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Intraspecific oophagy in *Hierophis viridiflavus* (Serpentes: Colubridae) during oviposition in a controlled environment

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Abstract

Intraspecific oophagy in *Hierophis viridiflavus* (Serpentes: Colubridae) during oviposition in a controlled environment. Following the observation of adult pairs of the Western Whipsnake, *Hierophis viridiflavus,* in a controlled environment, two distinct but related phenomena were observed: egg deposition and predation of freshly laid eggs by the male. Data about deposition, number and morphology of the eggs, hatching and offspring are presented and compared with the literature. The episode of oophagy is described, confirming the inclination to predate snake eggs and intraspecific oophagy in this species.

Keywords: Egg deposition, Intraspecific predation, Reproduction, Snakes, Western Whipsnake.

Oofagia intraespecífica em *Hierophis viridiflavus* (Serpentes: Colubridae) durante a ovipostura em um ambiente controlado. Após a observação de pares adultos de cobra-chicote-do-oeste, *Hierophis viridiflavus*, em um ambiente controlado, dois fenômenos distintos, mas relacionados, foram observados: deposição de ovos e predação pelo macho de ovos recém-depositados. Dados sobre a ovipostura, o número e a morfologia dos ovos, a eclosão e os filhotes são apresentados e comparados com os dados da literatura. O episódio de oofagia é descrito, confirmando a tendência dessa espécie de predar ovos de serpentes e praticar a oofagia intraespecífica.

Palavras-chave: Cobra-chicote-do-oeste, Ovipostura, Predação intraespecífica, Reprodução, Serpentes.

Introduction

Predation of bird eggs by snakes is widespread in hundreds of species, especially among colubrids (Schulz 1996). For most of these, eggs and nestlings are part of a vast food spectrum, while some species have a specialized diet based on bird eggs (Gans 1959, Bates and Broadley 2018).

Predation of reptile eggs by snakes is well known, but observations in the wild in European species are almost exclusively limited to the predation of saurian eggs. This phenomenon has been widely observed in *Coronella austriaca* Laurenti, 1768 (Galán 1988, 1991, Galán and Fernández Arias 1993, Amat 1998, Moreira *et al.*

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2011, Lunghi *et al.* 2015, A. Paterna unpubl. data), the congeneric *Coronella girondica* (Daudin, 1803) (Luiselli *et al.* 2001), *Zamenis scalaris* (Schinz, 1822) (Pleguezuelos *et al.* 2007), and the boid *Eryx jaculus* (Linnaeus, 1758) (Faraone *et al.* 2017). Data about predation on reptile eggs in snakes arise mainly from the dissection of roadkilled individuals or manipulation of individuals in which regurgitation is voluntarily or involuntarily induced.

Intraspecific oophagy in colubrids has been confirmed almost exclusively in captivity (Mitchell and Groves 1993), and the only European species in which this phenomenon has been validated with certainty is *Z. scalaris* (Laferrere 1970). Snake egg predation has been reported in *Hierophis viridiflavus* (Lacépède, 1789). A roadkilled adult male of this species that had four snake eggs in its stomach and intestine was found near a communal nesting site of the same species (Capula and Luiselli 1995). The ingested eggs were assumed to belong to the same species of the predator. A similar case involving *H. viridiflavus* has been recently reported by Bonnet *et al.* (2021) from western France.

The western whipsnake *Hierophis viridiflavus* is a colubrid snake widely distributed mainly in Italy and southern France. Within this species, two main phenotypes generally corresponding to the two subspecies are present: the nominal *viridiflavus,* with a characteristic dorsal pattern of yellow and black tones, and *carbonarius,* in which the adult specimens exhibit melanism. The northern apex of the distribution of this species reaches the southern portion of the Metz province, France, for *viridiflavus* and the southern Alpine arch for *carbonarius* (Kreiner 2007). Established populations outside their natural distribution have been recorded in western Switzerland in the Cantons of Vaud and Valais (Meier *et al.* 2022). In 2017, a small population of *H. viridiflavus* was found in a waste disposal area in southwestern Germany. The origin of this allochthonous colony is thought to have been the accidental introduction of some individuals through waste delivery from Italy, specifically from the province of Napoli. The local administration for the protection of nature opted to capture as many specimens of *H. viridiflavus* as possible, providing private breeders or organizations with the opportunity to adopt the collected specimens (H. Laufer, pers. comm.). This study is based on the data obtained from observations of the captive specimens of *H. viridiflavus* originating from the allochthonous colony and maintained at OPHIS Museo Paleontologico e Centro Erpetologico.

Materials and Methods

Four specimens of *Hierophis viridiflavus* recovered by the company Büro für Landschaftsökologie LAUFER (Offenburg, Germany) were adopted and kept at OPHIS Museo Paleontologico e Centro Erpetologico. These individuals were captured on 12 and 13 April 2022 and delivered to OPHIS on 25 April 2022. After capture, the specimens were kept separately. Two pairs were formed based on the size of the specimens and housed in two terrariums (100 × 60 × 50 cm) set up in a “naturalistic” way, with branches, bark, plants, and baltic peat as substrates. The snakes were observed daily from their arrival at the herpetological center to brumation (April–October 2022). Weight of the snakes was recorded periodically each month during their activity period. Throughout the observation period, the specimens refused any food that was offered them, and they were force-fed every seven days with thawed young *Mus musculus* Linnaeus, 1758. After 47 days in captivity oviposition occurred. Egg mass and size data were collected using an electronic scale and caliper, respectively. Eggs were transferred to an incubator, built with insulating materials, and warmed using an electronic thermostat and further controlled by digital probes. The incubator dimensions were 38 × 58 × 36 cm, and within it, eggs were contained in two plastic boxes (12 × 12 × 6 cm) with a transparent lid and moist vermiculite as medium.

Results

Egg Deposition

Throughout the breeding period of the housed specimens, there was a visible increase in the mass of all individuals with the exception of one male, which was distinguished from the beginning by its larger size. The female who shared the terrarium with this male molted on 31 May 2022. After this event, the possible presence of eggs in the female was noticed, although no copulation was observed. A closed container measuring 27 × 18 × 8 cm, with a circular entrance of 4 cm to allow the snake to enter, was prepared with moistened vermiculite for possible egg deposition.

On the morning of 13 June 2022, exactly seven weeks after the arrival of the specimens at the OPHIS Herpetological Centre, the female began laying eggs not in the dedicated container, but under bark that was used as a hiding place (Figure 1). At 09:00 h her third egg was expelled. Photographs were made quickly to minimize disturbance. Two hours later, the female was in the same position with eggs remaining inside her, but the eggs that had been laid could not be found. The terrarium, the nesting container, and the entire substrate were searched, but no traces of the eggs were found. The co-inhabitant male was then removed from the terrarium to determine if he had ingested the eggs. By manipulation it was possible to make the male regurgitate four intact eggs and one damaged egg (Figure 2). These were cleaned and placed in a separate container. The female was reintroduced to the container dedicated to egg-laying inside the terrarium, where, after five hours from the first check, she laid three additional eggs. Two days before egg deposition the female weighed 144 g; after oviposition she weighed 106 g.

Figure 1. Adult female *Hierophis viridiflavus* originating from the southwestern Germany allochthonous colony at the time of deposition at OPHIS Museo Paleontologico e Centro Erpetologico.

Figure 2. Adult male *Hierophis viridiflavus* originating from the southwestern Germany allochthonous colony after regurgitating the eggs freshly deposed by the conspecific female at OPHIS Museo Paleontologico e Centro Erpetologico.

Eggs

The clutch consisted of eight eggs. Three were laid by the female in the appropriate container (Figures 1 and 3A), while five, of which one was damaged, were recovered after regurgitation by the male (Figure 2). Weight of each egg was 6–7 g, lengths were 3.7–4.2 cm in length, and diameter1.5 cm. The eggs were white with a longitudinal striae-like pattern. Some eggs exhibited asterisk-shaped concretions inside a smooth circular recess that interrupted the rough pattern of the surface (Figure 3).

The uneaten and regurgitated eggs were placed in two separate containers with moist

Figure 3. (**A**) Details of the last three eggs deposited by the female *Hierophis viridiflavus* originating from the southwestern Germany allochthonous colony at OPHIS Museo Paleontologico e Centro Erpetologico. (**B**) Hatching of two *Hierophis viridiflavus* at OPHIS Museo Paleontologico e Centro Erpetologico. Clutch laid by a female originating from the southwestern Germany allochthonous colony.

vermiculite and placed inside the incubator at a temperature between 25.5 and 27.5°C. Three weeks after deposition, the regurgitated eggs were removed from the incubator following deterioration. This process was already evident after a few days but was slowed, in vain, by cleaning the surfaces of the eggs. During the incubation period, the three remaining eggs increased in size, expanding mainly transversally.

Hatch and Offspring

On the 56th day of incubation, two eggs were cut, and the hatchlings (Figure 3B) emerged from the eggs on the same day. The same was done for the third specimen on the following day. The hatchlings shed for the first time eight days following hatching. The hatchlings each weighed 5 g and measured 28, 28.5, and 30 cm. The male was the shortest and had the darkest pattern (Figure 4). The head was black with yellow lines; dorsum was greenish/grayish with small darker rectangles. The chin was white, fading to a light aqua-green on the ventral and subcaudal scales. The iris was orange in contact with the pupil, becoming darker at its outer edge.

Discussion

Reproduction

Information on the reproduction of *Hierophis viridiflavus* is plentiful in the literature. However, in most sources, descriptions of the eggs, their appearance, dimensions, and clutch size are identical, without any specific data (Bologna *et al.* 2000, Vanni and Nistri 2006a, b). The only data about gestation are found in Di Tizio *et al.* (2008), where the authors stated that the time from mating to egg-laying is 20–30 days. It is assumed that the data concerning the reproduction of *H. viridiflavus* found in these sources, identical and non-specific, all originated from Bruno (1984, 1998).

Original data on egg numbers are 5–10 (Luiselli 1995), 4–7 (Capula *et al.* 1997), 3–9 (Filippi *et al.* 2007), and 4–11 (Zuffi *et al.* 2007).

Figure 4. Portrait of a male *Hierophis viridiflavus* hatchling after its first shed at OPHIS Museo Paleontologico e Centro Erpetologico. Hatched from a clutch laid by a female originating from the southwestern Germany allochthonous colony.

Unfortunately, no information on morphology, features of the eggs, or times and methods of incubation were reported. Only two photos of eggs or hatchlings have been reported (Ferri 1992, 1993, Ferri and Soccini 2002).

In the present study, it was not possible to determine with certainty whether the fertilization of the eggs occurred in the wild or in captivity, as no copulation in captivity was observed. Observation of copulation in the controlled environment would have been difficult because the snakes are extremely elusive and rarely found outside their hiding places. The subjects in this study came into contact with each other for the first time 47 days before deposition, the earliest possible date on which the copulation in captivity could have occurred. Gestations with time frames identical to this have been observed in *Zamenis longissimus* in three consecutive years, where times of gestation were 48, 48, and 49 days, respectively (unpubl. data).

Egg-laying took place 13 days after the pre-deposition molt and resulted in the female losing just over 25% of her weight. The eggs were very similar in appearance to those of the sympatric Aesculapian snake *Zamenis longissimus,* although smaller (up to 5.5 cm long and 2 cm wide in *Z. longissimus,* personal observation). The appearance and size of the hatchlings are similar to those reported in a large number of *H. v. carbonarius* from a communal nesting site in Abruzzo (Paterna 2015).

Oophagy

Intraspecific oophagy in colubrids has been confirmed almost exclusively in captivity (Mitchell and Groves 1993), and the only European species in which this phenomenon has been documented with certainty is *Zamenis scalaris* (Laferrere 1970). In the present study, it was possible to document and confirm such behavior as the predation of conspecific eggs in *Hierophis viridiflavus* because the event took place in a controlled environment.

It is not known whether the instinct to swallow eggs is affected by captivity, but the male obviously recognized snake eggs as a food item. To date, all specimens of *H. viridiflavus* housed at the OPHIS Herpetological Centre have refused any food that has been offered to them (mice, baby quails, baby rats, fish) and have been force-fed for their sustenance. However, when oviposition took place in this study, the male found and swallowed the eggs in a short time. This demonstration that the male did not feed on the eggs randomly is confirmed by an analogous event, also occurring in a controlled environment, in a pair of *H. viridiflavus carbonarius* bred by researchers Tomaž Jagar and Erika Ostanek in 2022 (personal communication).

Adult *H. viridiflavus* in nature have a very broad food spectrum (Filippi *et al.* 2003, Lelièvre *et al.* 2012, Mondino *et al.* 2022), although there is no shortage of documented episodes of ophiophagy (Capula *et al.* 2014) and cannibalism. This broad food spectrum makes *H. viridiflavus* one of the most opportunistic European snakes and may suggest that the males of the two above-mentioned episodes likely fed on the eggs produced by the co-occupying females opportunistically, not distinguishing whether the deposited eggs belonged to their own species or not. An alternative hypothesis is that the males of this species are inclined to feed on the eggs of conspecific females (where they come into contact with them) for population control purposes. Males of *H. viridiflavus* are strongly territorial, and predation on conspecific eggs would reduce the number of rivals in the territory. This also raises the question of whether *H. viridiflavus* could differentiate its own eggs from those of other conspecifics and from other sympatric species.

Moreover, it must always be considered that the stimuli and stress to which these animals are subject in the wild are completely different from those in captivity, and, in the case of this species, much of its ecology remains unknown. Further investigations should be made in the future because the impact of such behavior is potentially influential not only in the areas where the species naturally occurs but also in introduced populations, where the opportunistic nature of the phenomenon could be interpreted as an additional indication of the invasiveness of the species in the allochthonous localities.

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