

Reproduction in Australian Elapid Snakes I. Testicular Cycles and Mating Seasons

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Abstract

Testicular cycles and mating seasons of eight species of elapid snakes are described. Four livebearing species (*Austrelaps superbus*, *Hemiaspis signata*, *Notechis scutatus* and *Unechis gouldii*) show spermiogenesis in summer, retain active spermatozoa in the vas deferens all year, and mate in both autumn and spring. *Hemiaspis daemeli* shows a similar testicular cycle, but its mating season is unknown. Another livebearing species, *Pseudechis porphyriacus*, shows spermiogenesis in spring, retains spermatozoa in the vas deferens only during spring, and mates only in spring. Two oviparous species (*Pseudonaja nuchalis* and *P. textilis*) show spermiogenesis in spring (and possibly also in summer), and retain spermatozoa in the vas deferens all year. Both *Pseudonaja* species mate in spring, and possibly through summer. These data are consistent with the hypothesis that the times of production of spermatozoa, and retention of spermatozoa in the vas deferens, are adapted to the timing of the mating season.

Introduction

Although a considerable literature exists on seasonal patterns of testicular activity in representatives of all the vertebrate classes, there are still many groups about which very little is known. One such family is the Elapidae (the proteroglyphous land snake—cobras and their allies). Members of this group are prominent elements of the venomous snake fauna in all continental tropical and subtropical land masses. Testicular cycles have been described in detail for only one species, and there is limited information on three others (Lofts *et al.* 1966; Saint Girons 1966; Saint Girons and Pfeffer 1971). The present study describes testicular cycles and mating seasons in eight species of Australian elapid snakes from two study areas.

Materials and Methods

Snakes were collected: (1) in the highlands New England region of New South Wales, in eastern Australia, within 100 km of the city of Armidale (30° 31' S., 151° 39' E., elevation 980 m); (2) 400 km west of Armidale, in a large swamp surrounded by semiarid plains, 20 km west of Quambone, N.S.W. (30° 58' S., 148° 23' E., elevation 180 m). The climate of Quambone differs considerably from that at Armidale; it is warmer (mean daily maximum air temperature 30.0°C in January, 18.2°C in June) and drier (mean annual rainfall 497 mm). Armidale climate, habitat types, and snake ecology have been described elsewhere (Shine 1975, 1977a). Female reproductive cycles are described in a following paper (Shine 1977b). Table 1 lists the nomenclature, body sizes, sample sizes and reproductive mode of the species studied.

Specimens were collected by hand, returned to the laboratory within 3 h of capture (60 h for Quambone specimens) and killed by intracranial injection of sodium pentobarbitone. Testes were removed, measured and weighed. Small macerated fragments were examined under phase-contrast

microscopy and spermatozoa density judged as rare, common or abundant. Vasa deferentia were examined for the presence or absence of spermatozoa in the same way as above.

A testis weight index was calculated as the ratio of the combined (wet) weight of both testes, to body weight, multiplied by 100. Such a ratio is invalid unless organ weights increase linearly with body weight. Regressions of testis weight indices against body length were calculated for the largest monthly sample of each species (if $n > 6$). In all species except *Austrelaps superbus* the slopes were not significantly different from zero ($P < 0.10$); for *A. superbus*, testis size was not significantly correlated with body size ($P > 0.10$), and testis weight was used instead of a ratio (Fig. 1).

Table 1. Species studied

Study areas: NE, New England; Q, Quambone. Mode of reproduction: L, live-bearing; O, oviparous

Scientific name	Vernacular name	Study area	N	Mean snout-vent length of mature males (cm)	Mode of reproduction
<i>Austrelaps superbus</i>	Copperhead	NE	110	76.6	L
<i>Hemiaspis daemeli</i>	Grey snake	Q	55	42.6	L
<i>H. signata</i>	Swamp snake	NE	53	43.1	L
<i>Notechis scutatus</i>	Tiger snake	NE	174	81.6	L
<i>Pseudechis porphyriacus</i>	Black snake	NE	225	111.6	L
		Q	66	126.2	L
<i>Pseudonaja nuchalis</i>	Brown snake	Q	52	123.9	O
<i>P. textilis</i>	Brown snake	NE	53	126.3	O
<i>Unechis gouldii</i>	Gould's snake	NE	129	36.2	L

Mating seasons were determined from: (1) field records of mating; (2) field records of male-male combat, which is restricted to the mating season (Fleay 1937); (3) the presence of motile spermatozoa in oviducts of recently collected females. Oviducts were dissected out and flushed with water, and this fluid then examined microscopically for motile spermatozoa.

Results

Testes

In all species, high testis weights were correlated with the presence of abundant spermatozoa in the testis (Figs 1, 2). Volsøe (1944) suggested that length would be a more reliable indicator of testicular activity than would weight; in these elapids the trends for both indices were the same (unpublished data). There were clear inter-specific differences in testicular cycles:

- (1) *Notechis scutatus*, *Hemiaspis daemeli*, *H. signata* and *Unechis gouldii* all showed relatively little seasonal variation in testis size. Spermiogenesis occurred in midsummer (December), and the testes contained abundant spermatozoa from then until late autumn (April). They did not contain spermatozoa in spring (Figs 1, 2). *Austrelaps superbus* showed the same cycle, but differed from other species in that large males were often found with inactive testes throughout the year (Fig. 1).
- (2) *Pseudechis porphyriacus*. Testes were small in summer and autumn, and lacked spermatozoa through this period. Recrudescence commenced in autumn but spermiogenesis was delayed until spring. Testicular cycles in the New England and Quambone populations were similar (Figs 1, 2), despite the climatic difference. The oviparous *Pseudonaja nuchalis* showed a testicular cycle similar to that of *Pseudechis porphyriacus*, except that spermiogenesis continued longer.

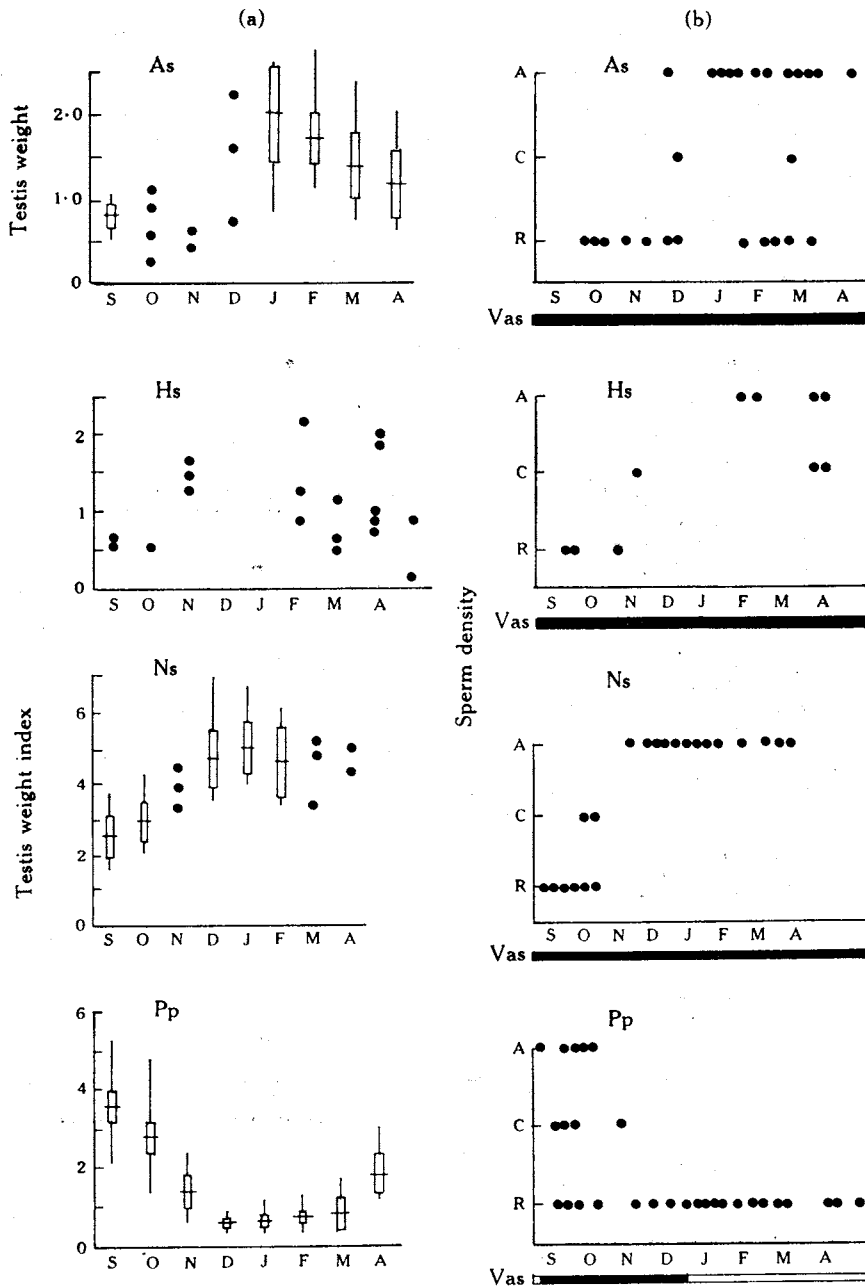


Fig. 1. Seasonal variation in relative testicular weight, and concentration of spermatozoa in the testes, in elapid snakes. (a) 'Testis weight index' versus date of collection. The index is given by the combined weight of both testes, divided by body weight, multiplied by 100. For *Austrelaps*, absolute testis weight is given (g), rather than an index (see text). Horizontal bars show monthly means, rectangles enclose two standard errors on either side of the mean (95% confidence limits), and vertical bars show range of values observed. For any month with $n < 6$, or if only one month in the entire sample had $n > 6$, individual points are plotted. (b) Seasonal variation in the concentration of spermatozoa in the testes: as 'rare' (R), 'common' (C) or 'abundant' (A). The bar below this graph indicates the presence (shaded) or absence (unshaded) of motile spermatozoa in the vas deferens throughout the year. As, *Austrelaps superbus*. Hs, *Hemiaspis signata*. Ns, *Notechis scutatus*. Pp, *Pseudechis porphyriacus* (New England).

Testis weights changed less markedly than in *P. porphyriacus*, and some testes with abundant spermatozoa were found in all parts of the year. The testicular cycle of *P. textilis* is probably similar to that of its western congener (Fig. 2).

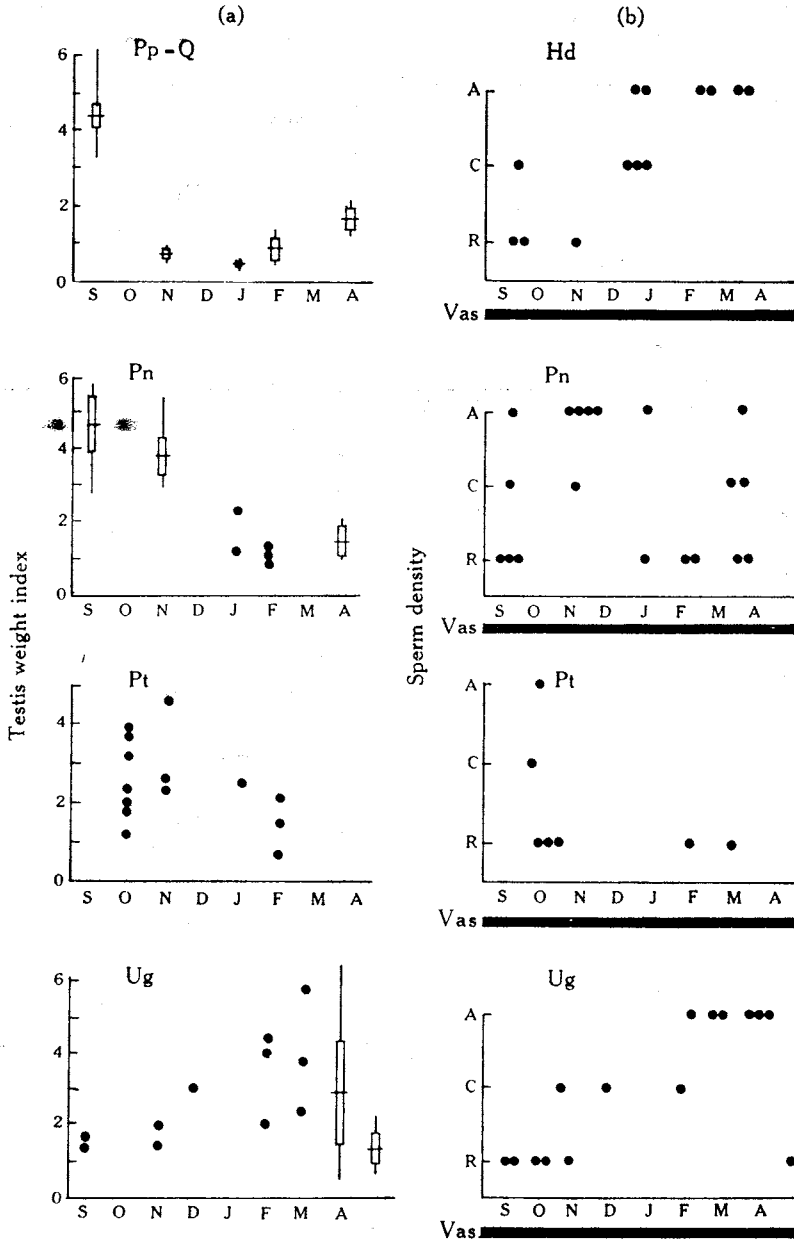


Fig. 2. Seasonal variation in relative testicular weight (a), and concentration of spermatozoa in the testes (b), in elapid snakes. Conventions as in Fig. 1. Pp-Q, *Pseudechis porphyriacus* (Quambone). Hd, *Hemiaspis daemeli* (Quambone). Pn, *Pseudonaja nuchalis* (Quambone). Pt, *Pseudonaja textilis*. Ug, *Unechis gouldii*.

Vasa Deferentia

The ductus deferens contained motile spermatozoa year-round in all species except *Pseudechis porphyriacus*. In this species, the vas deferens was empty of spermatozoa in summer, autumn, and early spring (Figs 1, 2). Some of the 'mature' *Austrelaps superbus* with inactive testes also lacked spermatozoa in the vas deferens in midsummer.

Mating Seasons

In all the elapids studied here, females ovulated in late spring (November). The oviparous (*Pseudonaja*) species oviposited shortly thereafter, but the live-bearers did not give birth until late summer (February–March; Shine 1977b). The presence of abundant motile spermatozoa in oviducts of autumn-collected females (Table 2) shows that mating commenced soon after parturition in all the live-bearing species except *Pseudechis porphyriacus*, in which it was restricted to spring. Absence of oviducal spermatozoa in four April-collected *P. porphyriacus* from Quambone suggests that mating is restricted to spring in this population also.

Table 2. Microscopical determinations for oviducal spermatozoa in seven species of elapid snakes
Letters are abbreviations for months of the year

Species	Sperm	Months								Winter
		S	O	N	D	J	F	M	A	
<i>Austrelaps superbus</i>	Absent	1	–	1	–	–	–	–	–	1
	Present	2	1	1	–	–	–	3	2	1
<i>Hemiaspis signata</i>	Absent	1	–	1	–	–	–	–	1	–
	Present	1	1	–	–	–	–	–	3	–
<i>Notechis scutatus</i>	Absent	1	3	–	2	–	1	1	1	1
	Present	2	3	1	–	–	–	1	1	2
<i>Pseudechis porphyriacus</i> (New England)	Absent	8	3	3	2	2	–	1	3	–
	Present	–	4	2	1	–	–	–	–	–
<i>Pseudonaja nuchalis</i>	Absent	4	–	1	–	1	2	1	–	–
	Present	–	–	5	–	–	–	–	–	–
<i>Pseudonaja textilis</i>	Absent	–	–	1	–	–	–	2	–	–
	Present	–	–	1	–	–	–	–	–	–
<i>Unechis gouldii</i>	Absent	1	–	1	–	–	–	1	1	3
	Present	2	3	5	–	–	–	1	1	–

The autumn-mating elapid species apparently mated in spring as well, because: (1) oviducts of spring-collected females often contained very abundant spermatozoa; this indicates recent mating (Fukada 1959); (2) many mature spring-collected females with enlarged ovarian follicles were uninseminated. The low incidence of follicular resorption in all species (Shine 1977b) suggests that these females must mate before ovulation. This conclusion may be incorrect if spermatozoa in specialized oviducal storage structures were not revealed by the oviducal flushing technique. However, the technique used (maceration of oviducts as well as direct flushing) renders this unlikely. Also, spermatozoa are usually found in the lumen of the oviduct, even in species with specialized spermatozoa storage structures (Rahn 1942).

Field observations support the conclusions drawn from oviducal examinations. I have recorded reproductive activity four times in *Pseudechis porphyriacus*, always in

spring (copulation on 22 September 1972; male combat on 1 October 1973, 12 October 1974 and 13 October 1974). Copulation has been recorded twice in *Austrelaps superbus*, both times in autumn (13 April 1975, G. Witten, personal communication; 10 March 1976, P. Webber, personal communication). Both spring and autumn copulation, and male combat, have been recorded in *Notechis scutatus* (C. Tanner, personal communication; S. Smith, personal communication). Male combat has not been reported previously for *N. scutatus*; both of my informants noted that the postures adopted by fighting males were similar to those described for *Pseudechis* and *Pseudonaja* by Fleay (1937).

I conclude that mating occurs only in spring in one live-bearing species, *Pseudechis porphyriacus*, and in both autumn and spring in four others, *Austrelaps superbus*, *Notechis scutatus*, *Hemiaspis signata* and *Unechis gouldii*. Mating seasons of the egg-laying *Pseudonaja* species are less certain. *P. nuchalis* mates in spring but not in autumn. At least some oviparous elapids produce more than one clutch of eggs per year (Shine 1977b), so they may also mate in midsummer.

Discussion

The spring spermiogenesis of *Pseudechis porphyriacus* is clearly an example of 'prenuptial spermatogenesis' (Volsøe 1944). This type of cycle also occurs in at least three other Australian elapids, *Pseudechis guttatus*, *P. australis* and *Acanthophis antarcticus* (the latter two suggested by Saint Girons (1966), all confirmed by my own unpublished data), and in the Asian elapid *Naja naja* (Lofts *et al.* 1966).

Testicular cycles of the oviparous *Pseudonaja* species are problematical, in that they resemble *P. porphyriacus* but with prolonged spermiogenesis (at least in *P. nuchalis*). Thus, they fall between the categories of 'prenuptial' and 'mixed' spermatogenesis (Saint Girons 1966); further data are needed on this point.

All the other (live-bearing) elapids studied show summer spermiogenesis (Volsøe's (1944) 'postnuptial spermatogenesis'), with little interspecific variation in seasonal timing. Another Australian elapid, *Suta suta*, is thought to show the same type of cycle (Saint Girons 1966).

The data presented here have been used elsewhere (Shine 1975) to test general hypotheses on the evolution of reptilian testicular cycles. The data are consistent with hypotheses that the seasonal timing of spermiogenesis, and the duration of retention of spermatozoa in the vas deferens, are adaptive to the seasonal timing of mating activity.

In spite of the probable importance of interspecific differences in mating seasons in determining interspecific differences in testicular cycles, the former phenomenon has received much less detailed attention than the latter. For example, despite the occurrence of autumn (and probably summer) mating in some of the most common Australian elapid species, recent works (e.g. Worrell 1963; Rawlinson 1965; Cogger 1967) give spring as the sole mating season for these snakes. Earlier workers were more accurate: Kellaway and Eades (1929) noted autumn copulation in *Austrelaps superbus* (but believed that *Notechis scutatus* mated only in spring) and Fleay (1937) mentioned autumn mating in both *A. superbus* and *N. scutatus*. The latter author also suggested that neither *Pseudechis porphyriacus* nor *Pseudonaja textilis* mate in autumn (confirmed by the present study).

These differences in mating season between sympatric species are surprising in view of the general similarity in the timing of the female reproductive cycle (Shine 1977b). Ultimately, any interpretation of these interspecific differences in mating seasons requires an understanding of the selective forces determining the times of onset and cessation of mating activity. This will be a complex answer because males and females are likely to have different optima (Trivers 1972), and because the observed mating season is likely to be a compromise between the two (Gibson and Falls 1975). Further detailed studies on reptilian mating seasons, particularly intraspecific geographic comparisons, would be valuable.

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