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Short Communication

Novel breeding habitat, oviposition microhabitat, and parental care in *Bokermannohyla caramaschii* (Anura: Hylidae) in southeastern Brazil

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Selection of novel breeding habitats and oviposition microhabitats are influenced by the plasticity of a species to adapt to habitat changes (Silva and Giaretta 2008, Campos *et al.* 2013). The mountainous region of the Brazil's Atlantic Forest provides a wide range of habitats and microhabitats. The Atlantic Forest has changed from continuous forest to smaller and isolated remnants, requiring anurans to cross over or even reproduce on human-modified habitats (Ferreira *et al.* 2016, Mageski *et al.* 2018).

Breeding habitat and oviposition microhabitat influence parental care in anurans (Storti *et al.* 2019), because parental care influences the survivorship of offspring (Furness and Capellini 2019). Anurans exhibit a variety of parental care strategies, from egg guarding to tadpole feeding (Wells 2007). More complex parental care requires more energy expenditure and physiological and morphological adaptations. Egg and tadpole guarding are present in 6% of the anurans in the world (Furness and Capellini 2019). Parental care is important for species that breed in water bodies with reduced capability for rainwater storage, such as bromeliads, bamboo, and cattle footprints (Gally and Zina 2013, Ferreira *et al.* 2019).

Endemic to the Atlantic Forest, the treefrog *Bokermannohyla caramaschii* (Napoli, 2005) is distributed across mountainous forests above 650 m in the states of Minas Gerais and Espírito Santo, southeastern Brazil (Napoli 2005, Frost 2020). Females lay egg masses in rocky cavities backwater areas of streams (Pezzuti *et al.* 2015). The present study characterizes a new type of

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breeding habitat and oviposition microhabitat, and, in addition, describes parental care in *B. caramaschii.*

The study was carried out in Parque Estadual do Forno Grande (PEFG; 20.311951° S, 41.64949° W, WGS 84; 1200 to 1535 m a.s.l.), municipality of Castelo, in a mountainous region of the state of Espírito Santo, southeastern Brazil. The PEFG has 913 hectares of Dense Montana Ombrophylous Forest with rough terrain that reaches up to 2039 m a.s.l. (Simonelli and Fraga 2007).

We conducted nocturnal sampling in October 2016 using active visual and auditory searches. We measured and characterized the oviposition microhabitat and breeding habitat of *B. caramaschii,* including diameter, depth, and distance to the nearest stream. We collected two specimens (MBML 10618 and MBML 11072; collection permits: IEMA 52838-1; SISBIO 50402) and deposited them at the Museu de Biologia Mello Leitão (MBML) from Instituto Nacional da Mata Atlântica, Santa Teresa, Espírito Santo state, Brazil.

We defined used pools by having spawning eggs and/or tadpoles. Those with eggs or tadpoles were defined as “used pools”. We evaluated the differences between used and unused oviposition microhabitats regarding diameter, depth, and distance to the nearest stream by using T-tests. We ran the Shapiro-Test Normality Tests and Two Sample T-tests using the package “vegan” 2.5-6 in the R 3.6.1 (R Core Team 2019).

We found *Bokermannohyla caramaschii* at two sites (Site A and B) at PEFG. Site A had adults, egg masses, and tadpoles in water-filled rocky cavities (usual oviposition microhabitat) along a stream (usual breeding habitat; Figure 1). Site B had adults, egg masses, and tadpoles in pools formed in cattle footprints (novel oviposition microhabitat) near a stream in a pasture (novel breeding habitat; Figure 2).

Site B (2.7 × 3.9 m) had 19 pools, of which nine (47%) pools had eggs and/or tadpoles (used pools) and 10 (53%) pools had no eggs and/or tadpoles (unused pools) of *B. caramaschii.* The used pools were larger (t = -2.10; df = 14.81; p < 0.05) and deeper (t = -3.17; df = 13.23; p < 0.05) than unused pools (Figure 3). The used pools had a mean diameter of 16.2 ± 3.7 cm and a mean depth of 6.4 ± 3.1 cm. The unused pools had a mean diameter of 11.9 ± 4.5 cm and a mean depth 2.6 ± 1.9 cm. However, both used and unused pools were approximately the same distance from the stream (t = 1.21; df = 13.40; p = 0.88). The used pools were 16.6 ± 6.2 cm from the stream and unused pools were 22.1 ± 12.6 cm (Figure 3).

At site A, adults of *B. caramaschii* were near egg masses (i.e., egg guarding; N = 2) and tadpoles (i.e., tadpole guarding; N = 2) in rocky cavities. Also at site A, carcasses of adults (N = 2) were preyed upon near a spawning site (Figure 4). At site B, no adult was observed near the eggs or tadpoles.

This report is the first to describe egg deposition by *B. caramaschii* in cattle footprints in a pasture. In addition, it is the first record of species of *Bokermannohyla* spawning in temporary pools. The congeners *B. napolii* Carvalho, Giaretta, and Magrini, 2012 and *B. izecksohni* (Jim and Caramaschi, 1979) also oviposit in water-filled backwater pools and rocky cavities near streams. *Boana pardalis* (Spix, 1824) and *B. faber* (Wied-Neuwied, 1821) are the most closely related species that use cattle footprints for oviposition (Bokermann 1968, Andrade *et al.* 2017). Our data demonstrate that *B. caramaschii* has the ability to utilize human-modified breeding habitats and oviposition microhabitats.

The selection of deeper and larger pools shows the ability of *B. caramaschii* to influence the survival of the offspring. Deeper and larger pools probably reduce the risk of desiccation because these pools may store water for a longer time compared to small shallow pools. This selection of microhabitats probably increases the chance of reproductive success. Oviposition in temporary water bodies such as cattle footprints and rock pools reduces the risk of predation of offspring compared to permanent water bodies

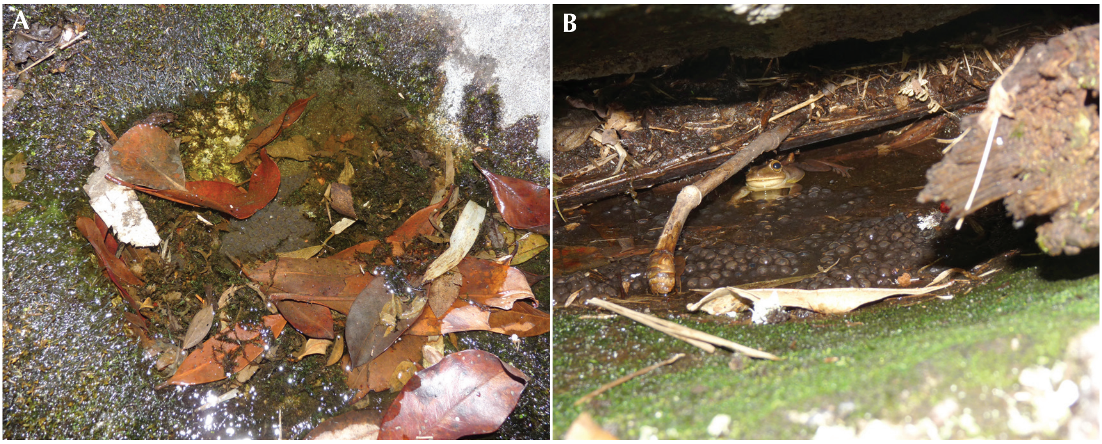

Figure 1. (**A**) Spawning of *Bokermannohyla caramaschii* in a water-filled rocky cavity (usual microhabitat) along a stream (usual habitat) and (**B**) adult guarding eggs and tadpoles (parental care) at Parque Estadual do Forno Grande, state of Espírito Santo, southeastern Brazil. Photos: TSS.

Figure 2. (**A**) Pools formed by cattle footprints (novel microhabitat) in a pasture (novel habitat) and (**B**) close-up of a pool with eggs and tadpoles of *Bokermannohyla caramaschii.* Photos: TSS.

where the density of fish increases predation of larvae (Rieger *et al.* 2004).

We provide the first report on parental care for species of *Bokermannohyla.* Egg guarding is the most common parental care in anurans, possibly because this behavior requires less energy compared to other types of parental care (Wells 2007). Egg guarding decreases the rate of egg predation, thus increasing reproductive success (Furness and Capellini 2019). *Boana faber* oviposits in temporary pools, and also displays egg and tadpole guarding (Martins *et al.* 1998). However, parental care can also increase the risk of predation of the caring parent because the parent remains immobile near the offspring (Clutton-Brock 1991).

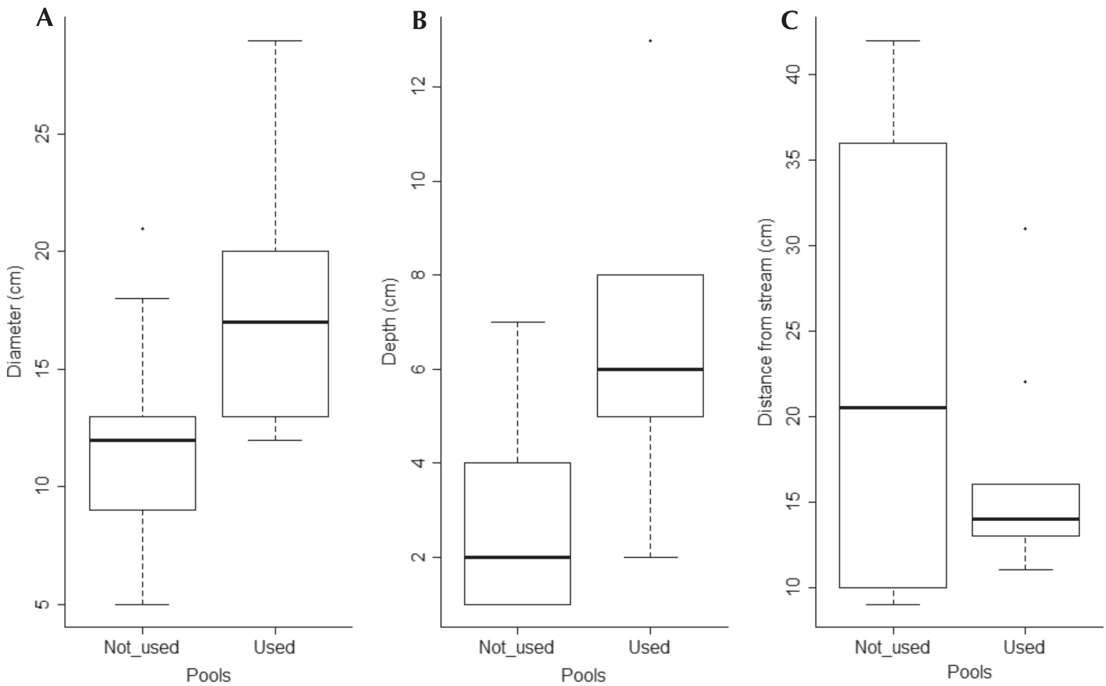

Figure 3. (**A**) Diameter, (**B**) depth and (**C**) distance from stream between non-used and used cattle footprints by eggs and tadpoles of *Bokermannohyla caramaschii.*

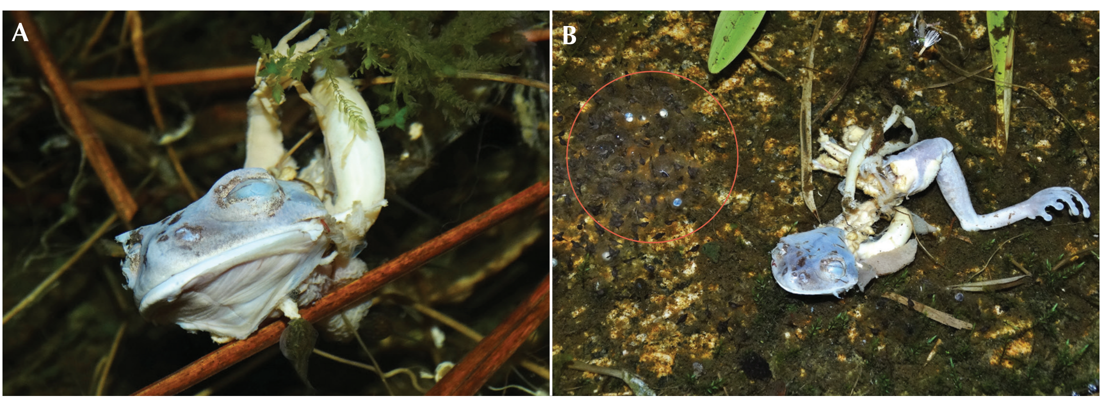

Figure 4. (**A**) Carcass of *Bokermannohyla caramaschii* preyed upon in a water-filled rocky cavity along a stream. (**B**) Carcass near a spawning. Photos: ATM.

Our study contributes to understanding reproductive plasticity of microhabitats and habitats used during egg deposition by *B. caramaschii.* It is not possible to determine the environmental pressure for *B. caramaschii* using a new oviposition microhabitat and habitat. Nevertheless, the use of oviposition microhabitat and habitat does not occur randomly and is probably critical to avoid desiccation of eggs and tadpoles. Egg and tadpole guarding are likely to increase survival of offspring. We recommend that future studies evaluate the influence of habitat and microhabitat selection on development and survival of eggs and tadpoles of *B. caramaschii.*

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