

Maternal care and defence of young by the plethodontid salamander *Speleomantes strinatii* (Aellen, 1951)

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Abstract. Egg brooding females of the North-west Italian Cave Salamander *Speleomantes strinatii* display a complex array of parental behaviours. Recently, prolonged skin contacts between mother and young were documented by means of infra-red video recording, up to 40 days after hatching. In this study the behaviour of two females attending their young in presence of a potential predator was experimentally tested. About seven days after hatching, two adult males of *S. strinatii* were introduced inside the terrarium, where two females (A and B) were both attending eight young. In one case, female A approached female B and her young, but was repelled away. The approaching male always elicited an aggressive behaviour of females that repelled the intruder by head pushes and bites. This behaviour, similar to the one displayed during egg brooding, is the first evidence of active defence of young in terrestrial salamanders.

Keywords. *Speleomantes strinatii*, infra-red video recording, post-hatching parental cares, young defence.

INTRODUCTION

Post-hatching parental cares provided by one or both parents have been documented in several species of amphibians (Wells, 2007). Anura display attendance, brooding, feeding, transport and defence of egg and tadpoles and transport of froglets (Lehtinen and Nussbaum, 2003; Wells, 2007). In Apoda, the female may provide food to the hatchlings through modified and specialized skin tissues (Kupfer et al., 2006). Aquatic Urodela display egg and larvae attendance (Nussbaum, 2003; Wells, 2007), while in terrestrial species such as *Plethodon cinereus* (Green, 1818) and *Aneides aeneus* (Cope and Packard, 1881) young remain in the proximity of the mother up to four weeks after hatching (Mathis et al., 1995). However, mother-young interactions were never described in detail to date. The first unambiguous evidence of post-hatching parental behaviour in terrestrial salamanders was recently documented in the North-west Italian Cave salamander *Speleomantes strinatii* (Aellen, 1958)(Oneto et al., 2010). In that study, a cave salamander brood-

ing female was recorded by infrared (IR) video technique in a terrarium kept in complete darkness inside an underground laboratory. The two hatchlings remained near and often climbed on the female back, resting motionless for hours without any contact with the substrate (Oneto et al., 2010, fig. 1F). This behaviour, comprising repeated physical contacts between female and young, lasted for about 40 days, and during this period the female rarely abandoned the nesting site. The female's post-hatching parental behaviour consisted in attendance at the nesting site and in long-lasting and repeated skin contacts with her young. This was interpreted as a true parental behaviour producing a selective advantage for the offspring, by providing protection against fungal or bacterial infections and by increasing skin hydration of the small-bodied young (Oneto et al., 2010). A possible protective role of the attending female against potential predators was also suggested, but no experimental evidence could be provided. This research, a prosecution of the previous study by Oneto et al. (2010), assessed experimentally the role of the female cave salamander in presence of her young, by introducing a conspecific potential predator inside the terrarium.

MATERIALS AND METHODS

Study species

The North-west Italian Cave Salamander, *Speleomantes strinatii*, is a completely terrestrial plethodontid endemic to SE France and NW Italy (Lanza, 2006). This species is found from the sea level up to about 2470 m a.s.l. along streams, on humid rocky outcrops, in humid forest floors and also in subterranean natural or artificial habitats (Lanza et al., 2005; Renet et al., 2012). Sexually mature males are recognized by the presence of a mental gland lacking in females and in immature individuals. In the wild, nest sites and egg clutches of *S. strinatii* remain unknown (Lanza, 2006), but females were observed laying 6-14 relatively large eggs in captivity (Durand, 1970; Salvidio et al., 1994; Oneto et al., 2010). During egg brooding the female coils around the clutch, maintaining skin contacts and actively defends it against approaching potential predators (Durand, 1970; Oneto et al., 2010).

Experimental setting and infrared recording

This study was conducted in the Biospeleological Station of San Bartolomeo (390 m a.s.l.) near the village of Savignone, 25 km N of the town of Genova (Liguria, NW Italy). An experimental Plexiglass terrarium (70×50×30 cm) was kept in semi-natural conditions, and, apart from movement restriction, all the experimental animals were exposed to the same environment as the salamander population living in the cavity (Salvidio et al., 1994). The terrarium was located at about 20 m from the cavity entrance, where solar illumination is lacking (0.0 Lux, measured by Deltha Ohm Luxmeter HD 8366, Padova, Italy), annual mean air temperature is 9.7 °C (range 6.7-12.5), and relative air humidity is 95% (Salvidio et al., 1994). Salamander behaviour was recorded by a hard-wired waterproof camera Sony Su-



Figure 1. Physical contacts occurring among female A and her young immediately after hatching. In front of the female the egg envelop is still present (photo by Fabrizio Oneto).

perHAD™ equipped with a Charge Coupled Device (CCD) optical sensor and with a built-in infrared (IR) 12-led illuminator (RE-BCC6L, Digital Surveillance Equipment, DSE Torino, Italy). The camera was suspended 1.2 m over the terrarium and connected to a 10-m distant PC. During the experiments, video recording was conducted in continuous with 12.5 frames per second. The video sequences were saved in the PC as 60-min packages and were visualized at normal speed or in slow-motion by a commercial software provided by DSE (Torino, Italy). The quality of the images was relatively low, but the moving sequences allowed to interpret the behaviours unequivocally. In November 2010, two apparently gravid females (A and B) were introduced inside the terrarium. IR recording began in October 2011, after the hatching of the young. All young were born by October 15 and 17 for clutch A and B, respectively. After about seven days from hatching, two adult males (X and Y) were introduced in the terrarium for a period of three days and their interaction with the resident females was recorded in complete darkness. Overall, 840 hours of recording were analysed.

RESULTS

Interactions between female and young

The analysis of the IR video recordings of the two females and their young confirmed the previous observations, conducted on one single female attending two young (Oneto et al., 2010). Both females shared the nesting site for about 30 days with their young, that repeatedly entered in physical contact with their moth-

er, climbed on it's body and rested motionless without any physical contact with the substrate for hours. Often, this behaviour was displayed by all young simultaneously (Fig. 1). These observations are a confirmation of the existence of intimate physical contacts between mother and her young in *S. strinatii* (Oneto et al., 2010).

Interactions between the attending female and a possible predator

On October 17, 2011 female B left her nesting site and approached female A and her young. The resident female touched with her head the intruding female, probably pushing away or maybe biting. After a contact lasting about 15 sec female B walked away and returned to her young.

Soon after being introduced in the terrarium, on October 25, both cave salamander males approached the two attending females. When the intruder approached and entered inside the nesting site, the attending female became vigilant, looking towards the intruder and walking to intercept and actively repel the male by head contacts, pushes and in some cases bites. In only one case, the male started the contact by walking upon the motionless female that reacted biting the male trunk and repelling the intruder (see Table 1). It was not easy to distinguish simple physical contacts from bites, because of the perpendicular angle of the video camera, but in some cases the contact between the interacting salamanders caused a rotation of the male body, who was “pulled” by the female (see Fig. 2, S5), or a rapid body twisting of the male that tried to free himself from the female's grip. These

Table 1. Synthesis of *Speleomantes strinatii* agonistic interactions between females attending their young and intruding male. All the interactions, with the exception of (*) were started by females. Bites were delivered only attending by females.

	Contact (N)	Duration time (sec)	Notes
Female A	1	13	1 bite
	2	7	-
	3	13	1 bite?
	4	60	-
	5	11	1 bite
	6	3	1 bite
Female B	1	7	-
	2	109	-
	3	15	-
	4	8	-
	5	149	1 bite
	6	11(*)	1 bite

(*) The duration time starts when the female bites the male



Figure 2. Stills (S1-S8) from infrared video footage recorded on October 25, 2011, showing the interactions between female A and intruding male Y. In still (S1) the following letters indicate: A and B the attending females; (a1) and (b1) their young; X and Y the males. This sequence is described in detail in the text.

agonistic behaviours were observed 6 times in both females, for a total of 107 and 299 sec respectively (Table 1). In all cases, the female forced the intruder to exit from the nesting site. An example of these interactions is given in Fig. 2, that was composed by eight video stills (S1-S8) taken from the full video footage. This sequence refers to the encounter occurred on October 25, 2011, between the two salamanders A (female) and Y (male). The complete action lasted about 3 min and is here described in detail:

- S1) at 16h 24m 57s: female A walks from the young (a1) on a straight line towards the intruder Y;
- S2) at 16h 25m 04s: female's head "touches" the intruder's neck, the physical contact was maintained for about 12 sec; at time 16 25 15 the two individuals detached;
- S3) at 16h 25m 26s: the female engages again in a physical contact that lasted 58 sec;
- S4) at 16h 26m 29s: the female bites the intruder's right hind-leg, pulling it with energy;
- S5) at 16h 26m 36s: the male is half turned on his right side by the pulling female;
- S6) at 16h 26m 46s: the male free himself and moves away from the female;
- S7) at 16h 27m 11s: the female "touches" (probably bites) the posterior part of the male's trunk;
- S8) at 16h 27m 17s: the male walks outside the nesting site, and the female-male interaction ends.

DISCUSSION

This study, as the one by Oneto et al. (2010), proves the importance of recording the social behaviour of *Speleomantes* salamanders in semi-natural settings and not in completely artificial laboratory conditions. In this study, IR video recording were obtained in total darkness, without disturbing the focal individuals, unless by restraining their movements inside the terrarium. Indeed, some of the previous described parental behaviour of *S. strinatii* were obtained in completely artificial settings and therefore were only partially described (Durand, 1967, 1970). The post-hatching parental behaviour recorded during this study (e.g. complete skin contacts of young that actively climbed on the mother), confirms the observations reported by Oneto et al. (2010) on a single female attending two young. Moreover, the female interactions occurring several days after hatching with a potential predator (i.e. an adult male introduced in the terrarium) gave new insights in the parental behaviour of the species. Both experimental females repeatedly displayed an aggressive behaviour towards the male intruder that was intercepted, pushed, sometimes bitten and always repelled away from the young. It is interesting to note, however, that in all cases the attending female was aggressive towards the intruder individual (male or female) but not vice versa. These results are of interest because territorial agonistic behaviour has never been demonstrated in *Speleomantes* (Zanetti and

Salvidio 2004; Sguanci et al., 2010) and therefore aggressive behaviour seems to be limited only to the defence of the egg clutch (Durand, 1970; Oneto et al., 2010) and young (this study). The presence of eggs and/ or young seems to give to the female a social status that is perceived as dominant by other approaching individuals of the species.

In any case, the agonistic behaviour recorded in this study clearly exposed the female to the possible risk of being bitten, wounded and possibly impaired by the approaching predator, that was an adult salamander of similar size. Costly behaviours, in terms of energy investment and of risk-taking, fulfil Trivers's (1970) definition of parental care, demonstrating that the attending *S. strinatii* female actively defended its young, up to about 10 days after hatching. These results provided the first evidence of "young defence" in Urodela (Jaeger and Forester, 1993; Duellman and Trueb, 1994; Nussbaum, 2003) and enlarge our knowledge on salamander parental behaviour that appear much more complex than previously assumed (Mathis et al., 1995).

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