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**Front Cover:** A barred grass snake *Natrix helvetica* photographed in Kent, England by Jason Steel. There is an article about this species on p. 38.

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# Herpetofauna of the Estação Biologia Marinha Augusto Ruschi, a coastal forest remnant in the Atlantic Forest, south-eastern Brazil

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**ABSTRACT** – A four-day herpetofauna survey was undertaken monthly from October 2018 to September 2019 at the Estação Biologia Marinha Augusto Ruschi (EBMAR) nature reserve, a coastal forest remnant in south-eastern Brazil. Each survey involved active visual and auditory searching at night, pitfall traps and incidental observations. This study revealed 31 species of anurans and 28 reptiles. Swamp forest was the most used habitat by these species. Marginal vegetation was the most used microhabitat, followed by leaf litter and bromeliads. Regarding temporal occurrence, 27 (47%) species were occasional, 20 (34%) semi-constant and 11 (19%) constant. Three species are listed as Data Deficient (*Chelonoidis carbonarius*, *Physalaemus signifer* and *Sphaenorhynchus pauloalvini*) and two (3%) as Endangered (*Ameivula nativo* and *Arcovomer passarellii*). Thirty-seven (63%) species were recorded during the active survey whereas 11 (17%) were recorded in pitfall traps. The herpetofauna of EBMAR is spatially isolated because the surrounding landscape has been converted to agriculture and human settlement.

## INTRODUCTION

Amphibians and reptiles are important components of biodiversity (Vitt & Caldwell, 2013), occupying different positions within the trophic chains (Teixeira & Coutinho, 2002; Alves et al., 2018; Zocca et al., 2017; 2019). The Brazilian Atlantic Forest biome is one of the world's biodiversity hotspots (Myers et al., 2000) with a high rate of endemism, harbouring about 720 species of anurans (Figueiredo et al., 2021) and 300 reptile species (Tozetti et al., 2017), with new species being described steadily (e.g. Mebert et al., 2020; Prates et al., 2020; Lacerda et al., 2021; Silva-Soares et al., 2021; de Sá et al., 2022; Neves et al., 2023; Zucchetti et al., 2023). Originally, the Atlantic Forest covered around 1,500,000 km², of which 76–89% were deforested (Ribeiro et al., 2009; Rezende et al., 2018). In recent decades, human-induced activities, mainly agriculture and urbanisation, have led to significant suppression and alteration of the Atlantic Forest (Tabarelli et al., 2010), causing local population extinction of amphibians and reptiles (Stuart et al., 2004; Becker et al., 2007; Böhm et al., 2013; Ferreira et al., 2016). Despite the high degree of urbanisation, coastal ecosystem remnants have rich herpetofauna, some of which have restricted distribution and are threatened with extinction (Tonini et al., 2011).

The Espírito Santo state, south-eastern Brazil is known to harbour 138 (22%) amphibian species (Rossa-Feres et al., 2017) and 134 (16%) reptile species (Guedes et al., 2023), but new species have been described in recent years (e.g. Cardozo et al., 2018; Taucce et al., 2018; Maciel et al., 2019; Silva et al., 2020; Lacerda et al., 2021; Silva-Soares et al., 2021). Furthermore, many areas of Espírito Santo state have not been adequately sampled (Almeida et al., 2011) such as the coastal forest remnant of the Estação Biologia Marinha Augusto Ruschi. The surrounding landscape of this private reserve is mostly deforested due to the development of the real estate, agricultural and tourist activities, and proximity to large urban centres. Our objective was to elaborate a species list of herpetofauna from the Estação Biologia Marinha Augusto Ruschi and provide data on habitat and seasonal preferences.

## MATERIALS & METHODS

### Study area

The Estação Biologia Marinha Augusto Ruschi (hereafter EBMAR; 19° 58'09" S, 40° 08'37" W; Fig. 1) is a small 50 ha private reserve. It is located in the district of Santa Cruz, municipality of Aracruz, Espírito Santo state, south-eastern Brazil at altitudes ranging from 0 to 100 m. The vegetation cover of EBMAR is coastal Atlantic forest, and the climate is typical of a tropical savanna with a dry winter (Köppen classification Aw), with annual precipitation ranging between 1,300 to 1,600 mm and mean annual temperature of 22–24 °C (Xavier, 1998; Alvares et al., 2013).

We sampled three habitats (swamp forest, restinga forest and human-altered areas) and their respective microhabitats (water, shrub, bromeliad, human construction, leaf litter,

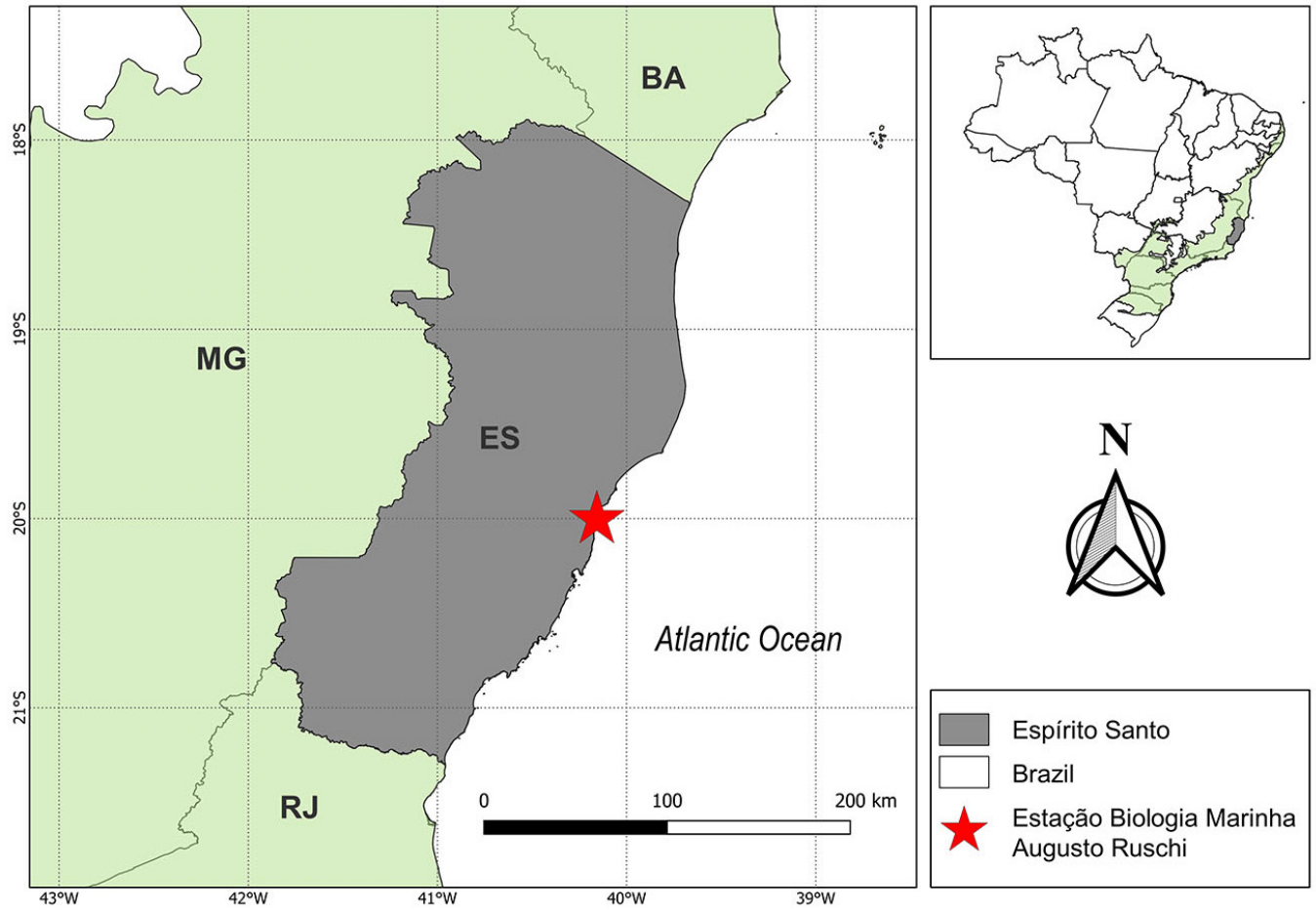


**Figure 1**. Location of the study site, Estação Biologia Marinha Augusto Ruschi (EBMAR), Santa Cruz district, municipality of Aracruz, south-eastern Brazil


**Figure 2**. Habitats and microhabitats sampled in the Estação Biologia Marinha Augusto Ruschi (EBMAR) - **A.** & **B.** Swamp forest, **C.** Restinga forest, **D.** Human-altered area

and marginal vegetation) (Fig. 2). The swamp forest is an ecosystem of the Atlantic Forest characterised by densely distributed vegetation, with tall trees and a large amount of decomposing plant material, forming a dense layer of leaf litter. Water availability in the swamp forest is greater than in other habitats, with temporary wetlands and periodically flooded areas. The restinga forest is an ecosystem of the Atlantic Forest characterised by vegetation ranging from shrubs to large trees with a thin layer of leaf litter, and mostly arenaceous. Human-altered areas consist of human housing and horticulture with sparse vegetation, strips of bare sand, high solar incidence, and low potential for water retention.

**Sampling**

A four-day survey of the herpetofauna of EBMAR was undertaken at monthly intervals from October 2018 to September 2019. On each sampling occasion data were collected through active auditory and visual search, and pitfall traps. We sampled the herpetofauna by walking ~2 km transects at night (18:00 to 23:00 h) with three researchers, using active auditory and visual search method (Crump & Scott, 1994), resulting in a sampling effort of 240 h per researcher (720 h total). We also installed five stations of pitfall traps in the three habitats: swamp forest (2 stations), restinga forest (2 stations) and human-altered areas (1 station). Each station had seven 40-litre buckets arranged in a Y shape, connected by 5 m long and 1 m high black tarp drift fences (Cechin & Martins, 2000). We monitored the pitfall stations once a day during the sampling period, corresponding to a sampling effort of 1680 buckets (35 buckets/5 stations/4 days/12 months). We also included incidental observations of species by third-party and outside the study period.

**Table 1**. Number of individual anurans recorded from October 2018 to September 2019 in the Estação Biologia Marinha Augusto Ruschi (EBMAR), south-eastern Brazil. Number of individuals: brown shading = <15 individuals; green shading = 15–30 individuals; red shading = >30 individuals; Temporal occurrence: O= occasional; S= semi-constant; C= constant

| Anuran species | Months | | | | | | | | | | | | Temporal occurrence |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Oct | Nov | Dec | Jan | Feb | Mar | Apr | May | Jun | Jul | Aug | Sep | |
| Bufonidae | | | | | | | | | | | | | |
| *Rhinella crucifer* | | | | | | | | | | | | | S |
| *Rhinella granulosa* | | | | | | | | | | | | | O |
| Craugastoridae | | | | | | | | | | | | | |
| *Haddadus binotatus* | | | | | | | | | | | | | C |
| Hylidae | | | | | | | | | | | | | |
| *Boana albomarginata* | | | | | | | | | | | | | S |
| *Boana faber* | | | | | | | | | | | | | C |
| *Boana pombali* | | | | | | | | | | | | | S |
| *Boana semilineata* | | | | | | | | | | | | | O |
| *Dendropsophus berthalutzae* | | | | | | | | | | | | | O |
| *Dendropsophus bipunctatus* | | | | | | | | | | | | | O |
| *Dendropsophus branneri* | | | | | | | | | | | | | C |
| *Dendropsophus elegans* | | | | | | | | | | | | | S |
| *Dendropsophus* sp. *(cf. haddadi)* | | | | | | | | | | | | | C |
| *Itapotihyla langsdorffii* | | | | | | | | | | | | | C |
| *Nyctimantis brunoi* | | | | | | | | | | | | | C |
| *Ololygon argyreornata* | | | | | | | | | | | | | S |
| *Phyllodytes luteolus* | | | | | | | | | | | | | C |
| *Phyllomedusa burmeisteri* | | | | | | | | | | | | | O |
| *Scinax alter* | | | | | | | | | | | | | C |
| *Scinax cuspidatus* | | | | | | | | | | | | | S |
| *Scinax fuscovarius* | | | | | | | | | | | | | O |
| *Sphaenorhynchus pauloalvini* | | | | | | | | | | | | | O |
| *Sphaenorhynchus planicola* | | | | | | | | | | | | | O |
| *Trachycephalus nigromaculatus* | | | | | | | | | | | | | O |
| Leptodactylidae | | | | | | | | | | | | | |
| *Leptodactylus fuscus* | | | | | | | | | | | | | O |
| *Leptodactylus latrans* | | | | | | | | | | | | | C |
| *Leptodactylus natalensis* | | | | | | | | | | | | | S |
| *Physalaemus signifer* | | | | | | | | | | | | | C |
| Microhylidae | | | | | | | | | | | | | |
| *Arcovomer passarellii* | | | | | | | | | | | | | O |
| *Chiasmocleis capixaba* | | | | | | | | | | | | | S |
| *Stereocyclops incrassatus* | | | | | | | | | | | | | O |
| Odontophrynidae | | | | | | | | | | | | | |
| *Proceratophrys laticeps* | | | | | | | | | | | | | S |

We classified the spatial distribution of the species according to habitat (swamp forest, restinga forest and human-altered areas) and microhabitat (water, shrubs, bromeliad, human housing, leaf litter and marginal vegetation). We classified temporal occurrence of species recorded during the sampling period as:

i) occasional (occurring from one or two months);
ii) semi-constant (occurring from three to nine months);
iii) constant (occurring from 10 to 12 months) (Prado & Pombal, 2005).

The conservation status of the species follows the international (IUCN, 2023), national (ICMBIO/MMA, 2018) and Espírito Santo (ES) state lists (Bérnils et al., 2019; Ferreira et al., 2019). We identified the species using photographic guides (Haddad et al., 2013) and comparison of specimens deposited in the Coleção de Zoologia do Museu de Biologia Prof. Mello Leitão (MBML). The voucher specimens were euthanised with lidocaine, fixed in 10% formalin, conserved in 70% alcohol (McDiarmid et al., 1994) and deposited in the MBML from the Instituto Nacional da Mata Atlântica (INMA) located in the municipality of Santa Teresa, Espírito Santo, Brazil. A list of these voucher specimens is provided in the Supplementary Material.

**Table 2**. Number of individual reptiles recorded from October 2018 to September 2019 in the Estação Biologia Marinha Augusto Ruschi (EBMAR), south-eastern Brazil, legends as per Table 1

| Reptile species | Months | | | | | | | | | | | | Temporal occurrence |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Oct | Nov | Dec | Jan | Feb | Mar | Apr | May | Jun | Jul | Aug | Sep | |
| Testudinidae | | | | | | | | | | | | | |
| *Chelonoidis carbonarius* | | | | | | | | | | | | | O |
| Amphisbaenidae | | | | | | | | | | | | | |
| *Leposternon* sp. | | | | | | | | | | | | | O |
| Dactyloidae | | | | | | | | | | | | | |
| *Dactyloa punctata* | | | | | | | | | | | | | S |
| *Norops fuscoauratus* | | | | | | | | | | | | | O |
| *Norops ortonii* | | | | | | | | | | | | | O |
| Gekkonidae | | | | | | | | | | | | | |
| *Hemidactylus mabouia* | | | | | | | | | | | | | C |
| Gymnophthalmidae | | | | | | | | | | | | | |
| *Leposoma scincoides* | | | | | | | | | | | | | S |
| Mabuyidae | | | | | | | | | | | | | |
| *Brasiliscincus agilis* | | | | | | | | | | | | | O |
| *Psychosaura macrorhyncha* | | | | | | | | | | | | | O |
| Phyllodactylidae | | | | | | | | | | | | | |
| *Gymnodactylus darwinii* | | | | | | | | | | | | | S |
| Polychrotidae | | | | | | | | | | | | | |
| *Polychrus marmoratus* | | | | | | | | | | | | | S |
| Teiidae | | | | | | | | | | | | | |
| *Ameiva ameiva* | | | | | | | | | | | | | S |
| *Ameivula nativo* | | | | | | | | | | | | | O |
| *Salvator merianae* | | | | | | | | | | | | | S |
| Tropiduridae | | | | | | | | | | | | | |
| *Tropidurus torquatus* | | | | | | | | | | | | | S |
| Boidae | | | | | | | | | | | | | |
| *Boa constrictor* | | | | | | | | | | | | | S |
| *Corallus hortulana* | | | | | | | | | | | | | S |
| Colubridae | | | | | | | | | | | | | |
| *Chironius foveatus* | | | | | | | | | | | | | O |
| *Drymoluber dichrous* | | | | | | | | | | | | | O |
| *Leptophis ahaetulla* | | | | | | | | | | | | | S |
| *Oxybelis aeneus* | | | | | | | | | | | | | O |
| *Spilotes sulphureus* | | | | | | | | | | | | | O |
| Dipsadidae | | | | | | | | | | | | | |
| *Dipsas indica* | | | | | | | | | | | | | O |
| *Dipsas neuwiedii* | | | | | | | | | | | | | O |
| *Oxyrhopus petolarius* | | | | | | | | | | | | | O |
| *Philodryas olfersii* | | | | | | | | | | | | | O |
| Elapidae | | | | | | | | | | | | | |
| *Micrurus corallinus* | | | | | | | | | | | | | O |
| Viperidae | | | | | | | | | | | | | |
| *Bothrops jararaca* | | | | | | | | | | | | | S |

## RESULTS

We recorded 3785 individuals of the herpetofauna comprising 31 species of anurans (17 genera and seven families) and 28 species of reptiles (26 genera and 15 families) at EBMAR; the species names and families are shown in Tables 1 and 2. Photographs of many of these species are shown in the Supplementary Material - Table 1S for anurans and Table 2S for reptiles. The details of the species, the habitats and microhabitats in which they were encountered, their endemicity and the sampling methods that revealed their presence are shown in Supplementary Material - for anurans

(Table 1S) and for reptiles (Table 2S). The most recorded anuran species were *Phyllodytes luteolus* (674 records; 19%), *Physalaemus signifer* (612 records; 18%) and *Scinax alter* (473 records; 14%). The most recorded reptile species were the lizards *Tropidurus torquatus* (102 records; 31%) and *Ameiva ameiva* (94 records; 29%).

Regarding habitat, 45 (76%) species were recorded in the swamp forest, 38 (64%) species in human-altered areas and 18 (30%) species in the restinga forest. The distribution and overlap of species between habitat types is shown in Figure 3. Twelve amphibian species were found in all three habitat types compared with only three reptile species. No amphibian and only one reptile species was restricted to the restinga.

Regarding microhabitat, 1364 (36%) individuals were recorded in the marginal vegetation of the water bodies, 1187 (31%) individuals were in the leaf litter, 785 (21%) individuals were in bromeliads, 234 (6%) individuals were in shrubs, 168 (4%) individuals were in the water, and only 47 (1%) individuals were recorded in human constructions. Thirty-seven (63%) species were recorded in only one microhabitat, 12 (20%) species were in two microhabitats, and eight (14%) species were in three microhabitats (Tables 1S and 2S). The highest number of species was recorded in the leaf litter (N = 28 species; 47%), followed by shrub (N = 20 species; 34%) and marginal vegetation (N = 18 species; 30%).

Thirty-seven (63%) species were recorded during active survey (visual and auditory), followed by 11 (19%) species recorded through pitfall trap. Six (10%) species of anurans and 15 (25%) species of reptiles were incidental records (Tables 1S and 2S). Active search (i.e. auditory and visual) recorded the highest number of individuals (N = 3629 individuals; 96%), followed by pitfall trap (N = 98 individuals; 3%), and incidental sightings (N = 58 individuals; 1%).

Regarding temporal occurrence, 28 (47%) species were occasional, 20 (34%) semi-constant and 11 (19%) constant (Tables 1 and 2). Overall, constant species had more individuals (>30 individuals) per month. *Itapotihyla langsdorffii*, *Nyctimantis brunoi*, *P. luteolus* and *S. alter* were active every month of the year. The lowest number of species (N = 13; 22%) was recorded in March, whereas the highest number of species (N = 31; 52%) was recorded in November (Tables 1 and 2).

We recorded 32 species in EBMAR (54% of the total) that are endemic to the Atlantic Forest, of which 24 were anurans (41%) and eight reptiles (14%) (Tables 1S and 2S). Regarding conservation status, 51 (86%) species were categorised as Least Concern, three (5%) as Data Deficient (*Chelonoidis carbonarius*, *P. signifer*, *Sphaenorhynchus pauloalvini*), and two (3%) as Endangered (*Ameivula nativo*, *Arcovomer passarellii*) from either IUCN, Brazilian or state redlists.

## DISCUSSION

Our survey of EBMAR provided a list of 31 anuran species and 28 reptile species. The 31 anuran species corresponds to 86% of the 36 species listed for the municipality of Aracruz (Almeida et al., 2011), 23% of the 138 species listed for Espírito Santo state (Rossa-Feres et al., 2017), 4% of the 720 species listed for Atlantic Forest (Figueiredo et al., 2021),

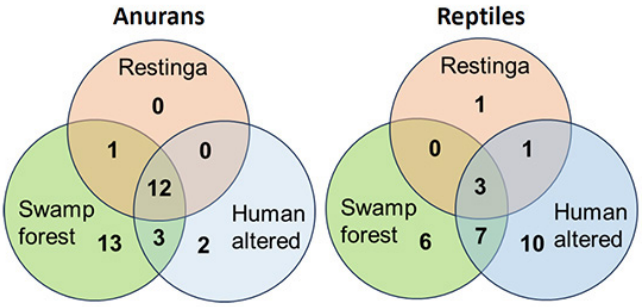


**Figure 3**. Number and overlap of herpetofauna species found in the three habitats at the Estação Biologia Marinha Augusto Ruschi (EBMAR)

and 3% of the 1188 species listed for Brazil (Segalla et al., 2021). The 28 reptile species corresponds to 21% of the 134 species listed for Espírito Santo state (Guedes et al., 2023), 9% of the 300 species listed for Atlantic Forest (Tozetti et al., 2017), and 3% of the 856 species listed for Brazil (Guedes et al., 2023). Studies at other sites in the Espírito Santo state have recorded both fewer species than EBMAR, such as Canal de Itaputanga (ca. 115 km south; N = 9 species; Maioli et al., 2018), Universidade Federal do Espírito Santo (ca. 38 km south; N = 20 species; Ferreira & Mendes, 2010), and more species than EBMAR, such as Reserva Biológica de Duas Bocas (ca. 50 km south; N = 76 species; Tonini et al., 2010) and Reserva Natural Vale (ca. 90 km north; N = 120 species; Bérnils et al., 2014; Almeida & Gasparini, 2015). However, most studies in Espírito Santo state were undertaken in large forest remnants and in protected areas. In contrast, EBMAR is a private, small and isolated forest remnant. The species richness of herpetofauna from EBMAR may be related to the high habitat heterogeneity, which includes permanent and temporary water bodies, an abundance of tank bromeliads, and forest with dense leaf litter.

Regarding habitat, the highest number of species (76%) were recorded in the swamp forest which is particularly rich in hylid species that use the permanent and temporary water bodies in this forest for breeding (Haddad et al., 2013; Silva et al., 2019). Surprisingly, human-altered areas were used by 38 (64%) of amphibian and reptile species of which 12 (20%) were found only in this habitat. Human-altered areas are found within the natural areas of EBMAR, thus species can move easily from natural to altered habitats. Furthermore, some species recorded in this habitat are considered to have high ecological plasticity, such as *Rhinella granulosa* and *T. torquatus*.

Regarding microhabitats, the marginal vegetation along the water bodies had the highest number of individuals. The high abundance of hylids was mostly associated with permanent and temporary water bodies surrounded by marginal vegetation in the swamp forest. Species of this family are characterised by adhesive discs present on the fingertips, which give them the ability to perch on vertical marginal vegetation (Cardoso et al., 1989; Zocca et al., 2014). Nine species (*Rhinella crucifer*, *Boana pombali*, *I. langsdorffii*, *N. brunoi*, *P. luteolus*, *S. alter*, *Ololygon argyreornata*, *Bothrops jararaca* and *Hemidactylus mabouia*) were recorded in bromeliads, showing the importance of this microhabitat for

the local species. *Phyllodytes luteolus* was the most recorded species in bromeliads, because it breeds in the accumulated rainwater between the leaves of these plants.

The use of multiple sampling methods may have increased the number of species recorded. Active survey recorded a higher number of individuals than either pitfall traps or incidental records. Active survey possibly detected more species because searching directly in refuges, burrows, bromeliads may reveal both active and inactive species (Auricchio & Salomão, 2002). Pitfall traps were responsible for recording species associated with leaf litter that generally have fossorial habits and are rarely observed during the active search or by incidental observation.

The EBMAR is a small forest fragment (ca. 4 km²), surrounded by plantations and human constructions. Despite this, EBMAR harbours higher species richness than other larger areas in the state of Espírito Santo, including threatened species such as *A. passarellii* (EN) and *A. nativo* (EN) (MMA, 2014; Bérnils et al., 2019). *Arcovomer passarellii* was abundant and widely distributed across the sampled habitats. On the other hand, only one individual of *A. nativo* was recorded, which was found in the restinga forest. Both species had preference for the leaf litter microhabitat, highlighting the importance of forest remnants to maintain these populations. It is noteworthy that records of the hylid *B. pombali*, the tortoise *C. carbonarius* and two snakes *Boa constrictor* and *Corallus hortulana* were the first for the municipality of Aracruz. Although a native of the Atlantic Forest, *Chelonoidis carbonarius* has become locally extinct in most of its range. It is likely *C. carbonarius* has been released in EBMAR by the local animal rehabilitation centre (CEREIAS).

Our study has shown that EBMAR supports a rich herpetofauna, highlighting the conservation importance of this isolated remnant of coastal Atlantic Forest, surrounded by a low permeable matrix (i.e. human settlements and agriculture). The understanding of the composition of the EBMAR herpetofauna fills knowledge gaps about habitat use, microhabitat selection and temporal occurrence. The long-term monitoring of these species should be undertaken as a basis for the elaboration of effective conservation plans.

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RESEARCH ARTICLE

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# Behaviour and activity pattern of a Kanchanaburi viper *Trimeresurus kanburiensis*

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**ABSTRACT** – Several conservation assessments have proposed that the Kanchanaburi pit viper *Trimeresurus kanburiensis* is threatened with extinction due to limited distribution, few documented sites and the collection of mature individuals for the international pet trade. No previous study has evaluated these threats comprehensively, nor investigated the natural history of the species in the wild. To address these knowledge gaps, we began with a brief review of wildlife trade reports and then during September–November 2020 we undertook a field study (nearly 100 surveyor hours) using fixed camera recording. We located only a single specimen, a male, whose behaviour, activity pattern and natural history appeared similar to those of other green pit viper species. The general habitat selected by this individual conformed to prior observations although close proximity to a paved road and use of a termite mound were novel. The review of wildlife trade reports did not reveal any seizures or observations, which was likely due to *T. kanburiensis* not receiving formal international protection and exclusion from stringent local legislation. We recommend further field study, identification of sites of occurrence, and investigation on popular websites and social media platforms that trade in *T. kanburiensis* and phenotypically similar Thai species. For a Thai language summary of this article see Supplementary Material on the BHS website.

## INTRODUCTION

Viper diversity is comparatively high in Thailand, with 23 species currently recorded in the country (Thai National Parks, 2023). Much attention and study has been devoted to finding and describing species, taxonomy and nomenclature, and venomics, with very little work being published describing basic natural history beyond initial species descriptions and personal observations supplementing peripheral topics. Conservation measures based on data from studies prioritising these topics tend to be unreliable and ineffective without a strong ecological foundation. In this study of the Kanchanaburi pit viper *Trimeresurus kanburiensis* Smith 1943, which has been categorised by the International Union for the Conservation of Nature (IUCN) as Endangered (Chan-Ard et al., 2022), we address a significant conservation knowledge gap with a novel natural history field study complemented by a discussion of conservation threats.

Two of the three justifications presented within the IUCN ENB1ab(v) listing for *T. kanburiensis* are habitat-related restricted-distribution of little more than 3,000 km² and presence in fewer than five locations. These have been indirectly addressed through surveys, but the third justification for listing the viper as endangered, trade (either wild-caught or captive), has not been investigated. Interestingly, *T. kanburiensis* was initially proposed to be of 'Least Concern' and 'Endemic' in the Thailand Red Data: Vertebrates list (Office of Natural Resources and Environmental Policy and Planning, 2007), in 2017 this was conservatively increased to 'Endangered' (Office of Natural Resources and Environmental Policy and Planning, 2017). The Thailand Red Data List does not provide justifications for each species assessed.

## MATERIALS AND METHODS

### Field observations

Field surveys (14 visual-encounter hikes plus 8 by road) to find *T. kanburiensis* were conducted during 4-day sampling sessions per month between September–November 2020 for a total of 96.9 surveyor hours at Khao Laem National Park (KLNP), Thailand (Fig. 1). This is a previously unpublished but locally well-known site for the Kanchanaburi pit viper. Local national park management were notified a minimum of two weeks in advance of every survey session, a ranger from the Department of National Parks, Wildlife and Plant Conservation (DNP) was present during every survey, and no animals were handled/captured nor were they or their habitat intentionally disturbed as per research and ethical permission requirements. The viper field research adhered to Guidelines for the Treatment of Animals in Behavioural Research (developed by the Association for the Study of Animal Behaviour; ASAB, 2012) and Guidelines for Research on Live Amphibians and Reptiles (American Herpetological Animal Care and Use Committee; HACC, 2004) ethical standards.

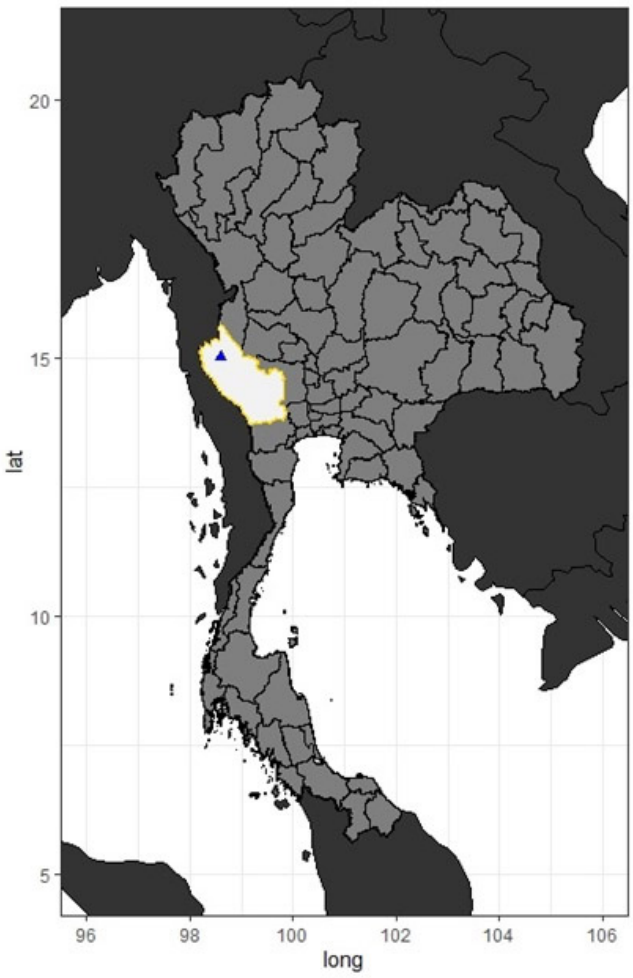


**Figure 1**. Kanchanaburi pit viper *Trimeresurus kanburiensis* study location at Khao Laem National Park (indicated by a blue triangle) study area within Thailand which is shaded in light grey. Within Thailand, provinces are partitioned in black, with Kanchanaburi outlined in yellow.

A continuous feed 'security video camera' (Hikvision model DS-2CE16C0T-IRF), set to record at a rate of 29 frames per second and mounted on a tripod, was used to monitor the behaviour, natural history and activity patterns of *T. kanburiensis*. The ethogram for this study, a set of terms and descriptions of the behaviours of an animal (Lehner, 1987), comprised behavioural states and events (ambush, rest and move states; headbob, probe, strike, tail undulation and gape events; as defined in Barnes & Knierim, 2019). Non-parametric circular kernel-density function analysis was employed to delineate temporal differences and similarities of behaviours to each other and also specifically to relate the presence of prey on camera to ambush behaviour displayed by *T. kanburiensis*. A coefficient of overlap (Δ) was used to measure the extent of overlap between two kernel-density estimates in the 'overlap' R package (Ridout & Linkie, 2009; Linkie & Ridout, 2011). General microhabitat notes were made, and canopy cover (%) was estimated using the CanopyApp smartphone application (Landert, 2016).

**Review of trade threats**

We sought to understand the threat (reflected by the IUCN listing) that international trade may pose to *T. kanburiensis*.

**Figure 2**. An adult male Kanchanaburi pit viper *Trimeresurus kanburiensis* observed initially moving in limestone karst habitat interspersed with bamboo in Khao Laem National Park, Kanchanaburi province, Thailand. Neither the viper nor its habitat was disturbed and just this one photograph was taken with flash, a second photo was taken without flash immediately after and then again later in the sampling session during the day.

A review of trade literature post-2001 was undertaken by accessing records publicly available from the Convention on International Trade in Endangered Species (CITES) and Law Enforcement Management Information System (LEMIS) using R packages, 'rcites' (Geschke et al., 2021) and 'lemis' (Eskew et al., 2020), and from the Thailand CITES Enforcement Division, DNP through their website (https://portal.dnp.go.th/p/WildlifeConserve), trade data files archived on OSF with other *T. kanburiensis* publication project code and data. Results from program R were confirmed through review of records publicly available on the LEMIS and CITES websites, and it is worth noting that no Thai pit viper species (including *T. kanburiensis*) are specifically listed in CITES Appendices 1–3.

The trade (CITES and LEMIS) program R code, list and pictures of reptile and amphibian species observed during KLNP field surveys can be found at osf.io/2sh3m. Video recordings have also been reposited at Movie Archives of Animal Behavior (http://movspec.mus-nh.city.osaka.jp/ethol/title-e.php) and on Youtube (https://www.youtube.com/channel/UCdRlzXz9YbUR2eWyEAEGU4g).

## RESULTS

**Field observations**

Only a single adult male *T. kanburiensis* was observed during the study period, which by its colouration, body plan and size (> 300 mm by visual estimation) was identified by reference to Cox et al. (2012). The observation was made on 3 November at 19:40 h when the snake was moving slowly as if searching for a potential ambush site (Fig. 2). The viper was spotted with a headlamp, one photograph was taken without flash and one with flash from a distance of >2 m, neither of which appeared to disturb the slowly moving viper as pace and direction did not appear to change. Surveyors moved away and briefly checked whether the viper was moving using the lowest headlamp settings (10 minutes

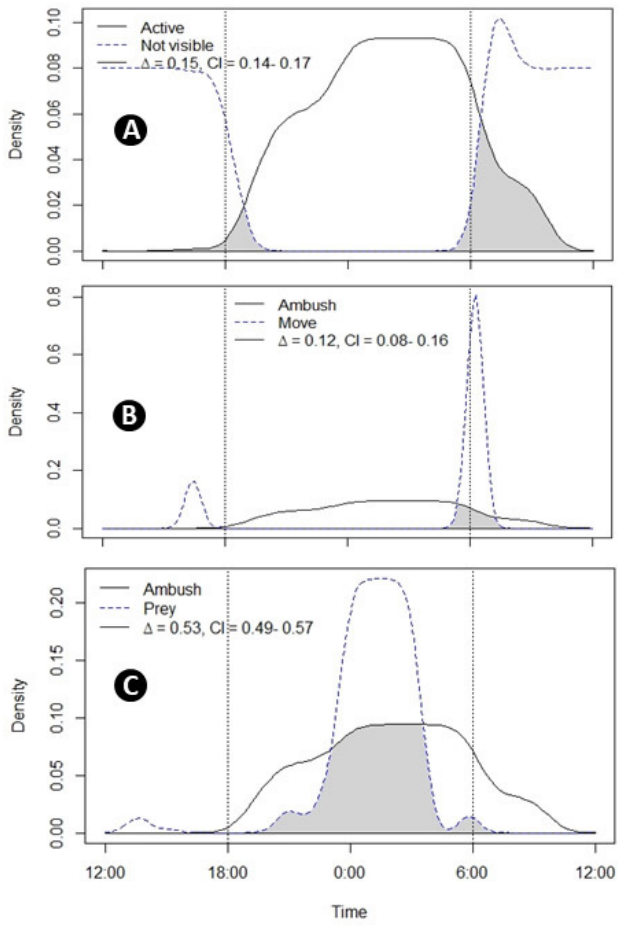


**Figure 3**. Temporal activity patterns observed for an adult male Kanchanaburi pit viper *Trimeresurus kanburiensis* observed on a continuous feed camera at Khao Laem National Park, Thailand, with the overlapping coefficient (Δ) represented by the grey shaded area - **A.** The viper was primarily active during the night (between 18:00–06:00 h) and usually not visible during the day, **B.** Move behaviour was primarily observed in the early morning and early evening close to the overlap periods of ambush and when the viper was not visible, **C.** Prey was primarily observed during the night, this matched the *T. kanburiensis* ambush foraging pattern.

and 35 minutes after initial observation). The video camera was then set (lights never being pointed directly at the viper during this process) about 2.5 m horizontal ground distance away from the viper at approximately 55 minutes after initial observation when the viper was observed in ambush posture on a termite mound. The camera recorded from then until 6 November morning (09:45 h, 61.5 hours), with SD card failure experienced the first night. When the camera was retrieved on the last morning the Kanchanaburi viper was observed in ambush posture and left undisturbed.

The *T. kanburiensis* was primarily observed in an ambush posture during the night and was usually not visible during the day, with limited movement between day and night (Fig. 3A& B). On 4 November, the viper was seen on the camera entering the termite mound for shelter. Behavioural events were infrequently observed, as were prey. No conspecifics or predators were visible on the camera. The most frequent behavioural event displayed by the viper was headbobbing, which was observed 85 times between 19:03–06:28 h. Only

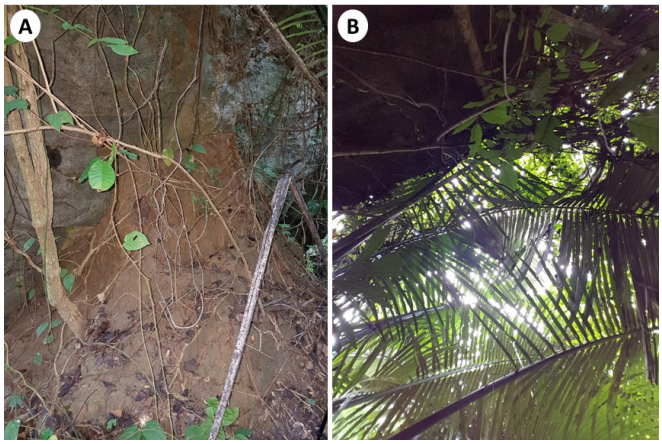


**Figure 4**. Microhabitat where a *Trimeresurus kanburiensis* was observed in the Khao Laem National Park, Kanchanaburi province, Thailand - **A.** Location where the viper was observed in an ambush posture on an inactive termite mound, **B.** The canopy almost (approximately 1.5 m) directly above the foraging site

four probes (at approximately the following times 20:02 h, 03:24 h, 03:25 h and 03:26 h) and two gapes were observed (approximately 21:25 h and 01:07 h). One strike was observed (approximately 20:19 h), but no prey was visible for more than four hours prior nor any other organism which could have elicited a defensive strike.

Potential prey observed on camera included geckos (*Cyrtodactylus* and *Hemidactylus* sp.), a squirrel, a bird (passerine) and rats. No previous predation observations of wild *T. kanburiensis* have been published, but captive specimens have been successfully fed lizards and pinky mice (Cox et al., 2012). Potential prey was primarily observed at night which largely overlapped the viper's foraging period (Fig. 3C), although it is worth noting that the squirrel, bird and rats were likely too large for the viper to ingest. Nine species of snake (including a road-killed adult female *Trimeresurus albolabris*) and six species of lizards (including signs of monitor lizards *Varanus nebulosus*) were observed in Khao Laem National Park during the study; adult *Cyrtodactylus* sp. (undescribed), a juvenile *Acanthosaura crucigera* and an adult *Lycodon davidsonii* were observed within 15 m of the *T. kanburiensis* (summarised in the Table at osf.io/2sh3m), and frog *Micryletta erythropoda* was observed within 5 m.

The immediate habitat of the *T. kanburiensis* matched previous descriptions for the species, i.e. limestone karst interspersed with bamboo. However, the viper's ambush site was within 30 m of a well-traveled paved road with one lane going in each direction (total width of the road approximately 10 m) which has not been previously reported. Similar habitat was also present on the opposite side of the road. The termite mound (Fig. 4A) where the *T. kanburiensis* was sheltering and waiting in ambush appeared to be old or abandoned, and no termites were visible; the overall appearance of the mound indicated that the termites might be of the genus *Macrotermes*. Two potential species are *Macrotermes cabonarius* or *Macrotermes annadalia*, which can be differentiated by soldier head color (black or red, respectively; personal communication, Dr. Warin Boonriam).

Canopy cover at the microhabitat level (Fig. 4B) was 66.2% approximately 1.5 m away from the viper. It was not possible to make an estimation directly above the viper due to it being present during monitoring at the same site; attempts would have potentially disturbed the viper, which was prohibited under the study research and ethics permissions. The canopy cover at the point of measurement appeared to be fairly similar to that directly overhead the viper.

**Review of trade threats**

The only record of any *Trimeresurus*/green pit vipers being reported or seized by Thai authorities (DNP) was in 2009 (56 live, 240 dead *T. trigonocephalus*; non-native to Thailand if correctly identified). Interestingly, there were 20 records provided by CITES of *Trimeresurus* between 2013–2020, one of which was listed as '*Trimeresurus* sp.' (labelled 'specimen') in 2017, the other 19 records being designated specifically as '*Trimeresurus mangshanensis*' which is listed under CITES Appendix II. LEMIS 2000–2014 records list '*Trimeresurus* sp.' (346 records), '*Trimeresurus albolabris*' (40 records), '*Trimeresurus gramineus*' (2 records), '*Trimeresurus hageni*' (2 records), '*Trimeresurus jerdoni*' (3 records), '*Trimeresurus popeiorum*' (3 records), '*Trimeresurus puniceus*' (5 records), '*Trimeresurus purpureomaculatus*' (2 records), '*Trimeresurus stejnegeri*' (1 record) and '*Trimeresurus trigonocephalus*' (4 records). Note that the nomenclature used by LEMIS does not reflect current taxonomy. Just 6 (all '*Trimeresurus* sp.') records listed Thailand as a country of origin, which were being shipped to 2 natural history museums (3 records) and a university in the United States (3 records).

Despite conservative categorisations in international (IUCN) and local (Thailand) Red Lists, it is worth pointing out that these assessments do not imply legal or enforcement capabilities. With few exceptions, Thai wildlife trade law appears to follow CITES for international trade. However possession is regulated by the Thai Wild Animal Reservation and Protection Act (WARPA B.E. 2535, 1992; and WARPA B.E. 2562, 2019)(Moore et al., 2016), which does not list *T. kanburiensis* (although there was documented discussion about it at the 2014 and 2015 CITES meetings; CITES AC27 Inf. 7; CITES AC28 Doc. 14.3). Personal communication does indicate potential inclusion in the WARPA edition currently being drafted.

## DISCUSSION

Our single observation of a *T. kanburiensis* did conform to the bamboo and limestone karst habitat use of the species, but the close proximity to a paved and well-used road does present unique future study opportunities in the form of genetics, mortality and general influence of human disturbance. Camera study, a relatively non-invasive method, appeared similar in success, namely in the identification of key ecological topics such as behaviour and potential prey, to prior study of green pit vipers in natural and human-dominated areas (Barnes & Knierim, 2019; Barnes et al., 2020). This success can be attributed at least partly to the fact that these snakes are ambush foragers and relatively resilient to human proximity disturbance, factors that may be important for the design of effective conservation strategies. They may also have a bearing on local snakebite management efforts given that there is at least one documented severe bite case attributed to *T. kanburiensis* in the wild (Warrell et al., 1992).

The number of sites known to be occupied by *T. kanburiensis* has more than doubled since the last IUCN Red List assessment, although new sites (including ours) on private land, protected land, temples and even a university campus in Thailand, remain largely in the realm of local knowledge and have not been published in western journals. Furthermore, the geographical range of the Kanchanaburi pit viper may be greater than currently stated as it has been suggested that it extends into Myanmar (Malhotra & Thorpe, 2004; which was repeated in Chan-Ard, 2022), but this is unconfirmed. The survey effort required during our study to find a *T. kanburiensis* was substantial, indicating that occupancy analysis with detection probabilities and population estimation would be difficult to calculate reliably.

To date no ecological study of wild Kanchanaburi pit vipers has been published beyond initial description and subsequent expeditions for study of presence, which coupled with limited quantification of effort and descriptions of observations provide at best only partial (at worst, biased) knowledge of the basic natural history of the species. Similarly, enforcement and legal protection for *T. kanburiensis* is currently unclear or non-existent. To address these disparities, we recommend a natural history and trade study focused on *T. kanburiensis* and two phenotypically similar, and potential habitat specialist species, *Trimeresurus kuiburiensis* and *Trimeresurus venustus*. This should be followed by clear legislation for protection of the species both locally and internationally that will reflect that research.

## ACKNOWLEDGEMENTS

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# Conservation recovery of *Batagur kachuga* and *Batagur dhongoka* turtles in India: Development and operation of a head-starting facility

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**ABSTRACT** - In 2006, a long-term conservation project was initiated for the recovery of endangered turtles *Batagur kachuga* and *Batagur dhongoka* along the lower Chambal river. This is based on the collection of eggs from the wild, incubation in a riverside hatchery, and then for most hatchlings release back into the wild within 24 hours of hatching. A sub-set of these hatchlings are reared at the Turtle Conservation Centre, in a specially designed head-starting facility until they weigh up to 1 kg, before release into the wild. A detailed description is given of the design, development and operation of the head-starting facility. Up to 2020, the project had successfully head-started 786 *B. kachuga* and 66 *B. dhongoka*. Moreover, this head-starting facility has supplied two conservation colonies, consisting of 50 subadult *B. kachuga* within the species' historical range, Kukrail Gharial Rehabilitation Centre and Kanpur Zoological Park, both under the aegis of the Uttar Pradesh Forest Department. The long-term aim of the project is to retore *B. kachuga* to its former range.

## INTRODUCTION

The Chambal River, within the National Chambal Sanctuary (NCS), holds the last known wild population of red-crowned roofed turtles *Batagur kachuga* (Fig. 1) and possibly the most viable wild population of the three-striped roofed turtle *Batagur dhongoka*, both Critically Endangered species. The drastic decline in the population of *Batagur* spp called for immediate recovery options for these two important turtles of the Gangetic plain. Following the recommendations of the 'Conservation Action Plan for Endangered Freshwater Turtles and Tortoises of India' (CFH/MCBT, 2006), recovery efforts were initiated in 2006 with a specific goal of raising juvenile survivorship through nest protection, hatch-and-release at riverside hatcheries, and a head-starting initiative at a nearby facility for future conservation and research initiatives.

A total of 0.35 acres of land within the protected area limits of the NCS, along the lower stretch of the Chambal River and vegetated with Mexican mesquite *Prosopis juliflora*, was procured for the development of the rearing facility and complimentary structures to support project activities (Fig. 2). The existing nursery facilitated the rearing-and-release of, on average, 100 hatchlings every year between 2006–2010. However, a larger facility was required to rear juvenile *B. kachuga* until they attained a weight of up to 1 kg, so that various size classes could be supplemented. Moreover, some of the individuals were used to develop conservation colonies within the distribution range of the species. This article focuses on our experiences of developing the field based retention and rearing facilities.

**Figure 1**. Adult female red-crowned roofed turtle *Batagur kachuga* from the lower Chambal river which supports the last known population of this species

## MATERIALS & METHODS

### Riverside Turtle Hatcheries

Between February to May every year, there is a *Batagur* nest protection programme. In 14 nesting seasons (2006 through 2020), the programme has successfully protected over 6,500 vulnerable nests of *B. kachuga* and *B. dhongoka*. All nests were translocated, as per appropriate procedures (Başkale & Kaska, 2005), and incubated in riverside hatcheries along the Chambal River. Methods were refined every year with the experiences gained from this project. At the hatchery,

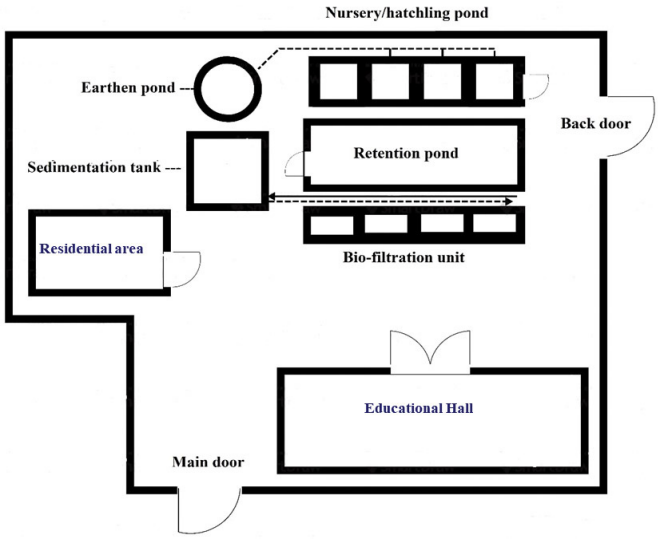


**Figure 2**. A diagram showing the final design of the ponds and facilities of the Turtle Conservation Centre (TCC) used for the head-starting programme, near Garhaita village, National Chambal Sanctuary, Uttar Pradesh

**Figure 3**. Hair-pin shaped hatchling nursery, this is the initial design which was superseded by the four separate tanks

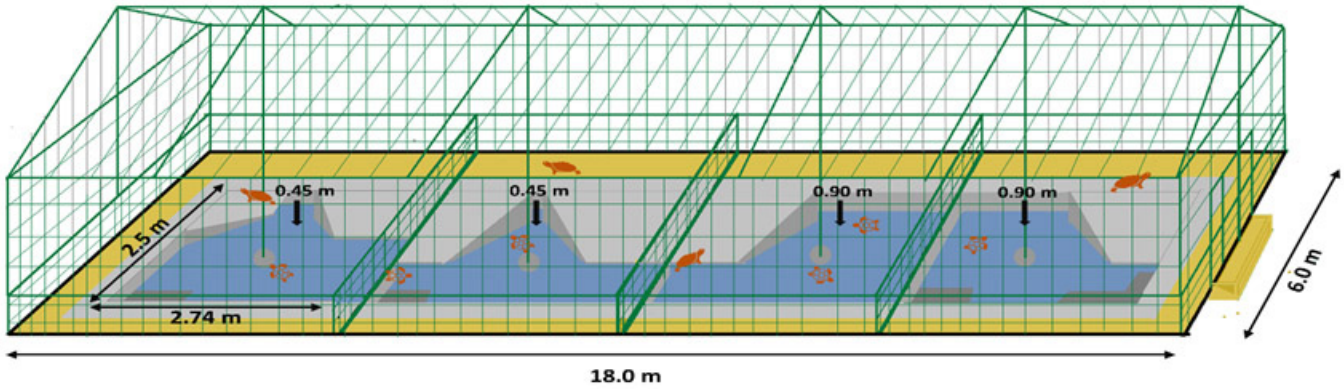


**Figure 4**. The improved hatchling nursery of four separate tanks with a margin of sand (in yellow) provided as a basking/burrowing area

clutches are incubated in holes excavated 1 m apart and buried at the same depth and placed in the same orientation as recorded for each wild nest. The incubation temperature in these outdoor hatcheries simply follows ambient conditions. Over 120,000 hatchlings have been released at the precise location of nest collection in the Chambal River. The majority of these hatchlings are released within 24 hours of hatching. After hatching, a subset of 50–100 *B. kachuga* hatchlings are moved to the head-starting facility of the Turtle Conservation Centre (TCC) (Fig. 1) for the short term (1–2 years) head-starting programme. In early years (2006–2007) an additional set of 100 hatchings of *B. dhongoka* were head-started at the facility.

**Head Starting Facilities**

On arrival at head-starting facility the hatchlings are placed in the hatchling nursery.

**Hatchling nursery**

The first hatchling nursery developed in 2006 was hairpin-shaped (Fig. 3) but in 2009 this was reconstructed as four traditional square tanks as it had been challenging to maintain the turtle cohort in the former structure due to its shape. A set of four tanks was developed within the enclosure with a surrounding strip filled with river sand (Fig. 4). All four tanks were constructed in a row with a single service entrance. Two of the tanks (2.5 m long x 2.7 m wide x 0.9 m deep) closest to the service access have a capacity of ~6.7 $m^3$ of water and are used for the retention of a maximum of 50 hatchlings (7.5 hatchlings/$m^3$) for two months. The other two tanks (2.5 m long x 2.7 m wide x 0.45 m deep) with a capacity of ~3.3 $m^3$ are used to isolate weak turtles, holding a maximum of 30 turtles each (9 hatchlings/$m^3$). All tanks are separated from each other using iron mesh (1.25 cm x 1.25 cm) frames to avoid mixing of hatchlings. The sand-filled areas (2.5 m x 1 m) by the tanks are used for basking/burrowing and access to them from the water is by mounting a stairway (Fig. 5) which itself allows turtles to rest at different levels in the water. In the initial year of the project, the hatchlings were observed burrowing in the sand areas during the first few weeks. We kept this sand moistened in order to avoid any unwanted desiccation of hiding hatchlings but, despite this

**Figure 5**. A stepway in each nursery tank allows turtles to thermoregulate at different water depths and to access the sand margins of the enclosure

care, seven of the hatchlings that stayed longer in the sand and did not return to water became desiccated and died. From 2016–2017, hatchlings were shifted to the retention pond after monsoon to provide them with a large area with varying depths to thermoregulate and swim prior to the winters. Since 2018, hatchlings were retained only for two weeks in nursery and have been prevented from leaving the water and burrowing in the sand margin by the construction of a temporary brick wall at the highest step of the stairway.

The water quality in nursery tanks is maintained by simple drain and fill processes. During the initial phase of the project (2006–2009) this was done once a month. The tanks are filled with ground water using a submersible pump. Each tank has a drain outlet (10 cm diameter) at its deepest end to allow complete drainage of waste water, which is collected in a shallow earthen pond created behind the facility. This water is used to grow aquatic plants such as water spinach *Ipomoea aquatica,* to feed to the turtles reared in the facility. Tanks are further equipped with an outlet to drain excess rainwater

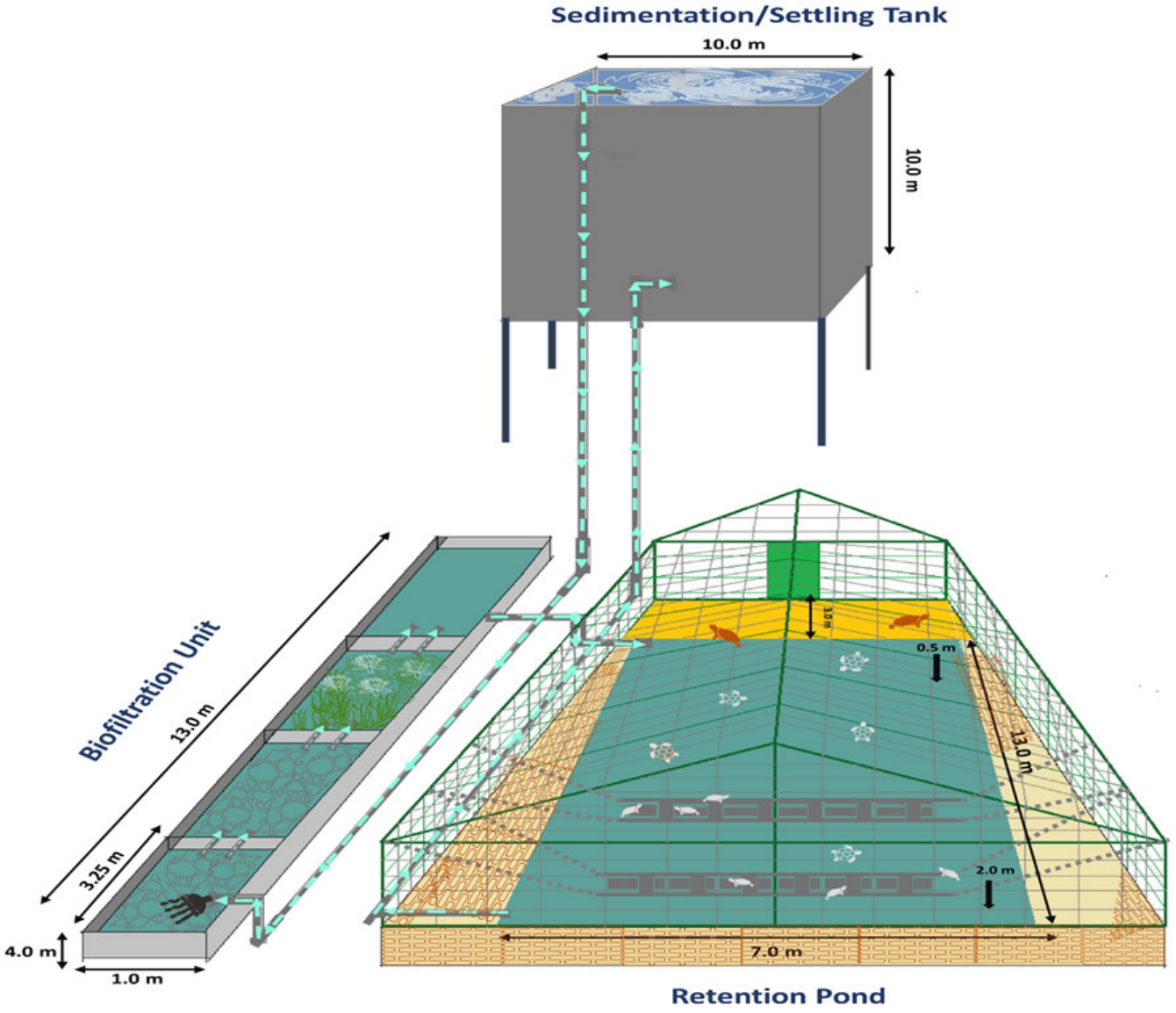


**Figure 6**. Large retention pond with a water circulation system to rear turtles for the head-starting programme, the far end of the enclosure has a sand filled area for turtle basking and a service entrance. For full layout see Figure 2.

during the monsoon. All pipework and the pump are provided with nets on both ends to prevent any turtles entering them accidentally. Since 2010 water quality in the tanks has been monitored using a Jal-TARA mini water testing kit (Development Alternatives) to test basic parameters such as pH, total dissolved solids (TDS), chloride, ammonia and nitrite/nitrate. This testing indicated that from June to September the water quality was dropping below optimum levels and so for these months of the year the water is changed three times a month instead of once.

**Retention pond**

A large enclosure with a retention pond (13 m long x 7 m wide x 0.5–2.0 m depth) was developed mainly to retain the larger hatchlings and yearlings turtles (Fig. 6). The pond can hold approximately 136 $m^3$ of water and houses between 150–200 hatchlings (0.68$m^3$/hatchling) (Kuchling, 2000; Whitman, 2009; Kanghae et al., 2016). To ensure a robust concrete structure, which would withstand the temperature extremes of the region, the enclosure was built below ground level with 10 cm thick exterior walls and a 22.5 cm thick concrete base. The enclosure is bounded by a 3.75 cm iron mesh cage to make it predator-proof. A depth gradient was created in the pond from 0.5 m to a maximum of 2 m to create a temperature gradient. During the summer, water temperatures at the shallow end range from 25–28 °C in the daytime to 20–22 °C at night time; the deep area remains more or less a constant 25–29 °C. Two floating platforms (3.6 m x 0.5 m), made out of split bamboo and stone tiles, are anchored in the middle of the pond where turtles can bask on top or hide beneath them (Fig. 7). These platforms are tethered and adjust themselves according to the prevailing water level. A sand-filled area (3 m x 4.5 m) is also allocated at the shallow far end of the pond as a disturbance-free basking space, the same end as the service entrance.

**Feeding the turtles**

The captive turtles are fed a balanced diet consisting of fish, soya and fresh vegetables, such as pumpkin, bottle gourd, cucumber, mulberry leaves, aquatic macrophytes (mainly *Chara* species, *Hydrilla* species), as well as a feed prepared from small fish and Pedigree© dog food to which is added multi-vitamin (Mayvit T Syrup) and calcium supplements (Calcwin T Syrup) at the rate recommended by the manufacturer.

**Water supply and circulation to the retention pond**

Regular water changes of the retention pond were problematic due to its large water capacity, the scarcity of water in this semi-arid region and the irregular supply of electricity. Consequently, it was necessary to maintain optimum water quality using a water treatment system. This is powered by a solar-powered 1 HP submersible pump (22 TECH 12v DC pump with 700 litres/hour pumping capacity) equipped with 6 solar panels; in sufficient sunlight one panel produces 250 watts/h (i.e. 250 x 6 = 1500 watts/h used to operate the 1 HP pump). The system is equipped with a battery pack that acts as both storage and additional start-up power to the unit. The solar panels are installed on the top of the sedimentation tank (Fig. 6). Water is pumped into the overhead settling/

**Figure 7**. Floating platforms suspended in the retention pond on which turtles could bask or alternatively hide beneath

sedimentation tank (3 m long x 3 m wide x 3 m high), through a 2.5 cm diameter pipe, for settling coarse suspended solids, prior to the treatment through the biofiltration system. Supernatant from the tank passes through an outlet pipe to the biofiltration unit. A valve operated at the base of the tank allows it to be drained for cleaning and an area below the tank is used occasionally to incubate turtle nests outside the *Batagur* hatchery season and prepare the turtle feed.

The biofiltration unit (13 m long x 1 m wide x 4 m high) has 22.5 cm thick walls and is divided into four chambers, built parallel to the retention pond (Fig. 6). The chambers are connected to each other by means of small circular openings. Supernatant from the sedimentation tank is discharged directly into the first tank and subsequently passes through the three interconnected tanks before returning to the retention pond. In order to improve aeration, water from the settling tank is trickled over the filter media in the first chamber using shower-heads. The first two chambers function as rough filters containing chiefly brick ballast and coarse gravel as filter media to retain suspended solids. The third and fourth chambers contain water hyacinth *Eichornia crassipes* which helps to remove excess nitrogen from the system and also contain small fish (guppy, zebra fish, gourami and Indian potasi) to be used as supplemental feeding for the turtles. At night, when solar panels are not functional, outflow is restricted to a 1.25 cm pipe that reduces the influx of filtered water and so maintains the overall circulation.

**Winter preparations**

The winter period starts in mid-November and by December–January temperatures drop to 1–2 °C at night and most days are foggy with limited sunshine; by mid-February ambient day time temperatures have returned to 29–30 °C (Singh S., personal observation, 2006–2010). At the beginning of the project during the cold period, we experienced mortalities and pneumonia, eye swelling and other infections in the turtles. To prevent these, the retention pond is insulated using thick plastic sheets from the beginning of November to retain heat during chilly nights. Overnight, the water temperature in the retention pond may drop by several degrees (10 to 11 °C). Additionally some freshwater was added on cold nights to achieve optimum temperature. Further, two room heaters of

1 kg watt are employed to regulate temperatures within the enclosure, especially on days with prolonged overcast skies (usually late December, early January). On sunlit days the sheets are removed so that turtles can bask in the sunlight, which also helps to increase the temperature of the pond water. Finally, the sheets are removed completely by mid-February.

**Summer preparations**

The peak summer (May–June) temperatures can often reach 48 °C. To prevent the pond water becoming too hot, the cage enclosing the western side of the retention pond is covered with a green woven sun cloth to prevent direct sunlight exposure, while on the eastern side the cage is not covered so that the turtles have access to morning sunlight for basking. The sun cloth is made of 'breathable' fibres that facilitate temperature exchange.

## RESULTS

**Impacts over the Years**

Initially, during the head-starting period, all individuals were kept together in nursery/hatchling ponds for the first two months and this practice of mixing hatchings of different age groups/size classes was compromising their health and growth. Larger individuals were outcompeting smaller, and therefore younger, individuals for resources (basking preference, food, etc.). This competition led to health issues such as shell softening, eye infections, and injuries from conspecifics, which may lead to a higher rate of mortality. On average, 15–20% of the annually-retained individuals showed shell softening and 1–2% had signs of eye infections in the initial years of the project. The solution to this problem was to separate individuals in different nursery/hatchling tanks based on the batch, size and whether or not they were healthy. Eye infections may often be linked to poor water quality, hence we attempted regular monitoring of water parameters which led us to change the water more frequently. These measures helped us achieve significant growth rate, improve health condition, reduce infection (50-60% reduction in shell softening and eye infection cases) and reduced mortality.

Over the 14-year period, the project successfully head-started and released into the Chambal 786 *B. kachuga* and 66 *B. dhongoka* 1–2 year-old turtles. The mortality rate during head-starting between 2014–2020 was low, only 1.9% (14 *B. kachuga* & 3 *B. dhongoka*), benefiting from the addition of the retention pond and improvements in husbandry practice. From 2017, weight gain (Fig. 8) and body dimensions were routinely recorded for *B. kachuga*. Between 2017–2020, during the six-month head-starting period (June–November), the average total weight gain (± SD) by hatchlings (n = 100 per year) in 2017 was 58.75 ± 27.10 g, in 2018 was 69.82 ± 26.04 g, in 2019 was 78.34 ± 31.11g and in 2020 was 95.07 ± 38.05 g. In the four years (2017–2020), the average total weight gain (± SD) of hatchlings (n = 100 per year) was 75.50 ± 13.27 g. Similarly, the average straight carapace length (± SD) of hatchlings (n = 100 per year) (SCL) increased by 32 ± 4.12 mm, and the average straight carapace width (SCW) increased by 25 ± 5.24 mm.

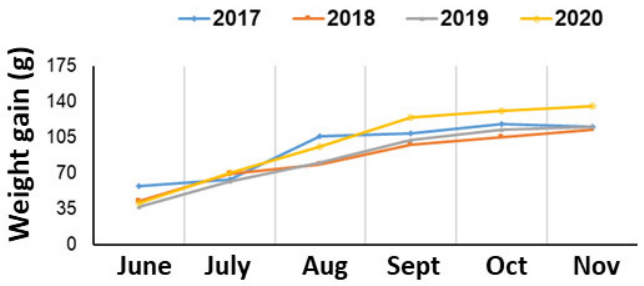


**Figure 8**. The average weight gain for *B. kachuga* hatchlings (n = 100) during the six month head-starting period from 2017–2020

**Establishment of *Batagur* Assurance Colonies**

In 2006, over 50 individuals were retained for the establishment of captive colonies at regional zoos (Lucknow and Kanpur) and a recognised captive breeding centre (Kukrail Gharial Rehabilitation Centre, Lucknow). Fifty subadults *B. kachuga* were selected – 30 went to Kukrail and 20 went to Kanpur Zoo in years 2015 and 2016, respectively.Of these subadults, only one was male, which was sent to Kanpur Zoo, suggesting that our stock has a significant female bias due to high temperatures during incubation. In 2020, ten animals from Kukrail were translocated to Nawab Wajid Ali Shah Zoological Garden, Lucknow, to create an additional conservation colony. These robust captive populations of this Critically Endangered species serve as a hedge against extinction and will aid in the recovery of its wild populations to their former strength. The Madras Crocodile Bank Trust has a successful conservation breeding of *B. kachuga* using the founders that are on breeding loan from Kukrail Gharial Centre, Lucknow. To redress the male imbalance the eventual goal of the project is to acquire more males from the Madras Crocodile Bank Trust for all three colonies in Uttar Pradesh and initiate captive breeding as part of an integrated conservation recovery programme for this species.

## DISCUSSION

In 2006, we initiated a head-starting programme with a basic rearing facility and subsequently developed it based on needs and experience. We made several important improvements including the development of a biofiltration unit to reduce water wastage and improve water quality. We diagnosed that the previously used hatching nursery needed to offer more housing space for the turtles and also that routine maintenance needed to be made easier in order to implement better hygiene. We did this by dividing the initial single facility into four square tanks that were easier to maintain and allowed an increase in stocking density. Additionally, a major issue faced during the initial years (2006–2010) was the high rate of mortality among rearing cohorts. To mitigate this problem, the husbandry practices were improved and individuals were separated in the nursery based on age class and size, and a balanced diet with micro-nutrient supplementation was provided. At the same time, regular monitoring of water parameters was started and both these steps helped to improve health and increase survivorship. In addition, health benefits have been obtained by maintaining the retention

pond in the optimum range of water temperatures (20–25°C) during the coldest and hottest times of the year.

The Chambal River has an estimated 500 adult nesting females (Sirsi et al., 2017; Praschag et al., 2019); if we assume a sex ratio of 1:1 then there should be around 1000 adult *B. kachuga* in the wild. In the last 14 years, we have successfully added a total of 722 individuals to the wild. A release of ten 8 year-old captive-raised *B. kachuga* along the middle Chambal indicated at least 70% survival for first two years. A release of 2.5 year-old *B. kachuga* individuals from this (TCC) facility in April 2023 shows 80% survival in the following four months (Singh et al., in preparation). Furthermore, our annual nest protection efforts indicate that there has been a gradual rise in numbers of nests in the area and an increase in the number of juveniles sightings.

Here, we have presented information based on the 14 years of work and the project is still in progress with new objectives and goals. Currently, we are evaluating the diet preference of hatchlings and juveniles under captive conditions, which will aid future captive propagation efforts. For *B. kachuga* the only surviving population is that on the Chambal River. The long-term aim should be to fully restore the species so we are currently scoping the reintroduction of *B. kachuga* to its historic range. Such reintroductions will follow strict guidelines and will be subject to prior assessment of the selected site with respect to food availability as well as potential population density. The biosecurity of the native species present in the release habitat will be safeguarded by comprehensive health assessment of release cohorts (Huber et al., 2022). To achieve all this will require further research. This should include investigation of the optimum duration of the head-starting/retention, the post-release survival rate and health performance in the wild, efficient method of release, either 'hard release' without pre-conditioning to the release environment or 'soft release' with pre-conditioning, and a suitable release site selection methodology.

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SHORT NOTE

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# Construction of a refuge wall with crevices to protect European leaf-toed geckos *Euleptes europaea* and young Turkish geckos *Hemidactylus turcicus* on the Ile du Levant, France

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## INTRODUCTION

Faced with the rise in invasive reptile species worldwide and the consequent threats to native species (Dubos et al., 2023), studying the requirements of the most vulnerable animals is crucial for their successful conservation management. Conservation translocations are being used increasingly for reptiles with positive results being reported (Evans et al., 2023) but a lack of understanding of their ecological requirements can lead to problems such as rejection of the release site that results in wandering, exhaustion or predation (Berger et al., 2020; Bradley et al., 2023). Two species of geckos, the European leaf-toed gecko *Euleptes europaea* (Gené, 1839) and the Turkish gecko *Hemidactylus turcicus* (L., 1758), have long been present on the Ile du Levant (Lantz, 1931; Ineich et al., 2019) a Mediterranean island in the Hyeres archipelago lying about 10 km from the south coast of France (43°01' N, 6°27' E). A third species, the Moorish gecko *Tarentola mauritanica* (L., 1758), which is considered to be problematic, has been introduced recently (Deso et al., 2018). In France, including Ile du Levant, these three gecko species are often the subject of translocation plans in response to land development projects. Although *T. mauritanica* is a nationally protected species (Arrêté national de protection du 8 janvier 2021), it is nevertheless considered a potentially invasive species in France (Zdunek, 2022a) and throughout the world (Rato et al., 2023). Its arrival on Mediterranean islands is being monitored (Médail et al., 2013; Deso et al., 2020), as its presence is a threat to native island species (Astruc et al., 2014). The colonisation of natural and man-made environments by *T. mauritanica* on the Ile du Levant has now reached the heart of the island (Deso et al., 2020).

Given the concerns about the potentially significant impacts of *T. Mauritanica* on small threatened species (Renet et al., in press), and the reported extinctions of isolated populations of *E. europaea* (Salvidio & Delaugerre, 2003), we undertook a rapid test of an experimental refuge wall for the two vulnerable gecko species as part of a development and translocation plan. In the case of the leaf-toed gecko, an abundance of deep, narrow crevices is a key factor in the species' survival (Salvidio & Oneto, 2008). Since September 2022, we have been supervising the construction of two 10-metre-long experimental walls with 200 narrow cracks that are conducive to the establishment of adult and juvenile *E. europaea*, as well as juvenile and subadult *H. turcicus*. At the beginning of December 2022, we relocated a total of 10 *E. europaea* into the experimental walls, these are the smaller and more threatened of the two species, to assess the site fidelity of this species and the visits and colonisation by other lizard species.

## MATERIALS & METHODS

It is known that *E. europaea* prefers refuges in which it would fit tightly such as narrow cracks in rocks (Salvidio & Oneto, 2008) or tightly packed tree bark (Deso et al., 2023). On the island of Port-Cros it has been reported that *E. europaea* can occupy particularly narrow cracks and interstices, ones to which *H. turcicus* does not have access (Delaugerre, 2003a). Given the depth of the skull of *E. europaea*, we considered that crevices of 0.4–0.7 cm would constitute a comfortable refuge for both young and adult specimens and from which it would be difficult for larger species of lizard to remove them.

To provide such crevices we constructed two walls each 10 m long and 70 cm high on 5–6 September 2022 using breeze blocks (50 x 25 x 20 cm) in three layers (Fig. 1). A numbering system was used to identify the layers (wall 1 A–C, wall 2 D–F) and the crevice positions (1, 2, 3 etc.). The centres of the breeze blocks were hollow and were filled with soil in which plants could grow (Fig. 1C). This plant cover provided additional hiding places for the geckos and food resources from the insects that the plants attracted. The walls were constructed with a south-south-west orientation, the same as the rock faces and low garden walls around the site, which were already heavily colonised by both gecko species. As favourable thermal properties of refuges are crucial for the persistence of populations (Bradley et al., 2023), we took care not only to reproduce an exposure appreciated by both gecko species, with the heat of the day making the walls warm and allowing the geckos to thermoregulate efficiently by thigmothermy during the night (Delaugerre, 1984; Salvidio & Oneto, 2008) but also embedded part of each breeze block, partially covered with soil, in the ground in order to form a barrier to strong thermal amplitudes (Fig. 1A). We inspected


**Figure 1**. Views of an experimental breeze block wall constructed on an excavated surface with the breeze blocks set apart to give standardised crevices between them, in which *Euleptes europaea* and young *Hemidactylus turcicus* could take refuge, as photographed in - **A.** September 2022, **B.** January 2023, **C.** June 2023

**Figure 2**. One of ten *Euleptes europae* located to an experimental wall

the area around the refuge wall to eliminate any shelters that might offer refuge to other larger lizards (and geckos) that could potentially be predators.

On 5 December 2022, ten adult *E. europaea* were captured during the demolition of an old asbestos building located 200 m from the experimental walls. The ten specimens were deposited in crevices of the walls (5 individuals in each wall) on the same day (Fig. 2). To monitor the crevices and observe the behaviour of the geckos in the crevices and around the walls, we used an L52 LED lamp and a video trap (Num'Axes Trail Camera PIE 1023) as well as an infrared thermal camera (Leica Calonox View). In total, we made one visit each month from January to December (= 12 visits), which involved a one-hour inspection of the numbered cracks to determine the presence/absence of geckos or other lizards. This protocol of inspecting numbered cracks, one after the other, means that there was no double counting of geckos and also allowed us to see whether the geckos were faithful to particular crevices.

## RESULTS

### Ageing of the wall

After four months of winter, the scrubland around the walls had regrown with many local plant species, such as strawberry tree *Arbutus unedo*, myrtle *Myrtus communis*, pistachio *Pistacia lentiscus*, thistles, etc. (Fig. 1C). The lower sections of the two walls, which were built on loose soil, had undergone movement and by April 2023 showed five crevices that had spread beyond 0.7 cm (in a localised sector of the wall) and 50 crevices that had closed completely, so that of the 200 original crevices 145 were still available as refuges for *E. europaea*.

### Monitoring of geckos

After the transfer of ten *E. europaea* at the beginning of December 2022, although we did not mark or photo-identify the individuals, we observed between four and eleven different adult individuals during 2023 (in approximately the same crevices) and five different juveniles that established themselves from August, September, October, November and December 2023 (Figs. 3 & 4). We noted the arrival of four different adult *H. turcicus* from May and three adult wall lizards *Podarcis muralis*, including a hibernating juvenile, in one of the cracks in December. The presence of other adult and larger lizard species is linked to the enlargement of the spacing of five crevices mentioned above, making it possible for them to be exploited by larger animals. Night-time video surveillance during the night of 21 June in a section of wall where only 0.4–0.7 cm crevices were present enabled us to observe their usage by *E. Europaea* and the prospecting of three adult *H. turcicus* unable to enter these crevices (BHS video, 2024). *Euleptes europaea* showed a certain willingness to retreat into the crevices in the face of repeated visits by

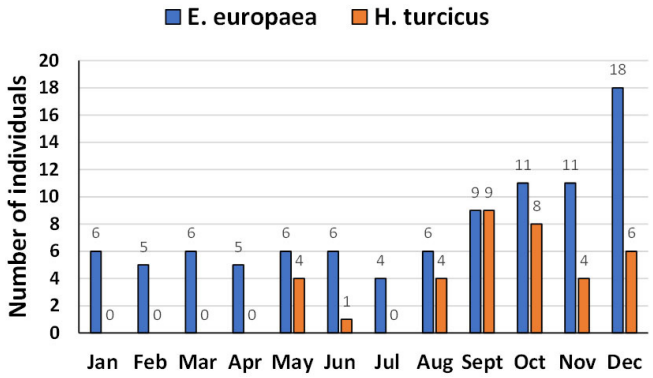


**Figure 3**. Monthly count of individual *Euleptes europaea* and *Hemidactylus turcicus* in the two experimental walls in 2023

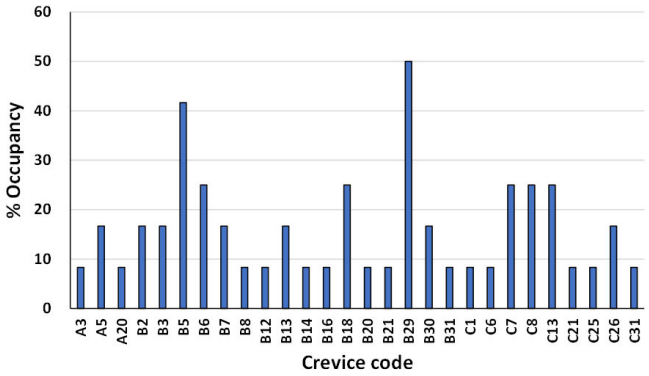


**Figure 5**. % occupancy by *Euleptes europaea* of the crevices of experimental wall #1 from January to December 2023

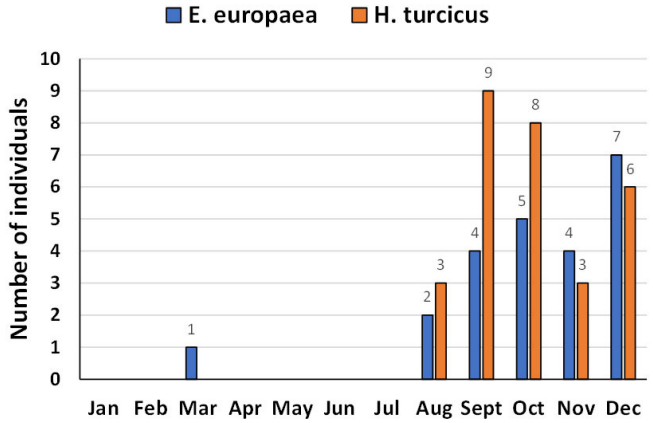


**Figure 4**. Monthly count of individual juvenile *Euleptes europaea* and *Hemidactylus turcicus* in the two experimental walls in 2023

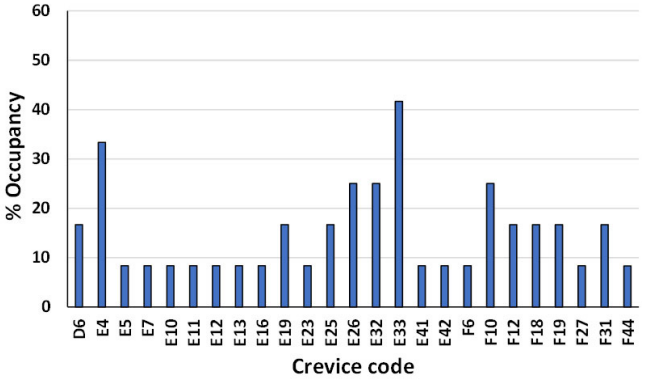


**Figure 6**. % occupancy by *Euleptes europaea* of the crevices of experimental wall #2 from January to December 2023

adult *H. turcicus*. On several occasions between 22:40 h and 00:30 h, *H. turcicus* attempted to interact by positioning itself around the same crevices but without being able to enter them (BHS video, 2024). Juveniles of both *E. europaea* and *H. turcicus* were observed at the walls mostly in August and September (Fig. 4). Out of 145 crevices considered favourable, 23.5% were occupied by six *E. europaea*. Crevice occupancy rates in the two experimental walls sometimes exceeded 40% (Figs. 5 & 6) and crevice B29 in wall 1 even reached an occupancy rate of 50% for *E. europaea* (Fig. 5).

## DISCUSSION

Sympatric lizard species may coexist by adopting different activity patterns and niche partitioning (Luiselli & Capizzi, 1999; Simbula et al., 2018; Radi & Zuffi, 2022; Zdunek, 2022b). In the case of the gecko species of concern in this study, when sharing the same habitat (syntopia), *T. mauritanica* may displace *H. turcicus* towards more vegetated areas (Lisičić et al., 2012) and it is suspected that *E. europaea* will move towards denser forest environments in response to *H. turcicus* (Deso et al., 2023). To facilitate monitoring of *E. europaea*, artificial refuges have already been built on certain Mediterranean islands, such as piles of stones on Port-Cros (Delaugerre, 2003b) and tiles on the island of Grand Rouveau (Cheylan et al., 2018) and the encouraging results of those studies have led to the construction of refuges using tiles and piles of stones on Ile du Levant (pers. comm.). However, it is now known that these refuge structures are easily damaged by wild boar following their arrival on the islands of Hyères (Ballouard et al., 2021), and potentially result in the predation of the geckos when they turn over the piles of stones and tiles. There are already wild boar present on Ile du Levant and we observed wild boar dung close to the walls throughout the year but they remained undamaged so we conclude that the breeze block walls are not vulnerable to wild boar.

Successful measures to manage gecko populations by creating appropriate crevices in solid walls have been implemented in south-east Australia (Webb & Shine, 2000). We have therefore opted for this strategy using a standardised crevice size (0.4–0.7cm) favouring *E. europaea* and also juvenile and subadult *H. turcicus*.

Our translocation of *E. europaea* at the beginning of winter was successful in that most of the geckos took up residence in the refuge walls and over the winter the flora gradually colonised the new habitats providing further cover. During the video capture of 21 June, *E. europaea* showed its eagerness to withdraw into the crevices in the presence of adult *H. turcicus* that attempted to interact by positioning themselves around the same crevices (but without being able to enter them). The nocturnal movements of three adult *H. turcicus* across the ground to the refuge wall, despite the lack of shelter

(caches for large geckos had been removed), indicates the considerable dispersal ability of this species.The number of juvenile *H. turcicus* that found and colonised the refuge walls was greater than that of *E. europaea*, which appears to corroborate this observation (Fig. 4). This avoidance of *H. turcicus* by *E. europaea* suggests limitations to cohabitation between these species that may result in the displacement of *E. europaea* (Lisičić et al., 2012). However, the persistence of adults and juveniles of *E. europaea* for a whole year demonstrates the advantage of a refuge that prevented the intrusion of adult *H. turcicus*. Habitat supplementation and revegetation are important parameters for increasing the densities of protected species (Goldingay & Newell, 2017; Mickael et al., 2018) but also for increasing the chances of successful translocations (Bradley et al., 2023). Here, we tested the effectiveness of a refuge wall to separate two gecko species whose nature of interaction is still uncertain, prior to the arrival of an even more aggressive invader to the site, *Tarentola mauritanica*. Although the walls showed a few points to be corrected, such as the fact that they should be established on a solid base to prevent them from moving over time, the initial results are encouraging for the development of different strategies depending on the life stages of the different gecko species living in syntopia. It is a positive sign that the experimental walls allowed the establishment of 11 *E. europaea* adults (Fig. 3) for a whole year (including one winter), as well as the colonisation of seven juveniles making a total of 18 European leaf-toed gecko with eight individuals in addition to those originally displaced. Some crevices in the two experimental walls reached a high occupancy rate of over 40%, and sometimes as high as 50% over the year. The strong colonisation by young *H. turcicus* (nine juveniles) in September 2023 shows that they also feel the need to exploit shelters that offer them protection at this stage of their lives. Given that adult *H. turcicus* were unable to penetrate the wall crevices and potentially dislodge adult and juvenile *E. europaea* or young *H. turcicus*, it may be assumed that the same crevices would also protect these species from *T. mauritanica*. Our results clearly show the usefulness of this type of structure for *E. europaea*, which showed an increase in the presence of individuals (18). The presence of 11 adults throughout the year, as well as the autumn colonisation and overwintering by juveniles (7), also show the usefulness of this type of refuge for the hibernation of this species.

Despite the presence of juveniles, at this stage we do not know whether the refuge walls may have served as an egg-laying site for *E. europaea* which has the habit of using cracks to lay its eggs (Delaugerre, 2003; Salvidio et al., 2010). This is of particular interest as the availability of egg laying sites is a key factor in the survival of gecko species such as *E. europaea* that lay hard-shelled eggs (Salvidio & Oneto, 2008); it would be useful to pay particular attention to this in further research.

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SHORT NOTE

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# Direct development of the Reza Khan's bush frog *Raorchestes rezakhani* in north-east India

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**ABSTRACT** - There are currently 76 species of frog in the genus *Raorchestes*, these undergo direct development, i.e. there is no free living tadpole stage, instead a froglet emerges from the egg. We documented the development of the recently described Reza Khan's bush frog, *Raorchestes rezakhani*. Twenty-three eggs were laid in a hole in a tree stump about 1.5 m above the ground. On average the eggs were 5.5 ± 0.26 mm in diameter and weighed 0.9 ± 0.07 g. We describe six stages of development at temperatures between 23–26° C and relative humidities of 73–86%. The froglets emerged from the egg 13 days after the embryo had developed head, tail, limb structures and body pigmentation.

## INTRODUCTION

Anurans have the greatest diversity of reproductive modes among tetrapod vertebrates, with at least 42 being currently recognised (Malagoli et al., 2021). The majority of frogs lay eggs from which free living aquatic larvae (tadpoles) hatch. These tadpoles metamorphose into froglets that resemble the adult. However, 27.2% of known anuran species (1,928) have adopted 'direct development' (Liedtke et al., 2022) where there is no free living larval stage, instead the embryo develops directly into a froglet that then hatches from the egg (De Lima et al., 2016; Schweiger et al., 2017; Goldberg et al., 2020). Direct development may be favoured where environmental conditions do not provide suitable aquatic habitats for tadpoles (Liedtke et al., 2017; Salica et al., 2023).

The family Rhacophoridae contains 76 species of small-bodied, terrestrial bush frogs in the genus *Raorchestes* (Biju et al., 2010), that are found from southern India to Nepal and in mainland south-east Asia (Biju & Bossuyt, 2009; Vijayakumar et al., 2016; Frost, 2023). There is little data on clutch sizes and development times for the few species that have adopted direct development. Here we report the direct development of the little known Reza Khan's bush frog *Raorchestes rezakhani* Al-Razi, Maria & Muzaffar, 2020, a small cryptic bush frog with a mean snout-vent length (SVL) of 20.06 ± 0.87 mm, originally described from north-eastern Bangladesh based on morphological characters, genetics and bioacoustics (Al-Razi et al., 2020). This species was later discovered in various locations in Mizoram, India (Decemson, 2021; Hlondo, 2021) where it was found mainly on the edge of streams and near man-made trails in primary and secondary forests. During the monsoon season, it frequently uses hilly slopes for calling, where individuals perch on leaves and branches of small trees and on bamboo trunks (Al-Razi et al., 2020; Decemson et al., 2021; Hlondo, 2021).

**Figure 1**. Adult female *Raorchestes rezakhani* (MZMU 2173)

## METHODS

On 25 May 2022, during a herpetofaunal survey along stretches of the Sipai stream, Lunglei, Mizoram, north-eastern India (22° 53'18.7908" N, 92° 45'37.1046" E, 817 m a.s.l.), an adult female *Raorchestes* (Fig. 1) sitting near a developing clutch of eggs was found and collected at a depth of 7 mm inside a hole in the stump of Chinese albizia *Albizia chinensis* about 1.5 m above ground (Fig. 2 A&B). The specimen was preserved and deposited in the Departmental Museum of Zoology, Mizoram University (catalogue number MZMU 2173). The site was marked and covered with nets to keep out predators. The air temperature and relative humidity were measured using KusamMeco KM-918 Thermo


**Figure 2**. Breeding site of *Raorchestes rezakhani* in a hole in a tree stump by the Sipai stream, Lunglei, Mizoram - **A.** Tree stump covered with net to protect the eggs from predators, **B.** Close up view showing the eggs within the hole in the tree stump

**Figure 3**. *Raorchestes rezakhani* eggs after transfer to a plastic terrarium under natural light with organic soil, mixed vegetation, and leaf litter to maintain humidity and temperature

Hygrometer between 25 May and 6 June 2022. Though we are not sure at what time and date these eggs were laid, it was confirmed that the microhabitat was recently utilised for breeding by this species, as calls of this species were heard frequently in this specific area during the previous survey. The study site was visited daily until 31 May 2022. Following that, eggs with developing embryos were collected and transferred to a plastic terrarium (Fig. 3) to observe and document the development of the species until froglets emerged. The plastic terrarium (23 x 20 x 12 cm) had natural lighting and was provided with organic soil, mixed vegetation and leaf litter to maintain the relative humidity at 73–86% to trigger development. Eggs were misted daily with water and humidity inside the terrarium was carefully monitored. Room temperature ranged between 23–26 °C as per ambient conditions. The stages of the eggs were classified by comparing them to images of the various stages provided by Bahir et al. (2005). The dimensions and weight of the eggs and frogs were recorded using Mitutoyo Vernier Caliper and MH Series pocket scale, respectively.

## RESULTS

The adult frog that we collected was confirmed as *R. rezakhani* based on the original description provided by Al-Razi et al. (2020) which includes - relatively small size with a SVL of 21.36 mm, indistinct tympanum with supratympanic fold weakly distinct, both inner and outer metacarpal and metatarsal tubercles absent; in addition, genetic studies (Gen Bank accession no. OR482067) showed only 1.80% genetic divergence from the holotype (Gen Bank accession no. MN072375). From the date of encounter until emergence from the eggs, *R. rezakhani* developmental stages were documented. Within the vitelline membrane the egg developed, and a juvenile frog emerged. There were a total of 23 eggs, with diameters ranging from 4.82–6.24

**Table 1**. Observations of embryonic development in *Raorchestes rezakhani* in May/June 2022. The stages of the eggs were categorised by comparing the eggs to photographs of the various stages provided by Bahir et al. (2005).

| Observation period day/date | Stage | Developmental Characteristics |
|---|---|---|
| Day 1<br>25 May | 1 | Unpigmented subdermal eyes visible; head become prominent; tail elongated and dorsal and ventral fin transparent; body pigmentation appears on dorsal and lateral body surface; forelimb elbows emerge. |
| Day 4<br>28 May | 2 | Cornea transparent; eyes pigmented; pigments denser on head, snout and dorsum; toe indentations visible. |
| Day 7<br>31 May | 3 | Toes separated; expanded abdominal folds on venter; pigmentation progresses from the head to the tail region of the body; eyes more defined; iris black. |
| Day 10<br>3 June | 4 | Pigmentation on abdomen; hands clearly distinguishable on forearms; head region longer; eyes larger; hindlimbs lengthen, tips of toes enlarged; tail fin fully developed; upper eyelid visible. |
| Day 12<br>5 June | 5 | Most parts of tail have resorbed; dorsal and ventral fins disappear; mouth fully developed; body greyish brown; abdominal folds disappear; head and jaw developed and fully structured; body is stouter; subarticular tubercles under fingers/toes developed. |
| Day 13<br>6 June | 6 | First hatchling emerged; dorsal colour greyish brown with some small dark brown/ grey spots; froglet has internal yolk sac; a small tail tip remains. |


**Figure 4**. Various developmental stages of *Raorchestes rezakhani* - **A.** Eggs with embryos in which the head and tail area demarcated and body pigmentation developed, **B.** Subarticular patches developed on fingers and toes, **C.** Embryo fully developed, yolk visible inside abdomen, **D.** A newly hatched froglet

mm (5.5 ± 0.26 mm) and weight 0.82–1.26 g (0.9 ± 0.07 g). The six major features of development of the froglet are described in Table 1, with some stages illustrated in Figure 4. At the time of observation, the embryo was encased in a clear gelatinous capsule and had already attained a well demarcated head and tail as well as limb structures. Body pigmentation developed and the tail increased in length with a transparent blade. Subsequently, the eyes developed a black iris and translucent cornea. Toes separated and sub-articular patches became visible. Finally, the tail shrunk and then the first froglet hatched on the 13$^{th}$ day from the start of observation; the last hatchlings emerged three days later. The tail remnant was resorbed within the next two to three days. From 23 eggs, 17 froglets hatched successfully and 12 of these were released in the forested area in the University campus, while the remaining five were preserved as reference specimens in the Departmental Museum. The SVL of the froglets range from 5.51–5.98 mm (5.7 ± 0.17 mm, N = 5).

## DISCUSSION

The embryonic development of only a few *Raorchestes* spp has been described due to the difficulty of finding their eggs in the natural environment. Like its congeners *Raorchestes huanglianshan* (Zhang et al., 2022) and *Raorchestes longchuanensis* (Yan et al., 2021), owing to direct development *R. rezakhani* embryos did not develop many of the structures associated with tadpoles, such as external gills, tadpole teeth or a coiled intestine, and the front and

hind limbs developed nearly simultaneously. Interestingly, the newly hatched froglet (Fig. 4D) still had a small tail tip.

*Raorchestes* spp are known to lay eggs in a variety of locations. Bush frogs such as *Raorchestes bombayensis* (Bossuyt et al., 2001), *Raorchestes glandulosus* (Krishnamurthy et al., 2002), *Raorchestes nerostagonaq* (Biju & Bossuyt, 2005a), *Raorchestes bobinger* (Biju & Bossuyt, 2005b) and *Raorchestes chalazodes* (Seshadri et al., 2015) have been observed laying eggs on the upper side of plant leaves or in deep holes in trees. *Raorchestes rezakhani* appears typical of this group by spawning in a hole in the stump of a tree. In contrast, some bush frogs have been recorded laying their eggs in grass clumps or in soil, e.g. *Raorchestes graminirupes* (Biju & Bossuyt, 2005b), *Raorchestes resplendens* (Biju et al., 2010), *Raorchestes ghatei* (Padhye et al., 2013) and *Raorchestes tinniens* (Princy & Kannan, 2018).

The clutch size of *Raorchestes* may vary between species. We observed a clutch size of 23 in *R. rezakhani* while *R. huanglianshan* has been reported laying 13 eggs (Zhang et al., 2022) and a pair of *R. longchuanensis* in amplexus laid eggs twice within one month; the first clutch included 12 eggs, and the second included ten eggs (Yan et al., 2021), *R. glandulosus* has been reported to lay a clutch of 14 eggs (Krishnamurthy et al., 2002), and *R. tinniens* a clutch of 30 eggs (Princy & Kannan, 2018).

In our present work, the developmental period of *R. rezakhani* is uncertain as we did not observe the eggs from the time of egg laying but only from a stage at which the embryos were already relatively advanced with head, limbs and tail. In other species under both natural and captive conditions the period for oviposition to hatching has been 22–28 days in *R. glandulosus* (Biju, 2003; Krishnamurthy et al., 2002), 17–19 days in *R. tinniens* (Princy & Kannan, 2018), in captivity alone 25–30 days in *R. longchuanensis* (Yan et al., 2021) and 31–35 days in *R. huanglianshan* (Zhang et al., 2022).

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# Discovery of the high-Andean anole *Anolis tetarii* (Squamata: Anolidae) in the Páramos of northern Colombia

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**ABSTRACT** - We present the fourth documented specimen and the first record of *Anolis tetarii* in Colombia, the least studied member of the *Phenacosaurus* clade of *Anolis* lizards. We showcase the first published photographs of the species, provide a succinct diagnosis, and discuss morphological variations compared to type specimens, and the species conservation status.

The genus *Anolis* Daudin, 1802 comprises the most diverse genus of terrestrial tetrapods, totalling 438 currently recognised species. These species are found across a geographical range that spans the south-eastern United States through Central America, South America and the Caribbean islands. Presently, 79 species of *Anolis* have been recorded in Colombian territory (Uetz et al., 2023). The *Phenacosaurus* series constitutes a clade of highland *Anolis* lizards within the northern Andes of Colombia, Ecuador and Venezuela. This group is characterised by distinct features including a large head, short limbs, varied scales on the flanks, a prehensile tail, a rectangular process of the ileum, and expanded toe pads extending from the base of the toes (Barbour, 1920; Dunn, 1944; Lazell, 1969; Moreno-Arias et al., 2023). This clade comprises 11 species, of which three were recently revalidated or described (Moreno-Arias et al., 2023). Previously, *Phenacosaurus* Barbour, 1920 was listed as a full genus, until Poe (1988) consolidated it within *Anolis* to maintain its monophyly. *Anolis euskalerriari* (Barros, Williams & Viloria, 1996), *Anolis nicefori* (Dunn, 1944) and *Anolis tetarii* (Barros, Williams & Viloria, 1996) constitute members of this species group that are found in the Serranía del Perijá. This region, forming the northern-most extent of the Andean mountain range, at its highest point marks the boundary between Colombia and Venezuela.

*Anolis tetarii* represents the most elusive member within the *A. heterodermus* group and is known solely through its type series, consisting of three specimens retrieved from the highest peaks of the Serranía de Perijá: Cerro Tetari or Cerro de la Teta (type locality) and Cerro Pintao or Cerro Pintado (paratype locality), situated on the Venezuelan side of the border. These specimens were collected at altitudes ranging from 2799 to 3020 m a.s.l. (Barros et al., 1996; Rivas et al., 2023). Records of this species are relatively old, with one paratype (MHNLS 664; Museo de Historia Natural La Salle, Caracas, Venezuela) having been collected in 1952, while the holotype (MBLUZ-R-215; Museo de Zoología de la Universidad de Zulia, Maracaibo, Venezuela) and the remaining paratype

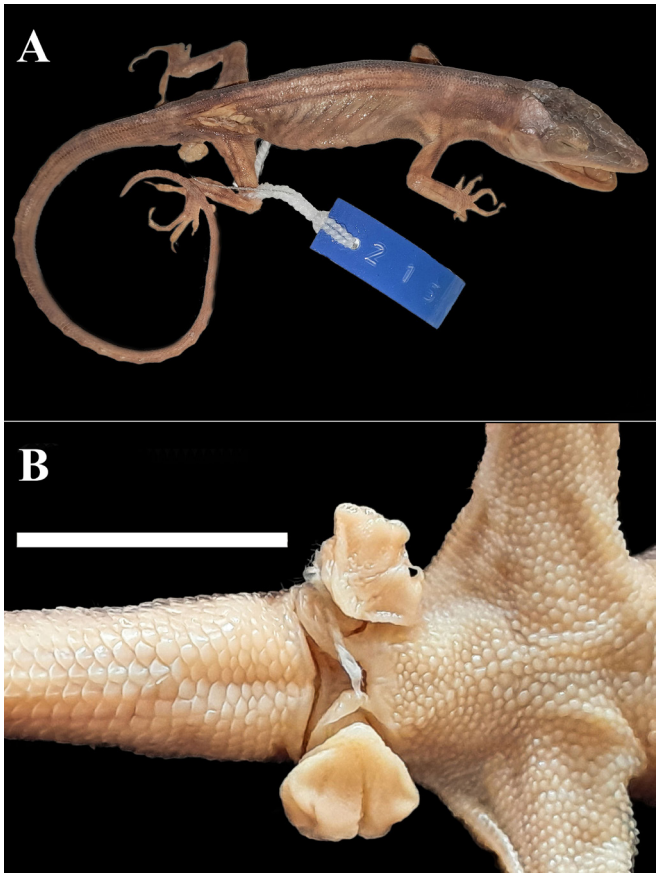


**Figure 1**. Holotype specimen of *Anolis tetarii* (MBLUZ-R-215) - **A.** Full body in dorsolateral view, and **B.** Postcloacal scales, scale bar = 10 mm

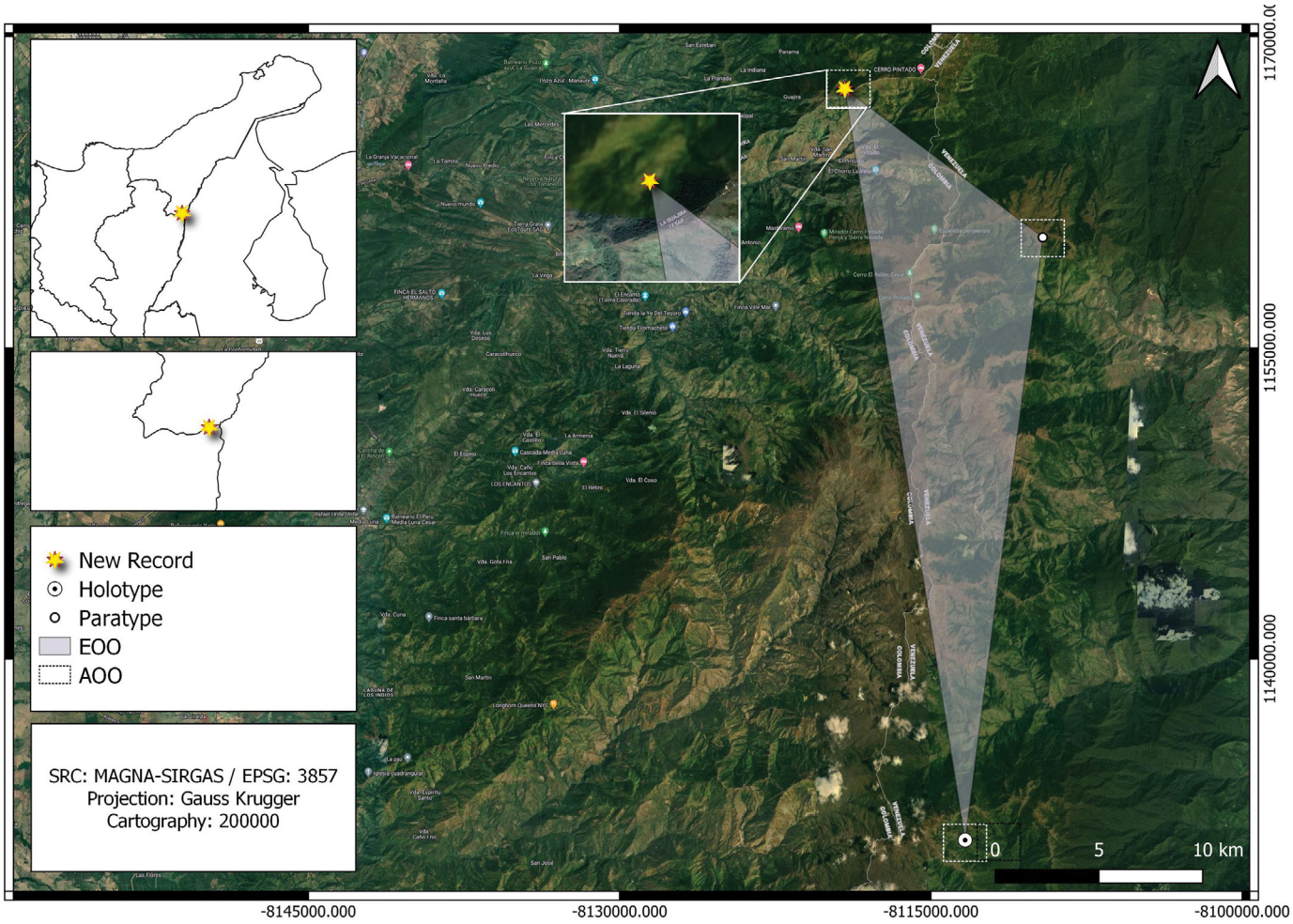


**Figure 2**. Known distribution of *Anolis tetarii*, EOO = extent of occurrence, AOO = area of occupation. The two line maps (top left) show the northern border of Colombia shared with Venezuela.

(MCZ: Herp:R-176474; Museum of Comparative Zoology, Cambridge, USA) were obtained in 1989. Despite the age of the records and the scarcity of available data, *A. tetarii* holds a classification of Least Concern on the IUCN Red List (Rivas & Schargel, 2020). However, it is important to exercise prudence in interpreting this categorisation, as many of the evaluation criteria for its conservation status remain undisclosed.

In September 2015, during an expedition to the Colombian side of the Serranía del Perijá, a single specimen from the *Phenacosaurus* clade was captured. This unidentified specimen was subsequently preserved and placed within the Reptile Collection of the Centro de Colecciones Científicas de la Universidad del Magdalena in Santa Marta, Magdalena, Colombia (CBUMAG:REP:00798). Upon closer examination, aided by the descriptions provided by Barros et al. (1996) and the morphological characterisation by Köhler (2014) concerning the genus *Anolis*, as well as the comparison with photographs of the holotype specimen (Fig. 1), we were able to definitively ascertain the identity of this lizard as the fourth recorded specimen of *A. tetarii*. This contribution serves a twofold purpose: to document, for the first time, the presence of this species within Colombian territory, and to describe certain facets of its external morphology, distribution, natural history, and conservation.

The specimen CBUMAG:REP:00798 was collected in the department (equivalent to state) of La Guajira, specifically in the municipality (analogous to county) of La Jagua del Pilar, within the village of El Espejo (coordinates: 10° 25’45.8” N, 72° 56’6.6” W; elevation: 2800 m a.s.l., as shown in Fig. 2). This location lies within the shrubland transitional zone that marks the interface between the high-Andean forest and páramo biomes. Notably, by the end of the 20th century, this area had undergone substantial degradation due to the illicit cultivation of opium poppy. Although these activities have purportedly ceased, the region still bears the brunt of degradation attributed to extensive cattle ranching. The specimen was encountered around 08:00 h while basking on a slender branch of a bush, positioned at a height of 230 cm.

This lizard was determined as *A. tetarii* by the following combination of characters: (1) the relatively large size (75 mm in snout-vent length), (2) heterogeneous dorsal squamation with (3) small granules surrounding the big and flattened scales, (4) continuous nuchal crest, (5) enlarged supralabials do not extend to the commissure of mouth, and (6) small yellow dewlap (note that this is visible even with the dewlap unextended, Fig. 3A). The count of canthals, loreals and postrostrals of the Colombian specimen (Fig. 3B, C) agree with that reported by Barros et al (1996). In the original description of the holotype it is stated that there is

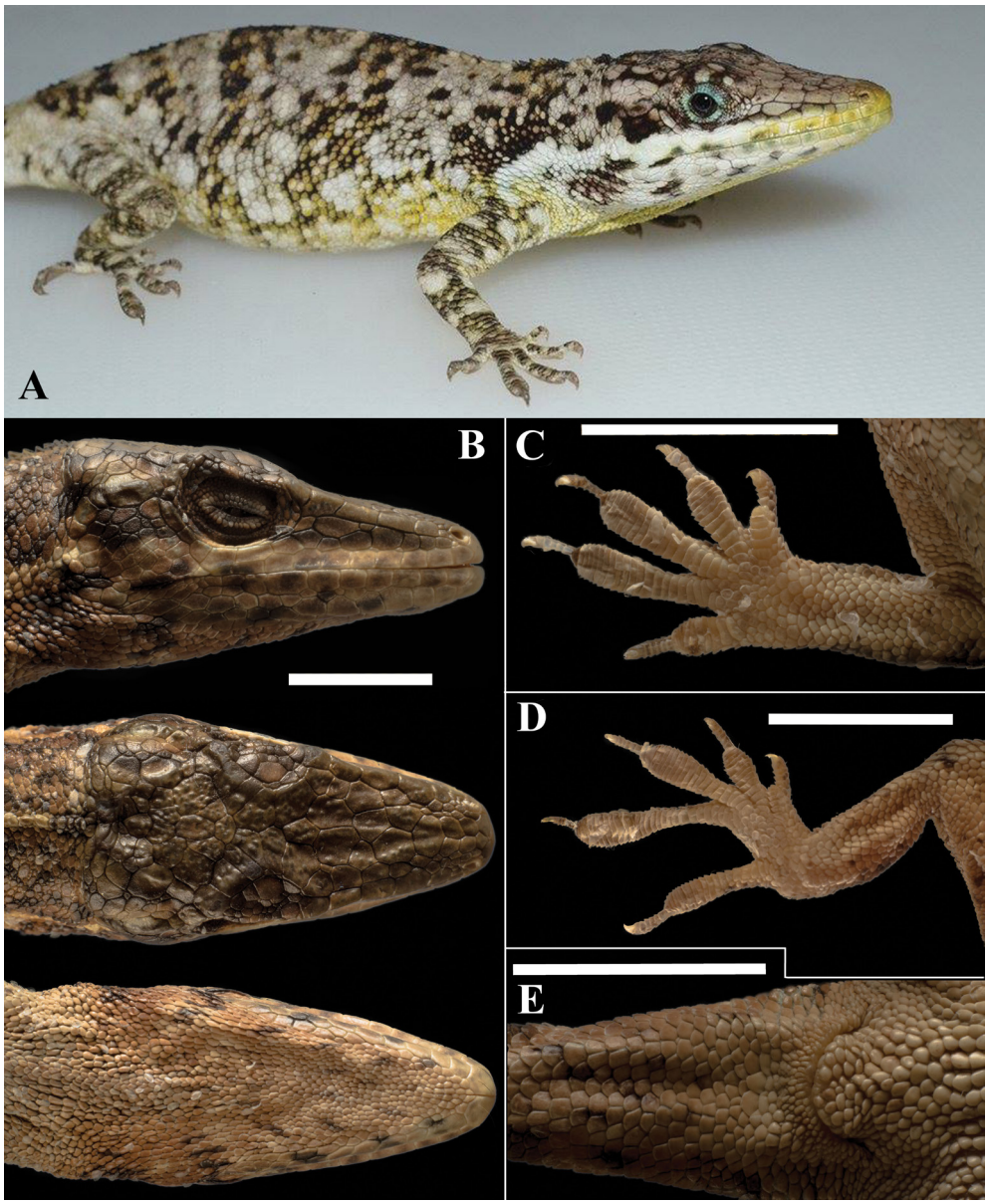


**Figure 3**. Detailed morphology of *Anolis tetarii* in life and of the specimen after preservation (CBUMAG:REP:00798)- **A.** Photograph in life, **B.** Head in lateral, dorsal, and ventral view, **C.** Right hand, **D.** Left leg, and **E.** Postcloacal scales, scale bars = 10 mm

a count of 21 lamellae under the entire fourth toe but a subsequent examination (made by Gilson Rivas, MBLUZ, correspondence dated 29 August 2023) determined that there are 35–36 lamellae under the fourth toe (phalanx I = 5, phalanx II-IV = 30–31, count for both feet), which is closer to the count of our specimen CBUMAG:REP:00798 for this character (phalanx I = 5, phalanx II–IV = 31, count for both feet; Fig. 3D). The preserved specimen was recognised as male by the direct test with a probe. Nevertheless, the significantly large postcloacal scales typical of male *Anolis* were not observed, neither in the holotype (Fig. 1B) nor in the Colombian specimen (Fig. 3E).

This lizard is easily distinguishable from the other two species of the *Anolis heterodermus* group which occur in the Serranía de Perijá, *A. euskalerriari* and *A. nicefori* (Lazell, 1969; Barros et al., 1996; Moreno-Arias et al., 2023), by the presence of dorsal crest (absent in *A. euskalerriari*), heterogeneous dorsal squamation (homogeneous in *A. euskalerriari*), small granules between flattened dorsal scales (absent in *A. nicefori*), and fewer canthals (four in *A. tetarii*, six in *A. euskalerriari*, six to eight in *A. nicefori*). In addition, *A. tetarii* can be distinguished from *Anolis richteri* (Dunn, 1944), which is the most similar species from the Cordillera Oriental (equivalent of Eastern Andes) of Colombia, by the dewlap coloration in males (white-cream to pink coloured dewlap), a greater count of supralabials at centre of eye (8–10 in *A. tetarii*, 7–8 in *A. richteri*) and the smaller size of postcloacal scale in males (enlarged in male *A. richteri*).

This discovery marks the first documented record of *A. tetarii* in Colombia and represents the fourth known specimen of this species. The locality of the Colombian specimen is at a distance of 11.6 km in straight line from the paratype locality and 35.8 km in straight line from

the holotype locality. When combined, these localities delineate an extent of occurrence (the area occupied by the convex polygon formed by the outermost points of the distribution range) spanning 146 km$^2$ and an area of occupation (the number of 2 km$^2$ plots containing at least one record of the species when the distribution map is gridded) covering 12 km$^2$. Based on the new evidence, the Least Concern classification proposed by Rivas & Schargel (2020) should be reconsidered. While they assert that the species thrives in remote, unpopulated and well-preserved regions, this dynamic changes markedly within Colombian populations. Notably, the Colombian páramos of Perijá experience a greater degree of degradation compared to their Venezuelan counterparts due to higher population densities, increased disturbance and a lack of protected areas (Rangel-Ch., 2007; Instituto Alexander von Humboldt, 2017). Furthermore, the lack of quantitative assessments concerning potential habitat loss and population status exacerbates the situation. In light of this, the known distribution does not provide sufficient information to support the current classification, prompting us to advocate for a more fitting categorisation of Data Deficient (DD) for the conservation status, as supported by the newfound evidence. Notably, this finding also contributes to a milestone for Colombian herpetology as it constitutes the 80$^{th}$ confirmed *Anolis* species within the country.

## ACKNOWLEDGMENTS

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SHORT COMMUNICATION

https://doi.org/10.33256/hb167.3436

# First report of a freshwater turtle species as a permanent resident of a cave

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Freshwater turtles are biological indicators of a healthy ecosystem, as they are important scavengers, prey and predators (Lovich et al., 2018; Santori et al., 2020). India is home to 34 of the 356 known tortoises and freshwater turtle species worldwide (Jadhav et al., 2018). The freshwater turtles in tropical regions use various artificial/natural, mainly freshwater and sometimes marine habitats (Ahmed et al., 2020; Das & Bhupathy, 2009). During our visit to a laterite cave in the northern Western Ghats of India, in Sindhudurg, Maharashtra, one of the Global Biodiversity Hotspots, we documented the first-ever resident population of a freshwater turtle inside a cave. Close observation identified the species as *Melanochelys trijuga*, the Indian black turtle or Indian pond terrapin.

The Indian black turtle is widespread across India, Bangladesh, Myanmar, Nepal and Sri Lanka (Ahmed et al., 2020). Its conservation status is considered to be of Least Concern in India but is generally considered threatened in Myanmar and Sri Lanka, and some subspecies (*coronata*, *edeniana*, *parkeri*) may be threatened (Ahmed et al., 2020). According to the IUCN's recent assessment, the population is declining (Ahmed et al., 2020) due to threats such as the pet trade, local consumption and medicinal use (Krishnakumar et al., 2009).

*Melanochelys trijuga* is primarily crepuscular or nocturnal and can be seen foraging after dark (Ahmed et al., 2020; Das & Bhupathy, 2009). Other turtle species have been reported to use subterranean habitats and may use caves as a temporary refuge to hide from predators (Acuña-Mesen, 1994; Tuberville et al., 2005); a fossil turtle has even been found in a cave in Texas (Milstead, 1956). We came across no reports of turtles as permanent inhabitants of caves, although there is a recent report of the Mediterranean stripe-necked terrapin *Mauremys leprosa* using an iron mine tunnel in Spain (Hinckley et al., 2015).

During our first exploration inside the cave on 24 October 2021, we recorded 11 *M. trijuga* roosting. In a further seven visits between November 2021 and May 2023 we made further records of the species. This laterite cave has a single entrance and runs horizontally for 100.2 m (the cave length continues further with inaccessible passages) with an average height of 6 m. The average temperature inside the cave is 30.8 °C with 100% relative humidity. The cave structure from the inside is highly uneven with several small voids, burrows and clastic sediments (fallen rocks/boulders). A perennial freshwater stream flows from inside the cave to the entrance where there is a pool of water all year round. Within the cave there is a significant population of frugivorous and insectivorous bats, so the stream water brings bat guano to the pool, which may be a food/nutrient source for the fauna and flora. As *M. trijuga* is known to feed opportunistically on various animals and plants (Das, 1991), it seem likely that the fauna (including dead animal bodies) and flora in the pool are their primary food source. The pool outside the cave entrance provides drinking water for cattle and possibly occasionally for humans. Along with

**Figure 1**. The Indian black turtle *Melanochelys trijuga* on a boulder in the laterite cave

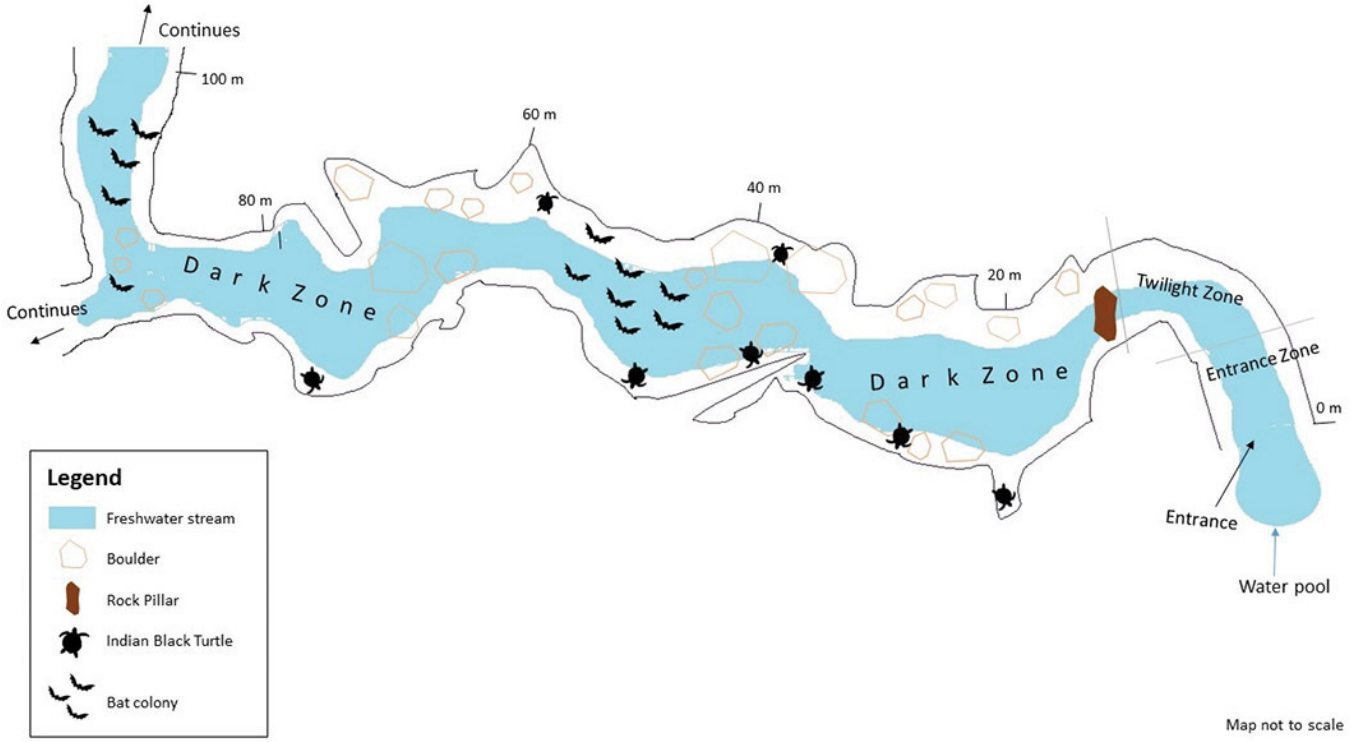


**Figure 2**. The locations of the Indian black turtle inside the laterite cave

its ecological significance, the cave has cultural importance and is worshipped and preserved by local people.

During our visits to the cave at various times of day, the turtles were mainly seen roosting in the small voids, burrows and under the boulders on the ground (Figs. 1 & 2) and at night were only in the dark zone of the cave. We did not attempt to measure them but based on what we have seen the turtles were of various body sizes. This may indicate that the species is breeding in this safe underground cave habitat. Given the stable microclimatic conditions and the availability of water and food (aquatic flora and fauna and dead animal bodies, including bats) within the cave and at the entrance (pool), it appears likely that the turtles are permanent residents of this laterite cave.

Caves as ecosystems are often overlooked due to their inaccessibility and the absence of light. Along with functioning as a unique habitat for diverse species, caves have an important role in the region's hydrology. Cave ecosystems provide unique habitat conditions for freshwater animals but remain under-explored and insufficiently studied (Culver & Pipan, 2019; Wynne et al., 2021). This note is the first to document a freshwater turtle species inhabiting a cave. Further, recognising this knowledge gap we recommend scientific studies to explore the ecology of the Indian black turtle using caves in its entire distribution range and the identification of the various threats faced in both subterranean and more usual habitats.

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# A possible case of insect-frog mimicry

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Predatory selection pressure has shaped the complex defensive strategies of amphibians and terrestrial arthropods. Warning colouration is among these strategies and is widespread among vertebrate and invertebrate species (Toledo & Haddad, 2009; Fieldler & Brehm, 2021; Kikuchi et al., 2023). Bright contrasting colours signal to potential predators the presence of toxins or other dangers, a strategy known as aposematism (Toledo & Haddad, 2009). Aposematic species are often the subject of mimicry by other species so that predators associate the mimics with the toxins or any other dangerous traits of their models. The best-known examples are Batesian mimicry (where only the model is toxic) and Müllerian mimicry (where both the model and its mimic are toxic). Frogs are a group well known for having species involved in mimicry rings where several species benefit from convergence in colouration. To date all records of frog mimics have involved other frogs (e.g. Darst & Cummings, 2006; Prates et al., 2012; Stuckert et al., 2014). However, we offer a possible example of an insect that mimics an aposematic frog.

Based on fortuitous field sampling, we found a weevil *Cratosomus* sp. (Coleoptera; Curculionidae) that could possibly be a mimic of the aposematic arrow poison frog *Ameerega trivittata* (Dendrobatidae) (Fig. 1 A&B). The species are sympatric and were recorded in the Amazon, in the city of Puerto Misahualli, province of Napo, eastern Ecuador (1° 01'51.5" S, 77° 39'48.0" W). The weevil used the same microhabitat as *A. trivittata*, and was found walking on the vegetation at 50 cm from the ground and on the soil. Also, both species are similar in size (about 4 cm in length). We do not know whether this weevil is toxic and so we cannot suggest the type of mimicry involved, i.e. Batesian or Müllerian. Such hypothetical morphological convergence would be selected for by visual predators, such as birds with colour vision that prey upon both frogs and insects. Our observation presents an opportunity to investigate this species pair more deeply and alerts us to the possibility of other insect mimics of frogs.

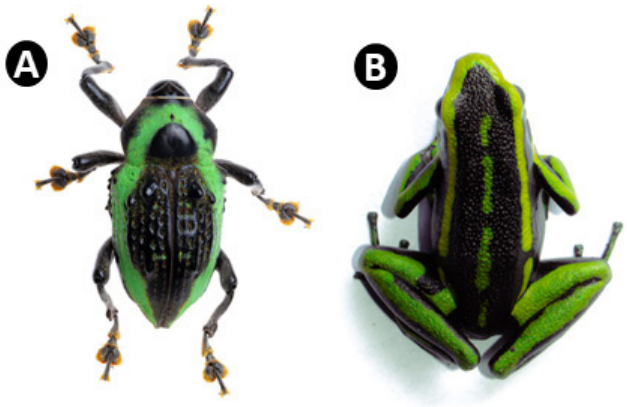


**Figure 1**. Similar colour patterning of - **A.** A weevil *Cratosomus* sp. and **B.** The arrow poison frog *Ameerega trivittata,* suggest that the weevil may be a mimic of this highly toxic frog

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NATURAL HISTORY NOTE

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# The barred grass snake *Natrix helvetica* as a potential predator of sand martin *Riparia riparia* chicks

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In Great Britain, the barred grass snake *Natrix helvetica* is widespread throughout most of England and Wales, with a limited distribution in Scotland (Cathrine, 2014). It is a semi-aquatic predator living in close proximity to water and in a site in southern England was found to feed mostly on amphibians and fish (73%), small mammals (25%) and just rarely on birds (1%) (Gregory & Isaac, 2004). A listing of ten bird species that have been observed as prey of *Natrix* spp in Britain and mainland Europe is given by Di Nicola & Zabbia (2021).

The sand martin *Riparia riparia* is the smallest of the European hirundines, these are agile flyers that feed when flying over water, mainly on insects (Holden & Gregory, 2021). They arrive in Europe during March and April where they reproduce, before returning to their African wintering grounds from August (Holden & Gregory, 2021). They can be distinguished from other hirundines by the presence of a distinctive dark chest bar, dark brown upper parts and pale under parts with a slightly forked dark tail. Sand martins breed in vertical sandy banks, often using artificial banks that have been especially created for them to nest in alongside lakes and canals. They are widespread throughout the British Isles during the breeding season (Holden & Gregory, 2021).

On 31 July 2023, at Blashford Lakes, New Forest, Hampshire (50.87510, -1.78610), an adult female grass snake was observed in a sand martin artificial nesting bank (Fig. 1). At approximately 11:15 h, a juvenile *R. riparia* fell into the water below the artificial nesting bank, which later tried to climb to a nearby concrete foundation after failing to climb the sheer face of the bank. It was at this time that the *N. helvetica* was first observed exiting one of the *R. riparia* nests, at which point the adults birds started to mob it. For almost 30 minutes, the snake was watched as it navigated the nesting bank, while the juvenile *R. riparia* was fed by its parents despite the mishap. Both species were acutely aware of one another, and even though no direct observations of feeding were recorded, the long-time that the *N. helvetica* remained within the nesting holes suggests that it was consuming chicks and/or eggs that would have been readily available.

This is the first report suggesting that *N. helvetica* may prey upon *R. riparia*. While the ingestion of avian prey is a

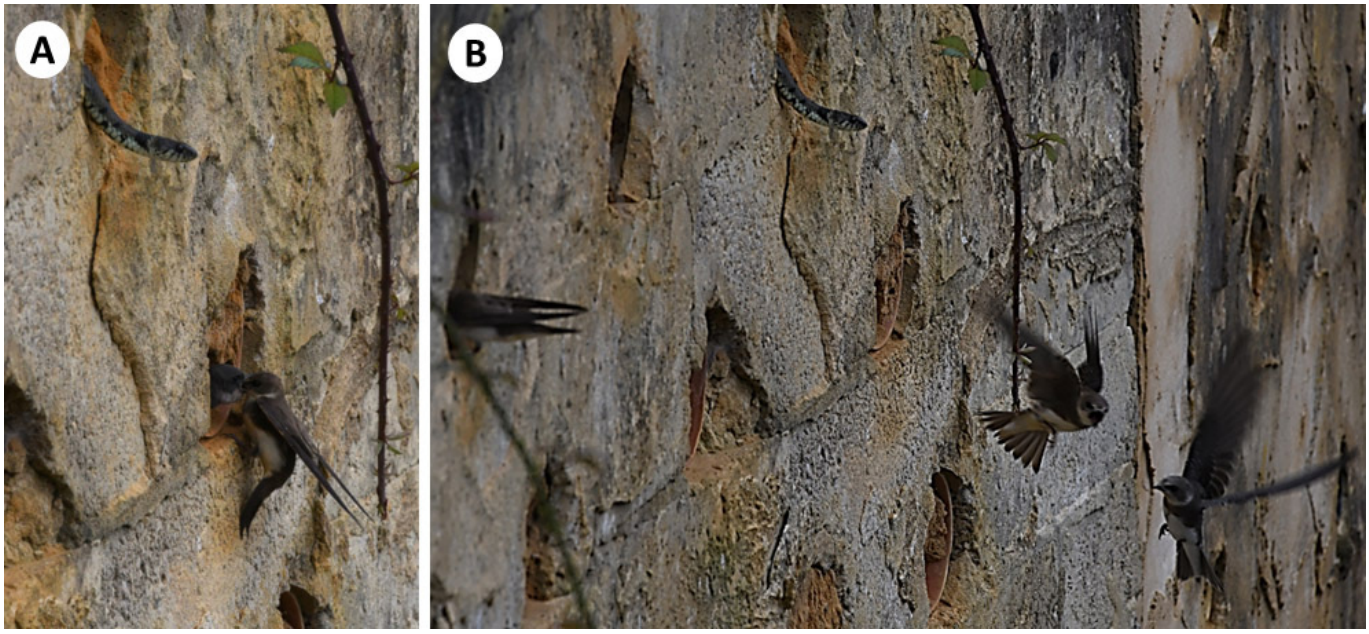


**Figure 1**. An adult female barred grass snake *Natrix helvetica* in a sand martin *Riparia riparia* artificial nesting bank- **A.** The snake emerging from one of the nesting holes, **B.** The sand martins are alerted to the snake's presence and start to mob it

seemingly relatively rare occurrence in *N. helvetica*, this observation and others like it suggest that *N. helvetica* is an opportunistic hunter that may feed on birds, when they are available. It is likely that *N. helvetica* preys on amphibians early in the year when they are plentiful, but then it may be forced to switch to other potential food sources when amphibians have dispersed away from their breeding ponds and are much harder to find. Our observation was made during the late summer when both adult and metamorph amphibians would have dispersed into the surrounding environments. Instead of expending energy trying to locate them around Blashford Lakes, the *N. helvetica* may instead have sought out more easily located prey in the artificial nesting bank.

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NATURAL HISTORY NOTE

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# Attempted predation of a chameleon *Chamaeleo gracilis* by Blanding's tree snake *Toxicodryas blandingii*

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Blanding's tree snake *Toxicodryas blandingii* is a nocturnal, rear-fanged, venomous and largely arboreal snake that has been recorded 20 m above the forest floor (e.g. Pitman, 1974). Its prey may include frogs, chameleons and other arboreal lizards, rodents and bats (e.g. Cansdale, 1961; Menzies, 1966; Wickler & Uhrig, 1969; Luiselli et al., 1998) but interestingly a quantitative study in southern Nigeria has shown that it undergoes an ontogenetic dietary change (Luiselli et al., 1998) with juveniles feeding only on lizards (Scincidae, Agamidae, Chamaeleonidae) of appropriate size, subadult stages adopting a wider diet, and adults eating mainly birds and mammals. Here we report a case of attempted predation by a juvenile Blanding's tree snake of a chameleon *Chamaeleo gracilis*. It should be noted that in this case we are using the traditional classification of the *T. blandingii* complex, although a recent article suggests that *T. blandingii* occurs only west of the confluence of the Congo and Ubangi rivers (Greenbaum et al., 2021), while our observation was made to the east.

On 24 October 2023 at 21:03 h in the Kaniyo Pabidi, Budongo forest, Uganda (0°.20018' N, 31°.71941' E) researchers observed a young Blanding's tree snake (determined by the juvenile colouration pattern) that was holding and biting a slender chameleon *C. gracilis* above the rear legs. Meanwhile, the chameleon was also fighting hard, opening its mouth wide and using its legs to turn. At an advantaged angle, the chameleon was able to detach itself from the snake's jaws and grasp the nape of the snake's neck in its own jaws for more than five minutes (Fig. 1) before releasing the snake, which then moved off deep into the bushes. The chameleon had a dark patch on its flank where it had been grasped by the snake suggesting that this may be the site of envenomation, assuming the chameleon had been envenomed. The next morning, we revisited the battleground, the snake was not visible but the chameleon was found dead with ants already feeding on its body (Fig. 2).

Our sighting confirms that lizards are food for juvenile Blanding's tree snakes. It also shows that once the snake had abandoned its prey it did not return to swallow it, despite having envenomed it (unlike terrestrial vipers that would be expected to seek out their envenomed prey). We speculate that the failure to return could have been due either to the trauma suffered by the snake or, perhaps more likely, the interruption of a behavioural sequence where venomous, rear fanged, arboreal snakes would normally hold their prey in their mouths, wait for the prey to die, and then swallow it.

**Figure 1**. A *Chamaeleo gracilis* biting a juvenile *Toxicodryas blandingii* that had attempted to prey upon it, red arrow indicates a dark patch where the snake had been holding the chameleon in its jaws

**Figure 2**. The dead *Chamaeleo gracilis* observed a few hours after the attempted predation event

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NATURAL HISTORY NOTE

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# The Asian giant toad *Phrynoidis asper*: a terrestrial bufonid observed in an arboreal habitat

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The Asian giant toad *Phrynoidis asper* is a terrestrial species usually found on river banks that prefers fast-flowing or cascading streams for breeding (Eprilurahman & Kusuma, 2011; Kusrini, 2013; Maghfiroh & Eprilurahman, 2019). It has a stocky body with short limbs, prominent webs on the hind feet, and very rough skin with rounded poison glands located behind the eye and above the ear (Haas et al., 2023). The species has previously only been reported from terrestrial habitats but here we report an observation of it in a tree.

On 13 October 2023 at 11:48 h, a juvenile *P. asper* (SVL 8 cm) was observed sitting on the branch of a weeping fig tree *Ficus benjamina* at a height of 2 m above a riverbank (Fig. 1). The observation was made at the junction of the Bedog and Konteng rivers, located in Kasihan District, Bantul Regency, Province of Daerah Istimewa Yogyakarta, Indonesia (-7.837148° S, 110.340091° E). The observation lasted for three minutes, following which we managed to photograph it at a distance of 50 cm, before it dived into the water below.

Traditionally arboreal anuran species (e.g. hylids and rhacophorids) have adhesive toe pads, which provide adhesion for climbing (Hanna & Barnes, 1991). However, *P. asper* is considered to be terrestrial and has no obvious morphological adaptations to climbing (Kusrini, 2013; Alhadi et al., 2021). The obvious advantages to terrestrial bufonids of selecting arboreal resting sites are that they may offer avoidance of predators or parasites, and better access to prey (Lindquist et al., 2007; Granda-Rodriguez et al., 2008; Petrovan et al., 2022). Recently reported similar examples of normally terrestrial bufonids climbing into trees/shrubs have been for *Rhinella margaritifera* and *Rhinella castaneotica* in Brazil (De Noronha et al., 2013) and *Bufo* in Britain (Petrovan et al., 2022). It would be of interest to make further behavioural observations on *P. asper* to establish whether the use of arboreal habitats is a common phenomenon.

**Figure 1**. Juvenile *Phrynoidis asper* (inset) sitting on a branch of a weeping fig tree *Ficus benjamina* above a riverbank (arrow indicates position of toad on branch)

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NATURAL HISTORY NOTE

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# A banded krait *Bungarus fasciatus* scavenging on a water snake

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The banded krait *Bungarus fasciatus* has a wide geographical range that extends through south and south-east Asia as far east as southern China (Ahmed et al., 2009; Knierim et al., 2019). It occurs up to 2,300 m above sea level and occupies numerous habitat types including forests, agricultural areas and domestic gardens. The species is primarily nocturnal and will feed on a wide array of prey but is well known as a frequent predator of other snakes, a cannibal and an occasional scavenger (Daniel, 2002; Kalita et al., 2021; Knierim et al., 2017). A wide range of snake species have been reported to feed by scavenging such as *Agkistrodon piscivourus* feeding on dead fish in an intertidal zone, *Bungarus caeruleus* scavenging on a road-killed conspecific, and *Malpolon monspessulanus* feeding on a putrescent rat (Lillywhite et al., 2002; Deshmukh et al., 2016; Deso & Bonnet, 2023).

On 3 August 2022 at around 19:16 h, we observed a *B. fasciatus* consuming the dead body of a water snake in Assam, India near the main street of Mayong village (26° 15'52.0" N 92° 02'05.7" E). The krait had nearly finished consuming the dead snake by the time we arrived at the location, since only the tail of the dead water snake was visible to us, which the krait took just three minutes to ingest fully before moving off. The presence of maggots on the tail of the dead snake indicated clearly that the specimen was decomposing. Based on the dorsal colouration of the tail we think that the ingested snake was a checkered keelback *Fowlea piscator*. It seems that the ingested snake had been trapped as the tail was wrapped around some fishing net that the krait consumed along with the tail. Observations such as this one help us to understand that scavenging behaviour might be more common in snakes than previously thought.

**Figure 1**. A banded krait *Bungarus fasciatus* scavenging on a water snake, believed to be *Fowlea piscator*, in Assam, India. Note that the dead snake is wrapped around a fishing net which the krait consumed along with the maggots that were on with the tail.

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NATURAL HISTORY NOTE

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# Observations of the smooth newt *Lissotriton vulgaris* and the great crested newt *Triturus cristatus* climbing drift fences in Britain

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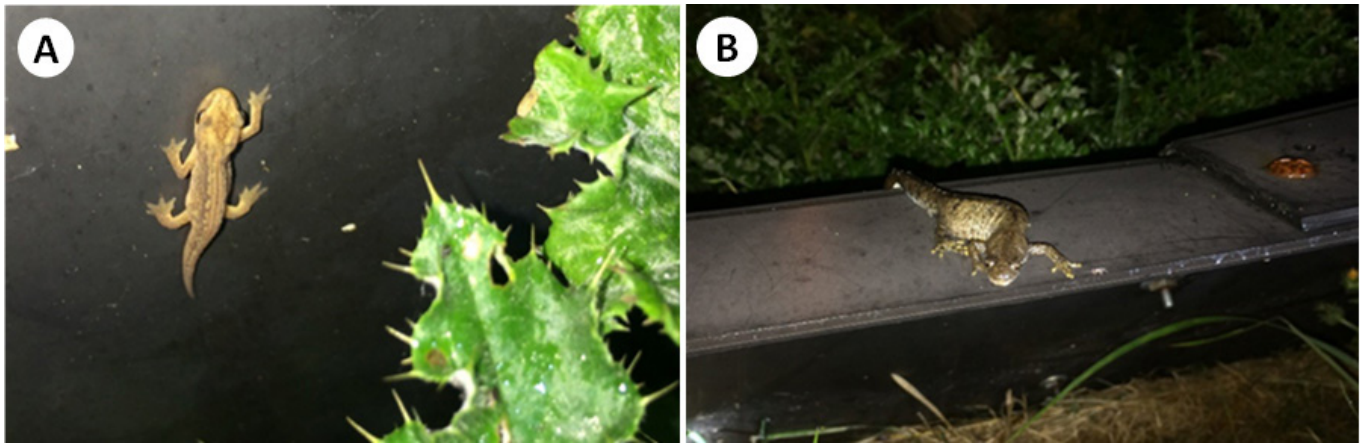


**Figure 1**. Newts climbing drift fences- **A.** Juvenile smooth newt *Lissotriton vulgaris* scaling a plastic drift fence, **B.** A male great crested newt *Triturus cristatus* climbing a concrete drift fence

In Denmark and Germany, the smooth newt *Lissotriton vulgaris* and great crested newt *Triturus cristatus* have been observed scaling vegetation and man-made structures in the form of pitfall traps (Bringsøe, 2013) and both species have been reported scaling vegetation in south-east England (Lynn & Allain, 2022). The feet of both species can adhere quite well to vertical surfaces by suction and surface tension but are not especially adapted for climbing, unlike those of truly arboreal amphibian species with adhesive toe pads (Stebbins & Cohen, 2021). This behaviour of *L. vulgaris* and *T. cristatus* generally goes unnoticed, as most of their activity is at ground level and so does not involve climbing. Herein, I report the first record of the smooth newt *L. vulgaris* and the great crested newt *T. cristatus* climbing drift fences in Britain.

On 29 September 2014, a routine check was made of drift fences deployed to divert amphibians away from roads along Nature's Way, Peterborough on the border of Hampton Nature Reserve. The fences were vertical and between 30–60 cm high. At 23:44 h, two *L. vulgaris* (Fig. 1A) were observed climbing over a plastic drift fence (52° 32'24.0" N, 0° 16'51.6" W). Additional observations were made on 6 October 2014 along the road. The first was at 22:17 h, where a male *T. cristatus* (Fig. 1B) was observed climbing over a concrete drift fence (52° 32'17.9" N, 0° 16'54.8" W), and the second at 22:29 h was of a *L. vulgaris* climbing the plastic drift fence (52° 32'24.0" N, 0° 16'51.6" W). In all instances, the observations were made at night, during favourable weather and temperature conditions for amphibians, and all newts were successful in climbing over the drift fences.

From these observations, it is clear that both *L. vulgaris* and *T. cristatus* can bypass drift fences. This has the potential to reduce both the recording rate in amphibian surveys and the efficiency of arrangements to avoid road kill. Further research to establish the proportion of individuals that climb drift fences would clarify the significance of this behaviour and alert practitioners to any need there might be for taking action to prevent it.

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BOOK REVIEW

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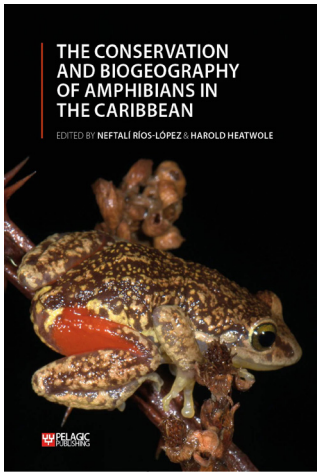
# The Conservation and Biogeography of Amphibians in the Caribbean

**Neftalí Ríos-López & Harold Heatwole (Eds.)**
**Pelagic Publishing, 2023**
**ISBN 978-1-78427-267-8 (Hbk), 604 pp.**




The 'Amphibian Biology' series was initiated in 1994, with a mission to publish volumes that provide detailed, contemporary accounts of work in different areas of the subject. The first eight volumes therefore covered topics such as social behaviour, palaeontology, endocrinology and disease. From volume 9 onwards the series has focused more on conservation aspects and regional accounts. The series has had its ups and downs and has had more than one publisher, but with Harold Heatwole leading the editing of every volume, a high standard of quality and consistency in approach has been maintained throughout. Volume 9 of the series - of which this book is part 5 - focuses on the "Status of Decline of Amphibians: Western Hemisphere". In producing this Caribbean volume the two editors have assembled some 29 contributors - drawn from across the Caribbean, North and South America and Europe - to assist them with the task.

After a dedication of the book to Stephen Blair Hedges, whose research on Caribbean amphibians is nudging some 300 publications, Professor Heatwole outlines the scope of the volume in a preface, summarising ongoing efforts to update the IUCN Red List status of the species in the region. He points out that the book goes a little beyond the Caribbean sensu stricto, as it includes the Bahamas and Turks and Caicos Islands, as well as some small islands close to the coasts of South and Central America.

The first chapter of the book is co-authored by the editors and provides an overview of insular biogeography and ecology. Building on the classical MacArthur and Wilson theory of island biography, this chapter takes the reader on a scientific and historical journey through the theories that have been developed since. Although neither explicitly about the Caribbean or amphibians, this opening chapter provides a comprehensive and up-to-date account of the ideas that have framed the work of subsequent chapters. The acknowledgements provide an intriguing personal perspective from Heatwole about his relationships with Robert MacArthur, E.O. Wilson and Richard Levins at the ground-breaking time of the 1960s and 1970s.

Chapters 2–13 provide accounts of the status and distribution of amphibians in different regions of the Caribbean. The larger islands, such as Cuba, Jamaica, and Trinidad and Tobago get dedicated chapters, while others, such as those comprising the Lesser Antilles are dealt with collectively in individual chapters. As the book runs to over 600 pages, it is difficult to provide a detailed review of each individual chapter, so some highlights will be presented here.

In Cuba, the amphibians have a higher level of endemism (95%) than reptiles, mammals and birds, with the number of frog species exceeded only on the island of Hispaniola. Indeed, Hispaniola is dealt with in two separate chapters focusing on the two countries it comprises – Haiti to the west and the Dominican Republic to the east. *Eleutherodactylus* has radiated into many endemic species across the Caribbean islands, with 17 species within the genus endemic to Haiti alone. In contrast, the much smaller islands that form the arc of the Lesser Antilles are relatively depauperate in terms of amphibians. Here just 47 out of several hundred islands contain amphibians, with the fauna comprising a mere ten native and six non-native species. The difficulty of colonising such small, often inhospitable habitats across saltwater is flagged as a driver of this pattern. *Eleutherodactylus johnstonei* plays an intriguing role in this story. In Chapter 9, Robert Powell and Robert Henderson describe this species as "the anuran version of a weed". Interestingly, the type specimen of *E. johnstonei* is from Grenada, yet it was introduced there from Barbados, which itself has a non-native population dating from about 1885. One of the reasons this species is so widespread across the region is its ability to capitalise on human initiatives to increase water supplies and irrigate what is an otherwise dry landscape. Cisterns, ornamental pools and ponds on golf courses can all support buoyant populations of this species. I can attest to this personally, as having stayed on an island where the only water supply was a rain-filled water tank, each bucket needed to be checked carefully for frogs before transferring the contents to the kettle or shower!

All of the chapters provide historical accounts of progress in the discovery of the species found on the island (or group of islands), along with taxonomic information and debates, distribution, climate, threats and conservation interventions. Several chapters discuss the role that captive breeding can play in the conservation of the species of the region. Indeed, some captive programmes, such as that for the Puerto Rican crested toad *Peltophryne lemur* have

been at the forefront of the field in developing in vitro fertilisation, cryopreservation of sperm and eggs, and wider assisted reproductive technologies.

A common theme running through the book is the complexity of threats facing amphibians of the region. Although the threats are well-known and certainly not confined to the Caribbean (i.e. habitat loss and fragmentation, introduced species, overexploitation, pollution, climate change and disease), there are clearly pockets of research that deserve further exploration. For example, an appendix to Chapter 8 that deals with Puerto Rico and the Virgin Isles provides a critical review of the evidence that the mongoose has had on amphibians. Intriguingly, the conclusion is that this non-native predator may not deserve the reputation that it has historically received. Earlier on in the same chapter, the authors highlight the importance of scrutinising "old literature" to provide context for the current distribution and status. In fact, what is clear from the book is that the whole of the Caribbean has a long history of herpetological research. Given the biological and geographical diversity of the region, the Caribbean has proved irresistible as a natural laboratory for exploring evolutionary patterns and processes. This book is as much about how that work has progressed over many decades as it as about describing present-day status and distribution.

Frogs of the genus *Eleutherodactylus* are not the only taxa to have been widely transported around the islands by human agency. As in other parts of the world, cane toads *Rhinella marina* have been introduced for pest control, and American bullfrogs *Lithobates catesbeianus* for commercial and aquaculture purposes: both species are considered by IUCN to be among the world's top 100 most invasive species. As its scientific name implies, the cane toad is tolerant of saltwater and able to swim short distances between islands. One of the more unusual arrivals was a fire salamander *Salamandra salamandra* that arrived in a plastic box of salad shipped from France to St Barthélemy in 2012. It died a couple of days after its discovery.

Pelagic Publishing is a growing publisher with a strong portfolio of quality herpetological books. It is therefore reassuring to see the 'Amphibian Biology' series firmly homed within this stable. 'The Conservation and Biogeography of Amphibians in the Caribbean' is a bulky volume, and although £120 may seem quite expensive, this is now standard for a scholarly book of this quality and size. Indeed, the standard of production is high. All of the photographs, maps and figures are reproduced in colour and the tables are clearly laid out. Although I reviewed the hardback copy, the book is also available as a PDF and e-book, which might suit researchers who have limits on luggage yet need this book for fieldwork. The book is comprehensively referenced throughout and includes a glossary and separate subject and taxonomic indices. This is likely to remain the standard reference volume for those undertaking research on amphibians in the Caribbean for some time to come, but it will also be of interest to those wishing to explore the rich herpetological history of the region.

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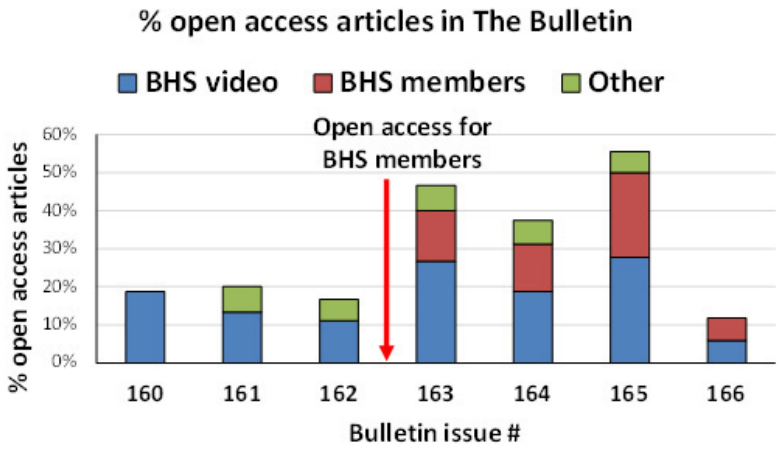
# HERPETOLOGICAL BULLETIN REPORT 2023

In 2023, issues 163–166 of The Bulletin were uploaded to the BHS website on the first day of each quarter. There were 110 manuscripts submitted in 2023, almost the same number as in 2022 (see Table). The overall acceptance rate for articles rose to 60% and all four issues in 2023 have been a full 48 pages. The time from acceptance to the publication of a manuscript is typically 4 to 5 months. Of the published articles, 11% have been illustrated with an accompanying video posted on the BHS YouTube site and consequently had full public access from the time of release.

**Table 1**. Submission and acceptance rates for manuscripts received in 2023 (2022)

| | **No. submitted** | **No. accepted** | **% accepted** |
|---|---|---|---|
| Full Papers | 15 (17) | 11 (10) | 73% (59%) |
| Short Notes | 13 (19) | 6 (8) | 46% (42%) |
| Short Communications & Natural History Notes | 82 (81) | 49 (43) | 60% (53%) |
| **Totals** | **110 (117)** | **66 (61)** | **60% (52%)** |

A major change implemented from Issue 163 onwards is that articles written by BHS members are open access from the time of release. Previously, articles would be open access only if they were connected to a BHS video, were book reviews, or were reports promoting BHS interests. It was anticipated that this change would result in the proportion of open access articles in any one issue of The Bulletin rising to about 50%. The figure below shows that this is what has happened with the exception of the most recent issue (166) which had an unusually low number with a link to a BHS video or written by BHS members.

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Rick Hodges
Scientific Editor

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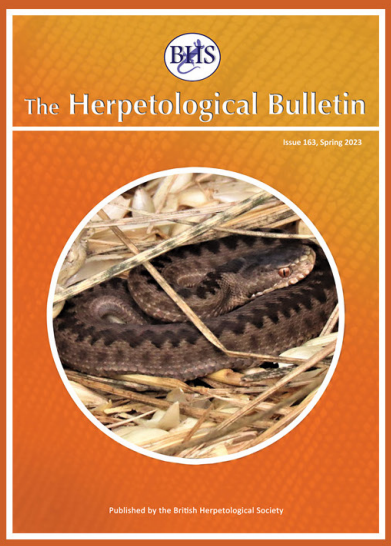
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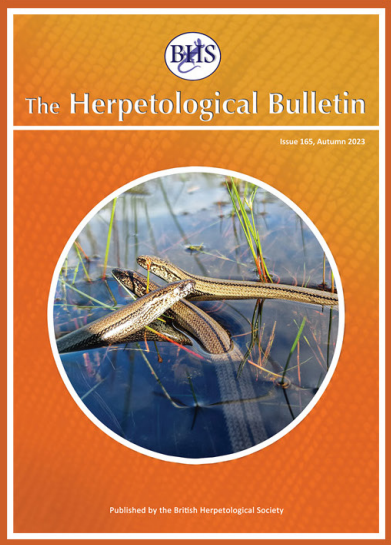
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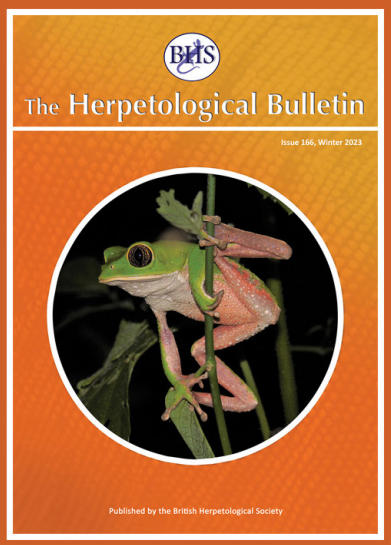
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