

## REPRODUCTIVE ACTIVITY AND SEXUAL DIMORPHISM OF *LIOLAEMUS MULTIMACULATUS* (SAURIA: TROPIDURIDAE)

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The lizard *Liolaemus multimaculatus* inhabits coastal sand dunes of the Buenos Aires Province of Argentina, and exhibits reproductive activity during the spring and early summer months. Females had vitellogenic follicles and oviductal eggs between September and December, and the size at maturity was 48.2 mm (snout-vent length). Five females with yolked follicles as well as eggs were collected in November, indicating that at least some females can produce more than one clutch per reproductive period. Mean clutch size was 4.2 eggs (range 3 to 7) and clutch size was positively correlated with female body size. In males, testicular diameter peaked in August (late winter) and September (early summer) and declined in January (mid-summer). Testis size increased gradually from January onwards. Individual males were seen more frequently during the reproductive period than females. Hatchlings were first seen in February and March and the smallest measured 26.7 mm (snout-vent length). *Liolaemus multimaculatus* males were larger than females in several morphological traits: snout-vent length, head length, distance between fore and hind limbs, tibio-fibulla, hand and foot lengths.

### INTRODUCTION

Reproductive patterns, growth rates and survivorship are the principal life history traits of an organism (Stearns, 1992). Recently, the reproductive cycles of temperate lizards in Argentina have begun to be studied (Fitzgerald *et al.* 1993; Aun & Martori, 1994; Martori & Aun, 1994; Cruz, 1994a, b, 1995), including some of the tropidurid *Liolaemus* species (Ramirez Pinilla, 1991a, 1992). Reproductive cycles of the *wiegmanni* group (Etheridge, 1995) which includes *L. multimaculatus* and other arenicolous forms of Argentina and South America are known, specifically for *L. wiegmanni* (Ramirez Pinilla, 1991b; Martori & Aun, unpublished data), *L. scapularis* (Ramirez Pinilla, 1994) and *L. lutzae*, a tropical lizard from south-eastern Brazil (Rocha, 1992). For all these species, the reproductive pattern was described as a cyclic spring-summer pattern with variations in clutch size or in the number of clutches per season. Recently, Ramirez Pinilla (1994) pointed out that in spite of its poorly known cycle, observations of male and female individuals of *L. multimaculatus* suggest they should also exhibit reproductive activity in spring and early summer.

The aim of this paper is to describe the reproductive activity, size at maturity and clutch size of *L. multimaculatus*, and also to examine sexual dimorphism in the population under study. Results will be compared to related species.

### MATERIAL AND METHODS

*Liolaemus multimaculatus* is a small lizard with a brownish dorsum, with small dark spots and blue scales between armpit and groin. It is an endemic species from

the coastal sand dune habitats of Buenos Aires and Río Negro Provinces of Argentina (Gallardo, 1977; Cei, 1993). In a recent taxonomic review (Etheridge, 1993) the forms *L. m. multimaculatus* and *L. m. riojanus* from the sandy flats of La Rioja and San Juan Provinces (Cei, 1979) were regarded as two different species.

### STUDY AREA

Fieldwork was carried out at Mar del Sur (38°21'S; 57°59'W) in the south-east of Buenos Aires Province, Argentina. Mean annual temperature is 13.5°C with maximum average in January (21°C) and minimum in July (7.5°C). Annual average rainfall is over 830 mm and there is no marked wet or dry season, although precipitation is at its highest level in March and lowest in August, with 105 mm and 50 mm respectively. The vegetation at the site consisted mainly of *Spartina ciliata* and *Panicum racemosum* on the foredunes and short grass communities of *Bothriochloa* sp., *Lagurus ovatus*, *Adesmia incana* and *Ambrosia tenuifolia* stabilizing backdunes. Another lizards in the study area was *L. gracilis*, which occupied foredunes along with *L. multimaculatus*. *Stenocercus pectinatus* and *Ophiodon vertebralis* occurred in the area, but were restricted to backdune habitats (Vega, 1994).

The study site was visited once every month, from September 1984 to December 1985. During each visit, a census between 11.00 and 12.00 hrs was made by walking a 15 ha strip along coastal sand dunes, searching for lizards. For each lizard, age (adult or juvenile) was recorded, but sex was difficult to identify. Before each census, air temperature (100 cm above ground) was measured with a Miller-Weber thermometer to the nearest 0.1°C. From January to December 1985, in ad-

dition to the census, an attempt was made to capture individuals by hand. A total of 73 specimens (adults: 33 males, 29 females; juveniles: 6 males, 7 females) was collected. All these specimens were killed with ether minutes after capture, and within a few hours were injected with 10% formalin and stored in 70% ethyl alcohol. Voucher specimens were deposited at the Herpetological Collection of Vertebrate Laboratory, Universidad Nacional de Mar del Plata. Measurements were made using a digital calliper (accuracy 0.01 mm) and included snout-vent length (SVL), head length (from posterior edge of auricular opening to rostral scale), head width (between corners of the mouth), distance between fore and hind limbs, humerus length (from elbow to axilla), radius-ulna length (from elbow to the internal angle between hand and forearm), hand length (including fourth toe-nail), femur length (from knee to groin), tibio-fibula length (from knee to the internal angle with the foot), foot length (including fourth toe-nail) and tail length.

Specimens were dissected and gonads removed. Sex and age class (juvenile or adult) were noted. Females were considered adults if they had yolked follicles (YF) over 2 mm that were opaque and yellow, oviductal eggs (OE) or distended oviducts, in which case left oviduct width (OW) was measured. For males, sexual maturity was based on the presence of enlarged testes and convoluted epididymes, and left testis length (TL) and width (TW) were measured. All these measurements were made using an ocular micrometer of a binocular microscope.

In statistical analyses, distribution and variances of the data sets were examined, and transformations were performed as necessary to ensure that the assumptions of the tests being applied were not violated. The effects of SVL were removed with ANCOVAs when slopes among groups were homogeneous at the  $P < 0.05$  alpha level. Means are given  $\pm 1$  SD (Zar, 1984).

## RESULTS

### FEMALE REPRODUCTIVE CYCLE AND CLUTCH SIZE

The smallest reproductive female with yolked follicles measured 48.2 mm SVL and the biggest measured 63.2 mm SVL, while the smallest female with eggs measured 51.2 mm SVL. Mean SVL of females showing evidence of reproduction was  $54.6 \text{ mm} \pm 3.97$  ( $n=29$ ). The smallest female size with distended oviducts (OW = 2.5 mm) but without yolked follicles or oviductal eggs, was 54.8 mm SVL.

Principal events of the adult (individuals  $> 48.2$  mm SVL) female cycle were the following: in September 100% of females ( $n=1$ ) had yolked follicles, in November, 62.5% ( $n=8$ ) of females had simultaneously yolked follicles and oviductal eggs while another 37.5% ( $n=8$ ) had oviductal eggs. In December, 12.5% ( $n=16$ ) of the females had oviductal eggs and 25% ( $n=16$ ) yolked follicles. In January and February ( $n=1$  and 3 respectively) there was no evidence of reproductive activity.

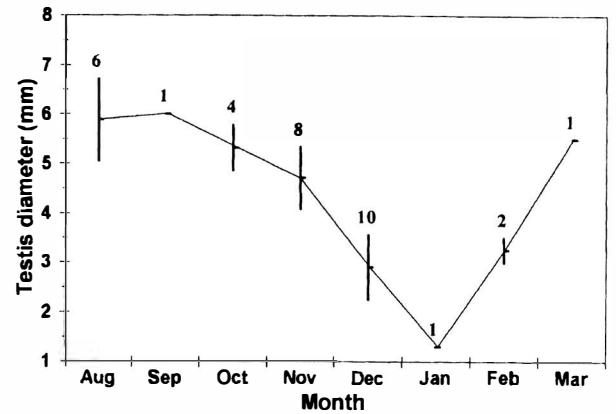


FIG. 1. Reproductive events of male *Liolaemus multimaculatus*. Mean values of testis diameters represented by bold lines. 1 SD represented by vertical lines.

Clutch size, based on the number of oviductal eggs in both oviducts, ranged from 3 to 7 (mean =  $4.2 \pm 1.22$ ,  $n = 10$ ). Clutch size was positively correlated with female size (SVL) (Spearman's  $r = 0.72$ ,  $n = 10$ ,  $P < 0.03$ ).

### MALE REPRODUCTIVE CYCLE

The smallest male with obviously enlarged testes was 47.5 mm SVL (TL = 4.5 mm, TW = 3 mm) and the largest reproductive male was 69.6 mm SVL (TL = 8 mm, TW = 6 mm). The mean SVL of males showing evidence of reproduction was  $59.1 \pm 4.42$  mm SVL ( $n=30$ ). The highest values of width and length of testis were observed in August and September, decreasing in the following months until testis sizes were at their minimum sizes in January. Testis size began to increase again in February and March (Fig. 1). There were significant differences in adjusted TW using least squares means, for the months evaluated (ANCOVA:  $F = 13.03$ ,  $P < 0.0001$ ,  $df = 4, 21$ ,  $n = 27$ ; homogeneity of slopes:  $df = 4, 17$ ,  $F = 0.01$ ,  $P < 0.05$ ) and Tukey Multiple Comparisons Test found significant differences ( $P < 0.05$ ) between December and all months except February, and between February and August. Values of September, January and March were not considered in the analysis because only one specimen was in the sample.

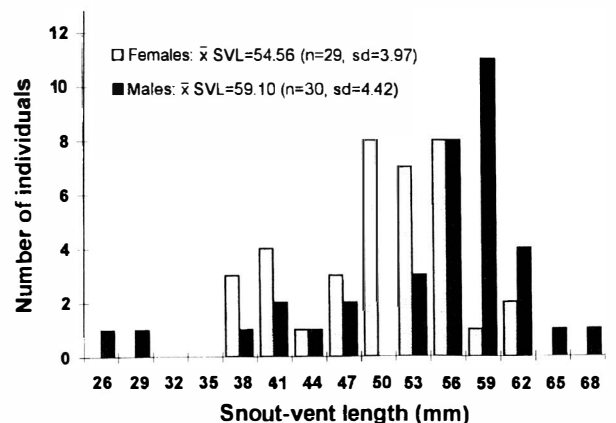


FIG. 2. Size-frequency distributions for male and female *Liolaemus multimaculatus*.

TABLE 1. Survey and capture data of *Liolaemus multimaculatus* at Mar del Sur. \* air temperature during census

Date	T°C*	Number of individuals				
		Censused		Captured		
		adult	juvenile	adult male	adult female	juvenile
September 1984	11	0	0	0	0	0
October 1984	17	0	0	0	0	0
November 1984	30	12	0	0	0	0
December 1984	17	0	0	0	0	0
January 1985	27	2	0	1	1	0
February 1985	26	9	1	2	3	1
March 1985	26	1	3	1	0	1
April 1985	18	0	0	0	0	0
May 1985	21	1	0	0	0	0
June 1985	18	0	0	0	0	0
July 1985	12	0	0	0	0	0
August 1985	21	7	0	6	0	0
September 1985	11	3	0	1	1	0
October 1985	28	6	0	4	0	0
November 1985	25	16	2	8	8	2
December 1985	27	38	9	10	16	9

## REPRODUCTIVE ACTIVITY

Individuals of *L. multimaculatus* emerged from over-wintering in August and ceased activity in late March. The numbers of individuals seen active during censuses were positively correlated with air temperatures on the census days (Spearman's  $r = 0.74$ ,  $n = 16$ ,  $P < 0.002$ ).

Lizards were caught in the same proportion as were seen (Table 1). Between April and July, lizard activity was not observed except for one individual seen in May, but no captures were made during these months. Captures in August and October were exclusively comprised of males. The highest number of females were caught in November and December, comprising 82% of total female captures (Table 1).

The two smallest sized individuals, probably hatchlings, were collected in February (male, SVL = 26.7 mm) and March (male, SVL = 29.5), but one individual of approximately the same body size was seen in January in Costa Bonita locality, 60 km south of Mar del Sur, though hatching period would seem to occur during mid-summer (January, February). Larger juveniles or subadult lizards collected ranged from 39.1 mm to 46.4 mm SVL ( $n = 11$ ).

## SEXUAL DIMORPHISM

*L. multimaculatus* exhibited sexual size dimorphism, with males attaining larger sizes than females (Mann-Whitney  $U$  Test:  $z_{30,29} = 3.74$ ,  $P = 0.0001$ ) (Fig. 2). Analyses of covariance used to remove the effects of SVL revealed that males had significantly larger heads than females (see Table 2 for statistics). Similarly, the distance between forelimbs and hindlimbs,

length of tibio-fibula, and lengths of hands and feet were significantly larger in males than in females. There were not found to be significant differences between sexes in morphological traits such as head width and lengths of humerus, radius-ulna, femur and tail (Table 2).

Sexual dichromatism was only evident in ventral surface. Females possessed an immaculate venter, whereas males showed brown spots on the throat and ventral surface. Ventral spots were more diffuse in juvenile males. Males had also yellowish precloacal pores that were lacking in females.

## DISCUSSION

The reproductive cycle of *L. multimaculatus* is seasonal in spring (September) and early summer (December). Testicular diameter was maximum in late winter (August) and early spring (September) and minimum in mid-summer; in late summer it began to increase gradually. Although the middle and late summer sample was small, it seems that testicular regression would be at a maximum at mid-summer and recrudescence would occur in early autumn. This testis cycle was also observed in other species of the *wiegmanni* group (Ramirez Pinilla, 1991b, 1994; Rocha, 1992; Martori & Aun, unpublished data). Maximum testicular diameters were synchronized with the presence of vitellogenic follicles in females through September to November, but not in December. First copulations probably occur in August and September.

The simultaneous occurrence of oviductal eggs and enlarged vitellogenic follicles in females captured in November suggests that some females produce more than one clutch per season. Multiple clutches are

TABLE 2. Results of tests for differences in the mean size of several morphological traits of male and female *Liolaemus multimaculatus*. The effects of SVL were removed using ANCOVA. All measurements are in mm.

	Mean (SD)	N	ANCOVA results	
			F-ratio	P
<i>Head length</i>				
males	14.16 (1.00)	30	5.22	0.026
females	12.93 (0.98)	29		
<i>Head width</i>				
males	10.51 (1.02)	30	3.64	0.061
females	9.54 (0.65)	29		
<i>Distance between limbs</i>				
males	26.00 (2.93)	28	25.48	0.000
females	25.87 (3.09)	25		
<i>Humerus length</i>				
male	5.64 (0.85)	28	0.13	0.719
females	5.12 (0.43)	25		
<i>Radius-ulna length</i>				
males	7.59 (0.80)	28	0.69	0.409
females	6.89 (0.61)	25		
<i>Hand length</i>				
males	10.27 (0.66)	30	50.74	0.000
females	8.80 (0.56)	29		
<i>Femur length</i>				
males	8.44 (0.88)	28	0.17	0.677
females	7.91 (0.75)	25		
<i>Tibio-fibula length</i>				
males	9.39 (1.01)	28	4.14	0.047
females	8.12 (0.92)	25		
<i>Foot length</i>				
males	16.88 (0.96)	30	21.78	0.000
females	15.14 (1.07)	29		
<i>Tail length</i>				
males	55.86 (7.69)	25	0.25	0.615
females	51.68 (5.59)	26		

known to occur in *L. wiegmanni* from central and chacoan regions of Argentina (Ramirez Pinilla, 1991b; Martori & Aun, unpublished data), in *L. scapularis* from north-western Argentina (Ramirez Pinilla, 1994) and in *L. lutzae* from south-eastern Brazil (Rocha, 1992). Mean clutch size of *L. multimaculatus* (4.2 eggs) was similar to that observed for *L. wiegmanni* from the chaco (4.5 eggs) (Ramirez Pinilla, 1991b) and *L. scapularis* (4.3 eggs) (Ramirez Pinilla, 1994), but about one egg less on average than in *L. wiegmanni* from central regions of the country (5.4 eggs) (Martori & Aun, unpublished data). Mean clutch size of *L. multimaculatus* at our site was greater than that observed for *L. lutzae* (2.0 eggs), but *L. lutzae* produces two or three clutches over a longer reproductive period (Rocha, 1992). For all these species, female size at maturity ranged from 40.2 mm to 51.5 mm SVL (Ramirez

Pinilla, 1991b, 1994; Rocha, 1992; Martori & Aun, unpublished data), and it is probably associated at the species body size. Clutch size was positively correlated with female body size, a pattern commonly observed in other tropidurid species of Argentina (Ramirez Pinilla, 1994; Cruz, 1995; Martori & Aun, unpublished data). The above mentioned and additional reproductive data of other *Liolaemus* species (Ceï, 1993) indicate that a seasonal cycle with an average clutch size of 4 to 6 eggs laid in more than one clutch per reproductive period, would seem to be a life history trait that emerges in oviparous *Liolaemus* species of subtropical and temperate regions of Argentina.

The activity period of *L. multimaculatus* extends through late winter (August), spring (September to December) and late summer (March), while autumn (April) and early and mid-winter (June, July) is the in-

active period. This seasonal and thermic pattern of activity is common in temperate reptiles (Huey, 1982). As the highest rates of observations and captures of females were in late spring, coinciding with their ovigerous condition, it is presumed that females were more active at this time, possibly searching for nesting sites. In contrast, the numbers of males captured monthly in spring and early summer were more constant. This kind of differential activity between sexes, especially the high activity rates of males during the reproductive season, was observed in many lizard species (Stamps, 1983).

Males of *L. multimaculatus* were larger than females in several body traits. This sexual dimorphism may be related to sexual selection, as occurs in most territorial iguanian lizards (Stamps, 1983), or to natural selection by means of intraspecific niche divergence (Schoener, 1977, 1986), but more behavioural and ecological data are needed to differentiate between these hypotheses. Ventral sexual dichromatism of *L. multimaculatus* contrasts with dorsal sexual dichromatism seen in most *Liolaemus* species, that may vary from a different to a paler or less strong pattern for the females (Ceí, 1993). In *L. multimaculatus* both males and females are highly dorsally cryptic and match the texture and colour of sandy substrate.

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