

Functional necrophilia: a profitable anuran reproductive strategy?

Thiago J. Izzo^{a,b}, Domingos J. Rodrigues^{a,b,c*}, Marcelo Menin^{a,d}, Albertina P. Lima^a and William E. Magnusson^a

^aCoordenação de Pesquisas em Biodiversidade, Instituto Nacional de Pesquisas da Amazônia, CP 478, CEP: 69011-970, Manaus, AM, Brazil; ^bInstituto de Biociências, Coordenação de Pós-graduação em Ecologia e Conservação da Biodiversidade, Universidade Federal de Mato Grosso, Av. Fernando Correa, s/n. CEP: 78060-900, Cuiabá, MT, Brazil; ^cPresent address: Instituto de Ciências Naturais, Humanas e Sociais, Coordenação de Pós-graduação em Ciências Ambientais, Universidade Federal de Mato Grosso, Av. Alexandre Ferronato, 1200. CEP: 78557-267, Sinop, MT, Brazil; ^dInstituto de Ciências Biológicas, Departamento de Biologia, Universidade Federal do Amazonas, Av. General Rodrigo Otávio Jordão Ramos 3000, 59077-000, Manaus, AM, Brazil

(Received 31 March 2012; final version received 23 August 2012; printed 6 December 2012)

Explosive breeding is a common reproductive strategy, especially in anurans. In some cases of explosive breeding in anurans, intense aggressive interactions occur among males attempting to gain access to females, and for terrestrial species that mate in water, females may die by drowning during long mating struggles. Such occurrences are obviously detrimental to females, and may also cause a decrease in the reproductive success of males through depletion of energy without obtaining access to a live female. However, the males of the small Amazonian frog *Rhinella proboscidea* can promote the ejection of oocytes from the abdominal cavities of dead females and fertilize them. This behaviour can minimize losses to both partners during explosive reproduction events. The existence of such a “functional necrophile strategy” shows that there may be possible selection in favour of stronger and more persistent males in explosive breeders.

Keywords: amphibians; davian behaviour; reproduction; mating balls

Introduction

A reproductive behaviour or strategy may be viewed as the combination of physiological, morphological and behavioural traits that act together to maximize the number of offspring produced under given environmental conditions (Stearns 1992). Different behaviours can increase the efficiency (fitness) or minimize the costs and risks associated with reproductive processes (Harvey and Pagel 1991; Stearns 1992; Sakai and Westneat 2001). Consequently, there are a myriad of different strategies among animals, including some that are considered “socially unacceptable” and impossible in human society, such as infanticide in insects and vertebrates (Hager and Johnstone 2004), necrophilia in lizards (Costa et al. 2010) and amphibians (this study) and post-mating clutch piracy (Vieites et al. 2004).

In general, the reproductive behaviour of animals can be divided into two basic temporal patterns: prolonged breeding and short-term “explosive” breeding (see Wells

*Corresponding author. Email: djmingo23@gmail.com

1977, 2007; Odendaal et al. 1985). Explosive reproduction is common in some animal groups, and is particularly well studied in some anuran families, such as Bufonidae, Hylidae, Microhylidae and Ranidae (Wells 1977, 1979; Zimmerman and Bogart 1988; Prado et al. 2005; Rodrigues et al. 2004, 2005, 2007). Explosive breeding is generally characterized by males remaining at breeding sites for only a few days, chorus formations, synchronous arrival of the female in the pond, and low selectivity of males (active search for female) (Wells 1977). The formation of ponds, amount of rainfall, larval competition and predator abundance at breeding sites, demographic factors such as age at first reproduction and frequency of reproduction of individual females could have led to the evolution of explosive breeding (Wells 1977; Aichinger 1987; Menge and Olson 1990; Prado et al. 2005). The males of species with explosive breeding generally form large mating aggregations. In this situation, the females are available for a short time (Menin et al. 2006) and the males occur in higher density. Hence, the opportunity for female choice should be limited by intense male–male competition (Wells 2007). Indeed, the males use a lot of energy searching for mates and struggling over females (Wells 1977, 2007). Unpaired males try to displace those already in amplexus, resulting in large mating balls and this behaviour is sometimes fatal to females (Verrell and McCabe 1986; Menin et al. 2006; Wells 2007). For some of these species, females may be drowned under the weight of several males attempting to mate (Duellman and Trueb 1994; Trauth et al. 2000; Wells 2007). Such occurrences imply a loss of fitness for males as well as females because if reproductive contests between males take too long, and females die, there is a decrease in the reproductive success of males through loss of energy in fights that do not result in reproduction. In this case, the evolution of a behaviour trait that allows this energy and fitness loss to be minimized should be favoured. *Rhinella proboscidea* (Anura: Bufonidae) is a small central Amazonian frog that exhibits intense male–male competition for mating opportunities. This species is an explosive breeder, and up to several hundred males congregate in small streamside ponds or headwaters of streams for 2 to 3 days (Lima et al. 2006; Menin et al. 2006; Rodrigues et al. 2010), where thousands of fertilized eggs are laid (Figure 1A). Here we show that males of *R. proboscidea* can extract and fertilize oocytes from dead females, which is a previously undescribed reproductive strategy for amphibians.

Material and methods

Study area

This study was conducted between June 2001 and June 2005 during the rainy seasons in the primary forest of Reserva Florestal Ducke (between 02°55' and 03°01' S, 59°53' and 59°59' W), adjacent to the city of Manaus, Amazonas state, Brazil. The reserve covers 10,000 ha of *terra-firme* rain forest, a well-drained forest not subject to seasonal inundation. The forest is characterized by a 30-m to 37-m tall closed canopy, with emergent trees growing to 40–45 m (Ribeiro et al. 1999). The understory contains abundant sessile palms (*Astrocaryum* spp. and *Attalea* spp.; Ribeiro et al. 1999). The climate is characterized by a rainy season from November to May and a dry season from June to October (Marques-Filho et al. 1981). Mean annual temperature is approximately 26°C (Marques Filho et al. 1981) and mean annual rainfall was 2489 mm between 1985 and 2004 (unpublished data from the Meteorological station of Reserva Ducke).

Methods

Observations of reproductive events were made sporadically in temporary ponds in headwater streams distributed over an area of 64 km² (Lima et al. 2006). Fifteen dead females were collected (10 females in June 2001 and five females in June 2005) in two different ponds during explosive reproductive events in which dozens of individuals had gathered for reproduction. All dead individuals collected were fixed in formalin and preserved in 70% alcohol. The females were dissected to check for the presence of mature ovarian oocytes. Observations of behaviour of amplexant pairs were made simultaneously with collection of dead individuals. Oocytes extracted and fertilized by males from dead females were maintained in transparent plastic basins until embryos and the developmental stage were defined following Gosner (1960). As the eggs of *R. proboscidea* are relatively large (c.1.5 mm) we could observe their development in the field without removing the eggs from plastic bags, using a (10 ×) magnifying glass. In all cases we observed at least half the eggs in a clutch. The eggs were observed until they reached stage 13 by which stage fertilization is certain (Gosner 1960); we then fixed the eggs in 10% formalin. The embryos were fixed in the field because the team did not have the facilities needed to allow continued development in the laboratory. The stage of development was confirmed later in the laboratory under a stereomicroscope. Voucher specimens were deposited in the Coleção Zoológica Paulo Bührnheim of the Universidade Federal do Amazonas (CZPB-UFAM 279, lot 141) in Manaus, Amazonas, Brazil.

Results and discussion

Twenty dead *R. proboscidea* females were found in the first aggregation, which involved about 100 males. Only 10 dead females were collected and dissected. The other 10 showed some decomposition and were not collected. During the second aggregation, involving about 50 males, we found five dead females. Dissection revealed that none of the 15 females contained abdominal oocytes. In the first aggregation (Figure 1B) we witnessed a male in axillary amplexus with one dead female (Figure 1C), and he extracted her oocytes by squeezing the sides of her belly with rhythmic movements of his front and hind limbs (Figure 1C). We did not check the fertilization of eggs because of lack of materials suitable for observations in the field. During the second reproductive event, four *R. proboscidea* males were seen extracting oocytes from dead females. Three of them were not interrupted by other males but, on one occasion, a male was observed pushing the amplexant dead female around the pond, apparently to avoid other males. This male continued to compress the dead female's abdomen during such displacements and the oocytes kept being expelled. All eggs were collected and maintained until they reached embryo stage 13 of Gosner (1960), so confirming fertilization. Fertilization in *R. proboscidea* is external and clutches of this species are bead-like gelatinous strings, with eggs disposed uniserially (Menin et al. 2006). This feature may make it easier for males to expel oocytes from dead females when compared with other groups such as hylids because of the provision of the eggs in the abdominal cavity.

Some females were observed leaving the pond after the second reproductive aggregation, indicating that some females survived the reproductive event. However, it is unknown if these females can survive and reproduce in the next breeding season. Necrophilic behaviour by males can, at least, minimize the female's loss of investment

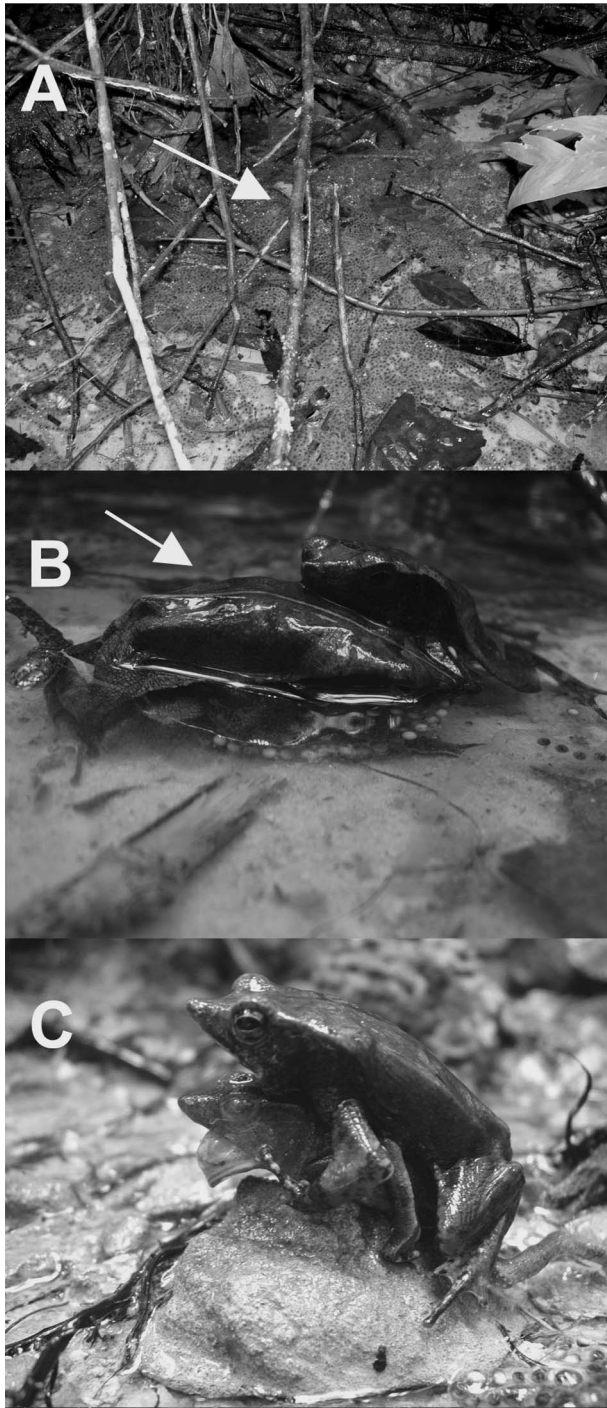


Figure 1. Necrophilia in *Rhinella proboscidea* in a central Amazonian headwater stream. Thousands of eggs (arrow) from a single reproductive event, in a small patch of a headwater stream (A). Two males in a battle for a drowned female. The larger (arrowed) is in amplexus and compressing the female's abdomen with his legs, which resulted in expulsion of the oocytes (B). Male compressing the abdomen of a dead female, which resulted in expulsion of her oocytes (C).

in oocytes if she dies during a mating aggregation. Once operational sex ratio is strongly biased to males (about ten males to one female; M. Menin, unpublished data), and the males spend a lot of energy in these battles (Beebee 1996), the probability of a given male finding and mating with another female, once the first female is dead, is probably small. It is therefore advantageous for the male to take advantage of oocytes from dead females. It brings to light the hypothesis that this low probability imposes a pressure on males to successfully mate with the first female found. Selection should favour behaviour in mating males that does not culminate in the death of females, unless some strategy is possible that minimizes the loss to the mating male's fitness when the female's death occurs. We suggest that the "necrophilia strategy" could solve the question of why there should be selection for stronger and persistent males (Duellman and Trueb 1994) if this reduces the number of live females available to a persistent male, and why there should be selection for morphology such as male size, keratinized spines on thumb and chest or development of arms as in some bufonid species, that facilitates dominance over the female's body, whether she is dead or not (Greene and Funk 2009), independent of selection for female traits. In the case of necrophilia, where there can be no future investment by the female, partial oviposition of a clutch is obviously of no selective advantage. As expulsion of oocytes from dead females probably can have fitness advantages for both partners, the behaviour may be more prevalent in anurans, or even other groups that rely on external fertilization, than present records indicate. However, studies of reproductive behaviour of species that are explosive breeders should be conducted to confirm this hypothesis.

Necrophilia has been reported in several vertebrate groups (mammals, birds, amphibians and reptiles; Costa et al. 2010). Although necrophilia has been reported in other species of anurans (Meshaka-Jr 1996; Bettaso et al. 2008; Sinovas 2009), this may be the first case where the necrophilia brings a direct fitness gain, generating descendants. In contrast to the conclusions of other studies (Sinovas 2009; Costa et al. 2010), necrophilia is not a behavioural mistake in *R. proboscidea*, but rather is a functional behaviour in terms of fitness, with positive effects on the reproductive success of both males and females.

Acknowledgements

We thank R. Macias-Ordóñez, G. Machado, M. Anciães, Cythia Prado, William Eberhard, T. Mott and an anonymous reviewer for comments on the draft. All authors received fellowships from Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq). The study benefited from the RAPELD infrastructure installed by the Brazilian PELD and PPBio programmes financed by CNPq and the Ministry of Science, Technology and Innovation (MCTI), respectively.

References

- Aichinger M. 1987. Annual activity patterns of anurans in a seasonal neotropical environment. *Oecologia* 71:583–592.
- Beebee TJC. 1996. Ecology and conservation of amphibians. London: Chapman and Hall.
- Bettaso J, Haggarty A, Russel E. 2008. *Rana boylii* (Foothill Yellow-legged Frog). Necrogamy. *Herpetol Rev.* 39:462–462.

- Costa HC, Silva ET, Campos PS, Oliveira MPC, Nunes AV, Santos PS. 2010. The corpse bride: a case of Davian behaviour in the green ameiva (*Ameiva ameiva*) in southeastern Brazil. *Herpetol Notes*. 3:79–83.
- Duellman WE, Trueb L. 1994. *Biology of amphibians*. Baltimore: The Johns Hopkins University Press.
- Gosner KL. 1960. A simplified table, for staging Anura embryos and larvae with notes on identification. *Herpetologica* 2:183–190.
- Greene AE, Funk WC. 2009. Sexual selection on morphology in an explosive breeding amphibian, the Columbia Spotted Frog (*Rana luteiventris*). *J Herpetol*. 43:244–251.
- Hager R, Johnstone RA. 2004. Infanticide and control of reproduction in cooperative and communal breeders. *Anim Behav*. 67:941–949.
- Harvey PH, Pagel MD. 1991. *The comparative method in evolutionary biology*. New York: Oxford University Press.
- Lima AP, Magnusson WE, Menin M, Erdtmann LK, Rodrigues DJ, Keller C, Hödl W. 2006. Guia de sapos da Reserva Adolpho Ducke, Amazônia Central: Guide to the frogs to Reserva Adolpho Ducke, Central Amazonia. Manaus: Atemma, Brazil.
- Marques-Filho AO, Ribeiro MNG, Santos HM, Santos JM. 1981. Estudos climatológicos da Reserva Florestal Ducke – Manaus – AM. IV. Precipitação. *Acta Amazôn*. 11:759–768.
- Menge BA, Olson AM. 1990. Role of scale and environmental factors in regulating of community structure. *Trends Ecol Evol*. 5:52–57.
- Menin M, Rodrigues DJ, Lima AP. 2006. The tadpole of *Rhinella proboscidea* (Anura: Bufonidae) with notes on adult reproductive behavior. *Zootaxa* 1258:47–56.
- Meshaka WE. 1996. Anuran Davian behavior: a Darwinian dilemma. *Fla Sci*. 59:74–75.
- Odendaal FJ, Iwasa Y, Ehrlich PR. 1985. Duration of female availability and its effect on butterfly mating systems. *Am Nat* 125:673–678.
- Prado CPA, Uetanabaro M, Haddad CFB. 2005. Breeding activity patterns, reproductive modes, and habitat use by anurans (Amphibia) in a seasonal environment in the Pantanal, Brazil. *Amphibia-Reptilia* 26:211–221.
- Ribeiro JELS, Hopkins MJG, Vicentini A, Sothers CA, Costa MAS, Brito JM, Souza MAD, Martins LHP, Lohmann LG, Assunção PACL, et al. 1999. Flora da Reserva Ducke: guia de identificação das plantas vasculares de uma floresta de terra firme na Amazônia Central. Brazil: Editora do Instituto Nacional de Pesquisas da Amazônia. Manaus: Brasil.
- Rodrigues DJ, Lima AP, Magnusson WE, Costa FRC. 2010. Temporary pond availability and tadpole species composition in Central Amazonia. *Herpetologica* 66:124–130.
- Rodrigues DJ, Uetanabaro M, Lopes FS. 2004. Reproductive strategies of *Physalaemus nattereri* (Steindachner, 1863) and *P. albonotatus* (Steindachner, 1864) at Serra da Bodoquena, State of Mato Grosso do Sul, Brazil. *Rev Espan Herpetol*. 18:63–73.
- Rodrigues DJ, Uetanabaro M, Lopes FS. 2005. Reproductive patterns of *Trachicephalus venulosus* (Laurenti 1768) and *Scinax fuscovarius* (Lutz 1925) from the Cerrado, Central Brazil. *J Nat Hist*. 39:3217–3226.
- Rodrigues DJ, Uetanabaro M, Lopes FS. 2007. Breeding biology of *Phyllomedusa azurea* Cope 1862 and *P. sauvagii* Boulenger 1882 (Anura) from the Cerrado, Central Brazil. *J Nat Hist*. 41:1841–1851.
- Sakai, A. K., and D.F. Westneat. 2001. Mating Systems. In: Fox CW, Roff DA, Fairbairn DJ, editors. *Evolutionary ecology: concepts and case studies*. New York: Oxford University Press, p. 193–206.
- Sinovas P. 2009. *Bombina variegata* (Yellow Fire-bellied Toad). Mating behavior. *Herpetol Rev*. 40:199.
- Stearns SC. 1992. *The evolution of life histories*. New York: Oxford University Press.
- Trauth SE, McCallum ML, Cartwright ME. 2000. Breeding mortality in the Wood Frog, *Rana sylvatica* (Anura: Ranidae), from Northcentral Arkansas. *J Arkansas Acad Sci* 54:154–156.

- Verrell PA, McCabe N. 1986. Mating balls in the common toad, *Bufo bufo*. Br Herpetol Soc Bull 26:28–29.
- Vieites DR, Nieto-Román S, Barluenga M, Palanca A, Vences M, Meyer A. 2004. Post-mating clutch piracy in an amphibian. Nature 431:305–308.
- Wells KD. 1977. The social behaviour of anuran amphibians. Anim Behav. 25:666–693.
- Wells KD. 1979. Reproductive behavior and male mating success in a Neotropical toad, *Bufo typhonius*. Biotropica 11:301–307.
- Wells KD. 2007. The ecology and behavior of amphibians. Chicago: The University of Chicago Press.
- Zimmerman BL, Bogart JP. 1988. Ecology and calls of four species of Amazonian forest frogs. J Herpetol. 22:97–108.

Copyright of Journal of Natural History is the property of Taylor & Francis Ltd and its content may not be copied or emailed to multiple sites or posted to a listserv without the copyright holder's express written permission. However, users may print, download, or email articles for individual use.