

November 2012


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### Recommended Citation

Goldberg, Stephen R.; Mahrtdt, Clark R.; and Beaman, Kent R. (2010) "Reproduction in Cope's Leopard Lizard, *Gambelia copeii* (Squamata: Crotaphytidae)," *Bulletin of the Southern California Academy of Sciences*: Vol. 109: Iss. 1.  
Available at: <http://scholar.oxy.edu/scas/vol109/iss1/3>

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## Research Note

### Reproduction in Cope's Leopard Lizard, *Gambelia copeii* (Squamata: Crotaphytidae)

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*Gambelia copeii* is endemic to Baja California, Mexico, and ranges from extreme southern San Diego County, California, south to Todos Santos on the west coast of the Cape Region, Baja California (Grismer 2002, Mahrtdt et al. 2010). Information on its reproduction is limited to brief accounts (Fitch 1970, McGuire 1996, Grismer 2002, Stebbins 2003, Lemm 2006, Mahrtdt and Beaman 2009). The purpose of this paper is to provide more information on the reproductive biology of *G. copeii* from a histological examination of museum specimens as part of an ongoing analysis of the reproductive patterns of reptiles from Baja California (e.g., Goldberg and Beaman 2003, 2004). The use of museum specimens in life-history studies is becoming increasingly valuable as it is often impossible to collect lizards from native populations. Elucidation of the reproductive strategy is fundamental in documenting the life history of a species and is important in the formulation of conservation policies.

We examined a sample of 39 *G. copeii* consisting of 16 males (mean snout-vent length, SVL = 94.8 mm ± 8.2 SD, range: 80–108 mm) and 23 females (mean SVL = 103.5 mm ± 11.1 SD, range: 83–133 mm) from San Diego County, California, and Baja California, Mexico from the California Academy of Sciences (CAS), Natural History Museum of Los Angeles County (LACM), Museum of Vertebrate Zoology (MVZ), and San Diego Society of Natural History (SDSNH) (Appendix I). Lizards were collected 1922–1978.

The left testis was removed from males and the left ovary was removed from females for histological examination (Presnell and Schreiber 1997). Enlarged follicles (> 5 mm) and/or oviductal eggs were counted. Tissues were embedded in paraffin, sectioned at 5µm, and stained with hematoxylin followed by eosin counterstain. Ovary slides were examined for yolk deposition or corpora lutea. Testes slides were examined to ascertain the stage of the testicular cycle present. Mean body sizes of male versus female *G. copeii* were compared using an unpaired *t*-test and the relationship between body size (SVL) and clutch size was examined by linear regression. Statistical tests were performed using Instat (vers. 3.0b, Graphpad Software, San Diego, CA).

Monthly stages in the testicular cycle of *G. copeii* are shown in Table 1. Two stages were present in our samples: (1) spermiogenesis (sperm formation), in which the seminiferous tubules are lined by clusters of mature spermatozoa and/or metamorphosing spermatids; and (2) regressed, in which sperm production has ceased, seminiferous tubules are reduced in size, and the dominant cells present are spermatogonia and Sertoli

Table 1. Monthly changes in the testicular cycle of *Gambelia copeii*.

Month	n	Regression	Spermiogenesis
March	1	0	1
April	4	0	4
May	2	0	2
May–June	1	0	1
June	7	0	7
August	1	1	0

cells. *Gambelia copeii* appears to follow a testicular cycle typical of other lizards from the North American temperate zone characterized by a spring-summer period of spermiogenesis followed by late summer regression (e.g., Goldberg 1974, 1975). One male from June was in a late stage of spermiogenesis in which the germinal epithelium was reduced to 5–6 layers, although sperm were still being produced. Spermiogenesis was almost complete in this male. The smallest reproductively active males in spermiogenesis were captured in June and measured 80 mm SVL (SDSNH 5078, 5079).

The mean female body size of *G. copeii* significantly exceeded that of males (unpaired *t* test,  $t = 2.66$ ,  $df = 37$ ,  $P = 0.0114$ ). Monthly stages in the ovarian cycle of *G. copeii* are shown in Table 2. Five stages were present in the ovarian cycle: (1) no yolk deposition (quiescent); (2) early yolk deposition with basophilic yolk granules present; (3) enlarged preovulatory follicles (> 5 mm diameter); (4) oviductal eggs with eggs in oviducts; and (5) corpora lutea, indicating an egg clutch had been deposited. Because of museum policy, clutch sizes (consisting of enlarged follicles > 5 mm) from specimens SDSNH 18946 (June) and SDSNH 26752 (March) were not counted. The oviductal eggs in CAS 56105 (July) were damaged and could not be counted. Mean clutch size ( $n = 10$ ) was  $5.0 \pm 1.4$  SD, range: 3–8 eggs. Lemm (2006) in his field guide, reported 11 as the maximum clutch size for *G. copeii*, although we suspect Lemm's statement is in reference to *Gambelia wislizenii* (see Parker and Pianka 1976). Linear regression analysis indicated the positive correlation between female SVL and *G. copeii* clutch number was not significant ( $P = 0.064$ ,  $r^2 = 0.366$ ). This may reflect our small female sample size. The smallest reproductively active female (follicles > 5 mm) was from May and measured 83 mm SVL (CAS 57865). We noted the presence of one *G. copeii* female from March with corpora lutea from a previous clutch (SDSNH 26753), two other females from March with enlarged follicles (> 5 mm), and one female from June with oviductal eggs and concomitant yolk deposition for a subsequent egg clutch (MVZ 37260). These observations provide evidence that *G. copeii* may produce multiple annual clutches. In addition, the female in March with corpora lutea had sufficient

Table 2. Seasonal changes in the ovarian cycle of *Gambelia copeii*; \* = oviductal eggs and concurrent yolk deposition for second clutch; \*\* one clutch was not counted.

Month	n	No yolk deposition	Early yolk deposition	Enlarged follicles > 5 mm	Oviductal eggs	Corpora lutea
March	4	0	0	3**	0	1
April	6	4	2	0	0	0
May	8	1	0	5	2	0
June	2	0	0	1**	1*	0
July	2	1	0	0	1**	0
October	1	1	0	0	0	0

time to produce a subsequent clutch, as did the other two March females with enlarged ovarian follicles. Fitch (1970) also reported two female *Gambelia sp.* from Baja California as being gravid in March. Our findings support Fitch (1970) and Lemm (2006), that two clutches may be produced by *G. copeii* in optimal years.

In view of the extensive geographic range of *G. copeii* (Grismer 2002), subsequent studies are needed to ascertain the degree of geographic variation in the reproduction of this species. Fitch (1985) reported clutch sizes of the congeneric *G. wislizenii* tended to be larger in the southern part of its range. Grismer (2002) observed a female with breeding coloration during late August near Todos Santos in the cape region of Baja California Sur. However, use of breeding coloration in museum specimens as an indicator of reproductive activity in natural populations, is not possible, as the pigments fade in alcohol.

#### Acknowledgments

We thank Jens Vindum (CAS), Christine Thacker (LACM), Carol Spencer (MVZ), and Brad Hollingsworth (SDSNH) for permission to examine specimens.

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#### Appendix I

*Gambelia copeii* examined from the California Academy of Sciences (CAS), Natural History Museum of Los Angeles County (LACM), Museum of Vertebrate Zoology (MVZ), and San Diego Society of Natural History (SDSNH). California, San Diego County: CAS 57865, SDSNH 46099, 55251; Baja California: CAS 11547, MVZ 31794, 50017, 50020, 140756, 140757, 161174, LACM: 4005, 4006, 4932, 15708, 126596, 126598, 137898, SDSNH 5078, 5079, 17470, 18118, 18945, 18946, 26752, 26753, 43007, 52959; Baja California Sur: CAS 56105, 65857, 90297, 90458, 147739, 147750, MVZ 13150, 13151, 37260, 37261, 37262, 50018.