

**DESMOGNATHUS CONANTI (Spotted Dusky Salamander). NESTING UNDER MANMADE COVER OBJECT.** At 1550 h on 17 August 2019, I overturned a manmade cover object colloquially called a “cookie” (a small circular piece of wood designed to aid in herpetological surveys) in a muddy streambank near a pond in a mesic hardwood forest in Knox County, Tennessee (35.956816 4°N, -83.868139°W) and found a female *Desmognathus conanti* guarding a clutch of 18 eggs. Also under the cover object were three conspecific adult *Desmognathus conanti* and one adult *Eurycea cirrigera*. The cover object was about 30cm in diameter and loosely embedded in the ground. In the direct vicinity were multiple natural cover objects such as small logs, rocks, and moss. In an assessment of the nesting cover object choice

of *Desmognathus fuscus* (Hom. 1988. Journal of Herpetology. 22: 247-249) suggests that females will preferentially lay eggs under moss while also laying eggs under logs and rocks. To the best of my knowledge this is the first recorded observation of a salamander in the genus *Desmognathus* using a manmade cover object as a nest site. It is also interesting to note that, though they often had adult *Desmognathus*, other cover objects in the direct vicinity had no nests. This may suggest that manmade cover objects could be preferentially chosen by female *Desmognathus* when available, though more research would be necessary to confirm this hypothesis.

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FIG. 1. A female *Desmognathus conanti* guarding her brood of eggs.

**PLETHODON WELLERI (Weller's Salamander). EARLY COURTSHIP.** At 1428 h on 16 March 2020, I overturned a trailside log along the Appalachian Trail between Indian Grave Gap and Beauty Spot (36.112952, -82.359852, elevation 1010 m) to reveal two *Plethodon welleri* in close contact. One individual was a gravid female while the other was a male with well-developed nasolabial projections, a temporary physical trait the male salamander develops during the breeding season (Organ 1960). These two individuals were likely a mating pair given the enhanced secondary sexual characteristics and position; however, I was unable to confirm the presence of a mental gland on the male or the presence of eggs in the female due to concerns of disturbing the pair. *Plethodon welleri* are considered to be both a fall and

spring breeding species with evidence of courtship documented in April and October from both Mt. Rogers, NC and under lab conditions (Organ 1960. *Copeia* 1960(4): 287-297; Thurrow 1963. University of Kansas Publications 44:87-108). To my knowledge this is the earliest recorded breeding event for *Plethodon welleri* and may stem from an unseasonably mild and wet winter. As anthropogenic climate change progresses, anomalously mild winter conditions will likely become increasingly common and may lead to the continued documentation of earlier courtship in *P. welleri*.

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Fig. 1. A breeding pair of *Plethodon welleri* from northeast Tennessee.

**DIADOPHIS PUNCTATUS** (Ring-Necked Snake). **COLORATION.** On 27 October 2019, we discovered a juvenile *Diadophis punctatus* underneath a discarded scrap of wood at Sandy Creek Nature Center in Clarke County, Georgia (33.9889°N, 83.3719°W; WGS 84), and we deposited a photo voucher on HerpMapper (HM 290368). This individual lacked nearly all the yellow pigment typical of *D. punctatus* in the region, with the venter and nuchal collar instead being a pale white (Fig. 1). This coloration appeared to be permanent, as the snake showed no signs of impending ecdysis (e.g., opaque eyes or lightened dorsal coloration). Other individuals of this species that we have encountered at this location exhibited typical coloration, and relatively few reports of similar coloration abnormalities are present in published literature. Jolley et al. (1983. Herpetol. Rev.

14:119–120) noted a *D. p. arnyi* from Missouri that likewise lacked “nearly all orange, yellow, and red pigmentation”, but described its nuchal collar as “obscure” and its venter as “light metallic gray”. These authors referred to this condition as “acarotenoidic”, although it might be more commonly referred to as “axanthic”—a general term used to describe the absence of yellow (and sometimes red or orange) pigments. Brust et al. (2020. Herpetol. Rev. 51:142) described an axanthic *D. p. edwardsi* from West Virginia which very closely matches the appearance of the individual we observed.

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FIG 1. An axanthic *Diadophis punctatus* from northern Georgia. PHOTO BY TODD W. PIERSON.

***NERODIA SIPEDON*** (Northern Water Snake). **SHEDDING.** In the summer of 2019, we collected 22 Northern Watersnakes from Cheatham, Putnam, and Rutherford counties in Tennessee (TWRA Scientific Collection Permit #1907). Snakes were placed into captivity at Middle Tennessee State University (IACUC approval 19-3012). The enclosures provided for these animals consisted of 66.24 L plastic storage totes (66 × 34 × 41 cm) with ventilation holes, hide box, climbing branch, and water dish. The experimental chamber housing the snakes was maintained at 23 °C with a 12 h light/dark cycle generated by white overhead lights. The daytime period began at 0800h and ended at 2000h each day. Snakes were housed in captivity to facilitate an experiment involving timeseries data. As part of this experiment, we observed snakes continuously (24 h/day) six days a week for a period of approximately three months.

During daytime hours, an observer was present in the experimental chamber who would continuously monitor snakes for signs of shedding behavior. Daytime observation did not involve active disruption of snake behavior as the provided hide boxes were translucent. This permitted observers to view individual snakes through the transparent walls of the enclosure without physical disruption or touching of the animal. During nighttime hours, an observer would enter the dark experimental chamber and observe each snake individually using a red headlamp positioned several centimeters from the side wall of the enclosure. Red light was chosen as it is generally considered to be less disruptive to sleeping or nocturnal animals than white light (Ouyang et al. 2017. *Global Change Biology* 23:4987–4994). We repeated this process once every thirty minutes for the extent of each nighttime period. Each nighttime observation period took between 5–10 minutes. During this time, each snake was individually inspected for between 30 seconds and 2 minutes without any physical disturbance. In the later part of the experiment, some snakes were exposed to intermittent red light via an

automated lighting rig to mimic observation periods by human observers.

Over the course of this experiment (which lasted from 31 May 2019 to 21 August 2019), ten shedding events took place among the captive snake population. Prior observations of snakes maintained in captivity for long periods of time have not reported a bias for nocturnal or diurnal shedding behavior (Lamonica et al. 2007. *International Journal of Morphology* 25:103–108). However, we observed eight shedding events that occurred during daytime hours and two shedding events that occurred during nighttime hours. Previous work has shown that ambient temperatures effect overall rates of ecdysis in snakes, but little is known about the mechanisms that influence a snake already in a pre-shed state, to begin the shedding process (Semlitsch. 1979. *Society for the Study of Amphibians and Reptiles* 13:212–214).

Between the dates of 25 June 2019 and 15 July 2019, seven shedding events took place. All of these shedding events, except one, took place during the daytime observation window. The one nighttime shedding event which took place during this period occurred over the weekend of 4 July 2019 when nighttime observation was temporarily suspended, hence, this snake was not exposed to intermittent red light, via observer activity, on the night of the shedding event. It was determined that this animal had shed when the lights were off as routine animal wellness checks were still conducted that evening (2000h, during which no shed was found) and the following morning (0800h, during which the shed was found). This led us to hypothesize that nighttime observation activities, particularly exposure to light, may alter snake shedding behavior.

To investigate this, we constructed a lighting rig that utilized red lights (n=2 light bulbs, UTILITECH Model #YGA08A54-8W-RED) placed 15 cm away from the enclosures and interval timers to automatically expose snakes in a pre-shed state (development and subsequent loss of cloudy eyes) to red light throughout the

night. The lights of this system came on for 30 minutes and turned off for 30 minutes in a repeating cycle throughout the nighttime period. We utilized this lighting rig to expose two snakes exhibiting a pre-shed state to red light without other observer activities. One animal was exposed to light using this system for one night before shedding during the day on 16 July 2019. Another animal was exposed to light using this system for two nights before shedding during the day on 17 July 2019. Thus, both snakes exposed to this automated lighting system shed during daytime observation hours.

The final shedding event recorded during this experiment occurred on the night of 20 August 2019. This was the last night before the experiment was concluded and, like the weekend of 4 July 2019, nighttime observation was suspended. As with the previous nighttime shedding events, health checks were conducted the evening before and the morning after the shedding event took place. This allowed us to determine that the shedding event had occurred when the overhead lights were off in the experimental chamber.

In our observations, every snake that shed during the day had been exposed to intermittent red light the night before the shedding event took place. Alternatively, the two snakes that shed at night had not been exposed to intermittent red light on the night that they shed. Recent research investigated the expression of three visual pigment genes across snakes and suggested that most snakes exhibit cone dichromacy resulting in the lack of an ability to perceive the color red (Simões et al. 2016. *Molecular Biology and Evolution* 33:2483–2495). However, Natricine snakes possess vision which is sensitive to wavelengths of light which correspond with the color red (Fenwick et al. 1992. *Journal of Comparative Physiology A* 170:701–707). Consequently, when exposed to a red light in an otherwise dark room, study snakes would perceive the illumination caused by the light

source. From the perspective of the snake, it might appear as a yellow light source rather than red.

Our observations suggest that intermittent exposure to red light may have an effect on when snakes begin the shedding process. Northern Watersnakes are known to display crepuscular activity cycles during summer months in some parts of their range (Ernst et al. 2012. *The Herpetological Bulletin* 121:23–28). Other members of the genus *Nerodia* display varying degrees of nocturnality depending on season (Mushinsky & Hebrard 1977. *Canadian Journal of Zoology* 55:1545–1550). Additionally, snakes are generally considered to be more vulnerable to predation during the shedding process (Loughran et al. 2015. *Northwestern Naturalist* 96:156–160). Thus, study snakes may have preferentially shed at night. However, when nighttime disturbance was induced, via intermittent exposure to red light, study snakes may have switched to shedding during the daytime, when only passive observation occurred. Further study may elucidate if shedding behaviors can be modified via exposure to distinct lighting regimens.

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TABLE 1. Overview of snake shedding behavior with regard to lighting exposure.

Red Light Exposure	Shedding Window	Exposure Method	Snake ID	Date	Shed Start Time
Yes	Diurnal	Observer Flashlight	C4	25-Jun	1530h
			E5	28-Jun	1230h
			E7	28-Jun	1300h
			E6	4-Jul	1000h
			E10	9-Jul	0900h
			C7	15-Jul	0930h
Yes	Diurnal	Lighting Rig	C10	16-Jul	0900h
			C4	17-Jul	1200h
No	Nocturnal		E9	7-Jul	2000h–0800h
			E10	20-Aug	2000h–0800h