



# Ecology of *Antricola* ticks in a bat cave in north-eastern Brazil

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Received: 25 July 2020 / Revised: 6 September 2020 / Accepted: 8 September 2020  
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## Abstract

Argasid ticks are a diverse group of acarines that parasitize numerous vertebrate hosts. Along with birds, bats serve as hosts for several argasid ticks, which are commonly found in bat caves. Argasid ticks have regained attention from tick taxonomists in recent decades, with a number of new species described in various zoogeographical regions. Nonetheless, studies on their ecology are still scarce. We conducted a 1-year longitudinal study to assess the presence of argasid ticks in a bat cave in the drylands of north-eastern Brazil and evaluate their possible response to abiotic factors. From July 2014 to June 2015, 490 ticks were collected (272 nymphs, 169 males and 49 females) in a cave chamber hosting a large colony of *Pteronotus* spp. bats, being relatively more frequent from July to December 2014. Adults were identified as *Antricola guglielmonei*, whereas nymphs were assigned to the genus *Antricola*. Almost all ticks (98%) were collected on the cave walls. Only 2% were on the ceiling and, surprisingly, no specimens were found on the floor and/or guano. Adults were usually clustered in the crevices and little mobile, whereas nymphs were dispersed and more active, moving over the walls or ceiling of the cave. Although present in most of the studied period, there was a significantly negative correlation between tick abundance and relative humidity, and *A. guglielmonei* was more frequent during the dry season. Moreover, there was no evident correlation between the abundance of ticks and bats. Further long-term studies will be able to verify whether this pattern is repeated over time, and even whether other variables can influence the population dynamics of *A. guglielmonei*.

**Keywords** *Antricola* · Argasidae · Bat · Cave · Ecology · Ticks

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**Electronic supplementary material** The online version of this article (<https://doi.org/10.1007/s10493-020-00544-9>) contains supplementary material, which is available to authorized users.

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## Introduction

Argasid ticks (family Argasidae) are the second largest group of ticks within the order Ixodida. To date, approximately 218 species of argasid ticks are considered as valid (Dantas-Torres 2018; Dantas-Torres et al. 2019; Sun et al. 2019; Muñoz-Leal et al. 2020). The generic classification of argasid ticks is still subject of debate (Dantas-Torres 2018; Mans et al. 2019), but the most widely used classification includes the genera *Antricola*, *Argas*, *Nothoaspis*, *Ornithodoros* and *Otobius*. *Ornithodoros* (ca. 133 spp.) and *Argas* (ca. 62 spp.) are by far the most speciose genera of argasid ticks (Dantas-Torres 2018; Mans et al. 2019; Sun et al. 2019; Muñoz-Leal et al. 2020). The third most diverse is the genus *Antricola*, which currently includes 17 species (Estrada-Peña et al. 2004).

Incidentally, *Antricola* spp. are very unusual ticks, whose larvae feed primarily on cave-dwelling bats (Estrada-Peña et al. 2004; Labruna et al. 2012). Information on the life cycle of most *Antricola* spp. remains virtually unknown, but it is acknowledged that adults do not feed on blood and it has been postulated that they derive their nutrition from bat guano (Ribeiro et al. 2012). Early nymphal instars of *Antricola delacruzi* feed on blood (Estrada-Peña et al. 2008), but information for other species is limited. Another interesting aspect of some *Antricola* ticks is the maternal care. For instance, the female of *Antricola marginatus* broods her clutch of eggs and carries the larvae on her dorsum, as demonstrated in Cuba and Mexico (Labruna et al. 2012).

The role of *Antricola* ticks as vectors of pathogens to bats and eventually humans remain unknown (Dantas-Torres et al. 2012). Nonetheless, a recent viral metagenomic investigation reported a high diversity of viruses in *A. delacruzi* in the western Amazonian region of Brazil (Blomström et al. 2019). Remarkably, several of these viruses presented low genetic sequence similarity to previously described viruses, suggesting the existence of several undescribed viral species (Blomström et al. 2019). These findings certainly points to the need for further studies to assess the diversity of microorganisms *Antricola* ticks may carry as well as to investigate their potential risk for bats and, eventually, for humans.

Three *Antricola* species are known in Brazil: *Antricola guglielmonei*, *A. delacruzi* and *Antricola inexpectata* (Estrada-Peña et al. 2004); the validity of *A. inexpectata* has been questioned (Dantas-Torres et al. 2019). These species have been originally described in north-eastern Brazil, but later on reported in other regions of the country, eventually in association with other argasid tick species (Labruna et al. 2008). Larvae identified as belonging to the genus *Antricola* have also been collected from cave-dwelling bats (Luz et al. 2016; Muñoz-Leal et al. 2018). Data on the ecology of *Antricola* spp. are still insipient, especially regarding the population dynamics of these cave-dwelling ticks.

On May 2014, during an occasional field expedition to a bat cave located in the Caatinga dry forest of Pernambuco, north-eastern Brazil, we found specimens of 10 ticks (six nymphs and four males), whose adults were morphologically identified by one of us (FDT) as *A. guglielmonei*. This finding motivated us to conduct a 1-year longitudinal study aimed at assessing whether these ticks would be present in this cave during the whole year as well as to correlate their presence with abiotic factors and abundance of bats.

## Materials and methods

### Study area

The Meu Rei cave, where the present longitudinal study was carried out, is located within the boundaries of the Catimbau National Park (hereafter CNP), in the municipality of Tupanatinga, state of Pernambuco, north-eastern Brazil (08° 29' 12.0'' S, 37° 16' 48.0'' W; 777 m above sea level; Datum WGS 84) (Fig. 1). CNP has a total area of approximately 62,200 hectares and is considered a priority area for the conservation of the Caatinga—the largest tropical dry forest region in South America (MMA 2002; Silva et al. 2017). The region's climate is classified as hot semi-arid climate (*BSh*, according to the Köpper-Geiger classification), with an average annual temperature of 23 °C and well-defined rainy (March–June) and dry seasons (July–February) ranging from 486 to 975 mm (Silva et al. 2011, 2017; Specht et al. 2019). Valleys and mountains mark the CNP's relief, with altitudes ranging from 400 m to 1,000 m above the sea level (SNE 2002).



**Fig. 1** Meu Rei cave, located in the Catimbau National Park, state of Pernambuco, north-eastern Brazil. **a** Cave overview. **b** Cave entrance. **c** Cave chamber where *Antricola* ticks were found. **d** Several aggregated *Antricola* ticks on the cave wall. Photos by Eder Barbier (**a** and **d**) and Narjara Tércia Pimentel (**b** and **c**) (colour figure online)

Meu Rei is a sandstone cave, ca. 162 m long, and can be subdivided into four main chambers, which differ mainly by temperature, humidity, bat species present and their concentrations. Meu Rei cave is recognized as having a high relevance for bat conservation (Azevedo and Bernard 2015). This cave shelters populations of at least 10 species of bats, which may reach up to 120,000 individuals at some times, with the big naked-backed bat *Pteronotus gymnonotus* (Mormoopidae) being the most abundant species (Otálora-Ardila et al. 2019). Expeditions in this cave for the study of bats and their ticks were authorized by MMA/ICMBio (SISBIO permits #43816-1 and #43816-2).

## Tick collection and identification

From July 2014 to June 2015, ticks were collected on a monthly basis in the Meu Rei cave. With the help of flashlights, walls, ceilings (areas up to about 2 m high), and accumulated guano on the cave floor were examined for ticks. We also shallowly excavated the substrate (ca. 10 cm deep) at random points to check for the possible presence of ticks hidden in the innermost part of the cave—between 120 and 150 m from the entrance—, where the greatest accumulation of guano occurs. These guano samples were examined *in loco*.

Ticks were collected with forceps and deposited in labelled vials containing 70% ethanol, with the location in the cave where they were found being recorded. In each sampled month, the search for ticks lasted 1 h (similarly divided among wall, ceiling, and cave floor) and it was always started after the bats left the cave (which often occurred between 7:00 pm and 7:30 pm).

Ticks were examined under a stereomicroscope and identified to genus and species level using taxonomic keys for Brazilian ticks (Dantas-Torres et al. 2019). Nymphs were identified to the genus level only, as presently there is no description for nymphs of *Antricola* spp. occurring in Brazil (Barros-Battesti et al. 2013; Dantas-Torres et al. 2019). Voucher specimens from this study were deposited in the ectoparasite collection of the corresponding author (FDT), under the accession numbers CVBD-0055 and CVBD-0056.

## Abiotic data collection

We installed two data loggers (model HOBO® U23-001 Pro V2) to monitor the temperature and relative humidity inside the cave, one ca. 60 m from the entrance, and the other in the deepest part of the cave, ca. 150 m from the entrance. The choice of those monitoring points was based mainly on the higher presence of bats at those parts of the cave. The data loggers were set to take temperature and humidity readings at 60-min intervals and data were downloaded monthly.

## Data analysis

We performed simple linear regression (Ordinary Least Squares) to verify whether the presence of ticks was correlated with environmental variables, using monthly mean of temperature and humidity (independent variables) and the abundance of ticks (dependent variable). Additionally, we tested a possible correlation between the abundance of ticks and the abundance of bats in the cave. For this, we used monthly bat abundance data between October 2014 and June 2015, available from Otálora-Ardila et al. (2019). We used the

**Table 1** Number of *Antricola* ticks collected monthly in Meu Rei cave, north-eastern Brazil, from July 2014 to June 2015, according to developmental stage

Year	Month	Nymphs	Males	Females	Total
2014	July	24	11	1	36
	August	59	21	0	80
	September	45	24	10	79
	October	30	30	2	62
	November	40	24	5	69
2015	December	44	8	2	54
	January	10	20	8	38
	February	15	24	16	55
	March	3	4	3	10
	April	2	3	2	7
	May	0	0	0	0
	June	0	0	0	0
Total		272	169	49	490

Males and females were identified as *A. guglielmonei*

**Table 2** Developmental stage and microhabitat exploited by *Antricola* ticks in Meu Rei cave, north-eastern Brazil, from July 2014 to June 2015

Tick stage	Wall (%)	Ceiling (%)	Total (%)
Nymph	265 (54.0)	7 (1.4)	272 (55.5)
Adult	215 (44.0)	3 (0.6)	218 (44.5)
Total	480 (98)	10 (2)	490 (100)

Adults were identified as *A. guglielmonei*

software SigmaPlot v.14.0 (Systat Software, San Jose, CA) for all analyses, with a significant level of  $\alpha=0.05$ .

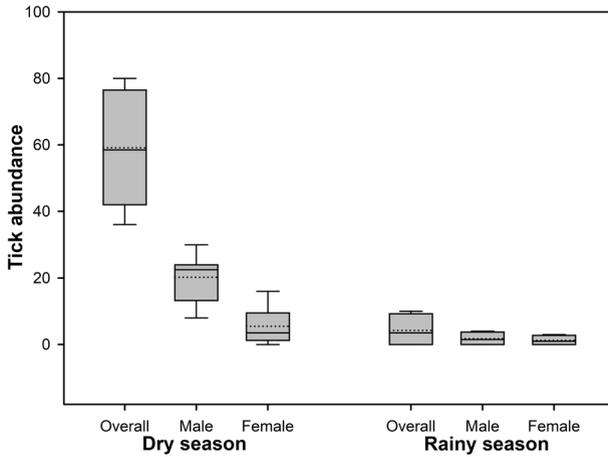
## Results

Four hundred and ninety argasid ticks were collected during the whole study period, including 272 nymphs and 218 adults (169 males and 49 females; sex ratio 3.5:1) (Table 1). All adults were identified as *A. guglielmonei*. All nymphs were identified as belonging to the genus *Antricola*.

Ninety-eight percent of the ticks were collected on the cave walls, between 80 and 120 cm above the ground level, and 2% on the ceiling. No specimens were found on the floor and/or guano (Table 2). Adults were usually clustered in the crevices and little mobile, whereas nymphs were dispersed and more active, moving over the walls or ceiling of the cave. All specimens were collected in the final portion of the cave, where the colony of big naked-backed bats was concentrated (Fig. 1).

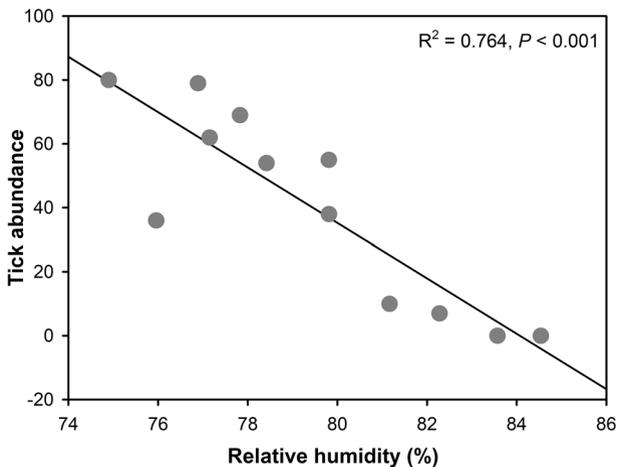
Ticks were present during all months, except in May and June 2015. Overall, ticks were more frequently collected during the dry season (July 2014–February 2015) (96.5%) as compared to rainy season (March 2015–June 2015) (3.5%) (Fig. 2; Table 1).

Considering the entire study period, the mean temperature and relative humidity in the middle region of the cave were 25.3 °C (range 24.7–32.2 °C) and 87.4%



**Fig. 2** Abundance of *Antricola* ticks ( $n=490$ ) sampled in the Meu Rei cave, north-eastern Brazil, in the dry (July 2014–February 2015) and rainy (March 2015–June 2015) seasons

(62.4–94.9%), respectively. In the deepest part of the cave, the mean values for these variables were 28.3 °C (27.6–31.5 °C) and 79.6% (72.5–89.2%), respectively (see Supplementary Material 1). Considering only the data obtained from the chamber where the ticks were found, monthly mean temperature did not correlate with tick abundance ( $N=12$ ,  $R^2=0.145$ ,  $P=0.22$ ). On the other hand, there was a negative correlation between monthly mean relative humidity and tick abundance ( $N=12$ ,  $R^2=0.764$ ,  $P<0.001$ ) (Fig. 3). The number of sheltered bats in the cave did not correlate with the abundance of ticks in the environment ( $N=09$ ,  $R^2=0.338$ ,  $P=0.10$ ).



**Fig. 3** Linear regression showing a negative correlation between the abundance of *Antricola* ticks and the relative humidity in a chamber of the Meu Rei cave, north-eastern Brazil, from July 2014 to June 2015

## Discussion

This study reports the presence and abundance of *A. guglielmonei* in a bat cave in the Caatinga drylands of Pernambuco, including aspects of its biology and ecology. This tick species was originally described from a holotype female and an allotype male collected in a cave in the state of Sergipe, north-eastern Brazil (Estrada-Peña et al. 2004), about 260 km from our study site (cave Meu Rei). In the original description of this species, the authors pointed out a higher number of males than females (129 and 56, respectively) (Estrada-Peña et al. 2004), a pattern also observed by us (169 and 49, respectively). Considering that females are larger than males (Estrada-Peña et al. 2004), the lower number of the former may suggest a distinct behaviour of females, which could make them more difficult to find, or a sexual disproportion suggesting that the populations of *A. guglielmonei* from north-eastern Brazil are male-biased. Apparently, populations of *A. delacruzi* that also occur in bat caves in north-eastern Brazil show the same trend towards male bias (Estrada-Peña et al. 2004). On the other hand, other authors reported the presence of 20 females and only five males of *A. guglielmonei* and 31 females and only eight males of *A. delacruzi* in a bat cave in Porto Velho, Rondônia state, northern Brazil (Labruna et al. 2008). Although the set of data available in the literature limits a more robust interpretation, this difference between regions can be an indication that different phenological patterns occur for the same species on a spatial scale.

Interestingly, in previous studies, *A. guglielmonei* was always found in association with *A. delacruzi* (Estrada-Peña et al. 2004; Labruna et al. 2008), which contrasts with our findings. This fact may be related, for instance, to the composition of bat species that occur in the cave. *Antricola guglielmonei* and *A. delacruzi* can be easily distinguished morphologically by a combination of characters (Estrada-Peña et al. 2004; Dantas-Torres et al. 2019). In particular, *A. guglielmonei* is smaller than *A. delacruzi* and presents the dorsum covered with tubercles as compared to the dorsum devoid of tubercles in *A. delacruzi*. A third species, *A. inexpectata*, was described from female specimens collected in Araripe municipality, Ceará, north-eastern Brazil, based on females resembling *A. guglielmonei* (Estrada-Peña et al. 2004). This species has been treated as valid in the last list of Brazilian ticks, but the authors stated that further study might reveal if it is conspecific with *A. guglielmonei* (Dantas-Torres et al. 2019).

Among argasid ticks, the occurrence of maternal care was previously reported in *A. marginatus* in Cuba and Mexico (Labruna et al. 2012). This type of behaviour is hypothesized as an adaptive facilitating mechanism, ensuring that the newly emerged larvae find their bat host for mandatory feeding in order to continue their life cycle (Labruna et al. 2012). We found no females carrying larvae, which suggests that maternal care may not be a common behaviour in all females of the genus *Antricola*.

The abundance of *Antricola* ticks was not correlated with the temperature in the studied cave, but this abiotic variable was very stable throughout the study period (see Supplementary Material 1). Nevertheless, it is known that *A. guglielmonei*—as well as *A. delacruzi* and *Ornithodoros rondonienseis*—tolerate higher temperatures than those recorded in the cave Meu Rei (Nava et al. 2010). These three species have been found in a cave with temperature between 33 and 38 °C, in the western Brazilian Amazonia (Nava et al. 2010). *Ornithodoros moubata* and *Ornithodoros savignyi*, other argasid ticks, tolerate air temperatures up to 63 and 75 °C, respectively (Lees 1947); but such tolerance may vary between genera and species. Due to the limited information

available on the biology of argasid ticks, species-specific temperature tolerance among other argasids remains unknown. Further studies, mainly in caves that offer a wide range of temperature variation, are recommended.

Unlike from what was observed with respect to temperature, the abundance of *Antricol* ticks was negatively influenced by cave relative humidity—i.e., as the cave became more humid, ticks became scarcer in the environment. In species distribution modelling studies including cave and non-cave areas in North America, Donaldson et al. (2016) found that precipitation or temperature alone were weak factors for predicting the occurrence of the argasid tick *Ornithodoros turicata*. Those authors also observed that only models including at least five variables (i.e., maximum temperature of warmest month, mean temperature of wettest quarter, mean temperature of driest quarter, precipitation seasonality, and precipitation of warmest quarter) were more accurate when predicting the distribution of this tick. In addition, Donaldson et al. (2016) argued that the composition and structure of the host community in a given location is likely to influence the ecology of argasid tick populations. However, as caves provide a more stable environment in relation to the external environment, it is possible that the predictors for the occurrence of argasid ticks associated with it are not the same for those that occur outside it. Furthermore, the ability to detect fluctuations in the population size of argasid ticks can also be influenced by their diapause behaviour and/or by hiding in difficult to access and unexamined sites, for example.

There was no information on the seasonality of cave-dwelling argasid ticks in Brazil, and our results are the first to provide some inputs in this sense. Ticks were present during most of the year, but more abundant during the dry season and absent in May and June 2015. Even though the number of ticks in the environment had no correlation with the number of bats sheltering in the cave, bats can alter cave microclimate due to their presence and, especially under very high densities, like in caves with *Pteronotus* spp., which may harbour more than 100,000 bats in them (Ladle et al. 2012). In such conditions, cave humidity and temperature varies as a by-product of the bats' metabolism (Kunz 1982). Extrapolating the magnitude of the effect of bat metabolic rates, a study showed that a colony with 100,000 individuals could raise the temperature of a cave by about 23 °C in an hour if it were a closed system (Baudinette et al. 1994). This increase in temperature can be even more pronounced in the case of maternity colonies (Davis et al. 1962; Baudinette et al. 1994).

As argasid ticks are often associated with caves where large populations of mormoopid bats are sheltering, it was expected that there would be a positive correlation in the abundance of these two groups. A recent 2-year study indicated large fluctuation in the population of big naked-backed bats, but still without a clear phenological pattern (Otálora-Ardila et al. 2019). The fact that population of big naked-backed bats present such dynamism, as reported by Otálora-Ardila et al. (2019), may be an indication that the same would occur with cave-dwelling argasid ticks. Due to the longevity of argasid ticks in the off-host period, it is likely that their abundance is, to some extent, asynchronous in relation to that of bats. That is, the abundance of argasid ticks in the environment in a given period would be related to the abundance of its host in the past. Therefore, long-term studies will help to shed light into this understanding.

Along with previous data (Estrada-Peña et al. 2004; Labruna et al. 2008; Nava et al. 2010), our results indicate that *A. guglielmonei* is a common tick species in bat caves in Brazil. In summary, our data also suggest that this species may be more abundant during the dry season and that it is negatively affected by high rates of relative humidity within the cave (> 80%). However, further research on the development, reproduction, life span, and

dispersal capacity of argasid ticks in Brazil should be encouraged in order to better appreciate their biology and ecology.

**Acknowledgements** This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior—Brasil (CAPES)—Finance Code 001, Fundação Grupo Boticário de Proteção à Natureza (Grant No. 0983-20132), and Anglo American. The first author received a doctoral scholarship from CAPES (2014–2018) and has a postdoctoral grant from CAPES and Fundação de Amparo à Ciência e Tecnologia do Estado de Pernambuco (FACEPE) (Process No. 88887.353052/2019-00). F. Dantas-Torres and E. Bernard are research productivity grantees from Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq; Process Nos. 313118/2018-3 and 305079/2017-4, respectively). We are grateful to the Catimbau National Park staff, and to the team of the Programa de Pesquisas Ecológicas de Longa Duração – PELD Catimbau, especially Marcelo Tabarelli and Inara Leal, for the logistical support.

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