

REPRODUCTIVE BIOLOGY OF *ATRACTUS RETICULATUS* (BOULENGER, 1885) (SERPENTES, COLUBRIDAE) IN SOUTHERN BRAZIL

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The reproductive biology of *Atractus reticulatus* was studied based on the analyses of 135 specimens from preserved in collections, and on field observations in Rio Grande do Sul State, southern Brazil. The snout-vent length (SVL) of mature females was significantly larger than the SVL of mature males, and the tail length (TL) of mature males was significantly longer than the TL of mature females. Analysis of neonate body size, hatching time and seasonal distribution of body size classes (SVL) suggests that males reach sexual maturity at eight to 10 months old (SVL c. 198 mm) and females at 11 to 12 months old (SVL c. 242 mm). The reproductive period begins in late August, with clutches laid in November, December and January, and hatching occurring from January to March. Fecundity is low: the number of vitellogenic follicles varied from 2 to 6, and the number of eggs from 1 to 3. Relative to female size, the eggs are large and the clutches are heavy, corresponding to approximately 27% of female body mass.

Key words: colubrid, fecundity, sexual dimorphism, snakes, reproduction

INTRODUCTION

The genus *Atractus* Wagler, 1828 comprises about 80 small fossorial species of snakes that occur in South America, from eastern Panama to western Argentina (Fernandes & Puerto, 1993; Fernandes, 1995; Giraudo, 2001). The reproductive biology of most species is poorly known, and available data are mainly from very small samples (e.g. Martins & Oliveira, 1993, 1998; Fernandes & Puerto, 1993; Murphy, 1997; Starace, 1998; Cassimiro *et al.*, 2002). *Atractus reticulatus* (Boulenger, 1885) corresponds to the southern distribution of the genus, occurring in southern Paraguay, north-eastern Argentina and south-eastern and southern Brazil (Fernandes, 1995; Giraudo & Scrocchi, 2000). This species inhabits forests, savannas, transitional environments and semi-urban areas (Giraudo & Scrocchi, 2000). The reproductive biology of *A. reticulatus* has been almost completely ignored. There is only anecdotal information on the incubation time of two clutches from south-eastern Brazil, and biometry of neonates from one of them (Fernandes & Puerto, 1993).

Herein we present data on sexual dimorphism in body size, sexual maturity, reproductive cycle, fecundity and recruitment of *Atractus reticulatus* in southern Brazil.

MATERIAL AND METHODS

STUDY AREA

All specimens examined were from eastern Depressão Central of Rio Grande do Sul State, southern Brazil. This area encompasses a zone of lowlands (10-300 m above sea level) with dark red podzol soil, giving rise to poorly drained land with high humidity. The relief is homogene-

ous, forming hillocks. Some disjunct hills are present and the presence of man is high in some areas. The climate is mesothermic temperate (IBGE, 1990).

MATERIAL EXAMINED

This study was based on the dissection of 135 specimens preserved in the collections of Museu de Ciências e Tecnologia of Pontifícia Universidade Católica do Rio Grande do Sul (MCP) and Museu de Ciências Naturais of Fundação Zoobotânica do Rio Grande do Sul (MCN) (see Appendix 1). Of these, three specimens were maintained alive and laid eggs in captivity, providing additional data on clutch size and time of oviposition.

PROCEDURES

The data recorded for each specimen were: snout-vent-length (SVL, mm), tail length (TL, mm), sex (analysis of gonads or hemipenis muscles), number of vitellogenic follicles or eggs, size of the largest vitellogenic follicle or egg (in mm), and condition of efferent ducts.

Females with follicles at least 6 mm in length or with eggs in the oviducts were considered mature (modified from Shine, 1977). Males with convoluted and opaque efferent ducts were considered mature (Shine, 1988). The age and size at maturation were estimated based on the seasonal size classes of males and females over time (as in Shine, 1988). This estimative is possible when reproduction (mainly hatchlings) is markedly seasonal. The size and sex of the neonates, and the time of hatchlings were recorded from eggs incubated artificially (see below). The time of maturation was considered to be the time interval between the hatchlings and the first appearance of mature males and females in the sample. Gravid females were kept in captivity until oviposition. Eggs were measured (largest

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length), weighed and incubated in vermiculite at about 25 °C. The relative clutch mass (RCM) was calculated by dividing the total clutch mass by the sum of female mass and total clutch mass. Only hatchlings hatched in captivity were considered as neonates. The degree of sexual size dimorphism (SSD) was the difference of the mean size (SVL) of the largest sex divided by the mean size of the smallest sex to 1.0 (Shine, 1993). Statistics were performed using SPSS for Windows v. 10. We employed a two-tailed Student's *t*-test to evaluate the statistical significance of sexual dimorphism.

RESULTS

BODY SIZE AND SEXUAL DIMORPHISM

Neonate SVL ranged from 77 mm to 117 mm (mean±SD = 107.7±12.9; *n*=10) and TL from 5 mm to 17 mm (mean±SD = 13.5±3.5; *n* = 10). Mature female SVL ranged from 242 mm to 370 mm (mean±SD = 300.1±30.0; *n*=43) and TL from 26 mm to 44 mm (mean±SD = 35.0±4.54; *n*=43). Mature male SVL ranged from 198 mm to 295 mm (mean±SD = 249.9±24.4; *n*=58) and TL from 26 mm to 51 mm (mean±SD = 38.2±5.8 *n*=58). The SVL of mature females was significantly larger than the SVL of mature males (*t*=1.98, *df*=99, *P*<0.001), and the TL of mature males was significantly larger than the TL of mature females (*t*=1.98, *df*=99, *P*<0.001). The degree of sexual size dimorphism was calculated as 0.20.

SEXUAL MATURITY

The combined analysis of neonate body size, birth time and seasonal distribution of body size classes (SVL) suggests that males reach sexual maturity at eight to 10 months old (SVL of nearly 198 mm, Fig. 1) and females at 11 to 12 months old (SVL of nearly 242 mm, Fig.2).

REPRODUCTIVE CYCLE

Vitellogenesis began in late August and lasted until late December (Fig.3). Females were collected together with mature males in November (*n*=2). Clutches were observed in November, December and January. Hatchlings were recorded from January to March (from three clutches). One female had vitellogenic follicles while carrying eggs in the oviducts. Another female was dissected immediately after laying eggs and also had vitellogenic follicles.

CLUTCH SIZE AND BIOMETRY OF EGGS AND NEWBORNS

Fecundity in *Atractus reticulatus* was low. The number of vitellogenic follicles varied from 2 to 6 (mean±SD = 3.9±1.5, *n*=9), and the number of eggs varied from 1 to 3 (mean±SD = 2.7± 0.8, *n*=6). Within the six females that contained eggs, five females (measuring from 242 mm to 333 mm SVL) had three eggs, and one female (measuring 286 mm SVL) had one egg. Relative

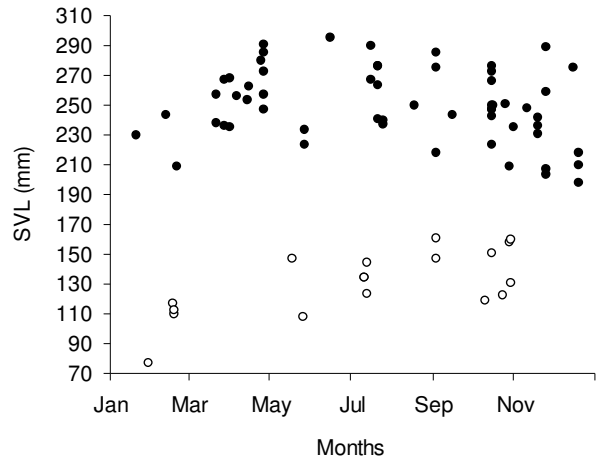


FIG. 1. Seasonal distribution of body size in males of *Atractus reticulatus* from southern Brazil. Open circles, immature males; closed circles, mature males.

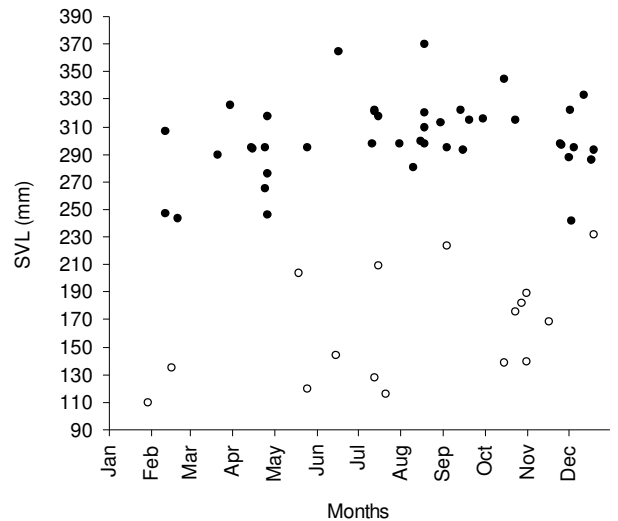


FIG. 2. Seasonal distribution of body size in females of *Atractus reticulatus* from southern Brazil. Open circles, immature females; closed circles, mature females.

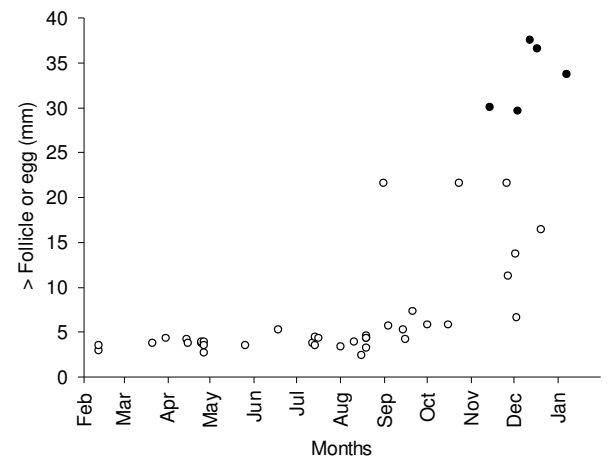


FIG. 3. Seasonal variation in the diameter of largest follicles (open circles) and eggs (closed circles) of mature females of *Atractus reticulatus* from southern Brazil.

to female size, the eggs are large (mean length \pm SD = 34.6 \pm 3.92 mm, mean mass \pm SD = 1.7 \pm 0.25 g, $n=10$), and the clutches are heavy, corresponding to approximately 27% of the female weight (calculated from means, $n=4$). After 66 days of artificial incubation, one clutch with three eggs produced three neonates measuring mean SVL \pm SD of 116.3 \pm 0.58 mm and mean \pm SD mass of 1.31 \pm 0.32 g.

DISCUSSION

BODY SIZE AND SEXUAL DIMORPHISM

The SVL of neonates of *Atractus reticulatus* is relatively large when compared to the SVL of conspecific adults, and also large when compared to the SVL of neonates of other Neotropical colubrids (Fernandes & Puerto, 1993). In snakes, female size and the number of eggs are usually correlated and this affects neonate size (Seigel & Ford, 1987). The small size of *Atractus reticulatus* and the low fecundity seem to be adaptations to the underground environment, as observed in other morphostructural characteristics of fossorial snakes (Cadle & Greene, 1993; Webb *et al.* 2000a, 2001). Lemen & Voris (1981) suggest that some small species of marine snakes (*Hydrophis torquatus*) may produce large offspring while reducing clutch size. Availability of food is higher to large neonates than to small neonates since the former can swallow larger prey. Madsen & Shine (1998, 2000) recorded mortality rates higher than usual among *Liasis fuscus* neonates that were born late and, hence, small in relation to their prey size.

Larger SVL in females corresponds to the most common pattern in snakes. It is recorded, as a rule, among species in which male-male combat does not occur (Shine, 1978, 1984). There is no record of male-male combat in *A. reticulatus* or in any other species of the genus, thus the pattern found here was as expected. Natural selection may also favour larger body size in females that incur high reproductive costs (Bonnet *et al.*, 1998). *Atractus reticulatus* seems to belong to this category since its clutches are small but comprise large and relatively heavy eggs when compared to female body size.

The sexual dimorphism in TL, with larger values in mature males than mature females, also corresponds to the most common pattern in snakes (King, 1989; Shine, 1993). Giraudo & Scrocchi (2000) analysed specimens of *Atractus reticulatus* combining all available sizes and ages and recorded sexual dimorphism in the number of ventrals (greater in males), subcaudals (greater in females), and in the ratio tail length / total body length (greater in males). Our data demonstrate sexual dimorphism in SVL (greater in females) and TL (greater in males) in mature specimens.

SEXUAL MATURITY

In snakes the energetic costs incurred while reaching sexual maturity seem to be smaller for males than for females with high reproductive costs (Shine, 1978). The

delayed sexual maturity in females of *Atractus reticulatus*, in relation to conspecific males, may have advantages by allowing females to reach a larger body size at time of reproduction, and to produce larger eggs and hatchlings. Due to their faster maturation, mature males are available in the population when the first females reach maturity, which may improve the reproductive success of the species. However, only neonates of *Atractus reticulatus* emerging at the beginning of the hatchling period (January) appear able to reach a similar SVL to mature individuals in the reproductive season subsequent to their birth. Late neonates (March) may remain immature until the second reproductive season, as recorded by Oliveira (2001) for the colubrid *Lystrophis dorbignyi* on the North Coast of Rio Grande do Sul - an area geographically close to this study.

REPRODUCTIVE CYCLE

In the study area, the reproduction of *Atractus reticulatus* is seasonal, with recruitment during the hotter months of the year. In south-eastern Brazil, Fernandes & Puerto (1993) recorded two clutches in January, which corresponds to the end of the oviposition period of this species in southern Brazil. In *Atractus pantosticus*, a typically subtropical species with a more northern distribution than *A. reticulatus*, the reproductive period is longer, with records of clutches from January to March (Fernandes & Puerto, 1993; Cassimiro *et al.*, 2002). Martins & Oliveira (1993, 1998) investigated the biology of eight species of *Atractus* (*A. alphonsehoguei*, *A. latifrons*, *A. major*, *A. poeppigi*, *A. schach*, *A. snethlageae*, *A. torquatus* and *A. trilineatus*) at low latitudes (Manaus city and adjacent regions), where the climate is typically tropical, and suggested aseasonal reproduction for two of them (*A. latifrons* and *A. torquatus*). The temperate climate in the southern region of Brazil seems to impose some physiological constraints that limit snake activity to the hotter months of the year, and this influences the natural history of the species. Thus, much of what is known for Brazilian snakes of tropical regions may not be useful as parameters for the same species (or closely related species) in southern Brazil (Di-Bernardo, 1998).

CLUTCH SIZE AND BIOMETRY OF EGGS AND NEONATES

The low fecundity of *Atractus reticulatus* may be related to the small size of adults (Shine, 1983, 1984), since the larger congeneric species, *A. major* and *A. torquatus*, produce larger clutches (Martins & Oliveira 1993, 1998). The fossorial habit of *Atractus reticulatus*, may also influence the number and shape of the eggs of each clutch. Clutches of *A. reticulatus* consist of long eggs, which probably limit the presence of additional eggs in the oviducts. The increase in width and decrease in length of eggs could allow accommodation of additional eggs with constant mass (Ford & Seigel, 1989).

However, wide eggs possibly restrain the locomotion of fossorial snakes inside galleries under the ground. Clutches with similar parameters to *Atractus reticulatus* (small and containing long eggs) seem to be common to other species of this genus (Martins & Oliveira 1993, 1998; Fernandes & Puerto, 1993; Murphy, 1997; Starace, 1998; Cassimiro *et al.*, 2002), and to reptiles of other genera with fossorial or semifossorial habits, even from phylogenetically distinct lineages such as *Tantilla* (Aldridge & Semlitsch, 1982; Marques & Puerto, 1998), and *Amphisbaenia* (*Monopeltis anchietae* and *Zygaspis quadrifrons*, Webb *et al.*, 2000b - but see Webb *et al.*, 2001 with regards to typhlopids from southern Africa). On the other hand, some non-fossorial colubrid snakes from southern Brazil, with SVL similar to that of *A. reticulatus*, may produce up to 13 eggs (e.g. *Liophis jaegeri*, Pontes & Di-Bernardo, 1988).

Fernandes & Puerto (1993) recorded mean length of neonate *Atractus reticulatus* similar to our records, but recorded hatchlings during a much longer period.

Although the fossorial lifestyle of *Atractus reticulatus* makes the study of live individuals challenging, we show that some interesting reproductive parameters can be accessed by dissecting preserved specimens. We propose that the fossorial lifestyle of *Atractus* drives its reproductive parameters, for instance, reducing the number of eggs and increasing their lengths, thus providing more mobility to the snake in the subterranean environment. However, additional research must be made to test the hypotheses proposed here. *A. reticulatus* is a common snake that can be found even in places of anthropogenic disturbance (e.g. the suburbs of large cities), and we suggest that it would make a good model to test such hypotheses.

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APPENDIX 1

SPECIMENS EXAMINED

Brazil. Rio Grande do Sul: Esteio: MCN 9438; Gravataí: MCN 2954, 3000; Porto Alegre: MCN 1015, MCP 1907, 4842, 5186, MCN 8083, 8084, 8570, MCP 9069, 9087, 9088, MCP 9318, 9388, 10129, 10130, 10131, 10209, 10210, 10211, 10212, 10509, 10510, 10515, 10965, 11258, 11259, 11356, 11901, 11903, 13178, 13179, 13180, 13281, 13299, 13300, 13363, 13364, 13365, 13382, 13383, 13384, 13385, 13386, 13407, 13408, 13409, 13410, 13463, 13464, 13465, 13466, 13527, 13528, 13529, 13530, 13531, 13532, 13533, 13534, 13716, 13792, 13793, 13794, 13795, 13803, 13804, 13805, 13977, 13978, 13979, 13980, 13981, 14121, 14179, 14234, 14235, 14236, 14237, 14238, 14280, 14295, 14303, 14304, 14305, 14306, 14307, 14393, 14398, 14399, 14402, 14403, 14433, 14434, 14435, 14436, 14437, 14438, 14439, 14440, 14441, 14471, 14472, 14473, 14474, 14570, 14596, 14597, 14598, 14599, 14600, 14601, 14639, 14640, 14641, 14642, 14643, 14644, 14645, 14646, 14647, 14675, 14676, 14697, 14698, 14699, 14700, 14701, 14702; Santo Antônio da Patrulha: MCP 4; São Leopoldo: MCN 7450; Taquara: MCP 11727; Viamão: MCP 8983.