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NOTES ON THE REPRODUCTION OF *ELAPHE SCALARIS* (SCHINZ, 1822) IN SOUTHERN SPAIN

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Reproductive aspects of the genus *Elaphe* have been studied in North American species (Gillingham, 1974, 1979, 1980; Seigel & Ford, 1987), but information on European species is scarce and lacking in quantitative data. References to the ladder snake *Elaphe scalaris* can be found in Butler (1991), Dupres (1991), Cheylan & Guillaume (1993) and Pleguezuelos (in press).

In this paper we present information on the reproduction of *E. scalaris*, as a part of a more extensive study on the ecology of the species which was carried out in Doñana National Park (Huelva). The study area is situated in the SW of the Iberian Peninsula (37° 7', 36° 48' N; 6° 12', 6° 30' W). The climate is Mediterranean with Atlantic influence, with mild and wet winters and hot and dry summers. Average annual rainfall is 560 mm. The mean monthly temperature during the study varied between 10.5°C in January and 24.5°C in July-August. The substratum is sandy with shrub-like vegetation and isolated *Quercus suber* trees. Descriptions of the study area may be found in Rivas Martínez, Costa, Castroviejo & Valdés (1980) and Blázquez (1993).

During 1988 and 1989, 14 adult individuals (six males and eight females) were force fed radio-transmitters (Blázquez, 1993). Prior to their release, the snakes were retained in the terrarium for one day for transmitter calibration and to ensure that ingestion had been successful. None of the snakes subsequently released were detected mating or became gravid. The reproductive behaviour described here was observed in the terrarium before the transmitters were inserted.

Each terrarium was a glass cage 1.5 m length, 75 cm wide and 0.5 m high with a movable wood and screen cover at top. The cages were provided with sand and grass on the floor, a bowl of water, and a piece of cork bark as refuge. They were placed outside the laboratory, in the field at ambient humidity, temperature, and light/dark diel rhythm.

Mating was observed on two occasions; one mating involved a male of 864 mm in snout-vent length (SVL), and 295 g weight, and a female of 1070 mm in SVL and 593 g. The pair was captured by hand on 25 and 26 May

respectively, and kept in a separate terrarium until 8 June. The final stage of another mating between a male of 995 mm in SVL and 419 g captured on 17 June and a female of 920 mm in SVL and 340 g captured on 15 June also was observed.

The mating season in captivity lasts from April to the beginning of June in Southern France (Cheylan & Guillaume, 1993). González de la Vega (1988) refers to mating dates of ladder snakes in captivity in Southern Spain as between May and July. In captivity with temperature and light/dark conditions controlled, mating occurred from mid April to early May (Butler, 1991; Dupres, 1991). Observed mating in Doñana occurred on 9 and 22 June, respectively. Moreover, on 24 and 25 June we found two ladder snake pairs in the field, suggesting they were in courtship.

The behaviour of both sexes during mating on 9 June followed the three stages that Gillingham (1979) defined in other species of the *Elaphe* genus. The male was put inside the female cage on 9 June at 10.00 hr, and apparently they showed no immediate interest in each other. At 13.00 hr the male moved close to the female and continuously touched her with his chin and tongue. The female responded to this stimulation by uncoiling and stretching out completely. The duration of this stage is unknown (in other species of the genus it lasts between 1 and 23 min.; Gillingham, 1979) as we were not able to observe the beginning, but after two minutes the individuals went to the second stage - "copulatory attempts" or TSCA - (Gillingham, 1979). This stage began with a coital bite from the male in the nape of the female, and a more intense rubbing with the whole body. During this stage the animals were stretched out (the male on top of the female), while both experienced rapid epidermic caudocephalic waves. The duration of this stage was 4 min. (between 1 and 43 min. in other species of *Elaphe*; Gillingham, 1979) and resulted in the insertion of the hemipenis in the cloaca of the female. The third stage or copulation was exceptionally long (40 min.) compared with other species of the genus (between 11 and 31 min.; Gillingham, 1979), finishing with the separation of the cloaca and releasing the female from the coital bite. During this stage the epidermic waves continued, while the individuals spun around raising their tails at an angle of 45° from the substratum. Later they remained almost motionless, with the epidermic waves coming slower. Finally, just before separation they experienced spasmodic shocks again from tail to head.

The copulatory behaviour of *E. scalaris* described here is similar to that noted by Dupres (1991) and that of *E. guttata* in the length of the first two stages and the existence of a "coital bite" (Butler, 1991). However, the length of the third stage in the Iberian species exceeded that of any of the species studied by Gillingham (1979) by almost ten min.

Three ovipositions occurred in the terrarium; one from the female of the first described courtship (female

1, hereafter) and kept in captivity since then; another, from a female captured on 11 July (female 2); and the last one from a female captured on 9 July (female 3). All the ovipositions lasted between two and four days.

Egg laying by female 1 took place on 25-28 July, starting 46 days after mating. According to information on specimens in captivity, the gravid period was considered to be between 41-71 days (Butler, 1991) and between 20 and 44 days (González de la Vega, 1988; Cheylan & Guillaume, 1993). The laying period in other areas was June-July in the centre of Spain (Pleguezuelos, in press), May-June and July in Southern France (Cheylan & Guillaume, 1993), and mid June to early July in strict captivity (Butler, 1991; Dupres, 1991).

During pregnancy, female 1 ate normally until 20 days before laying, when it began to shed. This pre-laying shedding was also recorded in strictly captive females (Butler, 1991; Dupres, 1991). During the week between shedding and starting to lay it did not feed, probably due to the physical impediment of the eggs on the digestive tract (Gregory & Stewart, 1975). From the moment laying began to two days later, the female remained hidden and coiled up next to the eggs, without eating. On leaving the nest it appeared very active, accepting food immediately.

Although the three clutches were maintained in the terrarium under the same "natural" conditions described above, only one hatching was observed, because the other two clutches failed. The average air temperature throughout incubation period inside the terrarium was $25.5^{\circ}\text{C} \pm 0.6$; (mean \pm SD, range 22-36°C). The oviposition dates and the sequence of laying from the terrarium clutches are showed in Table 1.

Clutch size data come from ovipositions observed in captivity and in the field; four clutches of ladder snakes were found inside abandoned rabbit warrens, hatched and presumably complete (Blázquez & Villafuerte, 1990).

Measurements of eggs were taken from 19 newly-laid eggs and from 23 well-preserved hatched eggshells from the field. The eggshell were humidified with a blend of water (70%) and alcohol (30%) to soften them before taking their measures.

Clutch size observed in the terrarium (Table 1) is rather constant around 12, perhaps because the females were similar in size, but the clutches found in the field, on 12 and 23 September 1988, and 19 September and 1 October 1989, were of 4, 5, 6 and 12 eggs, respectively. Total clutch size ranged between 4 and 13 eggs ($n = 7$), which is consistent with previous observations (Butler, 1991; Dupres, 1991; Cheylan & Guillaume, 1993; Pleguezuelos, in press).

The eggs of the ladder snake have a parchment-like appearance and are rounded and yellowish, with a delicate shell. Sizes of 19 just-laid eggs were (mean \pm SD) $50.9 \text{ mm} \pm 2.7$ (range 45.9-55.6 mm) long and $30.3 \text{ mm} \pm 1.5$ (range 26.8-33 mm) wide. The hatched shells

Female number	1	2	3
SVL (mm)	1070	1060	1050
Weight preoviposition	693	845	710
Date (July)	25 28	16 17	12 13
No. of eggs	10 1	10 2	11 2
Total clutch size	11	12	13

TABLE 1. Clutch size and sequence of laying in three *Elaphe scalaris* females.

were $44.2 \text{ mm} \pm 4.9$ (range 35-50.7 mm) long and $28.9 \text{ mm} \pm 2.4$ (range 22.7-33.1 mm) wide. The difference was significant in length ($t = 5.3$, $P < 0.001$) but not in width ($t = 1.9$, $P > 0.05$), probably because the newly laid eggs are more turgid. Other authors generally refer only to ranges, which usually correspond to eggs longer and thinner than those we found (González de la Vega, 1988; Cheylan & Guillaume, 1993).

The hatching of the clutch of 12 eggs occurred on 20-23 September, between 66 and 69 days after laying. The length of the incubation period, which is dependent on the temperature (Lillywhite, 1987; Burger, 1991), ranged between 51 and 55 days in Southern France (in nests maintained at temperature of 28°C; Cheylan & Guillaume, 1993); and between 52 and 63 days in captivity in Southern Spain (González de la Vega, 1988). Butler (1991) refers to a 50-day incubation period. The relatively longer incubation period observed in Doñana was probably due to daily fluctuating temperatures in the terraria.

The hatching began on 20 September when cracks appeared on two eggs; on 21 September eight had cracks, and in the two earlier the young could be seen beginning to emerge; on 22 September three young left their eggshell and on 23 September ten young were outside. One of them died that afternoon and the two remaining eggs failed to hatch. The healthy nine newborn spent the first few days together inside the nest, almost motionless and curled up against each other.

This situation changed after the first slough, which occurred in all the young between 9 and 13 days after hatching (Butler, 1991). The young left the nest to facilitate sloughing by rubbing against the ground, and finally scattered. This behaviour is similar to that observed in *Malpolon monspessulanus* (Hailey, 1982; Blázquez, 1993), but it had never been described for *E. scalaris*.

The average size of the nine young (75% of the clutch) at birth was 290 mm \pm 12.5 (range 280-315 mm) in SVL and 19.6 g \pm 2.2 (16-23 g) in weight. After the first slough, their weight was 19.4 g \pm 1.7 (range 17-22 g). The dead hatchling was smaller than the others at birth (SVL = 265 mm, weight = 9.5 g). The sizes of newborn snakes described here agree with those noted by Cheylan & Guillaume (1993) and Pleguezuelos (in press). Additional data were obtained from two juveniles captured in the field in April, after their first winter; they measured 320 and 340 mm in SVL, respectively.

Out of a total of 10 adult females captured during the potential gravid period (last week of May-first week of July) only four contained eggs, which implies that the proportion of females reproducing each year is low (approximately 40%) compared to other oviparous species of colubridae: 76.97% according to Seigel & Ford (1987), with only 20% of species showing less than 50% of females with oviductal eggs.

The number of gravid females and clutches of *E. scalaris* discovered during the favourable period of this study was rather low, in spite of the considerable number of traps set and active searches in the field. A small density of ladder snakes, their low reproductive rate in the area, and/or the fact that the species selects oviposition sites that we were unable to discover, may all explain the situation. In Doñana *M. monspessulanus* (Blázquez and Villafuerte, 1990) and other terrestrial colubrids (Blázquez, pers. obs.) usually select abandoned rabbit warrens to oviposit because there is a scarcity of suitable places, but surprisingly during the study we found few clutches of *E. scalaris* there. Probably the burrowing abilities of this species enables it to make holes under shrubs in less evident places.

As for the effort that breeding represents in terms of weight loss, the only information available refers to a female (female 1) of 1070 mm in SVL, weighing 693 g (100 g more than at mating time) a week before laying and 480 g just after laying (30.7% weight loss). Eleven eggs were laid weighing 223 g just after laying. The only other known reference to this subject comes from Cheylan & Guillaume (1993), which refers to a female 950 mm in SVL that after laying 10 eggs (260 g) had lost 46.6% of its weight.

Other parameters (clutch size, size of eggs and offspring) are within the normal variation range in oviparous terrestrial snakes of temperate zones (Seigel & Ford, 1987).

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