

HA File No. 2016.26 - Part I

Review, Recommendations and Support for Listing the Louisiana Pinesnake (*Pituophis ruthveni*) as a Federally Threatened Species



The Louisiana Pinesnake (*Pituophis ruthveni*).

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to

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INTRODUCTION

The U.S. Fish and Wildlife Service, proposes to list the Louisiana Pinesnake (*Pituophis ruthveni*), as a federally threatened species under the Endangered Species Act. I was asked to review the proposed rule document in the Federal Register (Department of the Interior, Fish and Wildlife Service 2016). As outlined in the proposed rule, there are several combined threats that have caused the decline of Louisiana Pinesnakes throughout their historic range and their greatly reduced current range in the southern United States (Rudolph *et al.* 2006; USFWS 2016).

The Louisiana Pinesnake (**Figure 1**), is a large, non-venomous snake that occurs in western portions of Louisiana and eastern sections of Texas (Conant and Collins 1991). *Pituophis ruthveni* is currently the accepted scientific name for this distinct species (Reichling 1995; Crother 2000; Rodriguez-Robles and Jesus-Escobar 2000; Collins and Taggart 2002; Rudolph *et al.* 2006).

Reptiles, as a group have been largely ignored in conservation efforts (Gibbons *et al.* 2000), yet they are likely to be affected by the same threats as other more popular wildlife species. This is true for pinesnakes in general and the Louisiana Pinesnake in particular, especially because of past and current illegal collecting, predation, habitat disruption or alteration, habitat fragmentation, and outright loss of habitat by commercial and residential development (Rudolph 2000; Himes *et al.* 2002; Duran 2010; Burger and Zappalorti 2011; Burger and Zappalorti 2016).

PERSPECTIVE AND SCOPE

Historically, Louisiana Pinesnakes occupied vast sections of the longleaf pine (*Pinus palustris*) forest ecosystem within their known range, but over the past 200-300-year period, tree farming by the logging industry drastically changed the forest landscape. Conservationists recognized this problem and there is now a major effort by most southern states to restore the longleaf pine forest ecosystem. In cooperation with state and federal forest agencies, the lumber industry is generally managing the longleaf pine forest habitat to help support rare plants and wildlife in conjunction with tree harvesting. It has been shown that longleaf pine forests in southeastern United States harbors the highest reptile and amphibian diversity in North America (Guyer and Bailey 1993).

Along with Louisiana Pinesnakes, there are other rare reptile and mammal species who are dependant upon the longleaf pine - wiregrass ecosystem, such as the Gopher Tortoise (*Gopherus polyphemus*), Black Pinesnake (*Pituophis lodingi*), Eastern Indigo Snake (*Drymarchon couperi*), and Baird's Pocket Gopher (*Geomys breviceps*). These rare species are also declining due to the loss of their longleaf pine, sand-hill habitat (Auffenberg and Franz 1982; Moler 1992; Frost 1993). Aside from past logging practices, other types of human disturbance, fragmentation, and habitat degradation are among the contributing factors towards the reduction of Louisiana Pinesnakes (Wilcove *et al.* 1998, Fernandez-Juricic 2000; Goebel *et al.* 2001).

BRIEF NATURAL HISTORY OF LOUISIANA PINESNAKES

A relatively small, low-density population of Louisiana Pinesnakes continues to persist in western Louisiana and eastern Texas, with some larger core populations in protected and managed longleaf pine forest habitats. Louisiana Pinesnakes select areas in the forest with an abundant ground-layer of herbaceous vegetation. Likewise, the Baird's pocket gopher (*Geomys breviceps*), also prefers the same habitat type. The pocket gopher is one of the Louisiana Pinesnake's primary prey items, which constitutes 75% of their prey biomass (Rudolph *et al.* 2012). Baird's pocket gopher depends mostly on various plant parts of a variety of herbaceous species (Pennoyer 1932; Sulentic *et al.* 1991), especially the roots and fruits of the plants. Pocket gopher abundance is associated with a low density of trees, an open canopy, and a small amount of woody vegetation cover, which allow greater sunlight and more herbaceous forage for these secretive mammals (Himes 1998; Melder and Cooper 2015).



Figure 1. Louisiana Pinesnake showing its long, black forked tongue.

Baird's pocket gophers are important "hole providers" because they excavate the network of underground burrows and connected tunnels where Louisiana Pinesnakes are most often found (Rudolph and Conner 1996; Rudolph and Burgdorf 1997; Himes 1998; Rudolph *et al.* 1998; Rudolph *et al.* 2002; Himes *et al.* 2006). Using their keen sense of smell, Louisiana Pinesnakes dig down into the burrow systems of pocket gophers and/or eastern moles (*Scalopus aquaticus*), and use them as nighttime shelters, to flee from forest fire, or to escape from high summer surface temperatures. Likewise, during cold spells in winter months they use the pocket gopher tunnels as a safe retreat and hibernacula (Rudolph and Burgdorf 1997; Rudolph *et al.* 1998; Ealy *et al.* 2004; Rudolph *et al.* 2007; Pierce *et al.* 2014).

The Louisiana Pinesnake is a large, secretive ophidian species that uses a variety of upland habitats including the longleaf pine ecosystem (Stull 1929; Franz 1992; Rudolph and Conner 1996; Rudolph and Burgdorf 1997; Himes 1998; Rudolph *et al.* 1998; Rudolph *et al.* 2002; Himes *et al.* 2006).

However, the once-dominant longleaf pine forest and associated sandhill-wiregrass habitat has been significantly reduced and altered (Frost 1993; Noss *et al* 1995). It was estimated that >90% of the original longleaf pine ecosystem has been converted to agriculture, pine plantations and urban human occupied areas. The few remaining extant upland tracts of longleaf pine forest are highly fragmented (Noss *et al* 1995). It is clear that the continued loss of upland longleaf pine habitats may reduce Louisiana Pinesnakes to dangerously low numbers which may lead to their extinction.

Although the Louisiana Pinesnake is a fairly well studied snake species (Rudolph and Conner 1996; Rudolph and Burgdorf 1997; Himes 1998; Rudolph *et al.* 1998), there is much more to be learned about their ecology and nest site selection. For instance, what are the best management practices to maintain current populations, and to increase the snakes to historic population levels? Our lack of knowledge regarding mating and nesting behavior or other poorly understood aspects of their natural history hampers proper conservation decisions. Knowing these important aspects of the Louisiana Pinesnake's life history is crucial to the planning and development of a successful recovery plan. This uncertainty is compounded because of the limited distribution of remaining wild populations.

RULE REVIEW AND RECOMMENDATIONS

There were some important questions that lacked specific life history details in the proposed rule document that will require additional research. These questions are:

- 1) Are there any additional reasons or predators that reduce free roaming adult, juvenile, hatchling and/or eggs of Louisiana Pinesnakes in their natural habitat?
- 2) Aside from Baird's Pocket Gophers, are there other important prey organisms selected and eaten by Louisiana Pinesnakes (including reptiles, mammals, and ground nesting birds, eggs and chicks)?
- 3) How, when and where do gravid female Louisiana Pinesnakes deposit their eggs (e.g., nest site selection, or do they only use Baird's Pocket Gopher tunnels)?
- 4) What is the hatching success and survivorship of Louisiana Pinesnakes in their natural habitat?

To that end, I provide life history information which was gathered on Northern Pinesnakes over a 46-year period that can be extrapolated to the Louisiana Pinesnake. While there are some differences between the two species such as geographic location of populations, climate and weather conditions, and the vegetation species in their forest habitat, their similarities far outweigh the differences. Both species have a similar body structure, shape and keeled scales, including the well developed snout and head shape. The rostral scale is pointed for their burrowing propensities related to foraging and/or nesting. They both select sandy or loamy soils in their preferred habitat where they excavate down into pocket gopher or mole tunnels. Both species are powerful constrictors when subduing their prey. In my opinion, what was learned about Northern Pinesnakes could be used to help with the conservation and management of the Louisiana Pinesnake.

History of a 7.5-year Northern Pinesnake Study in New Jersey

The Stafford Park Redevelopment property (hereafter SPR property) is 370-acres in size and is located in Stafford Township, Ocean County, New Jersey. This was a 7.5-year investigation that started in June of 2006 and terminated in December 2013. The framework for this project was guided by the June 28, 2006 Memorandum of Agreement which was made between Walters Homes, Inc., Ocean County, Stafford Township, and the New Jersey Pinelands Commission. As part of its responsibilities, Walters closed and excavated the old unlicensed landfill on site and used the excavated materials to properly close and cap the new licensed landfill located on the SPR property. This action was taken because the unlicensed landfill was contaminating ground water and the nearby Mill Creek.

On December 4, 2006, Herpetological Associates (HA) and the NJDEP, Endangered and Nongame Species Program submitted the final Management Plan to the Pinelands Commission entitled: “A Northern Pinesnake Management and Conservation Plan, and Radio-tracking and Monitoring Plan for Stafford Business Park and Stafford Forge WMA.” This was done on behalf of the Walters Homes, Inc. The Plan was approved by the Pinelands Commission in late December of 2006, and Walters was allowed to proceed with redevelopment and new landfill construction, provided the research and conservation plans were followed. Three Northern Pinesnake management fields with 6 artificial snake hibernacula (two on each field), were built in Stafford Forge WMA for the shifted portion of the Northern Pinesnake population (see **Table 1**). Walters began residential and commercial development in 2007 and various stages of building and reconstruction were ongoing through 2016. The monitoring program evaluated whether the pinesnakes shifted from the old landfill at the Stafford Business Park would use the artificial dens.

Table 1. Random Distribution of the 2006 Shifted Pine Snakes that were Released into Six Artificial Dens (Treatments A and B), and Non-random Assignment into Treatment C.

Winter Treatments	Den Number	Adult Males	Adult Females	Juveniles	Hatchlings	Totals
B = Two Winters	1	1	1	1	11	14
A = One Winter	2	2	2	0	13	17
A = One Winter	3	2	1	1	11	15
B = Two Winters	4	2	1	0	12	15
A = One Winter	5	1	1	2	11	15
B = Two Winters	6	2	1	0	13	16
C = One Winter in HA’s Laboratory		3	5	0	0	8
Three Treatments		13	12	4	71	100

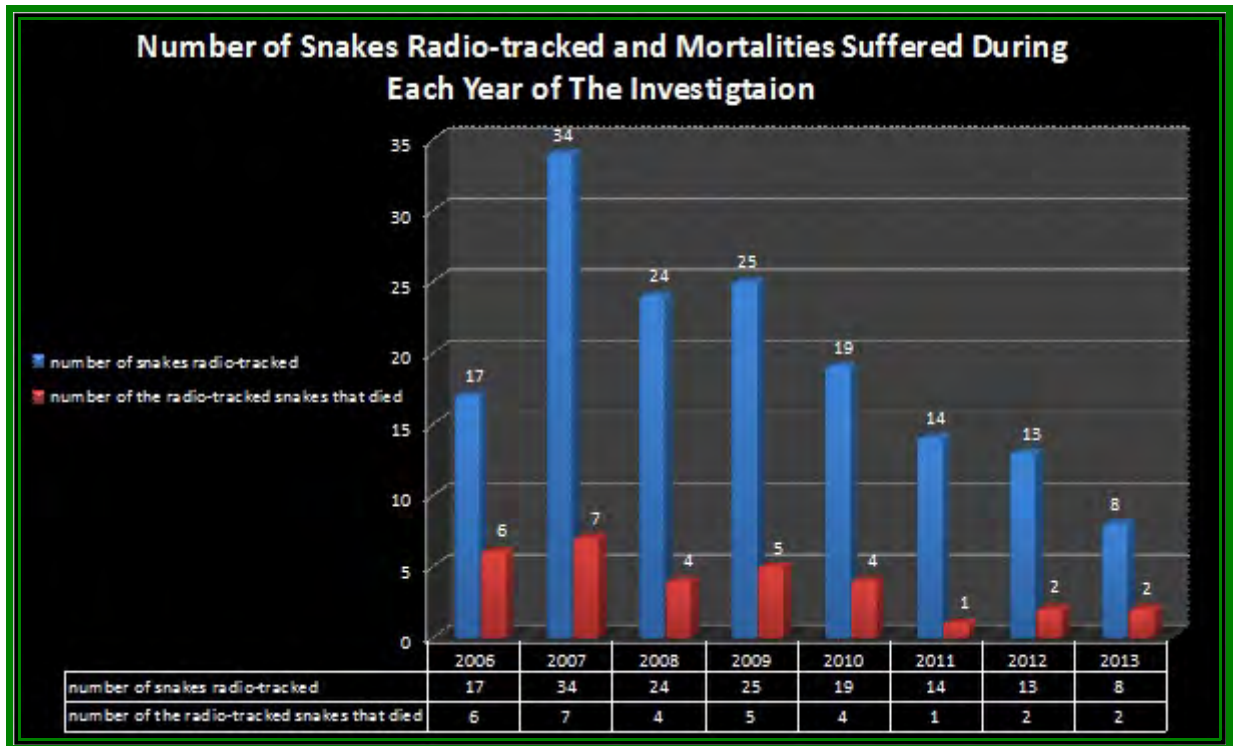


Figure 2. Graph showing the mortality of radio-tracked Northern Pinesnakes over a 7.5-year period.

Pinesnakes were randomly selected for distribution into Treatments A and B (A = one-winter treatment and/or B = two-winter treatment). The third, Treatment C (the laboratory treatment), was not originally planned, but was created out of necessity after it was determined that 8 of the pinesnakes were not healthy enough to be released in the fall of 2006 (Table 1). Unlike Treatments A and B, these snakes were not randomly assigned to a treatment because they suffered from poor health due to surgery by another consulting company. They were held in HA’s laboratory for the first winter (2006-2007). All 8 snakes from Treatment C were cared for and fed during the winter and were deemed healthy enough to be released into the two-winter treatments in the spring of 2007. They too were monitored via radio-telemetry during this investigation. Another phase of the conservation plan involved enhancing existing habitat within Stafford Forge WMA for pinesnakes. This included the construction of 6-foot high earthen berms along the edges of the three fields and large earth and wood debris piles in the center of the fields. The berms were constructed out of A-horizon sand, stumps, logs and brush. The fields provided pinesnakes with forest-edge habitat, suitable for basking and resting (Burger and Zappalorti 1988a; Zappalorti and Burger 1985). The fields are also open, with sandy areas that provide potential nesting habitat for female pinesnakes (Burger and Zappalorti 1986 and 1991). As part of the habitat enhancement, the Division of Fish and Wildlife’s Bureau of Lands Management planted grasses on the fields to replicate the lost landfill field habitat. Open grassy fields have been shown to be good nesting and foraging habitat for Northern Pinesnakes (Burger and Zappalorti 1986 and 1991).

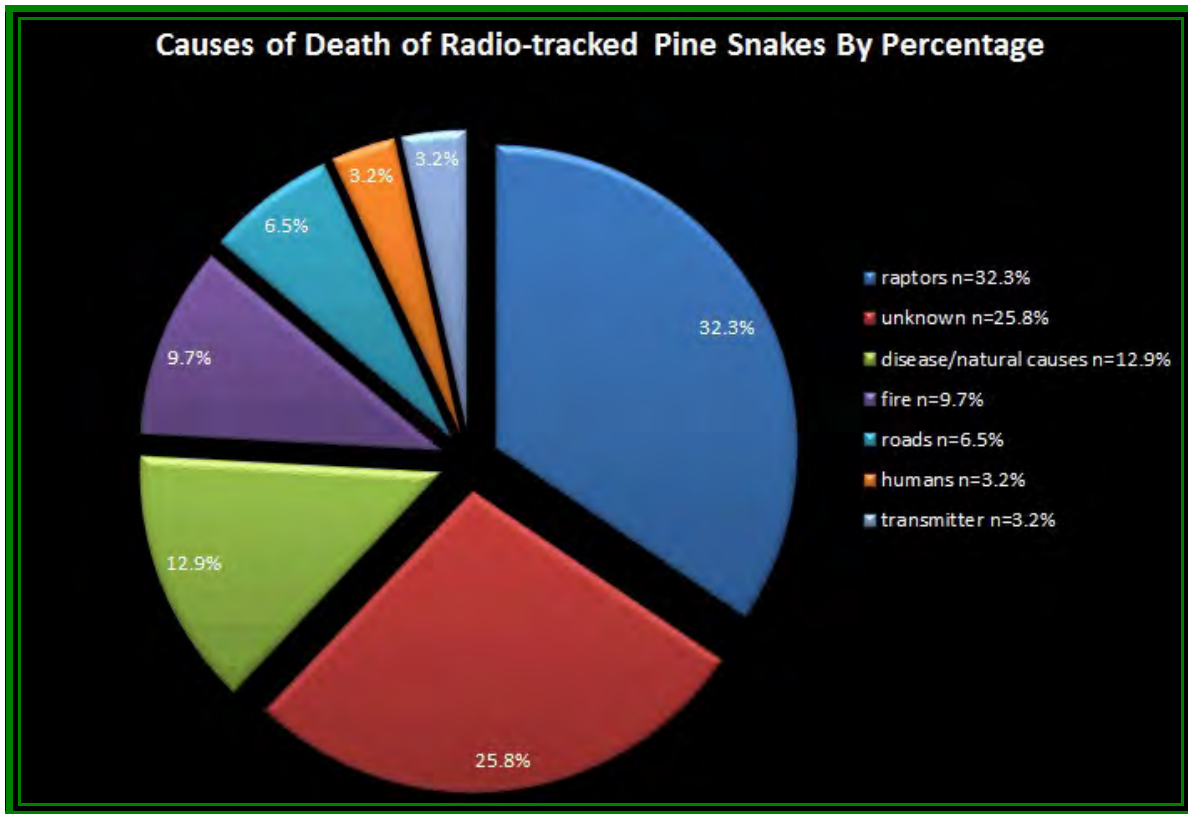


Figure 3. Chart depicting the percentage of radio-tracked snakes lost due to different causes of death.

After emerging from the artificial dens, snakes in the two-winter treatments were all released into the 3-acre corrals. On May 16, 2007, a severe crown forest fire devastated the entire pine snake study area and the three management fields, burning all the fabric off the fences and all the vegetative cover in the forest. The fire killed four of our radio-tracked adult study snakes and an unknown number of the PIT tagged hatchlings.

Due to damage related to the forest fire and the lack of vegetative ground cover to protect the snakes from hawk predation, these three corral fences could no longer be used. Instead, the two-winter treatment snakes were held within the 1-acre corrals for the duration of the treatment period (artificial dens 1, 4 and 6). After one winter of hibernation, snakes emerging from dens 2, 3, and 5 (the one-winter treatments) were allowed to disperse into the surrounding Stafford Forge WMA forest habitat. All adults were radio-tracked for the duration of the 7.5-year study or until they died from predation or other causes (Figures 2 and 3).

POSSIBLE REASONS FOR LOUISIANA PINESNAKE DECLINES

1) - Predation and Types of Natural Predators

Excessive predation may also be a contributing factor causing the decline of Louisiana Pinesnakes. Causes for mortality are only partially known in the western Louisiana and eastern Texas landscape. However, there are numerous documented records of natural predation of Pinesnakes from New Jersey that can be extrapolated to the Louisiana Pinesnake population, and may provide insight as to reasons for declines. During a 7.5-year radio-tracking investigation in Ocean County, New Jersey, HA documented Pinesnake mortalities. **Figure 2** above, shows the mortality rate among the radio-tracked Pinesnakes along with the mortality rate over the entire study (n=31). As **Figure 2** indicates, the mortality rate of the radio-tracked Pinesnakes was 66% for the entire 7.5-year study. In other words, nearly two-thirds of all the radio-tracked Pinesnakes died from predation, forest fire or other reasons during the investigation.

The mortality rate among the radio-tracked Pinesnakes during each year of the study varied from 13% to 35% (**Figure 3**). It should be noted that due to transmitter failure and the variation in the rate of new captures, a different number of Pinesnakes were radio-tracked each year. Therefore caution should be taken when drawing conclusions from the yearly mortality rate. The mortality rate of 66% over the entire 7.5-year investigation is probably a more accurate indicator of the death rate among free roaming Pinesnakes in their preferred habitat (Himes et al, 2002; Burger and Zappalorti 2016). In 2006, the majority of deaths (5 out of the 6), were due to red-tailed hawk predation when Pinesnakes were forced to hibernate within enclosed den corrals for one or two winters (**Figure 3**). Four of these deaths occurred in October of 2006. Even though sufficient ground cover was provided when the corrals were first erected, there was no protective netting placed over the top of the corrals to prevent raptors from taking Pinesnake when they emerged from the dens to bask. The corral walls restricted the snake's movements, and they relied on whatever ground cover that was provided inside the holding corrals. Unfortunately, when a Pinesnake crawled out in the open to bask, red-tailed hawks would kill and feed upon them. The hard lesson learned from this event was to always install suitable bird of prey netting over the holding corrals before placing snakes in them for "soft release."

To rectify this situation, HA and the NJDEP made arrangements to install sturdy netting to protect the snakes in the 6 artificial dens and holding corrals. In late March of 2007, our client Walters Homes, Inc. installed two 30-foot utility poles on opposite sides of each of the six holding corrals/artificial dens. Support cables were run between the two poles in a tent-like fashion and new heavy duty netting was installed over the 6 artificial dens and holding corrals. The new netting was 65-pound test, with 1-inch mesh openings that is commonly used near golf courses, in batting cages and game farms. The new bird netting proved effective in keeping red-tailed hawks and other birds of prey from killing the snakes and out of the corrals.

Unfortunately, a major forest fire in May of 2007 destroyed most of the netting. However, it was quickly replaced and no more snakes were lost due to raptor predation while they were inside the den corrals in the time it took to replace the netting. Once the Pinesnakes were released from the corrals, raptors continued to be a main cause of death among the free roaming radio-tracked snakes. This was no surprise to HA, since we have observed red-tailed hawks and other birds of prey feeding on various snake species in the wild (Zappalorti personal observations).

Aside from raptors, Pinesnakes (eggs, hatchlings and adults), have many other natural predators, including red foxes (*Vulpes vulpes*), raccoons (*Procyon lotor*), red squirrels (*Tamiasciurus hudsonicus*), northern short-tailed shrews (*Blarina brevicauda*), and other larger mammals. In addition to these historic predators, coyotes (*Canus latrins*) and the American black bear (*Ursus americanus*) have now become well established in New Jersey and their populations have risen over the past few decades (McConnell 1997; McBride 2006; NJ Fish and Game Council 2010). Black Bear may become an additional predator to Northern Pinesnakes and their eggs.

During the investigation, sightings of coyotes and evidence of their presence (tracks and scat) continually rose in the Pine Barrens. In fact in 2010, an HA staff member observed a coyote feeding on an adult Pinesnake alongside Hay Road in Ocean County, New Jersey. In addition, we also witnessed neonate pine snakes being eaten by coyotes when they emerged from their nests (Zappalorti, personal observations). It appears that the increase of coyotes in the Pine Barrens will have a significant negative impact upon the Pinesnake population.

The list of potential Louisiana Pinesnake egg predators in western Louisiana and eastern Texas is even larger than that of Northern Pinesnakes in New Jersey, because there are wild feral hogs, nine-banded armadillos (*Dasypus novemcinctus*) and the red imported fire ant (*Hymenoptera formicidae*), also in the mix. While armadillos chiefly eat invertebrates, they are also opportunistic in their feeding habits and have been documented to eat reptiles and amphibians, including snake eggs (Mengak 2005).

Fire ants have been documented to eat alligator eggs, turtle eggs, lizard eggs and snake eggs (Mount *et al.* 1981; Allen *et al.* 2001; Buhlmann and Coffman 2001; and Tuberville *et al.* 2000). In addition to coyote, gray fox, skunk, kingsnake and scarlet snake, red imported fire ants, and to a lesser extent, armadillos are all capable of eating eggs or hatchling Louisiana Pinesnakes (or any reptile or bird eggs), when they attempt to emerge from their eggs (while pipping).

With the aid of radio-telemetry, by monitoring adult gravid female Louisiana Pinesnakes in the wild, it could be determine where eggs are deposited, and if the eggs are indeed hatching or being preyed upon. Clearly, this type of research is needed in order to answer this important conservation question! All the above mentioned mammal and raptor species are present in the western Louisiana and eastern Texas landscape. It is likely that Louisiana Pinesnakes are also subject to predation by these animals.

Besides predation, three of the adult radio-tracked Pinesnakes in New Jersey, along with an untold number of the 2006 hatchlings, were killed in the May 16, 2007 major forest fire. While control burns are needed to manage longleaf pine forest, burning should only be done during the winter (December - February), when most snakes or other reptiles and amphibians are in active (hibernation). In addition to predators and natural occurring events, HA also lost radio-tracked Pinesnakes due to human intrusion. Two of the radio-tracked Pinesnakes were killed on roads (DOR's, one on the Garden State Parkway and one of Route 539 in Warren Grove).

Another adult Pinesnake was found dead behind a trailer complex located off of Stafford Park Boulevard. The type of wounds to this snake's body suggested it had died from blunt force trauma induced by wonton killing by a human. A construction work crew was in the same area the day the snake was found. **Figure 3** above, shows a pie-chart divided by the percentage of radio-tracked Pinesnakes lost due to different causes of death (Zappalorti, personal observations).

Among the various reasons for their decline is the construction of new roads through western Louisiana and eastern Texas, coupled with the advent of improved, well-traveled secondary roads. The once remote longleaf pine forest where Louisiana Pinesnakes persisted became accessible to all types of vehicles carrying hunters, loggers and tourists into their habitat. Most pinesnakes have large home ranges and an adult can travel up to 1.5-kilometers in a single 24-hour period (Zappalorti, personal observations). If Pinesnakes have to cross a paved or busy sand road, they are subject to impact with motor vehicles. Many of the documented Louisiana Pinesnake records are based upon live or dead on road (DOR) specimens (Rudolph et al 1999).

Other possible reasons for Louisiana Pinesnake declines would include the loss of Baird's Pocket Gopher populations. There is an ecological relationship between the two species. Aside from being a prey item of Louisiana Pinesnakes, Baird's Pocket Gophers provide underground tunnels that are suitable refugia and nesting habitat. Similar to Northern Pinesnakes, it is likely that gravid female Louisiana Pinesnakes seek the soft sandy substrate in the tunnels to excavate a nest burrow (a side chamber), to deposit eggs (Burger and Zappalorti 1986, 1991, 2011).

Nesting females not only need a relatively soft substrate to excavate a nest, but the nesting area must also have an open, sunny, treeless section of the forest that receives intense sunlight for several hours a day. Egg laying snakes and other reptiles depend upon the heat of the sun to incubate their eggs, so natural succession of trees and shrubs will cause too much shade in a once open, treeless habitat. Lack of prescribed burns in the longleaf pine forest would allow a once natural open area to eventually become closed canopy. Thus suitable nesting areas for Pinesnakes may be a limited habitat feature, that could directly effect hatching success and recruitment of Louisiana Pinesnakes (Burger and Zappalorti 1986, 1991, 2011).

2) - Prey organisms selected and eaten by Louisiana Pinesnakes

The diet preference of Louisiana Pinesnakes is somewhat different than that of Northern Pinesnakes, which is a function of southern mammal prey availability within their natural range. In other words, some small mammal species only occur in the south. In the New Jersey Pine Barrens certain small mammal species do not occur, such as Baird's pocket gopher, cotton rats (*Sigmodon hispidus*), harvest mice (*Reithrodontomys* sp.) among others. Baird's pocket gopher is the primary prey of the Louisiana pinesnake (Rudolph *et al.* 2002), comprising an estimated 53% of available individual prey records (75% of total prey biomass - Rudolph *et al.* 2012). The Louisiana pinesnake exhibits specialized prey handling behavior for the burrow-dwelling pocket gopher not common among most other constricting snake species (Rudolph *et al.* 2002). The Louisiana pinesnake is also known to eat eastern moles (*Scalopus aquaticus*), cotton rats, white-footed mice (*Peromyscus leucopus*), harvest mice (*Reithrodontomys* sp.), and turtle eggs of various species, including red-eared sliders (*Trachemys scripta* - Rudolph *et al.* 2002; Rudolph *et al.* 2012).

Foraging observations of Northern Pinesnakes are provided for comparison to what Louisiana Pinesnakes may eat in the wild. The various habitat types in the New Jersey Pine Barrens are rich with bird and small mammal resources which provide ample food supply for Northern Pinesnakes and other top predators (Burt and Grossenheider 1980; Arnold 1993 and Boyd 2000). HA operated a 3,500-foot perimeter drift fence trapping system (Enge 1997a and 1997b), in the ecotone habitat that separates the Stafford Business Park Redevelopment property from Stafford Forge WMA. The drift fence was operational for a four year period between April 15 and October 31 (2007 to 2010, the fence trapping was terminated on October 31, 2010). The following species of small mammals were seen or captured in the perimeter drift fence box funnel traps: short-tailed shrew (*Blarina brevicauda*), masked shrew (*Sorex cinereus* -



Figure 4. Masked Shrews caught in drift fence traps. Shrews are common in the Pine Barrens and serve as an important prey for hatchling Pinesnakes. Photo by Bob Zappalorti.

Figure 4), eastern mole (*Scalopus aquaticus* - **Figure 5**), star-nosed mole (*Cyanocitta cristata*), white-footed mouse (*Peromyscus leucopus*, red-backed vole (*Clethrionomys gapperi*), woodland vole (*Pitymys pinetorum*), meadow vole (*Microtus pennsylvanicum*), meadow jumping mouse (*Zapus hudsonius*), eastern chipmunk (*Tamias striatus*), red squirrel (*Tamiasciurus hudsonicus*), gray squirrel (*Sciurus carolinensis*) and eastern cottontail rabbit (*Sylvilagus floridanus*). All these small mammals were readily eaten by Pinesnakes being held in the laboratory or were seen eating in the field, while being radio-tracked (Zappalorti, unpublished data). The Pine Barrens has diverse habitat types including pine-oak, oak-pine, wetland sponges, and cedar swamp corridors.



Figure 5. An adult female Northern Pinesnake eating an Eastern Mole (*Scalopus aquaticus*). The snake constricted the mole in its burrow and came to the surface to swallow it as shown in the photo.

The snake management fields in the Stafford Forge Wildlife Management Area offered highly suitable habitat for an assortment of Pine Barrens wildlife species. Because of grassland habitat and forest-edge diversity there were ample prey resources available for the Pinesnake population (Burt and Grossenheider 1952, Reynolds and Scott 1982, Arnold 1993).

Additionally, many species of birds also occur in the Pine Barrens. Based upon HA's field observations and laboratory feeding experiments, the following birds were confirmed to be eaten by Pinesnakes: bobwhite quail (*Colinus virginianus* - eggs and chicks), whip-poor-will (*Caprimulgus vociferus*), eastern towhee (*Pipilo erythrophthalmus*), pine warbler (*Dendroica pinus*), wood thrush (*Hylocichla mustelina*), starling (*Sturnus vulgaris*), mourning dove (*Zenaida macroura*), American robin (*Turdus migratorius*), ovenbird (*Seiurus aurocapillus*), field sparrow (*Spizella pusilla*) house wren (*Troglodytes troglodytes*, and cedar waxwing (*Bombycilla cedrorum* - Zappalorti personal observations).

Table 2. Foraging and Feeding Observations of Radio-tracked Northern Pinesnakes in the New Jersey Pine Barrens between 2006 and 2016.

Snake ID	Date of Observation	Animal Species Consumed
2006.26	07/03/07	Bobwhite Quail eggs.
2007.05	08/09/07	White-footed Mouse
2007.04	06/26/08	Young Eastern Cottontail Rabbits.
2007.04	07/23/08	Young Eastern Cottontail Rabbit.
2007.15	08/25/08	Pine Vole in stump hole.
2007.10	09/18/08	White-footed Mice.
2007.05	05/22/09	Eastern Mole.
2007.05	04/22/10	Young White-footed Mice.
2008.03	05/05/10	Young Red Squirrels in nest.
2007.14	05/15/10	Young Eastern Cottontail Rabbits.
2007.14	06/07/10	Young Eastern Cottontail Rabbits.
2006.41	07/25/10	Eastern Mole
2007.11	08/21/10	Whip-poor-will (bird).
2007.10	09/22/10	Young Red Squirrels.
2007.07	05/09/11	Young Eastern Cottontail Rabbits.
2007.14	07/04/11	Cedar Waxwing Fledglings
2006.49	09/14/11	White-footed Mouse.
2007.10	04/20/12	Young Eastern Cottontail Rabbits.
2007.07	08/16/12	White-footed Mice.
2006.108	09/11/12	Raiding Red Squirrel nest inside the cavity of a standing dead pitch pine tree.
2006.46	04/24/13	Meadow Vole
2015.01	5/26/15	Eastern Mole
2016.05	6/1/16	Eastern Mole



Figure 6. Northern Pinesnake number 200.14, is attacking and consuming a Cedar Waxwing nestling. The young bird's head, neck and body can be seen hanging upside down from the snake's mouth. Photo by Dave Burkett, HA Staff.

By using rapid tongue flicking, Pinesnakes find their prey by following scent trails left by small mammals or birds (Gillingham and Clark 1981; Rudolph *et al.* 2002). During various radio-tracking studies, HA had the opportunity to observe different species of small mammals and ground nesting birds, many of which proved to be prey items for the Northern Pinesnake population (Zappalorti, personal observations). Ample prey availability and foraging success is an important part of a pinesnake population's survivorship (Arnold 1993; Rudolph *et al.* 2002).

In past studies, HA has witnessed radio-tracked snakes successfully subduing and eating wild prey items, such as the common mole, eastern chipmunk, young cottontail rabbits in a nest, white-footed mice and several bird species including bobwhite quail and their eggs. Several Pinesnakes were observed climbing shrubs and trees in search of bird eggs and chicks in nests (**Figure 6**). Within the Louisiana Pinesnake's range, aside from pocket gophers and moles, they probably eat the eggs, chicks and adults of many ground nesting bird species, but this remains to be confirmed.

HOME RANGE AND SEASONAL MOVEMENTS

Louisiana Pinesnakes studied in eastern Texas were found to be semi-fossorial and mostly diurnal, and were also relatively immobile (*i.e.*, moved less than 33-feet or 10-meters) on 54.5% of days monitored (Ealy *et al.* 2004). In one study, snakes spent on average, 59% of daylight hours (*e.g.*, sunrise to sunset) below ground, and moved an average of 541-feet (163-meters) per day (Ealy *et al.* 2004). These data compare favorably with movements and behavior of Northern Pinesnakes (Zappalorti *et al.* 2015; Burger and Zappalorti 2011).

In a Louisiana study by Himes *et al.*, snakes moved an average of 495-feet (150-meters) daily, with the longest movement being 3,802-feet (1,159-meters), whereas adult females moved on average 348-feet (106-meters), and juveniles moved 112-feet or 34-meters (Himes 1998). Himes *et al.* (2006), documented an average home range size of 82-acres (33.2-hectares) with a range of 16 to 267-acres (6.5 to 108-hectares) for the Louisiana Pinesnake. Himes (1998), also found that adult males had larger average home ranges of 145-acres (58.7-hectares), than adult females with 25-acre (14-hectare), and juveniles had average movements of 13-acres (5.5-hectares).

As a comparison to home range sizes for Louisiana Pinesnakes, HA studied home ranges and maximum dispersal distance from hibernacula in a population of Northern Pinesnakes (n= 40), at a 1,417.5-hectare preserve in Cumberland County, New Jersey, between 1993 and 2003. We discovered 22 different winter hibernacula that were used by this Pinesnake population. Of the 2 male and 8 female snakes monitored in hibernacula for 5-years, voluntary shifting was observed by 8 individuals. Seven snakes shifted dens between years once and one male shifted dens twice. In contrast, 2 females showed den philopatry for 5 consecutive years (Zappalorti *et al.* 2015).

Radio-tracked snakes were relocated in their habitat between 20 and 140 times. The average MCP home range size of 27 radio-tracked Pinesnakes was 69.26-hectares (170.71-acres). An adult male had the largest home range (184-hectares or 455.70-acres). The maximum distance traveled from its winter den was 1,609 meters (1.609 kilometers and/or 5,280 feet). The average maximum distance traveled by radio-tracked Pinesnakes, to and from their winter dens, was 952.9-meters (0.9529-kilometers). Of these, 50% (n=20) snakes traveled more than 1,000-meters, 20% (n=8) snakes traveled 1,100-meters, 10% (n=4) snakes traveled 1,200-meters, and 2.5% (n=1) snake moved 1,609-meters. Some snakes were only radio-tracked for one full year, while others were tracked for 3 to 5-years.

Snakes that were monitored for 2-years or more had larger home ranges than those individuals that were only radio-tracked for one year. Based upon these data, radio-tracking several adult snakes over a 3 to 5-year period is not only the most efficient method to find hibernacula locations of meta-populations, but reveals a more complete understanding of their ecology, secretive behavior and conservation needs (Zappalorti *et al.* 2015). Based upon the above home range data, it appears that Louisiana Pinesnakes have slightly smaller home ranges than Northern Pinesnakes. That aside, many other aspects of their life history are similar where they carry out all necessary biological activities which helps them to survive in their habitat.

Louisiana Pinesnakes are highly fossorial and seek shelter in pocket gopher burrows where they are sympatric. They otherwise occasionally use old pine tree stumpholes or hollow fallen logs as refugia. In New Jersey, Northern Pinesnakes were studied to determine their preferred habitat use and most frequent behavior patterns. They typically used pitch pine forest or pine-oak dominated forest with ample hollow fallen logs, abandoned mammal borrows or stump holes for shelter and nighttime retreats (Burger and Zappalorti 2011; Zappalorti *et al.* 2015). On occasion they used forest ecotone habitat and open sunny grassy fields for basking as shown in **Figures 7 and 8**.

HABITAT USE AND BEHAVIORAL ANALYSIS

Radio-tracking and monitoring of Northern Pinesnakes at the SPR property and the adjacent Stafford Forge WMA revealed some interesting habitat preferences (**Table 3**). **Figure 7** provides a comparison by percentage of habitat selected by the radio-tracked shifted, non-shifted, and 2006 hatchling pine snakes during the six and a half year investigation (McCormick 1970 and 1979, Burger and Zappalorti 1989a, Boyd 1991). **Figure 8** shows a comparison by percent of the most frequent behaviors observed by radio-tracked Pinesnakes within Stafford Forge WMA.

For the purpose of the investigation, habitat types selected and used by Northern Pinesnakes in Stafford Forge Wildlife Management Area are defined as follows:

Open Field - little or no trees, sandy soil often dominated by various native grass species.

Artificial Hibernaculum - artificial snake shelter or den, designed and constructed by HA and located in the management fields.

Barren Ground/Disturbed - habitat with little to no vegetative cover or was cleared and altered by human disturbance.

Ecotone Between Upland and Wetland - transitional edge between upland forest habitat and wetland habitat.

Forested Wetland - hardwood trees and/or cedar dominated wetland corridors.

Ecotone Between Forest and Barren Ground - transitional habitat between upland forest and disturbed or barren habitat (*e.g.*, the management fields, and SPR property).

Pine/Oak Forest - pitch pine dominated forest, but containing an oak component.

Oak/Pine Forest - oak dominated forest, but containing a pitch pine component.

Pine Forest - pitch pine forest with no other overstory tree species present.

Selectively Thinned Forest - area of forest within Stafford Forge Wildlife Management Area that was selectively thinned by the New Jersey Division of Parks and Forestry.

Emergent Wetland - open canopy wetland habitat with herbaceous vegetation dominating the saturated substrate.

Note: The above listed forest types and descriptions were modified from McCormick (1970 and 1979) and Boyd (1991).

Table 3. Habitat Preferences of Radio-tracked Pine Snakes at Stafford Forge WMA and the Stafford Park Redevelopment Property between 2007 and 2013.

Habitat Types	All Snakes (n = 6628)		Shifted Snakes (n = 3078)		Non-Shifted Snakes (n = 3274)		2006 Hatchlings (n = 276)	
	Number of Relocations	Percent of Total	Number of Relocations	Percent of Total	Number of Relocations	Percent of Total	Number of Relocations	Percent of Total
Developed Land	1	0.02%	0	0%	1	0.03%	0	0.0%
Habitat/Development Interface	2	0.03%	1	0.03%	1	0.03%	0	0.0%
Emergent Wetland	15	0.23%	11	0.36%	4	0.12%	0	0.0%
Selectively Thinned Forest	32	0.48%	2	0.06%	30	0.92%	0	0.00%
Open Field	50	0.75%	28	0.91%	18	0.55%	4	1.45%
Artificial Hibernaculum	115	1.74%	109	3.54%	0	0.00%	6	2.17%
Barren Ground/Disturbed	159	2.40%	96	3.12%	62	1.89%	1	0.36%
Ecotone Between Field and Forest	182	2.75%	146	4.47%	21	0.64%	15	5.43%
Ecotone Between Upland and Wetland	239	3.61%	90	2.92%	147	4.49%	2	0.72%
Forested Wetland	329	4.96%	168	5.46%	160	4.89%	1	0.36%
Ecotone Between Forest and Barren Ground	349	5.27%	250	8.12%	85	2.60%	14	5.07%
Oak/Pine Forest	1138	17.17%	518	16.83%	608	18.57%	12	4.35%
Pine/Oak Forest	1413	21.32%	577	18.75%	782	23.89%	54	19.57%
Pine Forest	2604	39.29%	1082	35.15%	1355	41.39%	167	60.51%
Total Relocations	6628	---	3078	---	3274	---	276	---

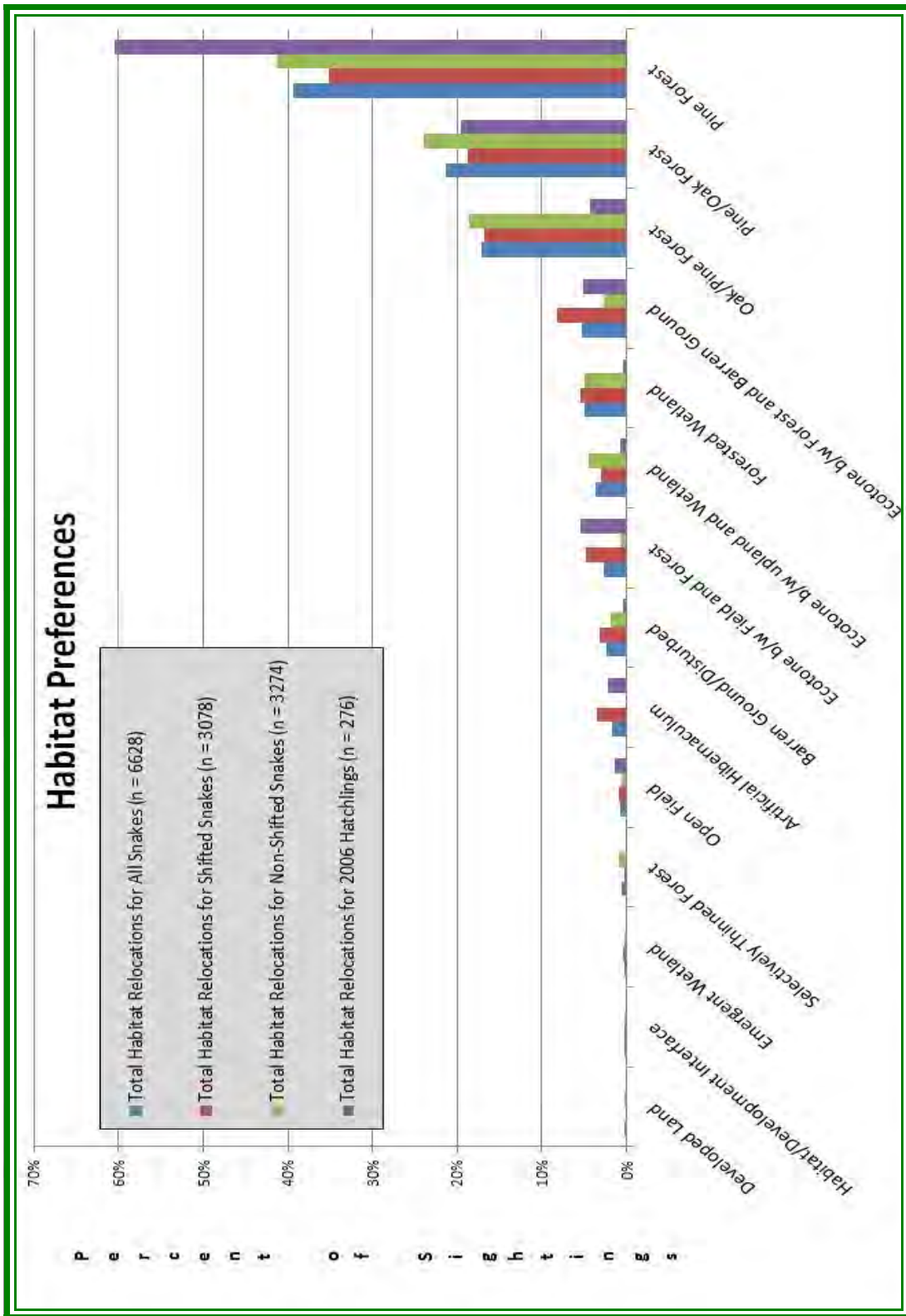


Figure 7.

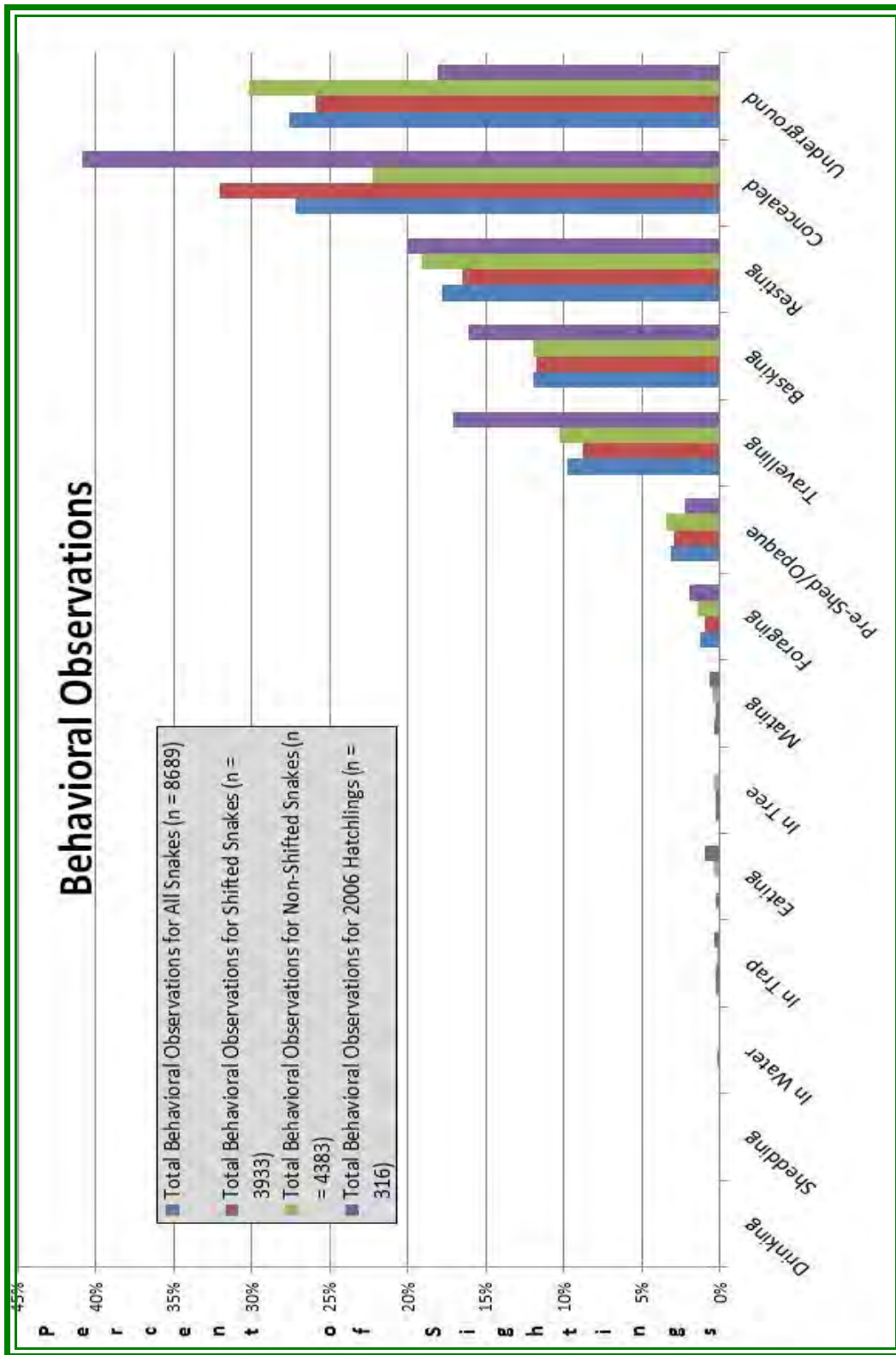


Figure 8.

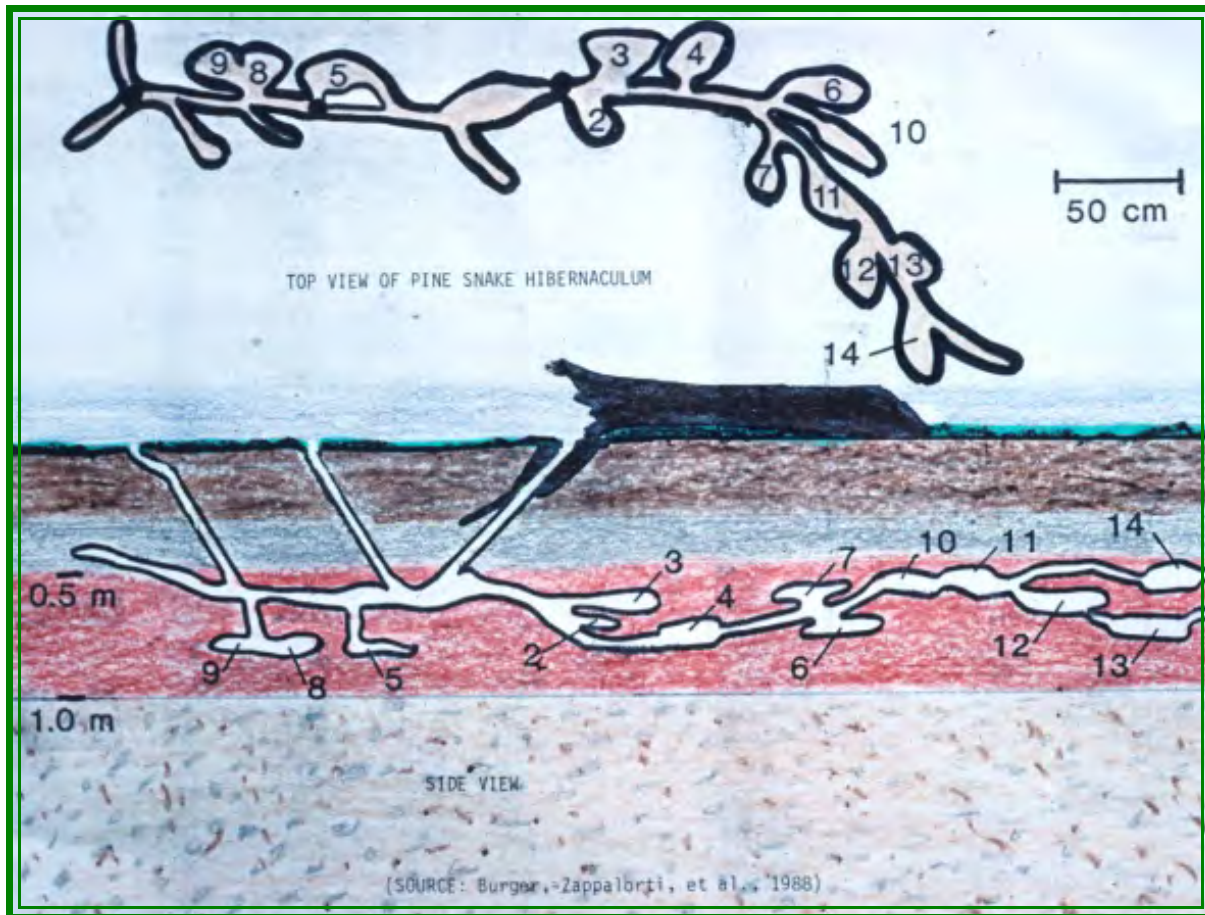


Figure 9. A diagrammatic drawing of a natural Pinesnake hibernaculum that was excavated by Joanna Burger and Bob Zappalorti and drawn to scale. There was one Northern Black Racer and 13 Northern Pinesnakes hibernating in this den. We found three entrances, one at the base of a fallen pine stump, and two on the surface at 60-cm and 90-cm from the stump hole. The snakes were down between 30 and 80-cm from the humus surface, in the B Horizon of the sandy soil (Burger et al, 1988).

WINTER HIBERNACULA

Based upon my habitat evaluations in western Louisiana and eastern Texas between 2000 and 2006, suitable winter hibernacula were available for Louisiana Pinesnakes (Rudolph et al 2007, Zappalorti personal observations). Additionally, active and abandoned pocket gopher burrows are also used as winter and/or summer retreats. Several other species of snakes, including timber rattlesnake, southern copperhead, gray rat snake, red rat snake, eastern hognose snake, black racer, and eastern coachwhip bask by stump holes and pocket gopher burrows. In New Jersey, old decaying pine and oak stump holes (**Figure 9**), and abandoned mammal burrows are most frequently used as winter hibernation sites by Pinesnakes (Burger et al 1988, Zappalorti *et al.* 2014). Pinesnakes often share their winter dens with several of the abovementioned species.

Because of the warmer climate within Louisiana and Texas, the lack of suitable hibernacula is not a limiting factor for safe overwintering sites. However, in a region where pocket gophers occur in low numbers, are completely absent, or there is a lack of suitable pine stump holes for Louisiana Pinesnake to use as winter dens, this problem could easily be remedied by constructing artificial shelters or hibernacula. They could be constructed in designated management fields (Zappalorti and Reinert 1986; Burger and Zappalorti 2011). In New Jersey, Pinesnakes hibernate either at individual den sites or at communal dens. Hibernacula are sometimes in old stump-hole root systems (**Figure 9**), or the snakes locate abandoned mammal burrows in which to spend the winter. Pinesnakes have been found hibernating with other conspecifics (up to 16 snakes in one den), along with other species of snakes such as black racer, hognose snake, coastal plains milk snake, corn snake, black rat snake and timber rattlesnake (Burger et al. 1988, 2000; Zappalorti, personal observations).

Most of the stump hole hibernacula are dug or expanded by the Pinesnakes themselves. The snakes follow old rotten tree roots and dig down to the B horizon. When they use abandoned mammal burrows, the snakes dig side chambers (Burger *et al.* 1988). There are usually several side chambers off the main tunnel, which can be as long as 4 to 6-meters.

Chambers with snakes can be between 50 and 120-cm below the ground surface, depending upon winter temperatures. Snakes tend to use the same hibernacula (or hibernacula system) in consecutive years (Burger *et al.* 1992 and 2007). When several hibernacula are in the same general area, snakes often shift from one den to the other in different years (Zappalorti *et al.* 2015).

Adult male Pinesnakes shifted between winter dens more frequently than females (Zappalorti *et al.*, 2014 and 2015). Hibernation sites are sometimes used in the summer for resting, avoiding predators or high surface temperatures (Burger *et al.* 1988; Zappalorti *et al.* 2015). Young and adults locate hibernation sites by following the chemical scent trails of other Pinesnakes, as clearly demonstrated by laboratory experiments (Burger 1989b), and they are capable of distinguishing the odors of conspecifics from predators (Burger 1989b, 1991b, Burger *et al.* 1991; Zappalorti *et al.*, 2014 and 2015).

USE OF THE MANAGEMENT FIELDS BY GRAVID FEMALES FOR NESTING

Twenty one of the radio-tracked Northern Pinesnakes were confirmed using the management fields during the course of the investigation. This included snakes from the shifted set, the non-shifted set and the radio-tracked 2006 hatchlings. While in the management fields the radio-tracked snakes were observed exhibiting multiple natural behaviors which was the intended purpose predicted in the management plan design (Zappalorti and Golden 2006). In addition to the radio-tracked snakes, HA researchers visually confirmed a non-implanted Pinesnake (one without a radio-transmitter) digging a nest in management field 2 during the 2008 nesting season (**Figure 10**). This gravid female was observed excavating a nest chamber at the base of a large earth mound. This was the only non-transmitted (radio-implanted) female Pinesnake observed nesting in the management fields during the 2008 investigation (Zappalorti, personal observation).

However, HA researchers would corral the artificial dens in the late summer and early fall of each field season in order to capture any snakes that were trying to enter the dens to overwinter. In 2011, HA captured 6 Northern Pinesnake hatchlings in the den traps. By looking at Table 3 below, one can see that there were no gravid radio-tracked females documented nesting in the management fields in 2011. Therefore, the fact that hatchling pine snakes were present in the management fields in the late summer and early fall, strongly suggests that one additional non-radio-tracked gravid female Pinesnake nested in 2011. Additionally, in 2012 only one of the radio-tracked gravid Pinesnake was observed nesting in the management field 3, resulting in 12 hatchling Pinesnakes being found in late fall (October 2012), or caught in the artificial den traps. Finding twelve hatchlings in the fields in late summer and early fall strongly suggests that more than one Pinesnake nested in the fields that year.

Table 4. Radio-tracked Pine Snakes that Nested in the Management Fields.		
Snake's Field Number	Management Field No. and Location	Year
2007.05	West berm of MF 2.	2008
2006.29	Earth mound in middle of MF 3.	2009
2007.04	North mound of MF 3.	2009
2007.15	Same earth mound as 2006.29 in MF 3.	2009
2007.05	West berm of MF 2.	2009
2007.07	East berm of MF 1.	2009
2007.07	MF 1.	2010
2006.19	West berm of MF 1.	2010
2006.46	East berm of MF 3.	2012
2006.46	East berm of MF 2. (Suspected)	2013
2007.07	Excavated nest in MF 3.	2013

Nesting Behavior

HA documented both radio-tracked study snakes and non radio-tracked snakes successfully nesting in the management fields during the investigation. Between 2008 and 2013, a total of six radio-tracked snakes were confirmed to have nested in the management fields. **Table 4** above, lists the radio-tracked gravid pine snakes that nested in the fields chronologically by year and which management field they were documented using.

3) - *Were do gravid female Louisiana Pinesnakes deposit their eggs?*

Clearly, it is difficult to capture Louisiana Pinesnakes in their habitat, and even more challenging to secure gravid females. However if a concerted effort was made, it is possible to capture them early in the season before they are bulging with eggs, so radio-transmitters could be safely implanted. Then, with the aid of radio-telemetry adult gravid female Louisiana Pinesnakes could be monitored carefully in the wild. Gravid snakes spend most of their time basking while the eggs are developing within their body, but in the south warm surface temperatures may allow them to remain hidden below ground. Early morning searches may help to find gravid females basking on the surface.

When ready, gravid female Louisiana Pinesnakes will select and excavate a nest site. The nest site may be in a pocket gopher burrow, a mole tunnel, or stump hole. Once the female snake goes underground to deposit her eggs, researchers could find the nest, record data and monitor it. Decisions could be made whether to protect the eggs in the field or to hatch the eggs in the laboratory. This type of research could help determine where Louisiana Pinesnakes deposit their eggs. More important, it could be learned if the eggs are indeed hatching or are the eggs being preyed upon. Clearly, this type of research is needed in order to answer this important conservation question.

Although there is a captive-breeding population of Louisiana Pinesnakes being maintained across 18 AZA accredited institutions and 2 non-AZA partner institutions, this important conservation effort does not protect Louisiana Pinesnake eggs and hatchlings in the wild. This captive population, established in 1984, has been managed under an AZA Species Survival Plan (SSP) since 2000. As of March 2016, this captive-breeding population consists of 111 individuals (51 males, 53 females, and 7 unsexed). Since 2010, this population has provided 77 captive bred Louisiana pinesnakes for release into the wild. However, the survivorship of these snakes is poorly understood.

4) *What is the hatching success and survivorship of Louisiana Pinesnakes?*

Another important conservation measure would be to protect eggs from predators. If Louisiana Pinesnake eggs were hatched in the laboratory and the young were immediately released back to the wild after collecting data and PIT tagging them with micro-chips, this action would help ensure hatching success and a possible better chance of survivorship. However, I do not recommend head starting hatchling snakes, because they lose the ability to imprint to their nesting area, do not learn how to successfully forage for prey, lose the ability establish a home range, are not equipped to avoid predators and are subject to become easy targets for a host of mammal and bird predators (Zappalorti, personal observations).

Instead, the hatchlings, shed skins and egg shells should be returned to the exact site from where they were found. By returning the eggs and shed skins, it is more natural and helps the hatchling snakes imprint to the nesting area (Burger and Zappalorti 1986; Burger 1989; Burger and Zappalorti 1992). Once the hatchling snake emerges from the nest site to disperse into the habitat, the learning process begins for survivorship. The nest site is the center of the home range.

However, finding gravid female Louisiana Pinesnakes (or their eggs) in Pocket Gopher tunnels is difficult, so great effort should be invested to identify where, how and when gravid females deposit their eggs. To that end, one method that should be considered is to use specially trained detector dogs to locate Louisiana pinesnakes and their nests (egg deposition sites) in their natural habitat (Stevenson *et al.* 2010). I have worked on two snake projects using detector dogs, one with Pinesnakes in New Jersey and one with Indigo Snakes in Georgia and Florida. The results were positive because the dog was able to find Pinesnake eggs underground and Indigo Snakes in Gopher Tortoise burrows (Dave Golden NJDEP, personal communication; Stevenson *et al.* 2010).

Louisiana pinesnakes are highly secretive serpents and spend much of their time hidden in various animal burrows, hollow logs, stump holes and under human debris dumped in the forest. Therefore, they are not easily found in densely vegetated habitats unless they are trapped, seen crawling in the open or crossing roads. With the help of an organization known as *Working Dogs for Conservation*, the USF&WS should experimentally field test dogs in western portions of Louisiana and eastern sections of Texas (the current known critical habitat), to investigate if dogs can be used as a helpful tool to locate Pinesnakes in the wild. Dogs could be trained using live Pinesnakes and/or freshly shed skins for chemical scent recognition and memory imprinting. Additionally, nest site selection by gravid female Louisiana Pinesnakes is currently poorly understood. Dogs could be trained to locate Louisiana Pinesnakes eggs to learn where gravid females typically select their nest sites.

CONSERVATION AND MANAGEMENT RECOMMENDATIONS

Aside from prescribed burning of the longleaf pine forest ecosystem every 3 to 5-years, some other additional habitat management may be required. For instance, creating protective brush pile shelters built from cut hardwood and pine trees would provide suitable and safe basking habitat. Managing open sandy portions of the forest with an abundant ground-layer of herbaceous vegetation would be important for Baird's pocket gopher populations. Keeping a rich array of abundant grasses and other herbaceous vegetation would provide suitable habitat for both the Louisiana Pinesnakes and Baird's pocket gophers. The USF&WS may want to consider experimental translocation and repatriation of captive bred hatchling Louisiana Pinesnakes to protected habitat within their historic range. Ideal recipient sites should be selected that have suitable forest structure, ample prey and foraging opportunities, the presence of Pocket Gophers (or ample pine stump holes), suitable soils for nesting, open grassy basking areas, and overwintering sites that could be managed specifically to benefit Louisiana Pinesnakes. Release sites should be a minimum of 1,500-acres (607-hectares), because of the large home range size of Louisiana Pinesnakes. Himes *et al.* (2006), documented an average home range size of 82-acres (33.2-hectares). Himes *et al.* (1998 and 2006) also found that adult males had larger average home ranges of 145-acres (58.7-hectares) than adult females with a home range of 25-acres (14-hectares). Given the large home range size of Louisiana Pinesnakes, any proposed release area should be large enough to allow free roaming adult snakes to move about in their habitat without restrictions. Additionally, the proposed recipient sites should not have any paved roads within 1.0-mile of the core managed habitat.

SUMMARY

I commend the U.S. Fish and Wildlife Service for compiling a detailed historical chronology and a thorough life history account of the Louisiana Pinesnake in the Federal Register (proposed rule document - USFWS 2016). There were only a few important life history details that seem to be lacking in the proposed rule document that require additional research. These are: 1) predation and types of predators of free roaming Louisiana Pinesnake, 2) the selection and types of prey organisms eaten by Louisiana Pinesnake, and 3) additional data on nest site selection, hatching success and survivorship of Louisiana Pinesnakes in their natural habitat.

To that end, I have provided information which was gathered on Northern Pinesnakes over a 46-year period that can be extrapolated to the Louisiana Pinesnake. While there are differences in geography, climate, and forest vegetation species, there are many similarities in behavior, habitat structure and burrowing propensities. In my opinion, what we have learned about Northern Pinesnakes could be used to help with a conservation and management of the Louisiana Pinesnake. Herpetological Associates, Inc. recommends and fully supports the USF&WS's proposed listing of the Louisiana Pinesnake for the reasons provided above in this review document. Clearly, Pinesnakes are declining throughout their range in the eastern United States.

A good example of this decline is in the state of Virginia, where the Pinesnake has not been documented in over 30-years (J.D. Kleopfer, personal communication). They are listed as threatened or special concern species in most states where Pinesnakes still remain. Obviously, a Louisiana Pinesnake Conservation and Management Plan is needed that is based upon current science and best management practices. HA would like to be part of the planning and conservation team because we have the knowledge and experience working with Pinesnakes throughout their range in the eastern United States. The USF&WS's recovery goals can only be achieved with proper funding and by working with partners.

I agree with the U.S. Fish and Wildlife Service, that the remaining longleaf pine forest ecosystem which occurs within the known (and potential) longleaf pine landscape areas in western Louisiana and eastern Texas are critical habitat for the Louisiana Pinesnake. Confirmed Louisiana Pinesnake areas on private lands with highly suitable habitat should be purchased if funding is available. Or if outright purchase is not possible, conservation easements should be sought from the land owners to protect these disjunct meta-populations. I am ready, willing, and able to help the USF&WS develop a conservation, management and recovery plan for the Louisiana Pinesnake.

Respectfully Submitted,

Robert T. Zappalorti

Herpetological Associates, Inc.