The amplexus, that is, the behaviour of a male embracing a female just before egg deposition, is widely distributed among anurans (Duellman and Trueb, 1986), with few exceptions reported (e.g. Zhang et al., 2012). Inguinal amplexus, in which a male clasps a female’s waist, is considered the ancestral condition and together with axillary amplexus are the most common types of anuran amplexus (Duellman and Trueb, 1986). Different types of amplexus imply different types of contact between cloacae and therefore are considered to optimize external fertilization (Duellman and Trueb, 1986; Wells, 2007).

*Aplastodiscus* is a Neotropical frog genus which comprises 15 nominal species with occurrence in the Atlantic Forest and Brazilian Savanna, in South America (Berneck et al., 2016, 2017). The reproductive biology of *Aplastodiscus leucopygius* (Cruz and Peixoto, 1985) was comprehensively described for the first time on the basis of 12 pair formations by Haddad and Sawaya (2000). After years of natural history observations the reproductive mode of several species of *Aplastodiscus* are known as being very similar with that of *A. leucopygius*, in which the male excavates an inconspicuous burrow in the mud near the stream edge, then, usually perched on vegetation above the ground level, finds a spot and starts calling for females (Haddad and Sawaya, 2000). When a female approaches the male, both exchange a complex sequence of tactile stimuli, such as: skin contact of the hind limbs and snout with hands, chin on head, and snout with snout (Haddad and Sawaya, 2000). The sequence of mutual touches is exchanged while the male guides the female to the burrow entrance, a journey that can last from thirty minutes to several hours (see Lutz and Lutz, 1938; Cochran, 1955; Cruz and Peixoto, 1985, 1987; Haddad and Sawaya, 2000; Hartmann et al., 2004; Haddad et al., 2005; Carvalho et al., 2006; Zina and Haddad, 2006a,b, 2007). However, the female may still reject the burrow after inspecting it (Hartmann et al., 2004; Zina and Haddad, 2007; Carvalho et al., 2006; see also Berneck et al., 2016 for a review). Despite all the research available, the amplexus in *Aplastodiscus* remains unknown because reproduction takes place inside the excavated burrow and the pair is wary, fleeing after any disturbance in the nest, like the removal of dome for observation.

During fieldwork carried out on October 23, 2009 (from 10 pm to 4 am) at Parque das Neblinas, Bertioga, São Paulo, Brazil (23° 43.874’ S - 46° 10.368’ W), we observed a pair of *A. leucopygius* from the moment when the female approached the male (Figure 1A) until the arrival at the burrow (Figure 1L). The sequence and types of mutual touches was the same of that already described for *A. leucopygius* by Haddad and Sawaya (2000): the male vocalizes the courtship call (Figure 1D); the male and female touch each other snout (Figure 1E); then male touches female’s head with his snout (Figure 1F); then female touches male’s leg with her hand and snout (Figure 1G–I); male touches female head with his foot (Figure 1J) However, after they entered the burrow (Figure 1R), we collected and transferred them to a plastic bag and then we observed their amplexus. The male initiated the behaviour over the female, with its hands at the female’s jaw (Figure 2A), then he proceeded downward, still over the female body, and held the female’s flanks or thighs (Figure 2D). Then the female moved and started to switch her position while the male waited or while he was still holding her (Figure 2G). In some movements, the female stretched her body and put her hands forward as if she was pushing something. Then the pair repeated the same behaviour over and over, until eggs were laid (Figure 2).
Figure 1. Courtship behaviour of *A. leucopygius* at Parque das Neblinas, Bertioga, Brazil. A. Male calls for females. B. Female approaches the male. C. Female starts to follow the male. D. Male vocalises courtship call to guide female. E. Male and female touch each other snout. F. Male touches female’s head with his snout. G and H. Female touches male’s leg with her snout. I. Female touches male’s leg with her hand. J. Male touches female’s head with his leg. K. The pair proceeds the way to the burrow. L – R. The pair enters the burrow. S – U. Burrow’s interior.
The amplexus, as we had observed, is quite difficult to categorize. At different moments, it was cephalic (Figure 2A), inguinal (Figure 2D), and also axillary (Figure 2E).

The movements displayed by the female of *Aplastodiscus leucopygius*, of stretching her body, putting her hands forward as if she was pushing something, were quite similar to that described for the related *Boana faber* (Wied-Neuwied, 1821), in which the female renovates the nest by bringing clay from its bottom to the walls (Martins and Haddad, 1988; Martins, 1993; Martins et al., 1998). However, it was not possible to confirm this behaviour in *Aplastodiscus* because the pair was removed from the burrow.

We recognize that our observation is based on an unnatural situation: a pair in a plastic bag. However, the pair deposited approximately 140 viable eggs (Figure 2I), what may indicate that the observed behaviours occur inside the burrow. The eggs were kept in laboratory to observe their development but they got infected by a fungus and died. Additionally, we tentatively kept gravid females of *Aplastodiscus* with males in plastic bags where no laying of eggs occurred. Still, we note that the pair we observed had entered the burrow and the female had accepted the male, thus evidence suggests that we possibly observed a situation most similar to a natural one. Since 2009, we have tried to observe the process of amplexus several times without success, so we strongly

---

**Figure 2.** Amplexus behaviour and eggs of *A. leucopygius*; note that the pair has been shifting positions, presenting different amplexus types during the moments that preceded the oviposition (see the text for details of sequence of events). Parque das Neblinas, Bertioga, Brazil.
recommend for future researches of Aplastodiscus reproductive behaviour, the use of infrared endoscope cameras, unavailable to us at the time.

This pattern of amplexus has been observed for other Hylids and authors has postulated that these positions would improve egg’s fertilization or prevent the female being injured by male’s prepollical spines (e. g. Camurugi and Juncá, 2013; Martins and Haddad, 1998). Brunetti et al. (2014) described the reproductive biology of Boana punctata (Schneider, 1799) and raised an alternative hypothesis, where the skin contact of mental and lateral glands would allow the male deliver chemical signals to female during amplexus (see also Brunetti et al., 2015). As Aplastodiscus bear no prepollical spines and male and female not preset sexual size dimorphism (Haddad and Sawaya, 2000) we consider this alternative hypothesis preferable to explain the biological role of this amplexus behaviour.

Specimens observed: female: CFBH 24176, body mass of 3.21 g, CRC 4.17 cm; male: CFBH 24177, body mass of 4.2 g, CRC 3.99 cm.

Acknowledgements. We thank the Instituto Chico Mendes for license (IBAMA, #17168) and the Instituto Ecoturismo for allowing access to the Parque das Neblinas. To São Paulo Research Foundation (FAPESP) for grants #2008/55235-4, #2013/18807-8, and #2013/50741-7. Cynthia Prado kindly pre-reviewed this note. Rodrigo Lingnau and Alessandro Morais made helpful suggestions and improved this note. BVMB thank J. Faivovich for academic support.

References


Accepted by Alessandro Morais