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REPRODUCTION IN THE BLACKNECK GARTER SNAKE, *THAMNOPHIS CYRTOPSIS* (SERPENTES: COLUBRIDAE)

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Abstract.—Reproductive tissue was examined from 120 museum specimens of *Thamnophis cyrtopsis* from Arizona, New Mexico, Texas and the Mexican state of Sonora. This species of garter snake appears to follow a seasonal reproductive cycle in which maximum spermiogenesis occurs in summer-autumn. Females with enlarged follicles or developing embryos were found March-June. The mean clutch size for 25 females was 8.4 ± 4.3 SD (range 3-24). The maximum clutch size of 24 is apparently a new record for this species. Neonates were found June to August.

The blackneck garter snake *Thamnophis cyrtopsis* occurs from southeast Utah to Guatemala and from central Texas to central and southern Arizona from sea level to around 2700 m (Stebbins 1985). The biology of this species is summarized by Webb (1980). There are only anecdotal reports on reproduction in this species (Sabath & Worthington 1959; Vitt 1975; Tennant 1984; Stebbins 1985; Jones 1990; Degenhardt et al. 1996). The purpose of this paper is to provide information on the seasonal ovarian and testis cycles of *T. cyrtopsis* and to report additional clutch sizes.

MATERIALS AND METHODS

A sample of 120 specimens of *Thamnophis cyrtopsis* (64 females) Mean Snout-Vent Length, SVL = 508.6 mm ± 74.1 SD, range 378-705 mm; (56 males) Mean SVL = 441.2 mm ± 64.3 SD, range 338-677 mm from Arizona, New Mexico, Texas and Sonora, México was examined from the herpetology collections of Arizona State University, Tempe and the University of Arizona, Tucson. Counts were made of enlarged follicles (> 6 mm diameter), oviductal eggs or embryos. The left testis, epididymis, vas deferens and part of the kidney were removed from males; the left ovary was removed from females for histological examination. Tissues were embedded in paraffin and cut into sections at 5 μm. Slides were stained with Harris' hematoxylin followed by eosin counterstain. Testes slides were examined to determine the stage of the male cycle; ovary slides were examined for the presence of yolk deposition. Epididymides and vasa deferentia were examined for sperm. Slides of kidney sexual segments were examined for secretory activity.

Kidneys and vasa deferentia were not available for examination from some road-killed males.

Material examined.—The following specimens of *Thamnophis cyrtopsis* were examined from the herpetology collections of Arizona State University, Tempe (ASU) and the University of Arizona, Tucson (UAZ).

ARIZONA: APACHE COUNTY, 2 specimens (ASU 903-904); COCHISE COUNTY, 7 specimens (ASU 24232, UAZ 34524, 39933, 41604, 42342, 42478, 43802); GILA COUNTY, 6 specimens (ASU 2245, 2340, 2365, UAZ 30947, 35976, 44766); GRAHAM COUNTY, 11 specimens (ASU 7012, 7015, 7018, 7772, 7776-7778, 7784, 13293, 22465, UAZ 45889); GREENLEE COUNTY, 3 specimens (UAZ 26499, 42711-42712); MARICOPA COUNTY, 8 specimens (ASU 1364, 2208, 8844, 9163, 13889-13890, UAZ 37037, 43170); MOHAVE COUNTY, 1 specimen (ASU 27845); NAVAJO COUNTY, 2 specimens (ASU 3160, UAZ 35975); PIMA COUNTY, 35 specimens (UAZ 26483, 26487, 26490-26493, 26496, 26498, 26500, 26504, 26506-26508, 26512-26513, 26523-26524, 26528, 26531, 26533, 26542, 26548, 26567, 26569, 27274, 32925, 41597-41598, 41600, 41603, 41605, 41607, 42713, 44976, 47141); PINAL COUNTY, 3 specimens (ASU 906, 2388, 27391); SANTA CRUZ COUNTY, 13 specimens (UAZ 26511, 26517, 26526, 26560, 26565, 28602, 35534, 36072, 40501, 41590, 41594, 47324, 49999); YAVAPAI COUNTY, 2 specimens (ASU 950, UAZ 39927). NEW MEXICO: GRANT COUNTY, 1 specimen (UAZ 26509); HIDALGO COUNTY, 2 specimens (UAZ 32771-32772) TEXAS: JEFF DAVIS COUNTY, 1 specimen (UAZ 41819); VAL VERDE COUNTY, 1 specimen (UAZ 42256).

MÉXICO: SONORA, 22 specimens (UAZ 26687, 26690, 26692, 28066, 28138, 32626, 38944-38947, 39284, 39297, 42359, 45133, 45135, 45137, 45139-45140, 45994, 46459, 46461, 46671).

RESULTS AND DISCUSSION

Testicular histology was similar to that reported by Goldberg & Parker (1975) for two colubrid snakes, *Masticophis taeniatus* and *Pituophis catenifer* (= *P. melanoleucus*). In the regressed testes, seminiferous tubules contained spermatogonia and Sertoli cells. In recrudescence, there was renewal of spermatogenic cells characterized by spermatogonial divisions; primary and secondary spermatocytes and spermatids may have been present. In spermiogenesis, metamorphosing spermatids and mature sperm were present. Males undergoing spermio-

Table 1. Monthly distribution of conditions in seasonal testicular cycle of *Thamnophis cyrtopsis*. Values shown are the numbers of males exhibiting each of the three conditions.

Month	N	Regressed	Recrudescence	Spermiogenesis
March	2	2	0	0
April	6	5	0	1
May	10	3	3	4
June	9	1	3	5
July	12	1	2	9
August	5	0	1	4
September	8	0	2	6
October	3	0	0	3
November	1	0	0	1

genesis were found April-November; regressed testes were found March-July (Table 1). Males with testes in recrudescence were found May-September. The smallest spermiogenic male measured 338 mm SVL. To avoid bias from including immature males in my analysis, males smaller than this size were excluded from the study. At least part of each monthly male sample was undergoing spermiogenesis April-November. Epididymides of all spermiogenic males contained sperm. Sperm were present in the vasa deferentia during all months (March-November) indicating *T. cyrtopsis* has the potential of breeding throughout the year. However, because 11/27 (41%) males had regressed testes in spring and 10/12 (83%) of September-November males were undergoing spermiogenesis (Table 1), the male cycle appeared to fit the postnuptial type of breeding pattern (Saint Girons 1982; Seigel & Ford 1987) with maximum spermatogenic activity in late summer and fall and mating occurring the following spring utilizing sperm stored overwinter in the vasa deferentia. One must also consider the possibility of some *T. cyrtopsis* mating occurring during autumn. Fall matings have been reported in three species of *Thamnophis* (*T. atratus*, *T. ordinoides*, *T. sirtalis*) and suggested to occur in two additional species (*T. radix* and *T. elegans*) (see Rossman et al. 1996). Kidney sexual segments were enlarged and contained secretory granules in 23/26 (88%) of males undergoing spermiogenesis, 5/6 (83%) males with regressed testes and 2/4 (50%) of males with recrudescent testes. Mating coincides with hypertrophy of the kidney sexual segment (Saint Girons 1982).

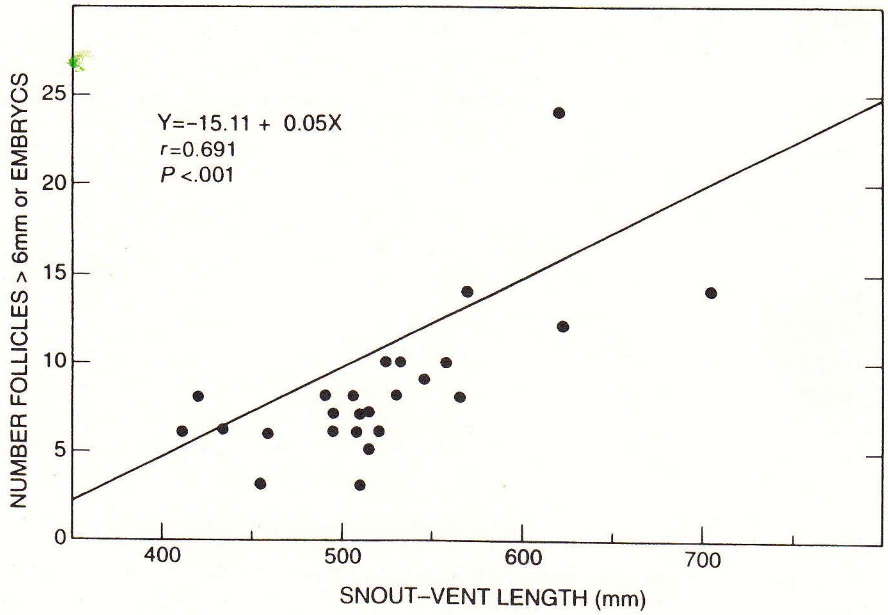

Figure 1. Linear regression of enlarged follicles (> 6 mm) or developing embryos on snout-vent length for 25 *Thamnophis cyrtopsis* from Arizona and Sonora, México, including two Arizona specimens from Vitt (1975).

The smallest reproductively active female (follicles > 6 mm diameter) measured 412 mm SVL. All females smaller than 412 mm SVL were excluded from the study to avoid bias from including immature females in my analysis of the ovarian cycle. Mean clutch sizes for 25 females, including clutches of 6 and 10 from Vitt (1975), averaged 8.4 ± 4.3 SD (3-24 range). There was a significant positive correlation between female body size and enlarged follicles > 6 mm, or embryos (P < .001) (Fig. 1). Other litter sizes from the literature were from Arizona and Texas: (Arizona) eight young were born from one female on 3 July (Vitt 1975) and four females gave birth, 29 June (14 young), 11 July (19 young), 17 July (21 young), 19 July (22 young) (Jones 1990); (Texas) seven young were born from one female on 14 August (Sabath & Worthington 1959) and six young were born from one female on 14 July (Tennant 1984). Young of *T. cyrtopsis* were born the second week of July in New Mexico (Fleharty 1967). Stebbins (1985) reported a range of about 7-25 young. Apparently, the litter of 25 young attributed in the literature to *T. cyrtopsis* was actually from *T. eques* (see Degenhardt et al. 1996). Thus, the clutch of 24 reported herein may represent the largest clutch known for *T. cyrtopsis*.

Table 2. Monthly distribution of conditions in seasonal ovarian cycle of Thamnophis cyrtopsis. Values shown are the numbers of females exhibiting each of the four conditions.

Month	N	Inactive	Yolk deposition	Enlarged follicles (> 6 mm diameter)	Developing embryos
March	5	1	1	2	1
April	17	4	0	12	1
May	6	1	0	0	5
June	10	7	1	0	2
July	12	12	0	0	0
August	3	3	0	0	0
September	8	8	0	0	0
October	1	1	0	0	0
November	2	2	0	0	0

The female reproductive cycle appears to follow that of other species of *Thamnophis* (see Rossman et al. 1996) in having prenuptial or Type 1 secondary vitellogenesis (*sensu* Aldridge 1979) with females entering hibernation with small, previtellogenic follicles. Secondary vitellogenesis (yolk deposition) occurs in spring. Females with enlarged follicles > 6 mm diameter or developing embryos were found March-June (Table 2). The presence of some inactive females during the period when gravid females were found (March-June) suggests that not all mature females produce litters each year. Data from Table 2 indicate 23/38 (61%) of females from March-June produced litters. It is not known if the one March and one June female *T. cyrtopsis* that were undergoing yolk deposition would have produced litters. This figure (61%) is within the range of gravid females (46-100%) for six species of *Thamnophis* in Seigel & Ford (1987). Data on reproductive frequency for other species of *Thamnophis* suggests most females reproduce each year, although some females may not breed every year (Rossman et al. 1996). This appears to be the case for females of *T. cyrtopsis*.

Neonates were found in June to August. The date of birth, size of neonates at birth and growth rates likely show geographic and yearly variation.

CONCLUSIONS

Thamnophis cyrtopsis appears to follow a seasonal reproductive cycle in which maximum spermiogenesis occurs in late summer-autumn.

Sperm is stored overwinter in the vasa deferentia. Mating presumably takes place in spring with the possibility of some occurring in autumn. Gravid females were found March-June. Mean clutch size was 8.4 ± 4.3 SD (range 3-24). The maximum clutch size of 24 may represent a new record for this species.

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