

SHORT COMMUNICATION

Mating behavior of *Anolis punctatus* (Squamata: Dactyloidae) in the Brazilian Amazonia

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Lizards perform a broad behavioral repertoire during their courtship and mating events that allows recognition of the opposite sex, males to assess female receptivity, and females to choose a mate. These behaviors ultimately maximize mating success (Tokarz 1995, 2007, Simon 2011). Studying such behavior is particularly interesting in diurnal, highly visually oriented and territorial lizards, such as those of the genus *Anolis* Daudin, 1802 (Jenssen 1978, Bull 2000, Losos 2009, Reedy *et al.* 2017). During courtship events, anole lizards emit a diverse array of visual displays, including head bobbing, push-ups, tail lifting, throat dewlap extension, and/or changing color (Losos 2009, Simon 2011, Driessens *et al.* 2014, Steffen and Guyer 2014, Beltrán *et al.* 2016). Most of these visual displays are stereotypic and emitted in other social contexts, such as during territorial interactions

(Jenssen 1978, Losos 2009, Reedy *et al.* 2017, Horr 2019). Regarding the behaviors performed during mating events, males of anole lizards most commonly bite the nape of females, grasp them, and consummate mating by inserting one of their hemipenes (Losos 2009).

The duration of lizard courtship and mating events is highly variable and influenced by a trade-off between the benefits of reproduction and exposure to a greater predation risk (Lima and Dill 1990, Cooper 1999, Simon 2007, Gerhardt 2014). Mating events of anoles follow this pattern and can vary in duration among species from less than one second to more than one hour, but events of several minutes are more commonly reported (Losos 2009, Alfonso *et al.* 2014, Beltrán *et al.* 2016). Mating duration can also differ among breeding pairs of a single anole species, increasing throughout the breeding season (Losos 2009) or in response to the presence of a predator or an observer (Beltrán *et al.* 2016). Nevertheless, observing and gathering behavioral data to improve knowledge of these

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events can be particularly challenging, especially considering species that usually mate perched on trees, such as anoles. As a result, much of the knowledge associated with reproductive events of these lizards comes from observations of laboratory experiments or captive animals (e.g., Stamps 1975, Lima and Souza 2006, Pandav *et al.* 2007, 2010, Driessens *et al.* 2014).

During recent fieldwork in Amazonian Brazil, JAO observed a complete reproductive event of the Amazon Green Anole (*Anolis punctatus* Daudin, 1802). This anole is a thermoconforming species, mainly arboreal, and distributed in both Amazonia and the Atlantic Forest (Ávila-Pires 1995, Vitt *et al.* 2003). Its mating behavior was briefly described based on a casual observation by Silva-Neto *et al.* (2019) in central Amazonia (as *Anolis philopunctatus* Ávila-Pires, 1995, currently synonymized with *A. punctatus*). To our knowledge, a complete description of its reproductive behavior and mating duration remain unknown. Herein we describe this reproductive event and compare it to the known behavior for this species and its congeners.

Individual behaviors were sampled through focal animal and all-occurrence sampling methods (Altmann 1974, Lehner 1996). To avoid interfering with the emitted behavioral signals, the observer remained silent and at least 3 m from the individuals during the entire reproductive event. The reproductive event was recorded using photos and video footage with the aid of a digital camera (Canon t3i; Tokyo, Japan) and a telephoto lens (Canon EF 70-300 mm USM; Tokyo, Japan). Individuals were sexed by analyzing sexually dimorphic characteristics of the external morphology. Males of *A. punctatus* have larger body size, more elongated snouts, bright orange-colored throat dewlaps, and thicker tail bases (hemipenial pouches) (Ávila-Pires 1995, Vitt *et al.* 2003). Females have thinner bodies and tail bases, shorter snouts, and no dewlaps (Ávila-Pires 1995, Vitt *et al.* 2003).

This observation occurred on 08 September 2020 at the peak of the regional dry season along

one of the trails of the Cristalino Lodge, which is part of the Cristalino Private Natural Heritage Reserve. This ecotourism lodge is located on the western bank of the Cristalino River in southern Amazonia (Alta Floresta, Mato Grosso state, Brazil; 09°35'51" S, 55°55'53" W, datum WGS 84). The regional climate in this locality is hot and humid with a pronounced seasonality. The annual means of temperature and rainfall reach 26°C and 1,950 mm, respectively (Alvares *et al.* 2013). Most of the regional landscape is dominated by *terra firme* forest that is not susceptible to the seasonal flooding of the Cristalino River. *Anolis punctatus* typically is found in this habitat (Ávila-Pires 1995, Vitt *et al.* 2003). The air temperature was around 24°C during the observation, which lasted approximately two hours.

At 09:00 h, two adult male *A. punctatus* were spotted at an observation station within the forest. These males were perched on distinct trees about 10 m apart, where they adopted survey postures, observing their surroundings by laterally moving their heads, and walking around for short distances. Sporadically, these males used visual displays during territorial interactions, such as flexing the anterior body by doing push-ups and alternating body color between greenish and brownish tones. At 09:45 h, one of these males approached a tree trunk with a diameter of about 15 cm, showing clear signs of agitation by slightly extending its dewlap (Figure 1A), head bobbing, and acquiring a stronger brown color. This male jumped on a nearby tree, disappearing from the field of vision of the observer. When going around the tree to see the lizard, the observer noticed that the male was then mating with a female (Figure 1B), suggesting that the previous behaviors were visual displays emitted in a courtship context. Because the female was out of the initial field of vision, the observer did not obtain information on the displays emitted by her prior to mating or on the behaviors that triggered the observed mating position. When the breeding pair was spotted, the male was curving its body upon the female, immobilizing

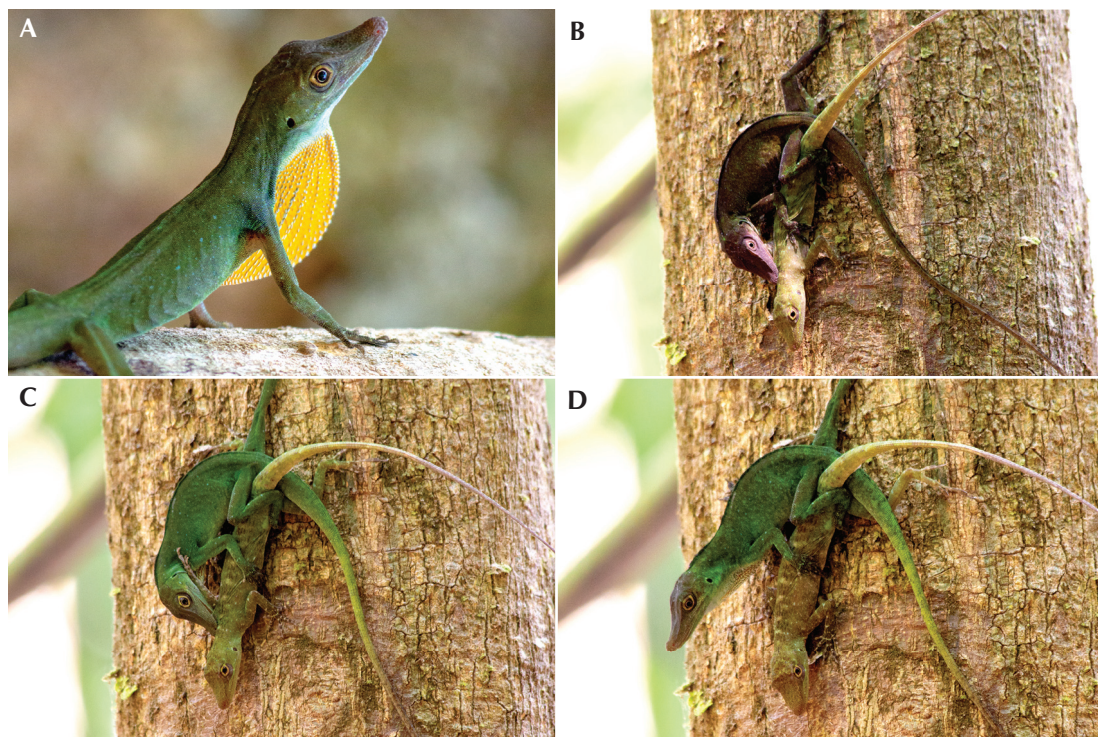


Figure 1. Mating behavior of *Anolis punctatus*, recorded in southern Brazilian Amazonia (Mato Grosso state, Alta Floresta, RPPNs Cristalino). (A) Male extending its dewlap. (B) Mating, with male immobilizing the female and acquiring an intense and uniform brown color. (C–D) Near the end of the mating, female trying to disengage, and male acquiring a bright green color and later stopping biting her neck.

her using a firm bite at the nape (right side), the left arm holding her at the midbody, and the left leg around her inguinal region (Figure 1B). The right limbs of the male were propped on the trunk, giving him support. Assuming this position, the male was most likely inserting its left hemipenis into the female. Both individuals had their heads toward the ground (Figure 1B).

Once in this position, the individuals remained almost immobile during the entire mating event, with some exceptions listed below. We noticed subtle movements of the female's tail and the base of the male's tail at least 19 times throughout the mating event, suggesting insertion of the hemipenis and insemination. After about 36 min, the female became agitated and displayed random

movements, apparently trying to disengage herself from the male on three occasions. The male continued to immobilize her through the same previously described strategies (Figure 1C). After 71 min (near the end of the mating event) the female made a new attempt to disengage, and the male stopped biting her nape (Figure 1D). After stopping the bite, the male immediately extended its dewlap and performed rapid head bobbing. Approximately 7 min later, the female shook her body again, and the male repeated the head bobbing. A few seconds later, the female made a new attempt to disengage herself, slightly moving in a short distance and dragging the lower body region of the male, as he continued immobilizing her in the inguinal

region with his left leg. After 6 min, the male began to disengage himself from the female, apparently retracting most of the hemipenis after the female lifted the tail. Both individuals moved separately a short distance, the male toward the canopy, and the female toward the ground, thus ending the 84-min mating event at 11:09 h. After disengaging, the male remained with the tail slightly raised, rubbing the cloaca toward the substrate while completing the retraction of the hemipenis. Both individuals remained immobile until the end of the observation period (87 min).

During the mating event, the overall color of both individuals varied. The male started the mating with a brownish green color and acquired a considerably intense and uniform brown color over time (Figure 1B). During the female's attempts to disengage, the male abruptly changed to a greenish color, returning to intense brown when she became passive. After about 60 min the male, still immobilizing the female, acquired a bright green color typical of the species, and remained that way until they disengaged (Figure 1C, D). The color of the female remained relatively stable throughout the mating event, ranging from light brown at the beginning to brownish green at the end (Figure 1B–D).

Although we did not observe the entire courtship event, behaviors recorded from the breeding pair of *A. punctatus* prior to the mating were congruent with those displayed by most anole species (i.e., male head bobbing, push-ups, and extending dewlaps; Rodrigues 1988, Losos 2009, Simon 2011, Cook *et al.* 2013, Beltrán *et al.* 2016). According to the literature, the behavior and positioning of individuals during mating, as well as the chosen substrate, may vary among anole species (Losos 2009). The positioning of the breeding pair of *A. punctatus* and the chosen substrate were almost entirely consistent with that previously observed *in-situ* for this species in central Amazonia (Silva-Neto *et al.* 2019). The single notable divergence was the direction of the heads of the individuals: toward the ground in this observation and toward the canopy in the report of Silva-Neto *et al.* (2019).

Compared to congeneric species, the observed mating position of *A. punctatus* resembled that reported for *Anolis smallwoodi* Schwartz, 1964 (Alfonso *et al.* 2014) and *Anolis notopholis* Boulenger, 1896 (Beltrán *et al.* 2016), although the latter has only been recorded mating on the ground.


The observed behavior of several disengagement attempts by the female *A. punctatus* was not reported for this species by Silva-Neto *et al.* (2019), but it was reported for the congeneric sympatric anole *Anolis fuscoauratus* D'Orbigny, 1837 (Rodrigues 1988). Rodrigues (1988) suggested that this behavior could derive from female dissatisfaction with exposure during mating and substrate choice (a tree trunk), as she remained immobile in a subsequent mating attempt, with the breeding pair sheltered under a palm leaf. Alfonso *et al.* (2014) also reported abrupt movements of females of *A. smallwoodi* during mating events but attributed them as a response to hemipenial insertion (copulatory phase). This idea was supported by the fact that they did not observe abrupt movements during the rest of the mating events (Alfonso *et al.* 2014). Considering these reports for congeneric species, it is impossible to determine whether the “disengagement behavior” observed here for the female *A. punctatus* was triggered by discomfort with mating conditions such as the chosen substrate, a reaction to hemipenial insertion and insemination, or even some additional factor.

Changing color from hormonal control is a common feature of several anole species and occurs in a stereotypic manner in many social contexts, such as exploration, territorialism, escape, aggressiveness, and stress (Jenssen *et al.* 1995, Greenberg 2002, Horr 2019). Color changes during mating events have not been widely reported for these lizards, except for *A. notopholis* (Beltrán *et al.* 2016). In fact, Silva-Neto *et al.* (2019) did not report color change for mating individuals of *A. punctatus*, but it is possible that such behavior went unnoticed during the short period they observed the mating event. Our observation corroborates those of

Beltrán *et al.* (2016) by showing that for *A. punctatus*, mating is an additional social interaction that may lead to a color change in anoles. We hypothesize that the change in a male's color to an intense brown may be a result of the combined effect of his excitement during the hemipenial insertion and insemination once it progressively intensified during mating, and a strategy to reduce his conspicuousness through disruptive camouflage at the most vulnerable time of mating (Boyer and Swierk 2017). Because color change is a stereotypic behavior for anoles, we cannot discard the hypothesis that this change may have been triggered by alternative social stimuli.

Mating events of anole species usually is of long duration, ranging from ca. 10–50 min (Losos 2009, Beltrán *et al.* 2016) to up to 64 min for *A. smallwoodi* (Alfonso *et al.* 2014), potentially exposing the breeding pairs to greater predation risk. The long mating duration recorded here for *A. punctatus* (84 min) is noteworthy as it is considerably above this range. Since Silva-Neto *et al.* (2019) observed a mating event of *A. punctatus* for only ca. 20 min, it is impossible to draw a parallel between our observations regarding mating duration. Nevertheless, our combined evidence supports the suggestion that long mating events are common for *A. punctatus*, and that they range from at least about 21–84 min. The long mating duration in our observation may be a result of different environmental conditions throughout the natural reproductive seasonality of the species (Losos 2009), or even a disturbance effect triggered by the presence of a nearby observer, as suggested by Beltrán *et al.* (2016). Only further studies, ideally isolating most of the extrinsic factors and involving various breeding pairs, can elucidate whether the mating duration reported here is part of the typical behavioral repertoire of *A. punctatus*.

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