Evaluation of Movement Patterns and Space Use in Reintroduced Eastern Indigo Snakes
(Drymarchon couperi) in the Florida Panhandle

by

Sara E. Piccolomini

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Approved by

Dr. Mary Mendonca, Chair, Professor, Department of Biological Science
Dr. Lora Smith, Co-chair, Jones Center at Ichauway
Dr. David Steen, U.S. Fish and Wildlife Research Institute, Florida Fish and Wildlife Conservation Commission
Abstract

The Eastern Indigo snake (*Drymarchon couperi*; EIS) is a federally threatened species of colubrid snake endemic to the U.S. Southeastern Coastal Plain for which reintroduction efforts are currently ongoing to reestablish populations where EIS are known to have been extirpated. To evaluate reintroduction success, post-release monitoring of movement patterns is considered essential. However, methods for quantifying animal movement patterns and space use are typically restricted to two-dimensional metrics, which ignore topographic variation in space use. Moreover, most monitoring studies do not attempt to account for the influence of demographics (i.e., sex and age class) on post-release movement behavior in reintroduced populations—despite the fact that interactions with conspecifics may influence spatial decision-making. In this study, we examined percent survival, home range size, rate of dispersal and habitat use of reintroduced EIS at Apalachicola Bluffs and Ravines Preserve (ABRP), a reintroduction site in the Florida Panhandle. Following two groups of released EIS in 2017 and 2018, a total of 32 snakes (19 males, 13 females) were radio-tracked 1-2 times a week. We recorded fate of all snakes up to one year post-release to estimate survival as a proximate, short term measure of success. We used location data to calculate two-dimensional 100\% minimum convex polygon (MCPs) home range estimates and looked at the effect of sex on home range size to determine if patterns of space use aligned with those reported for wild populations of EIS, and we calculated three-dimensional 100\% MCP home ranges to look at how inclusion of topographic variation of the unique landscape of ABRP might influence home range sizes of EIS. Kernel density estimates (KDEs) were calculated to describe areas of core use. We also calculated an index of dispersal to
investigate the potential influence of demographic factors (i.e., age class, sex) and release year on post-release movements.

The Kaplan-Meier survival estimates for EIS in 2017 was 45% after 49 weeks, and only 2 of the 12 snakes released were known to have survived (the fate of 4 additional EIS was not confirmed because transmitter signals were lost). In 2018, survival estimates for EIS were 46%, where 7 of the 20 snakes released were known to have survived after 42 weeks, and an additional 5 EIS could not be located (signals were lost). At the end of the two year study, 9 of the 32 snakes released were confirmed alive, resulting in a 28% estimate of survival.

Two-dimensional MCP home ranges for 16 EIS (5 males and 11 females) using the first 11 relocations averaged 15.0 ha, but varied among individuals (1.6 ha to 34.8 ha). There was a significant effect of sex on home range size (F= 4.129; df= 9; p=0.01) with males, on average, maintaining larger home ranges (ranging from 9.2 to 34.8 ha and averaging 23.2 ha) than females (ranging from 1.6 to 28.1 ha and averaging 11.56 ha), respectively. Three-dimensional MCP home ranges for 28 EIS (15 males, 13 females) using all relocations from snakes tracked 1-2 times per week ranged from 1.6 ha to 243.82 ha with a mean of 61.78 ha (Appendix 1). Our paired t-test to compare whether 2D and 3D revealed no difference (df = 54; p = 0.9551). Average 95 and 50% kernel density estimates and areas of core use for 8 EIS (2 males, 6 females) were 96.1 m (ranging from 15.2 to 345.8 m) and 16.0 m (ranging from 3.0 to 46.7 m), respectively (Table 3). We found a three-way interaction of sex, age class and release year (F= 4.961; df = 9) where young males released in year 2 dispersed more quickly (32.3 m/day) than all other groups (p = 0.004).

Snakes tracked throughout the study exhibited a seasonal shift in habitat use during the winter months, where EIS were exclusively found in upland sandhills habitat as opposed to the
rest of the year when they were found using both upland sandhills and ravine/wetland habitat. These data align with season shifts in habitat use reported for wild populations of EIS, suggesting reintroduced EIS in our study exhibiting similar habitat use similar to wild snakes.
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List of Abbreviations

2D    Two-Dimensional
3D    Three-Dimensional
ABRP  Apalachicola Bluffs and Ravines Preserve
AIC   Akaike Information Criterion
CFZ   Central Florida Zoo
CNF   Conecuh National Forest
DEM   Digital Elevation Map
EIS   Eastern Indigo Snake
ESA   Endangered Species Act
KDE   Kernel Density Estimates
MCP   Minimum Convex Polygon
OCIC  Orianne Center for Indigo Conservation
TIN   Triangular Irregular Networks
TNC   The Nature Conservancy
USFWS United States Fish and Wildlife Service
Chapter 1

INTRODUCTION

Conservation background

Loss of habitat is one of the primary causes of ongoing biodiversity declines around the world (Wilson 1999; Noss et al. 2006). Reduction of natural habitat by anthropogenic land-use often leaves imperiled species unable to persist—thus, furthering their already- eminent risk of extinction (Bruner et al. 2001; Ervin 2003; Gibbons et al. 2000.) In the United States, habitat loss has been identified as the primary cause of decline in >97% of imperiled herpetofauna (Wilcove et al. 1998). In the U.S. Southeastern Coastal Plain, which supports the highest diversity of herpetofaunal species in the country, an estimated 70% of amphibian and reptile species are in peril—many of which are intimately tied to longleaf pine (Pinus palustris) ecosystems (Trani-Griep 2002; Guyer and Bailey 1993; Means 2006).

Originally comprising nearly 30 million hectares, longleaf pine forests were once dominant throughout the U.S. Southeastern Coastal Plain (Frost 1993; Landers et al. 1995; Van Lear et al. 2005). Having suffered a near 97% loss, the longleaf pine is now considered one of the most imperiled ecosystems in the world (Noss and Peters 1995). In addition to severe loss in area, the exclusion of fire from the landscape led to further degradation of remaining intact longleaf habitat (Frost 1993; Van Lear et al. 2005). Without frequent, low-to-moderate-intensity fire, hardwood encroachment produces unsuitable habitat for many endemic species (Engstrom et al. 1984; Mushinsky and McCoy 1985; Van Lear et al. 2005).

Protection and maintenance of high-quality habitat has been identified as the primary focus of conservation for species in decline (Van Lear et al. 2005), which is based on the
assumption that restoration of degraded ecosystems promotes a return of native biotic communities to the landscape (Litt et al. 2001). However, in some cases, densities may be too low to rebound naturally or populations may be completely extirpated (Dodd 1987). As a result, establishment of additional populations may be necessary to aid recovery efforts.

Species reintroductions have become a popular tool to conserve imperiled species (Germano and Bishop 2009) and have been employed for a variety of vertebrates in an attempt to delist many species, spanning from smaller taxa like crested toads (Peltophryne lemur) to large megafauna like the Arabian oryx (Oryx leucoryx (Jachowski et al. 2016)). The term ‘reintroduction’ refers to the translocation of individuals to areas within their native range where they have been previously extirpated (Fischer and Lindenmayer 2000; IUCN/SSC 2013); the end-goal of such projects is reestablishment of self-sustaining populations. However, achievement of this benchmark can be difficult (Dodd and Siegel 1991) as reintroductions are complicated, expensive, and laborious.

For reintroduction to be considered an appropriate tool to help combat defaunation, habitat at sites being considered for release must first be evaluated in order to determine whether they are suitable to support the species. Once habitat has been deemed suitable and threats that originally lead to the species becoming extirpated have been alleviated, sites are therefore considered to be suitable for reintroduction.

Following release into the wild, reintroduced animals may exhibit abnormal/excessive dispersal—which can result in reduced fidelity to release sites and decreased population viability. In an attempt to minimize excessive dispersal, soft-release methods (i.e., enclosing animals at release sites for some period of time before freeing them (e.g. Wallace 2000: Kingsbury and Attum 2009)) are often employed. It has been suggested that these methods
increase interactions with conspecifics thought to influence settlement behavior (Campomizzi et al. 2008). In contrast, when animals are released directly into the wild it is considered to be a hard-release. However, despite the popularity of soft-release methods, they can be expensive to implement and individuals may escape enclosures—making it difficult for researchers to accurately evaluate their influence on post-release movements.

Once reintroductions have been implemented (i.e., individuals have been released into the wild), post-release monitoring is essential to evaluate the status of these efforts as they work towards establishing self-sustaining populations (Nichols and Armstrong 2012; Gitzen et al. 2016). It is often considered a sign of success in the short term when reintroduced individuals not only survive, but are observed behaving similarly (i.e., via movement and habitat selection) to wild individuals (Stiles et al. 2013); while signs of reproduction at recipient sites are considered to be indicative that long-term goals (i.e., reestablishment of self-sustaining populations) of the project are being met (Dodd and Siegel 1991). However, for species with certain life history and behavioral characteristics, monitoring for success can be difficult. For example, reproductive behavior in long-lived species may be delayed (Mertz 1971) and the large spatial requirements of elusive, free-ranging species such as snakes can result in large movements and overall decreased fidelity to release sites (Steen 2010; Durso et al. 2011)—further compromising the ability of researchers to accurately monitor for success.

**Study species**

The Eastern Indigo Snake (*Drymarchon couperi*; EIS) is a large-bodied (>2m), bluish-black colubrid native to the U.S. Southeastern Coastal Plain, most notable for its extensive movements and large home ranges (Hyslop et al. 2009). EIS exhibit male-biased sexual size
dimorphism and females reproduce annually once they reach sexual maturity [(3-4 years) Speake et al. 1978; Stevenson et al. 2009]. The breeding season for this species spans from October through March (Groves 1960; Speake et al. 1978; Stiener et al. 1983) and gestation lasts 100-150 days (Smith 1987; O’Connor 1991) with eggs typically being laid in May and June (Moulis 1976; Stiener et al. 1983; Moler 1992; Speake et al. 1987). The EIS is a diurnal, active-foraging species that preys on a multitude of vertebrates year-round and is considered an apex predator in longleaf pine ecosystems; however, a large portion of their diet consists of other snakes (Stevenson et al. 2010; Steen et al. 2016).

Historically, EIS were widespread throughout the U.S. Southeastern Coastal Plain (Enge et al. 2013) with their occurrence often overlapping that of the Gopher Tortoise (Gopherus polyphemus) in xeric longleaf pine sandhills (Diemer and Speake 1983). EIS use different vegetation types such as oak sandhills, pine and scrub flatwoods, dry prairie, freshwater wetlands, and mixed hardwood forests in addition to xeric sandhills (Speake, McGlincy and Colvin 1978; Hyslop 2009; Stiles 2013). During the winter months the presence of thermally stable refugia, such as Gopher Tortoise burrows, drives their almost exclusive use of upland sandhills habitat; for the remainder of the year EIS largely shift to utilizing hydric bottomlands (i.e., wetlands, creek drainages, floodplain swamps (Smith 1987; Hyslop et al. 2014)). Further, the breeding season drives increased movements in male EIS, as they must expand their home ranges in order to acquire access to multiple mates (Layne and Steiner 1996; Hyslop et al. 2014).

The sizeable home ranges that EIS maintain are thought to be largely attributable to both seasonal shifts in habitat use and reproductive behavior (Hyslop et al. 2014).

By 1978, EIS had experienced dramatic population declines (USFWS, 1978) with habitat loss, over-collection for the pet trade, and mortality associated with gassing of Gopher Tortoise
burrows for the collection of Eastern Diamondback Rattlesnakes (*Crotalus adamanteus* (Speake and McGlincy 1981)) ultimately leading to their listing as a threatened species by the U.S. Fish and Wildlife Service under the U.S. Endangered Species Act (ESA) (Enge et al. 2013; USFWS, 1978). In 1982, a recovery plan was established to maintain and protect existing populations of EIS. The recovery plan outlined the need to establish additional EIS populations in portions of the historic range (Speake et al. 1982). Despite listing under ESA and development of a recovery plan, EIS populations continued to decline (USFWS 2008). As a result, implementation of reintroductions for EIS were put into action to help recover the species in areas where populations were believed to be extirpated.

In accordance with recovery plans for the species, sites being considered for reintroduction must be comprised of large, intact longleaf pine habitat to support the spatial requirements of EIS and must also contain established populations of Gopher Tortoises, with which EIS are closely tied (USFWS 1978; Hyslop et al. 2014). In the late 1970’s-1990’s, captive EIS were raised and released throughout parts of southern Alabama and Georgia thought to be suitable for reintroduction; however, the number of snakes released was small and no snakes have recently been recaptured from this effort (Guyer et al. 2019). More recently, a second attempt to reestablish populations of EIS in Alabama began in Conecuh National Forest (CNF) in 2010. The CNF reintroduction effort is ongoing, and evidence of persistence have been reported (i.e., the recapture of multiple individuals at the release sandhills during the winter/breeding months, recent evidence of successful breeding (J. Godwin, Alabama Natural Heritage Program, personal communication).

Following the framework of the CNF reintroduction, an attempt to reestablish a population of EIS was initiated at The Nature Conservancy’s (TNC) Apalachicola Bluffs and
Ravines Preserve (ABRP) in Liberty County, Florida. As with the CNF reintroduction program the intention at ABRP was to release of 30 captive born EIS every year for the next 10 years beginning in 2017. The last known records of EIS in ABRP occurred more than 25 years ago; the last confirmed individual observed in the Florida Panhandle occurred approximately 150km from ABRP in 1999 (Enge et al. 2013). The extirpation of EIS from ABRP and the surrounding lands was likely caused by the conversion from natural longleaf sandhills to pine plantation, and the suppression of fire (Enge et al. 2013). Following its acquisition by TNC, ABRP has undergone intensive restoration over the last two decades. The return of frequent fire, establishment of longleaf pine and native ground cover, especially wiregrass (*Aristida stricta*), has resulted in a recent increase in Gopher Tortoise abundance on the property (D. Printiss, The Nature Conservancy, personal communication), making it suitable for EIS reintroduction. In this thesis, I report survival (snakes known to be alive approximately 1 year post-release), seasonal habitat use and examine movement patterns and space use of EIS as a part of a management effort to reintroduce snakes at TNC’s ABRP; I use these data to judge preliminary measures of reintroduction success.
Chapter 2

Post-release monitoring of reintroduced Eastern Indigo Snakes (*Drymarchon couperi*) to the Florida Panhandle: Evaluating survival, dispersal and space and habitat use of reintroduced snakes

INTRODUCTION

Habitat loss has been identified as the primary driver of imperilment in herpetofaunal species worldwide (Wilcove et al. 1998; Alford and Richards 1999). In the United States, the near 97% loss of intact longleaf pine (*Pinus palustris*) forests in the southeastern Coastal Plain has resulted in a decline in abundance of roughly 70% of southern herpetofaunal species, (Gibbons et al. 1997) including the federally threatened Eastern Indigo Snake (*Drymarchon couperi*; EIS).

EIS is the largest (>2m in length) native snake species in the United States and is considered an apex predator in longleaf pine ecosystems where the species consumes a multitude of prey (Stevenson et al. 2010). In 1978, the EIS was listed as threatened under the U.S. Endangered Species Act (ESA) after experiencing significant population declines resulting from extensive habitat loss, overcollection, and mortality associated with the gassing of Gopher Tortoise burrows (Speake 1993), where EIS frequently seek shelter in winter—most notably in the northern portions of their range. Restoration of large parcels of intact longleaf pine habitat is critical to conservation of the EIS, however in some cases it may be too late for these efforts to promote populations to rebound naturally. Therefore, reintroducing individuals to areas within their range where they have been previously extirpated is one strategy being implemented for recovery of EIS (Fischer and Lindenmayer 2000) in hopes of reestablishing self-sustaining populations in these areas.
In order to assess success of a reintroduction, in addition to measuring survival of individuals, implementation of monitoring programs aimed at quantifying movement and activity patterns following release into the wild is essential (Nichols and Armstrong 2012; Gitzen et al. 2016). Movement and activity patterns are considered fundamental characteristics of an animal’s life history (Nathan et al. 2008), often reflecting the availability of resources (i.e., food, mates and shelter) throughout the landscape (Waser and Wiley 1979). When reintroduced animals exhibit similar movement behavior to those of wild animals (i.e., home range size, dispersal patterns and habitat use) it is often considered a sign of preliminary success (Armstrong and Seddon 2008) whereas abnormal movement patterns (i.e, excessive dispersal) often lead to decreased population viability at release sites—making attainment of established self-sustaining populations difficult.

One of the biggest reasons thought to be responsible for excessive dispersal following release into the wild for reptile reintroduction efforts is poor habitat quality at release sites (Germano and Bishop 2009; Stiles 2013). EIS not only have large spatial requirements, but they also use multiple different habitat types (e.g., wetlands, floodplains and upland sandhills, pine forests, or scrubby flatwoods). If either upland or wetland habitat is lacking at release sites, it may drive abnormal dispersal behavior. However, demographic factors (i.e, presence/absence of others) are also thought to influence dispersal and settlement behavior (Campomizzi et al. 2008; Gitzen et al. 2016) by mediating intraspecific interactions.

Recent studies suggest that snakes can determine the sex and body size of conspecifics from information in chemical cues (Shine et al. 2003; Scott et al. 2013) eliciting social interactions such as kin recognition (Clark et al. 2012), reproductive tactics (Shine and Mason 2012; Doody et al. 2013), and cryptic social organization (Shine et al. 2005). Despite the
growing body of literature pertaining to social interactions in snakes, few studies have formally investigated how decisions regarding space use are affected by the presence/absence of conspecifics; however there is some evidence to suggest that the presence of conspecifics may have significant impacts on movements and habitat use in snakes (Gregory et al. 1987; Carpenter 1984). Further, it has been suggested that interspecific interactions (i.e., competitive exclusion) may influence patterns of co-occurrence in snakes (Steen et al. 2014) –suggesting that snakes may have the ability to detect the presence of others and thus, inform spatial decision-making. This may be of particular importance in regard to reintroduction efforts for species like the EIS, where some snakes are released in areas that lack the presence of conspecifics (i.e., first release group), while others are released in an areas heavily used by conspecifics (i.e., following release groups) and thus may result in differences in dispersal and settlement behavior following release into the wild.

Differences in release methods employed for EIS (i.e., soft vs. hard releases) have also suggested that presence of conspecifics may have an influence on post-release dispersal behavior. For example, data from the CNF reintroduction suggest that dispersal of reintroduced female EIS are not sensitive to the type of release method employed but that in order to encourage males to settle, the presence of established females in the release area may be needed (Stiles, J. 2013). Specifically, Stiles (2013) speculates that if snakes of both sexes are released in multiple groups over time, males in the first release group will disperse more quickly than males in the second group because first-group males encounter few, if any, females, whereas second-group males could experience physical encounters and chemical trails of newly established first-group females. While sex of conspecifics may play a role in encouraging settlement behavior in
male EIS, other demographic factors such as differences in age class (i.e., sexually mature vs. immature) of released individuals may also influence dispersal.

Recently it has been suggested to release juvenile (ages 1-2) EIS instead of sub-adults (ages 3-4) in an attempt to eliminate financial burdens associated with rearing snakes in captivity (C. Guyer, Pers. Communication). However, releasing individuals of this age class poses challenges to monitoring efforts. For example, if the end-goal of reintroduction efforts for EIS is the establishment of self-sustaining populations—the longer animals must survive before being able to participate in reproductive behavior, the more delayed achievement of this goal becomes. The longer it takes to achieve reproductive success, the more expensive monitoring efforts become (Folt et al. 2019). As a result, additional data are needed to evaluate the effect of demographic factors, such as age class (i.e., juvenile vs. sub-adult) on post-release dispersal and establishment.

While quantifying patterns of space use to evaluate behavior of reintroduced animals is critical to monitoring efforts, methods of home range estimation considered suitable for herpetofauna are highly debated with minimum convex polygons (MCPs) being recommended for behavioral reasons (Row and Blouin-Demers 2006) and kernel density estimates (KDEs) being recommended for statistical reasons (Calange 2011) –along with whether assumptions that influence these estimations are biologically meaningful (White and Garrot 1990). Despite their popularity in use, each have their own setbacks. For example, 100% MCPs which encapsulate all animal locations have a tendency to over-estimate home ranges, because they can incorporate extreme, atypical movements and areas not used by the animal, while KDEs, which are weighted by intensity of use of particular areas, can be biased for animals snakes, which tend to use the same sites repeatedly (Row and Blouin-Demers 2006).
As typically implemented in studies of snakes, both of these methods are typically restricted to two-dimensional (2D) estimates of space use, despite the fact that three-dimensional (3D) estimates may be more appropriate for landscapes that are topographically diverse (Greenberg and McClintock 2008)
While the incorporation of topography has been used in home range estimation for many species of mammals (i.e., flying squirrels, woodrats, and deer (Stone et al., 1997; Castleberry et al., 2001; and Campbell et al., 2004)) the third dimension has received considerably less attention among herpetofauna (Greenburg and McClintock 2008). As a result, relying on standard two-dimensional estimates of space use alone may limit our understanding of spatial ecology across a multitude of vertebrates, including large, active foraging snakes that potentially use a variety of topographically diverse habitats, like the EIS.

This study is part of a range-wide recovery effort for EIS and is the first attempt to reestablish a population of EIS in the state of Florida. Following the framework of the CNF reintroduction, we used radio-telemetry to track post-release survival, dispersal patterns, and habitat and space use of hard-released EIS to ABRP. Our objectives were to report annual survival of reintroduced EIS, determine whether male EIS had larger home ranges than females, to explore the use of three-dimensional home range estimation, and to evaluate the influence of demographic factors (i.e., sex, age class) and release year on dispersal rate. We hypothesize that:

1. Male EIS will have larger home ranges than females

2. EIS will traverse across a topographic gradient unique to our field site (i.e., ravine slopes during the active season to access wetland/swamp habitat) causing 3D home ranges to be larger than 2D home ranges.

3. EIS in the first release group will disperse more than EIS in the second release group because first group snakes will be released onto a landscape lacking conspecifics
METHODS

Study Area

This study was conducted at The Nature Conservancy’s (TNC) Apalachicola Bluffs and Ravines Preserve (ABRP) in the Florida Panhandle. This site consists of 2,548 ha of upland sandhills and steephead ravines bordering Torreya State Park (5,600 ha) and Beaver Dam Creek Wildlife Management Area (533 ha), for a total of 8,681 ha of habitat thought to be suitable for the reintroduction of EIS. Over the last 30 years, the sandhills of ABRP have been transformed. The return of prescribed fire coupled with novel groundcover restoration tactics implemented by TNC allowed for establishment of longleaf pine and native groundcover, such as wiregrass (*Astrida stricta*). Additionally, recent increases in Gopher Tortoise abundance (David Printiss, The Nature Conservancy, pers. Communication) have been observed throughout the property, with which EIS are intimately tied in North Florida. Having addressed the reasons thought to be responsible for the historic extirpation of EIS on the property, ABRP was selected as the second reintroduction site as a part of a range-wide recovery effort for the species.

Telemetry

Thirty-two captive-bred Eastern Indigo Snakes were surgically implanted with Holohil SI-2 radio-transmitters (Holohil Systems Ltd., Ontario, Canada) and PIT tags (AVID, Norco, California; Reinert and Zappalorti 1982) in 2017 (12 snakes) and 2018 (20 snakes). These surgeries were performed by Dr. James Bogan, the Central Florida Zoo vet at the Orianne Center...
for Indigo Conservation (OCIC) when snakes were approximately 944 grams in weight. Surgeries occurred 1-2 months prior to release onto ABRP, to allow for a heal time of at least 2 sheds. Transmitters weighed 13.5 grams, were 50 mm in length, and 11 mm in width, and contained batteries designed for 24 months of signal transmission. Twelve snakes (8 males, 4 females) were released between July and August of 2017 and 20 snakes (12 males, 8 females) of varying release ages (2 to 4) were released between June and August of 2018 (Appendix 1). Transmitter weight was less than 2.5% of the snake’s weight. All snakes were released during the day into tortoise burrows selected within the release area (Unit 2; Figure 1) which met the following specifications: adult-sized (>20 cm), signs of recent tortoise use, and at least 20 m distance from any other release burrow. All burrows selected for the 2017 release were reused for the 2018 release. All procedures that involved handling of live snakes were covered under the following Florida Fish and Wildlife permit: LSSC-16-00041.

A Communications Specialist, Inc. R-1000 Telemetry Receiver (Communications Specialist Inc., Orange, CA) coupled with a directional 3 element folding Yagi antenna (Advanced Telemetry Systems, Inc. Isanti, MN) were used to locate snakes via homing 1-2 times per week. The transmitter range was approximately 300 m. All snake locations were recorded to the nearest 5m using a hand-held Garmin GPS unit (Garmin Ltd., Olathe, KS). Upon location, time, date, habitat and refuge type (if below ground) were recorded along with notes on any behavior observed when snakes were visible.

**Sampling Effort**

Radio-telemetry studies often suffer from serially autocorrelated data because observations (or relocations) are not independent in time or space and therefore these data
violates assumptions for statistical inferences (De Solla et al. 1999). As a result, many reptile home range studies advocate for the attempted removal of autocorrelation (Swihart and Slade 1985; Worton 1987). In order to account for this in our data, we used the method of O’Conner et al. (1994) in a post-hoc analysis to assess the number of recaptures needed to eliminate spatial autocorrelation allowing us to select a minimum number of locations to achieve a reasonable estimate of annual home range size for EIS. While the methods of O’Conner et al. (1994) differ from most conventional methods to handle serial autocorrelation in the data, we chose it because it was best suited to account for the inconsistencies often associated with exploratory movements (i.e., short movements interspersed with infrequent, longer movements). We used a custom program (BASIC programming language) to randomize the first 20 relocations for 10 EIS (8 females, 2 males), generating 10 randomized data sets for each individual. Snakes that had fewer than 20 relocations were excluded from this analysis, as that was our cut-off for inclusion. We then calculated 100% minimum convex polygons (MCPs) for each set of randomized locations for each animal. The program then selected 10 random starting points within the observed sequence of 20 locations registered for each of the 10 EIS. From each random starting point, a sequence of 20 locations was generated by concatenating the locations from the random starting point to the end of the observed sequence and the locations from the first observed location to the random starting point. Each concatenated sequence was used to calculate 100% MCPs (10 sequential estimates of home range size per animal).

Means and 100% confidence limits were calculated for home ranges generated from the randomized and concatenated localities of each sample size (Fig. 2). The sample size of sequential localities large enough to be unaffected by serial autocorrelation was estimated to be the sample size for which home ranges estimated from concatenated samples no longer were
smaller than those estimated from randomized samples of comparable size. Confidence limits for male and female EIS start to overlap at 11 locations (Fig. 2) which indicates an appropriate number of locations for sampling effort to truncate our data, leaving us with a total of 16 EIS (10 females, 6 males) to be included in home range analyses. Because no evidence of an asymptote in home range size was indicated as relocation number increased beyond 11 locations (Fig. 2), all comparisons of home range size were made for the first 11 relocations.

Survival

In order to examine annual survival for EIS following release, we generated Kaplan-Meier survival curves for snakes released in each year (2017 and 2018). In 2017, we released a total of 12 snakes (8 males, 4 females) and censored 5 individuals because they were MIA at the end of our study and thus we were unable to determine whether they were alive or not. In 2018, we released 20 snakes (11 males, 9 females) and censored 5 individuals because they were MIA at the end of our study and again we were unable to determine whether they were alive or not.

Home Range Size

To examine home range size of reintroduced EIS, we calculated 100% minimum convex polygon (MCP) home ranges (Mohr 1947) for 16 individuals and restricted our analysis to only include the first 11 relocations which allowed us to maximize our sample size while minimizing the effects of serial autocorrelation in the data (Figure 2). We also calculated 95% kernel density estimates (KDEs) and 50% areas of core use (i.e., established areas of familiarity within a home range where a majority of time is spent (Girard et al. 2002)). EIS with less than 30 relocations were excluded from these calculations to decrease bias from individuals with fewer observations.
Girard et al. 2002), leaving a total of 8 EIS (6 females, 2 males). Program R (R Foundation for Statistical Computing 2018) was used to calculate 100% MCP home ranges, 95% KDEs and core areas using 50% UD with the package adehabitatHR (Calenge 2011). We used a reference bandwidth (h ref) for our smoothing parameter to generate utilization distributions because the h ref is more likely to include areas used but may have gone undetected due to a lower sampling regime (Bauder et al. 2015) like the 1-2 times per week tracking regime implemented in our study for EIS.

Linear regression of 100% MCP (dependent variable) on 95% KDEs (independent variable), evaluated against a null hypothesis of a slope of 1.0, was used to determine the degree to which the two methods consistently measured the target entity (Appendix 4) justifying our use of MCPs instead of KDEs in other analyses. We then used a linear regression to test for an effect of sex on home range size in EIS using 100% MCP home range estimates. We then plotted our data, which showed a significant fit to a normal distribution, justifying our use of parametric statistics.

We also calculated 3D home range estimates (Greenberg and McClintock 2008) using 1/3 arc-second Digital Elevation Model (DEM) data for the study area which were retrieved from the USGS National Map 3DEP Downloadable Data Collection (https://viewer.nationalmap.gov/basic/) on April 19th, 2019. The raster DEM layer was imported into ArcMap 10.5.1 and converted to a Triangular Irregular Networks (TIN) layer using the Raster to TIN tool. The MCPs for 28 EIS (15 males, 13 females) using all relocations were used in conjunction with the TIN layer of the study site to determine the three-dimensional home range area of each snake using the Polygon Volume tool. A custom model was used to iterate the process for each feature class containing a snake’s home range and to calculate two-dimensional
and three-dimensional area statistics, including percent increase of the three-dimensional area (Appendix 2). A paired t-test was used to determine if home range estimators were significantly different.

**Dispersal**

To quantify the rate at which reintroduced EIS dispersed following release, we first used Program R (R Foundation for Statistical Computing) to calculate 100% MCP (hereafter, MCP) home ranges for 24 EIS (12 males, 12 females) using the package adehabitatHR (Calenge 2011: Table 1). MCPs encompassed all locations of individual snakes, excluding their release location. We included all locations here because we wanted to base our calculation of dispersal on the entirety of space used by EIS during our study. We then calculated the centroid of each MCP polygon, and the distance between the MCP centroid and release location for each snake, and divided this value by the number of days (ranging from 38 to 512 days) over which each snake was tracked. We considered this value to represent the rate each snake dispersed from its point of release to create its MCP polygon over the total number of days it was tracked. EIS that had less than 6 relocations were excluded from these analyses because 6 is the minimum number of relocations required for the MCP centroid calculation (Calenge 2011). We used three-factor anovas to look at the effect of sex (male and female), release age (age 2 = young; ages 3-4 = old) and release year (2017 and 2018) on rate of dispersal, and used a corrected sample size Akaike Information Criterion (AICc) to determine the best model because our sample sizes were small among our groups. We then ran a post-hoc analysis (TukeyHSD) to test for differences among all groups.
Habitat Use

To summarize habitat use of reintroduced EIS, we extracted landcover data for Apalachicola Bluffs and Ravines Preserve, in Liberty County, FL from USGS and then used the landcover data to delineate two habitat types for our field site; uplands and ravine/wetlands. Habitat types included in the uplands category were forest regeneration areas and upland mixed-coniferous/hardwood, whereas habitat types included in our ravine/wetlands category were titi swamps, gum swamps, mixed wetland forest, and bottomland swamps. We calculated the percent of relocations for all snakes (N = 32) found in either habitat type throughout the course of our study (July 17th 2017 through January 5th 2019) by month.

RESULTS

Survival

Kaplan-Meier survival estimates for 12 EIS released in 2017 (8 males, 4 females) was 45%, whereas survival estimates for 20 EIS released in 2018 (12 males, 8 females) was 46% (Figure 6). In 2017, we had 2 females snakes known to be alive, 1 female that died in a prescribed burn, and 1 female that was removed from the study due to severe injury. We also had 4 males known to have died, 3 more males for which we found bare transmitters for but no body/remains, and 1 male still remains MIA. In 2018, we had 5 female snakes known to be alive, 3 females known to have died, and 1 female that was considered MIA. We had 1 male known to be alive, 3 males that died getting hit by cars, 2 other males known to be dead for unknown reasons, and 5 males considered MIA.
Home Range Size

Minimum convex polygon home ranges for EIS using the first 11 relocations (n = 16) ranged from 1.6 and 34.8 ha, with a mean of 15.0 ha. Female MCPs (n = 11) ranged from 1.6 to 28.1 ha, with a mean of 11.4 ha. Male MCPs (n = 5) ranged from 9.2 to 34.8 ha, with a mean of 23.2 ha (Fig. 3). Our data indicate that MCP home range sizes of reintroduced EIS varied by sex (F=4.129; df=14; p=0.01) with males on average using larger home ranges than females. Further, regression of 100% MCP on 95% KDE area was strongly linear (R² = 0.99) with a slope that did not differ from 1.0. Similarly, regression of 50% areas of core use on 95% KDE area was strongly linear (R² = 0.85) with a slope of 0.15. Thus, the core use area of the home range was 15% of the 100% MCP. Fifty percent areas of core use and 95% KDE areas were calculated for 8 EIS with 30 relocations or more (2 males, 6 females). 50% areas of core use ranged from 3.0 to 46.7 ha, with a mean of 16.0. 50% areas of core use for females (n = 6) ranged from 3.0 to 46.7 with a mean of 15.4. 50% areas of core use for males (n = 2) ranged from 16.0 to 22.7 with a mean of 19.4. 95% KDE areas ranged from 15.2 to 345.8 with a mean of 96.1. 95% KDEs areas for males (n = 2) ranged from 83.5 to 98.8 with a mean of 92.

Three-dimensional 100% MCP home ranges calculated for EIS 28 EIS (15 males, 13 females) using all relocations for snakes tracked 1-2 times per week between July 17th 2017 and January 5th 2019 ranged from 1.62 ha to 271.13 ha, with a mean of 61.78 ha whereas two-dimensional estimates ranged from 1.6 ha to 266.79 ha with a mean of 60.68 ha (Appendix1). The paired t-test returned a significant value (df = 54; p = 0.9551) indicating the two home range estimators are not significantly different from each other.
Dispersal

Dispersal rate was best predicted by a three-way interaction between sex, release age and release year (Table 2). Our post-hoc analysis (TukeyHSD) of the best-fitting ANOVA model (p=0.004) revealed that younger males released in year 2 dispersed away from the release site more rapidly than all other groups (Figure 4).

Habitat

Percent use of each habitat type by EIS showed that reintroduced EIS utilized both the upland sandhills and ravine/wetland habitat on ABRP (Figure 5). However, from December through May EIS exhibited a shift to exclusive use of sandhills habitat.

DISCUSSION

Two-dimensional 100% MCP home range sizes of EIS released onto ABRP varied by sex (p = 0.01), with males, on average, maintaining larger home ranges than females (mean: 20.99 vs. 11.38 ha) than females (mean of 11.38 ha) aligning with patterns of space use reported for EIS from wild populations and for those reintroduced into CNF (Hyslop et al. 2014; Stiles, J. 2013; Breininger et al. 2011). However, our home range estimates only included the first 11 relocations due to constraints associated with serial autocorrelation in our data. Therefore, our estimates may not be entirely representative of what constitutes a more conventionally determined home range calculation. This may be in part why estimates for EIS in our study were
considerably smaller than those reported by Stiles (2013) for both soft and hard released EIS into CNF (mean: 155.7 vs. 61 ha for males and females, respectively) and for those reported by Breininger et al. (2011; e.g. 156 vs. 44 ha for males and females, respectively). However, the Breininger et al. study had a much larger sample size of snakes with a much higher number of fixes for EIS, which may have influenced their results.

Radio-tracked EIS in our study show a seasonal shift in habitat use, explicitly using upland sandhills habitat during the winter months (December—May), and using a combination of upland sandhills and ravine/wetland habitat during the summer and autumn months (November—June). This is similar to the findings of Hyslop (2014) which indicated that while the presence of thermally stable refugia such as tortoise burrows might drive their exclusive use of sandhills habitat during the winter and breeding months, that sandhills habitat is used consistently across every month of tracking in addition to wetland/ravine habitat in the summer months. However, it is important to note that our release strategies for EIS were structured around releasing snakes in the middle of summer into tortoise burrows, when they typically only use these burrows in the winter months. This may have influenced our results, because snakes were forced to start off in the uplands habitat before they could find their way to wetland habitats. While we did not do any formal habitat analyses, the data show that the habitat of ABRP is suitable to support both the wetland habitat needs as well as the overwintering needs of reintroduced EIS which is vital for snakes.

A paired t-test did not reveal a significant difference in 2D and 3D home range estimates for EIS in our study (df = 54; p = 0.9551) with a mean of 60.68 ha for 2D estimates and a mean of 61.78 ha for 3D estimates. One potential explanation for this may be that the use terrain modeling is more appropriate for the topographic diversity of mountain slopes, much like the
Greenburg and McClintock study (2008) whereas ABRP consists of more lower-elevations (i.e., floodplain and ravine bottoms) which may be more difficult to represent. However, there may be fine scale differences in habitat use that could be highlighted in 3D estimations of home range size—which could be particularly valuable in helping to characterize topographic complexity in landscapes that snakes occupy (Heard et al. 2004; Moore and Gillingham 2006).

Dispersal data for EIS in our study indicated that young males released in year two dispersed away from the release sandhills more quickly than all other snakes (p = 0.004). One possible explanation for this may be that younger, subordinate males released in year 2 of our study were avoiding the release sandhills that were occupied by larger, more dominant snakes (Powell 1979; Bauder et al. 2016). Multiple studies have demonstrated that snakes can ascertain information about conspecifics (i.e., sex and body size) from information in chemical cues (Powell 1979; Shine et al. 2003; Scott et al. 2013) potentially influencing how individuals distribute themselves in relation to the presence/absence of others. This may be of particular importance when thinking about release strategies for EIS in which changes in abundances of conspecifics (i.e., sex and age class) occur with each release. For example, individuals in the first release group for a reintroduction effort will experience an “empty” environment (no conspecifics present), whereas, individuals in subsequent release groups may be exposed to varying abundances of conspecifics (i.e., age class and sex) (Mihoub et al. 2011; Richardson and Ewen 2016) that may influence settlement behavior. However, it is important to note that our dispersal analysis was not set up to formally evaluate intraspecific interactions and that our sample size was small due to constraints on the assemblage of snakes able to be reintroduced each year (i.e., sex, age class and total number of individual snakes available for release) –and therefore our results must be interpreted with some caution.
Another potential explanation may be due to differences in body sizes. For example, many studies indicate a relationship between body size and dispersal behavior in snakes (Hyslop et al. 2009; Hyslop et al. 2014). Unfortunately we do not have specific size data for EIS in our study, but if older animals are thought to be larger in size, the EIS we saw with the greatest dispersal were the younger (potentially smaller) individuals. Further, the associated risks of exploratory behavior may also be playing a role in dispersal of EIS reintroduced at our field site. It is common for animals to spend some time exploring their surroundings following release into the wild before they start to meaningfully move throughout it. As a result, snakes that had longer periods of exploratory behavior would have inherently dispersed at a greater rate away from the release sandhills, and it is possible that younger males may exhibit longer exploratory periods following release when conspecifics are present on the landscape.

Monitoring for survival can be challenging for many wildlife populations. This is especially true for elusive species like snakes that often occur in low densities, and whose secretive nature enables them to evade detection (Hyslop et al. 2009; Parker and Plummer 1987). While the use of radio-telemetry has drastically improved our ability to estimate survival by allowing researchers to consistently monitor individuals, the behavioral challenges that are associated with monitoring large, active-foraging snakes like the EIS may still yield incorrect assessments of survival as the large movements that EIS are capable of making may subject researchers to lose track of individuals easily and thus being unable to confirm fate of snakes. Further complicating these assessments are the associated complications with transmitter surgeries (White and Garrot 1990). For example, some snakes in our study were believed to have expelled their transmitters, while others had antennae become broken on the inside of snakes – both of which can influence movement behavior. Further, there may be residual infection as a
result of transmitter surgeries (C. Guyer, Pers. Communication) which may further compromise animal movements.

Kaplan-Meier survival estimates of reintroduced EIS in our study were 45% for 12 EIS (8 males, 4 females) released in 2017, and 46% for 20 EIS (11 males, 9 females) released in 2018. However, these estimates censored all snakes with unknown fates (i.e., MIA snakes) and in both years we had a total of 5 individuals that fell under this category. We often assume that snakes we can no longer detect are likely fine but have just dispersed far from where they were released. However, these individuals can no longer contribute to reproductive behavior needed in order to achieve the long-term goal of establishing self-sustaining populations. In comparison to other studies of snake survival, estimates for EIS at ABRP are considerably lower. For example, Parker and Plummer (1987) report annual survival of 70% for late-maturing colubrids and 77% for late-maturing viperids. One potential explanation for why survival estimates in our study are so low might be due to increased exploratory behavior in reintroduced snakes. For example, two males in our study that were released in 2018 were hit by cars within a month of release (probably as a result of abnormal dispersal behavior following release). The more exploratory movements there are, the more likely snakes are to encounter risky situations (i.e., encounters with predators, crossing roads) and because these snakes were released into novel environments, we might speculate that initial exploratory dispersal from the release site can be linked with the low survival for EIS in our study. It is also important to consider that while we treat MIA snakes as alive in these survival estimates, they are functionally dead in regards to the main goal of the reintroduction: to establish self-sustaining populations at release sites. If snakes are dispersing so far from the release site that we are unable to detect them, they are not contributing to achievement of this goal.
In contrast, survival data reported for EIS by Hyslop (2009) indicated a disproportionately higher mortality in female snakes while our study documented a disproportionately higher mortality of males. Physiological stressors brought on by breeding behavior (i.e., gestation and oviposition) have been identified as contributing factors to higher female mortality in some snakes (Bonnet et al. 1999; Parker and Plummer 1987). Despite the higher female mortality reported by Hyslop (2009) they were unable to report direct observations of mortality related to reproductive behaviors. However it is important to note that even with the help of radio-telemetry EIS is an incredibly elusive species that spends most of its time in tortoise burrows during the breeding season, which could make it difficult for researchers to appropriately collect meaningful data on deaths occurring during these times. This is important to consider as we think about what success looks like for EIS reintroductions. For example, because male survival has been almost non-existent (only 1 male known to be alive) the opportunities for females to participate in reproductive behavior are incredibly low. Therefore, the risks associated with gestation and oviposition that have been identified as driving factors of lower survival in breeding female snakes are not coming in to play in our study yet. Once males survive long enough to breed with females, we might speculate that female survival would suffer decreases as a result. Therefore, we should identify a number of female snakes that need to be released every year in order to best account for this in the beginning stages of EIS reintroductions to ensure continued reproductive success. Further, Hyslop (2009) data also indicated lower survival associated with larger snakes. While we did not collect data on EIS body size per se, if we assume that body size and age are correlative (i.e., the older the animal the larger it should be) our survival data suggest the opposite to be true. Of all known surviving EIS at ABRP, all survivors except one were reproductively mature individuals.
Table 1. Data summarizing radio-telemetry tracking efforts for two groups of reintroduced Eastern Indigo Snakes (*Drymarchon couperi*) to Apalachicola Bluffs and Ravines Preserve, Liberty County, FL in 2017 (8 males, 4 females) and 2018 (11 males, 9 females). All snakes were tracked 1-2 times a week between July 17*th* 2017 and January 5*th* 2019. Snakes considered to be “young” were released at age 2, and snakes considered to be “old” were released between the ages of 3 and 4. Fates for all MIA snakes and bare transmitter snakes are considered to be unknown.

<table>
<thead>
<tr>
<th>Sex</th>
<th>Release Age</th>
<th>Release Year</th>
<th>Days Tracked</th>
<th>Number of Relocations</th>
<th>Fate</th>
</tr>
</thead>
<tbody>
<tr>
<td>F</td>
<td>Young</td>
<td>2017</td>
<td>347</td>
<td>40</td>
<td>Alive</td>
</tr>
<tr>
<td>F</td>
<td>Young</td>
<td>2017</td>
<td>209</td>
<td>25</td>
<td>Removed</td>
</tr>
<tr>
<td>F</td>
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<td>2017</td>
<td>291</td>
<td>31</td>
<td>Dead</td>
</tr>
<tr>
<td>F</td>
<td>Old</td>
<td>2017</td>
<td>512</td>
<td>70</td>
<td>Alive</td>
</tr>
<tr>
<td>M</td>
<td>Old</td>
<td>2017</td>
<td>110</td>
<td>15</td>
<td>Bare Transmitter*</td>
</tr>
<tr>
<td>M</td>
<td>Young</td>
<td>2017</td>
<td>53</td>
<td>10</td>
<td>Bare Transmitter*</td>
</tr>
<tr>
<td>M</td>
<td>Old</td>
<td>2017</td>
<td>85</td>
<td>17</td>
<td>Dead</td>
</tr>
<tr>
<td>M</td>
<td>Old</td>
<td>2017</td>
<td>38</td>
<td>17</td>
<td>MIA*</td>
</tr>
<tr>
<td>M</td>
<td>Old</td>
<td>2017</td>
<td>33</td>
<td>6</td>
<td>Dead</td>
</tr>
<tr>
<td>M</td>
<td>Old</td>
<td>2017</td>
<td>209</td>
<td>12</td>
<td>Bare Transmitter*</td>
</tr>
<tr>
<td>M</td>
<td>Young</td>
<td>2017</td>
<td>117</td>
<td>16</td>
<td>Dead</td>
</tr>
<tr>
<td>F</td>
<td>Old</td>
<td>2018</td>
<td>100</td>
<td>19</td>
<td>Dead</td>
</tr>
<tr>
<td>F</td>
<td>Young</td>
<td>2018</td>
<td>24</td>
<td>5</td>
<td>Dead</td>
</tr>
<tr>
<td>F</td>
<td>Old</td>
<td>2018</td>
<td>166</td>
<td>30</td>
<td>Dead</td>
</tr>
<tr>
<td>F</td>
<td>Old</td>
<td>2018</td>
<td>168</td>
<td>30</td>
<td>Alive</td>
</tr>
<tr>
<td>F</td>
<td>Old</td>
<td>2018</td>
<td>168</td>
<td>33</td>
<td>Alive</td>
</tr>
<tr>
<td>F</td>
<td>Old</td>
<td>2018</td>
<td>165</td>
<td>11</td>
<td>Alive</td>
</tr>
<tr>
<td>F</td>
<td>Old</td>
<td>2018</td>
<td>165</td>
<td>30</td>
<td>MIA*</td>
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<tr>
<td>F</td>
<td>Old</td>
<td>2018</td>
<td>163</td>
<td>16</td>
<td>Alive</td>
</tr>
<tr>
<td>F</td>
<td>Young</td>
<td>2018</td>
<td>164</td>
<td>13</td>
<td>Alive</td>
</tr>
<tr>
<td>M</td>
<td>Young</td>
<td>2018</td>
<td>88</td>
<td>10</td>
<td>Dead on Road</td>
</tr>
<tr>
<td>M</td>
<td>Young</td>
<td>2018</td>
<td>30</td>
<td>6</td>
<td>Dead on Road</td>
</tr>
<tr>
<td>M</td>
<td>Old</td>
<td>2018</td>
<td>168</td>
<td>11</td>
<td>Dead on Road</td>
</tr>
<tr>
<td>M</td>
<td>Young</td>
<td>2018</td>
<td>50</td>
<td>10</td>
<td>Dead</td>
</tr>
<tr>
<td>M</td>
<td>Old</td>
<td>2018</td>
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<td>17</td>
<td>Dead</td>
</tr>
<tr>
<td>M</td>
<td>Old</td>
<td>2018</td>
<td>165</td>
<td>30</td>
<td>Alive</td>
</tr>
<tr>
<td>M</td>
<td>Young</td>
<td>2018</td>
<td>43</td>
<td>4</td>
<td>MIA*</td>
</tr>
<tr>
<td>M</td>
<td>Young</td>
<td>2018</td>
<td>36</td>
<td>5</td>
<td>MIA*</td>
</tr>
<tr>
<td>M</td>
<td>Young</td>
<td>2018</td>
<td>30</td>
<td>2</td>
<td>MIA*</td>
</tr>
</tbody>
</table>
Table 2. Comparison of linear models for rate of dispersal in Eastern indigo snakes (*Drymarchon couperi*) reintroduced to Apalachicola Bluffs and Ravines Preserve, Liberty County, FL in 2017 and 2018 using an Akaike Information Criterion (AICc). 12 snakes were released in 2017 (8 males and 4 females) and 20 snakes were released in 2018 (11 males and 9 females). Snakes ranged from 2-4 years of age at release. Dispersal was estimated using the average distance between the release site and the geometric mean of a 100% minimum convex polygon of each snake, and dividing this value by the number of days each snake was tracked. Snakes that had less than the minimum required number of relocations (6) were excluded from this analysis, leaving a total of 24 snakes (12 males and 12 females).

<table>
<thead>
<tr>
<th>Model</th>
<th>K</th>
<th>AICc</th>
<th>Delta_AICc</th>
<th>AICcWt</th>
<th>Cum.Wt</th>
<th>LL</th>
</tr>
</thead>
<tbody>
<tr>
<td>release age * sex * release year</td>
<td>9</td>
<td>151.49</td>
<td>0</td>
<td>0.79</td>
<td>1</td>
<td>-60.32</td>
</tr>
<tr>
<td>release age * release year + sex</td>
<td>6</td>
<td>154.13</td>
<td>2.64</td>
<td>0.21</td>
<td>0.21</td>
<td>-68.59</td>
</tr>
<tr>
<td>release age + release year + sex</td>
<td>5</td>
<td>166.52</td>
<td>15.03</td>
<td>0</td>
<td>0</td>
<td>-76.59</td>
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<tr>
<td>release age * sex + release year</td>
<td>6</td>
<td>168.09</td>
<td>16.6</td>
<td>0</td>
<td>0</td>
<td>-75.57</td>
</tr>
<tr>
<td>release age + release year * sex</td>
<td>6</td>
<td>169.38</td>
<td>17.89</td>
<td>0</td>
<td>0</td>
<td>-76.22</td>
</tr>
</tbody>
</table>

Table 3. Comparison of 100% MCP & 94% KDE home range sizes, and 50% areas of core use reported in hectares for 8 EIS (2 males, 6 females) tracked between July 17th 2017 and January 5th 2019 that had a minimum of 30 relocations.

<table>
<thead>
<tr>
<th>Sex</th>
<th>100% MCP</th>
<th>95% KDE</th>
<th>50% KDE</th>
</tr>
</thead>
<tbody>
<tr>
<td>F</td>
<td>356.06</td>
<td>345.85</td>
<td>46.74</td>
</tr>
<tr>
<td>F</td>
<td>119.88</td>
<td>97.72</td>
<td>23.79</td>
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<td>F</td>
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Figure 1. Map of ABRP and bordering protected areas of land (Torreya State Park & Beaver Dam Creek Wildlife Management Area) creating a total of 8,681 ha of suitable habitat for EIS. Each Unit indicates an area proposed as a release site within ABRP. All EIS released between 2017 and 2018 were released in Unit 1.
Figure 2. Effect of sample effort on home range area of 8 female (A) and 2 male (B) reintroduced EIS from Apalachicola Bluffs and Ravines Preserve, Florida. Sample effort is based on random (filled triangles, independent) and consecutive (filled circles; serially autocorrelated) tracking locations (see text). Vertical lines represent 95% CI.
Figure 3. Box and whisker plot comparing mean 100% MCP home range sizes of 16 EIS (6 males, 10 females).
Figure 4. Three-way interaction of release year (1= 1st year, 2= 2nd year), release age (young= age 2, old= ages 3+) and sex (m=male, f=female) on predicted values of rate of dispersal (meters moved per day) in reintroduced EIS. A indicates the significantly different group (young males released in year 2) and Bs indicate all other groups that did not differ significantly in their dispersal rates.
Figure 5. Percent use of habitat by reintroduced Eastern Indigo Snakes (N=32) on Apalachicola Bluffs and Ravines Preserve in Liberty County, FL. Landcover data was delineated for all relocations collected for snakes tracked from July 17th 2017 through January 5th 2019.
Figure 6. Kaplan-Meier Survival curves for reintroduced Eastern Indigo Snakes onto Apalachicola Bluffs and Ravines Preserve, FL in 2017 and 2018. In 2017, 12 EIS were released (8 males, 4 females) and survival was 45%. In 2018, 20 EIS were released (11 males, 9 females) and survival was 46%. All snakes that were considered to be MIA at the completion of our study along with snakes for which bare transmitters were found were censored in our curves to be treated as alive.
References


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Appendix 1. Summary of 2D 100% MCP home ranges for 16 EIS (6 males, 10 females) using only the first 11 relocations to account for serial autocorrelation, and 2D-3D percent increase in 100% MCP home range area using all relocations for 28 EIS (15 males, 13 females) radio-tracked 1-2 times per week from July 17th 2017 to January 5th 2019 at Apalachicola Bluffs and Ravines Preserve, Florida.

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Appendix 2. Comparison of 2D-3D home range sizes in male and female EIS

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Appendix 3. Comparison of home range size using 100% minimum convex polygon (MCPs) and 95% and 50% kernel density estimates (KDEs) with an h ref bandwidth for 8 EIS (2 males, 8 females) 30 relocations or more tracked from July 17th 2017 through January 5th 2019.