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Reproduction in the Great Basin Collared Lizard, *Crotaphytus bicinctores* (Squamata: Crotaphytidae)

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Abstract.—The reproductive cycle of the Great Basin Collared lizard, *Crotaphytus bicinctores* was studied by a histological examination of museum specimens. Mean clutch size (n = 13) was 3.46 ± 1.1 SD, range: 2–6. Histological evidence indicates that two clutches may be produced in the same year. The reproductive season includes spring and early summer. There was a significant positive correlation between female body size (SVL) and clutch size (P = 0.03). The smallest reproductively active male and female C. bicinctores measured 81 mm and 78 mm SVL, respectively.

Crotaphytus bicinctores frequents xeric rocky habitat in southeastern and extreme northeastern California, much of Nevada, western and northern Arizona, western and central Utah, southeastern Oregon and western Idaho (McGuire 1996). Information on its reproduction is limited and consists of reports of juveniles, gravid females and adult males from southeastern California on 2 May (McGuire 1996); neonates observed in eastern Oregon in August (Brooking 1934); mean clutch size of 5.38, (range: 3–7), egg deposition in June in Millard County, Utah (Andre and MacMahon 1980). Ryan (2009) reported clutch sizes of 3–7, the possibility of 2 clutches and hatchlings appearing in August (Ryan 2009). The purpose of this paper is to examine the reproductive cycle of *C. bicinctores* in the southern portion of this species range. Information on the reproductive life history including period of sperm production, timing of yolk deposition and number and sizes of clutches produced, provides critical information for formulating conservation policies of lizard species populations. Due to the difficulty in justifying collections of monthly lizard samples, utilization of museum collections for obtaining reproductive data has become increasingly important.

A sample of 135 *C. bicinctores* consisting of 57 adult males (mean snout-vent length [SVL] = 94.5 mm ± 7.7 SD, range: 81–117 mm; 47 adult females (mean SVL = 88.5 mm ± 5.9 SD, range: 78–98 mm); 31 subadults consisting of 15 males (mean SVL = 67.8 mm ± 6.5 SD, range: 58–78 mm SVL) and16 females (mean SVL = 67.1 mm ± 6.5 SD, range: 56–75 mm SVL) from Arizona (n = 5), California (n = 114), Nevada (n = 14) and Utah (n = 2) was examined from the herpetology collection of the Natural History Museum of Los Angeles County (LACM: Appendix 1). Lizards were collected from 1929–1980.

The left testis was removed from males and the left ovary was removed from females for histological examination (Presnell and Schreibman 1997). Enlarged ovarian follicles (> 5 mm) and/or oviductal eggs were counted. Tissues were embedded in paraffin, sectioned at 5 μm and stained with hematoxylin followed by eosin counterstain. Slides of testes were examined to ascertain the stage of the testicular cycle. Slides of ovaries were

Cover Page Footnote: We thank Christine Thacker (LACM) for permission to examine specimens.

Table 1. Monthly stages in the testicular cycle of 57 *Crotaphytus bicinctores*.

Month	*n*	Regression	Recrudescence	Spermiogenesis
March	5	1	2	2
April	18	1	5	12
May	17	1	4	12
June	9	1	0	8
July	4	1	1	2
August	4	4	0	0

examined to ascertain the stage of the ovarian cycle. Histology slides were deposited in the herpetology collection of LACM. Mean SVL of male and female *C. bicinctores* were compared using an unpaired *t*-test and the relationship between female SVL and clutch size was examined using linear regression analysis (Instat vers. 3.0b, Graphpad Software, San Diego, CA).

The mean SVL of males of *C. bicinctores* significantly exceeded that of females (unpaired *t* test, $t = 4.4$, $df = 102$, $P < 0.000l$). Monthly changes in the testicular cycle of *C. bicinctores* are shown in Table 1. Three stages were present: (1) Regression, the germinal epithelium is reduced to 1–3 cell layers in thickness and consists of spermatogonia and Sertoli cells; (2) Recrudescence, a proliferation of germ cells for the next period of sperm formation is underway. In early recrudescence, primary spermatocytes predominate, whereas in late recrudescence, secondary spermatocytes and spermatids are most abundant; (3) Spermiogenesis, lumina of the seminiferous tubules are lined by clusters of sperm or metamorphosing spermatids. The period of sperm production encompassed March to July. The smallest reproductively active (spermiogenic) male of *C. bicinctores* (LACM 16870) measured 81 mm SVL, and was collected May 1954 from Los Angeles County. Males < 81 mm SVL had not attained reproductive maturity.

Monthly changes in the ovarian cycle of *C. bicinctores* are presented in Table 2. Four stages were present in the ovarian cycle of *C. bicinctores*: (1) No yolk deposition (quiescent); (2) Early yolk deposition with basophilic granules present in the ooplasm; (3) Enlarged preovulatory follicles (> 4 mm); (4) Oviductal eggs. Gravid females were noted April to June. Mean clutch size ($n = 13$) was 3.46 ± 1.1 SD, range: 2–6. Linear regression analysis revealed a significant positive correlation between female SVL (n = 13) and clutch size ($Y = -4.95 + 0.095X$, $r = 0.57$, $P = 0.04$). A significant positive correlation between female SVL and clutch size was also reported for *C. bicinctores* from Millard

Table 2. Monthly stages in the ovarian cycle of 47 *Crotaphytus bicinctores*, * = one female with oviductal eggs and concurrent early yolk deposition for a second egg clutch; ** = some enlarged follicles in one female were crushed not allowing clutch determination.

	n	Quiescent	Early yolk deposition	Enlarged follicles > 5 mm	Oviductal eggs
April	8	6	1	0	1
May	23	9	8	4	2
June	6	0	0	5**	1*
July	3	3	0	0	0
August	6	5	1	0	0
October	1	1	0	0	0

County, Utah (Andre and MacMahon 1980). The smallest reproductively active female (LACM 26840) measured 78 mm SVL, contained three oviductal eggs and was collected June 1958 in Churchill County, Nevada. Females < 78 mm SVL had not attained reproductive maturity. One female (LACM 16865) from Los Angeles County collected 14 June (SVL 93 mm) contained three oviductal eggs and concurrent early yolk deposition for a subsequent clutch indicating select females may produce two clutches in the same reproductive season. A single female collected in August exhibited early yolk deposition (Table 2), but it is not known if she would have produced a clutch or if the follicles would have undergone atresia. Follicular atresia is commonly observed near the close of the reproductive season when follicles that did not complete vitellogenesis degenerate (Goldberg 1973).

Andre and MacMahon (1980) reported a mean clutch size of 5.38, (range 3–7) for six females from northern populations from Idaho and Utah. Fitch (1985) reported a mean value of 5.0, range 3–8 for six northern *C. bicinctores* females from southern Idaho and Millard and Tooele counties, Utah. Both values for northern *C. bicinctores* females were greater than those of Fitch (1985) for seven females from southern populations in California, Nevada and Utah (3.9, range 2–5), suggesting larger clutch sizes at higher latitudes. The value provided by Fitch (1985) for southern populations approximates our value ($n = 13$, 3.46 ± 1.1 SD, range 2–6) reported herein. Whether the larger clutch sizes in *C. bicinctores* from northern populations reflect a shorter activity season with fewer females producing two clutches, warrants further study.

Other species of *Crotaphytus* exhibit similar reproductive life histories. *Crotaphytus vestigium* exhibited a spring to early summer period of sperm production and ovarian activity (Goldberg and Mahrdt 2010) as did *Crotaphytus collaris* (Fitch 1956, Trauth 1978, 1979). Two clutches were produced by *C. collaris* in Arkansas (Trauth 1978). Based on anecdotal information such as time of egg deposition and appearance of hatchlings, other species of *Crotaphytus* also appear to exhibit similar timing of events in their breeding cycle, even though their reproduction has not been studied in detail. The report of a gravid female of *Crotraphytus grismeri* from September (McGuire 1996) merits further investigation. Detailed studies of the breeding cycles of *C. antiquus, C. dickersonae, C. grismeri, C. insularis* and *C. nebrius* (see McGuire 1996, Grismer 2002, Babb 2009) are needed to ascertain similarities in the timing of the reproductive cycles, and also to determine if significant geographic variation in their clutch sizes exists.

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Appendix I

Crotaphytus bicinctores examined from the Natural History Museum of Los Angeles County (LACM) by state and county: **Arizona**: Coconino 134075; Yuma 16892, 16893, 26835, 26836; **California**: Inyo 26824–26832, 36666, 36667, 36670, 52887, 122468, 123321–123325; Kern 63829–63835, 63837, 63838; Los Angeles 3994–3996, 3998, 3999, 16865, 16867, 16870, 16872, 26811, 26812, 26814, 26815, 63188–63190, 94595–94600, 132436, 132437; Riverside 16876, 16885, 16888, 16889, 26820, 26822, 62448, 94601–94624, 94626; San Bernardino 16877, 16882, 16883, 21651, 23245, 26816–26819, 63179, 63180, 63184–63186, 64007, 94633–94638, 94641–94643, 94645–94648, 137887, 137890; **Nevada**: Churchill 26840–26843; Clark 76458, 107189 Lyon 26837, 26838; Mineral 126915; Nye 61454, 94658, 94659, 133205; Pershing 112790; **Utah:** Washington 94678, 147476.