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# Notes on Reproduction of the Mexican Garter Snake, *Thamnophis eques* (Serpentes: Colubridae), from Mexico

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## ABSTRACT

Reproductive tissue was examined from museum specimens of *Thamnophis eques* from Mexico. This species of garter snake appears to follow a seasonal reproductive cycle in which spermiogenesis begins in summer and ends in autumn. *Thamnophis eques* starts enlargement of ovarian follicles in autumn whereas other species of garter snakes delay this until spring. *Thamnophis eques* females with oviducal eggs or developing embryos were found May-July. Mean litter size (n = 6) was 8.0 ± 2.3 SD, range: 6-12. Young appear June-August.

## INTRODUCTION

The Mexican garter snake, *Thamnophis eques* ranges from central Arizona and southwestern New Mexico south to Veracruz, Mexico (Stebbins, 2003). Information on reproduction of *T. eques* is available in a number of papers (Degenhardt et al., 1996; Ernst and Ernst, 2003; Macías García and Drummond, 1988; Ramirez-Bautista et al., 1995; Rosen and Schwalbe, 1988; Rossman et al., 1996; Manjarrez, 1998; Conant, 2003). The purpose of this paper is to present information on reproduction in *T. eques* from a histological examination of museum specimens collected in Mexico. The first information on the timing of the testicular cycle is given.

## MATERIALS AND METHODS

A sample of 55 specimens of *Thamnophis eques* (20 males) mean snout-vent length, SVL = 455.4 mm ± 62 SD, range: 350-590 mm; (21 females) mean SVL = 504.2 mm ± 70 SD, range: 403-666 mm; (14 juveniles) mean SVL = 166.5 mm ± 12 SD, range: 150-188 mm from Mexico was examined from the herpetology collections of the Natural History Museum of Los Angeles County, (LACM), Los Angeles, California and the University of Arizona, (UAZ), Tucson, Arizona (Appendix). The left testis and vas deferens 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 $\mu$m. Slides were stained with Harris' hematoxylin followed by eosin counterstain. Slides of testes were examined to determine the stage of the male cycle; slides of ovaries were examined for

the presence of yolk deposition (secondary yolk deposition *sensu* Aldridge, 1979). Enlarged ovarian follicles (> 6 mm length) or oviducal eggs were counted and not sectioned. An unpaired *t*-test was used to compare male and female mean body sizes (SVL's).

## RESULTS AND DISCUSSION

Testicular histology was similar to that reported by Goldberg and Parker (1975) for two species of colubrid snakes, *Masticophis taeniatus* and *Pituophis catenifer*. In the regressed testis, seminiferous tubules contained mainly spermatogonia and Sertoli cells. In testes undergoing recrudescence there was a proliferation of germ cells including spermatogonia, primary and secondary spermatocytes. Occasionally, small numbers of spermatids were present. In spermiogenesis, metamorphosing spermatids and mature sperm were present. Males undergoing spermiogenesis were found June to September (Table 1). Males with testes in recrudescence were found in May and June. Three males with regressed testes were found in September. The smallest mature male (UAZ 34755, spermiogenesis in progress) measured 360 mm SVL. Because males in recrudescence were found in May and June and spermiogenesis ends in September, spermatogenesis in *T. eques* appeared to fit the "aestival spermatogenesis" of Saint Girons (1982). The testicular cycle of *T. eques* appears similar to those of *Thamnophis cyrtopsis* and *Thamnophis rufipunctatus* (Goldberg, 1998, 2003) in which males undergoing spermiogenesis were also found during the summer months. Sperm was present in the vasa deferentia during May-September indicating *T. eques* has the potential to breed during this time period. Rosen and Schwalbe (1988) reported that courtship and mating have not been observed in *T. eques* from Arizona, however high male activity levels in September along with the onset of follicular enlargement indicate mating occurs in autumn. A pair of captive *T. eques* mated in fall and spring (Degenhardt et al.,1996).

Female *T. eques* were significantly larger than males ($t = 2.4$, $df = 39$, $P = 0.02$) as was reported by Degenhardt et al. (1996) in New Mexico. The smallest reproductively active female (UAZ 34179, oviducal eggs present) measured 460 mm SVL. This size is smaller than the 550-700 mm SVL at which female *T. eques* start reproduction in Arizona (Rosen and Schwalbe, 1988). Mean litter size for six females was 8.0 ± 2.3 SD, range: 6-12. This is within the range of 4-26 in Ernst and Ernst (2003) and 5-36 in Conant (2003). Mean litter size for twelve female *T. eques* from Toluca, Mexico was 10.8 ± 2.9 SD, range = 6-17; there was a significant positive correlation between female body size and number of offspring (Manjarrez, 1998). Females with oviducal eggs or developing embryos were found May-July (Table 2). Conant (2003) reported gravid *T. eques* from July and August. The one female undergoing early yolk deposition (secondary yolk deposition *sensu* Aldridge 1979) was found in May (Table 2). Females with enlarged follicles for next year's litters were found in September and December. This supports the statement of Rosen and Schwalbe (1988) that follicular enlargement begins in autumn for next year's litter. *Thamnophis eques* therefore differs from other species of *Thamnophis* in which females enter hibernation with small, previtellogenic ovarian follicles (Rossman et al. 1996). By starting follicular enlargement in autumn, young *T. eques* can presumably complete development earlier in the summer and have a longer period to feed and grow. The ovarian cycle of the congener *T. cyrtopsis* differs as enlarged ovarian follicles were not found in autumn (Goldberg, 1998).

Rosen and Schwalbe (1988) reported that in Arizona young are born from very early June to early July and two broods of neonates averaged 137 mm ± 4 SD and 194 mm ± 8 SD in SVL. Three newborn *T. eques* measured between 150-203 mm SVL (Degenhardt et al. 1996). Fourteen neonates collected 16 June-13 August from Mexico had a mean SVL of 167 mm ± 12.2 SD, range =150-188. The exact time of parturition is unknown although *Thamnophis eques* females from Toluca, Mexico state, Mexico gave birth gave in the laboratory birth between 30 April and 2 November (Manjarrez, 1998). Conant (2003) observed neonates during July 1959 in Jalisco; young were born in captivity July-November.

Rosen and Schwalbe (1988) reported only half of *T. eques* females from southern Arizona produce young each year. The presence of 5/7 (71%) non-reproductive *T. eques* females in June (Table 2) when other females are gravid suggests that not all females bear litters each year. Macías García and Drummond (1988) similarly found this to be the case for *T. eques* females from Hidalgo, Mexico.

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## APPENDIX

*Thamnophis eques* from Mexico examined from the herpetology collections of the Natural History Museum of Los Angeles County (LACM) and The University of Arizona (UAZ).

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CHIHUAHUA (LACM 75675, 21418; UAZ 26815, 26836, 30925, 31255-31257, 34118, 34121-34123, 34125, 34127, 34128, 34150, 34151, 34153, 34156, 34179, 34181, 34182, 34186, 34187, 34431, 34432, 35009, 35010, 35243-35245, 35247, 35248, 35921, 36299). DURANGO (LACM 51624, 51626, 51627, 104187, 104188, 104190-104194; UAZ 26814, 26816, 26912). JALISCO (LACM 37350, 37351; UAZ 34755). MICHOACÁN (UAZ 26808). SONORA (UAZ 26837, 38863). ZACATECAS (UAZ 26811).

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

| Month | N | Regression | Recrudescence | Spermiogenesis |
|---|---|---|---|---|
| May | 2 | 0 | 2 | 0 |
| June | 6 | 0 | 2 | 4 |
| July | 4 | 0 | 0 | 4 |
| August | 3 | 0 | 0 | 3 |
| September | 5 | 3 | 0 | 2 |

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

| Month | N | Inactive | Early yolk deposition | Enlarged follicles (> 6 mm length) | Oviducal eggs or developing embryos |
|---|---|---|---|---|---|
| May | 2 | 0 | 1 | 0 | 1* |
| June | 7 | 5 | 0 | 0 | 2 |
| July | 4 | 2 | 0 | 0 | 2 |
| August | 2 | 2 | 0 | 0 | 0 |
| September | 4 | 3 | 0 | 1 | 0 |
| December | 2 | 1 | 0 | 1 | 0 |

*damaged oviducal eggs could not be counted.