blunt snouts, dorso-ventrally flattened bodies and thick tails that are covered with hard spiny ornamented scales (Smith 1935). Locally known as "*Sanda*", these diurnal lizards excavate long curved burrows with an elliptical mouth (Figure 1a) in gravel plains, which leads into a tunnel that gradually slopes downward (Ramesh & Sankaran 2013). They live in clustered colonies with multiple burrows scattered in close vicinity (Abdulali 1960; Minton 1966) and close their burrows with sand soon after the sun sets or during rains (Figure 1b). The species represents an important prey base in this particular landscape as they are fed upon by the Great Indian Bustard *Ardeotis nigriceps,* Laggar Falcon *Falco jugger*, Tawny Eagle *Aquila rapax*, Steppe Eagle *Aquila nipalensis*, Short-toed Snake Eagle *Circaetus gallicus*, Cattle Egret *Bublcus ibis*, Indian Fox *Vulpes bengalensis*, Desert Fox *Vulpes Vulpes pusilla*, Golden Jackal *Canis aureus*, Bengal monitor Lizard *Varanus bengalensis,* Desert Monitor Lizard *Varanus griseus*, and Red Sand Boa *Eryx johnii,* to name a few (Gupta 1975; Dutta & Jhala 2007; Pardeshi et al. 2008; Home & Jhala 2009; Maurya et al. 2009; Jhala et al. 2012; Ramesh & Sankaran 2013). *S. hardwickii* is 'vulnerable to extinction' (Molur & Walker 1998) owing to the decline in population due to loss of habitat and poaching pressure (as a result of superstitious medicinal properties of the lizard's body fat; Ramesh & Ishwar 2008).

The Red Sand Boa *Eryx johnii* (Russell 1801), of the family Boidae, are a widely distributed species found throughout the Indian subcontinent, and are common within the limits of Desert National Park Wildlife Sanctuary in Rajasthan (Whitaker & Captain 2004; Uetz et al. 2020). *E. johnii* is a nocturnal boa species, which inhabits burrows in dry and sandy soil found across Peninsular India (Marimuthu & Asokan 2014). An adult individual of *E. johnii* lengths between 600–1200 mm. The species has been reported to feed on rodents, birds, lizards, and insects (Whitaker & Captain 2004; Sharma 2007; Ingle 2011). Though this species has been observed to exit abandoned *S. hardwickii* burrows (Ramesh & Sankaran 2013), no direct observation of any defence strategy used by the lizard has been reported.

On September 22, 2017 at 21.18 hours, within the compound of the forest department check post at the Desert National Park Wildlife Sanctuary at Sam village [26.83526°N 70.50914°E; ca. 225 m a.s.l.], Rajasthan, the authors spotted a Red Sand Boa trying to enter the burrow of *S. hardwickii* (Figure 2a, b). The lizard was observed to have its entire upper body (Snout-to-vent) inside the burrow, with only its spiny tail protruding out of the burrow (Figure 2c, d). The snake made multiple attempts to enter the burrow, but was never seen biting the tail of the defensive lizard. The lizard also made several whip-like swipes with its tail, presumably to deter or injure the predator trying to enter. We hypothesize that the spiny ornaments on the tail of the lizard adds an additional tier of difficulty for the predator to push past the burrow opening.

**Figure 1.** a) Elliptical mouth of burrow of Spiny-tailed lizard *Saara hardwickii* (Gray, 1827); b) Closed mouth of burrow of Spiny-tailed lizard

**Figure 2.** a) & b) Red Sand Boa *Eryx johnii* trying to dig out the sand from Spiny-tailed lizard's burrow; c) & d) Defensive tail whipping action used by Spiny-tailed lizard.

The entire predatory episode lasted about three hours and prolonged well into the night.

There have been multiple speculative discussions suggesting the defence benefits offered by the spiny tail of *S. hardwickii* (Ramesh & Sankaran 2013; Hashmi et al. 2014), but no study has so far provided evidence confirming the use of such a defence strategy. Therefore, we report the first evidence to support the defensive strategy employed using the spiny tail of *S. hardwickii* against *Eryx johnii.*

The authors have in multiple instances observed half eaten bodies of the lizard discarded across the landscape with the tails totally intact suggesting that predators avoid consuming the heavily ornamented tail, and that the tail ornamentations are an evolutionarily selected anti-predatory defensive trait.

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## Anurophagy in *Indirana chiravasi* (Anura: Ranixalidae) from Goa, India

**CITATION.** Shinde, A., Joshi, P., Velguenkar, A. P. and Shinde, G. (2022). Anurophagy in *Indirana chiravasi* (Anura: Ranixalidae) from Goa, India. *Hamadryad: 39, 82-85*.

**KEYWORDS.** Anurophagy, Diet, Feeding, Ranixalidae, Leaping frog, Amphibian

The Western Ghats endemic anuran genus *Indirana* Laurent, 1986 is presently constituted of 14 species (Frost, 2022). Members of this genus are small to moderate-sized, bear a large and distinct tympanum, Y-shaped terminal phalanges, and extensive to moderate webbing between the toes (Modak et al. 2015). This ancient anuran lineage is known to have an unusual reproductive mode (Gaitonde & Giri 2014) and larval development (Annandale 1918). The group also shows cryptic diversity (Nair et al. 2012; Ramesh et al. 2020). Modak et al. (2018) described *Indirana leithii* Boulenger, 1888 as primarily insectivorous after finding large proportions of insect appendages in the gut; they also reported the presence of arachnids, annelids, and a large amount of plant material in the diet of post-metamorphic *I. leithii*. Subsequently, Kulkarni et al. (2020) revealed the presence of cannibalistic tendencies in *Indirana leithii*. The diet composition of other members of *Indirana* is presently unknown and is poorly studied. We herein report an instance of anurophagy in *Indirana chiravasi* Padhye, Modak, and Dahanukar, 2014, a quite distantly related congener of *I. leithii* (Padhye et al. 2014).

March is a dry period in Goa with relatively low frog activity. During day surveys we came across bush frogs (mainly *Pseudophilautus* sp.) perched on tree branches, bushes, and also on the ground; and other individuals of *Indirana chiravasi* and *Minervarya* sp. along the stream and in the adjoining riparian zone. On 2 March 2021 at 0925 h, an adult *Indirana chiravasi* of unknown sex was found under a laterite rock along a dried-up stream near Mollem village, Goa (15°22’22.6”N 74°13’03.2”E). The individual was put into a plastic jar for further observations, where it regurgitated a partially digested bush frog about 10 minutes after its collection. We suspect the ingestion occurred the previous night and although unlikely, we do not rule out the possibility of this being a scavenging event. The predator and its prey were carefully removed and placed on a rock for photography (Figure 1). Due to the unavailability of instruments, no attempt was made to measure the size and weight of the specimens. The *Indirana* stayed at this spot near the bush frog

**Figure 1.** *Indirana chiravasi* and the regurgitated prey (bush frog). Photo by Varad Giri

carrion for a long time and did not attempt to eat it. The regurgitated frog was then transferred to a leaf to take pictures of its dorsum (Figure 2). The *Indirana*, a few minutes after regurgitating the carrion, hopped away on its own accord.

The predator was identified as *Indirana chiravasi* based on the following set of morphological characters: dark brown dorsum, head longer than wide with a brown stripe running from the tip of the snout to shoulder, distinct canthus rostralis, moderate webbing, and elongated inner metatarsal tubercle (See Padhye et al. 2014). Despite the specimen matching the descriptions of *Indirana salelkari* Modak, Dahanukar, Gosavi, and Padhye (2015) as well, we would like to retain the identification as *Indirana chiravasi* Padhye, Modak, and Dahanukar, 2014, which was reported from Bondla Wildlife Sanctuary, Goa (ca. 13 km from Mollem aerially) by Ramesh et al. (2020) under the voucher ID, CESF2288. *Indirana salelkari* on the other hand has been confirmed only from its type locality (Tanshikar Spice Farm, Netravali), which lies 33 km from Mollem aerially. Modak et al. (2015) did not sample their new species from other locales in Goa, nor did they provide distribution limits for *I. chiravasi* and *I. salelkari.* Garg and Biju (2016) further tagged three specimens collected from Jog falls, Unchalli falls, and Dandeli, all from the state of Karnataka as *Indirana salelkari*. Therefore, we assume this species' distribution to be stretching southwards from its type locality. The partially digested prey was identified as a bush frog using the following set of morphological characters: thin and slender hindlimbs, dilated digit tips, webbing between the toes, and granular belly.

Measey et al. (2015) found anurophagy in frogs to be more common than previously perceived, and to be influenced by the synergy of species diversity in habitat, invasiveness potential of an anuran, and its body size. The observation reported by Kulkarni et al. (2020) might be an outcome of increased density of foraging frogs during breeding (in Matheran) leading to competition among conspecifics for space and food. A similar report made by Modak et al. (2018) during the monsoon, suggests that cannibalism or even anurophagy is not a common event in the *Indirana leithii* populations of Matheran. Our observation, if not a scavenging event, could presumably be a case of "opportunistic predation" (as defined by Toledo et al. 2007), wherein the predator (*Indirana chiravasi*) had a relatively larger body and gape size than the bush frog. No concrete conclusions can be made without understanding the species' diet composition across seasons; this report stands to be the first diet record of *Indirana chiravasi*. Studies on the diet of *Indirana* frogs are essen-

**Figure 2.** Bush frog with partially digested and smudged dorsum. Photo by Varad Giri.

tial to understand the spectrum of prey items involved. Isolated natural history reports are often considered trivial and less 'impactful', but they go a long way toward developing an overall understanding of the species' ecology. Documentation of a species' diet, predation, reproduction, morphology, habitat use, behaviour, time of activity and so on — in short, natural history notes — have been found to be extremely valuable in macroecological studies and systematic reviews (Maritz et al. 2021).

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## Confirmation of *Liopeltis rappi* (Günther, 1860) from Himachal Pradesh, India

**CITATION.** Kuttalam, S., Santra, V., Owens, J.B., Ahmed, M.T., Bharthi, O., Selvan, M., Mukherjee, N. and Malhotra, A. (2022). Confirmation of *Liopletis rappi* (Günther, 1860) from Himachal Pradesh, India. *Hamadryad: 39, 85-88*.

**KEYWORDS.** Himachal Pradesh, Stoliczka, Liopeltis, rappi, Chamba

*Liopeltis* Fitzinger, 1943, is an Asian genus consisting of small, slender, and non-venomous colubrid snakes. It is characterized by a cylindrical body bearing smooth scales in 13, 15 or 17 rows, without the presence of apical pits. Ventrals are rounded and the tail is long with paired subcaudals. These snakes do not exceed 800 mm in total length (Smith 1943). Smith placed the members of the genus in two distinct groups, one with the head distinct from the neck and midbody scales uniformly in 15 rows, and the second with the head not or scarcely distinct from the neck, a dark bar across the neck, and nostrils large, between two nasals. Currently, *Liopeltis* is represented globally by six species, four of which are present in India (Poyarkov Jr et al. 2019).

*Liopeltis rappi* (Günther 1860) is distributed sporadically across the Himalayan region in India and Nepal (Bhattarai 2017). It was described by Günther (1860) from two specimens found in Sikkim and Nepal. It was reported from India thereafter by Stoliczka (1870) from Shimla, Himachal Pradesh; Anderson (1871) from the Jurta Valley, Darjeeling; Boulenger (1890) from Sikkim and Darjeeling; Wall (1909) from Peshok and Tindharia, Darjeeling; and Chettri et al. (2011) from Sikkim. Smith (1943) and Anderson (1871) both mention that the snake is not uncommon in the Darjeeling area. Bhattarai et al. (2017) provide a list of reported localities in eastern and central Nepal.

Stoliczka's 1870 report of a specimen from the "neighbourhood of *Simla*" (Shimla, Himachal Pradesh) is the only known record of this species in the Western Himalayas, although it has subsequently been included in checklists of the state herpetofauna on this basis (Saikia et al. 2007). Smith (1943) believed that the locality given for the Shimla specimen may have been an error since no additional specimens had been reported from the Western Himalayas since, and that remained true until August 2018, when we collected a fresh road-killed specimen of *L. rappi* in Bhanjraru, Chamba district, Himachal Pradesh (N 32º53.945' E 076º09.123'), at an elevation of 1690 m. The habitat that surrounded the highway the specimen was found on was mainly dense pine forests with a shrub undergrowth. The forests were also punctuated by some large boulders and rock faces. The elevation at which it was found also falls within the confirmed elevational distribution of the species in Himachal Pradesh (Saikia et al. 2007). This is the first report of this species in the 148 years since Stoliczka's documentation from Himachal Pradesh. The specimen (IM8370) described by Stoliczka was cross verified at the Indian Museum, Kolkata, and confirmed to be *Liopeltis rappi.* It verifies a distribution of this species into the Western Himalayas and extends the range of the species 210 km further west within the state of Himachal Pradesh.

The specimen (Figure 1) is in good condition with 5-6 small puncture wounds on the ventral side of the body not interfering with scale counts, and the dorsal surface is untouched. It was preserved in 70% ethanol and deposited in the collection of the High-Altitude Regional Centre, Zoological Survey of India, Solan, Himachal Pradesh.

**Figure 1.** Photograph of the freshly dead specimen of *Liopletis rappi* from Chamba district, Himachal Pradesh. (Photo: Vishal Santra)

**Table 1.** Scalation details (where available) of specimens of *Liopeltis rappi* reported in the literature in comparison to the specimen described in this study.

| Locality | Sex | MBD | ND | VD | VEN | SUB | Source |
|---|---|---|---|---|---|---|---|
| Sikkim | M | 15 | 15 | 15 | 185 | 63 | Boulenger, *Proc. Zool. Soc.* 1860 - 1890 |
| Sikkim | Na | 15 | 15 | 15 | 184 | 60 | Boulenger, *Proc. Zool. Soc.* 1860 – 1890 |
| Sikkim | F | 15 | Na | Na | 191 | 60 | Günther, 1860 |
| Darjeeling | M | 15 | 15 | 15 | 189 | 74 | Boulenger, *Proc. Zool. Soc.* 1860 – 1890 |
| Darjeeling | M | 15 | 15 | 15 | 178 | 68 | Boulenger, *Proc. Zool. Soc.* 1860 – 1890 |
| Darjeeling | M | 15 | 15 | 15 | 180 | 71 | Boulenger, *Proc. Zool. Soc.* 1860 – 1890 |
| Darjeeling | M | 15 | 15 | 15 | 182 | 69 | Boulenger, *Proc. Zool. Soc.* 1860 – 1890 |
| Darjeeling | M | 15 | 15 | 15 | 184 | 75 | Boulenger, *Proc. Zool. Soc.* 1860 – 1890 |
| Darjeeling | Na | na | na | na | 190 | 60 | Anderson, 1871 |
| Darjeeling | Na | Na | Na | Na | 190 | 75 | Anderson, 1871 |
| Darjeeling | Na | Na | Na | Na | 194 | 65 | Anderson, 1871 |
| Darjeeling | Na | Na | Na | Na | 198 | 50 | Anderson, 1871 |
| Darjeeling | Na | Na | Na | Na | 191 | 73 | Anderson, 1871 |
| Darjeeling | Na | Na | Na | Na | 190 | 71 | Anderson, 1871 |
| Darjeeling | Na | Na | Na | Na | 190 | 77 | Anderson, 1871 |
| Darjeeling | Na | Na | Na | Na | 196 | 70 | Anderson, 1871 |
| Nepal | Na | 15 | Na | Na | 198 | Na | Günther, 1860 |
| Nepal | F | 15 | 15 | 15 | 195 | Na | Boulenger, *Proc. Zool. Soc.* 1860 – 1890 |
| Chitwan, Nepal | Na | 15 | 15 | 15 | 176 | 60 | Bhattarai et al. 2017 |
| Shimla | Na | Na | Na | Na | 196 | 67 | Stoliczka, 1870 |
| Chamba, Himachal Pradesh | M | 15 | 15 | 15 | 176 | 65 | Present study, 2018 |

*MBD: Number of mid-body dorsal scale rows, ND: Number of dorsal scale rows around neck, VD: Number of dorsal scales by vent, VEN: Ventral scales (measured according to Dowling (1951), SUB: Number of subcaudal scales. Na: not available.*

The specimen was identified as *Liopeltis rappi* from the following combination of characteristics: a slightly depressed head that is not distinct from the neck, large nostril between two nasals, long tail, body uniformly dark coffee brown with a faded but distinct nuchal collar and a few dark spots on the forebody. This specimen possessed a dark lateral stripe extending from two head lengths behind the head till the anal shield; the lateral stripe uniformly spanned three dorsal scales on either side of the body and the colour below is a yellowish-white.

Scalation is as follows: dorsal scales 15:15:15, ventrals 176, anal paired, subcaudals 65 paired, temporals 1+1, postoculars 2, preoculars 1, loreal singular and longer than high, supralabials 6 on right side and 5 on left, 3 and 4 entering eye on both sides and 5th being the largest, on the left side supralabials 5 and 6 appear to be fused forming one long shield, infralabials 6 on left side and 7 on the other, 4 infralabials in contact with anterior genials. A comparison with other specimens of this species reported in the literature is presented in Table 1.

The present record emphasizes the need for more surveying efforts in the intervening areas between Himachal Pradesh in India and Central Nepal, where there are currently no records of this species.

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Hamadryad Vol. 39 (1&2), pp. 88–94, 2022.

## Confirmation of the occurrence of *Calotes irawadi* Zug, Brown, Schulte & Vindum 2006 (Squamata: Agamidae) in Mizoram, Northeast India

**CITATION.** Tariang, A. D., Malsawmdawgliana, F., Biakzuala, L., Decemson, H., Muansanga, L., Rinsanga, L., Vabeiryureilai, M. and Lalrensanga, H. T. (2022). Confirmation on the occurrence of *Calotes irawadi* Zug, Brown, Schulte & Vindum 2006 (Squamata: Agamidae) in Mizoram, Northeast India. *Hamadryad: 39, 88-94.*

At present, the agamid genus *Calotes* (Cuvier, 1817) consists of 29 species, out of which

**Table 1.** 16S rRNA sequences of *Calotes* species used in the present study.

| **Sl. No.** | **Species** | **Specimen vouchers** | **NCBI Accession Nos.** | **Grid references** | **Locality** | **References** |
|---|---|---|---|---|---|---|
| **1** | *Calotes irawadi* | MZMU2385 | OL691118 | 23.73772°N, 92.66773°E | Mizoram, India | This study |
| **2** | *C. irawadi* | AG 855 | MW901300 | 23.5743°N, 95.7376°E | Tripura, India | Gowande et al. 2021 |
| **3** | *C. irawadi* | CAS 204862 | MW901301 | 23.5743°N, 95.7376°E | Sagaing, Myanmar | Gowande et al. 2021 |
| **4** | *C. irawadi* | CESL 1089 | MW901302 | 23.5743°N, 95.7376°E | Arunachal Pradesh, India | Gowande et al. 2021 |
| **5** | *C. versicolor* | L036C | MH844713 | 8.62448°N,77.1364°E | Kerala, India | Gowande et al. 2016 |
| **6** | *C. versicolor* | L182C | MH844729 | 9.97136°N,77.2328°E | Kerala, India | Gowande et al. 2016 |
| **7** | *C. versicolor* | L190C | MH844730 | 11.9664°N,79.7633°E, | Tamil Nadu, India | Gowande et al. 2016 |
| **8** | *C. versicolor* | NCBS AT 102 | MW901307 | 12.02909°N, 79.8503°E; | Puducherry, India | Gowande et al. 2016 |
| **9** | *C. emma* | NCBSAQ | MH844707 | - | - | Pal et al. 2018 |
| **10** | *C. emma* | NCBS | MK789847 | - | Meghalaya, India | Pal et al. 2018 |
| **11** | *C. emma* | USNM:Herp:587023 | MG935747 | 12.4359°N, 98.5941°E | Tanintharyi,Myanma | Mulcahy et al. 2018 |
| **12** | *C. farooqi* | DJ 10247 | MW901314 | - | Pakhtunkhwa, Pakistan | Gowande et al. 2021 |
| **13** | *C. farooqi* | DJ 10228 | MW901313 | - | Pakhtunkhwa, Pakistan | Gowande et al. 2021 |
| **14** | *C. grandisquamis* | L120C | MH844722 | - | Kerala, India | Pal et al. 2018 |
| **15** | *C. grandisquamis* | L035C | MH844712 | - | Kerala, India | Pal et al. 2018 |
| **16** | *C. mystaceus* | NCBSAQ | MH844750 | - | Nagaland, India | Giri et al. 2019 |
| **17** | *C. mystaceus* | WII | MK789848 | - | Nagaland, India | Giri et al. 2019 |
| **18** | *C. nemoricola* | L038C | MH844715 | - | Tamil Nadu, India | Pal et al. 2018 |
| **19** | *C. nemoricola* | L555C | MH844745 | - | Tamil Nadu, India | Pal et al. 2018 |
| **20** | *C. calotes* | L037C | MH844714 | - | Tamil Nadu, India | Pal et al. 2018 |
| **21** | *C. calotes* | L374C | MH844738 | - | Tamil Nadu, India | Pal et al. 2018 |
| **22** | *Psammophilus dorsalis* | L326P | MH844734 | - | India | Pal et al. 2018 |

12 species have been recorded in India and six species are known to occur in Mizoram, namely, *Calotes emma* Gray, 1845*, C. jerdoni* Günther, 1870*, C. maria* Gray, 1845*, C. geissleri* Wagner, Ihlow, Hartmann, Flecks, Schmitz & Böhme, 2021*, C. versicolor* (Daudin, 1802) and *C. zolaiking* Giri, Chaitanya, Mahony, Lalronunga, Lalrinchhana, Das, Sarkar, Karanth & Deepak, 2019 (Gowande et al. 2021; Uetz et al. 2021; Wagner et al. 2021). In the present note, we report the occurrence of the Ayeyarwady crested lizard, *Calotes irawadi* in Mizoram. Prior to this communication, the species was known from its type locality in Sagaing Division, Myanmar (Zug et al. 2006), Western Yunnan in China (Liu et al. 2021), and Arunachal Pradesh and Tripu-

**Table 2.** Morphometric and meristic measurements of the collected specimens from Mizoram.

| Museum No. | MZMU0166 | MZMU1110 | MZMU2611 | MZMU2385 | MZMU2596 |
|---|---|---|---|---|---|
| **Sex** | Male | Female | Male | Female | Female |
| **Locality** | Tamdil | Hmuifang | Durlui | Mizoram University campus | Sihphir |
| **GPS location** | 23.74112°N,<br>92.95005°E,<br>818 m a.s.l. | 23.45536°N,<br>92.75234°E,<br>1459 m a.s.l. | 23.89684°N,<br>92.65187°E,<br>106 m a.s.l. | 23.73772°N,<br>92.66773°E,<br>849 m a.s.l. | 23.83348°N,<br>92.74365°E,<br>1327 m a.s.l. |
| **Date** | 14 April 2007 | 4 August 2011 | 24 March 2013 | 11 October 2021 | 23 October 2021 |
| **Morphometric measurements (in mm)** | | | | | |
| **Eye-ear length** | 7.4 | 4.3 | 4.6 | 6.0 | 6.2 |
| **Head Height** | 19.5 | 10.9 | 12.6 | 13.5 | 16.7 |
| **Head Length** | 24.0 | 16.3 | 19.1 | 21.8 | 21.8 |
| **Head Width** | 21.4 | 12.0 | 14.2 | 15.9 | 16.7 |
| **Interorbital** | 13.5 | 9.8 | 12.5 | 11.7 | 12.9 |
| **Snout vent length** | 98.0 | 72.5 | 83.5 | 88.9 | 90.3 |
| **Snout to Forelimb** | 36.4 | 28.6 | 29.7 | 30.6 | 33.9 |
| **Tail Length** | 251.0 | 223.0 | 255.2 | 241.5 | 246.3 |
| **Tail Width** | 9.9 | 5.4 | 5.8 | 10.7 | 10.2 |
| **Trunk Length** | 43.4 | 39.8 | 35.8 | 45.4 | 43.5 |
| **Meristic features** | | | | | |
| **Head scales** | 12 | 12 | 11 | 12 | 12 |
| **Infralabial** | 10 | 9 | 10 | 11 | 10 |
| **Supralabial** | 11 | 10 | 11 | 11 | 10 |
| **4th Finger Lamellae** | 20 | 19 | 19 | 21 | 22 |
| **4th Toe Lamellae** | 24 | 26 | 26 | 25 | 27 |
| **Dorsal scales** | 50 | 51 | 57 | 50 | 53 |
| **Midbody scales** | 44 | 48 | 45 | 47 | 45 |

ra in northeastern India (Gowande et al. 2021). Although earlier studies like Pawar & Birand (2001), Mathew (2007), Lalrinchhana et al. (2011) and Decemson (2021) reported *Calotes versicolor* from various parts of Mizoram, there is no proper taxonomic confirmation on the identity of this species in the state. Thus, to test the specific status of *C. versicolor* in Mizoram, the present work was carried out based on morphology and molecular analysis.

Five specimens of adult *Calotes cf. versicolor* from different localities of Mizoram catalogued in the Departmental Museum of Zoology, Mizoram University (MZMU) were morphologically examined following Zug et al. (2006). The sex of each individual was determined by checking the presence or absence of a hemipenis, and later confirmed through dissection. Details of specimens with their morphometric and meristic measurements are shown in Table 1. Our specimens agreed with the diagnostic features of *Calotes irawadi* provided in Zug et al. (2006), and the adults' morphological features in Liu et al. (2021) (in parentheses). For example, snout-vent length 72.5–98.0 mm (64.3–106.8 mm); head scales 11–12 (10–15); 4th finger lamellae 19–22 (17–24); 4th toe lamellae 24–27 (22–29); dorsal scales 50–57 (36–59) and midbody scales 44–48 (40–51). Prior to the preservation of the specimen (MZMU2385), a liver tissue sample was excised and stored in PCR grade absolute EtOH at -4 °C for molecular analyses. Genomic

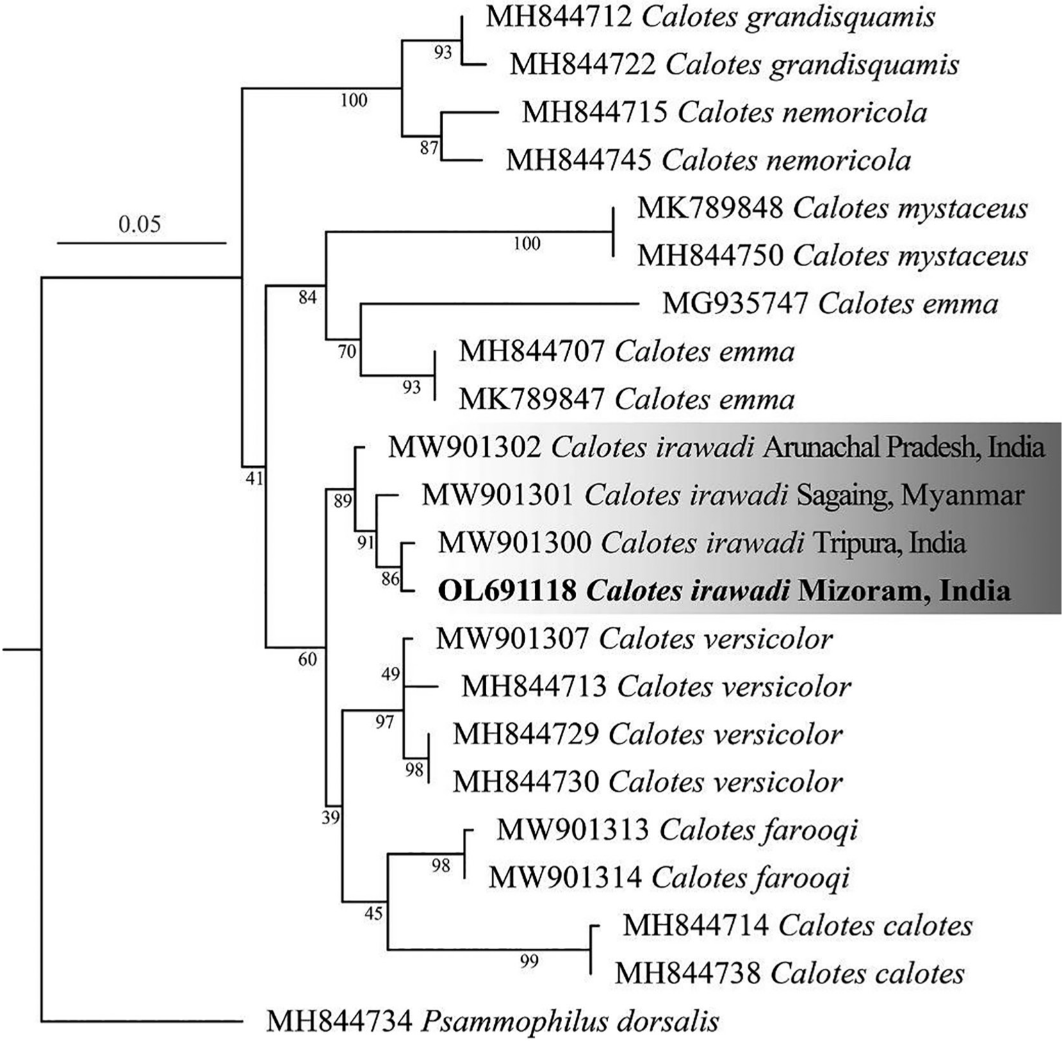

**Figure 1.** Maximum Likelihood phylogeny of the nominate *Calotes* species inferred from 16S rRNA gene sequence. Numbers at each node represent bootstrap support values. The sequence of *C. irawadi* from Mizoram, Northeast India is given in bold.

DNA was extracted using QIAamp DNA Mini Kit (Cat No. ID: 51306) following the manufacturer's instructions. The PCR was performed in a 20 µL reaction mixture containing 2 µL of 1X amplification buffer, 14.3 µL of water, 0.5 µL of 2.5 mM MgCl2, 1 µL of 0.25 mM dNTPs, 0.5 µL of 0.2 pM each of forward and reverse primers, 1µL of extracted DNA (approximately 50 ng/µL), and 0.2 µL of 1U Taq DNA polymerase. Sequences were generated using the 16S rRNA primers: forward (L02510 — CGC CTG TTT ATC AAA AAC AT, Palumbi 1996) and reverse (H03063 — CTC CGG TTT GAA CTC AGA TC, Rassmann 1997). The PCR thermal regime for amplification was 5 minutes at 95 °C for initial denaturation, followed by 35 cycles of 1 minute at 95 °C for denaturation, 30 seconds for annealing at 50.3 °C, elongation for 1 minute at 72 °C, and a final elongation for 5 minutes at 72 °C. PCR products were sequenced using Sanger's dideoxy method at Barcode Bioscience, Bangalore, India. To form a dataset, the newly generated sequences were combined with GenBank (Benson et al. 2017) sequences of *Calotes* species along with *Psammophilus dorsalis* as the outgroup (Table 2). A sequence alignment (458 bp) was generated using the MUSCLE algorithm (Edgar 2004) and p-distances estimation was performed in MEGA X (Kumar et al. 2018) using default

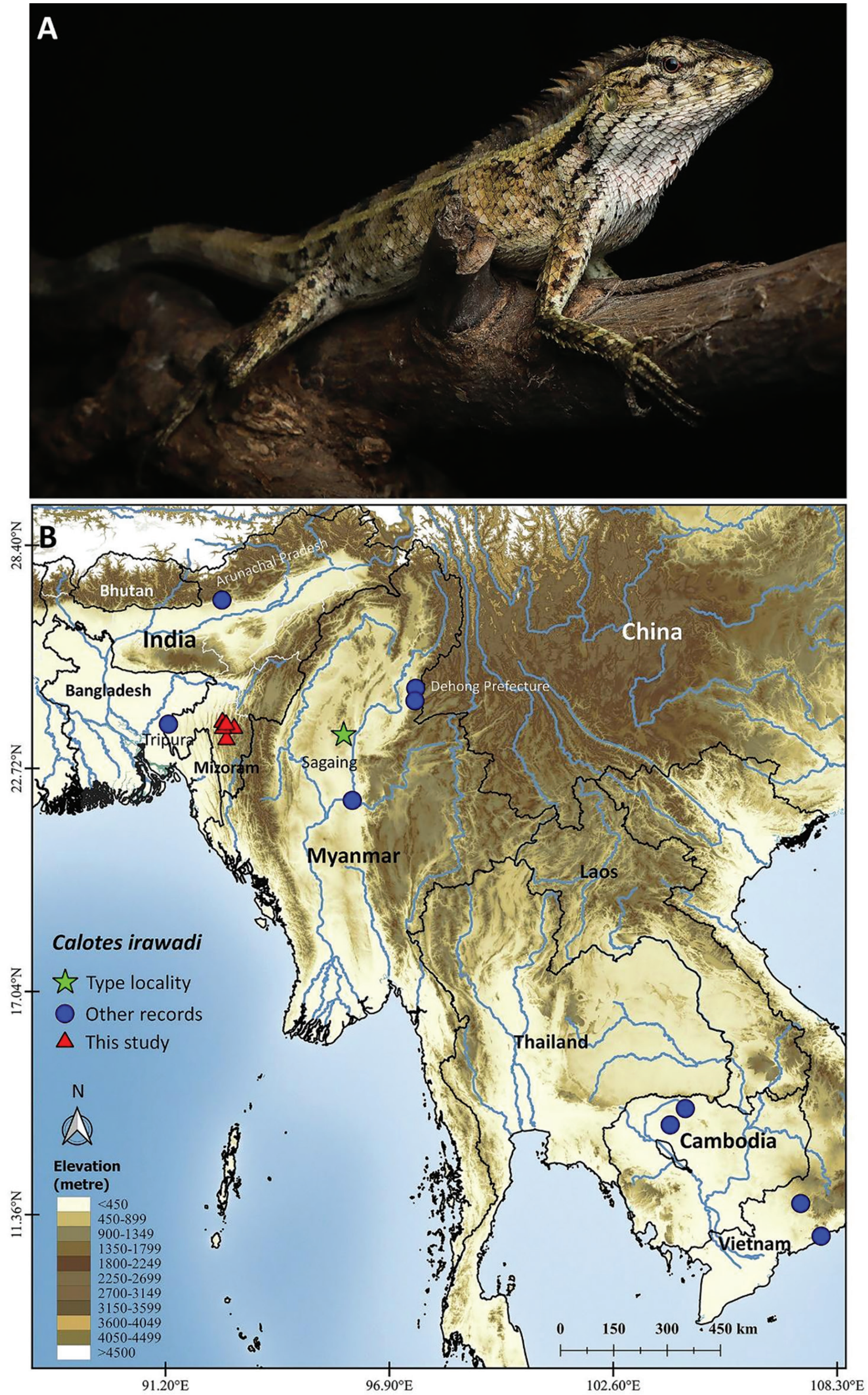

**Figure 2.** (A) Live individual of female *Calotes irawadi* (MZMU2385), (B) Map showing distributional records of *Calotes irawadi*: type locality in green star (Zug et al. 2006), other records in blue circles (Gowande et al. 2021; Liu et al. 2021), and new records from this study in red triangles.

parameter settings. The Maximum Likelihood (ML) phylogeny was reconstructed in IQ-TREE (Nguyen et al. 2015) with 10,000 Ultrafast bootstrap replicates using the best nucleotide substitution model (TIM2+F+1+G4) selected by ModelFinder (Kalyaanamoorthy et al. 2017) based on Bayesian Information Criterion. The ML phylogeny showed that the sequence from Mizoram (Voucher MZMU2385; GenBank OL691118) is grouped with the sequences of *C. irawadi* from Tripura, India (MW901300; Bootstrap value 86; p-distance 0.7%), Sagaing, Myanmar (MW901301; 1.4%), and Arunachal Pradesh, India (MW901302; 1.9%) (Fig. 1). Gowande et al. (2021) reported the intraspecific 16S divergence among *C. irawadi* was up to 2.4%. Interspecific genetic divergences of 3.3 – 4.1% are also seen between the Mizoram sequence and *C. versicolor* sequences from Puducherry (Neotype, MW901307, 3.3%), and Kerala, India (MH844713, 3.6%; MH844729 & MH844730, 4.1%).

Our results confirm the occurrence of *Calotes irawadi* in Mizoram (Figure 2A). Earlier reports on *Calotes versicolor* in Mizoram might be due to misidentification. The aerial distance from the present record at Durlui to the nearest locality at Unakoti, Tripura is approximately 70 km to the west. The present records bridge the other records from Northeast India and the type locality at Sagaing in Myanmar (Figure 2B). We suggest more extensive surveys for a taxonomic reassessment of species earlier identified as *Calotes versicolor* from the other states of northeastern India, which will provide a more accurate distributional range of *C. irawadi.*

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## A new locality record of the king cobra *Ophiophagus hannah* (Cantor, 1836) from Western Himalayas, Himachal Pradesh, India

**CITATION.** Kapoor, R., Bhardwaj, V. K. & Sharma, V. (2022). A new locality record of the king cobra *Ophiophagus hannah* (Cantor, 1836) from

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**KEYWORDS.** Elapidae, monotypic, state record, diurnal, longest venomous snake, Serpentes

The king cobra *Ophiophagus hannah* (Cantor, 1836) is a widespread elapid snake species discontinuously distributed across South and Southeast Asia. The species is distributed throughout the Oriental tropics, subtropics and temperate areas, from Western and Eastern Ghats of Peninsular India and the Himalayan foothills of northern India (Uttarakhand), east across Northeast India to southern China and southeast across Myanmar, Thailand, Vietnam, Peninsular Malaysia to Sumatra, Borneo, Java, Bali, and Sulawesi, with its eastern-most extent in Mindanao in the Philippines (Das, 2010; Wallach et al, 2014).

The king cobra is the world's longest venomous snake and grows up to 5.49-5.79 m (Aagard, 1924; Mehrtens, 1987; Daniel, 2002). It is designated as 'Vulnerable' under the IUCN Red List of Threatened Species Assessment (IUCN, 2012). Due to its wide distribution, the snake shows considerable geographic variation in colouration, size and scalation. Recent studies confirm at least four lineages of this species with unique haplotypes and morphological similarities evolving independently, which can be considered as confirmed species candidates (Shankar et al, 2021).

In the month of April 2021, an adult king cobra (Figure 1) was seen crossing the road and slithering across the soil heaps (30°30'33.1"N 77°25'08.0"E, 900 m asl) near a village called Phandi Boriwala in Sirmour district of Himachal Pradesh, which is at a linear distance of around 64 km from Dehradun, Uttarakhand. The location where the snake was seen is in the vicinity of the Girinagar forest range of Paonta forest division. The climate in the Girinagar forest range is sub-tropical monsoon with *Shorea robusta* and *Pinus roxburgii* being the dominant tree species (Bhardwaj & Kapoor 2017). Total length of the individual was estimated to be around 3 m by measuring the piece of land on which it was photographed. Sirmour district is adjacent to Uttarakhand, so the species could easily cross the boundaries of Uttarakhand and enter into the confines of Himachal Pradesh. Despite its wide distribution, the king cobra is rare in most parts of its range (Stuart et al. 2012) and that could be one of the reasons for this species going unnoticed so far in Himachal Pradesh, including in the parts adjacent to Uttarakhand. The species is well documented across several districts of Uttarakhand, boundaries of which coincide with Himachal Pradesh. The report presented herein constitutes a new locality record for the species and the first from Himachal Pradesh. Further surveys of the area are required to ascertain if the species is breeding in the area or the individual was a transient one.

**Figure 2.** King cobra *Ophiophagus hannah* (Cantor, 1836) from Western Himalayas, Himachal Pradesh, India

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## Contributions to the morphology and molecular phylogenetics of *Gonyosoma prasinum* (Blyth, 1854) (Reptilia: Squamata: Colubridae) from Mizoram, India

**CITATION.** Lalrinsanga[1], H.T. Lalremsanga[1], L. Ht. Decemson[1], V. Mathipi[1], L. Tanpuii[1], L. Muansanga[1], L. Biakzuala[1] (2022). Contributions to the morphology and molecular phylogenetics of *Gonyosoma prasinum* (Blyth, 1854) (Reptilia: Squamata: Colubridae) from Mizoram, India. *Hamadryad: 39, 96-103*.

**KEYWORDS.** Green Trinket Snake, morphometry, natural history, Northeast India, systematics

The Green Trinket Snake *Gonyosoma prasinum* (Colubridae) is arboreal (Smith 1943; Das 2012) and predominantly diurnal in habit (Whitaker & Captain 2008). Individuals of *G. prasinum* have a uniformly greenish dorsum (Blyth 1854; Smith 1943; Chan-Ard et al. 2015; Das 2012). Recent work indicates that the snake is distributed in India and Myanmar (David et al. 2022) and is listed as a species of 'Least Concern' (Wogan et al. 2021). Within India, they are found in the states of Arunachal Pradesh, Assam, Mizoram, Manipur, Nagaland, Meghalaya, and West Bengal. The snake is typically encountered in sub-montane or montane forests, near water in forest hills at elevations between 80–2650 m (Das 2012). Although distribution records are available, little else is known about the snake and no genetic data was hitherto available from India (see Blyth 1854; Smith 1943; Grossmann 2002; Whitaker & Captain 2008; Das 2012, 2018; Chan-Ard et al. 2015; Das & Das 2017). In this report, we provide morphological data of *G. prasinum* from Mizoram (Northeast India) and mitochondrial DNA sequences (*16s rRNA* and *Cyt b*). Additionally, we use the mitochondrial DNA to reconstruct a molecular phylogeny of the members of the genus *Gonyosoma*.

In this work, we documented the species from a total of 20 localities (Fig. 1), out of which the snake was known only from one locality (Mizoram University campus; Laltanpuia et al. 2008). A total of seven specimens (three males and four females) were collected from six different localities (Table 1). The morphology of these snakes largely conforms to the descriptions available in published literature (e.g., Smith 1943; Grossmann 2002). However, many of our specimens differed from the morphology detailed in published literature. We found differences in the number of subcaudal scales in two of our specimens (MZMU2551 and MZMU2620) wherein, they had 113 and 116

**Table 1.** Morphological data of *Gonyosoma prasinum* specimens from Mizoram, India.

| **Museum vouchers** | **MZMU0024 Juvenile** | **MZMU920 Juvenile** | **MZMU2619** | **MZMU2630** | **Mean±SD N=4** | **MZMU2009** | **MZMU2551** | **MZMU2620** | **Mean±SD N=3** |
|---|---|---|---|---|---|---|---|---|---|
| Date of collection | 26/9/2008 | 5/9/2016 | 8/11/2021 | 16/11/2020 | | 10/9/2020 | 22/7/2021 | 30/9/2021 | |
| Locality | Mizoram University campus, Aizawl | Mizoram University campus, Aizawl | Tanhril, Aizawl | Samtlang, Aizawl | | Maubuang, Aizawl | Murlen, Champhai | Durtlang North, Aizawl | |
| Sex | F | F | F | F | F | M | M | M | M |
| Snout-vent length | 263 | 394 | 1050 | 635 | 585.50±345.88 | 609 | 525 | 588 | 574.00±43.72 |
| Tail length | 92 | 95 | 305 | 196 | 172.00±100.99 | 217 | 186 | 205 | 202.67±15.63 |
| Eye-nostril | 3.66 | 3.00 | 7.65 | 5.07 | 4.85±2.06 | 5.98 | 5.36 | 4.86 | 5.4±0.56 |
| Eye diametre | 4.30 | 2.80 | 5.88 | 3.80 | 4.20±1.28 | 4.63 | 4.39 | 3.64 | 4.22±0.52 |
| Snout length | 5.50 | 4.90 | 11.34 | 7.41 | 7.29±2.91 | 8.38 | 7.61 | 7.90 | 7.96±0.39 |
| Snout width | 4.27 | 2.90 | 6.78 | 6.25 | 5.05±1.79 | 5.23 | 5.73 | 5.15 | 5.37±0.31 |
| Head length | 15.03 | 15.50 | 33.51 | 23.90 | 21.99±8.70 | 24.86 | 22.42 | 22.95 | 23.41±1.28 |
| Head width | 8.50 | 8.10 | 11.30 | 12.27 | 10.04±2.06 | 13.51 | 10.56 | 11.66 | 11.91±1.49 |
| Ventrals | 196 | 202 | 207 | 199 | 201.00±4.69 | 196 | 195 | 199 | 196.67±2.08 |
| Subcaudals | 112 | 111 | 111 | 96 | 107.50±7.68 | 109 | 113 | 116 | 112.67±3.51 |
| Dorsal scale rows | 19:19:13 | 19:19:15 | 19:19:15 | 17:19:17 | | 19:19:17 | 19:19:17 | 17:19:15 | |
| Supralabials | 9/9 | 9/9 | 9/9 | 9/9 | | 9/9 | 9/9 | 9/9 | |
| Supralabialstouching eye | 4-6th/4-6th | 4-6th/4-6th | 4-6th/4-6th | 4-6th/4-6th | | 4-6th/4-6th | 4-6th/4-6th | 4-6th/4-6th | |
| Infralabials | 9/10 | 10/10 | 10/10 | 10/10 | | 9/9 | 9/9 | 9/9 | |
| Loreals | 0/0 | 1/1 | 1/1 | 1/1 | | 1/1 | 1/1 | 1/1 | |
| Temprals | 2+2/2+2 | 3+3/3+2 | 2+3/2+3 | 2+2/2+2 | | 2+2/2+2 | 2+2/2+2 | 2+2/2+2 | |
| Preoculars | 1/1 | 1/1 | 1/1 | 1/1 | | 1/1 | 1/1 | 1/1 | |
| Postoculars | 2/2 | 2/2 | 2/2 | 2/2 | | 2/2 | 2/2 | 2/2 | |
| Anal shield divided | No | No | No | No | | No | No | No | |

**Table 2.** Detailed information of 16S and Cytb sequences used in this study.

| Species | Voucher | 16S | Cytb | Location | Reference |
|---|---|---|---|---|---|
| ***Gonyosoma prasinum*** | MZMU2630 | ON533525 | ON548552 | Mizoram, India | This study |
| ***G. prasinum*** | MZMU2009 | OL442122 | - | Mizoram, India | This study |
| ***G. prasinum*** | SEABRI2019120043 | - | MZ322864 | Htamanthi, Sagaing, Myanmar | Liu et al. 2021 |
| ***G. prasinum*** | SEABRI2019120075 | - | MZ322863 | Htamanthi, Sagaing, Myanmar | Liu et al. 2021 |
| ***G.* cf. *prasinum*** | CHS298 | MK194035 | MK201383 | China | Li et al. 2020 |
| ***G. boulengeri*** | CHS243 | MK194009 | MK201361 | Hainan, China | Li et al. 2020 |
| ***G. boulengeri*** | CHS242 | MK194008 | MK201360 | Mengzi, Yunnan, China | Li et al. 2020 |
| ***G. frenatum*** | CHS139 | MK193938 | MK201290 | Huangshan, Anhui, China | Li et al. 2020 |
| ***G. frenatum*** | CHS138 | MK193937 | MK201289 | Huangshan, Anhui, China | Li et al. 2020 |
| ***G. oxycephalum*** | ROM37622 | KX694646 | KX694870 | Not specified | Alencar et al. 2016 |
| ***G. oxycephalum*** | No voucher | - | AF471084 | Not specified | Lawson et al. 2005 |
| ***G. margaritatus*** | No voucher | - | KM870886 | Not specified | Chen et al. 2014 |
| ***G. jansenii*** | No voucher | - | DQ902113 | Sulawesi | Burbrink & Lawson 2007 |
| ***G. coeruleum*** | KIZ2019028 | - | MZ322867 | Mengla, Yunnan, China | Liu et al. 2021 |
| ***G. coeruleum*** | KIZ2019025 | - | MZ322870 | Mengla, Yunnan, China | Liu et al. 2021 |
| ***G. coeruleum*** | KIZ20200729 | - | MZ322866 | Zhenyuan, Yunnan, China | Liu et al. 2021 |
| ***G. coeruleum*** | KIZ20200904 | - | MZ322865 | Menglian, Yunnan, China | Liu et al. 2021 |
| ***Coelognathus radiatus*** | CHS556 | MK194066 | MK201411 | China | Li et al. 2020 |

scales, respectively. The scale counts are slightly higher than the previously known upper limit of the subcaudal scale (111 scales; Smith 1943; Grossmann 2002; Whitaker & Captain 2008). One specimen (MZMU0024) lacked a loreal scale which was present in all other specimens observed and the presence of the loreal scale is a key diagnostic feature *fide* Smith (1943). Some of our specimens also had a variable number of dorsal scale rows (Table 1) and we consider this character as a variable trait rather than an anomaly. We also found that some of the specimens were marginally longer compared to published literature (MZMU2619, Fig. 2, 135 cm vs. 120 cm.; Whitaker & Captain 2008; Chan-Ard et al. 2015; Das 2012). In addition to the information provided by Smith (1943) we observed that the hemipenis of the examined male specimens extended up to the 7th–8th caudal plate; the calyces are elongated longer in the apical part (partially everted) than that of the basal part with spinous points; apical notch with tiny spines; the proximal plicate area is smooth and without spine; sulcus spermaticus single, prominent, and runs up to the tip (Fig. 3).

**Table 3.** Partitioning schemes and nucleotide evolutionary models selected for the Bayesian Inference (BI) phylogenetic analysis.

| Partitions | Sites | Models |
|---|---|---|
| P I | 16S, Cytb 3rd codon pos | GTR+G |
| P II | Cytb 1st codon pos | HKY+I |
| P III | Cytb 2nd codon pos | TIM+G |

In our study, individuals of *G. prasinum* were encountered in terrestrial habitats (e.g., three individuals from the vicinity of a metalled road among bushes or shrubs, and one from the ground in a homestead garden) but they were occasionally seen in arboreal habitats as well. In captivity, we observed three individuals that were housed separately. One individual, (MZMU2009) preferred feeding on *Hemidactylus frenatus* than frogs, such as *Fejervarya multistriata, Minnervarya asmati, Microhyla berdmorei,* and *Sylvirana lacrima* when offered together. The other two individuals were not

**Table 4.** Uncorrected p-distance of Gonyosoma species estimated based on Cytb fragment. Sequence generated in this study is indicated by asterisk (*).

| | **Species** | **1** | **2** | **3** | **4** | **5** | **6** | **7** | **8** | **9** | **10** | **11** | **12** | **13** | **14** | **15** | **16** |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | *Gonyosoma prasinum* MZMU2630* | | | | | | | | | | | | | | | | |
| 2 | *Gonyosoma prasinum* SEABRI2019120043 | 0.022 | | | | | | | | | | | | | | | |
| 3 | *Gonyosoma prasinum* SEABRI2019120075 | 0.022 | 0.000 | | | | | | | | | | | | | | |
| 4 | *Gonyosoma prasinum* CHS298 | 0.118 | 0.118 | 0.118 | | | | | | | | | | | | | |
| 5 | *Gonyosoma boulengeri* CHS243 | 0.137 | 0.125 | 0.125 | 0.133 | | | | | | | | | | | | |
| 6 | *Gonyosoma boulengeri* CHS242 | 0.142 | 0.133 | 0.133 | 0.150 | 0.027 | | | | | | | | | | | |
| 7 | *Gonyosoma frenatum* CHS139 | 0.135 | 0.123 | 0.123 | 0.140 | 0.079 | 0.072 | | | | | | | | | | |
| 8 | *Gonyosoma frenatum* CHS138 | 0.135 | 0.130 | 0.130 | 0.152 | 0.077 | 0.077 | 0.032 | | | | | | | | | |
| 9 | *Gonyosoma oxycephalum* ROM37622 | 0.147 | 0.147 | 0.147 | 0.150 | 0.149 | 0.156 | 0.154 | 0.164 | | | | | | | | |
| 10 | *Gonyosoma oxycephalum* | 0.149 | 0.149 | 0.149 | 0.150 | 0.149 | 0.156 | 0.154 | 0.164 | 0.007 | | | | | | | |
| 11 | *Gonyophis margaritatus* | 0.125 | 0.130 | 0.130 | 0.135 | 0.123 | 0.128 | 0.126 | 0.120 | 0.164 | 0.164 | | | | | | |
| 12 | *Gonyosoma jansenii* | 0.161 | 0.157 | 0.157 | 0.150 | 0.154 | 0.152 | 0.152 | 0.159 | 0.067 | 0.070 | 0.168 | | | | | |
| 13 | *Gonyosoma coeruleum* KIZ2019028 | 0.118 | 0.121 | 0.121 | 0.022 | 0.128 | 0.142 | 0.138 | 0.147 | 0.149 | 0.149 | 0.128 | 0.154 | | | | |
| 14 | *Gonyosoma coeruleum* KIZ2019025 | 0.123 | 0.126 | 0.126 | 0.029 | 0.135 | 0.145 | 0.142 | 0.150 | 0.157 | 0.157 | 0.135 | 0.161 | 0.012 | | | |
| 15 | *Gonyosoma coeruleum* KIZ20200729 | 0.123 | 0.123 | 0.123 | 0.024 | 0.125 | 0.142 | 0.138 | 0.154 | 0.147 | 0.147 | 0.128 | 0.156 | 0.010 | 0.012 | | |
| 16 | *Gonyosoma coeruleum* KIZ20200904 | 0.121 | 0.121 | 0.121 | 0.024 | 0.126 | 0.140 | 0.133 | 0.149 | 0.150 | 0.150 | 0.135 | 0.150 | 0.014 | 0.015 | 0.010 | |
| 17 | *Coelognathus radiatus* CHS556 | 0.178 | 0.174 | 0.174 | 0.198 | 0.183 | 0.186 | 0.181 | 0.179 | 0.197 | 0.197 | 0.190 | 0.202 | 0.193 | 0.195 | 0.195 | 0.191 |

seen to feed in captivity. During the winter months (November to January), the one individual we observed, was found immersed inside the provided water bowl, particularly when the terrarium air temperature fell below 20 °C; the head was held outside while the remaining body was coiled inside the water with temperature ranging between 21 °C and 22 °C. But, during the summer months (March and May), the other two individuals we had observed so far in captivity did not show this kind of behaviour, instead, they were seen to climb vegetation but would mostly coil on the ground and hide underneath rocks.

Specimens were euthanized using MS-222 following Conroy et al. (2009); then, fixed in 10% buffered formalin solution and subsequently stored in 70% ethanol. Liver tissues were dissected for DNA extraction and stored in 95% ethanol at -20 °C. We extracted genomic DNA from two specimens (MZMU2009 and MZMU2630) using the QIAamp DNA Mini Kit. The fragments of 16S rRNA (*16S*) and Cytochrome *b* (*Cyt b*) loci were amplified in a PCR reaction using published *16S* primers pairs L02510 (Palumbi1996) and H03063 (Rassmann 1997), and the primers (Snk) from Dubey et al. (2009) was utilized for *Cyt b* gene. Amplicons were subjected to Sanger's sequencing at Barcode BioScience Pvt Ltd., Bangalore, India. The raw sequences were checked for the quality score by Sequence Analyzer v2 and high-quality sequences were included in the analysis. Both datasets (*16S*

**Table 5.** Uncorrected p-distance of Gonyosoma species estimated based on 16S fragment. Sequences generated in this study are indicated by asterisk (*).

| | Species | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 |
|---|---|---|---|---|---|---|---|---|---|
| 1 | *Gonyosoma prasinum* MZMU2630* | | | | | | | | |
| 2 | *Gonyosoma prasinum* MZMU2009* | 0.000 | | | | | | | |
| 3 | *Gonyosoma boulengeri* CHS242 | 0.051 | 0.051 | | | | | | |
| 4 | *Gonyosoma* cf. *prasinum* CHS298 | 0.043 | 0.043 | 0.055 | | | | | |
| 5 | *Gonyosoma boulengeri* CHS243 | 0.053 | 0.053 | 0.013 | 0.055 | | | | |
| 6 | *Gonyosoma frenatum* CHS139 | 0.055 | 0.055 | 0.017 | 0.060 | 0.019 | | | |
| 7 | *Gonyosoma frenatum* CHS138 | 0.053 | 0.053 | 0.019 | 0.058 | 0.021 | 0.002 | | |
| 8 | *Gonyosoma oxycephalum* ROM37622 | 0.064 | 0.064 | 0.072 | 0.081 | 0.070 | 0.066 | 0.068 | |
| 9 | *Coelognathus radiatus* CHS556 | 0.077 | 0.077 | 0.060 | 0.087 | 0.060 | 0.062 | 0.062 | 0.090 |

and *Cyt b* loci) contain the newly generated sequences and other sequences of *Gonyosoma* species along with one *Coelognathus radiatus* sequence (outgroup) retrieved from NCBI database (Table 2). We aligned sequences using the default parameters of MUSCLE (Edgar 2004), implemented in MEGA X (Kumar et al. 2018). Subsequently, the two aligned datasets were concatenated in SequenceMatrix (Vaidya et al. 2011) and partitioning (P) was done by gene and codon positions (pos). To reconstruct the Bayesian Inference (BI) phylogeny, the best partitioning schemes and models (Table 3) were

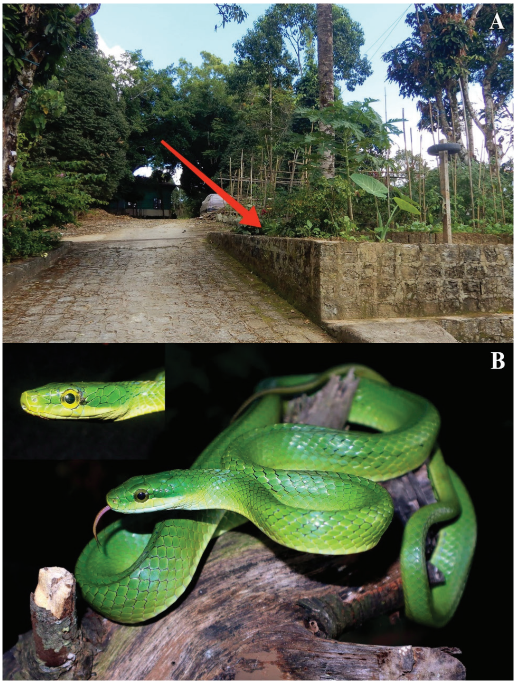

**Figure 1.** (A) Microhabitat of *Gonyosoma prasinum* (MZMU2619) at a homestead garden, Tanhril Vengpui, Mizoram, India. (B) Female of *G. prasinum* (MZMU2619) in life; inset showing the antero-lateral view of the head lacking a loreal scale (MZMU0024) in life.

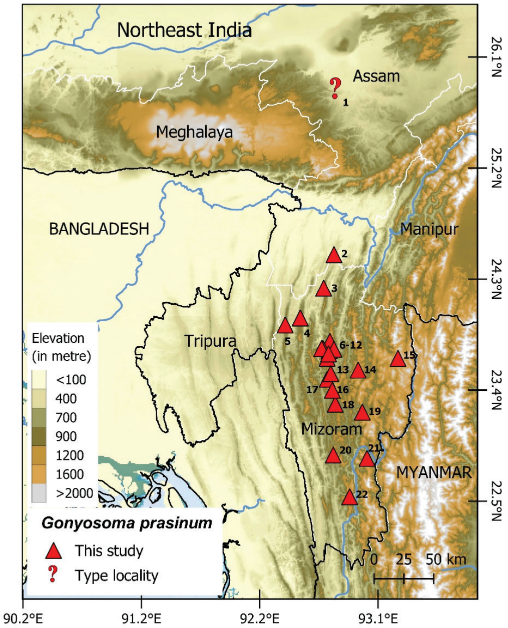

**Figure 2.** Map showing the imprecise type locality (question mark) and new distributional records (red triangles) of *Gonyosoma prasinum* from Mizoram, Northeast India: 1. Imprecise type locality (question mark was put at the approximate position of the unverified spotting in Assam by Liu et al. (2021)); 2. Kolasib; 3. North Sabual; 4. Tuidam; 5–11. Republic Vengthlang, Durtlang North, Tanhril, MZU campus, Berawtlang, Saikhamakawn, Samtlang; 12. Phulpui; 13. Tawizo; 14. Murlen National Park; 15. Maubuang; 16. Sialsuk; 17. Thenzawl; 18. Khawlailung; 19. Lunglei; 20. Muallianpui; 21. Lawngtlai.

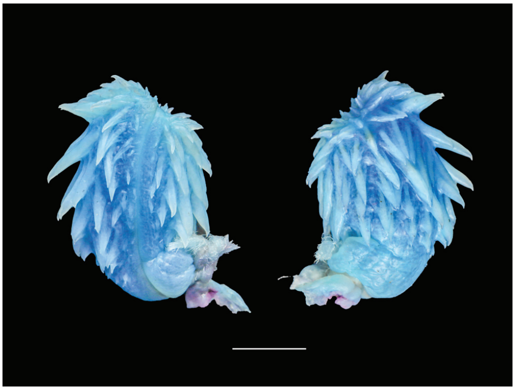

**Figure 3.** Hemipenis of MZMU2009, sulcal (left) and asulcal (right) views. Scale bar = 5 mm.

selected using PartitionFinder version 2 (Lanfear et al. 2017) based on the lowest Bayesian Information Criterion. Using the selected models, the BI phylogenetic analysis was subsequently performed by running four chains of Markov Chain Monte Carlo for 10 million generations by sampling every 10,000 generations in Mr.Bayes v3.2.5 (Ronquist et al. 2012). As a burn-in, the first 25% of the sampled trees were discarded, and Bayesian posterior probabilities (PP) were used to assess the nodal support. We visualized the BI tree in FigTree v1.4.4 (Rambaut 2019). In the *Cyt b* loci-based estimation of uncorrected p-distances, a genetic distance of 2.2% was detected between a specimen from Mizoram, India (MZMU2630) and those from Sangaing, Myanmar (SEABRI2019120043; SEABRI2019120075). A relatively high genetic distance (11.8%) was recovered between the *G.* cf. *prasinum* specimen from China (CHS298) and the other *G. prasinum* samples (India + Myanmar) (Table 4). Using the *16S* loci dataset, we found that the two specimens (MZMU2009; MZMU2630) from Mizoram, India were not genetically different from each other (genetic distance of 0.0%) but found a genetic distance of 4.3% between the Mizoram specimens and *G.* cf. *prasinum* from China (CHS298). The *16S* sequences of *G. prasinum* from Mizoram also depicted interspecific genetic divergence ranging between 5.1% (with *G. boulengeri*; CHS242) and 6.4% (with *G. oxycephalum*; ROM37622) (Table 5). Our BI phylogenetic relationship showed that populations from Mizoram are immediate sister to the population from Myanmar having strong nodal support values (PP=1.0), while the specimen of *G.* cf. *prasinum* from China (CHS298) is grouped with the type specimens of the recently described *G. coeruleum* (Fig. 4). In addition, we speculate that the specimen from China (CHS298) can be elevated

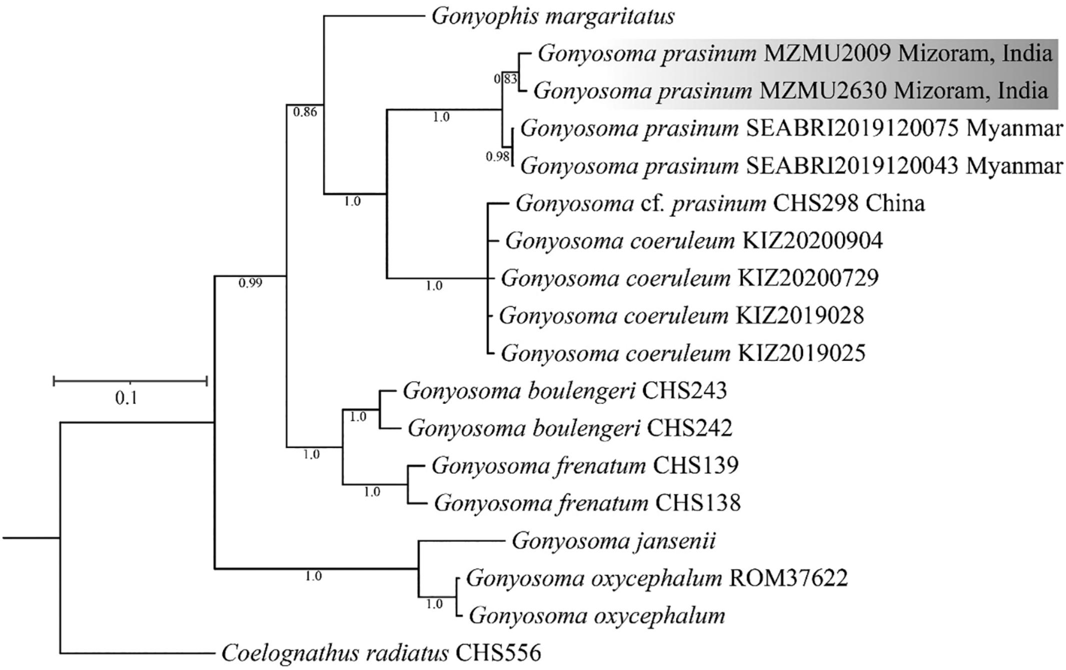

**Figure 4.** Bayesian inference phylogeny of Gonyosoma species inferred from concatenated sequences of 16S and Cytb. Bayesian posterior probability values are given at each node.

either as a new lineage or *G. coeruleum* by integrating morphological data.

The identity of *G. prasinum* was unresolved because of the lack of morphological and genetic data for comparison. Liu et al. (2021) pointed out that the original description of the species is insufficient for identifying the species. Moreover, the original description by Blyth (1854) does not specify the precise type locality and only states it as "Asám" (Blyth 1854) which comprised the whole of Northeast India during the colonial era. Our findings, where we provide detailed morphological measurements and genetic information of this species would bolster our understanding of this species across parts of Northeast India.

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# Report of Polyandry and Necrophilia in *Duttaphrynus himalayanus* (Günther, 1864) (Anura: Bufonidae) from Sikkim, India.

**CITATION.** N. G. Patel and A. Das[1] (2022). Report of Polyandry and Necrophilia in *Duttaphrynus himalayanus* (Günther, 1864) (Anura: Bufonidae) from Sikkim, India. *Hamadryad: 39, 104-109*.

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Many species of anurans have what is called "explosive breeding," which is a short, intense male-male scramble competition to mate, where males only spend a few days at breeding sites (Wells 1977; Pough et al. 2015). Explosive breeding has been documented in anuran families such as Bufonidae, Hylidae, Microhylidae, Bombinatoridae, Dicroglossidae, Pyxicephalidae, Leptodactylidae, Rhacophoridae and Ranidae (Wells 1979; Zimmerman & Bogart 1988; Seidel 1999; Prado et al. 2005, Rodrigues et al. 2007; Izzo et al. 2012; Vivek et al. 2014; Valetti et al. 2014; Khongwir et al. 2016; Abraham et al. 2013 &2018; Cai et al. 2019). The time window for such explosive breeding is relatively short and ranges from a few days to weeks (Ayres 2008). In explosive breeding, the Operational Sex Ratio (OSR) is strongly male biased as male frogs congregate at the breeding site and stay there after the mating is over in an attempt to mate with females, whereas females usually leave the breeding site after laying eggs. This results in higher male density (Menin et al. 2006, Hartel et al. 2007, Ayres 2008, Kovács & Sas 2010). Some males use non-agonistic strategies to acquire access to females associated with the most attractive males or a most attractive breeding site occupied by dominant male. Such males are called "satellite males" (Waltz 1982). These satellite males are often attracted to the amplecting pair and attempt to displace the amplecting male resulting into a large mating ball of multiple male frogs (Wells 2010). In *Isthmohyla pseudopuma* (Günther, 1901), 16 males have been recorded in a single multiple-male amplexus (Crump & Townsend 1990). Such interactions often lead to the death of the female due to drowning. (Sztatecsny et al. 2006; Zamudio & Chan 2008). Female fatality by drowning after they were clasped by scrambling males is reported in *Lithobates sylvaticus* (LeConte, 1825) and *Bufo bufo* (Linnaeus, 1758) (Howard 1980; Verrell & McCabe 1986). An observation of amplexus with a roadkill female was recorded on *Duttaphrynus melanostictus* (Schneider, 1799) from western India (Patel et al. 2016) and one record of *Duttaphrynus himalayanus* from Nepal (Rai 2022). So far, 47 cases of necrophilia have been reported worldwide, including 14 in North America,14 in South America, 10 in Asia, eight in Europe and one in Africa (Rai 2022; Serrano et al. 2022). Bufonidae have the most records of necrophilia, followed by Ranidae and Hylidae (Bufonidae: 18, Ranidae: 16, Hylidae: eight, Ascaphidae: two, Bombinatoridae: one, Leptodactylidae: one, and Pyxicephalidae: one) (Rai 2022; Serrano et al. 2022). However, possible cases of polyandry and necrophilia have hitherto been unknown from South Asia. Herein, we provide the observation of female mortality in *Duttaphrynus himalayanus* due to aggressive male scrambling competition for mating.

*Duttaphrynus himalayanus* is uniformly greyish brown to dark brown from dorsal region with diffuse large dark spots or without spots. The venter is yellowish to dark grey. Dark grey spots on the venter are also observed in some specimens. Spots on the venter are small in size. Dorsally, the spots are larger and diffuse. Both the sexes develop characteristic red spots which are more prominently visible in the females, Maximum SLV is 127 mm. Adult males attain only 70% of the female body size (Schleich & Kästle 2002). This species is widely distributed along the Himalayas (Sikkim, Darjeeling, Meghalaya, Arunachal Pradesh, Manipur, Assam, Himachal Pradesh, Uttrakhand, Jammu and Kashmir, (Kashmir) Pakistan, southern slopes of the Himalayas in central and eastern Xizang (China), Tibet, Nepal) between 1000 to 3500 m altitudinal gradient (Schleich & Kästle 2002; Patel pers. Obs.).

We observed the reproductive behaviour of *Duttaphrynus himalayanus* near Rahbangla village, Sikkim, India (27.30079167 N, 88.35187778E, 1770 m a.s.l.) on 22 April 2018. The toads were amplecting in an artificial tank (dimension: 10 m × 5 m) surrounded by agricultural fields. NP observed the amplexus event from 00:33 hrs until the female drowned in the water at 02:57 h. We attempted to photograph with a digital camera (EOS 60D; Canon Corporation), at one-minute intervals during the mating activity. From 145 minutes, a total of 117 minutes were documented.

At the beginning of our observation at 00:33 h, we recorded six males in amplexus with a single female. The males were clasping female from dorsal region in axillary position, from ventral region in axillary position, from dorsolateral region, on the hind limbs and cephalic region (Fig. 1A: Fig. 2). At a time, we recorded a maximum of eight males and a minimum of two males during the multiple amplexus (Fig. 1A &;1B). We documented a *D. melanosticus* that approached the mating event and attempted to clasp the female in the multiple amplexus (Fig. 2). The *D. melanosticus* is large sized and sturdy toad (Maximum SVL of 129 mm) with a head that is noticeably broader than long (Schleich & Kästle 2002). The snout is slightly pointed and the head and back are densely covered with warts. The warts on the sides of the body and venter are ash grey, similar to the basic body colour (Schleich & Kästle 2002). Supraorbital ridges are strongly developed, high, and black, which is absent in *D. himalayanus* (Schleich & Kästle 2002). The *D. melanostictus* individual was not able to compete with other males and left the amplexus. This *D. melanostictus* individual was there for about a minute in the amplexus. Males involved in mating were continuously pushing and kicking the other toads with their hind limbs. Due to this scrambling competition number of males were frequently changing from two to eight (Fig. 3). During the first 30 minutes of our observation, the amplecting male numbers ranged from two to eight. The amplecting male toad numbers ranged from six to eight from 30 to 60 minutes. During this time, the female struggled to stay afloat and remained motionless most of the time. Between the 60th minute to 144th minute, as time progressed, the

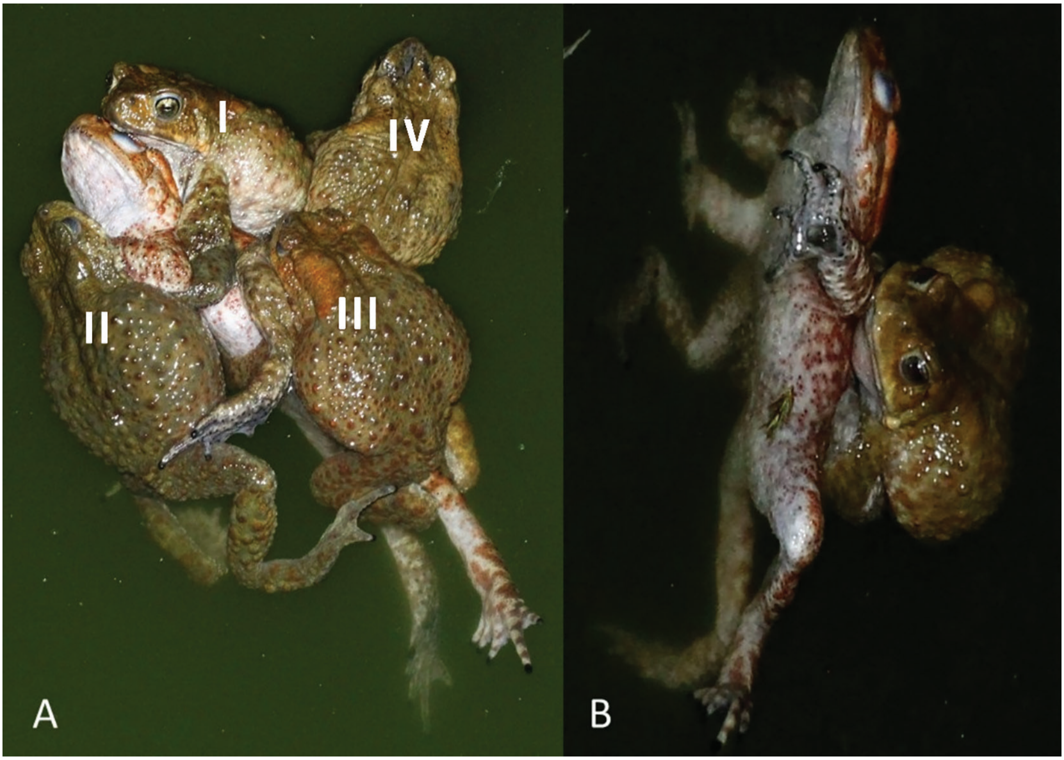

**Figure 1.** A) Four individual males trying to clasp the female from the dorsal axillary (I), ventral axillary (II) and dorsolateral (III) inguinal region. The fourth (IV) male clasped the first(I) male from the dorsal region B) Female was found dead at 2:24 am (112 minutes of the observation), at this moment two males were recorded clasping the female.

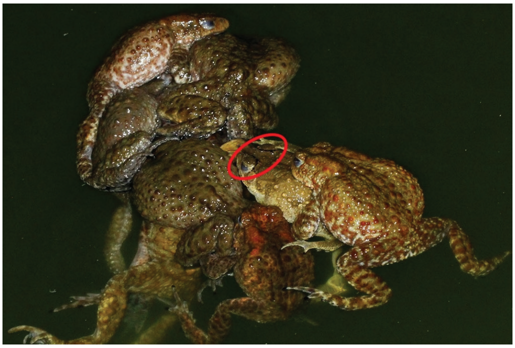

**Figure 2.** *Duttaphrynus melanostictus*, denoted by the red circle, was only seen once during the breeding event.

number of male toads kept changing from three to six individuals. After 112 minutes, the female died; at this moment only two toads were clasping the female (Fig. 1 B). We suspect that female died because of drowning. Males did not make any mating call or release call during our observation. We could not observe further after the female drowned. During the entire event eggs were not released by the female.

As time progressed, there was a constant change in the number of males attempting to clasp the female (Fig. 3). Most of the time smaller males were displaced by larger males (Wells 1979; Howard & Kluge 1985) due to the advantage of their size. In the present study, *D. melanostictus* got displaced by other males possibly due to bigger size of the amplecting males. The occurrence of mating events between *D. melanostictus* and *D. himalayanus* is possible as the hybrid between *D. melanostictus* and *D. himalayanus* was reported by Nanhoe & Oubotter (1987) in the Himalayan region. In the lower elevational limit of their distribution (~1000 m a.s.l.), *D. himalayanus* share their geographic range with *D. melanostictus* (Schleich & Kästle 2002). However, we encountered amplexus of *D. himalayanus* and *D. melanostictus* at the elevation of 1700 m a.s.l.

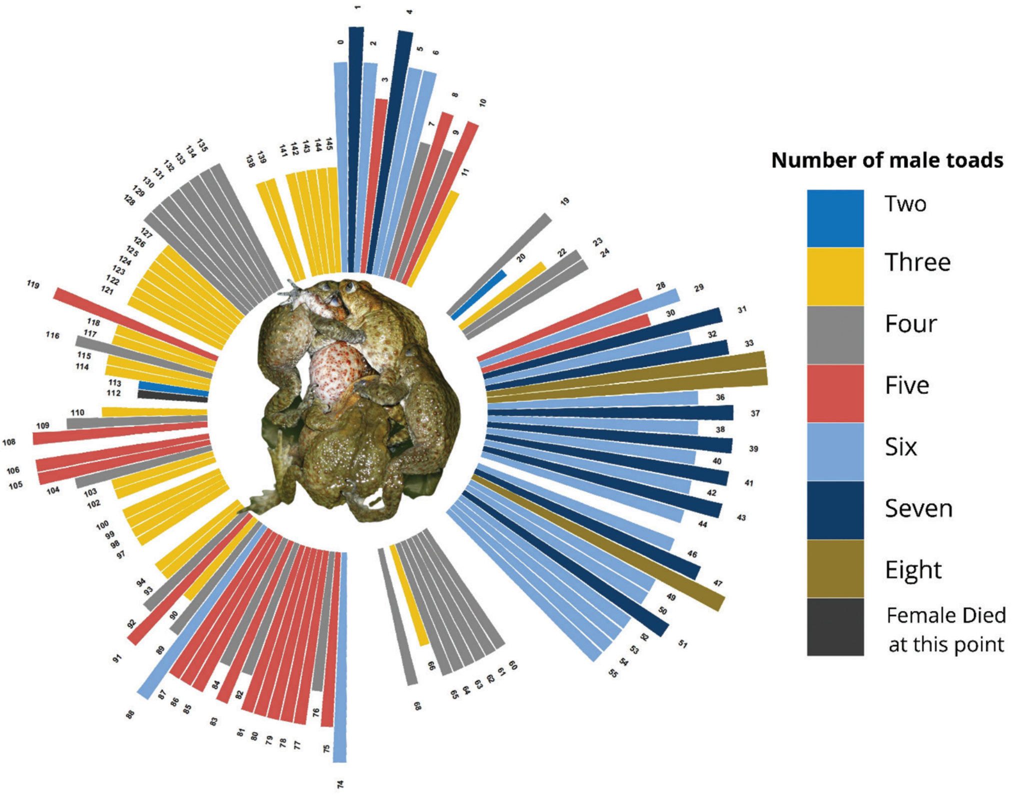

**Figure 3.** Number of male toads of *D. himalayanus* involved in the multiple-male amplexus as time progressed. A circular bar graph from 0 minute to 145 minutes showing number of male toads of *D. himalayanus* involved in the multiple-male amplexus.

Three males were clasped to the female most often during the breeding observation. Generally, the dorsally amplecting male tends to have an advantage in such scrambling competition over other males involved in the multiple-male amplexus (Wells 1979). Females are bigger in size compared to males (Schleich & Kästle 2002), hence the amplexus with a single individual male would not cause the female to drown. However, in the case of a mating ball where multiple males are trying to form an amplexus with the female, it becomes difficult for the female to stay afloat with other scrambling individuals. In this study, we did not observe the time at which the mating process started. We presume that this event might have started four to five hours (around 7 :30 pm or 8:30 pm) before we started recording it. Pintanel et al. (2021) suggested that necrophilia is primarily due to two causes: (1) failure in recognising live females by males, or (2) drowning to death during amplexus. (Marco & Lizana 2002; Đorđević & Simović 2014). In the case of necrophilia, the death of the female individuals in the population without successful reproduction is deemed maladaptive (Ayres 2010). Males who engage in necrophilia are deprived of time, energy, and the opportunity to mate with other potential females; they also face an increased risk of predation, roadkill, and disease transmission. (Meshaka 1996; McLister 2003; Ayres 2010; Costa et al 2010, Beebee 2012; Carmona-González et al. 2020). The possible evolutionary reason behind necrophilia could be selection for the stronger and persistent males, as the males having a morphological advantage in size or keratinized spinules will dominate the multiple-male amplexus (Izzo et al. 2012). However, there have been reports of female drowning to death because of intense multiple-male amplexus, leading to reproductive failure (Marco & Lizana 2002). As observed in this study once the female died, there was a decline in the number of male toads involved in the multiple-male amplexus, which suggest that males may need some feedback stimulation from the females in order to continue the amplexus, as observed in *Dryophytes versicolor* (LeConte, 1825) (McLister 2003). Our understanding of necrophilia is still in its infancy, and any additional research could clarify whether environmental and climatic conditions may promote these events and/or how sampling biases such as fieldwork funding and inventory completeness influence these spatial patterns (Serrano et al. 2022).

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- Andaman Science Association
- Asiatic Herpetological Research
- Bombay Natural History Society
- Central Zoo Authority of India
- Flora and Fauna International
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