Reintroduction of the Eastern Indigo Snake (*Drymarchon couperi*) into Conecuh National Forest

2008-2011 Final Report

submitted to

The Alabama Department of Conservation and Natural Resources

and

The Orianne Society

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Introduction

Terminology

Using translocation as a conservation tool to establish a new population or enhance an existing population has been used with varying success since the 1970s (Germano and Bishop 2008). Terminology pertaining to species translocation or reintroduction efforts may be unclear, and multiple definitions may apply to various facets of a translocation project. The following terms, as defined below, will be used in this report.

Translocation – Translocation, as originally defined by the IUCN (1987), is "the movement of living organisms from one area with free release in another." An expanded definition is "the intentional release of individuals of a species at a *within-range* location different from their capture location in order to "establish, reestablish, or augment a population" (Griffith et al. 1989; Tuberville et al 2005). Thus, as broadly defined, translocation applies to the current eastern indigo snake project in which a population is being re-established. "Reintroduction" falls under the general definition of "translocation" but with a more refined application.

Reintroduction – As used by Reinert (1991) and the IUCN (1998), a reintroduction is defined as "the intentional release of individuals of a species into an area formerly occupied by that species" and "an attempt to establish a species [population] in an area of historical distribution from which it is extinct [extirpated]." This latter definition conforms to the original as presented by the IUCN (1987): "Re-introduction of an organism is the intentional movement of an organism into a part of its native range from which it has disappeared or become extirpated in historic times as a result of human activities or natural catastrophe." For clarity, Armstrong and Seddon (2007) recommend following the original IUCN 1987 definition. The release of immature eastern indigo snakes onto Conecuh National Forest represents a reintroduction as the general belief is that the species has been extirpated from the region for several decades.

Repatriation – The term "repatriation" is found in Dodd and Seigel (1991) and defined as "the release of individuals of a species into an area normally or currently occupied by that

species." From the Merriam-Webster Dictionary the definition is given as "to restore or return to the country of origin, allegiance, or citizenship" and, to extend this to a species, the implication is that the individuals removed from a site are returned to that site. For example, the Georgia females from which eggs have been obtained were repatriated upon their return to their point of capture.

Relocation – Dodd and Seigel (1991) define relocation as "displacement of wild-caught animals from their habitat to avoid immediate threats such as development"; relocation does not apply to the current project.

Augmentation (supplementation, reinforcement) - This is the process of adding additional individuals to an existing population (Fischer and Lindenmayer 2000; IUCN 1998). Although multiple releases of eastern indigo snakes are scheduled over a period of years each annual release is considered to be a reintroduction, not augmentation, as the currently released snakes have yet to achieve the status of a viable or stable population.

Head-starting – Head-starting is the rearing of hatchlings (either from natural nests or eggs incubated in the lab) in captivity until they have outgrown their period of greatest vulnerability to predators, thus conferring increased survivorship (Buhlmann et al. in review). The releases of eastern indigo snakes in 2010 and 2011 may be considered to have been head-started but for reasons other than stated above, i.e. the need of growth to implant transmitters although an additional benefit has been a reduction of vulnerability to predators.

Project Background

The historic range of the eastern indigo snake (*Drymarchon couperi*) includes the entirety of Florida, southern Georgia, and the southern tier of counties in Alabama. The eastern indigo snake is most often associated with landscapes that include longleaf pine forest-sandhills-wetlands habitat matrix (Godwin 2004; Moler 1992; Mount 1975; Stevenson et al. 2008) within the longleaf pine ecosystem, an ecosystem now recognized as one of the most imperiled in the southeastern United States (Frost et al. 1986; Noss 1989; Stout and Marion 1993). Within these habitats, adult indigo snakes utilize gopher tortoise (*Gopherus polyphemus*) burrows as cool

season refugia and nest sites (Stevenson et al. 2003). In both Alabama and Georgia the gopher tortoise has experienced both population declines and local extirpations due to habitat alteration and degradation (Aresco and Guyer 2004; Birkhead and Tuberville 2008), and tortoise declines may possibly have been a factor in declines of the eastern indigo snake. In 1978 the eastern indigo snake was listed as Threatened by the U.S. Fish and Wildlife Service and a recovery plan was approved in 1982 (USFWS 1982). Reasons given for the listing included declines due to habitat degradation, fragmentation and loss, over-collection, road fatalities, and gassing of gopher tortoise burrows (Hyslop 2007, Speake 1993).

In 1976 a captive propagation program for the purpose of recovery of the indigo snake was established under the direction of Dr. Dan Speake of the Alabama Cooperative Wildlife Research Unit at Auburn University; this program continued through 1987. The purpose of the program was to supplement existing populations in the Southeast that were low in eastern indigo snake numbers or restock areas that experienced extirpation. Adult snakes used for captive breeding were obtained from zoos, U.S. Fish and Wildlife Service confiscations, and by wild capture in Florida and Georgia. These snakes formed the nucleus of the captive breeding population. Offspring of this population were used in restocking and reintroduction efforts, but in some cases adults were also released to the wild. Over a period of 10 years the project housed approximately 168 adult indigo snakes that were bred in captivity producing approximately 300 young. Snakes were released in 20 locations across the southeast, nine in Alabama (Speake and Smith 1987; Speake et al 1987, unpublished data). Between 1978 and 1986, 285 juveniles and 34 adults, for a total of 318 snakes, were released at the Alabama locations (unpublished data). Of the nine Alabama release sites, two were outside the historic range of the eastern indigo snake, although during the Pleistocene the eastern indigo snake ranged further north into the Black Belt of Dallas County, as did the gopher tortoise (Dobie and Leary 1996).

In recent years the Alabama Department of Conservation and Natural Resources (ADCNR) has conducted multiple searches of the Alabama release locations (Clay 2006; 2007) with no eastern indigo snakes or evidence of snakes being found at any release site. Sightings of presumed eastern indigo snakes are occasionally reported, and a number have been from credible sources (Hart 2002), but to date none of the sightings have been confirmed.

In 2006 ADCNR revived the prospect of reintroducing the eastern indigo snake to southern Alabama. Following a feasibility study, a collaborative project between Auburn University, the Alabama Department of Conservation and Natural Resources, and The Orianne Society was begun. The main goal of the project is to establish a viable population of the eastern indigo snake (*Drymarchon couperi*) on Conecuh National Forest, and secondarily to test for an appropriate release technique.

At the outset of the project a captive breeding population of eastern indigo snakes was to be housed at Auburn University; this approach was dropped in favor of bringing wild gravid females into the lab, holding them until oviposition, and then returning females to the site of capture. Eggs from the females would then be incubated in the lab with the resulting offspring being used as the basis for the release.

The release design incorporates a hard and soft release technique with an equal number of snakes scheduled for both approaches. Snakes that are hard released are taken to a site with appropriate cover, such as a brush pile or gopher tortoise burrow, placed on the ground and allowed to crawl away without interference; this is done in an area without movement-inhibiting structures. Snakes that are soft released are treated in exactly the same manner except that they are being released inside large enclosures (snake pens) approximately 1 ha in size (Figure 1). The snake pens have been designed to prevent wide-ranging movements of released snakes (Figure 2 a, b, c). Survivorship of snakes from each release technique is being compared to test for the better method for future snake releases. Radio telemetry is being used to track snake movements, activity patterns, habitat selection, and survivorship.

Figure 1. Diagram of the snake pens used for the soft-released snakes, the size of each one, and their location in Conecuh National Forest relative to nearby roads.

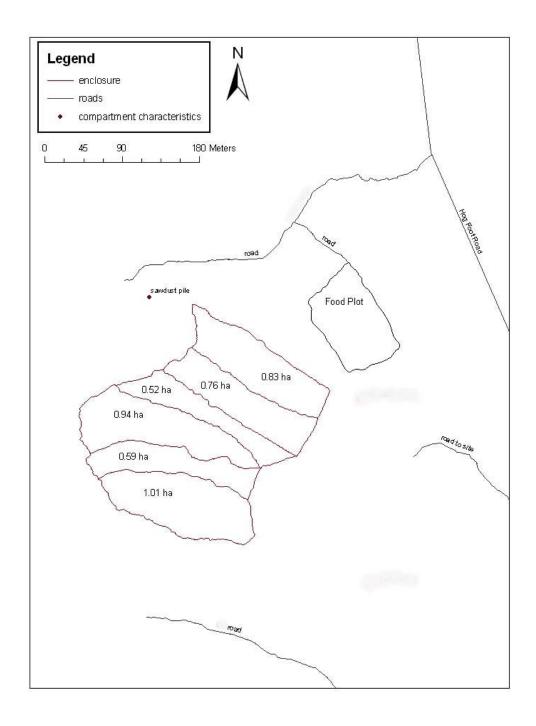


Figure 2a. The soft release pens are posted as a wildlife research area to discourage tampering. The use of eastern indigo snake specific terminology was avoided to avoid drawing unwanted attention to the snake pens.



Figure 2b. View of the pens showing the general habitat type that is encompassed.



Figure 2c. Wading pools, approximately 1.5 m x 0.25 m in sizes, were placed in the pens. Each pen received four pools. Brush piles were constructed at various sites within the pens to provide additional cover for the snakes.



Project Partnership and Collaboration

In late-summer 2006 the Alabama Department of Conservation and Natural Resources (ADCNR) approached Auburn University (AU) with the prospect of a second attempt to reintroduce the eastern indigo snake in Alabama. The result of this meeting was a two year feasibility study (2006-2008) supported through a State Wildlife Grant. At this time Project Orianne (now The Orianne Society) was in the early stages of creation, organization, and establishment. Positive outcome of the feasibility study led to the current three year implementation project and at this time The Orianne Society (TOS) joined the partnership of AU and ADCNR.

Contributions of ADCNR to the eastern indigo snake project have been through the State Wildlife Grant that receives pass-through U.S. Fish and Wildlife funds, logistical support with use of the ADCNR Cessna to locate wayward snakes, education and promotion through the website and *Outdoor Alabama* magazine, the posting of signage within the national forest to alert the public of the presence of the indigo snake, and continuing interest and support from nongame biologists.

TOS has contributed funds and expertise. Funding from TOS has been used to meet the non-federal match requirements of the State Wildlife Grant and with the additional funding the project has advanced more rapidly. TOS staff has provided irreplaceable expertise and logistical support with the acquisition of gravid female snakes from southeastern Georgia. The Orianne Center for Indigo Conservation (OCIC), located in Eustis, FL, is to be the breeding, research, and educational facility of TOS. From the clutches of snakes hatched at AU, a minimum of one snake per clutch has gone to the OCIC to serve as breeding stock. In 2014 the OCIC will begin providing snakes to be released in Conecuh National Forest.

US Fish and Wildlife Service is involved through two regional offices. Listing and recovery lead of the eastern indigo snake is held in the Mississippi Ecological Services Field Office in Jackson, MS. Daphne Ecological Services Field Office in Daphne, AL coordinates with ADCNR on instate State Wildlife Grants.

A snake reintroduction project is dependent upon having a suitable site for release and the U.S. Forest Service (USFS) responded favorably with the request to use an area in the Conecuh National Forest (CNF). USFS staff at CNF has provided on-the-ground support through the prescribed burns around and within the snake pens, and with a precision that preserved the fence lines and plastic wading pools.

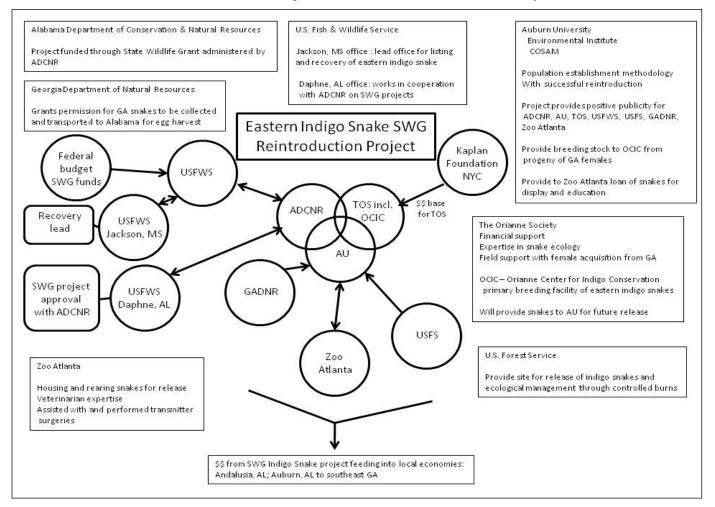
Collection permits allowing the use of female snakes captured in Georgia to be used were obtained through the Georgia Department of Natural Resources (GADNR), and GADNR staff participated in collecting females.

Zoo Atlanta (ZA) joined with the project in 2010 with their offer to house, feed, and rear snakes. ZA took on the task with the 2009 cohort and with the finalization of the captive phase at AU will have reared three cohorts of snakes. ZA has a room dedicated to the indigo snakes and isolated from other reptiles, a measure to reduce the likelihood of disease transmission. ZA is promoting eastern indigo snake conservation with a poster on public display in the reptile house (Figure 3). Two male eastern indigo snakes, provided by AU, are on loan to ZA for display and education. Figure 4 outlines the connections and contributions of the various partners and collaborators.

Figure 3. Poster that is on public display in the reptile house at Zoo Atlanta promoting conservation of the eastern indigo snake in the southeastern United States.



Figure 4. The breadth of partnership and collaboration that has developed around the reintroduction efforts of the eastern indigo snake into Alabama is illustrated by this flow chart.



An unexpected educational opportunity and collaboration developed shortly after the 2010 release. Blue Lake Methodist Camp across the Pond Creek drainage from the release site has been readily used by the reintroduced snakes from both years with as many as four snakes having been observed on camp property on the same day. The camp director was contacted to inform her about snake use on the camp's property and she expressed support and enthusiasm for the project. They welcomed access to the camp property to track snakes whenever needed. According to her, encounters by their guests with venomous snakes are a regular occurrence including one bite from a copperhead in 2011. The director hopes the indigo presence will eliminate encounters with venomous snakes. To help camp personnel have posted signs and posters around the camp to inform and educate patrons on the ecological importance and protected status of the eastern indigo snake.

Project Status

Females

In 2008, 2009, 2010, and 2011 gravid female eastern indigo snakes were collected from localities in southeastern Georgia (Table 1, Figure 5) and transported to Auburn. In the lab females were retained until oviposition, and once deemed healthy enough for release were returned to the site of capture. Three snakes have come from Atkinson County, five from Bryan, two from Coffee, four from Evans, one from Long, one from Sands, three from Telfair, three from Wayne, and two from Wheeler; no female has been used more than once.

Table 1. A listing of the localities by county, capture and release dates of wild-caught gravid females from Georgia.

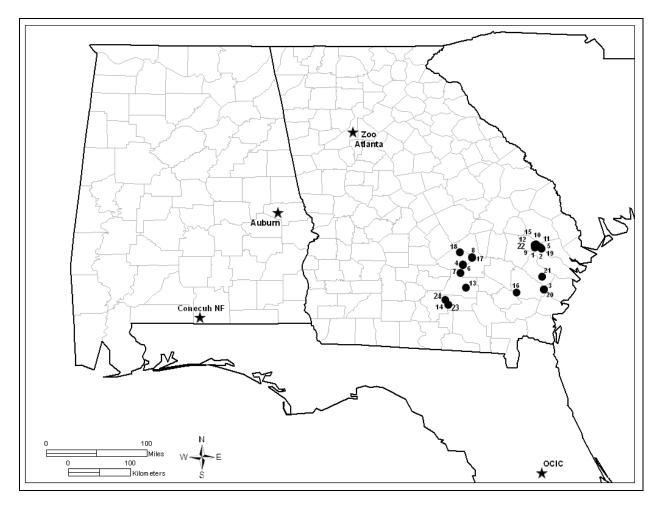
Female	Capture date	Release date	Capture location	Release location
			(County)	
1	29 Jan 2008	21 May 2008	Evans	same as capture
2	29 Jan 2008	21 May 2008	Bryan	same as capture
3 ¹	1 Feb 2008	3 Apr 2008	Wayne	na
4	5 Mar 2008	14 July 2008	Telfair	same as capture
5	29 Jan 2009	10 Apr 2009	Bryan	same as capture
6	7 Jan 2009	10 Apr 2009	Telfair	same as capture
7	29 Jan 2009	30 Apr 2009	Coffee	same as capture
8	29 Jan 2009	30 Apr 2009	Wheeler	same as capture
9 ²	27 Jan 2009	28 Jan 2009	Evans	same as capture
10	27 Jan 2009	30 Apr 2009	Bryan	same as capture
11	27 Jan 2009	30 Apr 2009	Bryan	same as capture
12	31 Jan 2009	10 Apr 2009	Evans	same as capture
13	30 Jan 2009	30 Apr 2009	Coffee	same as capture
14	16 Feb 2010	25 May 2010	Atkinson	same as capture
15^{3}	16 Feb 2010	25 May 2010	Evans	same as capture
16	16 Feb 2010	25 May 2010	Wayne	same as capture
17	4 Mar 2010	25 May 2010	Wheeler	same as capture
18	4 Mar 2010	25 May 2010	Telfair	same as capture
19	4 Mar 2010	24 June 2010	Bryan	same as capture
20	8 Mar 2010	25 May 2010	Wayne	same as capture
21	8 Mar 2010	25 May 2010	Long	same as capture
22	5 Feb 2011	25 May 2011	Sands	same as capture
23	5 Feb 2011	25 May 2011	Atkinson	same as capture
24	15 Mar 2011	25 May 2011	Atkinson	same as capture

¹Died in captivity; deposited in AUM herpetological collection

²Exchanged for larger female

³Not gravid

Figure 5. Map illustrating the location of Auburn University, the general collection sites in Georgia of the 22 gravid female eastern indigo snakes from which eggs have been obtained, Zoo Atlanta, Conecuh National Forest, the site of the reintroduction and the Orianne Center for Indigo Conservation.



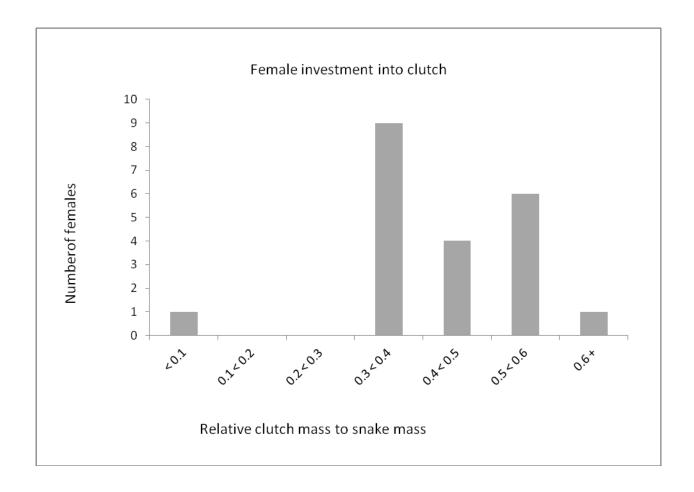
Once in the lab, total length and mass of each female was recorded, as was mass following oviposition. Relative clutch mass (clutch mass/female mass following oviposition) was calculated for all females (Table 2, Figure 6). Our data indicate that females are investing an average of 41% of their body mass into egg production with the normal range spanning 31% to 64%, although a low of 8% was recorded for one female.

Table 2. Female snake length and mass measurements taken before and after oviposition, total clutch mass and relative clutch mass to female snake mass.

Female	Total	Mass (g)	Mass (g)	Clutch	Relative clutch		
	Length	pre-oviposition	post-	mass ¹	mass/snake		
	(cm)		oviposition		mass		
			_		(post-		
					oviposition) ²		
1	172	1714.6	1450	723.8	0.50		
2	198	2459.8	1800	663.2	0.37		
4	147	1698.8	1373.7	554.2	0.40		
5	186	2426.7	1541.8	820.2	0.53		
6	169	1846.7	1212	614.4	0.51		
7	149	984.4	724.6	327.8	0.45		
8	179	1706.1	993.5	421.8	0.42		
10	186	1689.6	1145.4	92.6	0.08		
11	177	1430.8	896.1	336.3	0.38		
12	162	1700.7	950.5	613.1	0.64		
13	186	1579	875.4	274.6	0.31		
14	153	1921	1478	554.1	0.37		
16	142	1484	1232	434.5	0.35		
17	149	1689	1360	464.4	0.34		
18	159	1684	1255	428.3	0.34		
19	125	1223	817	322.5	0.39		
20	131	1130	738	320.6	0.43		
21	131	1258	674	381.3	0.57		
22	166	1780	1252	475.3	0.38		
23	169	1364	842	461.6	0.55		
24	165	1414	937	488.9	0.52		
mean	162±20.1	1627.8±364.8	1121.3±309.4	465.4±166.7	0.42±0.12		
±std							
i. 1 1	polydos all aggs of alutah unlags abytiquely inviable at time of avinagition						

¹includes all eggs of clutch unless obviously inviable at time of oviposition ²proportion of clutch mass/snake mass = clutch mass/(snake mass – clutch mass)

Figure 6. Distribution of relative clutch mass of 21 female eastern indigo snake, following oviposition, from southeastern Georgia.



Body condition (BC) statistics for 2008, 2009, 2010, and 2011 females were calculated using the following formula:

Body Condition (BC) = snake mass (SM)/total length (TL).

Female BC was calculated for both pre-oviposition and post-oviposition. In calculating the pre-oviposition metric the mass of the clutch was deducted from the female mass (Table 3).

Table 3. Body conditions of female snakes calculated before and after oviposition.

Female	ВС	ВС	
	pre-oviposition*	post-oviposition	
1	5.8	8.4	
2	9.1	9.1	
4	7.8	9.3	
5	8.6	8.3	
6	7.3	7.2	
7	4.4	4.9	
8	7.2	5.6	
10	8.6	6.2	
11	6.2	5.1	
12	6.7	5.9	
13	7.0	4.7	
14	12.6	9.7	
15	5.0	5.5	
16	10.5	8.7	
17	11.3	9.1	
18	10.6	7.9	
19	9.8	6.5	
20	8.6	5.6	
21	9.6	5.1	
22	7.9	7.5	
23	5.3 5.0		
24	5.6 5.7		

^{*}excludes mass of clutch

Mean female body conditions were not significantly different among years (Figure 7). Preovipositional females that have a lesser body condition score tend to lay oblong shaped eggs
(Figure 8). Using the pre-ovipositional body condition score is a better predictor for egg shape
than the post-ovipostional score (Figure 9). Female body condition exerts a positive influence
upon clutch size whereby females with better body condition produce more eggs per clutch
(Figure 10). Longer females also lay larger clutches (Figure 11). During each of the three years
the females used represented a wide range of pre-oviposition body conditions. But what cannot
be determined from these data is if body conditions of individual females fluctuate between
years, or if the individual female exhibits a similar body condition from year to year.

Figure 7. Mean body condition (and 95% confidence limits) for female eastern indigo snakes during the four years of study. Body condition of females taken from field does not differ among years (square is mean body condition; vertical bar is 95% confidence limits). Note that female condition during 2010 cold winter did not differ from other years.

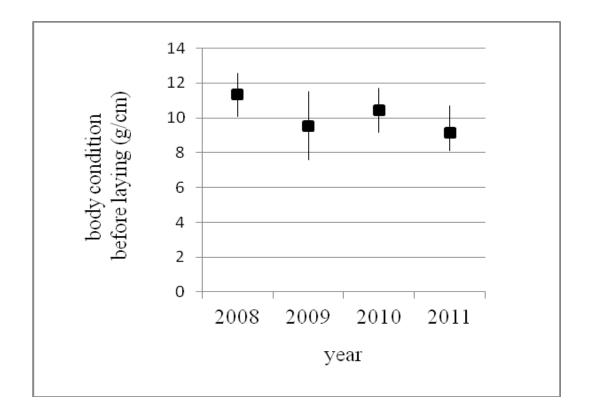


Figure 8. Linear regression of egg shape on female body condition before females have oviposted. Adult females that are light for their body length (before oviposition) tend to produce eggs that are, on average, oblong. This relationship is much stronger than a similar relationship based on body condition after oviposition.

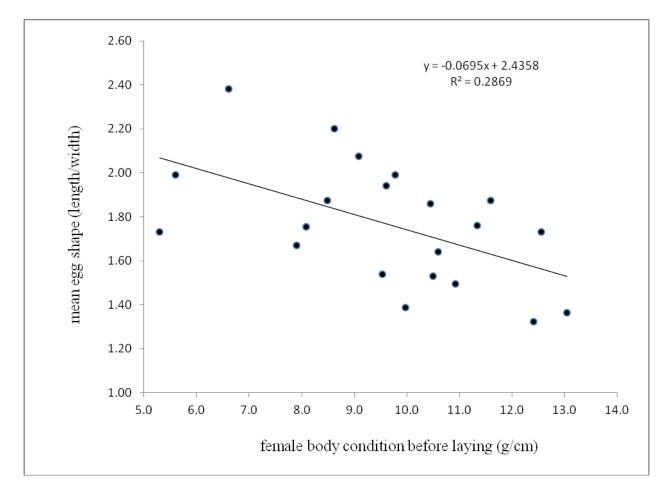


Figure 9. Linear regression of egg shape on female body condition after oviposition. Adult females that are light for the body length (after oviposition) tend to lay eggs that, on average are oblong.

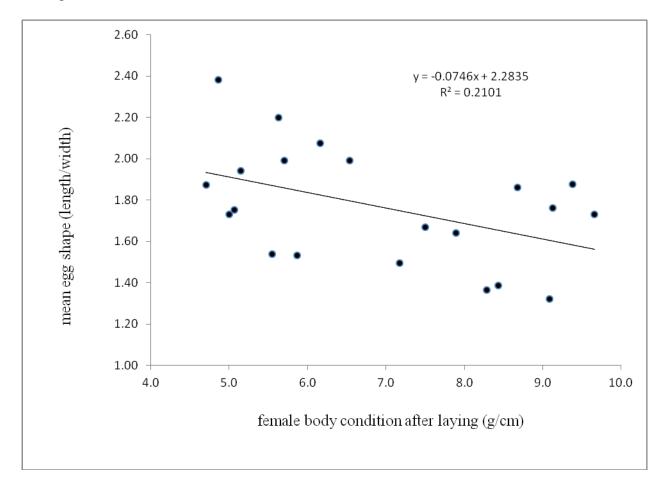


Figure 10. Linear regression of clutch size on female body condition before oviposition. Clutch size increases with increased body condition.

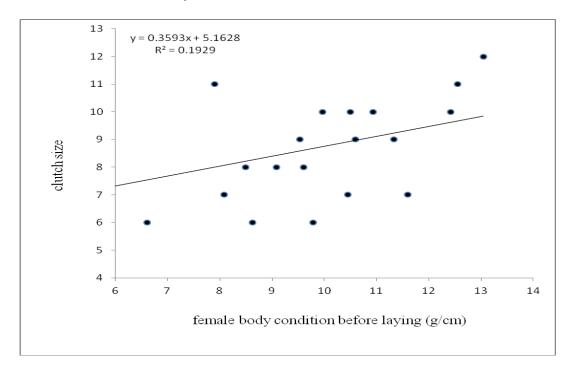
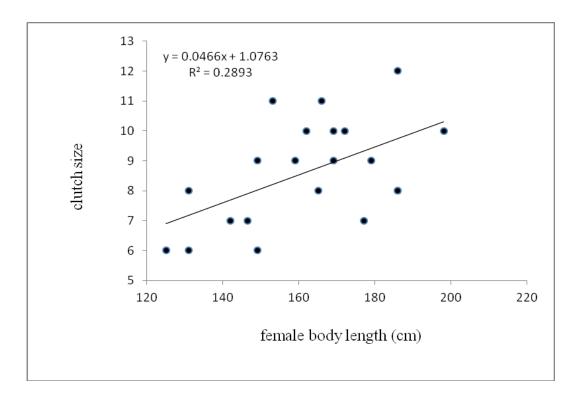


Figure 11. Linear regression of clutch size on female body length. Clutch size increases with increasing female length, as it does for many snake species.



Eggs

Eggs (Figure 12) were collected from 21 snakes; three clutches in 2008, eight in 2009, seven in 2010, and three in 2011. Clutch size ranged from six to 12, number of hatchlings from 0 to 11, clutch viability from 0% to 100%, and incubation times from 97 to 121 days (Table 4).

Figure 12. Eggs have a cream-white coloration with a granular surface.



Table 4. A listing of the 20 clutches of eggs from 2008, 2009, 2010, and 2011 with oviposition and hatch dates, clutch size, and viability.

Female	Clutch	Year	Oviposition	Hatch	Incubation	Clutch	No.	% Clutch
	ID		date	date(s)	duration	Size	Hatchlings	viability
					(days)			-
1	A	2008	6 May	13-17	99-103	10	10	100
			-	Aug				
2	В	2008	13 May	18-21	97-100	10	10	100
				Aug				
4	C	2008	15 May	20-23	97-100	7	6	85.7
				Aug				
5	F	2009	31 Mar	14-18	105-109	12	11	91.7
				July				
6	D	2009	25 Mar	13-21	110-118	10	9	90.0
				July				
7	I	2009	12 Apr	30 July	109-112	6	4	66.7
				-3 Aug				
8	K	2009	25 Apr	4-8 Aug	101-105	9	9	100
10	J	2009	20 Apr	na	na	8	0	0
11	Н	2009	12 Apr	30 July	109-113	7	6	85.7
				-3 Aug				
12	G	2009	5 May	22-29	78-84	10	10	100
				July				
13	Е	2009	29 Mar	27-28	120-121	8	2	25.0
				July				
14	L	2010	8 May	18-23	102-107	11	7	63.6
				Aug				
16	N	2010	13 May	27 Aug	106-112	7	7	100
				-3 Sept				
17	O	2010	16 May	4-19 Sept	110-125	9	9	100
18	P	2010	23 May	6-14 Sept	105-113	9	9	100
19	Q	2010	12 June	27 Sept –	136-141	6	5	83.3
				1 Oct				
20	R	2010	6 May	27-30	113-116	6	6	100
				August				
21	S	2010	16 May	27 Sept	133-137	8	8	100
				– 1 Oct				
22	T	2011		20-22	91-93	11	10	91
			21 Apr	July				
23	U	2011		22-24	86-88	9	9	100
			28 Apr	July				
24	V	2011	18 May	8-11 Aug	83-87	8	8	100
mean±std						8.6±1.8	7.4 ± 2.8	84.9±26.7

Overall clutch mass ranged from 92.6 g to 820.2 g with a mean of 494.7 g, and as expected total clutch mass was dependent upon number of eggs in the clutch. Individual egg mass ranged from 43.1 to 86.0 g with a mean of 64.0 g, mean egg length ranged from 5.7 to 9.0 cm with a mean of 6.6 cm, and egg width ranged from 3.0 to 5.0 cm, with a mean of 4.2 cm (Table 5).

Table 5. Egg mass and dimensional measurements for the clutches laid by wild collected female eastern indigo snakes in 2008, 2009, 2010, and 2011.

Clutch	Female	N	Clutch mass	Mean egg	Mean egg	Mean egg
			(g)	mass (g)	length (cm)	width (cm)
A	1	10	723.8	72.4 ± 4.8	6.4 ± 0.5	4.6±0.1
В	2	9	663.2	73.7±5.1	6.2±0.2	4.7±0.2
С	4	7	554.2	79.2±3.4	7.8±0.5	4.2±0.1
D	6	10	614.4	61.4±4.1	6.4±0.3	4.3±0.14
Е	13	5	274.6	57.9±2.3	6.8±0.2	3.6±0.1
F	5	12	820.2	68.4±2.6	6.2±0.4	4.6±0.4
G	12	10	613.1	61.3±3.1	6.4±0.4	4.2±0.2
Н	11	7	336.3	48.0±2.7	6.5±0.4	3.7±0.3
I	7	6	327.8	54.6±4.1	8.0±0.7	3.4±0.2
J	10	2	92.6	46.3±10.9	6.9±0.1	3.3±0.1
K	8	7	421.8	60.3±2.0	6.5±0.2	4.2±0.3
L	14	11	554.1	55.1±2.7	7.0±0.5	4.0±0.1
N	16	7	434.5	62.1±6.1	7.3 ± 0.8	4.0±0.2
О	17	9	464.4	51.6±1.2	6.7±0.3	3.8±0.1
P	18	9	428.3	47.6±1.5	6.3±0.2	3.8±0.1
Q	19	6	322.5	57.1±2.3	7.5±0.4	3.8±0.1
R	20	6	320.6	53.4±3.4	7.8±0.6	3.6±0.2
S	21	8	381.3	47.7±2.2	6.9±0.7	3.6±0.1
T	22	11	522.9	47.5±1.7	6.0±0.4	3.6±0.1
U	23	9	461.6	51.3±2.0	6.5±0.4	3.7±0.2
V	24	8	488.9	61.1±2.4	7.4±0.6	3.7±0.1
mean±std			467.7±167.2	58±9.4	6.8±0.6	3.9±0.4

Figure 13 presents the relationship of egg mass to width and as mean egg width increases mean egg mass increases. Figure 14 presents the relationship of mean egg mass to mean egg length and as mean egg length increases mean egg mass increases.

Figure 13. Regression of egg mass on egg width.

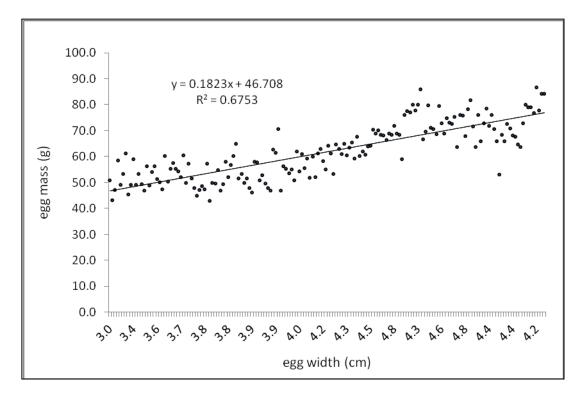


Figure 14. Regression of egg mass on egg length.

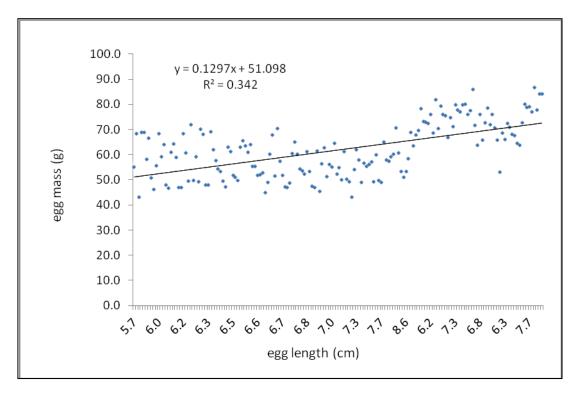
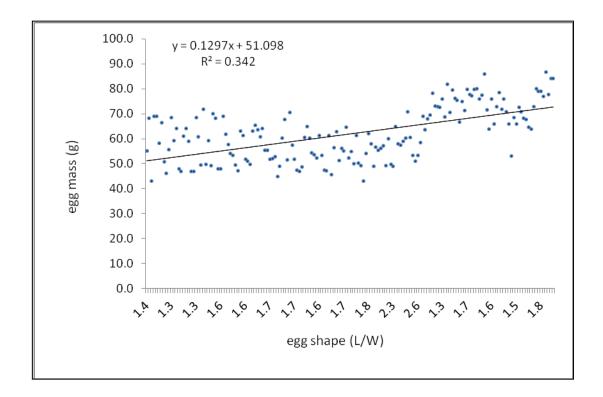


Figure 15 illustrates the relationship of egg shape to egg mass. While egg mass increases or decreases with a concomitant change in the dimensions of width and length (Figs. 13 & 14) egg mass is not influenced by egg shape (length/width). A minimum egg mass, intuitively, is required for full-term egg viability and any increase over this minimum may confer initial survivability advantages to the hatchling snake. But the female's body size, condition, and the number of eggs produced will impose constraints upon egg shape and mass as seen in these preliminary results.

Figure 15. Regression of egg mass to shape (length/width) egg.



Incubation temperatures of eggs were similar between years, ranging from 21.7 to 27.2 °C in 2008, 20.6 to 27.8 °C in 2009, 21.1 to 26.7 °C in 2010, and 22.2 to 27.5 °C in 2011 (Table 6).

Table 6. Egg surface temperatures ($^{\circ}$ C) recorded during incubation for 2008, 2009, 2010, and 2011.

	2008	2009	2010	2011
n	390	1072	1430	267
low	21.7	20.6	21.1	22.2
high	27.2	27.8	26.7	27.5
mean ± std	24.7±0.8	24.2±1.0	24.1±0.1	26.1±1.7

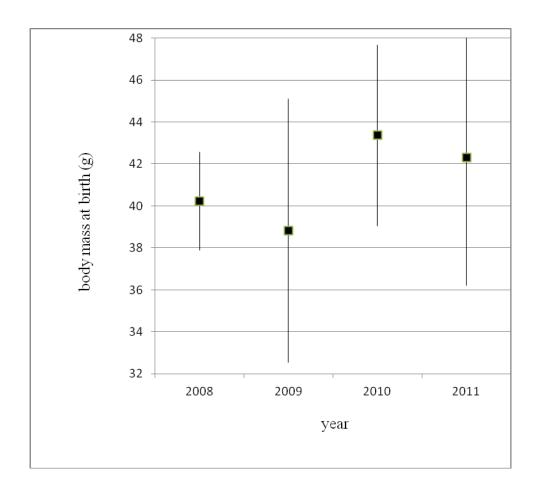
Hatchlings 2008-2011

Body mass at hatching (Figure 16) is presented in Figure 17. Mass of the 2010 cohort was greater than that of the other three years, but not significantly so.

Figure 16. Hatchling eastern indigo snake from eggs laid by wild caught gravid Georgia female.

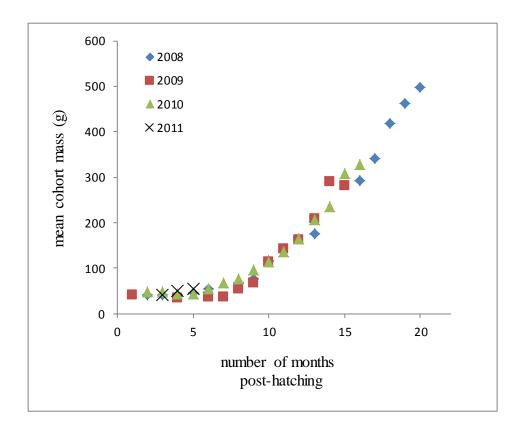


Figure 17. Body mass of offspring larger in 2010 than previous two years, or subsequent year. This was the cold year and these eggs hatched over a longer time range (8/18-10/1) than in 2008 (8/26-9/2), 2009 (7/19-8/8), or 2011 (7/20-8/11).



Growth data based on snake mass, comparing cohorts, is presented in Figure 18. Young snakes exhibit a gradually increasing growth curve until about 1 year of age at which time snake mass accumulates at an increasing rate. Rates between 2008 and 2009 were similar; the depressed growth curve for 2010 is due to a protracted hatching period. Few data points are available for the 2011 hatchlings, thus no protracted growth curve can be plotted.

Figure 18. Blue diamonds are 2008 cohort, red squares 2009 cohort, green triangles 2010 cohort, and black X 2011 cohort. All cohorts, while in captivity, have had similar mean growth trajectories.



Sex ratio of young snakes is only available at this time for the 2008, 2009, and 2010 cohorts. Seven clutches are skewed toward females, eight toward males, and one has equal numbers of males and females. The overall sex ratio is nearly equal at 1 female to 1.1 males (Table 7).

Table 7. Sex ratio of the 17 clutches from the 2008-2010 snake cohorts.

Clutch	Sex ratio FF:MM
A	5:3
В	3:6
С	3:2
D	6:1
Е	1:1
F	2:8
G	3:6
Н	1:2
I	0:4
K	2:6
L	3:4
N	2:5
O	3:6
P	5:4
Q	4:1
R	4:1
S	7:1
total	55:61
overall	0.9 FF:1 MM

In general the natal shed occurs within 10-12 days, but as quickly as 8 days, following hatching, as observed in all four cohorts. An examination of shed intervals reveals a faster shed interval for the 2011 cohort compared to the 2008, 2009, and 2010 cohorts (Table 8).

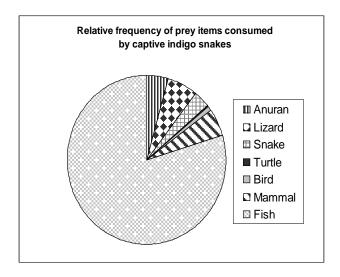
Table 8. The shed intervals of the four clutches (2008-2011) of eastern indigo snakes hatched at Auburn University. Natal shed is the first shed following hatching. Shed intervals are represented in days.

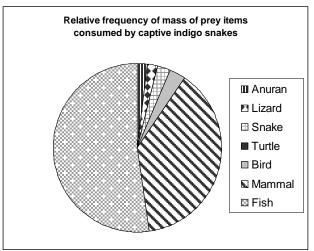
Cohort	2008	2009	2010	2011
natal shed	10.6±1.8	12±6.4	10.9±1.5	8.5±1.2
(n)	(25)	(50)	(50)	
shed	43.8±18.0	26.3±3.3	40.7±8.9	34.85±5.0
(n)	(193)	(24)	(40)	(39)

A wide variety of food items were offered and accepted as prey to the young indigo snakes, with 41 species of prey having been eaten. The list of taxa consumed includes fish (5 spp.), frogs and toads (10 spp.), squamates (5 lizards and 10 snakes), turtles (3 spp.), birds (3 spp.) and mammals

(4 spp.) Figure 19 is a graphical representation of food items consumed. Fish made up the greatest percentage of food accepted both numerically and by mass. Regarding prey mass, mammalian prey was the second largest category of prey accepted. The preponderance of the prey categories fish and mammal were due to their being default prey items because of ease of availability. Fish were readily available from aquaculture sources, and rodents were lab or commercially reared.

Figure 19. Food items accepted by the juvenile captive indigo snakes. The first chart illustrates the relative frequency of prey items by category based on the number of items eaten. The second chart presents the data based on the total mass of each prey item category.





Medical Care

Eastern indigo snakes, while being housed at Auburn University, received medical care by Dr. Marie Rush. Adult females were given physical examinations, a body condition assessment, weighed and measured, as well as a complete blood count and blood chemistry profile. A blood sample was taken from each female and banked. For females in which the body condition or level of hydration was of concern she was given supplemental oral nutrition with fluids, and individuals showing signs of illness were treated accordingly. Prior to release, they were fed, or if needed given supplemental nourishment, and given a physical examination and reassessment of the body condition score.

Eggs were examined for physical health, weighed, and measured. Their health was monitored

throughout incubation. If any concern was noted, it was dealt with through treatment of the individual egg and isolation of that egg from the clutch, if needed. All eggs that were non-viable (including slugs) were given postmortem evaluations.

Once hatched, each juvenile snake received a full physical examination, and a weight and measurement was taken. PIT tag placement in the 2008 and 2009 cohorts was subcutaneous, but in the 2010 and 2011 cohorts, coelomic. Blood samples were taken for complete blood estimates and for banking. Snakes are sexed via probing but not until approximately 1 ½ years of age.

The 2009 and 2010 snake cohorts were moved to Zoo Atlanta for growth and maintenance (Figure 20 & 21), and, to maintain consistency of care, a medical and necropsy protocol accompanied them in accordance with the care given at Auburn.

All snakes that expire or are euthanized are given a full necropsy with retention of tissue samples, and evaluated with culture and histopathology. This has led to several research studies and publications currently in process by Dr. Rush and Dr. Roberts with respect to integumentary and gastrointestinal pathogens in this species.

Figure 20. Racks and boxes used at Zoo Atlanta to house the eastern indigo snakes.



Figure 21. An individual box with one of the young eastern indigo snakes. Newspaper is used as the main substrate with wood shavings in the hide box.



Transmitter Implantation Surgery

Implantation of radio-transmitters: Original Indigo snake group A-C adults: 14 March 2011 for

re-release in Conecuh National Forest Site, Alabama.

Project Veterinarian: Elizabeth Marie Rush, BS, DVM

Assisting with Surgeries:

John Roberts, DVM, DACVP (Alabama State Veterinary Diagnostic Lab)

Mike P. Wines (project technician)

Drs. Sam Rivera and Brad Lock from Zoo Atlanta

Jim Godwin, Environmental Institute, PI

Eric Wheeler, AU pre-veterinary medicine student

Prior to the procedure all snakes were given a physical examination and found to be in sufficient

body condition for surgery; snakes had been fasted over the previous five days. Blood samples

were collected and stored for future research.

Snake B2 was noted the week prior to surgery to have significant pustular and crusting

dermatitis. Amikacin sulfate was started prior to surgical procedures to assist with secondary

infections.

Each snake was masked in a snake tube with isoflurane inhalant anesthesia until at plane II

anesthesia and then given a local block of lidocaine 1% (0.15 ml total), subcutaneous, at the

transmitter insertion incision site. Plane III was maintained throughout surgery. To prevent

hypothermia snakes were maintained on circulating warm water blankets throughout procedures.

An assistant monitored viability throughout the procedure through visual evaluation of heartbeat

and respiration. Isoflurane was maintained at the lowest percentage necessary for surgical

intervention with proper anesthesia for PLANE III safely. Snakes, if not naturally ventilating,

were manually ventilated (IPPV) at a minimum of 5-7 bpm.

The surgical site was prepared with chlorhexidine 2% surgical scrub and rinsed with sterile

saline 3 consecutive times. Sterile gloves and cold sterile instruments were used for each

procedure (in chlorhexidine disinfect baths). Each transmitter was immersed in chlorhexidine

2% solution for several minutes prior to implantation and then rinsed thoroughly with sterile

saline prior to placement.

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The chosen surgical site was located on the right side, approximately 15 cm cranial to the cloaca, a more caudal placement than in other species, but based on the notation of extended avascular lung (airsacs) in this species. A 1-2 cm long incision was made between lateral and ventral scales, with care to avoid damage to scales themselves, and through skin only. This allowed visualization of the coleomic wall membrane, which was easily lifted with forceps and pierced with the blunt tip of curved Kelly forceps allowing for the removal of the old transmitter. Transmitters from last year were located, removed, and evaluated for adhesion, inflammatory changes, etc. (none were noted with the exception of small grade 1 adhesion to the lateral wall of the uterus in female A4, who was gravid).

The replacement transmitter was placed in a caudal direction and tacked into place with a single looped suture around the base of the transmitter/antennae attachment and firmly affixing the transmitter around a rib to prevent migration. A 2mm (6Fr.) coated stylet was fed cranial under the skin cranial to the transmitter and through a 10 Fr. urinary catheter. Prior to surgery the catheter was cut to the length of the antenna. After tunneling cranially to maximum length, a small incision was made over the cranial end to allow for exposure and the stylet was removed, leaving the catheter behind. The antenna of the transmitter was then fed forward through the catheter, and the catheter was grasped at the cranial most aspect, pulled through the incision and thus allowing the antenna to lie flat in the tunnel. The small cranial incision was closed with 3-0 PDO in a single everting suture or with tissue glue. Over the transmitter the coelomic wall was closed with one simple interrupted or cruciate 3-0 PDO suture and the skin was closed in an everting horizontal mattress pattern with 3-0 PDO suture. Tissue glue was used to seal the area. The animal was reversed on room air to stimulate natural respiration and an ambu bag was used, if needed, to ventilate during recovery at several times a minute.

After recovery, each snake was given a 5mg/kg dose of amikacin intramuscularly and this was continued at a 2.5 mg/kg dosage every 72 hours for 3 doses. Animal were recovered in the surgery area before return to the housing area. Each snake was given a single dose of ketoprofen 0.05 mg/kg for inflammation intramuscularly (10mg/ml solution) 48 hours post-operatively. For the two gravid females 1 mg was administered at 48 hours post-operative to help prevent

adhesions to the serosal wall of the uterus, which is in direct contact with the transmitters. No complications were noted other than one snake exhibited prolonged recovery from anesthesia, but she was difficult to anesthetize as she held her breath. Manual ventilation was needed to gain plane III anesthesia, with the result of a longer recovery. She recovered fully.

Snakes were held for a full shed cycle prior to return to Conecuh National Forest to allow for sealing of the incision sites. Two females were suspected to be gravid on surgical intervention, A4 (confirmed by x-ray) and A6. This was discussed with both Dr. Guyer and Jim Godwin, as the potential for adhesion to the lateral serosal wall of the uterus would be increased in these animals with uterine expansion and adjacent positioning. This could lead to adhesions in the area of the transmitter and with potential long-term affect upon reproduction. An alternative to placement of transmitters would have been to delay until oviposition and, after regression of the reproductive tissues, then implant the transmitters. It was elected by the PI to proceed with surgeries. No major adhesions were noted from using this surgical procedure last year on these individuals; however it is noted that the transmitters at that time were significantly smaller and allowed for a much lower load ratio. Animals were rechecked at 48 and 72 hours and monitored daily for any signs of illness or problems. Food was offered after 72 hours post-transmitter implantation. At two weeks post-implantation all snakes were recovering well with no signs of transmitter rejection of or incision dehiscence.

Implantation of radio-transmitters: Juveniles for release 2011: 16 March 2011 for release in Conecuh National Forest Site, Alabama.

Project Veterinarian: Elizabeth Marie Rush, BS, DVM

Assisting with Surgeries:

John Roberts, DVM, DACVP (Alabama State Veterinary Diagnostic Lab)

Mike P. Wines (project technician)

Drs. Sam Rivera and Brad Lock from Zoo Atlanta

Laura Ellsaesser, 2nd year NCSU veterinary student

Eric Wheeler, AU pre-veterinary medicine Student

Zoo Atlanta veterinary technicians

Snakes were given a physical examination prior to surgery, and all were found to be healthy and in good body condition. A blood sample from each individual, collected from the caudal vein, was taken for CBCs, and serum and erythrocyte pellet for future studies. Snakes had been fasted for 5 days. Transmitters were Holohil with whip antennas and an approximate 10 month lifespan.

In preparation for surgery each snake was masked in a snake tube with isoflurane inhalant anesthesia until at plane II anesthesia and then given a local subcutaneous block of lidocaine 1% (0.15 ml total) at the transmitter insertion incision site. Plane III was maintained throughout surgery. Snakes were maintained on circulating warm water blankets to prevent hypothermia. Viability, throughout the procedure, was visually monitored by an assistant through evaluation of heartbeat and respiration. If not naturally ventilating, snakes were manually ventilated (IPPV) at least 5-7 bpm, and isoflurane was maintained at the lowest percentage necessary for surgical intervention at PLANE III. The surgical site was prepared with chlorhexidine 2% surgical scrub and rinsed with sterile saline 3 consecutive times. Sterile gloves and cold sterile instruments were used for each procedure (in chlorhexidine disinfect baths). Each transmitter was gas sterilized with ethylene oxide and allowed 48 hours to breathe prior to placement. This site was located on the right side and approximately 15 cm cranial to the cloaca. This is a more caudal placement than in other species based on the notation of extended avascular lung (airsacs) in this species. A 1-2 cm long incision, through skin only, was made between lateral and ventral scales, exercising care to avoid damage to scales. This allowed visualization of the coleomic wall membrane, which is easily lifted with forceps and pierced with the blunt tip of curved Kelly forceps. The transmitter was then placed in a caudal direction and tacked into place with a single

looped suture around the base of the transmitter/antenna attachment and was firmly affixed around a rib to prevent migration of the transmitter. A 2mm (6Fr.) coated stylet was fed cranial under the skin and cranial to the transmitter and through a 10 Fr. urinary catheter that had been cut to the length of the antenna prior to surgery. After tunneling cranially to maximum length, a small incision was made over the cranial end to allow for exposure, and the stylet was removed leaving the catheter behind. The antenna of the transmitter was then fed forward through the catheter, and the catheter was grasped at the cranial most aspect, pulled through the incision allowing the antenna to lie flat it the tunnel. The small cranial incision was closed with 3-0 PDO in a single everting suture or with tissue glue. Over the transmitter the coelomic wall was closed with one simple interrupted or cruciate 3-0 PDO suture and then the skin was closed in an everting horizontal mattress pattern with 3-0 PDO suture. Tissue glue was used to seal the area.

The animal was reversed on room air to stimulate natural respiration, but, if needed, the animal was ventilated several times a minute with an ambu bag to facilitate recovery. After recovery, each snake was given a 5mg/kg dose of amikacin, intramuscularly, and this was continued at a 2.5 mg/kg dosage every 72 hours for 3 doses. Animals were recovered in the surgery area and then returned to the housing area. Each snake was given a single dose post-operative, intramuscular dose of ketoprofen 0.05 mg/kg for inflammation (10mg/ml solution); no complications were noted.

Two snakes received 24 month transmitters, given their larger body size and capacity that would accommodate this equipment. Snakes were held for a full shed cycle prior to release to allow for sealing of the incision sites. Animals were rechecked at 48 and 72 hours and monitored daily for signs of illness or problems, with food being offered after 72 hours post-transmitter implantation.

Update: Animals healed well and showed no signs of transmitter rejection or incision dehiscence, and recovery progression was as expected. One animal was noted to have some mineralization of the liver on a radiograph taken at Zoo Atlanta, but no other abnormal findings were noted on diagnostics. Medical concerns were discussed and this animal was released with others as scheduled.

Transition Tubs

Because the hatchling snakes had been maintained in a lab setting, confined to a cage that offered limited room for movement, and had been fed a diet primarily of pre-killed, lab-produced rodents, we became concerned that the released snakes may be at a disadvantage once placed in a wild environment. Therefore, prior to release, we exposed the snakes to outdoor conditions by placing each into one of twenty-six fiberglass tubs that were set to acclimate the snakes to outside environmental conditions. The tubs were located north of Auburn on land owned by the Auburn University Fisheries Department, behind locked gates, and away from public view. The circular shaped tubs measure 1.52 m (5 ft) tall and 3.1 m (10 ft) across (Figure 22). They are constructed of fiberglass and were placed on a slight incline to promote drainage of water in the event of rain. Each tub had a series of holes drilled into the bottom for drainage. The holes were covered with hardware cloth to prevent the escape of snakes. Tops of the tubs were covered by hardware cloth, screwed on around the edge, and overlapping sides woven together. Hardware cloth covered approximately 2/3 of the top, with the remaining 1/3 section being covered by a door flap constructed of black plastic ground cloth material. The door flap also served as a shade cloth, and was woven to the hardware cloth top with plastic zip ties and held tight to the lip of each tub by 12 to 14 large metal binder clips, forming a tight seal (Figure 23). Each tub has one underground refugium connected to a hole drilled approximately 2.5 cm (1 in) off the ground. The refugia are buried at the highest point of the slope for each tub to prevent water from draining into them. These refugia were constructed of 7.6 cm (3 in.) inner diameter PVC tubes each with three chambers for different levels underground (Figure 24). They were buried 76-91 cm (30-36 in) underground to simulate a gopher tortoise burrow or stump hole (Figure 25). Sand was used as a substrate in the bottoms of the tubs, with pine straw, leaf litter and sticks being added as additional natural cover. Each tub contained a large water tub, a 0.6 m x 0.6 m (2 ft x 2 ft) piece of tin, several cinder blocks, and an upside down plastic tub, this allowed for multiple choices of cover for the snake (Figure 26). Only one snake was placed per tub.

The snakes were introduced into the tubs on 29 April 2009. They were brought back into the lab on 13 May 2009 for the implantation of radio transmitters. They were then returned to the tubs 21 May 2009 where they remained until 14 June 2009. The snakes used all types of hiding places, including the refugia, ate well while in the tubs, consuming both frozen/thawed items as

well as wild caught prey, and some individuals exhibited defensive behaviors previously not seen while in the lab.

Figure 22. Fiberglass tubs used to expose snakes to outdoor conditions prior to release.



Figure 23. Hardware cloth and plastic ground cloth used as coverings of the tubs. The black plastic ground cloth served as a shade and door for access into the tubs.



Figure 24. A PVC refugium.



Figure 25. PVC refugium set in place extending 76-91 cm (30-36 in) underground. Hole surrounding the refugium is filled with soil once refugium has been set in place and secured to the tub.



Figure 26. Bottoms of the transition tubs were covered with several cm of sand and pine litter, hide structures, and a water dish were added.



Conecuh Release - 2010

The reintroduction of the eastern indigo snake has become a high profile project for the Alabama Department of Conservation and Natural Resources, the U.S. Fish and Wildlife Service, Auburn University, and The Orianne Society. Because this project is known to the public and will continue to be one of public interest the first snake release was planned to raise awareness of the plight of the indigo snake, deliver the message that the species is protected at the state and federal level, that the indigo snake fits into the larger picture of longleaf pine forest ecosystem restoration, and to publicly acknowledge the partners and collaborators. To that end a publicity release was prepared in cooperation with media specialists with ADCNR and USFWS. Initially an invitation was to be extended to newspapers, radio and TV stations, but because of the remote setting no coverage by radio or TV was expected.

The first release for this project of the eastern indigo snake onto the selected site in Conecuh National Forest took place on 16 June 2010. In attendance were representatives from the Alabama Department of Conservation and Natural Resources (Figure 27), Auburn University (Environmental Institute, Department of Biology, and Society for Conservation Biology), The Orianne Society, U.S. Fish and Wildlife Service, U.S. Forest Service, Georgia Department of Natural Resources, and Zoo Atlanta. Dr. Dan Speake (Figure 28) was invited to attend and released the first snake. Seventeen snakes were released with eight going into the soft release pens and nine being hard-released (Table 9 under **Telemetry**).

Figure 27. Mark Sasser, Non-game Biologist with the Alabama Department of Conservation and Natural Resources, releasing an eastern indigo snake.

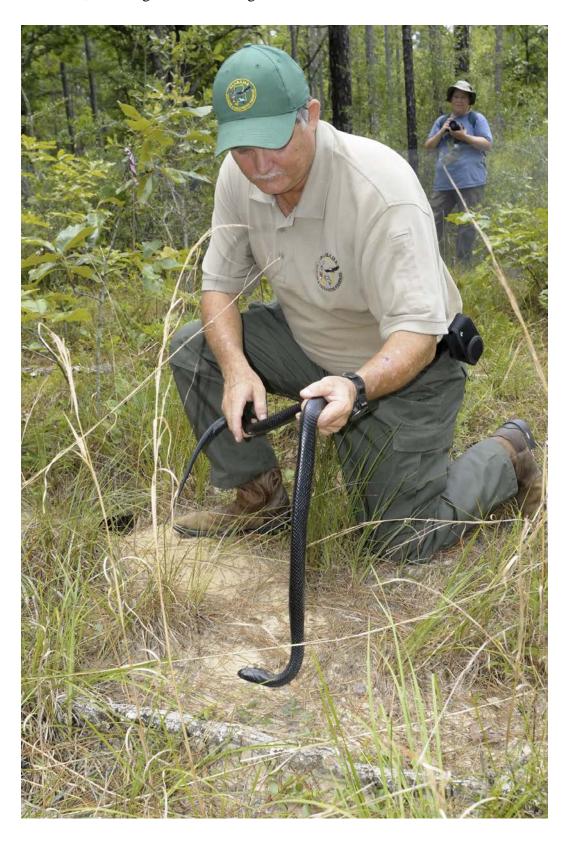
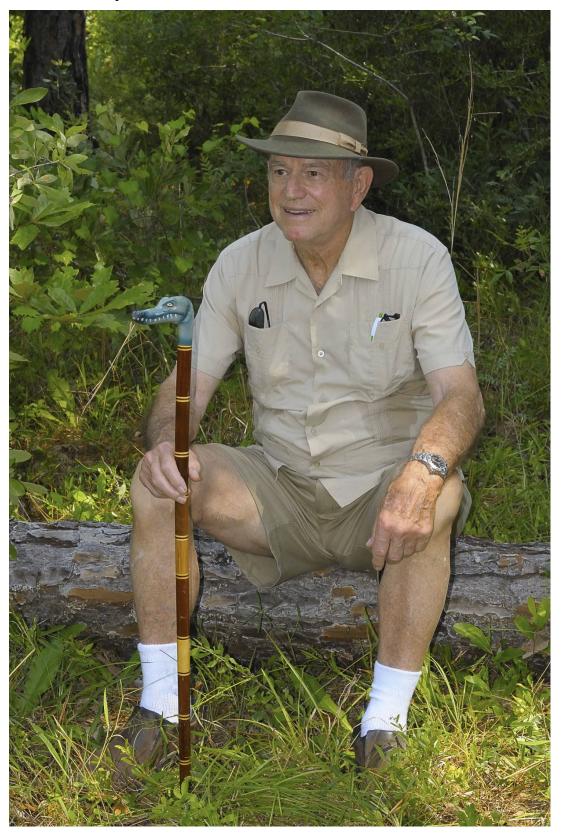


Figure 28. Dr. Dan Speake.



On-hand to cover and document the event was the local Andalusia Star-News newspaper, ADCNR photo and video crew (Figure 29), and Discovering Alabama video crew. The release received poor media attention as the major news item at the time was the Deep Water Horizon oil spill in the Gulf of Mexico. A partial listing of news articles and internet sites of the release include:

http://www.outdooralabama.com/news/release.cfm?ID=820

http://www.outdooralabama.com/OAOnline/indigo10.cfm

http://www.facebook.com/note.php?note_id=406956122556

http://alaparc.blogspot.com/search/label/Indigo%20Reintroduction

http://us1.campaign-

archive.com/?u=946679e7fe51bbf81ce578cc1&id=89dc4bb383&e=aa8d2c9d2b

http://www.accessatlanta.com/atlanta-events/zoo-atlanta-helps-to-440245.html

http://wireeagle.auburn.edu/news/1680

http://www.fs.fed.us/fstoday/100820/about-us/indigo.html

http://www.andalusiastarnews.com/2010/06/17/endangered-snakes-are-returned-to-forest/

http://davidasteen.blogspot.com/2010_06_01_archive.html

Figure 29. Alabama Department of Conservation and Natural Resources video crew on hand to videotape the release of the eastern indigo snakes in Conecuh National Forest.



Conecuh Release - 2011

On 3 May 2011 we re-released the six snakes from the 2008 cohort that were known to survive 2010. Snakes from the 2009 cohort were released on 16 May 2011. A total of 30 snakes were released, 21 with a radio transmitter (Table 9 under **Telemetry**). Ten snakes (five male, five female) with a radio transmitter were soft released within the pens. Four pens received two snakes, two pens with one male and one female, one pen with two females, and one pen with two males. The two remaining pens only received one snake each (one male and one female). Also, 11 snakes (five male, six female) with a radio transmitter were hard released north of the pen along a fire line separating a previously burned forest patch from an unburned forest patch. The remaining nine snakes (all male), with PIT tag only, were hard released south of the pen on an old logging road. Mean size of female snakes was 129.6±10.9 cm total length and 505±149.6 g mass, with a range of 118-154 cm, and 310-796 g. Male snakes were similar in size with a mean total length of 131.8±7.3 cm (range 119-141) and mean mass of 491.3±69.4 (range 369-583) (Table 9). Collection of location data for both hard and soft released snakes began immediately upon release.

In attendance were representatives from the Alabama Department of Conservation and Natural Resources, Auburn University (Environmental Institute, Department of Biology, and Society for Conservation Biology, and College of Science and Math), U.S. Fish and Wildlife Service, U.S. Forest Service, Georgia Department of Natural Resources, and Zoo Atlanta (Figure 30).

Figure 30. Dr. Brad Lock, Zoo Atlanta, preparing to release an eastern indigo snake hatched in 2009. Jimmy and Sierra Stiles are collecting data on the release of this particular snake.



Telemetry

The age of the released snakes has been approximately 22 months, an age old enough to allow

snakes to reach a body size that would allow for implantation of a transmitter with a minimum 6

month lifespan yet small enough not to impede normal bodily functions.

implantation was performed on 14 May 2010 at the Auburn University School of Veterinarian

Medicine under the direction of Dr. Marie Rush and with assistance by Dr. John Roberts, and 14

and 16 March 2011.

The first group of snakes to receive radio transmitters was the 2008 cohort, with surgery being

performed approximately one month prior to release. Transmitters used were Holohil SB-2

temperature calibrated with the following specifications:

weight: 5.0g

length x diameter (mm): 19 x 9.5

nominal lifespan: 10 months

lifespan range: 6-12 months.

Transmitter surgery for the 2009 cohort was performed approximately 2 months prior to release;

snakes were held in captivity longer than planned due to unseasonally cool late-spring weather.

Holohil SB-2T transmitters were implanted. Six snakes of the 2008 cohort were recaptured to

replace failing transmitters with ones providing longer battery life. Replacement transmitters

used were Holohil SI-2T with the following specifications:

weight: 13.5g

length x diameter (mm): 50 x 11

nominal lifespan: 24 months

lifespan range: 12-30 months.

We are using a Communications Specialist R-1000 Telemetry Receiver coupled with a

directional hand held antenna. This allows a reception range of approximately 300 meters.

In 2010, 17 snakes were released, nine female, and eight male. Mean size of female snakes was

129.6±10.9 cm total length and 505±149.6 g mass, with a range of 118-154 cm, and 310-796 g.

Male snakes were similar in size with a mean total length of 131.8±7.3 cm (range 119-141) and

mean mass of 491.3±69.4 (range 369-583) (Table 9). In 2011, 30 snakes were released, 11

female, and 19 male. Mean size of female snakes was 129.6±10.9 cm total length and

55

 505 ± 149.6 g mass, with a range of 118-154 cm, and 310-796 g. Male snakes were similar in size with a mean total length of 131.8 ± 7.3 cm (range 119-141) and mean mass of 491.3 ± 69.4 (range 369-583) (Table 9). Collection of location data for both hard and soft released snakes began immediately upon release (Figure 31).

Figure 31. Jimmy Stiles beginning the process of gathering location data on the first group of radio-transmittered eastern indigo snakes released at Conecuh National Forest.



Table 9. Listing of the 17 snakes hatched in 2008 and released on 16 June 2010, and the 30 snakes hatched in 2009 and released on 16 May 2011 in Conecuh National Forest. Data on each snake includes identification label, sex, length and mass measurements at time of release. Additional measurement data is given for snakes released in 2010 and recaptured in 2011 for transmitter replacement.

		2010		2011			
Snake	C	SVL^1	TL^2	Mass	SVL^1	TL^2	Mass
ID	Sex	(cm)	(cm)	(g)	(cm)	(cm)	(g)
A1	M	99	119	408			
A2	F	101	121	392	130	1525	802
A4	F	130	154	796			1243
A5	M	100	121	369			
A6	F	110	134	572	136	164	1221
A8	F	103	124	538	128	154	723
A9	F	100	122	339			
B1	M	110	133	535			
B2	F	105	128	460	133	160	1043
B4	M	115	141	496			
B6 ³	F	98	118	310			
B7	M	110	134	503			
B8	M	110	135	583			
B10	M	110	132.5	456			
C2	F	113	136	580			
C4	M	108	132	557			
C5	F	108	129	558	134	161	1052
C6	M	114	139	515			
D1	F				110	132	507
D2	F				104	127	437
D4	F				112	134	565
D5	F				99	119	422
D9	F				92	111	360
F2	F				127	153	850
F3	M				120	147	700
F5	M				107	131	528
F6	M				117	143	610
F7	M				119	145	513
F8	M				109	134	554

F10	M			109	133	452
F11	F			107	130	535
G2	F			113	134	468
G9	M			111	133	570
H1	M			109	134	504
H4	F			112	134	502
H5	M			114	139	679
K2	F			113	135	650
К3	M			117	141	537
K9	F			114	138	612
D3	M			130	152	406
E2	M			100	120	315
F9	M			109	112	420
G4	M			100	122	364
G5	M			101	123	465
I1	M			94	116	323
I2	M			104	127	430
K5	M			97	117	355
K6	M			112	137	530
1	4.1	41				

In the 2010 release, eight snakes (four male, four female) were soft released into the pens. Two pens received two snakes each (one male and one female) and four pens received only one snake each (two male and two female). During the 2010 release, nine snakes (five males and four females) were hard released outside the pens about 50 meters apart from each other.

¹snout-vent length ²total length ³died in lab day prior to release

Telemetry-based Studies

Survival

During 2010-2011 radio telemetry was used to determine the fates of the snake after their release. A table listing the fates of released snakes is below (Table 10).

Table 10. Listing of the 38 snakes released with radio-transmitters in 2010 and 2011 in Conecuh National Forest. Data on each snake includes identification label, sex, hatch year, release year, release type (hard or soft), and fate (alive, died, transmitter failure or unknown). Snakes listed as unknown fate are assumed to have had transmitter failure or emigrated out of the study area.

Snake ID	Sex	Hatch Year	Release Year	Release Type	Fate
A1	M	2008	2010	soft	died
A2	F	2008	2010	soft	alive
A4	F	2008	2010	hard	Transmitter failure
A5	M	2008	2010	soft	died
A6	F	2008	2010	hard	alive
A8	F	2008	2010	soft	transmitter failure
A9	F	2008	2010	hard	died
B1	M	2008	2010	soft	died
B2	F	2008	2010	soft	alive
B4	M	2008	2010	soft	died
В7	M	2008	2010	hard	transmitter failure
В8	M	2008	2010	hard	unknown
B10	M	2008	2010	hard	unknown
C2	F	2008	2010	soft	died
C4	M	2008	2010	hard	unknown
C5	F	2008	2010	hard	alive
C6	M	2008	2010	hard	unknown
D1	F	2009	2011	hard	alive
D2	F	2009	2011	hard	alive
D4	F	2009	2011	soft	alive
D5	F	2009	2011	hard	transmitter

					failure
D9	F	2009	2011	soft	died
F2	F	2009	2011	hard	died
F3	M	2009	2011	hard	transmitter failure
F5	M	2009	2011	soft	unknown
F6	M	2009	2011	soft	died
F7	M	2009	2011	hard	alive
F8	M	2009	2011	soft	unknown
F10	M	2009	2011	hard	alive
F11	F	2009	2011	soft	alive
G2	F	2009	2011	soft	died
G9	M	2009	2011	hard	died
H1	M	2009	2011	hard	died
H4	F	2009	2011	hard	alive
H5	M	2009	2011	soft	alive
K2	F	2009	2011	hard	alive
К3	M	2009	2011	soft	unknown
К9	F	2009	2011	soft	died

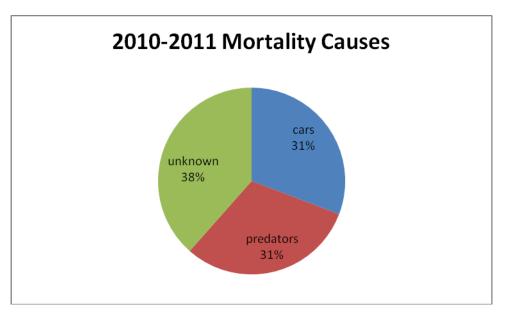
Annual survival was estimated for 2010 and 2011. These estimates were derived using a Kaplan-Meier Annual Survival Calculator for censored data. For the following estimates snakes with transmitter failure or those that emigrated out of the area were censored. For the 17 individuals released in 2010, there were six known mortalities, five were censored and six were known to have lived. These data produced a survival probability of 0.5 (95% confidence interval (CI) 0.90-0.1). For the 21 released in 2011, seven snakes are known to have died and five were censored. Nine are still known to be alive, producing a survival probability of 0.56 (95% CI 0.89-0.24). No snakes from the 2008 cohort that survived 2010 and were re-released are known to have died; although two were censored due to transmitter excretion. This produced a survival probability of 1 (95% CI 1-1). While the data set was small, indications are that after the first year post-release probability of survival increases.

Survival estimates between hard and soft release types are confounded by small sample size and heavy censoring. In 2010, for the hard release one snake died, five were censored, and three

survived producing a survival probability of 0.75 (95% CI 1.24-0.26). The 2010 soft releases had 5 snakes die, none censored and three survived with a survival probability of 0.375 (95% CI 0.92-0.17). So far for 2011, the hard releases had three die, three censored, and five are still surviving, producing a survival probability of 0.625 (95% CI 1.04-0.2). For the soft releases of 2011, four snakes died, three were censored, and three are still alive. This produced a survival probability of 0.429 (95% CI 0.98-0.13). Survival estimates between soft and hard releases indicate that hard releases trend toward having higher survival probabilities. However, further data is needed to help refine our efforts to elucidate how release type affects survival, as well as the survival of reintroduced indigo snakes in general for this project.

During 2010-2011, thirteen known mortalities were recorded. Four snakes were killed by cars, four snakes were killed by predators, and five snakes were found dead of unknown causes. Of the four snakes suspected of being predated upon, three mortalities are believed to be from avian predators, and one mortality is suspected to be from a mammalian predator. Figure 32, below, shows a proportional representation of causes. These are also known to be major mortality agents of wild indigo snakes (Hyslop 2009).

Figure 32. 2010-2011 Snake Mortality Causes.



Movements

The telemetry locations and ArcGIS were used to determine the farthest distance observed from the release site for each snake released. These distances were calculated as straight line distance in kilometers (km) (Table 11). The furthest distance recorded from a release site was snake B10, in 2010 it moved 5.94 km.

Table 11. Table showing the snakes identifying number, sex of the snake, release type, the area of its MCP and the farthest distance the snake moved from its release point.

Snake ID	Sex	Release	Home range	Distance to
		method	estimate	farthest point
			(ha)	(m)
A1	M	S	2.9	398
A2	F	Н	69.7	1876
A4	M	S	58.7	912
A5	F	S	30.4	945
A6	M	S	89.2	1290
A8	M	S	3.9	302
A9	F	Н	276.5	2097
B1	M	Н	137.7	2258
B10	F	S	119.8	1417
B2	M	Н	494	3354
B4	M	Н	466.5	5944
B7	M	Н	83.1	1411
B8	M	Н	219.9	2544
C2	F	Н	62.6	1100
C4	F	Н	75.8	736
C5	F	S	77	1165
C6	F	S	197.5	1853
D1	F	Н	25.3	815
D2	F	S	8.7	418
D4	F	Н	28	1296
D5	F	S	0.9	187
D9	F	Н	30.5	1168
F10	F	Н	51.9	1191
F11	F	S	2	249
F2	F	S	46.7	779
F3	F	Н	34.6	789
F5	F	S	8.9	572
F6	F	Н	11.4	427
F7	M	Н	161.7	2099

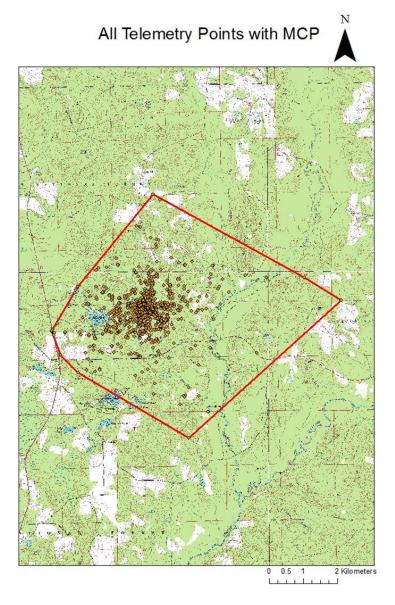
F8	M	Н	81	1853
G2	M	S	80.3	1903
G9	M	S	312.6	3807
H1	M	Н	65.2	1148
H4	M	Н	164.9	3320
H5	M	S	152.4	1904
K2	M	S	1.31	137
K3	M	S	274.9	3579
K9	M	Н	108.3	2513

The mean distance moved for all the snakes combined was 1.57 km (range 0.14-5.94 km). In both years, the males moved further distances than females. The mean distance moved from their release for females was 1.00 km (range 0.19-1.88 km) and for males was 2.14 km (range 0.14-5.94 km). Also hard released snakes tended to move farther from their release points than did soft releases. Mean distance moved for hard releases was 1.9 km (range 0.43-5.94 km) and for soft release 1.26 km (range 0.14-3.817 km). This difference was dramatically increased for males with mean distances of 1.58 km (range 0.14-3.81 km) for soft release and 2.64 km (range 1.15-5.94 km) for hard release.

Home Range Size

To examine home range size 100% Minimum Convex Polygons (MCPs) were created. GPS point data (1445 locations) collected during radio telemetry (16 June 2010 - 14 September 2011) was loaded into AcrGIS 9.3.1 and used to create a geodatabase. The geodatabase was then used to create point shapefiles for data sets. A 100% MCP was created for all the locations in a data set by using Hawth's Tools (Beyer 2004). The areas of the MCP polygons were calculated in hectares (ha). An MCP of all the 1445 locations encompasses an area of 3,344 ha (Figure 33).

Figure 33. Map of 1445 points used for interpretations and their Minimum Convex Polygon.

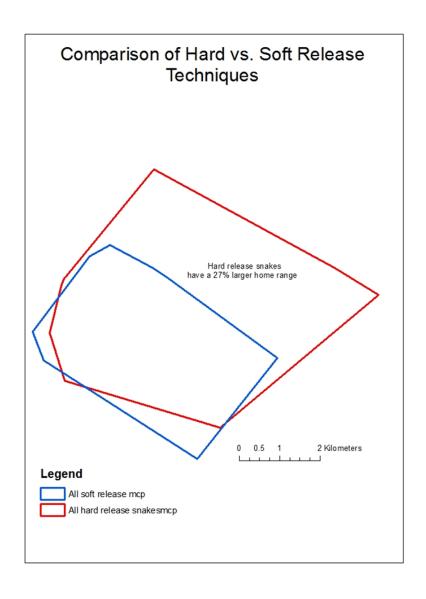


All of the locations for each snake were used to create MCPs that were used to calculate home range areas for each snake (Table 11). The mean area for all the snakes combined was 108.3 ha (range 0.9-494 ha). The mean area used by males was 155.7 ha (range 1.3-494 ha) and by females was 61 ha (range 0.9-276.5 ha). This shows that male home ranges are larger than females. This phenomenon is also seen in wild indigo snakes (Hyslop 2009). One of the reasons in using the soft release technique is to attempt to decrease the active ranges of released snakes. The mean area used by soft releases was 81.6 ha (range 0.9-312.6 ha) and for hard releases 132.4

ha (range 11.4-494 ha), this suggests that penning reduces active ranges.

MCPs were also created by combining point data for all the hard and soft released snakes. These areas were then used for comparison. The area of MCP calculated for all the soft release point data was 1711 ha. Combined hard release data yielded an MCP area of 3040 ha. This is 27% larger than the soft release MCP (Figure 34).

Figure 34. The MCPs of combined soft and hard release data.



Soft Release

Time spent in the pens was calculated for each soft released snake. Number of days in the pens was figured from the time of release to the first time the snake was observed outside the enclosed area. In 2010, it was five days before the first snake escaped from an enclosure. In 2010, the mean number of days spent in the pens was 51 days. In 2011, it was 11 days before the first snake escaped from the enclosure and the mean number of days spent in the pens was 71 days. Maximum time spent in the pens was in 2011; H5 is still in the pens (> 190days). The mean number of days spent in the pens for both 2010-2011 snakes combined was 63 days (range 5-190 days). Both years, there were no noticeable breaches in the fences and it appears the snakes are finding their way out through underground passages. While in the pens, the snakes utilized upturned tree root balls, burned out stump holes, gopher tortoise and small mammal burrows for refugia similarly to snakes outside the pens. While in the pens the snakes utilized the large brush piles created for them. However, snakes were never documented using metal roofing material that was placed in and around the pens. Other snake species were observed under the roofing metal pieces on occasion.

Male/ Female Overlap

The overall goal of the reintroduction is to establish a viable population, therefore male snake ranges need to overlap female snake ranges. To investigate how release technique is possibly affecting this interaction ArcGIS was used compare overlap between hard and soft release males and the female snakes. Since 2010 is the only data that includes the time frame when potential breeding occurs, shapefiles were made for all the 2010 female snakes combined and for the 2010 hard release males and 2010 soft release males. An MCP was created for each of these shapefiles. The males of the two release types were then overlaid and the mutual area clipped out. This area was converted to hectares to allow the examination of overlap. Soft released males overlapped females by 140 ha (Figure 35). Hard released males overlapped females by 395 ha (Figure 36). Male/ female overlap was greater in hard released males, whose overlap with females was 255 ha or 47% more. However, these results do not necessarily mean that hard release males will have more chances to mate. In 2010, only one male was observed in burrows

with female snakes during the breeding season. This male was a soft release and was found in burrows with several of the female snakes during the winter of 2010-2011. Interestingly, so far in 2011, male and female snakes have been observed together twice and both the males in this case were hard release.

Figure 35. Map showing area of overlap between soft release males and all females in 2010.

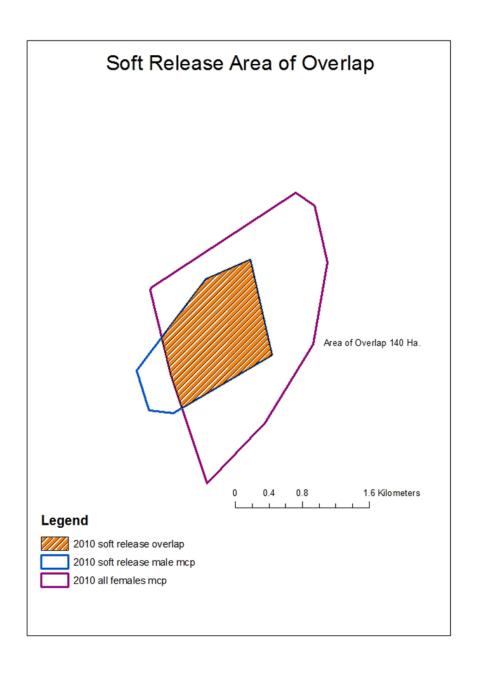
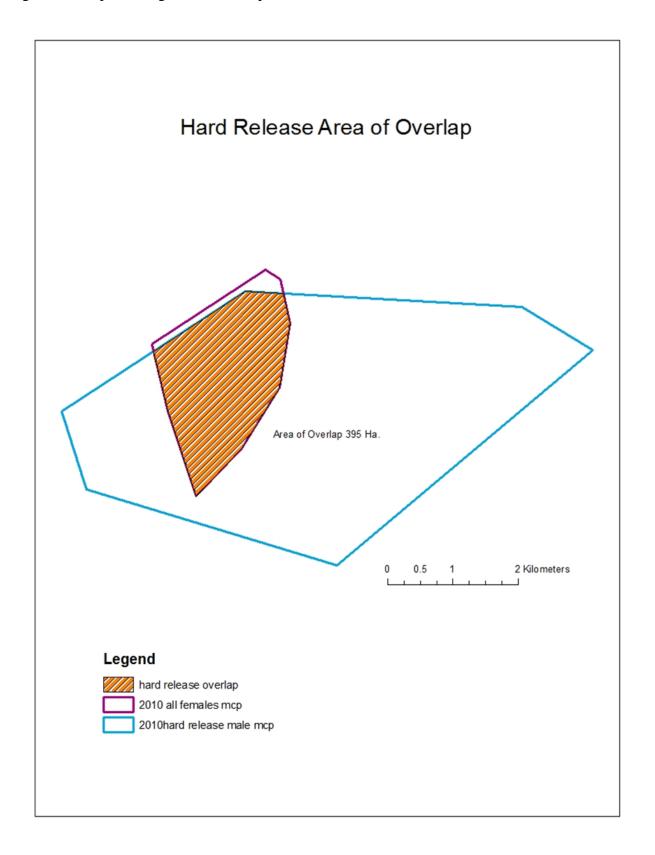


Figure 36. Map showing area of overlap between hard release males and all females in 2010.



Habitat

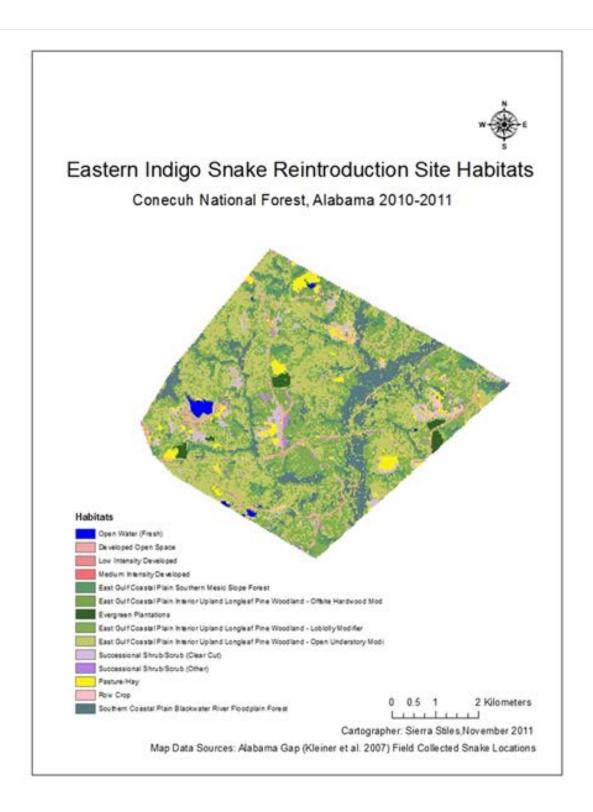
Habitat use by the reintroduced snakes has been diverse, with snakes utilizing a variety of habitats available in the area including both xeric upland and wet lowland habitats that are also known to be used by free-ranging snakes near the source populations (Hyslop 2007). Refugia use has also been similar to that observed near source populations (Hyslop 2007) and has included gopher tortoise, armadillo and small mammal burrows, root balls, stump holes, log piles, rubbish piles and road culverts. Gopher tortoise burrows were used by five of the seven snakes that we radio-tracked through the winter in Conecuh National Forest. The other two radioed snakes used stump holes for overwintering. Several times, snakes located and used the same burrows that had been used by snakes the previous year. This was observed in both soft and hard released snakes and some of the locations were a considerable distance from the initial release site. Snakes have often used habitats and refugia in close proximity to roads and have occasionally been encountered crossing both dirt and paved roads. "Do Not Harm" signs have been placed around the area to encourage drivers to avoid running over the snakes (Figure 37).

A map of the habitats accessible to snakes in the release area (Figure 38) was created using location data collected for the 38 reintroduced snakes by radio-tracking individuals during 2010-2011. All radio location waypoints were uploaded into ArcGIS 9.3 and were used to create a point shapefile. A 100% minimum convex polygon or MCP was created using Hawth's Tools (Beyer 2004). Land cover data from the Alabama Gap Analysis Program (AL-GAP) (Kleiner et al. 2007) was downloaded from the AL-GAP website (www.auburn.edu\gap) and uploaded as an ArcGIS 9.3 layer. Spatial Analyst was used to extract by mask only the portion of the raster layer that overlapped with the MCP representing the area where snakes were located.

Figure 37. Signage placed in key areas within Conecuh National Forest to alert motorists to the presence of the eastern indigo snake. Signs were sponsored by the Alabama Department of Conservation and Natural Resources, U.S. Forest Service, and U.S. Fish and Wildlife Service.

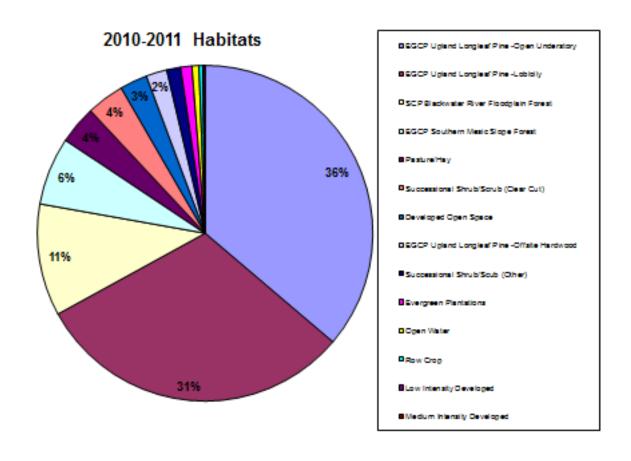


Figure 38. Map showing the habitats present at the reintroduction site for Eastern Indigo snakes (*Drymarchon couperi*) in Conecuh National Forest, Alabama.



AL-GAP metadata was used to identify the pixel size of the raster layer (30 meters x 30 meters) and calculate the area of one pixel (900 square meters). Data in the attribute table was used to determine the number of habitat types present in the area used by the snakes. By multiplying the area of each pixel by the pixel count for each habitat, I determined habitat composition and proportion of occurrence (Figure 39).

Figure 39. Pie chart illustrating the proportion of habitat types present on the 3,334 hectare reintroduction site in Conecuh National Forest, Alabama.



Behavior

Many interesting behaviors have been observed in the released snakes. After release, snakes have been observed rattling their tails, hissing and inflating their bodies in a threat pose when approached. Snakes have often been seen laying right inside the entrance to their underground retreat. They have also been observed "periscoping", a behavior where a snake pokes up only its head from the retreat and looks around. Tree climbing behavior, up to 10 meters off the ground, was observed in a few snakes in 2010, but has not been documented in 2011. However, several times snakes were observed in the shrub layer up to three meters off the ground.

Feeding

To ensure that snakes inside the pens had access to food, soft release snakes were given a snake as a prey item approximately once a week while they were in the pens. These prey items were set in front of the indigo snakes if they were on the surface. If the indigo snake was in a retreat, the prey snakes were encouraged to go into it as well. All feedings in which the indigo snake was observed resulted in the indigo snake successfully catching and consuming the prey. After a snake left the pens for the first time, feedings for that snake were discontinued. Snakes were chosen as the preferred prey since they could be left without undue harm to the prey or the indigo snake and indigo snakes outside the pens showed a propensity for eating them.

Radio tracking the snakes began with observing a feeding event. The very first indigo snake tracked in 2010 was found consuming a copperhead (*Agkistrodon contortrix*). The crew of the television show, Discovering Alabama, was there and shot footage of the feeding event for use in an upcoming episode that will feature indigo snake conservation. Including that first day we have observed feeding nine times (See Table 12). Prey items were primarily other snake species however one indigo snake was observed eating a southern toad (*Anaxyrus terrestris*). Snakes encountered during tracking on the surface are often observed slithering around flicking their tongues rapidly and prodding under logs and in holes in what seems like a foraging behavior. One indigo snake exhibited this foraging behavior in a failed attempt to catch a black racer (*Coluber constrictor*). Snakes in the pens also exhibit this behavior when approaching a prey

item given to them.

Table 12. Table showing observed prey items and number of observations captured by indigo snakes outside pens.

Species of Prey Item	Number of			
	observed predations			
Southern copperhead (Agkistrodon contortrix)	3			
Gray rat snake (Pantherophis spiloides)	3			
Corn snake (Pantherophis guttatus)	1			
Pygmy Rattlesnake (Sistrurus miliarius)	1			
Southern toad (Anaxyrus terrestris)	1			

Education

Education programs about indigos snakes in general and the reintroduction project in specific have been ongoing in both formal, (ALAPARC, Auburn University Chapter of the Society for Conservation Biology, Auburn University Chapter of the Wildlife Society, Gopher Tortoise Council, Montgomery Sierra Club, and SEPARC) and informal, (over lunch at the country store or standing around the check station during hunts) settings. Public feedback from both arenas has been surprisingly positive.

Fire-driven Ecological Restoration

The US Forest Service fire team at Conecuh National Forest conducted two prescribed burns in 2011. In order to maintain the habitat quality that the release site was chosen for, a 2-3 year burn interval was recommended for the compartment when the release site was chosen (Guyer and Johnson 2007). The first burn excluded the soft release pens and occurred 29 January 2011. The objective was to reduce fuel loads around the pens allowing greater flexibility in scheduling an early growing season burn within the pens. A total of 476.7 hectares (1178 acres) in Compartment 28 were burned (Figure 40). The last burn in this compartment prior to this had occurred on 17 February 2007.

In addition to maintaining habitat quality at the release site, another objective of this burn was to reduce fuel loads around the pens, allowing greater flexibility in scheduling an early growing season burn within the pens. To protect the fence from damage a fire line was raked around the perimeter of the fence; excessive heating of the fence would destroy the galvanized coating leading to premature rusting and deterioration (Figures 41 & 42). Outcome of the fire was a success in reducing fuel loads and encroaching woody midstory in the release area and surrounding the pens (Figure 43). Four radio-tagged snakes were known to be in the vicinity during the burn and once the forest floor had cooled all were tracked and found to have survived without injury; all were noted to have been underground at the time. The day of the burn was sunny and warm and one snake was observed active on the ashy forest floor within 2-3 hours following the fire (Figure 44).

Pen interiors were burned 10 and 11 May 2011. Forest floor debris was raked away from fence perimeters and wading pool edges in the weeks preceding the burn and a leaf-blower was used to clear additional accumulations on the day a pen was burned to prevent damage to these structures. Three pen interiors were burned on day one with the remaining three being burned the second day. These burns took place approximately one week prior to the 16 May 2011 snake release. No snakes were using the pens at this time.

Prescribed burns were conducted to promote the restoration of the longleaf pine forest that once dominated the Southeastern United States (Figures 45 & 46).

Figure 40. U.S. Forest Service personnel using drip torches to ignite forest floor fuel around the perimeter of the snake pens.



Figure 41. Successful ignition of leaf litter and small woody debris outside the snake pens.



Figure 42. Fire moving through the pine-hardwood forest at the snake realease site.



Figure 43. Ashy blackened forest floor covered immediately after the fires from the prescribed burn had died out.



Figure 44. An eastern indigo snake active on the surface of the blackened forest floor within 3 hours after the area had been burnt, January 2011. This snake was known to reside in a nearby root channel.



Figure 45. Longleaf pine are adapted to survive fires that sweep through forests.



Figure 46. Longleaf pine forest immediately following a prescribed fire. Young longleaf pine that have survived a prescribed fire stand in stark contrast to the dark ashy forest floor.



Population growth projection

The purpose of captive husbandry is to facilitate the production of a sufficient number of young snakes so that a viable population of the eastern indigo snake may be established. But what is the time line for this and how many snakes may be required? To examine these questions a projection of population growth has been calculated based on a series of assumptions that were formed on data gathered to date. As the project proceeds and data are gathered these projections will be refined.

Assumptions:

- 1) First year snake survivorship is 50% i.e. within the first year following release approximately 50% of the young snakes will have succumbed to some type of mortality. Based on survival estimates calculated for overall survivorship for 2010 releases.
- 2) Subsequent year survivorship is 90%. By the time the snakes have reached their 2nd year post release they have increased survival according to the 2008 cohort's survival estimates from 2011.
- 3) Sex ratio of hatchlings is 1:1.
- 4) Maturity for both female and male snakes is reached in four years. This point of maturity is more important for the female demographic than the male.
- 5) Average female reproductive output was calculated at 7.4 hatchlings per female. This is based on the 2008-2011 mean hatchlings to female ratio.

A total of 30 hatchling snakes may be reasonably attained and thus the calculations for the remaining 6 years of husbandry produced snakes is based on this figure. During the first year of release mortality is expected to be high, around 50% but after the first year the rate is expected to drop to about 10% and survivorship calculations are based on these numbers. The two year old snakes will reach maturity by five years of age (table 13, year 3) and females from the first release should then begin contributing young to the population at that time "Population

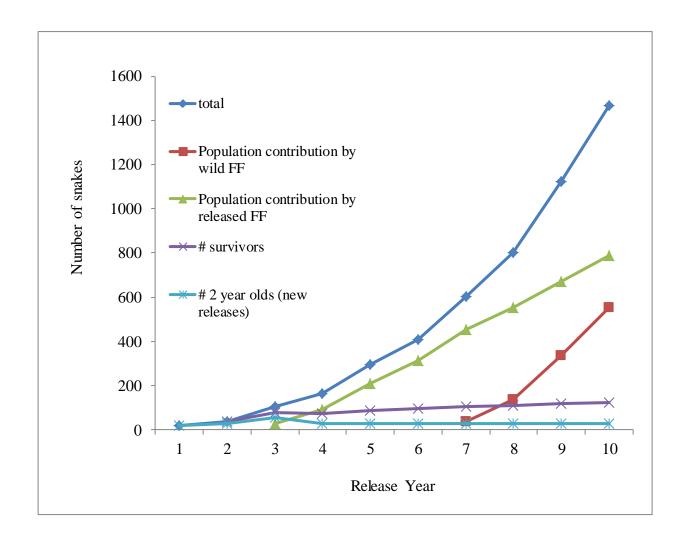
contribution by released FF". By year 7 offspring produced from this first reproductive cycle in the wild will be maturing and reproducing, this is represented by the row entitled "Population contribution by wild FF".

Significant population growth begins around year six and climbs steadily thereafter. In year six 32 mature females will be present and by year 10 the number will be 342 and the population, by this time, is expected to be self-sustaining (Table 13, Figure 47). These numbers serve as a model and starting point upon which modifications can be made. Calculations are based on a static increase in the population and a static mortality rate. In reality these rates will change from year to year as determined by the myriad of ecological factors under which these snakes will be required to live. Stochastic weather events, fluctuating prey base, shifting predation dynamics, variable indigo snake densities, occupation of optimum and marginal habitats, disease and parasitism, and other aspects of the ecology and natural history of indigo snakes will affect population growth and expansion.

Table 13. Population growth projection for the eastern indigo snakes released onto Conecuh National Forest over a 10-year time span.

Indigo snake population projections beginning with Spring 2010 (year 1)											
year	1	2	3	4	5	6	7	8	9	10	
Husbandry produced											
snakes											
# 2 year olds (new											
releases)	17	30	54	27	30	30	30	30	30	30	
# survivors (old											
releases)		8.5	15	27	13.5	15	15	15	15	15	
bold = cohort maturity			7.7	13.5	24.3	12.2	13.5	13.5	13.5	13.5	
				6.9	12.2	21.9	10.9	12.2	12.2	12.2	
					6.2	10.9	19.7	9.8	10.9	10.9	
						5.6	9.8	17.7	8.9	9.8	
							5.0	8.9	15.9	8.0	
								4.5	8.0	14.3	
									4.1	7.2	
										3.6	
#/year by age and class											
# immature	17	38.5	69	54	43.5	45	45	45	45	45	
# mature FF + MM			7.7	20.4	42.6	50.5	66.6	66.6	73.4	79.5	
# mature FF			3.8	10.2	21.3	25.3	33.3	33.3	36.7	39.8	
" matare i i			0.0			20.0	00.0	00.0		00.0	
Population contribution											
by released FF											
reproductive input*											
(hatchlings)			28.3	75.4	157.8	187	246.3	246.3	271.5	294.2	
# immature				14.2	37.7	78.9	93.5	123.1	123.1	135.8	
					12.8	33.9	71	84.2	110.8	110.8	
					_	11.5	30.5	63.9	75.7	99.7	
bold = cohort maturity						1110	10.3	27.5	57.5	68.2	
bold – concit matarity							10.0	9.3	24.7	51.8	
								3.3	8.4	22.3	
									0.4	7.5	
										7.5	
// income a to una			00.0	00.0	000.0	044.0	444.0	F47.F	F04.0	040.5	
# immature	-		28.3	89.6	208.2	311.3	441.3	517.5	581.2	640.5	
# mature FF + MM							10.3	36.8	90.6	149.7	
# mature FF							5.2	18.4	45.3	74.9	
B 10 10 10											
Population contribution by wild FF											
Reproductive input 2											
(hatchlings)							38.3	136.2	335.3	554.0	
snake totals by year	1	2	3	4	5	6	7	8	9	10	
# immature	17	38.5	97.3	143.6	251.7	356.3	524.6	698.6	961.5	1239.5	
mature FF + MM			7.7	20.4	42.6	50.5	76.9	103.4	164.0	229.2	
total	17	38.5	105.0	164.0	294.4	406.8	601.5	802.0	1125.5	1468.7	

Figure 47. Population growth curve of eastern indigo snakes released onto Conecuh National Forest over a 10-year time horizon. Data used to generate the growth projections may be found in Table 13.



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Appendix 1. Eastern indigo fact sheet prepared for the media event.

Eastern Indigo Snake Fact Sheet

Scientific Name: Drymarchon couperi

Other common names: blue indigo snake, gopher snake, blue bull snake



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Is the eastern indigo snake dangerous or venomous?

No. The eastern indigo snake is not venomous. It is harmless. Indigos seldom bite people, but they do bite their prey, enemies, or each other during aggressive competition between males. Although the indigo snake is harmless, because of its protected status this is a snake that you are not allowed to handle without a permit.

How do you identify the eastern indigo snake?

The most notable feature of the eastern indigo snake is the lustrous, glossy, iridescent, blue-black coloration of the head and body. The eastern indigo snake is the longest snake in North America and may reach a size of 8.5 ft (2.6 m) and a weight of 11 lbs (5 kg) for males; females are smaller (6.5 ft (2 m); 6.5 lbs (3 kg)). While the large size of the indigo snake is impressive most individuals seen are around 5-6.5 ft (1.5-2 m) in length. Snakes may be blue-black over the entire body or may have red, reddish-orange, or cream coloration on the chin, throat or cheeks. Scales of the body are smooth.

Other snakes with black coloration include the black pine snake, black racer, kingsnake, coachwhip, and eastern hognose. The black pine snake (*Pituophis melanoleucus lodingi*) is found only in extreme SW Alabama and adjacent SE Mississippi and is often entirely black, but a duller black. Black pine snakes may grow to be up to 6 ft (1.9 m) in length, but lengths of 4-5 ft (1.2-1.6 m) are more typical. Scales of the black pine snake have a keel down the center.

The black racer (*Coluber constrictor*) may reach a length of 6 ft (1.9 m) but is often shorter (4-5 ft), and has an entirely black body with a white chin. Black coloration of the racer tends to be a slaty-gray, dull black, and the body is thin. As with the indigo, body scales of the racer are smooth.

The eastern kingsnake (*Lampropeltis getula getula*) has an overall black coloration but is easily distinguished from the indigo snake by the yellow to cream-colored bands traversing the body. Kingsnakes may be large, up to 6 ft (2 m), but are generally 3-4 ft (0.9-1.2 m) in length, and have smooth scales. Entirely black kingsnakes are unknown.

Eastern coachwhip (*Masticophis flagellum*) rivals the indigo snake in length (maximum reported size of 8.5 ft (2.6 m)), but most seen are no larger than 5 ft. (1.5 m). Coachwhips are thinner than indigos and the body is bi-colored. The head, neck, and fore-quarter of the body is black but then grades into brown for the remaining three-quarters of the length of the snake.

Eastern hognose (*Heterodon platirhinos*) is a shorter, chunkier snake (20-33 in.; 51-84 cm) and some, but not all, individuals may be black. The hognose has keeled scales and an upturned snout.

Where do eastern indigo snakes live?

Historically, the eastern indigo snake lived throughout Florida, the coastal plain of southern Georgia, extreme south Alabama, and extreme southeast Mississippi. Today the indigo snake survives in peninsular Florida and southeast Georgia, persists in the Florida panhandle, but in low numbers, but has been extirpated from Alabama and Mississippi.

What do they eat?

Indigo snakes are known to feed mainly upon other snakes, turtles, mammals, frogs, birds, and lizards. Unusual food items, in comparison to that of other snakes, include small tortoises and all venomous snake species native to the Southeastern US.

How do they kill their prey?

Eastern indigo snake is not a constrictor therefore it overpowers its prey with its muscular jaws, and often larger size, consuming the prey head-first.

Do they get bitten when they eat rattlesnakes or other venomous snakes?

Indigo snakes may on occasion be bitten by the venomous snake on which it is feeding.

Are they immune to the venom?

Observations of biologists that have seen indigo snakes being bitten by eastern diamondback rattlesnakes suggest that the indigo snake is mostly immune to rattlesnake venom.

What is the habitat of the eastern indigo snake?

In peninsular Florida a wide variety of habitats are used as cold winter temperatures are unusual. In more northerly portions of the range the indigo snake occupies sandhills during the winter, using gopher tortoise burrows as a retreat from cold temperatures. During the warmer months snakes move to nearby wetlands.

How big is their territory?

Eastern indigo snake has a large territory based on studies conducted on male snakes. Range

of southeastern Georgia snakes were found to be as large as 3,000 acres (1,600 ha), and one male was noted to move a distance of about 13 miles (22 km).

When do indigo snakes breed?

Eastern indigo snakes breed from October to February. During this time the snakes are found in sandhills and, although this is the peak of winter, indigo snakes are active at temperatures (50-60 F) that are typically too cool for other snake species.

When do indigo snakes lay eggs, where do they lay eggs?

Eggs are typically laid during May or June.

How many eggs are laid and how large are the eggs?

Females lay a single clutch of eggs of 4 to 12 eggs. Little information is known on nest sites but gopher tortoise burrows are suspected to be a favored nesting location. The eggs are large, nearly 3 in. (75 mm) in length, are elongate-oval, cream-colored, with a granular surface.

How large are hatchling indigo snakes?

Large eggs produce large hatchling snakes. Hatchling snakes are about 16 in. (40 cm) long, and about 1.5 oz. (40 g).

What do young snakes eat?

Diet of young snakes is very similar to that of the adult snakes; the small snakes simply eat smaller prey.

Why have they disappeared from Alabama?

Throughout the southeastern United States the eastern indigo snake has been associated with the longleaf pine ecosystem, and this ecosystem has experienced severe reductions and alterations. The loss, fragmentation, and alteration of the longleaf pine ecosystem are most likely the major underlying cause for the disappearance of the eastern indigo snake in Alabama. As the forests were converted, gopher tortoises were reduced, and the gopher tortoise burrows, upon which the indigo snakes depend, were reduced. Eastern indigo snakes were an unintended casualty of the persecution of the eastern diamondback rattlesnakes too. The now-outlawed practice of gassing gopher tortoise burrows to drive out rattlesnakes also killed or seriously debilitated many other organisms occupying the burrow, including the docile indigo snake.

The eastern indigo snake is protected. What does this mean?

Under the Endangered Species Act the US Fish and Wildlife Service has listed the eastern indigo snake as "threatened." With this designation, individuals who harm or harass the indigo

snake may be subject to fines and jail terms. The Alabama Department of Conservation and Natural Resources includes the eastern indigo snake on the non-game species regulation list, thus providing an additional level of protection.

What is the importance of the eastern indigo snake, and why release snakes into the wild?

The generic name *Drymarchon* means "forest ruler" and that is a fitting title for this snake. Being at the top of its food chain, a healthy population of eastern indigo snakes in a longleaf pine forest setting is an indication of an ecologically functional forest. The loss of the indigo snake from Alabama and other areas is the loss of a significant part of the biodiversity of the forest. Longleaf pine forest restoration is underway in many localities across southern Alabama, thus establishing a foundation for the reintroduction of the eastern indigo snake. To return the eastern indigo snake to the south Alabama landscape is to restore a piece of the natural history of the state. Releasing snakes into the wild is a step toward establishing populations, or augmenting populations and addressing recovery objectives within the recovery plan of the USFWS. Once recovery objectives have been met, the eastern indigo snake would be considered recovered and be removed from the list of threatened species.



For more information or to report a sighting contact:

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