



## Insights on the sleep ecology of *Tropidurus hispidus* (Spix, 1825) (Squamata: Tropiduridae)

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We report preliminary data on the sleep ecology of *Tropidurus hispidus* (Tropiduridae) lizards based on six observations of sleeping lizards in natural settings, and highlight how standard monitoring techniques can be useful to study lizard sleep behaviour. We studied the sleeping habits and substrate use of three adult male lizards, monitored using thread bobbins, along with three adult lizards we found during active search (total  $n = 6$ ). Monitored and non-monitored lizards were observed sleeping during the night with varied postures at variable heights. Regarding sleeping sites or substrates, we observed for the first time burrowing behaviour in sandy ground under thorny vegetation. Besides the use of the ground, four lizards used woody branches of shrubs for sleeping. We did not observe intraspecific aggregations during the sleep period. These preliminary results indicate that *T. hispidus* may rely on hiding and thermoregulating during sleep hours, which differs from other lizard lineages that are commonly found sleeping perched in thin vegetation or on leaves. We also highlight that the use of standard monitoring techniques, such as thread bobbins to study lizard sleep behaviour, enhances our understanding of the natural history of lizards and other reptiles.

**Keywords:** behaviour, Caatinga, lizard, sleeping, tracking

While the interplay of lizards' behaviour and resource use has been extensively studied under the perspective of foraging and reproduction (Vitt & Pianka, 1994; Pianka & Vitt, 2003), the potential effect of sleep on the ecological niche remains largely underexplored. Sleep is a behavioural state involving a trade-off between physiological benefits and increased vulnerability to predators (Lima et al., 2005; Lakhiani et al., 2023), that has been systematically underexplored in herpetological studies (Mohanty et al., 2022). Key gaps in lizard sleep ecology, such as sleeping site preferences, body posture and intraspecific interactions, include limited taxonomic

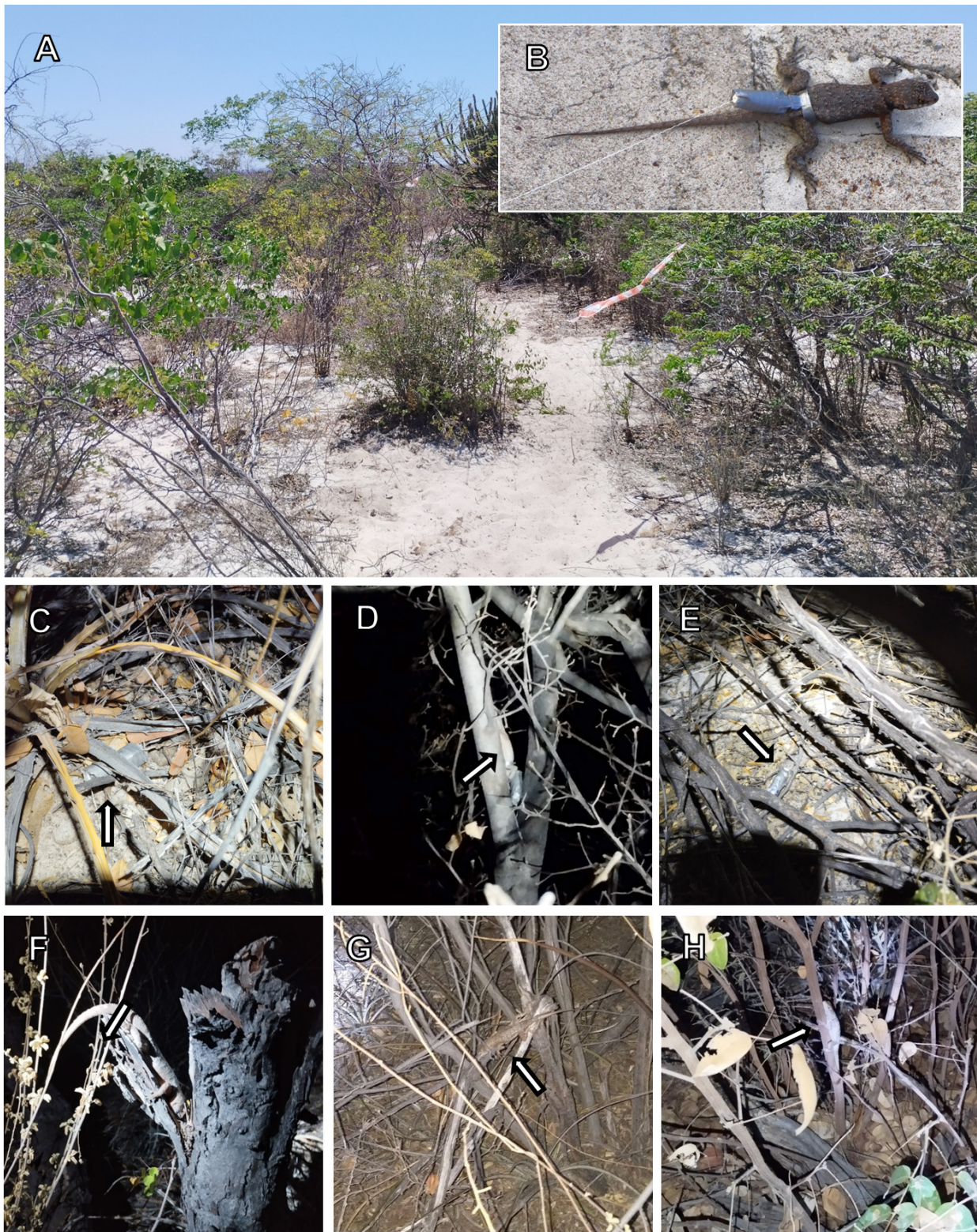
and geographic coverage and a lack of appropriate methods for observing sleep in the wild. For the South American lizard family Tropiduridae, for example, data are restricted to a few forest-dwelling species (Rand & Rand, 1966; Stebbins et al., 1967; Avila-Pires, 1995; Mohanty et al., 2022).

Observing animals sleeping in the wild is particularly challenging due to the frequent use of concealed sites and their tendency to flee when approached, underscoring the need for methods that enable observation from a safe distance (Rattenborg et al., 2017). Tracking techniques often implemented in movement ecology but rarely used in sleep studies could offer valuable tools for addressing some of these challenges (Carvalho et al., 2014), for example the thread bobbins that allows to track fine-tune movement and locate the individual with precision (Waddell et al., 2016).

Here, we used thread bobbins complemented by active search to describe the sleeping habits of the lizard *Tropidurus hispidus*. *Tropidurus hispidus* is a small to medium-sized lizard that inhabits both open and forested landscapes (Vanzolini et al., 1980; Albuquerque et al., 2018). The species has restricted diurnal activity, sleeping throughout the night (Vanzolini et al., 1980; Vitt et al., 1996). We studied *T. hispidus* from a population located at the community of Jurema, municipality of Casa Nova, Bahia, Brazil (9.34 S; 41.19 W; Datum: WGS84) during the dry season (October 2023). The area is classified as BSh (hot semi-arid climate) according to Köppen's climate classification (Ab'Saber, 1974). The soil of the study site is majorly sandy dunes alongside the São Francisco River and the vegetation is composed of xeromorphic plant species, like bromeliads, thorny scrubs of medium height (ca. 2m) (Fig. 1A).

Three adult males *T. hispidus* were sampled using pitfall traps with drift-fences (Cechin & Martins, 2000) on 24 and 29 October 2023. Attached to the pitfalls we placed thermo-hygrometer data loggers (HOBO U23-004 Pro) to monitor air temperature and humidity

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**Figure 1.** *Tropicidurus hispidus*, study area and sleeping postures. **A.** Phytophysiognomy of the study area (Caatinga biome); **B.** adult male individual of *T. hispidus* with a thread bobbin system attached for monitoring; **C.** lizard burrowed under sand covered by thorny vegetation; **D.** lizard perched on tree branch; **E.** lizard burrowed under the sand covered by thorny vegetation; **F.** lizard perched on dead trunk; **G.** and **H.** lizard perched on bush branch.

throughout the day. After capture, we attached a thread bobbin over the pelvic region (Fig. 1B) of lizards that were then released at the capture site for monitoring. Each thread bobbin originally held 250 metres of nylon thread and weighed ca. 4.8 to 5 g (HILTEX Ind. e Com.

de Fios LTDA), ensuring a weight proportion of less than 10% of body weight (following Pareja-Mejía et al. (2023) and Waddell et al. (2016) with adaptations to *T. hispidus* morphology). Each device comprised four components: a thread-line bobbin, a plastic film wrap, a silver tape

**Table 1.** Description of the sleeping behavior of six *Tropidurus hispidus* individuals observed in the Caatinga biome, in the municipality of Casa Nova, Bahia. Temperature and relative humidity were recorded using data loggers (HOBO U23-004 Pro) placed within the same area where the lizards were observed. All environmental measurements were taken within a maximum interval of 10 minutes before or after each sleeping observation.

Observation	Day	Time of observation (GMT-3)	Time of reading (GMT-3)	Temperature (°C)	Relative humidity (%)	Source	Sleeping site	Position height (cm)	Posture
Figure 1C	24 October 2023	18:23 h	18:29 h	29.6	39.1	Monitored	Burrowed in sand	0	Prone
Figure 1D	29 October 2023	18:30 h	18:29 h	31.4	31.6	Monitored	Live vegetation	240	Prone with head up
Figure 1E	29 October 2023	18:50 h	18:49 h	30.8	33.4	Monitored	Burrowed in sand	0	Prone
Figure 1F	23 October 2023	19:11 h	19:09 h	28.3	48.2	Not monitored	Dead vegetation	40–50	Prone with head down
Figure 1G	23 October 2023	19:26 h	19:29 h	27.6	48.3	Not monitored	Live vegetation	40–50	Prone with head down
Figure 1H	23 October 2023	19:12 h	19:09 h	28.3	48.2	Not monitored	Live vegetation	40–50	Prone with head up

covering and a silver tape belt securing it to the lizard's pelvis. The belt fit snugly to prevent injury or detachment (Fig. 1B). The device was considered suitable for lizards when individuals were unable to remove it, exhibited normal locomotor behaviour (e.g. walking and climbing) and showed no visible signs of injury shortly after attachment.

All three lizards were released at 08:30 h and actively monitored until 18:30 h (GMT-3) – sunset started at 18:00 h – or until the lizard remained completely immobile after sunset. During daylight hours, the thread was followed at two-hour intervals to verify its integrity, monitor for any potential entanglement or injury and record the lizard's location and overall movement patterns. Based on previous knowledge about the daily activity pattern of the species, all animals found resting at night with relaxed body posture that did not immediately escape once approached (a proxy for increased arousal threshold) were considered to be at the sleeping site and sleeping posture (Ayala-Guerrero & Mexicano, 2008; Mohanty et al., 2016). Additionally, we conducted night field searches for observations of sleeping lizards from 12 to 29 October 2023, after 18:30 h. At the end, our sampling effort consisted of 30 hours of active monitoring, in addition to 17 hours of active night search. The combined use of active searches and thread-bobbin tracking allows a more accurate characterisation of microhabitat use. Thread bobbin tracking is a well-validated method in movement ecology and is particularly effective for monitoring fine-scale movements (Vieira et al., 2005; Waddell et al., 2016; Silva et al., 2020). Compared with active searches alone, this method offers clear advantages because it enables the detection of microhabitat use beyond human visual capacity, such as underground retreats or sites located above eye level (Dodd, 2016). In addition, its low cost and adaptability to different model organisms make thread-bobbin tracking a practical and easily implemented technique for field studies.

We successfully monitored three individuals of *T. hispidus* using thread bobbins during the daylight hours and recorded their sleeping sites. In addition, we observed three adult *T. hispidus* sleeping at night (sex unidentified). One individual was monitored on 24 October 2023, and the other two on 29 October 2023. We only recorded one individual per sleeping site and no evidence of inter or intra-specific aggregation was observed. The lizards used different types of sleeping sites and expressed variable position and posture (Fig. 1C-H). We observed two lizards sleeping on the ground with their bodies semi-concealed with sand under thorny bromeliad leaves and four on vegetation perched at different heights, from approximately 40 cm to 240 cm above the ground (Table 1). The vegetation used to sleep above ground was on one occasion dead (Fig. 1F), the other occasions the plant was alive but without leaves and did not present thorns. The perches used to sleep were never thin and flexible. All individuals were in a prone position, with the body completely in contact with the substrate and with apparent use of the claws to grip the vegetation wood. Two presented the head oriented upward (Fig. 1D & H) whereas the other two were oriented downward (Fig. 1F & G).

Our findings suggest that thread bobbins can be a useful tool to collect data on microhabitat use associated with sleeping in *Tropidurus* lizards. Other methods that allow minimal interaction with animals at fine-tuned geographical scales, e.g. GPS collaring, are effective for larger mammals but impractical for small reptiles (Dodd, 2016). Small tags are an alternative and a widely implemented possibility but they are much more expensive compared to thread bobbins (Van Winkel & Weihong, 2014; Pašukonis et al., 2017). In sleep studies, the thread line enables the investigation of pre-sleep behaviour, such as sleep site selection and facilitates spotting a sleeping animal at a secure distance without disturbing the animal or initiating escape response.

This contrasts with occasional encounters with sleeping animals that depend on the experience (and chance) of the observer not awakening the animal before seeing it. The only requirement in the use of thread bobbin is to adapt the attachment system according to the organism studied (Waddell et al., 2016; Pareja-Mejía et al., 2023).

Sleeping behaviour was not consistent across individuals, but it generally differed from behavioural strategies reported for other lizards. For example, *Anolis* and *Coryphophylax* lizards are often found sleeping on thin perches, and frequently point their head to the branch node (Singhal & Johnson, 2007; Mohanty et al., 2016). These are hypothesised as anti-predator strategy that facilitate state reversal and escape response. Sleeping with the head oriented to branch node is hypothesised to facilitate the detection if a climbing predator should (e.g. snakes) approach, whereas sleeping perched on leaf and branch tips should facilitate waking through substrate vibration (Ikeuchi et al., 2012).

Although *T. hispidus* may not rely on similar early detection strategies, evidenced using wide substrates, our observations along with previous observation of *Tropidurus torquatus* (WIED-NEUWIED, 1820) in Belém, Brazil, suggest that *Tropidurus* may cope with predation pressure in different ways, including hiding and crypsis. A similar sleeping behaviour was observed for the diurnal lizard *Microlophus albemarlensis* (BAUR, 1890) in the Galapagos Island, which buries in the loose ground during the night hours (Stebbins et al., 1967). In addition to burrowing, the *T. hispidus* behaviour of burrowing under thorny vegetation adds another level of defense against predation.

Nonetheless, another explanation for the burrowing behaviour could be attributed to thermoregulatory strategies. Because during the night the air temperature decreases (Fig. S1 in supplementary materials), lizards could benefit from higher thermal inertia in the sand, maintaining higher body temperatures for longer periods of time. On the other hand, lower air temperatures provide metabolic decrease, which is beneficial for energetic savings in ectotherms. Hence, lizards could benefit from sleeping on vegetation and exposed to air. More studies are necessary to clarify the contrasting benefits and costs of different microhabitats during sleeping.

In addition to the ecological relevance of the microhabitat used, the nocturnal burrowing behaviour observed in *T. hispidus* represents a novel finding, as it has not been previously reported for this species. In contrast to daylight hours, when individuals are relatively easy to detect, the use of vegetation and subterranean sites as sleeping locations contributes to the discrepancy in sampling efficiency during the night hours.

Our findings demonstrate that conventional tracking techniques could be effectively applied to investigate the sleep ecology of lizards, and potentially other small vertebrates. We acknowledge that our findings are preliminary and restricted to the population studied and temporal range (dry season). We advocate for future research to formulate specific questions that evaluate how

seasonality, vegetation structure and other microhabitat properties affect sleep behaviour of *T. hispidus*. Ecology in reptiles varies dramatically across seasons due to temperature fluctuations affecting metabolic rates, photoperiod changes influencing circadian rhythms, prey availability affecting energy budgets, reproductive cycles altering behavioural priorities and seasonal predator activity patterns (Pianka & Vitt, 2003). Future studies could also clarify how climate change affects sleep behaviour. For instance, comparative studies between urban and natural environments could shed light on the effects of urbanisation on sleep architecture and selection of sleeping sites. Finally, the interplay of sleep and the environment could represent an underestimated aspect of lizard life and ecology.

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### Author contributions

Gabriel Spanghero and Thaís Barreto Guedes conceived and designed the study; Gabriel Spanghero and Josivan Bernardo da Silva conducted fieldwork and collected the data; Gabriel Spanghero performed the formal analyses and prepared the figures; Guarino Rinaldi Colli, Daniel Oliveira Mesquita and Thaís Barreto Guedes secured funding and provided resources; Thaís Barreto Guedes supervised the project; Gabriel Spanghero and Thaís Barreto Guedes wrote the first draft of the manuscript; all authors contributed to revisions and approved the final version.

### Data accessibility

All data is provided in the main text; however, any additional information can be provided upon request to the corresponding author.

### Ethical statement

All procedures involving animals were conducted in accordance with the ethical standards approved by the Comissão de Ética no Uso de Animais (CEUA; 6350-

1/2023) of the host institution and under authorisation no. 86246-1 issued by the Instituto Chico Mendes de Conservação da Biodiversidade (ICMBio/SISBIO), linked to the Brazilian Ministry of the Environment (Ministério do Meio Ambiente, MMA).

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