

1-1-2010

Effect of Snake Populations on Salamanders as a Result of Forest Fragmentation

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Effect of Snake Populations on Salamanders as a Result of Forest Fragmentation

Thesis submitted to
The Graduate College of
Marshall University

In partial fulfillment of the
Requirements for the Degree of
Master of Science
Biological Sciences

By
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Marshall University

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May 2010

ABSTRACT

Effect of Snake Populations on Salamanders as a Result of Forest Fragmentation

Forest fragmentation is one of the main causes for the loss of native biodiversity. One consequence is increased proportion of edge habitat that introduces new “edge” species, and makes habitat for interior forest-living species less-suitable. This study was conducted at three sites in Tucker County, West Virginia and included one downhill ski slope, one cross country ski slope, and one gravel road. The main objectives of this study were to determine relative abundance of snake communities, how far species move from edge habitat into the forest and to determine whether snakes are a predatory threat to salamanders, specifically the federally protected Cheat Mountain Salamander (*Plethodon nettingi*). Area constrained surveys were conducted at each site from June through mid-October 2009. Three transects were placed at each study site and a vegetation analysis was conducted to quantify changes in plant communities. Snakes found were measured for snout-to-vent length and total length. Gender was determined and each specimen marked for recapture data. Results show the majority of snakes were found along the forest edge under cover objects where direct sunlight heats the ground and rocks. The highest concentration of all salamander species were found along transects deeper into the forest. Snake species observed included 44 Northern Ring-necked Snakes (*Diadophis punctatus edwardsii*), 15 Red-bellied Snakes (*Storeria o. occipitamaculata*), 35 Eastern Gartersnakes (*Thamnophis s. sirtalis*) and 5 Smooth Greensnakes (*Opheodrys vernalis*), all of which include a diet of salamanders to some extent. Results from this study will not only provide data on the implications of forest fragmentation, but will also provide the US Fish and Wildlife Service vital information on revising the Cheat Mountain Salamanders recovery plan.

ACKNOWLEDGEMENTS

I would first like to thank my advisor, Dr. Thomas K. Pauley for giving me the opportunity to be part of his lab. I have learned more in the past two years not only about the importance of natural history, conservation and herpetology, but probably more importantly life lessons that will stick with me forever. I am extremely thankful that Dr. Pauley took me on as a graduate student and that I had the amazing opportunity to learn from one of the best herpetologists of the day. His patience, humor and ability to pick up the pieces when we, as graduate students, think our lives are falling apart are the reasons I made it through my graduate career. I would also like to thank my committee members, Dr. Bill Sutton, Dr. Evans and Dr. Strait for all the suggestions and revisions. Especially Dr. Sutton and Dr. Jayme Waldron for help with data analysis, without their help I would still be lost in the wonderful world of statistics.

I want to especially thank my mom, Deb, and brother, Carl, for telling me to follow my heart when it came to finding a career. I want to thank my mom for encouraging me to become a biologist at a young age; letting me play in the mud, constantly catching and bringing home animals, and having patience with me when I didn't understand why I couldn't keep turtles in our only bathtub. I'm thankful for the rest of my family and friends have been nothing but supportive in every adventure I have found thus far, and I wouldn't have made it without them. I am very thankful for Tyler, my fiancé, whose patience and support during the past two years has been unwavering. He made a long distance relationship work even though I wasn't the most pleasant person to talk to on the phone many days, and I can't wait to spend the rest of my life with him.

Several fellow graduate students assisted with field work: Kevin Messinger, Nathan Shepard, Scott Jones, Aaron Gooley, Doug Horchler, Amanda Spriggs, Tyler Hern, Katie Murphy and Sarah Miloski. I want to especially thank Katie for her growing friendship and more importantly, her endless supply of peanut butter sandwiches in the field. Without them I would have starved to death. And, Sarah, for making our field experiences such an array of adventures and laughs, never having a dull moment. Kevin, thank you for teaching me basically everything I know about snakes and letting me use some of your equipment. I am very thankful for Amanda, and the friendship we have gained over the last two years. I want to thank Tyler Hern for helping me with ArcMap, statistics, taking me fishing all the time and playing racquetball constantly with me when I just needed to hit things. I want to also thank former

graduate student Casey (Boy) for taking me into the field my first year of grad school and teaching me so much about WV herps and birds, and of course for being my gym buddy.

Wildlife biologist, Ken Sturm, was a big help during my field season and everyone else from Canaan Valley NWF. I was extremely thankful to them for letting me use their field house and have permission to conduct research on their land. Lastly, I want to thank the Forest Service for allowing me to work in the Monongahela National Forest.

This study was completed under Marshall University Animal Use Protocol Numbers 424-1, 424-2, 424-3, 424-4, 424-5, 424-6, and 424-7. Animals were collected under West Virginia Collecting Permit #2009.176

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INTRODUCTION

Habitat loss and fragmentation are posing severe consequences to natural biodiversity around the world (Wilson, 1992). Habitat loss reduces available living space which causes fluctuations in species richness and abundance (Blouin-Demers and Weatherhead, 2001). Fragments of forest are often separated by highly modified or degraded habitat (i.e. roads, ski slopes, powerlines) and edges often times have an altered set of conditions which is referred to as edge effect (Primack, 2006). Fragmentation increases edge habitat, which can introduce entirely new environments and change existing habitats. Edges can experience microhabitat changes in light, temperature, wind and humidity (Laurance, 2000). This change may affect predator-prey relationships (Blouin-Demers and Weatherhead, 2001) and cause direct consequences to prey species that would normally be protected by virtue of habitat characteristics. In areas with high degrees of fragmentation, species that live in interior sections of the forest may experience population declines due to a less suitable environment (Blouin-Demers and Weatherhead, 2001). Conversely, increased proportions of edge habitat may introduce edge-associated predators such as snakes (Blouin-Demers and Weatherhead, 2001). Animals that use the same habitat, such as snakes and salamanders, are equally vulnerable to habitat loss and destruction and are susceptible to these environmental changes (Gibbons et al., 2000). Fluctuations in animal species is common, however it can be difficult to determine whether fluctuations are occurring due to natural causes or human impact (Pechmann et al, 1991). How animals use provided habitat is important in determining interactions within the environment. Space use can be described by home range sizes, movement patterns within home ranges and if those home ranges are shared with other individuals (Andrassen et al., 1998).

In many areas, small snakes make up a great percentage of vertebrate biomass and play an important role in predator-prey interactions (Willson and Dorcas, 2004). Increasing the degree of edge habitat increases direct exposure to the sun. Snakes are exothermic animals that maintain their body

temperature via microhabitat and daily activity (Blouin-Demers and Weatherhead, 2001). Thermal demands for snakes may be higher when females are gravid, digesting food, or after shedding (Carfagno and Weatherhead, 2006). Fragmented forested areas are excellent habitat for snakes because they provide areas exposed to direct sunlight as well as sections of shady forest (Blouin-Demers and Weatherhead, 2001, Carfagno and Weatherhead, 2006), both of which are important in maintaining ideal body temperatures. It is important to note that not all species of snakes behave identically in the same habitat; different species may utilize different areas in the same study site (Reinert, 1984). Sex of snakes and whether or not a female is gravid may also change how snakes use habitat (Carfagno and Weatherhead, 2006b). Abundance of prey in an area may be less important for species of snakes with a general diet compared to those with a more specialized diet (Carfagno et al., 2006a).

It is vital that studies be conducted to determine the use of habitat edges by predators such as snakes to understand the relationship between landscape configuration and predation on prey items such as salamanders (Blouin-Demers and Weatherhead, 2001). Snake studies can be difficult because many of them have patchy distributions, are difficult to find when active and are often times nocturnal (Lind et al., 2005). It is important to gather data on snake's diet preference due to their strong effects in the food web and lack of published data (Lind et al., 2005).

Management of snakes and salamanders is key to maintain stable interactions in environments. Studies that provide information on forest fragmentation and predators' use of edge habitat as well as interior sections of the forest will help improve conservation efforts for many species of wildlife. There may be conservation needs we are unaware of for threatened or endangered species negatively affected by forest fragmentation, but first it must be determined if an introduced conflict between snakes and salamanders exists. To gather enough information to decide whether or not a species is imperiled is a time consuming and detailed process (Gibbons et al., 2000). Research is required to help

determine whether species are declining due to natural fluctuations in the environment or human influence (Pechmann et al., 1991). Herpetofauna around the world are currently being exposed to known and unknown pressures that will continue to increase extinction rates and extirpated species if biologists conduct research that informs us of all habitat requirements (Gibbons et al., 2000). To manage an area properly it is vital to understand the requirements and relationships between plants and animals (Uhler et al., 1939).

Plethodon nettingi (Cheat Mountain Salamander) is a species that has experienced population declines and limited range in West Virginia. The study was prompted in hopes of gaining a better understanding of snakes movement and foraging habits in areas where *P. nettingi* reside. The main objectives of this study were to determine relative abundance of snake communities, how far species move from edge habitat into the forest and to determine whether snakes are a significant predatory threat to the federally protected Cheat Mountain Salamander (*Plethodon nettingi*).

Species Accounts

Salamanders found throughout my study sites belong to the family Plethodontidae (lungless salamanders) which is the largest family of salamanders in the eastern United States. Lungless salamanders respire through the skin and lining of the mouth (Conant et al., 1998), and can inhabit a variety of habitats from completely terrestrial to entirely aquatic (Hulse et al., 2001). The genus *Plethodon* (woodland salamanders) contains the largest number (43) of salamander species in the United States. They are found in forested habitats and are most active when humidity is high (Hulse et al., 2001). Plethodontid salamanders are divided into two subfamilies: Plethodontinae (woodland salamanders) and Desmognathinae (dusky salamanders) (Conant et al., 1998).

Dusky salamanders include 12 species found in the eastern United States (Hulse et al., 2001). They are extremely variable in coloration and patterns between adults and juveniles as well as individual

populations (Conant et al., 1998). These salamanders are semi aquatic and are found under cover objects near water and small streams. They tend to avoid large streams that house predatory fish (Hulse et al., 2001).

Cheat Mountain Salamander (*Plethodon nettingi*)

The United States Fish and Wildlife Service listed the Cheat Mountain Salamander as a threatened species on September 28, 1989 (Pauley, 1991). These salamanders are particularly affected by forest fragmentation, including ski slopes and these areas of fragmentation may be separating gene pools, which could potentially results in subpopulations (Pauley, 2008). Pauley has suggested that construction and development in *P. nettingi* habitat should remain at least 90m from preferred habitat in areas where they are known to occur (Pauley, 2008a).

Cheat Mountain Salamanders are small, woodland salamanders that reach a total length of 10.2cm. Dorsal coloration is dark black or brown with gold or silvery flecking. The venter is dark, usually gray or black (Green and Pauley, 1987) (Figure 1).

This species is endemic to West Virginia, only found in 5 counties (Randolph, Pendleton, Pocahontas, Tucker and a small portion of western Grant) in the high elevations of the Allegheny Mountains (Pauley, 2007). Total range from north to south is around 92km from Blackwater River Canyon to Thorny Flat on Cheat Mountain (Pauley, 2007). From east to west, range extends only 3.2km at the southern portion and 31km in the north (Pauley, 2008a) (Figure 2).

Habitat includes areas that were primarily Red Spruce (*Picea rubens*) forests mixed with yellow birch (*Betula alleghaniensis*). After logging and fires between 1890-1920, these Red Spruce dominated forests now consist of mixed deciduous trees with occasional Eastern Hemlock (*Tsuga canadensis*) and disjunct *P. nettingi* populations can be found in these areas above 610m (Pauley 2008b). Ground cover

usually consists of the liverwort, *Bazzania trilobata*. *Plethodon nettingi* prefer cover objects such as logs, rock, leaf litter or even cover boards during the day and appear on the surface at night foraging for prey. Mating most likely occurs in April or May and females will lay eggs under cover objects such as logs or rocks (Pauley, 2008).



Figure 1: Adult male *Plethodon nettingi*.

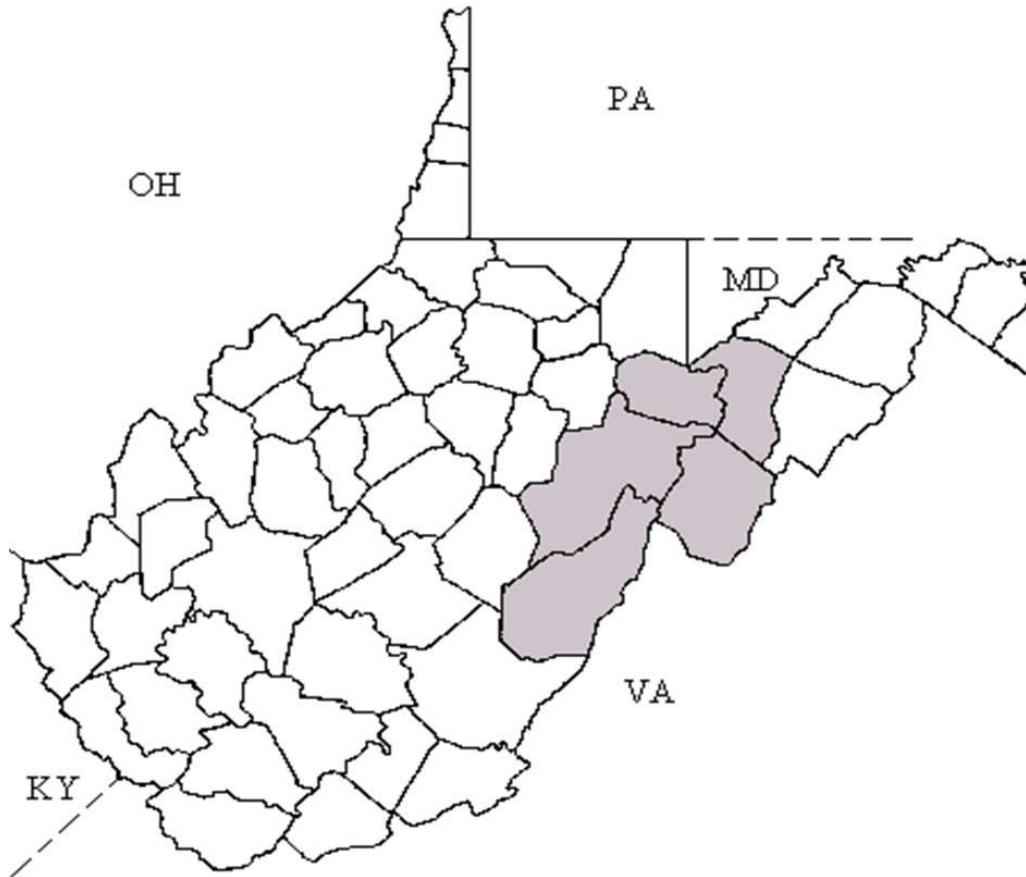


Figure 2: Total range of *Plethodon nettingi*, includes Tucker, Grant, Pocahontas, Randolph and Pendleton counties of West Virginia (Map by T.K.Pauley).

Allegheny Mountain Dusky Salamanders (*Desmognathus ochrophaeus*)

Allegheny Mountain Dusky Salamanders (Figure 3) are small salamanders in the family Plethodontidae. They have a broad dorsal stripe extending from the head to tail which varies in color from yellow, to orange, red or shades of brown. As with most salamanders from the genus *Desmognathus*, they have a pale stripe that extends from the posterior margin of the eyes to the mouth and the back legs are almost twice the size as the front legs. They normally have chevron-shaped spots that appear within the stripe. The belly is usually pale and sometimes lightly mottled (Hulse et al., 2001). Average length for mature individuals is around 38.2mm snout-vent length (SVL) and 76.7mm total length (TL) (Hulse et al., 2001).

Of all the salamanders in the genus *Desmognathus*, Allegheny Mountain Dusky Salamanders are the most terrestrial (Green and Pauley, 1987). They are normally found under cover objects such as logs, bark and rocks in both moist mixed deciduous and coniferous forests. As with most salamanders, they are usually active during wet or humid nights. They eat primarily small invertebrates (Hulse et al., 2001).

In West Virginia, *D. ochrophaeus* are found in the mountainous regions of the state, in Monongalia, Preston, Randolph, Monroe, Mercer and McDowell counties (Green and Pauley, 1987).



Figure 3: Adult *Desmognathus ochrophaeus*.

Red-backed Salamander (*Plethodon cinereus*)

Red-backed Salamanders (Figure 4) are one of the most common salamanders in West Virginia (Pauley, unpublished data). They are small, slender salamanders that normally display a red stripe that extends the length of the body and most of the tail. Red-backed salamanders live primarily in cool, moist mixed deciduous and coniferous forests in areas with sufficient moisture and cover objects. They can be found under leaf litter, fallen trees, logs and rocks (Hulse et al., 2001; Green and Pauley, 1987).

Plethodon cinereus remain under cover objects during the day and emerge at night to forage for food (Green and Pauley, 1987). They are important to the structure of the forests because they can reach high population densities and predate on small soil and leaf litter invertebrates that other predators do not consume (Hulse et al., 2001).



Figure 4: Adult *Plethodon cinereus*.

Wehrle's Salamander (*Plethodon wehrlei*)

Wehrle's Salamanders (Figure 5) are a moderately sized salamander with a brown to black dorsal color with the sides of the body covered in white spots that can extend all the way to head and jaws. Often this species can be distinguished by the presence of a small orange blotch just behind the shoulders (Hulse et al., 2001). The average length for adults is 66mm SVL. They are most often found in upland forests with dense canopy cover, whether it is mixed deciduous, mixed hardwood-conifer or conifer forests. In West Virginia, they are found in high elevations of the Allegheny Mountains in primarily Red Spruce forests mixed with yellow birch, and lower elevations of the Plateau in mixed deciduous forest (Green and Pauley, 1987). As with most salamanders, they can normally be found under cover objects during the day such as rocks and occasionally logs, bark and leaf litter, emerging at dusk to forage (Green and Pauley, 1987, Hulse et al., 2001). They seem to eat mostly ants, beetles and insect larvae (Green and Pauley, 1987).



Figure 5: Adult *Plethodon wehrlei*.

Northern Slimy Salamander (*Plethodon glutinosus glutinosus*)

Northern Slimy Salamanders (Figure 6) are a large *Plethodon* that have a black base color on the back and sides with white spots. The degree of spotting varies among individuals (Hulse et al., 2001). Females are larger than males, with an average of 73.5mm SVL and 150.3mm TL. Males are normally 68.6mm SVL and 136.4mm TL. This species is found primarily in mature woodland forests, moist ravines, shale banks, flood plains, and cave entrances where cover objects such as rocks, logs and vegetation are abundant. In West Virginia, this species is statewide (Green and Pauley, 1987). These salamanders feed on prey items such as millipedes, centipedes, worms, beetles, ants and insect larvae (Hulse et al., 2001).



Figure 6: Adult *Plethodon glutinosus glutinosus*.

Snakes found throughout my study site belong to the family Colubridae (harmless snakes) which includes around 270 genera and over 2,000 species, making it the largest family of snakes and representing three- fourths of known snake species (Hulse et al., 2001; Green and Pauley, 1987). Colubrids can inhabit all terrestrial and aquatic habitats within their geographic range. Snakes are always carnivores and eat their prey whole. Colubrids vary in dietary preference from generalized or opportunistic feeders to having extremely specialized diets (Hulse et al., 2001; Green and Pauley, 1987).

Eastern Gartersnake (*Thamnophis sirtalis sirtalis*)

Two subspecies of gartersnake are present in the Northeastern United States, the Eastern Gartersnake (*Thamnophis sirtalis sirtalis*) and the Maritime Gartersnake (*Thamnophis sirtalis pallidulus*). In West Virginia, the Eastern Gartersnake (*Thamnophis sirtalis sirtalis*) is present (Figure 7). Eastern Garter Snakes are heavy-bodied snakes with females reaching an averaging 439mm SVL, 550.3 TL and males averaging 339.6mm SVL and 444.5 TL (Hulse et al., 2001). There are a wide variety of markings and colors on these snakes. Most commonly a yellow or green medial stripe and two lateral stripes are present with a dorsal color ranging from dark green to brown or black. When stripes are present, dark spots are normally located between them. Ventral color usually varies between yellow and green with a lateral row of spots which may be difficult to see (Hulse et al., 2001). Scales are keeled.

This is not only the most common snake encountered in West Virginia (Green and Pauley, 1987), but it is also the most common snake in the Northeastern United States that can live in a variety of habitats. They are habitat generalists and are found in areas from open fields, deciduous and coniferous forests with both open and closed canopy, marshes, bogs, swamps to very dry upland forests (Hulse et al., 2001).

Eastern Gartersnakes have been found active every month out of the year in Pennsylvania (Hulse et al., 2001). Because my study sites were located in high elevations of West Virginia, it is likely

to correspond to environmental conditions of northern latitudes (i.e., Pennsylvania). Activity varies with each season; during the spring and fall, they are mostly diurnal and active during the middle of the day. During the hot parts of the summer, these snakes are active primarily in the morning or late afternoon/evening. They may even become nocturnal during the summer (Hulse et al., 2001).

Gartersnake movement seems vary depending on environmental conditions, abundance of food and areas for hibernation (Ernst and Barbour, 1989). Carpenter (1952) found they can travel at a speed of 0.75 – 2.00 miles per hour. Using mark recapture, a male gartersnake was found to move 1800ft in one hour (Blanchard and Finster, 1933). Carpenter (1952) found Eastern Gartersnakes moving up to 300 m, with average distance moved under 183 m. He also determined home ranges for this species is approximately 183 x 48 m.

Not only are gartersnakes habitat generalists, they also have a generalized diet eating both invertebrates and vertebrates. Prey items include “cold-blooded” organisms such as frogs, toads, salamanders, fish, earthworms and even some insects (Green and Pauley, 1987). Studies have indicated that within a certain geographic range, their diets become more specialized. A study done in the George Washington National Forest found that salamanders alone constituted 31.37% of an average meal, while other amphibians (toads especially) made up 25.01% of their diet (Uhler et al., 1939).

Some gartersnakes may show an aggressive behavior (such as flattening head and body, exposing teeth and striking repeatedly) when handled for periods of time, but this vary between individuals (Hulse et al., 2001).

Eastern garter snakes are viviparous and litter size varies according to female body size. Their range extends from northwester Quebec and central Ontario to the Florida keys and eastern Texas (Hulse et al., 2001).



Figure 7: Adult *Thamnophis sirtalis sirtalis*.

Northern Red-bellied Snake (*Storeria occipitomaculata occipitomaculata*)

Northern Red-bellied Snakes (Figure 8) are small species that show extreme variations in color. The dorsum can be tan to dark brown, reddish brown, gray or black with shades of brown being the most common. Many specimens exhibit a midvertebral stripe that is usually lighter than the dorsal color. A distinguishing characteristic are the three light spots on the back of the neck. As the name suggests, the ventral color of this species is most often a shade of red, however there are many variations to this, including shades of yellow, pink (Hulse et al., 2001) and even black. They have keeled scales.

Habitat includes areas where there is deciduous hardwood forests and open canopy near the forest (Blanchard, 1937; Hulse et al., 2001). They can be found in old fields, meadows, sedge habitats, borders of swamps and other open places (Palmer and Braswell, 1995; Green and Pauley, 1987). In West Virginia, it is one of the few species that can be observed in higher elevations in mountainous terrain (Green and Pauley, 1987). They are mostly nocturnal and secretive, rarely found basking or moving in the open. They are most often found under logs, rocks, bark or other natural cover (Palmer and Braswell, 1995; Hulse et al., 2001, Green and Pauley, 1987). Many times two or more of these snakes can be found under the same cover object (especially gravid females) and it is not known whether they are attracted to each other or the microhabitat conditions under specific cover (Hulse et al., 2001).

Snake movement in general has been understudied in literature. Movement in these snakes was discussed by Blanchard (1937), where 150 snakes captured, marked and released had not been recovered in later years, compared to other local species. However, a large female released was recovered a day later almost 1207meters away from where she was released (Blanchard, 1937). A few other individuals were recaptured within a week of their release.

In the Northeast, diet consists of mostly slugs and earthworms (Palmer and Braswell, 1995; Hulse et al., 2001; Green and Pauley, 1987). They have also been known to eat snails by extracting them from their shell (Hulse et al., 2001). Northern Red-bellies are inoffensive and rarely bite, however they emit musk and are known to curl the upper lip in a sneer, exposing teeth (Hulse et al., 2001; Palmer and Braswell, 1995; Green and Pauley, 1987).

Northern Red-bellied Snakes show sexual dimorphism with adult females averaging 245.9mm SVL and 308.5 TL, while males average 213.1mm SVL and 283.3mm TL. As in most snakes, males have a significantly longer tail than females. Females are viviparous and have young from late July through the early parts of September (Hulse et al., 2001; Green and Pauley, 1987).

This species range extends from Maine and the Maritime Provinces of Canada west to Manitoba and eastern North Dakota. The range moves southward to Oklahoma and Arkansas moving east to South Carolina and Georgia. It is found throughout the Northeast (Hulse et al., 2001) and probably throughout the state in West Virginia.



Figure 8: Gravid adult female *Storeria occipitomaculata occipitomaculata*

Northern Ring-necked Snake (*Diadophis punctatus edwardsii*)

Northern Ring-necked Snakes (Figure 9) are a medium-sized snake with a consistent gray, blueish-gray or brown dorsal color without any pattern. Around the neck, there is a yellow to orange colored ring that is bordered on either side with a black band. The ventral is variable from pale yellow to orange and occasionally a single row of black spots are present along the posterior section of the body (Hulse et al., 2001).

The entire range is from Nova Scotia and Newfoundland west to Wisconsin and south to Alabama and northern parts of Georgia (Hulse et al., 2001). These snakes are extremely common in West Virginia (Green and Pauley, 1987). Habitats vary from primary and secondary deciduous forests, old fields, rocky hilltops, grassy fields and borders of streams and rivers (Hulse et al., 2001; Green and Pauley, 1987). Normally found beneath flat rocks in forested areas (Palmer and Braswell, 1995) but can be found under logs, bark or man-made cover objects. They are secretive species and are rarely found in the open (Blanchard et al., 1979; Hulse et al., 2001). When found under cover objects, they are often coiled and not in motion (Blanchard et al., 1979). Studies have suggested the two main habitat requirements for this species include open areas with direct sunlight can warm the surface and a large quantity of rocks. Ring-necked snakes are thought to be nocturnal (Hulse et al., 2001), with greatest activity and movement during twilight or night (Blanchard et al., 1979). Blanchard and colleagues (1979) found one ring-necked snake actively crawling by flashlight, which was the only snake of this species he found to be out from under cover.

Many times more than one ring-necked snake can be found under the same cover object. Nine ring-necked snakes were discovered under the same board (Blanchard et al., 1979). Dundee and Miller (1968) found that these snakes tend to pick cover objects based on soil water content (not too dry but not soaking wet) and snakes will continually use the same by locating it based on olfactory cues. Studies

show they chose areas with protection from too much heat, light and desiccation (Blanchard et al., 1979).

Across their range, Northern Ring-necked snakes have been known to eat a variety of food including not only salamanders, but also other snakes, lizards, frogs, earthworms, slugs and insects (Hulse et al., 2001). However, amphibians probably constitute their primary food source. Brown (1979) found only salamanders had been eaten by 8 individuals, including *Plethodon cinereus* and *Plethodon glutinosus* (both which occur at my study sites). *Desmognathus fuscus* and *Eurycea cirrigera* have also been found in the guts of these animals (Palmer and Braswell, 1995). In Pennsylvania, *Plethodon* salamanders seem to be their primary food source. It has been observed that where salamanders are abundant, they are the main food item. Blanchard's study (1979) on captive snakes showed that they responded positively when fed *P. cinereus*, which was abundant in the woods of my study site. Even juvenile snakes preferred *P. cinereus* when given a choice of smaller prey. In this study, prey items (i.e. frogs, toads, Jefferson's salamanders, or insects) were offered to snakes, and they would examine prey for a half hour; then *P. cinereus* would be added and the snakes would quickly attack and eat it (Blanchard et al., 1979). In the George Washington National Forest, studies have shown that 80% of their diet constituted salamanders (Uhler et al., 1939). In areas where there are no salamanders, earthworms seem to be the primary food source (Hulse et al., 2001).

Females lay 2 to 10 eggs under cover objects (such as rocks, logs etc) from late May until early June in West Virginia (Hulse et al., 2001; Green and Pauley, 1987). At 210mm SVL, males are considered sexually mature and females near 230mm (Hulse et al., 2001).

Movement and foraging behavior in ring-necked snakes is understudied. Although Blanchard (1979) noted that snakes were never found at two of his site locations in the same day, a distance of 60 feet was traveled on consecutive days. Along with that, a female moved 300 ft in 17 days. A few weeks

later the same snake was found 400ft from the previous location (Blanchard et al., 1979). Overall, 10 snakes were recorded to have moved nearly 400ft within a few days, although none of these snakes were seen actively moving from location to location (Blanchard et al., 1979).



Figure 9: Adult *Diadophis punctatus edwardsii*.

Smooth Greensnake (*Opheodrys vernalis*)

Smooth Greensnakes (Figure 10) are long slender species of snake with a solid brilliant green dorsum and yellow to white venter (Hulse et al., 2001). As their name suggests, they exhibit smooth scales rather than keeled.

This species is terrestrial and found in a wide variety of habitats, including old fields, pastures and other agricultural areas and in fragmented sections in forests. They are normally found under cover objects such as rocks and logs (Hulse et al., 2001). This species is mostly diurnal with a diet primarily consisting of insects such as crickets, grasshoppers, caterpillars and ground dwelling spiders (Hulse et al., 2001).

These snakes exhibit sexual dimorphism with males averaging a length of 269.6mm SVL and 414.8mm TL, while females average 323.8mm SVL and 450.3mm TL. Males have longer tails than females (Hulse et al., 2001). Females will lay 5 - 11 eggs under rocks exposed to sunlight in late June to early July and hatch in early August to September (Green and Pauley, 1987; Hulse et al., 2001).

Populations of these snakes appear to be declining, perhaps due to pesticide use on their prey items. The total range of this species extends from eastern Nova Scotia to Saskatchewan, south through New England into Pennsylvania and West Virginia and into northern parts of Virginia. Along with that, it occurs in northeastern Ohio, Michigan, Illinois and the majority of Wisconsin. Through the plains and Texas it has a disjunct distribution (Hulse et al., 2001). In West Virginia, these snakes are more common in the mountains and can be found with species such as *S. o. occipitmaculata* or *T. s. sirtalis* (Green and Pauley, 1987).



Figure 10: Hatchling *Opheodrys vernalis*.

METHODS

Site Selection:

Sites were chosen based on the presence of *P. nettingi* as well as accessibility. I wanted to choose 3 sites that could be surveyed around the same area. Then I chose sites that had a high degree of fragmentation, leaving a forest edge somewhat exposed to the sun with transects deeper into the forest with high percentage of canopy cover. Three sites were chosen to ensure replication and proper sample sizes. All 3 sites are located in Tucker County, West Virginia (Figures 11 and 12).

Study Area

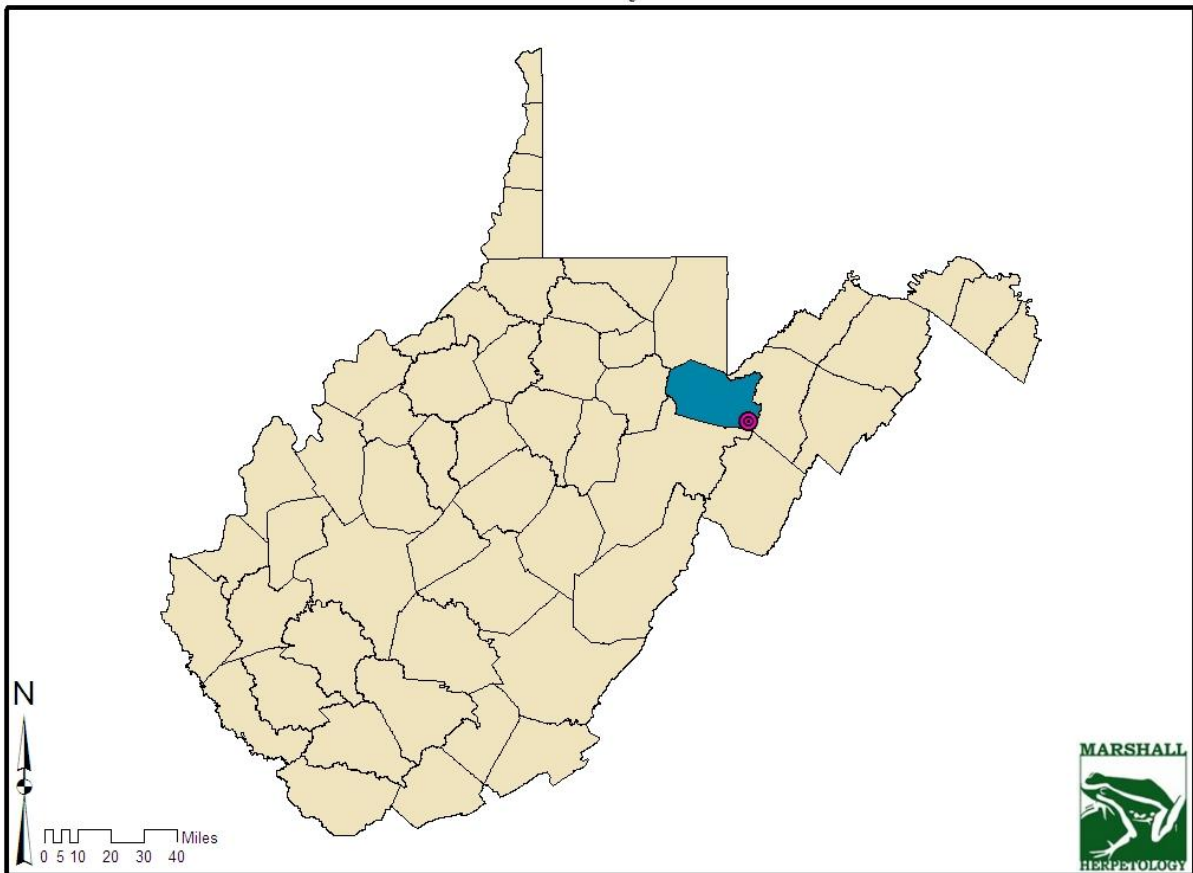


Figure 11: Map of West Virginia with Tucker County shaded blue and site location marked in pink.

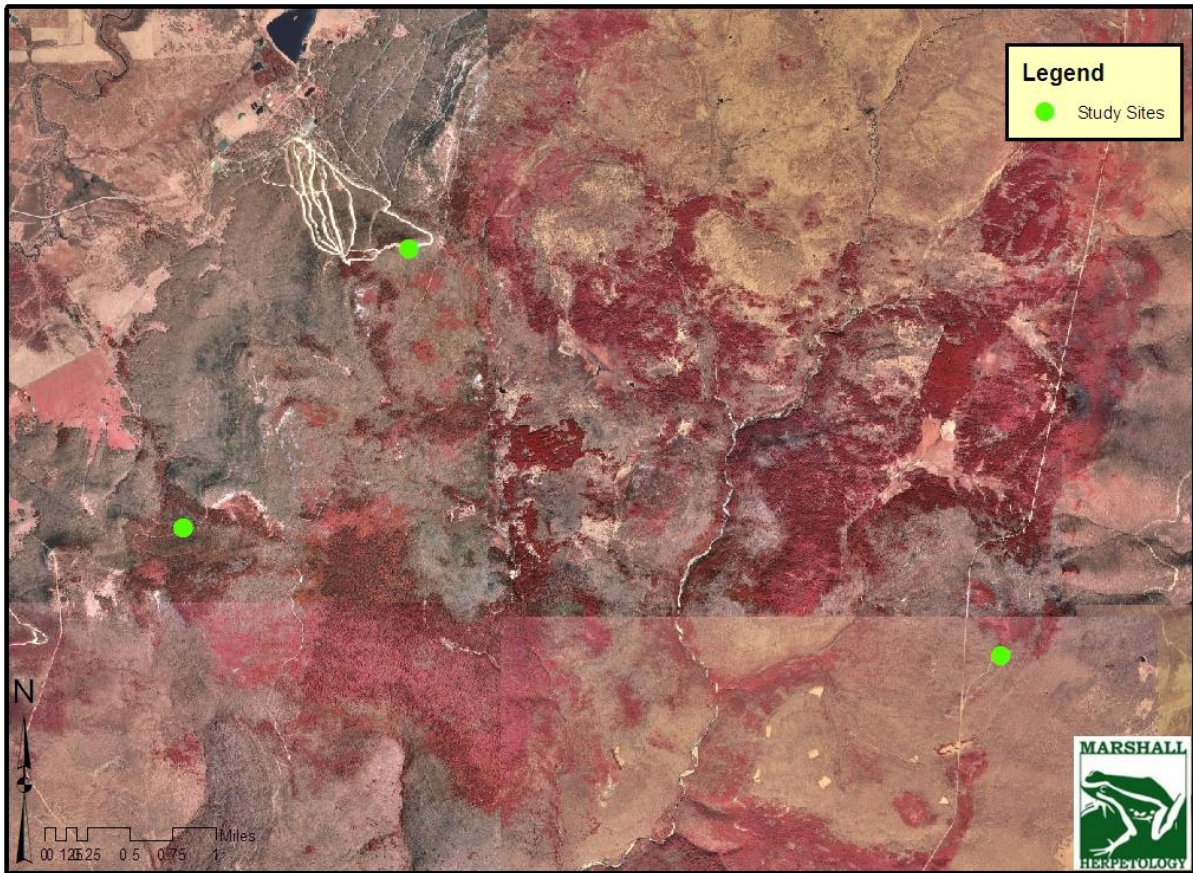


Figure 12: Aerial view of site locations, which are marked in green. Timberline is top, Whitegrass is left and Dolly Sods is lower right.

Experimental Design:

Six transects were set up at each site. Transects were 183 meters long by 1.5 meters wide. Transect length was first determined to encapsulate the majority of *P. nettingi* habitat and width to ensure the possibility of accurately surveying sites in a timely manner without overlooking the presence of study species and to eliminate habitat disturbance. Three transects were placed parallel, on either side of the forest fragmentation. Transect A was placed along the forest edge, Transect B was placed 8 meters from the forest edge, and Transect C was placed 17 meters from the edge of the forest (Figure 13).

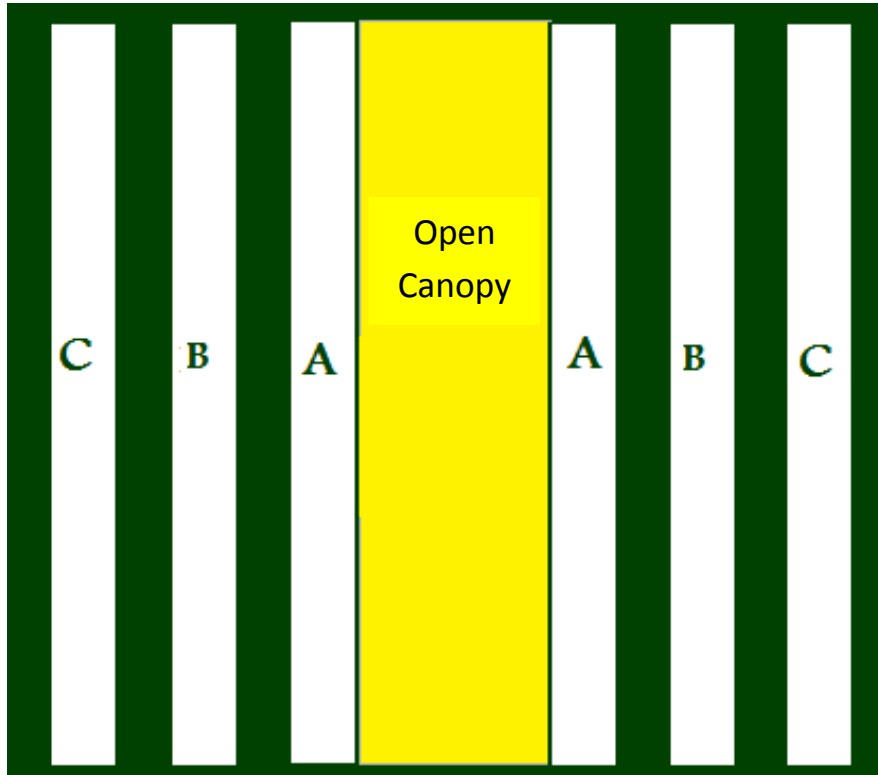


Figure 13: Diagram of experimental design of project. Yellow indicates open canopy with Transects A, B and C on both sides.

Studying amphibians and reptiles requires searching under natural or man-made cover objects, which can disturb and destroy habitat (Engelstoft and Ovaska, 2000), therefore limited my site surveys to twice a month to reduce both destruction of snake and salamander habitats. Along each transect, rocks, logs and coverboards were flipped over and returned to their original position during each survey. Rocks and logs flipped were counted and categorized according to size to eliminate bias between the amounts of available cover objects at each site. Coverboards were also flipped over at sites where they were being used for other *P. nettingi* studies (Timberline and Dolly Sods) and were put into the “medium” size category for cover object analysis. Artificial cover has been used to survey both reptiles (Engelstoft and Ovaska, 2000) and amphibians. Artificial cover objects were not specifically used in this study, however, to help the possible prevention of snakes in the area preying upon salamanders that may have used the same cover object as refuge. It is important to note two of my sites did have coverboards present which were being used for another study, and these boards were flipped when they appeared in transects.

Vegetation analysis was conducted at each site to determine dominant tree species and their densities, frequencies, basal area (BA), relative abundance and importance values. Basal area was calculated for each species at each site ($BA \text{ (m}^2 \text{ ha}^{-1}) = DBH \text{ (cm)}^2 * 0.00007854$). Species frequency was determined by counting the number of plots each species is found, divided by the total number of plots. Species density was calculated by counting the number of trees per species, divided by the total area surveyed. Relative BA is the sum of the basal area for each tree species divided by the total area surveyed in hectares. Relative frequency and density was calculated using the same equation as relative BA, using the sum of specie frequencies and densities. Importance values give a relative contribution of a species in a community and are the summation of relative density, relative frequency and relative basal area (Stohlgren, 2007). Trees in this study were ranked based on importance values to give the dominant species in the study transects (Stohlgren, 2007). Three plots, 4.5 meters x 1.5 meters were

set on each transect at the lower, middle and upper points on Transects A and C. Transect B had 3 plots set up as well, but they were staggered between the lower/middle section, middle, and upper/middle section to eliminate a bias between trees perpendicular to transects. DBH (diameter at breast height) and tree species was recorded for each tree that was >4cm DBH in the plot area.

Environmental data were recorded along each transect. Air temperature (AT) at the surface, soil temperature (ST), and relative humidity (RH) were taken at three locations along each transect. Two instruments were used to take the measurements, and the resulting values were averaged.

Data Collection:

Salamanders found in each transect were handled as little as possible to prevent unnecessary stress to the animal. Each potential capture was identified to species and reproductive status was noted if males showed a swollen cloaca or square chin, and if female salamanders were gravid.

Snakes were identified to species and measured for snout to vent length (SVL) and total length (TL) using a Stanley tape measure, or 30.5cm ruler if snakes were small enough. Gender was determined via probing the first two months in the field until I felt comfortable sexing them visually. Snakes can be sexed based on the thickness of the tail directly after the cloaca. Female's tails are as wide as the body immediately posterior to the anal scale, but after the second or third subcaudal scale they taper off very quickly. Male's tails are just as wide, if not wider than the body due to the presence of hemipenes and associated muscles. If there was any confusion about gender, snakes were probed. This involves inserting a tiny probe into the cloaca toward the caudal end of the snake. If the snake is female, the probe can only be inserted 2-3 subcaudals. Snakes were given a unique number using a standard numbering system and marked with a medical cauterizing unit on the ventral scale (Figure 14 and 15).



Figure 14: Medical cautery units used to heat-brand snakes. Different size units have different temperature ranges. The cauterizer used for this study had high temperature range and used 2 AA batteries (Winne et al., 2006).

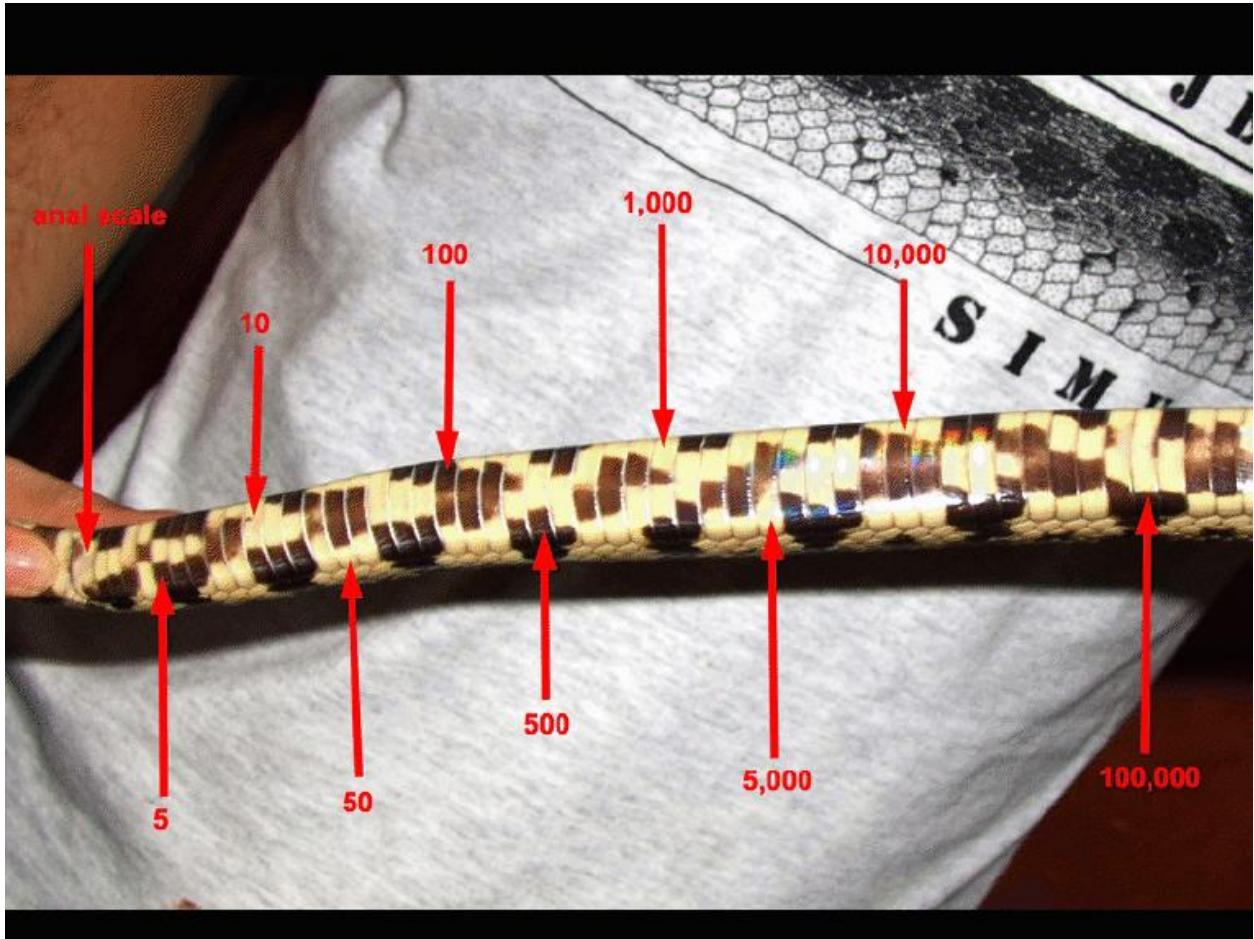


Figure 15 : Photograph representing the numbering system used to marked snakes. Numbering begins by counting the anal scale as 0, moving towards the head of the snake counting each scale. At scale 10, count by 10's until you reach 100. At scale 100, to count by hundreds, etc... This numbering system allows for tens of thousands of individuals in a single species to be marked without repeating a number.

Photograph and diagram by Kevin Messinger

Mark-recapture data can give abundance estimates over a period of time as well as survival rates (Lind et al., 2005). Although I am not extending my study into the next field season, marking each snake leaves the opportunity to continue this study or for other experiments to be conducted in the future. Studies have shown that heat-branding can last as long as 1058 days over a variety of species and snake recapture numbers can still be observed even after rapid growth in both length and weight (Winne et al., 2006). Additionally, cauterization is a quick and sterile method, it is effective for ongoing mark-recapture studies on snakes, it is field portable and reliable (Winne et al., 2006).

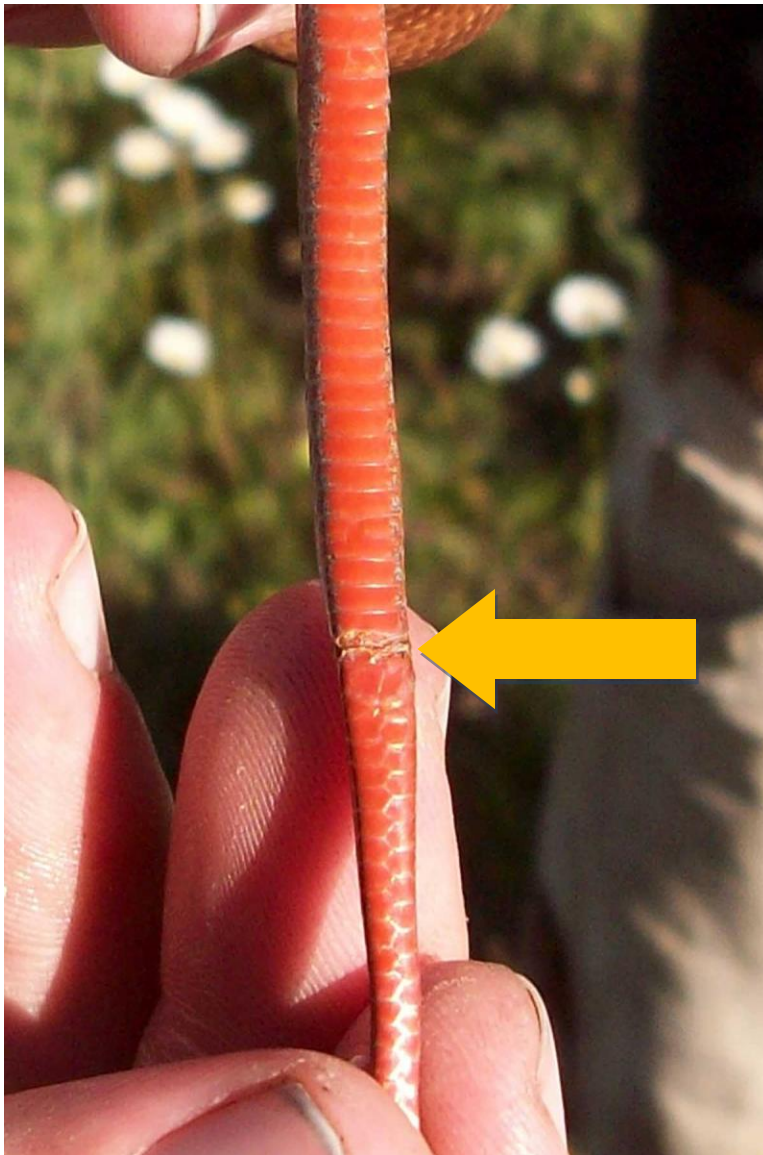


Figure 16: Ventral side of *S. o. occipitamaculata* after cauterization.

Data Analyses

A Pearson correlation analysis was conducted using SPSS (Statistical Package for Social Sciences) for air temperature, soil temperature, and relative humidity to determine if the environmental variables were correlated. Only values of >0.90 were considered significantly correlated when interpreting the results. ANOVA (analysis of variance) was used to analyze environmental variables for variance within and between transect means. We used $\alpha=0.05$ and p-values of less than 0.05 were considered significant. Furthermore a Tukey test was used to indicate significance among the statistically significant variables from the ANOVA results.

ANCOVA (Analysis of covariance) was used to analyze the significance of cover objects along each transect using cover objects as the covariate, snakes salamanders and total captures as the dependent variable with fixed variables as my transects.

SITE DESCRIPTIONS

The first study site was at Timberline Four Seasons Resort on a downhill ski slope called Salamander Run ($39^{\circ}01'59.67''N$, $79^{\circ}23'13.69''W$). This location was once a continuous forest dominated by Red Spruce and mixed deciduous trees. In 1986, the resort fragmented the forest by establishing many downhill ski trails. The ski slope is approximately 30 meter wide with mixed grass species and direct sunlight. Along the forest edge at Site 2, rocks and logs are present. At site 1 there are few cover objects for daytime refugia, a lower percentage of Red Spruce and fewer emergent rocks. During the summer months, the ski trail is used for a variety of outdoor activities including horseback riding, hiking, running and biking competitions (Figure 17).



Figure 17: Photograph of Salamander Run ski slope at Timberline Four Season Resort. Transects were located on both sides of the ski slope.

Photo by Casey Bradshaw

The second site was on property owned by the USFWS where Whitegrass maintains the trails during the winter for cross-country skiing. My study sites were located on Three-Mile and Powderline Ski Trials on the Canaan Valley National Wildlife Refuge which are approximately 1 mile from the nearest road (39°00'35.79"N, 79°24'43.29"W). These trails have been used as cross country ski trails for nearly 30 years. During the spring and fall, regular maintenance is conducted which includes removing brush and fallen trees and trimming vegetation to ensure the ski trails remain close to the same width. Water drainage is maintained by waterbars, which are cleaned at least once a year. In the fall, small wooden "bridges" are placed over some of the larger drainages so people can ski over them. In 1999, the WMA took over this land and since then no additional seeding of the trails has taken place. Prior to WMA use, many of the ski trails were used as logging and skid trails from previous logging operations (Pers.Com.: Ken Sturm).

During the summer at Whitegrass, the public is not allowed to enter the trails. Wildlife biologists and field technicians from Refuge conduct wildlife assessments, but other than that only those the Refuge has issued special permits are allowed to enter. A known population of *P. nettingi* are known to occur southeast of transects at Site 1 with an elevation of 1249m, which is around 30.5m higher than the study transects.

Even during the hottest parts of the summer, this trail still remained damp and had almost complete canopy cover over the ski trails. The habitat at my site was primarily a Red Spruce and mixed deciduous forest (Figure 18).



Figure 18: Photograph of Powderline cross country ski trail at Whitegrass. Transects were set up on either side of the ski trail.

Photo by Casey Bradshaw

The last site was located at Bell Knob at Dolly Sods in the Monongahela National Forest on the border of Tucker and Grant counties (39°00'00.35"N, 79°19'25.84"W).

This area was at a Forest Service gated road, therefore a locked gate was at the entrance of the road so public vehicles were not allowed through (Figure 19).

At one time, Dolly Sods contained some of the greatest stands of Red Spruce in the world (West Virginia Conservation Commission, 1908). At the end of the gated road, Bell Knob Fire Tower still stands which was used as a lookout for wildfires during the 1930's. The first fire to sweep through that area is thought to have occurred in 1863, and a more devastating fire went through in 1930 which burned for 6 months (Turner, 2001). This fire destroyed over 24,000 acres of old growth forest and has been repopulated primarily by Red Maple, American Beech, Rhododendron and patches of blueberries (Turner, 2001).



Figure 19: Photograph of Bell Knob gated road at Dolly Sods after all the leaves had fallen. Transects were set up on either side of this road, which fragments the forest.

Photograph by Kevin Messinger

Results

Between 9 June 2009 and 23 October 2009, 287 salamanders and 99 snakes were observed at the 3 study sites.

Timberline Site 1

Vegetation analysis shows, American Beech (*Fagus grandifolia*) had the highest importance values (198.42) at both sites, followed by Black Cherry (*Prunus serotina*) and Red Maple (*Acer rubrum*). Red Spruce (*Picea rubens*) had the lowest relative frequency, density and basal area, therefore the lowest importance value (Table 1).

There was no statistical correlation between air temperature, soil temperature and relative humidity (>0.90) among all transects in the study (Table 7). When considering only Timberline Transects there was no significance difference when analyzing the environmental factors (Table 18). Along with that, Transect A had the highest mean air temperature (20.6C) and mean soil temperature (17.5C), although differences between transects is not statistically significant (Table 10).

Nineteen salamanders were caught during the field season, the highest number were caught in June (n=9). No snakes were found at this site throughout the entire field season. Only 1 *P. nettingi* was found in any transects and 1 *D. ochrophaeus* was found in all 3 transects during the entire field season. Compared with Timberline Site 2, the relative humidity was slightly lower (Table 10). Figure 23 shows both Timberline site 1 and 2 with salamander and snake captures from the entire field season using spatial reference. This site was surveyed 9 times throughout the field season, with a capture rate of 2.5 salamanders per survey.

A positive correlation is found between number of snake captures and number of cover objects (Figure 33; $R^2 = 0.70$ and Figure 32; $R^2 = 0.17$). This site had 97 cover objects. Broken down, Transect A

had 16 cover objects, transect B had 37 cover objects and Transect C had 44, the majority of those were logs.

Timberline Site 2

Male to female ratios at Timberline were 1:2 for *D.p. edwardsii*, 1:0.6 for *S. occipitomaculata* and a ratio is not given for *T. s. sirtalis* because no females were captured.

Vegetation analysis shows, American Beech had the highest importance value of 95.7 (Table 2). Results show no correlation between air temperature, soil temperature and relative humidity (>0.90) (Table 7). Transect A, again, had the highest mean air temperature of the 3 transects (22.9C), highest mean soil temperature (19.4C) and relative humidity (60.5%) although differences between transects are not statistically significant (Table 11). The highest number of salamanders were caught in June (n=26) (Figure 21) and the highest number of snakes in October (n=9) (Figure 22). This site was surveyed 10 times throughout the field season with a capture rate of 0.38 snakes and 10.1 salamanders per survey.

A positive correlation is found between number of captures and number of cover objects (Figure 33; $R^2 = 0.70$). This site had 242 cover objects. Broken down, Transect A had 92 cover objects, 86 of those were rocks and this is the only transect where snakes were found. Transect B had 59 cover objects and Transect C had 91, the majority of those were coverboards and logs.

A spatial representation of species of snakes and salamanders observed is shown in Figure 23. Individual locations of each animal are not 100% accurate given the error on GPS unit and scale of the map. Due to accuracy of GPS and scale of map points were adjusted to better display spatial locality of salamander and snake locations.

Dolly Sods Site 1

Yellow Birch (*Betula alleghaniensis*) had highest importance value of 63.30 followed by White Oak (*Quercus alba*) and Mountain Holly (*Ilex montana*) (Table 3). Results show a correlation between air temperature and soil temperature (Table 8). Transect A had the highest mean value for air temperature (20.2C), soil temperature (17.5C) and relative humidity (61.8%). Although these values are not statistically significant, they do show a negative correlation between air temperature and soil temperature toward the interior of the forest (Table 12). When considering only Dolly Sods Transects there was no significance difference when analyzing the environmental factors (Table 19).

The highest number of salamanders was caught in October (n=18) and 31 salamanders were caught throughout the entire field season (Figure 26). The highest number of snakes was caught in August (n=24) and 47 snakes were caught throughout the entire field season (Figure 25). Sex ratios for snakes were 1.3:1 for *D. p. edwardsii* and 1:2 for *T.s.sirtalis*. Figure 28 shows both Dolly Sods site 1 and 2 with salamander and snake captures from the entire field season using spatial reference. This site was surveyed 9 times throughout the field season with a capture rate of 5.1 snakes and 3.6 salamanders per survey.

This site had 304 cover objects, most of them rocks. Broken down, Transect A had 209 cover objects, Transect B had 34 and Transect C had 61.

Dolly Sods Site 2

Vegetation analysis show Red Maple had the highest importance value of 92.11, followed by Mountain Holly and Yellow Birch (Table 4). There was a correlation between air temperature and soil temperature (Table 8). Transect A had the highest mean value for air temperature (19.4C) and soil temperature (16.6C). Average relative humidity was the same in both Transect A and C (65.2%).

Although these values are not statistically significant, they do show a negative correlation in air temperature and soil temperature farther into the core of the forest (Table 13).

Seventy-one salamanders were caught over the field season, the highest number caught was in October (n=41) (Figure). Twenty-eight snakes were caught throughout the entire field season, the highest number caught in July (n=15) (Figure). The sex ratio of snakes was 2:1 for *D.p. edwardsii* and 1:0.8 for *T. s. sirtalis*. This site was surveyed 9 times throughout the field season with a capture rate of 8.1 snakes and 3 salamanders per survey.

Three hundred and fifty-three cover objects were found at this site, the majority were rocks. Broken down, Transect A had 207, Transect B had 61 and Transect C had 85 cover objects.

Whitegrass Site 1

Vegetation analysis shows Striped Maple (*Acer pensylvanicum*) had the highest importance value of 62.86 followed by Red Spruce and Red Maple (Table 5). There was a correlation between air temperature and soil temperature (Table 9). Transect A had the highest mean value for air temperature (18.1), soil temperature (16.9C) and relative humidity (70.5%). Although these values are not statistically significant they do show a negative correlation in air temperature and soil temperature farther into the core of the forest (Table 14). Although when considering only Whitegrass transects there was a significant difference among Relative humidity (p-value=.049) and Soil temperature (p-value=.033) between transects 1 and 3. The Tukey results showed the mean difference between transects 1 and 3 to be .800 degrees Celsius with a significance value of .045. When considering Soil temperature; the mean difference between transects 1 and 3 was 1.342 degrees Celsius with a significance value of .035. This site in general had 2.69C lower average air temperature than Timberline and Dolly Sods. It also had an 8% higher average relative humidity than the other 2 sites.

Thirty-five salamanders were caught throughout the entire field season, the highest number were caught in August (n=14) (Figure 29). This site was surveyed 7 times throughout the field season with a capture rate of 4.86 salamanders per survey. This site had fewer cover objects compared with Dolly Sods and Timberline with 97, most were logs. Broken down, Transect A had 34, Transect B had 29 and Transect C had 34 cover objects.

Whitegrass Site 2

Vegetation analysis shows Black Cherry had the highest importance values of 66.38, followed by Striped Maple and Red Spruce (Tables 6). There was a correlation between air temperature and soil temperature (Table 9). Transect A had the highest mean value for air temperature (18.4C), soil temperature (16.6C) and relative humidity (68%). Although these values are not statistically significant they do show a negative correlation in air temperature and soil temperature farther into the core of the forest (Table 15). This site in general had a 2.35C lower average air temperature than Timberline and Dolly Sods. It also had a 6% higher average relative humidity.

Twenty-eight salamanders were caught throughout the entire field season, the highest number were caught in September (n=11) (Figure 30). This site was surveyed 4 times throughout the field season with a capture rate of 7.25 salamanders per survey. No live snakes were found, although a Gartersnake shed skin was found under a rock in Transect A on September 5th. Figure 31 shows a spatial representation of all salamanders caught throughout the field season.

Seventy-six cover objects were found at this site. Broken down, Transect A had 38, Transect B had 15 and Transect C had 23 cover objects.

Overall Statistical Analysis

Snake recapture intervals were too small to run statistical analysis. ANOVA results between environmental factors showed that soil temperature is significantly different between transects (Table 16). Results from the Tukey test showed that soil temperature in Transects A are significantly different from Transects B and Transects C (Table 17).

DISCUSSION

Timberline Site 1:

Over the course of the field season, no snakes were observed at this location and 19 salamanders (Figure 23). This is probably due to lack of cover objects and thick cover of ferns at the eastern end of each transect. Only 10 rocks were found in Transect A, which could be the reason no snakes were found. With the exception of 2, all salamanders caught in these transects were *P. cinereus*, which can tolerate a wider variety of habitats (Adams et al., 2007; Pauley, 2008b). During June, 9 salamanders were caught which was the highest for the entire season. This is not surprising given that many plethodontid salamanders breed in spring and fall. During June in the high elevations, temperatures can still be cool and vegetation remains moist, increasing salamander activity.

This habitat differs from the north side of the slope in both number of cover objects, presence of the liverwort *Bazzania tribulata* and higher frequency of Red Spruce.

Timberline Site 2

Both snakes and salamanders were caught frequently at this site. The greatest number of *P. nettingi* was caught at this site and were present on all 3 transects. This site provides a dense forest compared to Site 1 with American Beech still the primary tree species. However, Red Spruce is more common on this side, as well as the amount of cover objects in each transect. This site included medium

sized (9 x 9cm – 38 x 38cm) wooden coverboards that are used for a 23-year long term study by Dr. Thomas K. Pauley. Hundreds of these boards are scattered throughout the woods with 11 present in Transect B and 30 in Transect C of my site. These boards have been on the ground for 2 years, and most are covered by leaf litter, and even soil, providing excellent refuge for salamanders. No snakes were found under any cover boards.

On October 23, 2009, 2 male *P. nettingi* were found south of Transect A on the eastern end of the transect, under a rock, at least 10 meters from the forest edge. Coincidentally, the highest number of snakes was caught on this same day. Although most salamanders were found in Transects B and C, deeper into the forest, finding salamanders on Transect A may indicate niche overlap between snakes and salamanders. It could also indicate that male salamanders are simply moving through the area. Studies have shown in *Plethodon* salamanders, that during dry periods, salamander movement decreases, which also decreases prey intake. After events of rain, however, salamanders move to the surface to forage for food (Fraser, 1979). This can cause an increase in competition between salamanders at this site, if in fact food is a limited resource. Space (i.e. cover object to take refuge under) may also be a limited resource (Fraser, 1979) which could be the reason male salamanders was found moving from the forest to the ski slope. Although it was a cool, rainy day when the 2 *P. nettingi* were found, the rock they were found under would have been in direct sunlight with virtually no other refuge around the rock.

The most frequent species of snake caught was *Storeria o. occipitmaculata* (Northern Red-bellied Snake). Males, females and gravid females were found throughout the summer; indicating a stable breeding population. Literature states *S. o. occipitmaculata* eat primarily slugs and earthworms, and I found no record of them eating salamanders. However, most salamanders observed along these transects were small, no more than 10 cm in total length (i.e. *P. nettingi* and *P. cinereus*) making them

easy prey items for even small snakes. *Thamnophis s. sirtalis* and *D. p. edwardsii* were also observed, but in lower numbers than *S.o. occipitamaculata*. Because of abundance of salamanders on these transects, they are probably a primary food item of *T. s. sirtalis* and *D. p. edwardsii*. Foraging distances for *D. p. edwardsii* (Blanchard et al., 1979) and *T. s. sirtalis* encompasses all 3 transects (Carpenter, 1952; Ernst and Barbour, 1989).

Dolly Sods Site 1

Both snakes and salamanders were caught frequently at this site. The forest was dense with Great Rhododendron (*Rhododendron maximum*) at the northeast portion of transects and where deciduous trees were more abundant in the south west section of transects. In areas where Rhododendron was dominant, cover objects were scarce and the vegetation was so thick little sunlight could penetrate the forest floor.

Dolly Sods site 1 and 2 had the highest number of cover objects. This site had medium sized cover boards in Transect B that were being used for a salamander study. Coverboards were only flipped over where they occurred in transects. *Plethodon nettingi* found at this site were only under coverboards. All snakes except 1 were found in Transect A. An active Eastern Gartersnake was found moving through Transect C on 17 August 2009 around 4:00pm. This was the only active snake found all season; all other snakes were found under rocks. Whether other snakes were missed actively foraging or they were active at different times of the day is unknown. *Diadophis punctatus edwardsii* are primarily nocturnal (Blanchard et al., 1979) and *T. s. sirtalis* are as well during the hot parts of the summer (Conant, 1938) so they may have been active when surveys were not occurring. However, Dolly Sods site 1 and 2 were being used for a *P. nettingi* study, and night surveys for salamanders were conducted for 1 week every month from June through August 2009. In a 30-year study done by Blanchard (1979) on *D. p. edwardsii*, he noted that twilight and night are probably the greatest times of

activity, although he only saw 1 snake away from cover and active at night. These snakes are secretive and difficult to find without active searches under cover objects.

The most common snake found at this site was *D.p.edwardsii*, followed by *T.s. sirtalis*, both of which constitute a primary food source of salamanders (Uhler, 1939, Green and Pauley, 1987; Hulse et al., 2001). Salamanders were found in all transects under cover, normally in Transect A when the weather was either raining or very damp with cloud cover.

Dolly Sods Site 2

This site had the highest number of snakes caught throughout the field season. Red Maple and Mountain Holly had the highest importance values and little Red Spruce was present. This site also had the highest number of cover objects which provided refuge for both snakes and salamanders. All snakes were found under rocks in Transect A except for a large male *D. p. edwardsii* which was found under a rock on October 23rd at the edge of Transect B in a break in the canopy Whitegrass Site 1 & 2.

Both site 1 and 2 had presence of Red Spruce, moist habitat, closed canopy and more trees/hectare than any other site. However, no *P. nettingi* were found throughout the entire field season and very few salamanders were found compared to other sites. A population of *P. nettingi* is known to be located further from the area of fragmentation and into the forest southeast of transects at Site 1, at an elevation 30.5 meters above the location of transects. The lack of cover objects and presence of fragmentation could have limited the distribution of *P. nettingi* throughout the study site.

It is important to note that these sites were surveyed less than both Timberline and Dolly Sods due to rain and thunderstorms.

Consideration of Cover Objects

Thermoregulation is probably a key factor (Blouin-Demers and Weatherhead, 2001) for snakes and in areas of high elevation where temperatures stay somewhat cool even during the summer months, and rocks provide excellent refuge. Results from this study do indicate the importance of cover objects. Snake observations, statistically, were significantly increased with the higher number of cover objects. Snake observations, statistically, were significantly increased with the higher number of cover objects. It is also important to note besides 2, all snakes were found on Transects A (forest edge) where most cover objects were rocks. Snakes found under rocks were normally curled up, inactive and found during the day. Studies suggest that gartersnakes take advantage of thermoregulatory opportunities and often use burrows and rocks to reach desired temperatures (Huey, 1987). A suitable rock to take refuge under depends largely on thickness; thin rocks would cause snakes to overheat during the day and become too cold at night and too large of rocks (>43cm) would inhibit snakes to ever reach preferred body temperatures (Huey, 1987). However, snakes under intermediate-thickness rocks can spend the longest amount of time in preferred ranges (Huey, 1987). Rocks can also help regulate body temperatures by avoiding snakes critical temperatures (too low and too high). Snakes on the surface exposed to full sun for periods of time would succumb to heat stress, while snakes remaining below critical body temperatures would have slowed locomotion, making them incapable of defending themselves. Snakes may also choose certain refugia depending on their state (i.e. fasting versus digesting) (Huey, 1987). Dolly Sods Site 1 and 2 provided many different sizes, thicknesses and positions of rocks that could meet snakes physiological needs. Timberline Site 2 also had an array of rock types that acted as retreat sites for the snakes. How snakes utilize rocks depending on their own optimal body temperatures could be one reason snakes were not always found under the same rocks, especially at different times of the day.

Total number of captures (snakes and salamanders) was also significantly related to the total amount of cover objects; however salamanders alone, statistically were not related to total cover.

Almost all salamanders found throughout the study were under refuge, normally rotting logs or cover boards that remained moist and damp further from the edge habitat (Transects B and C).

Conclusion

The main objectives of this study were to determine (1) relative abundance of snake communities; (2) how far species move from edge habitat into the forest; (3) and to determine whether snakes are a significant predatory threat to the federally protected Cheat Mountain Salamander (*Plethodon nettingi*).

Overall results from the study showed that snakes are primarily confined to Transects A, which lie directly at the forest edge. Salamander abundance was greatest in Transects B (8 m into the forest) and C (17 meter into the forest) at each study site. Studies suggest snakes occur more frequently at edge habitat because it is less costly to reach optimal body temperatures in open habitat versus shaded forest (Patrick and Gibbs, 2009). A study in Australia also showed that snakes may pick specific habitat based on prey abundance as well as optimal areas for thermoregulation (Madsen and Shine, 1996). In areas where snakes were found (Timberline Site 2, Dolly Sods Site 1 and 2), salamanders were abundant in one or more transects. Cover objects were also abundant providing refugia for snakes to reach optimal body temperatures and remain protected while thermoregulating, then potentially move farther into the forest to forage for food (i.e., salamanders).

Because this study was only done for one field season, conclusions on whether or not snakes are a significant predatory threat to *P. nettingi* must be interpreted with caution. Further studies on mark recapture should be continued as well as analysis on snakes to determine whether they are eating a diet of primarily salamanders, as literature suggests. Although surveys were conducted at night at Dolly Sods 1 week of every month during the summer and no snakes were found active, twilight or night time surveys should also be conducted at each site in addition to daytime surveys to better understand

movement and foraging preferences of snakes. However, results from this study showed that in fragmented forests where *P. nettingi* reside, 2 snake species that eat salamanders (i.e., *D. p. edwardsii* and *T. s. sirtalis*) are abundant where cover objects are available on the forest edge. Habitat fragmentation has no doubt increased populations of *T. s. sirtalis* throughout its entire range given that it can thrive on forest edge habitats as well as close contact with humans (Conant, 1938).

Transect distances from edge habitat to the inner sections of the forest should also be increased, due to the foraging ranges for the snakes found. Number of site replications should be increased for better data analyses and sites that have the similar habitat should be chosen, whereas in this study Whitegrass had a difference in degree of fragmentation and presence of *P. nettingi* compared with Timberline and Dolly Sods.

Plethodon nettingi is a species that needs to be considered carefully. How habitat fragmentation is affecting these animals is vital to understand why only 80 disjunct populations have been found (Pauley, 2008a). It is also important to consider past disturbances (i.e., forest fires) as a cause for differences between sites and *P. nettingi* populations. Species with small populations are vulnerable and at an increased risk of extinction (Primack, 2006). Fragmenting the remaining habitat for this species could cause an increased number of predators as well as environmental changes that could cause further population declines. Conservation plans thus far for *P. nettingi* proposed by Pauley (1991) recommend limiting construction of structures which fragment the forest at least 90m from known populations.

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APPENDIX

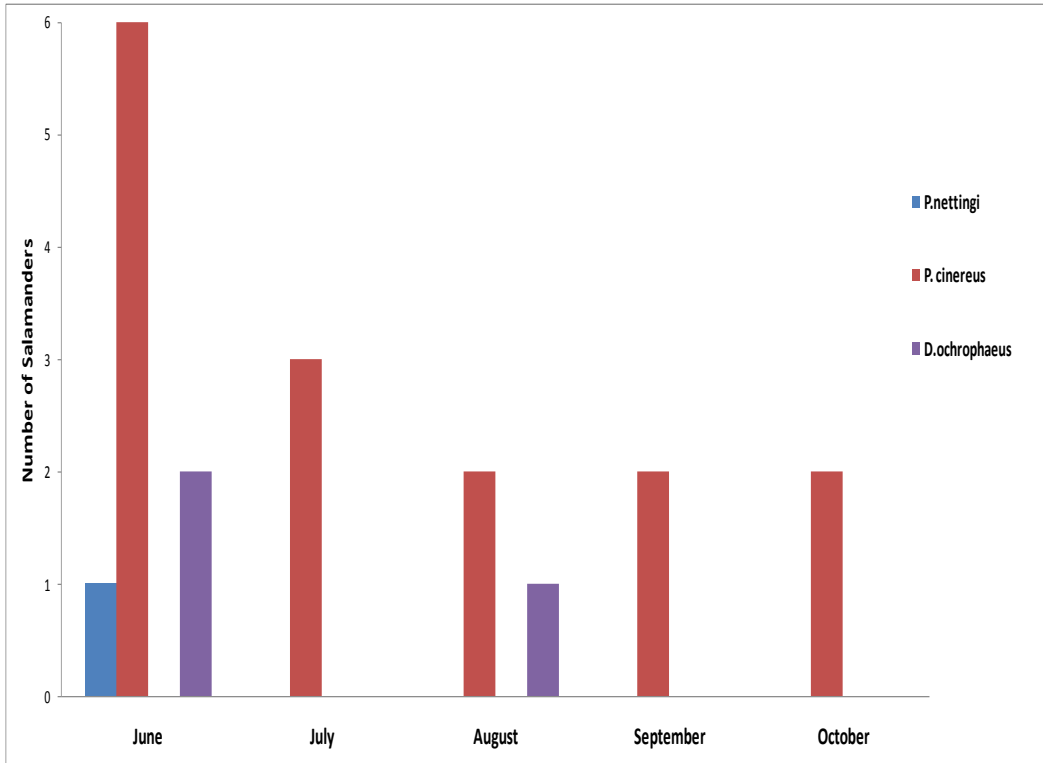


Figure20 : Number of salamanders caught monthly at Timberline Site 1 from June to October 2009

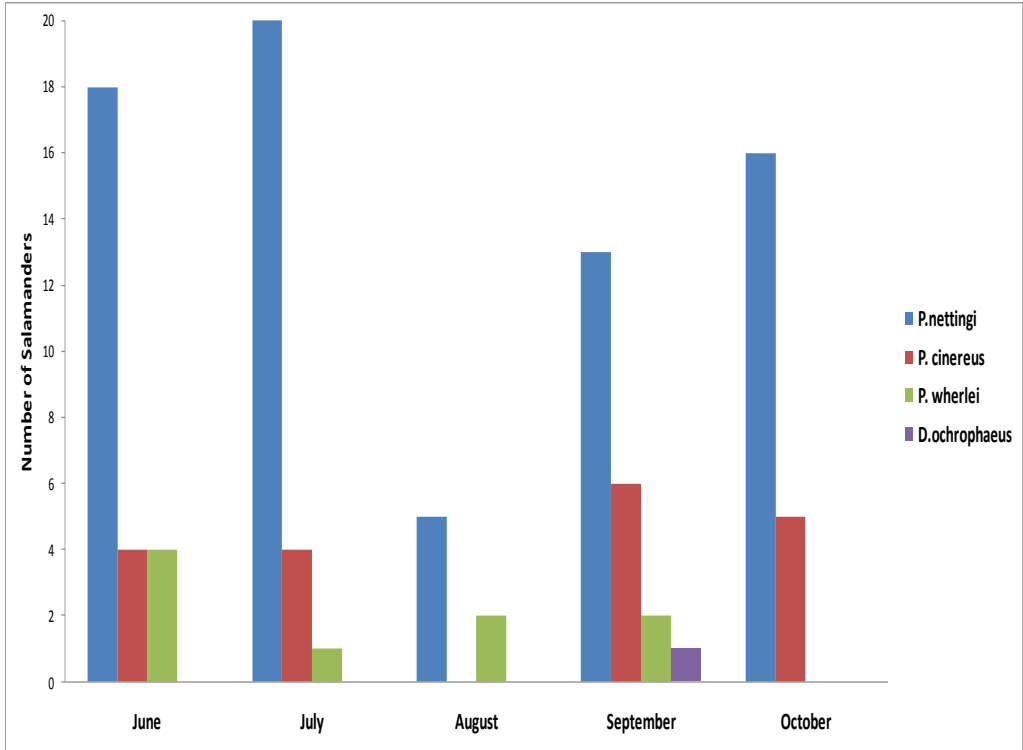


Figure 21: Number of salamanders caught monthly at Timberline Site 2 from June to October 2009.

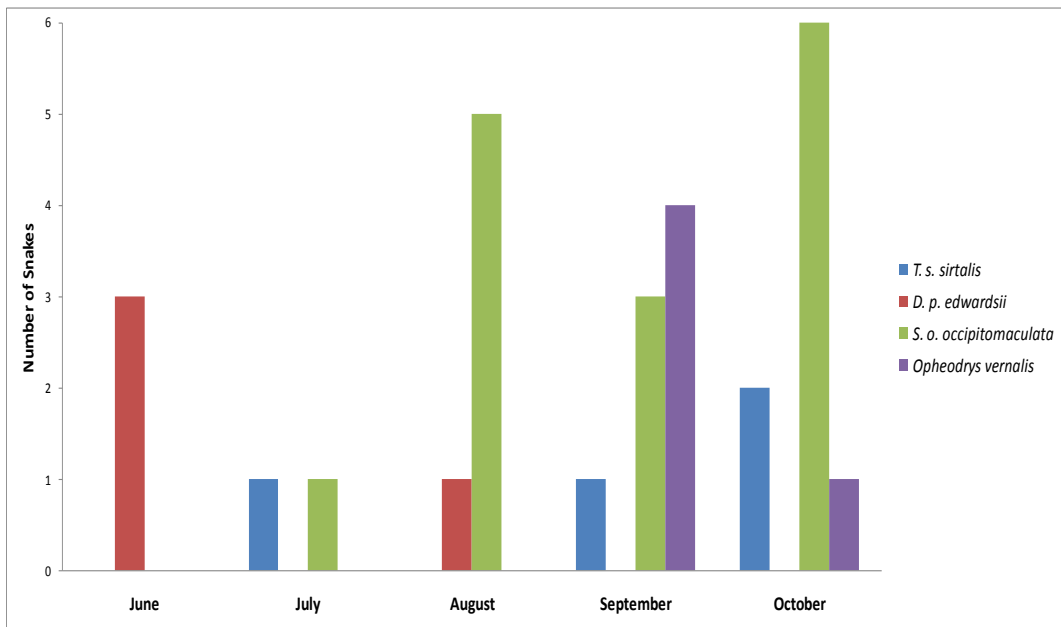


Figure 22: Number of snakes caught monthly at Timberline Site 2 from June – October 2009.

Timberline Site

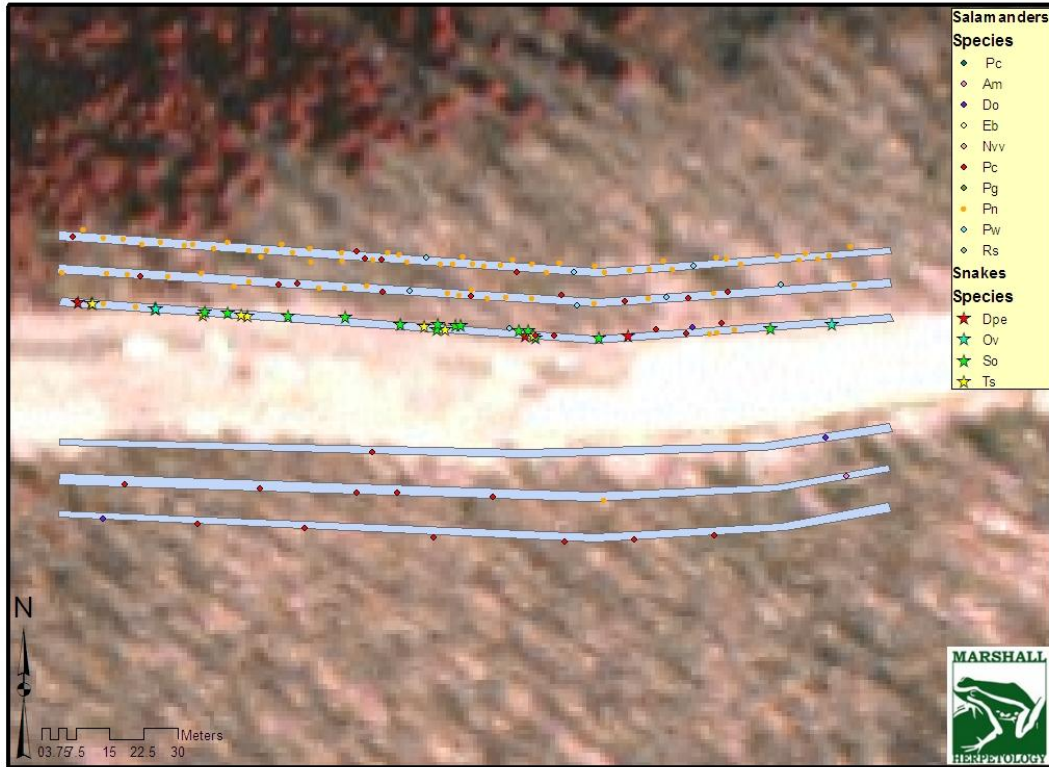


Figure 23: Aerial view of all 6 transects at Timberline with salamander by species (represented with circles) and snake by species (represented as stars) captures from June – October 2009.

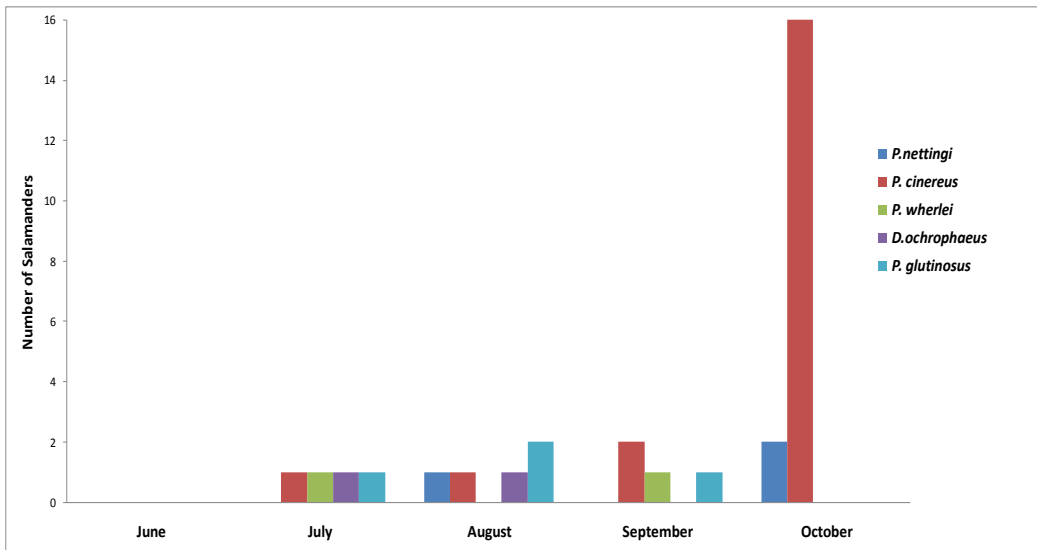


Figure 24: Number of salamanders caught monthly at Dolly Sods Site 1 from June to October 2009.

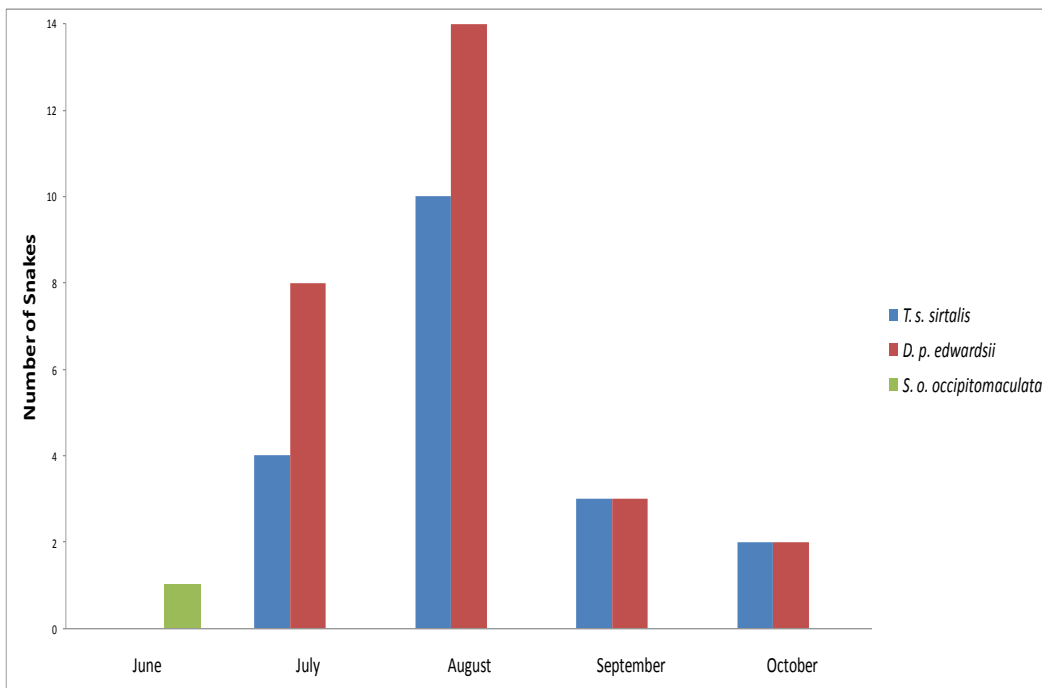


Figure 25 : Number of snakes caught monthly at Dolly Sods Site 1 from June – October 2009.

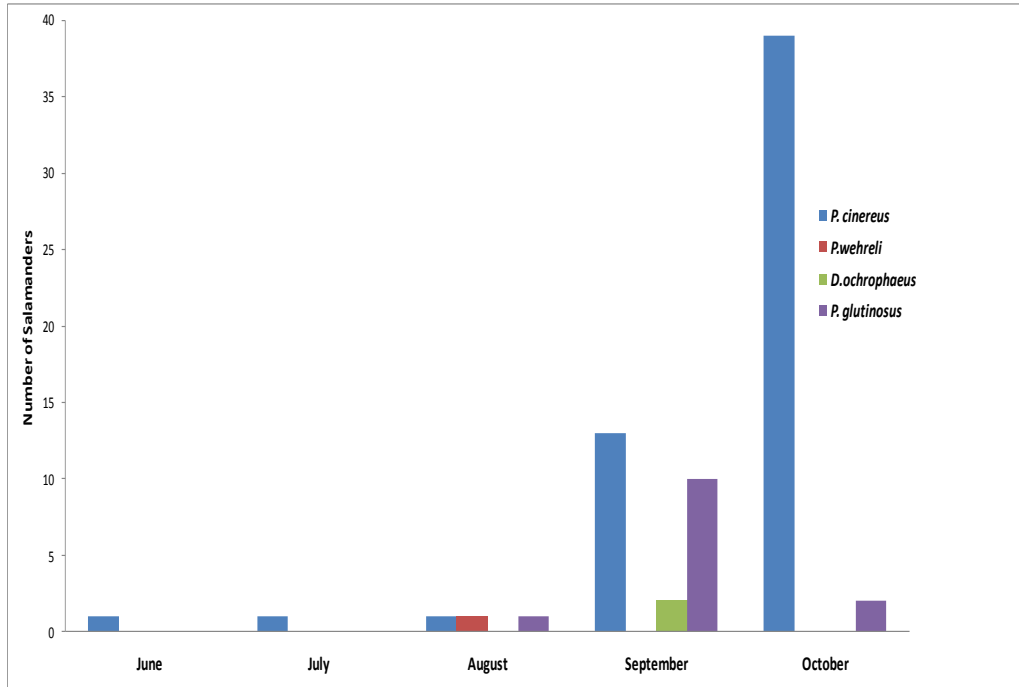


Figure 26: Number of salamanders caught monthly at Dolly Sods Site 2 from June to October 2009.

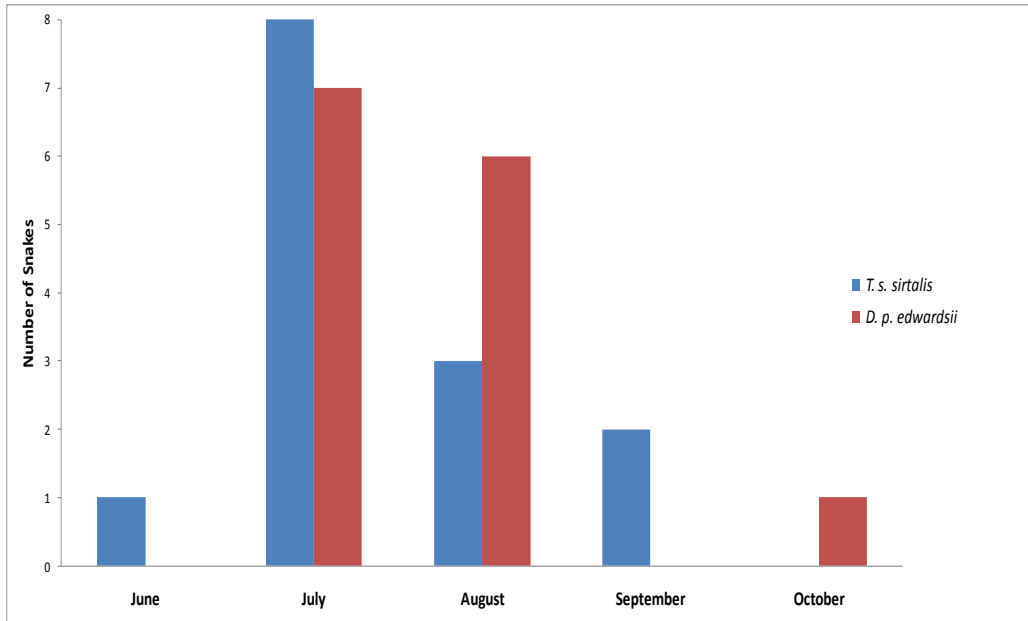


Figure 27: Number of snakes caught monthly at Dolly Sods Site 2 from June – October 2009.

Dolly Sods Site

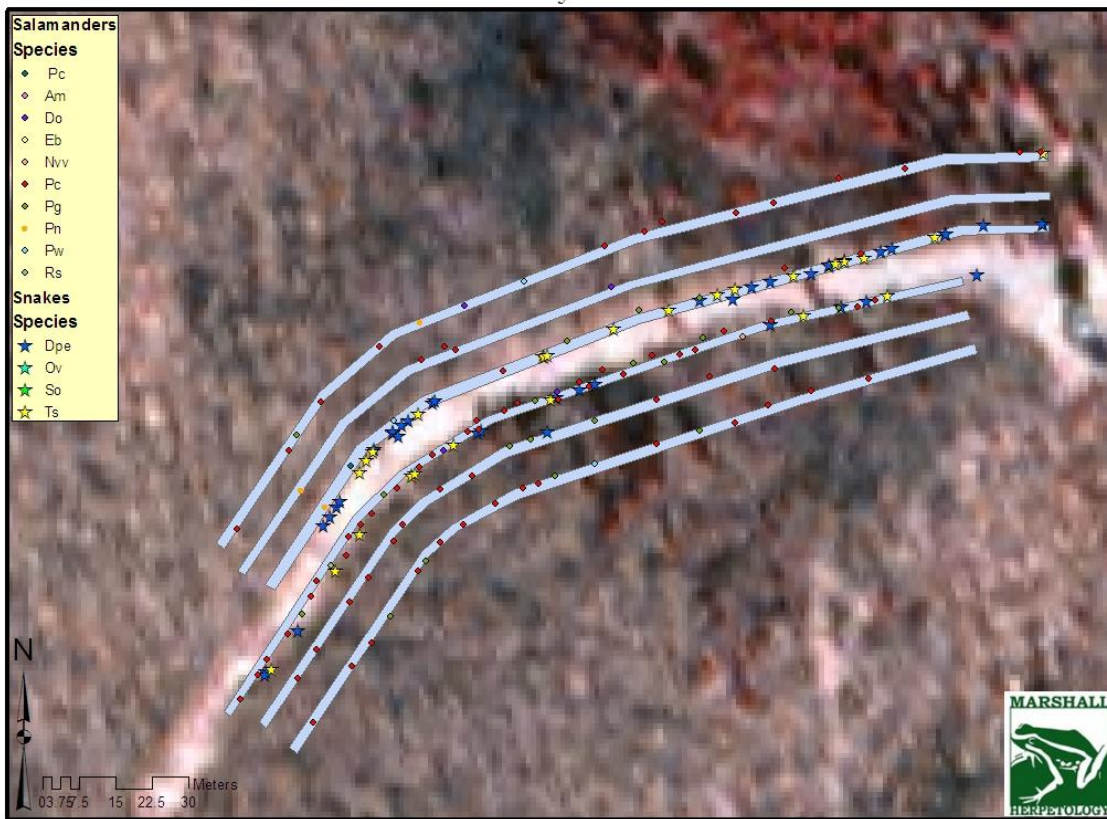


Figure 28: Aerial view of all 6 transects at Dolly Sods with salamander by species (represented with circles) and snake by species (represented as stars) captures from June – October 2009.

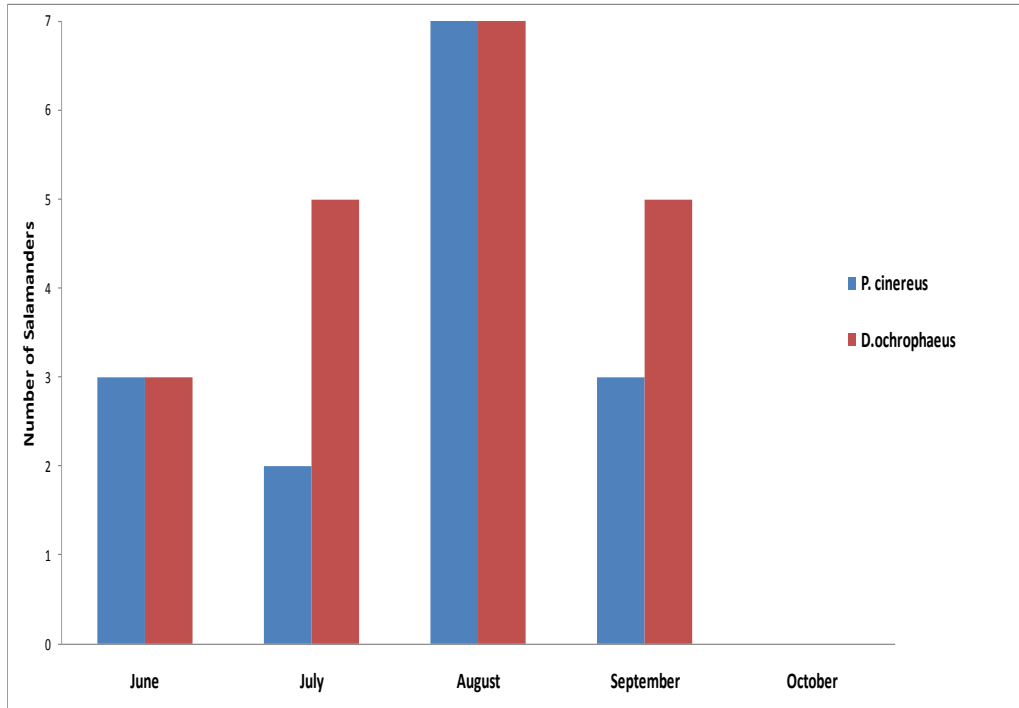


Figure 29: Number of salamanders caught monthly at Whitegrass Site 1 from June to October 2009.

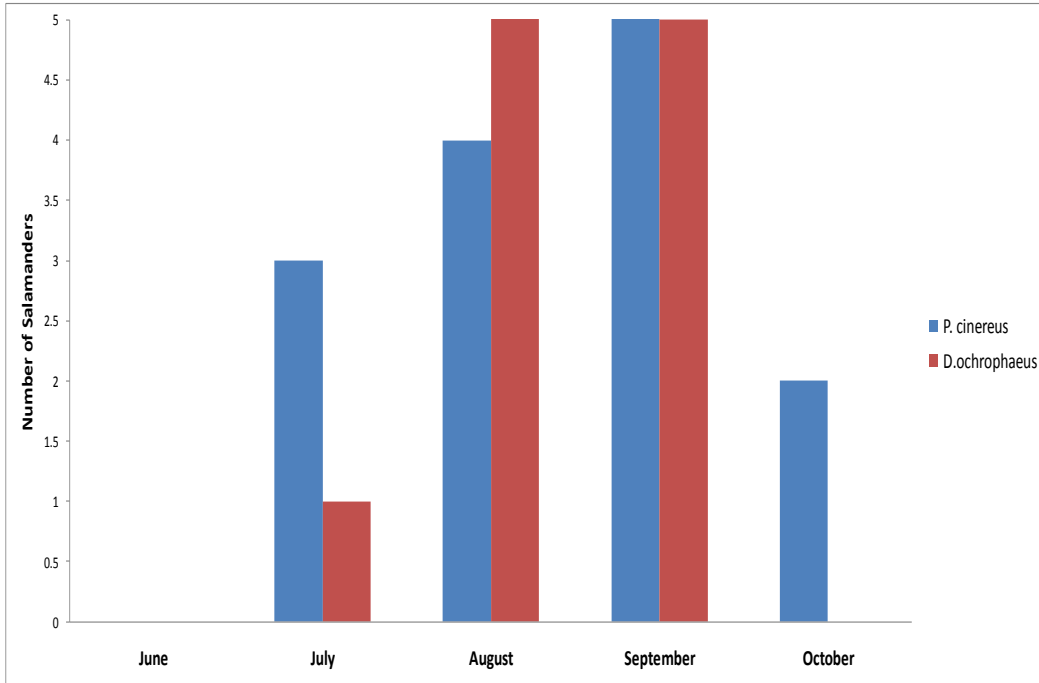


Figure 30: Number of salamanders caught monthly at Whitegrass Site 2 from June to October 2009.

Whitegrass Site

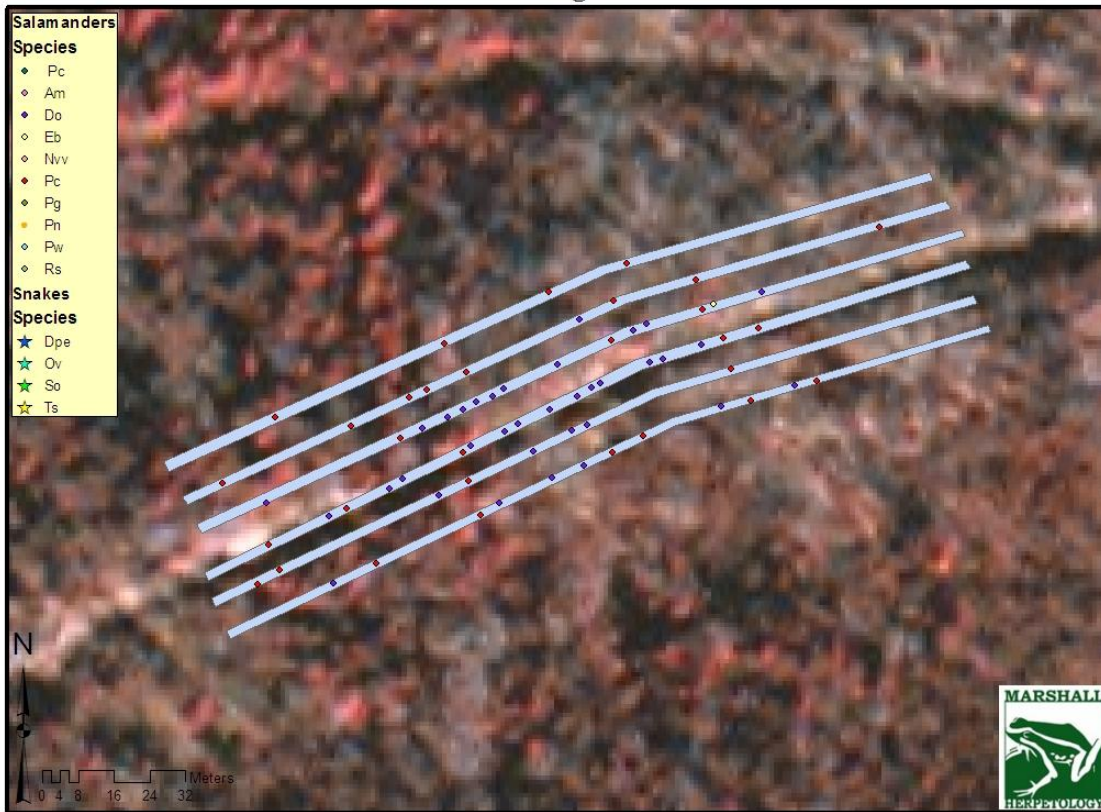


Figure 31: Aerial view of all 6 transects at Whitegrass with salamander by species (represented with circles) captures from June – October 2009.
No snakes were found at these sites.

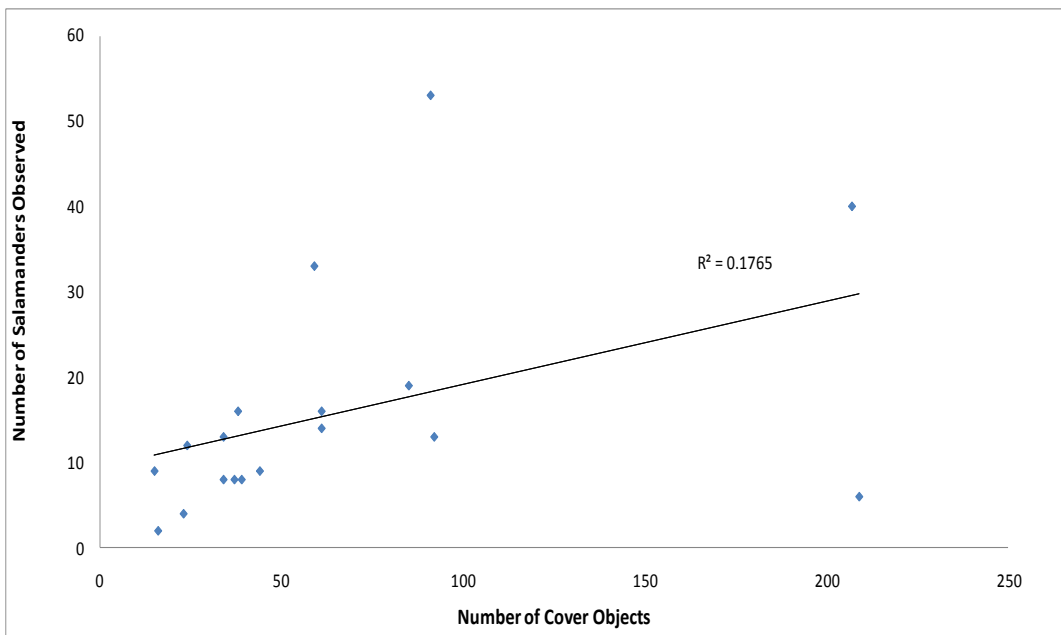


Figure 32: Number of cover objects versus number of salamanders caught during the field season. R² value = 0.1765.

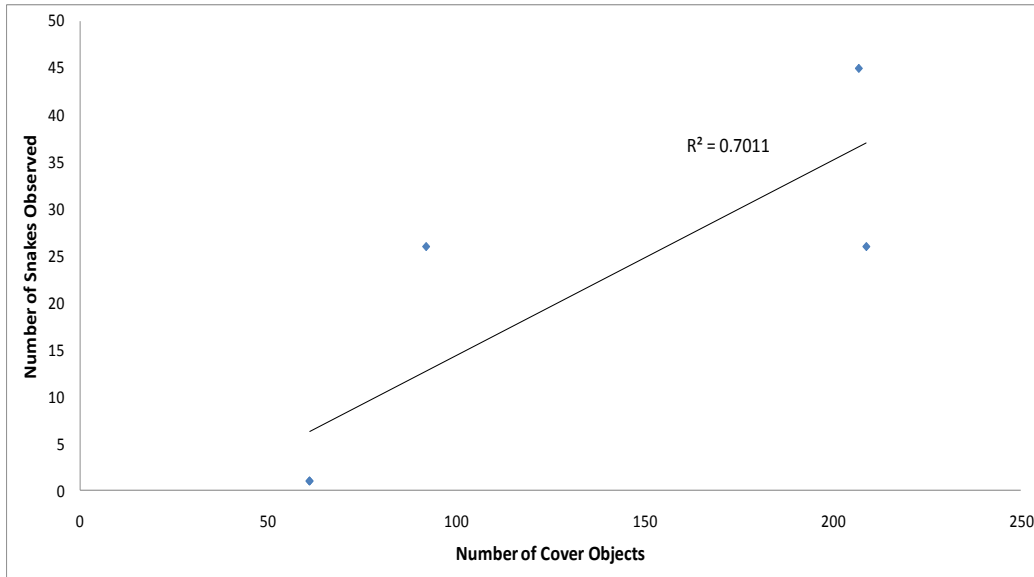


Figure 33: Number of cover objects versus number of snakes observed throughout the field season. R^2 value = 0.7011 showing a positive correlation. Values of each site were only taken where snakes were encountered.

Table 1: Timberline Site 1 Tree Species Analysis. Sp. Name = species name, C. Name = Common Name, Sp. Den. = Species Density, Sp. BA = Species Basal Area, Rel. Freq. = Relative Frequency, Rel. Den. = Relative Density, Rel. BA = Relative Basal Area and Imp. Val. = Importance Value.

Sp. Name	C. Name	Sp. Den. (Stems ha ⁻¹)	Sp. Freq.	Sp. BA (m ² ha ⁻¹)	Rel. Freq.	Rel. Den.	Rel. BA	Imp. Val.
<i>Fagus grandiflora</i>	American Beech	3968.25	1.00	113.02	56.24	78.13	64.05	198.42
<i>Prunus serotina</i>	Black Cherry	317.46	0.22	31.28	12.50	6.25	17.73	36.47
<i>Acer rubrum</i>	Red Maple	317.46	0.22	15.98	12.50	6.25	9.06	27.81
<i>Acer saccharum</i>	Sugar Maple	158.73	0.11	9.09	6.25	3.13	5.15	14.52
<i>Betula alleghaniensis</i>	Yellow Birch	158.73	0.11	4.99	6.25	3.13	2.83	12.20
<i>Picea rubens</i>	Red Spruce	158.73	0.11	2.11	6.25	3.13	1.19	10.57

Table 2: Timberline Site 2 Tree Species Analysis. Sp. Name = species name, C. Name = Common Name, Sp. Den. = Species Density, Sp. BA = Species Basal Area, Rel. Freq. = Relative Frequency, Rel. Den. = Relative Density, Rel. BA = Relative Basal Area and Imp. Val. = Importance Value.

Sp. Name	C. Name	Sp. Den. (Stems ha ⁻¹)	Sp. Freq.	Sp. BA (m2 ha-1)	Rel. Freq.	Rel. Den.	Rel. BA	Imp. Val.
<i>Fagus grandiflora</i>	American Beech	3492.06	0.78	58.22	30.43	46.81	18.46	95.70
<i>Acer saccharum</i>	Sugar Maple	476.19	0.33	119.29	13.04	6.38	37.82	57.24
<i>Picea rubens</i>	Red Spruce	1587.30	0.67	23.35	26.08	21.28	7.40	54.76
<i>Acer rubrum</i>	Red Maple	317.46	0.22	73.03	8.69	4.26	23.15	36.10
<i>Betula alleghaniensis</i>	Yellow Birch	952.38	0.22	28.26	8.69	12.77	8.96	30.42
<i>Prunus serotina</i>	Black Cherry	158.73	0.11	8.43	4.35	2.13	2.67	9.15
<i>Ilex montana</i>	Mountain Holly	317.46	0.11	0.81	4.35	4.26	0.26	8.86
<i>Betula lenta</i>	Sweet Birch	158.73	0.11	4.04	4.35	2.13	1.28	7.76

Table 3: Dolly Sods Site 1 Tree Species Analysis. Sp. Name = species name, C. Name = Common Name, Sp. Den. = Species Density, Sp. BA = Species Basal Area, Rel. Freq. = Relative Frequency, Rel. Den. = Relative Density, Rel. BA = Relative Basal Area and Imp. Val. = Importance Value.

Sp. Name	C. Name	Sp. Den. (Stems ha ⁻¹)	Sp. Freq.	Sp. BA (m ² ha ⁻¹)	Rel. Freq.	Rel. Den.	Rel. BA	Imp. Val.
<i>Betula alleghaniensis</i>	Yellow Birch	2222.22	0.02	0.78	4.87	30.43	28.00	63.30
<i>Quercus alba</i>	White Oak	634.92	0.11	0.33	34.00	8.70	12.00	54.69
<i>Ilex montana</i>	Mountain Holly	1904.76	0.00	0.56	0.22	26.09	20.00	46.31
<i>Picea rubens</i>	Red Spruce	476.19	0.08	0.22	23.86	6.52	8.00	38.39
<i>Acer rubrum</i>	Red Maple	793.65	0.04	0.33	12.02	10.87	12.00	34.89
<i>Quercus rubra</i>	Red Oak	158.73	0.06	0.11	19.47	2.17	4.00	25.64
<i>Acer pensylvanicum</i>	Striped Maple	317.46	0.01	0.22	1.59	4.35	8.00	13.94
<i>Rhododendron maximum</i>	Great Rhododendron	634.92	0.00	0.11	0.40	8.70	4.00	13.09
<i>Acer spicatum</i>	Mountain Maple	158.73	0.01	0.11	3.58	2.17	4.00	9.75

Table 4: Dolly Sods Site 2 Tree Species Analysis. Sp. Name = species name, C. Name = Common Name, Sp. Den. = Species Density, Sp. BA = Species Basal Area, Rel. Freq. = Relative Frequency, Rel. Den. = Relative Density, Rel. BA = Relative Basal Area and Imp. Val. = Importance Value.

Sp. Name	C. Name	Sp. Den. (Stems ha ⁻¹)	Sp. Freq.	Sp. BA (m ² ha- 1)	Rel. Freq.	Rel. Den.	Rel. BA	Imp. Val.
<i>Acer rubrum</i>	Red Maple	2380.95	0.05	0.78	42.54	25.42	24.14	92.11
<i>Ilex montana</i>	Mountain Holly	2698.41	0.00	0.89	1.57	28.81	27.59	57.97
<i>Betula alleghaniensis</i>	Yellow Birch	1111.11	0.01	0.44	7.61	11.86	13.79	33.27
<i>Sorbus americana</i>	Mountain Ash	476.19	0.03	0.11	20.39	5.08	3.45	28.92
<i>Picea rubens</i>	Red Spruce	158.73	0.02	0.11	18.19	1.69	3.45	23.33
<i>Prunus serotina</i>	Black Cherry	476.19	0.01	0.22	5.10	5.08	6.90	17.08
<i>Rhododendron maximum</i>	Great Rhododendron	1111.11	0.00	0.11	1.01	11.86	3.45	16.32
<i>Betula lenta</i>	Sweet Birch	476.19	0.00	0.22	1.57	5.08	6.90	13.56
<i>Acer pensylvanicum</i>	Striped Maple	317.46	0.00	0.22	1.01	3.39	6.90	11.29
<i>Hamamelis virginiana</i>	Witch Hazel	158.73	0.00	0.11	1.01	1.69	3.45	6.15

Table 5: Whitegrass Site 1 Tree Species Analysis. Sp. Name = species name, C. Name = Common Name, Sp. Den. = Species Density, Sp. BA = Species Basal Area, Rel. Freq. = Relative Frequency, Rel. Den. = Relative Density, Rel. BA = Relative Basal Area and Imp. Val. = Importance Value.

Sp. Name	C. Name	Sp. Den. (Stems ha ⁻¹)	Sp. Freq.	Sp. BA (m ² ha ⁻¹)	Rel. Freq.	Rel. Den.	Rel. BA	Imp. Val.
<i>Acer pensylvanicum</i>	Striped Maple	2857.14	0.00	0.67	0.33	33.96	28.58	62.86
<i>Picea rubens</i>	Red Spruce	2857.14	0.00	0.56	0.91	33.96	23.81	58.68
<i>Acer rubrum</i>	Red Maple	476.19	0.09	0.22	41.90	5.66	9.53	57.09
<i>Prunus serotina</i>	Black Cherry	317.46	0.08	0.11	37.12	3.77	4.76	45.65
<i>Betula alleghaniensis</i>	Yellow Birch	634.92	0.04	0.33	19.17	7.55	14.29	41.01
<i>Fagus grandiflora</i>	American Beech	1269.84	0.00	0.44	0.58	15.09	19.05	34.72

Table 6: Whitegrass Site 2 Tree Species Analysis. Sp. Name = species name, C. Name = Common Name, Sp. Den. = Species Density, Sp. BA = Species Basal Area, Rel. Freq. = Relative Frequency, Rel. Den. = Relative Density, Rel. BA = Relative Basal Area and Imp. Val. = Importance Value.

Sp. Name	C. Name	Sp. Den. (Stems ha ⁻¹)	Sp. Freq.	Sp. BA (m ² ha- 1)	Rel. Freq.	Rel. Den.	Rel. BA	Imp. Val.
<i>Prunus serotina</i>	Black Cherry	952.38	0.11	0.56	36.09	11.76	18.52	66.38
<i>Acer pensylvanicum</i>	Striped Maple	2857.14	0.00	0.89	0.42	35.29	29.63	65.35
<i>Picea rubens</i>	Red Spruce	2380.95	0.00	0.56	0.42	29.41	18.52	48.35
<i>Tsuga canadensis</i>	Eastern Hemlock	158.73	0.11	0.11	36.09	1.96	3.70	41.76
<i>Betula lenta</i>	Sweet Birch	476.19	0.05	0.33	15.19	5.88	11.11	32.18
<i>Betula alleghaniensis</i>	Yellow Birch	158.73	0.03	0.11	10.55	1.96	3.70	16.21
<i>Fagus grandiflora</i>	American Beech	634.92	0.00	0.22	0.42	7.84	7.41	15.67
<i>Ilex montana</i>	Mountain Holly	317.46	0.00	0.11	0.42	3.92	3.70	8.05
<i>Acer spicatum</i>	Mountain Maple	158.73	0.00	0.11	0.42	1.96	3.70	6.09

Table 7: Timberline Pearson Correlation of Environmental Factors. Correlation is significant at >0.90.

	Air Temperature	Soil Temperature	Relative Humidity
Air Temperature	1	.898**	-.561**
Soil Temperature	.898**	1	-.350**
Relative Humidity	-.561**	-.350**	1

Table 8: Dolly Sods Pearson Correlation of Environmental Factors. Correlation is significant at (>0.90).

	Air Temperature	Soil Temperature	Relative Humidity
Air Temperature	1	0.9	-.605**
Soil Temperature	0.9	1	-.661**
Relative Humidity	-.605**	-.661**	1

Table 9: Whitegrass Pearson Correlation of Environmental Factors. Correlation is significant at (>0.90).

	Air Temperature	Soil Temperature	Relative Humidity
Air Temperature	1	0.924	-.566**
Soil Temperature	0.924	1	-.556**
Relative Humidity	-.566**	-.556**	1

Table 10: Timberline Site 1 Average Air Temperature, Soil Temperature and Relative Humidity with Standard Deviations from June – October 2009.

	Mean (C°) AT	Standard Deviation	Mean (C°) ST	Standard Deviation	Mean RH	Standard Deviation
Transect A	20.68	4.00	17.57	4.00	62.59	16.89
Transect B	20.10	3.66	16.15	3.32	62.52	16.63
Transect C	19.77	3.52	16.08	3.56	63.51	15.72

Table 11: Timberline Site 2 Average Air Temperature, Soil Temperature and Relative Humidity with Standard Deviations from June – October 2009.

	Mean (C°) AT	Standard Deviation	Mean (C°) ST	Standard Deviation	Mean RH	Standard Deviation
Transect A	22.89	5.92	19.41	5.53	60.96	15.46
Transect B	21.66	4.84	17.28	4.54	60.13	13.97
Transect C	21.44	4.65	16.57	4.25	59.09	14.60

Table 12: Dolly Sods Site 1 Average Air Temperature, Soil Temperature and Relative Humidity with Standard Deviations from June – October 2009.

	Mean (C°) AT	Standard Deviation	Mean (C°) ST	Standard Deviation	Mean RH	Standard Deviation
Transect A	20.15	5.20	17.47	4.58	61.80	14.10
Transect B	19.96	5.02	16.64	4.55	61.07	13.89
Transect C	19.74	4.93	16.15	4.75	61.65	13.69

Table 13: Dolly Sods Site 2 Average Air Temperature, Soil Temperature and Relative Humidity with Standard Deviations from June – October 2009.

	Mean (C°) AT	Standard Deviation	Mean (C°) ST	Standard Deviation	Mean RH	Standard Deviation
Transect A	19.36	5.66	16.59	4.43	65.22	14.17
Transect B	19.36	5.71	15.76	4.18	64.79	14.84
Transect C	18.99	5.32	15.56	4.13	65.22	15.04

Table 14: Whitegrass Site 1 Average Air Temperature, Soil Temperature and Relative Humidity with Standard Deviations from June – October 2009.

	Mean (C°) AT	Standard Deviation	Mean (C°) ST	Standard Deviation	Mean RH	Standard Deviation
Transect A	18.07	5.08	16.93	4.45	70.53	12.58
Transect B	17.76	4.82	15.69	3.92	69.67	12.72
Transect C	17.59	4.66	15.72	3.95	69.24	12.44

Table 15: Whitegrass Site 2 Average Air Temperature, Soil Temperature and Relative Humidity with Standard Deviations from June – October 2009.

	Mean (C°) AT	Standard Deviation	Mean (C°) ST	Standard Deviation	Mean RH	Standard Deviation
Transect A	18.42	5.53	16.60	4.75	68.00	10.53
Transect B	17.72	4.76	15.65	4.10	68.45	9.72
Transect C	17.30	5.07	15.13	4.16	70.85	7.95

Table 16: ANOVA Results of Environmental Factors comparing all Transect A's, B's and C's at alpha = 0.05.

		Sum of Squares	df	Mean Square	F	Sig.
RH	Between Groups	.831	2	.415	.025	.975
	Within Groups	249.571	15	16.638		
	Total	250.402	17			
ST	Between Groups	8.143	2	4.072	6.813	.008*
	Within Groups	8.965	15	.598		
	Total	17.108	17			
AT	Between Groups	1.922	2	.961	.372	.695
	Within Groups	38.710	15	2.581		
	Total	40.632	17			

Table 17: Tukey results of environmental factors between transects. Transects A are represented as 1, Transects B = 2 and Transects C = 3 at alpha = 0.05.

Dependent Variable	(I) Site_num	(J) Site_num	Mean Difference	Significance
RH	1	2	0.413	0.983
		3	-0.076	0.999
	2	1	-0.413	0.983
		3	-0.489	0.977
	3	1	0.076	0.999
		2	0.489	0.977
ST	1	2	1.235*	0.036
		3	1.562*	0.009
	2	1	-1.235*	0.036
		3	0.327	0.748
	3	1	-1.562*	0.009
		2	-0.327	0.748
AT	1	2	0.5	0.853
		3	0.791	0.677
	2	1	-0.5	0.853
		3	0.291	0.947
	3	1	-0.791	0.677
		2	-0.291	0.947

Table 18: ANOVA Results for Timberline Site 1 and 2 Comparing Environmental Variables

		Sum of Squares	df	Mean Square	F	Sig.
RH	Between Groups	1.531	2	.766	.455	.672
	Within Groups	5.052	3	1.684		
	Total	6.583	5			
AT	Between Groups	.288	2	.144	.031	.970
	Within Groups	13.957	3	4.652		
	Total	14.245	5			
ST	Between Groups	5.339	2	2.670	3.291	.175
	Within Groups	2.434	3	.811		
	Total	7.773	5			

Table 19: ANOVA Results for Dolly Sods Site 1 and 2 Comparing Environmental Variables

		Sum of Squares	df	Mean Square	F	Sig.
RH	Between Groups	.164	2	.082	.319	.749
	Within Groups	.771	3	.257		
	Total	.935	5			
AT	Between Groups	.396	2	.198	.031	.970
	Within Groups	19.172	3	6.391		
	Total	19.568	5			
ST	Between Groups	1.466	2	.733	2.318	.246
	Within Groups	.949	3	.316		
	Total	2.416	5			

Table 20: ANOVA Results for Whitegrass Site 1 and 2 Comparing Environmental Variables

		Sum of Squares	df	Mean Square	F	Sig.
RH	Between Groups	.655	2	.327	9.707	.049
	Within Groups	.101	3	.034		
	Total	.756	5			
AT	Between Groups	1.079	2	.540	.309	.755
	Within Groups	5.237	3	1.746		
	Total	6.317	5			
ST	Between Groups	2.038	2	1.019	13.066	.033
	Within Groups	.234	3	.078		
	Total	2.272	5			

Table 21: Tukey Results of Environmental Factors at Whitegrass Site 1 and 2. Transects A are represented as 1, Transects B = 2 and Transects C = 3 at alpha = 0.05.

Dependent Variable	(I) SiteNum	(J) SiteNum	Mean Difference	Sig.
RH	1	2	0.502	0.139
		3	.800[*]	0.045
	2	1	-0.502	0.139
		3	0.298	0.363
	3	1	-.800[*]	0.045
		2	-0.298	0.363
AT	1	2	0.206	0.987
		3	-0.779	0.835
	2	1	-0.206	0.987
		3	-0.985	0.757
	3	1	0.779	0.835
		2	0.985	0.757
ST	1	2	1.093	0.059
		3	1.342[*]	0.035
	2	1	-1.093	0.059
		3	0.249	0.682
	3	1	-1.342[*]	0.035
		2	-0.249	0.682

Table 22: ANCOVA results between cover objects and animals observed at alpha = 0.05.

Source	Dependent Variable	df	F	Significance
Total Cover	Snakes Observed	1	34.435	0.000
	Total Captures	1	39.432	0.000
	Salamanders Observed	1	4.069	0.063

Table 23: Multivariate results showing total cover is significantly related to the amount of animals captured.

Effect		Value	F	Significance
Total Cover	Wilks' Lambda	0.123	46.501 ^a	0.000