Population and Nesting Ecology of Painted Turtles (*Chrysemys picta*) in Pennsylvania

Lily M. Bieber-Ham

Submitted in partial fulfillment of
Honors Requirements for the
Department of Environmental Studies

Scott Boback, Supervisor
Gene Wingert, Reader
Greg Howard, Reader

May 16, 2010
Dickinson College

ABSTRACT

Monitoring the population ecology of organisms is necessary for understanding the longevity of populations and for developing wildlife management plans. Painted turtles (*Chrysemys picta*) are a species commonly found in aquatic environs in Pennsylvania and eastern North America. An ongoing painted turtle mark-recapture study has been conducted at the Huntsdale Fish Hatchery Pond in Cumberland County, Pennsylvania since 2009. We estimate the population size to be 977±79 individuals, based on the Petersen estimator assuming a closed population. Turtle nesting was monitored during the summer of 2010. Nest site characteristics (depth, width, canopy coverage, distance from pond) and egg/clutch characters (length, width, mass, clutch size) were recorded. Additionally, we monitored internal nest temperatures using iButton™ temperature recording devices. Four nests were found. One hatchling painted turtle emerged in September 2010. A hatchling predation study began in the fall of 2010 to examine the hatchling predation intensity at this study site and to explore the reasons for overwintering in the nest. Previous work suggests that lower levels of predation during spring relative to the fall could be one benefit for hatchlings to emerge in the spring. Camera traps were set on hatchling replicas in fall 2010 and spring 2011 to compare levels of predation. In total, five predator species were detected, but there was no difference in estimated predation levels between fall and spring. These results suggest that at this site, reduced predation intensity is not a benefit afforded to hatchlings that overwinter in the nest. Because turtles are long-lived and exhibit high juvenile mortality, understanding factors that influence hatchling survivorship remains critical in understanding their population dynamics.

INTRODUCTION

Various organisms have the ability to act as indicators of environmental changes and ecosystem health. In order to observe certain environmental fluctuations, long-term research must be conducted on such indicator species. Ongoing ecological research can provide baseline and comparative data that the local and national community can use to make management decisions. Turtles remain a unique indicator organism due to their long life spans and historical resilience to withstand environmental changes.

Chrysemys picta (painted turtle) is one of the most well-studied reptile species due in part to their large geographic distribution and environmental tolerance (Ernst and Lovich 2009). This large body of previous research provides data for comparative analyses across geographic regions. By comparing data from multiple populations we can discover the ways in which this species has responded to environmental variation. Painted turtles have a distribution spreading across Southern Canada and the United States. Their ability to thrive in anthropogenic sites and contaminated waterways is well documented (Ernst and Lovich 2009). However, more typical habitats of these turtles consist of slow moving bodies of water such as ponds, lakes, small creeks, and marshes (Costanzo et al. 2004).

Painted turtles have a complex life-history with particularly mysterious reproductive behaviors. Female turtles venture out of the pond during the Spring-Summer months to nest. In Pennsylvania, the nesting season typically occurs between June and early July (Ernst 1971). During this time females leave the pond on nesting forays. The energetic cost of leaving the pond is extremely high for these aquatic reptiles, and their risk of terrestrial predation increases with increased time on land (Spencer 2002). Female turtles must therefore optimize the time that they spend out of the water. Upon leaving the pond females have been observed rubbing their heads against the soil in what could be attempts at detecting soil type, temperature, or moisture content suitable for nesting (Ernst and Lovich 2009). Nest site selection is especially critical for this species as a result of their unique biology. Painted turtles have temperature-dependent sex determination (TSD). Unlike most organisms in which sex is determined by genes, sex of the hatchling painted turtles is determined by the temperature within the nest during the middle third of development (Janzen, 1994). Painted turtles have TSD Type Ia in which males are produced at lower temperatures and females are produced at higher temperatures (Etchberger et al. 1992). Internal nest temperatures ranging from 29-32°C result in all female hatchlings whereas nest temperatures between 21.5°C and 27°C result in a clutch of entirely male hatchlings (Etchberger et al. 1992).

Nest temperature is influenced by various microhabitat characteristics including soil type, vegetative cover, aspect, and slope (Costanzo et al. 2008). Thus, female nest site selection directly influences the outcome of both the sex of the hatchlings and their survivorship (Schwanz et al. 2010). Female preference for certain microhabitat characteristics such as vegetative cover and for certain thermal environments could influence sex ratios over time (Schwanz et al. 2010; Morjan 2003). For instance, prior research has shown that individual females prefer specific levels of canopy coverage at their chosen nesting sites (Janzen and Morjan 2000). Female painted turtles at a site along the Mississippi River nest in sites with an average of 44% canopy coverage (Janzen and Morjan 2000). A wide range of canopy coverage values was recorded at this site, but the results showed that there is a positively correlation between the level of canopy coverage and the number of male hatchlings (Janzen and Morjan 2000).

Understanding why females nest in certain sites is a critical step in providing conservation plans for the species. As a result of temperature-dependent sex determination, painted turtles also have the potential to act as unique indicator species for global climate change. For instance, a 2°C increase in mean temperature could affect nest temperatures enough to alter the sex ratio, and a 4°C increase in mean temperature could result in 100% female offspring production (Janzen 1994). Small increases in temperature could skew the sex ratio of hatchling turtles and therefore impact population demography and lead to extinction (Schwanz et al. 2010; Hulin et al. 2009). Long-term monitoring will enable researchers to observe potential changes in nesting behavior as a result of a changing climate. It is possible that over time females could compensate for the shift in temperature by altering their nesting behaviors (Hulin et al. 2009). For instance, females could nest in cooler areas to reduce the overall temperature of the nest and level the sex ratio. After laying her clutch the female turtle returns to the pond exhibiting no parental care over her young. Therefore, nest site selection is of paramount importance in determining hatchling sex and survivorship (McGaugh et al. 2010).

The importance of nest site selection is significant given the proclivity of painted turtles to overwinter underground in the nest. Females must maximize the probability of hatchling survival by choosing ideal nest sites. Painted turtles in Pennsylvania and throughout their northern range frequently hatch underground at the end of incubation (65-80 days) but do not to emerge until the following spring (Ernst 1971). The turtles have the unique ability to survive freezing temperatures and hostile conditions (Costanzo et al. 2004). The ability to circumvent freezing enables the hatchlings to survive temperatures as low as -15°C (Costanzo et al. 2008). Extracellular body fluids and blood plasma of hatchling painted turtles do not contain ice nucleating agents, and thus the hatchlings can avoid freezing by supercooling (Constanzo et al. 2008). The young turtles sustain themselves with a diet of calcium rich shell remnants and surrounding soil particles for a brief period after hatching (Costanzo et al. 2008). The turtles remain in the nest for the duration of the winter after which they emerge in early to late spring (Costanzo et al. 2008).

Multiple reasons for overwintering have been proposed (Table 1). It has been suggested that both variability and uncertainty regarding resources contribute to overwintering behavior (Costanzo et al. 2008). The tendency for some female turtles to have multiple clutches throughout the nesting season could result in delayed emergence from the nest (Gibbons and Nelson 1978). Not all of the clutches are developmentally ready to emerge from the nest at the same time, and thus overwintering provides a buffer period for all hatchlings to complete their growth (Gibbons and Nelson 1978). In northern regions, the climatic variability also increases.

Lower temperatures in the fall and winter, variable precipitation levels, and limited food supplies all contribute to the uncertainty surrounding the ideal time for nest emergence (Costanzo et al. 2008).

There are various positive and negative implications of overwintering that can be explained using a cost/benefit analysis (Gibbons and Nelson 1978). Significant costs associated with overwintering in the nest include the inability to feed in the late summer to fall and increased chance of death by drowning (due to nest flooding), dehydration, and freezing (Gibbons and Nelson 1978; Costanzo et al. 2003). Potential benefits of overwintering include less exposure to bleak winter conditions (low temperatures, decreased food supply, and limited shelter), emergence in softer soil following spring rainfall, and refuge from predation (Wilbur 1975; Costanzo et al. 2008). In the fall, local predator populations may be at maximum levels and migratory predators are often concentrated at waterways (Costanzo et al. 2008). Remaining in the nest would allow hatchlings to avoid travelling to the water during the peak of pre-winter feeding activity (Costanzo et al. 2008). Further research regarding predation levels during the fall and spring emergence periods would provide valuable insight into hatchling turtle behavior.

For a major part of this study, I chose to focus on female painted turtle nesting ecology and behavior. I tested the possibility that decreased predator exposure during the fall has influenced emergence in this species. If overwintering is a mechanism for predator avoidance then hatchlings should be exposed to a greater density of predators in the fall compared to the early spring:

***Hypothesis*: Hatchling *Chrysemys picta* delay emergence into the spring as a way to minimize predation pressures.**

***Prediction*: There will be a significantly greater amount of predation of hatchling *Chrysemys picta* in the fall compared to in the spring.**

Painted turtles are the most vulnerable during the first few years of their life, and their mortality rate decreases with age (Wilbur 1975). Nest predation can reach 95-100% in certain years (Ernst and Lovich 2009). Mortality rates are also high following emergence. Past research shows that mortality rates can reach 92% between the developmental period in the nest and the time that a hatchling reaches the pond (Wilbur 1975).

The mortality rate of hatchlings is greatly impacted by predator species. Their small size, soft carapaces, and slow speed make them susceptible to a variety of larger and faster organisms. Numerous painted turtle nest and hatchling predators have been classified (Ernst and Lovich 2009). These predators depend on both visual and olfactory cues to prey on young turtles and nests (Strickland et al. 2010). Common raccoons (*Procyon lotor*) are considered the most devastating predator throughout the *C. picta* life cycle (from nest to maturity; Ernst and Lovich, 2009).

I utilized camera traps to monitor predator species without human disturbance at the site. This non-invasive technique provides a useful way of detecting evasive predator species (Rowcliffe et al. 2008). Animal replicas have been successfully used in a variety of predation studies (Brodie 1993; Stuart-Fox et al. 2003). These replicas are typically made from soft materials such as clay and plasticine. Predation on these soft-bodied replicas is typically assessed via replica displacement and/or surface imprinting by predators (Brodie 1993; Steffen 2009; Stuart-Fox et al. 2003). This study will combine the use of camera traps and plaster casted hatchling replicas to examine hatchling painted turtle predators in Pennsylvania. The plaster replicas have advantages over plasticine or clay replicas in that they can be painted to more accurately mimic real hatchlings. The game cameras will enable predator species identification and determination of predation attempt frequency.

Observing local hatchling predator species will contribute to our overall understanding of painted turtle overwintering behavior. Understanding the early life-history of the painted turtle and how females choose nest sites is necessary for monitoring long-term changes in population dynamics. These turtles have the potential to act as environmental indicators due to their long life spans and ability to withstand harsh environmental conditions. It is thus important to monitor their habitats and population ecology as a means of observing the impact of long-term environmental changes on vertebrate species.

METHODS

Study Site

This study took place surrounding the Huntsdale Fish Hatchery pond in Cumberland County, Pennsylvania (40.106355°, -77.297616°). The Huntsdale Fish Hatchery, consisting of 80 acres, was built in 1932. The study site consists of one 2.8 hectare man-made pond, surrounding satellite ponds, wooded areas, and the adjacent railroad tracks. The Yellow Breeches Creek runs alongside the study site and flows into the pond via a drainage tunnel. The anthropogenic pond was built after 1950 as a holding area for various fish species. The pond no longer functions as a component of the fish hatchery. This area is now a recreational destination for fishing, hunting, and hiking. A walking pathway around the pond's edge provides convenient seasonal access. The pond is dominated by mixed emergent vegetation such as cattails (*Typha sp.*), aquatic grasses, and algae species. The surrounding site is dominated by foxtail grasses (*Seteria sp.*), switch grass (*Panicum spp.*), multiflora rose (*Rosa multiflora*), Tartarian honeysuckle (*Lonicera tatarica*), and a variety of tree species. The dominant tree species include Norway maple (*Acer plantanoides*), red maple (*Acer rubrum*), and ash (*Fraxinus sp.*). The railroad adjacent to the pond consists of one Norfolk Southern Railroad line with granite track ballast. A single lane path next to the tracks provides truck access for railroad maintenance. This path consists of crushed coal and stone soil. This coarse soil is a known nesting habitat for various turtle species at this site.

Population Ecology

An ongoing painted turtle mark-recapture study has been conducted at this study site since 2009. In addition to painted turtles, this site is home to at least three aquatic turtle species (*Sternotherus odoratus, Clemmys guttata, and Chelydra serpentina*). Mark-recapture sessions have been conducted in the spring of 2009, summer-fall 2009, and spring-summer 2010. Turtles were captured using basking traps and sardine baited hoop traps. Each turtle was processed by measuring carapace length, plastron length, tail length, and claw length. Turtles were also aged and sexed. Age was determined by counting the number of rings on the pectoral and abdominal plastron scutes. Sex was determined by cloacal distance from carapace (male cloacas extended beyond the carapace) and by claw length. Passive integrated transponders (PIT tags) were inserted into the posterior upper hind limb of all turtles that were large enough and the insertion wound was sutured with super glue. All turtles were marked via a secondary marking technique in which a unique series of notches were filed on the margin of each turtle's carapace. Turtles were released back into the pond following processing. A population estimate was generated using the Petersen estimator assuming a closed population. Sample number one consisted of all turtles captured in 2009, and sample number two consisted of all turtles captured in 2010.

Nesting Ecology

Nesting Activity

Visual encounter surveys (VES) were conducted for eight weeks during June and July 2010. Surveys were conducted one to two times per day between 0730 and 1700 hours. A perimeter around the pond, and path along the adjacent railroad tracks, were followed daily. Nesting females were observed at a distance and undisturbed until nesting was complete (Figure 1A). Binoculars were often used to observe females at a distance. The nest was approached upon completion of laying. When possible, spent females, or females suspected of recent nesting, were collected and measured. The survey region was also monitored for previously completed nests in which the female was no longer present. Disturbances in the soil were observed and carefully inspected for eggs. The location of all nest attempts was recorded using a hand-held GPS unit (Trimble, Sunnyvale, CA). Nest attempts were classified as sites in which a female began digging but abandoned the site prior to laying. Distinct characteristics of turtle nest attempts include shallow holes with smooth plastron marks flattening the surrounding soil on one side. Often these cavities would terminate at a large rock or root. This is presumably were females had attempted but failed to excavate nests.

Nest Data and Measurements

Each nest was carefully examined following the departure of the female. Nests were excavated and eggs were removed, individually measured, and weighed using a portable battery powered scale from Denver Instrument (Bohemia, NY, Figure 1B). Exact position of each egg was noted in order to ensure that eggs were put back into their original positions. The depth and width of each nest was recorded after the eggs were removed. One to two iButton® temperature probes (Maxim, CA) were placed into each nest chamber along with the eggs (Costanzo et al., 2003). In the laboratory iButton® temperature probes were set and coated in Plasti Dip™ for water proofing purposes. The iButtons® were set to record the temperature within the nest every four hours for 365 days. After processing the eggs were replaced, iButtons placed, and nests were refilled and covered.

We obtained a variety of environmental parameters for each nest. The canopy coverage at each nest was measured using a Forestry Suppliers convex spherical densitometer (Jackson, MI). Measurements were taken at ground level directly over the nest.

Predator exclusion cages were then placed over each nest to minimize predation and disturbances (Figure 1C). Cages were 30 cm^2 and constructed with green hardware cloth (Figure 1C). The location of each nest site was recorded using a handheld Trimble GPS device. The distance from each nest to the pond was then recorded using ArcGIS software.

Nest Monitoring and Emergence

Nests were monitored for hatchling emergence in fall 2010 (between August 23rd and November 14th) and for spring 2011 emergence starting on March 27. Nest cages were checked every other day for signs of hatchling activity. Hatchlings were removed from cages when they were discovered. Individuals were measured, marked, and photographed. Hatchling sex will be determined using geometric morphometric analysis of the carapace photographs (Valenzuela et al. 2004). Unique notches were cut into the carapace for future identification. Hatchlings were then released at the pond.

Camera Trap Study

Hatchling Painted Turtle Predation

Hatchling replicas were set at two forest edge locations along the railroad tracks, sites known to be used by painted turtles for nesting. The replicas were designed to accurately portray hatchling painted turtles with all significant markings and coloration patterns. The first replica was made using a plastic turtle toy (2½ cm carapace) from Century Novelty (Century Novelty.com) (Figure 2A). Acrylic paint was used to add detail. The next three replicas (2-3½ cm carapaces) were obtained from Morgan Reptile Replicas (Liberty, NC). These replicas were made of plaster and hand painted. Each replica was set directly onto the ground and was unobstructed from surrounding vegetation and debris (Figure 2B). A wire was attached to an eye-hook on the plastron of the second replica to prevent removal by predators. The wire was then obscured with vegetative debris. Reconyx, INC. game cameras (PC800 HyperFire™, Holmen,MI) were attached to tree trunks within one meter of the replica. These cameras have an infrared sensitive trigger that takes photos when heat radiation is emitted from an organism moving within camera range. All camera triggers were set at sensitive rapidfire mode in which five images were recorded for every trigger. The cameras were fastened 45 cm off of the ground. In an attempt to delineate visual and olfactory predators, cameras were set on replicas with and without turtle scent. Our turtle scent consisted of water from an aquarium housing a painted turtle hatchling, and this was sprayed on and around the replica (Marchand et al. 2002). Cameras and replicas were checked every three days at which compact flash cards were replaced and scent was refreshed. All photographs were analyzed after being downloaded onto a computer. Animals captured in photographs were identified to species. The proportion of photographs per species was determined (Nielson and McCollough 2009). Using photos three levels of behaviors were classified.

Predation Attempt: Organism had oral contact with the replica visible in the photograph.

Possible Predation Attempt: An event in which the predator's head, or the replica, was obscured in the photograph, but the predator was directly next to the replica and/or the replica had been physically moved upon inspection the following day.

Interacting with Replica: Any organism photographed sniffing or observing the replica.

The cameras were set on hatchlings for 48 camera-days (1,152 hours) in the fall. This study design was relicated in the spring for a comparative trial. The cameras were set from April 5th-25th on a replica with no scent, and between April 15th and 25th cameras were set on replicas with scent. A total of 40 camera-days were analyzed (960 hours) for the spring trial. The cameras were set in the fall and the spring for a combined total of 2,112 camera-hours.

RESULTS

Population Ecology

A total of 826 of turtles have been captured, marked, and released at the study site between the spring of 2009 and the summer of 2010. In 2009, 648 turtles were marked and released. In 2010, 850 turtles were captured between March and July. This included 178 new unmarked turtles. The population is estimated to be 977±79 individuals. The sex ratio of the population is 1.2:0.84.

Nest Characteristics

A total of four painted turtle nests were found between June 12 and June 28, 2010. Nests were detected between the morning and early afternoon (0830-1200). Ten females were observed out of the pond between June and early July (Table 2). Some of these females were encountered while digging nests, but all but one female abandoned the site without laying her clutch. Further inspection of many of these attempt sites revealed large rocks or root systems that could have inhibited the female from digging a hole deep enough for laying. Clusters of nest attempts were typically found in close proximity to each other. A total of 22 nest attempts were observed. One female was observed in the process of laying (Table 2).

Her nest was examined and excavated after she had left the site. Three completed nests were found adjacent to the railroad tracks bordering the Huntsdale property, and one nest was found along the edge of the pond (Figure 3).

Nest data from the Huntsdale site was compared to data from throughout the species range (Table 3). The mean nest depth and width were 5.9cm and 4.0cm respectively (Table 3). Distances from each nest to the pond were variable, ranging from 0.4m to 68.9m (Table 3). The nest at the edge of the pond (0.4m from water) was made in soft loamy soil. All other nests were dug in rocky charcoal filled soil along the railroad tracks. No clutch consisted of fewer than 4 eggs or more than 7 eggs (5.3±1.3) (Table 3).

One hatchling emerged in the fall on September 28, 2010 after 108 days underground. The hatchling emerged after previous day of rainfall. This was one hatchling from a clutch of seven laid adjacent to the railroad tracks. The temperature probe will remain in this nest until the spring emergence period has ended, and it can then be determined if the remaining four eggs successfully overwintered.

Predator detection during the fall

During the fall trial, the cameras recorded a total of 1,965 animal images including eleven different species throughout the experimental period. The observed organisms consisted of a variety of mammal and bird species (Table 4 & Figure 5). Three predator species were photographed (Table 4 & Figure 4). These predator images accounted for 3% of the total number of animal images. One definitive predation event was recorded. A Virginia opossum (*Didelphis virginiana)* was observed picking up a hatchling replica (with scent) and inserting it into its mouth (Figure 4A &B). A second possible predation attempt was recorded.

A striped skunk (*Mephitis mephitis*) was photographed in front of the same hatchling replica. It appeared to move the replica though this was out of view because the skunk was facing away from the camera. The most frequently photographed organism, accounting for 41% of the total of recorded images, was the white-footed mouse (*Peromyscus leucopus*) (Table 4).

Predator detection during the spring

During the spring trial 597 animal images were recorded over a 40 camera-day period. A total of eight different species were observed (Table 4). Five predator species were photographed, accounting for 9% of the total number of animal images. One possible predation attempt occurred involving a striped skunk *(Mephitis mephitis)*. This event occurred on the hatchling replica without scent. The skunk had its back to the camera in all of the five photographs that were taken (Figure 4D). White-tailed deer were the most frequently photographed organism for the spring trial (Figure 5D). Six animal species that were detected in the fall were not detected in the spring, and three new organisms were first photographed during the spring trial (*Corvus brachyrhynchos, Procylon lotor, Marmota monax*) (Table 4). The least frequently photographed animal in both the fall and the spring was the red fox. This species accounted for 0.2% of the images in both seasons. All of the cameras remained functioning without technical problems throughout the study.

DISCUSSION

Population Ecology

Understanding the population ecology of painted turtles is critical for monitoring the health and vitality of the species over time. Consistent mark-recapture programs are especially important as a result of the long life span of freshwater turtle species (Frazer et al. 1990). In general, individual turtles can exceed 35 years of age, but age is site dependent (Frazer et al. 1990). As this pond is manmade, it could provide practical comparative data for looking into the ability of this species to function in various habitats over time. Based upon our given population estimate of 977 turtles, there are approximately 349 turtles per hectare in the Huntsdale Fish Hatchery Pond. This estimate is relatively conservative compared to the density of painted turtles at a study site in Michigan in which the density was 828 individuals per hectare (Frazer et al. 1990). Abundant freshwater habitats in Cumberland County may provide alternative territories for painted turtles in the region.

Nesting Ecology

Painted turtles essentially determine the fate of their young with their nest site choice. Many hypotheses have been made regarding female nest site selection as a result of the predominantly environmental control over the sex ratio of a clutch. The preliminary data collected on painted turtle nests during the summer of 2010 will provide a baseline for continued research at this site. During the nesting season, females had the greatest levels of terrestrial activity during the morning (before 1200). This differs from observations that indicate that females most actively seek nest sites in the afternoon and evening (Ernst and Lovich 2009). The majority of the discovered nests and nest attempts were found along the railroad tracks north of the pond. This suggests that females exert more energy to travel further from the pond to seek this nesting environment. Females may also be restricted to nesting along the southern side of the railroad tracks as a result of the inability of turtles to cross this anthropogenic impasse. The track provides a physical barrier that limits the potential nesting area for gravid females. This highlights the importance of monitoring nesting as a means of understanding the impact that human infrastructure may have on the reproductive behaviors of local species.

The comparison of nest site characteristics at various study sites across the United States enables us to begin monitoring long-term changes that may vary geographically. Our average clutch size was consistent with one previously recorded average clutch size for a population in Southeastern Pennsylvania. The mean clutch size of the two painted turtle populations in New Mexico and Illinois was greater than at our study site. The lowest end of the clutch size range at both the New Mexico and Illinois site was 6 eggs (Morjan 2003). This is greater than the average at both Pennsylvania sites. The differences in clutch size enable us to confirm reproductive variation between *Chrysemys picta* subspecies. The New Mexico and Illinois populations consist of *C. picta bellii* (Morjan 2003). The subspecies at our Pennsylvania site are *C. picta picta* and *C. picta marginata*. The larger body size of *C. picta bellii* (maximum carapace length of 25.4 cm compared to 19.0 cm for *C. picta picta* and 19.5 cm for *C. picta marginata*) generally results in larger clutch sizes (Ernst and Lovich 2009).

The average nest depth of 10.4 cm at the other Pennsylvania site was considerably deeper than the average of 5.6 cm at our site (Ernst and Lovich 2009). The minimum nest depth reported by Ernst and Lovich (2009) was 3.2 cm deeper than our maximum reported depth. This variation in nest depth may be a result of differences in soil characteristics at the two sites. Although the soil type was not described for the population in the Southeastern Pennsylvania site, at our site soil along the railroad tracks is composed primarily of rocky charcoal soil. Denser and rockier soil may inhibit the females from digging deeper nests at this site. Digging shallower nests may also be a means of influencing the internal nest temperatures during hatchling development (Morjan 2003). Shallower nests have higher internal nest temperatures and thus could impact the sex ratio of the hatchlings (Morjan 2003). It has been found that Southern populations of painted turtles dig deeper nests, possibly as a way to ensure that the nest does not overheat in warmer climates (Morjan 2003).

The distance between nests and the water source is also a condition that could impact the success of hatchlings (Morjan 2003). Hydric conditions within the nest are influenced by the nest's distance to water and others have found that soil moisture content is reduced within the nest as the distance from water increases (Morjan 2003). Additionally, nesting close to the pond may result in an increased chance of mortality due to nest flooding or predation (Morjan 2003; Marchand et al. 2002). It has been suggested that females may specifically use soil moisture content as a means of determining optimal nesting locations (Morjan 2003). All of these factors may influence female nest site selection. The average distance from each nest to the site specific pond varies greatly across painted turtle populations. Our wide range of recorded distances (0.4-68.9 m) is consistent with observations in Illinois (0-86.3 m; Morjan 2003).

The thermal environment within the nest is directly correlated with canopy coverage over the nest site (Janzen and Morjan 2001). There was a relatively high amount of canopy coverage above nests at our site (70-90%). The temperature data provided from our iButtons® will enable us to compare the canopy coverage, internal nest temperatures, and hatchling sex ratio of each nest. The high levels of canopy coverage could reduce the internal nest temperatures of the shallower nests found at this site. The positive correlation between canopy coverage and the relative maleness of the clutch could have long-term implications on painted turtle populations (Janzen and Morjan 2001). Females in an Illinois population nested non-randomly in regards to canopy coverage (Janzen and Morjan 2001). This suggests that females may be able to adapt to long-term climate changes and alter the sex ratio of certain populations. Since females consistently preferred specific levels of canopy coverage it is possible that this nesting behavior may be able to evolve and compensate for skewed sex ratios (Janzen and Morjan 2001).

The nests at Huntsdale will be continually monitored for hatchling emergence. The nests will be excavated in June 2011 if hatchling movement has been detected. At that time iButtons® will be collected and temperature data will be compared to hatchling sex ratio within each nest. Long-term monitoring of painted turtle nesting at this site could provide insight into the ability of a species to indicate ecosystem changes as a result of a changing climate.

Camera Trap Study

The results of this study demonstrate the success of game cameras and plaster replicas at capturing the potential predators on painted turtle hatchlings. Additionally, we can detect non-predators. Cameras provide an unobtrusive way of observing organisms without the influence of direct human presence. Organisms with a wide range of sizes and locomotory speeds were photographed throughout the study period. This increases our confidence that all predator species would have been recorded if present in the camera range. The cameras also allow for extensive periods of uninterrupted data collection. The images that were collected enabled the classification of a variety of local species. This leads to a greater understanding of the study site and the organisms which *C. picta* may encounter. At this point there is limited support for the hypothesis that hatchling painted turtles delay emergence into the spring as a way to minimize predation pressures. There were very few predation events and possible predation events in both the fall and spring. One predation attempt and one potential predation attempt occurred in the fall, and one potential predation attempt occurred in the spring. Overall, there were a greater number of predator species detected in the spring than in the fall.

The five predator species that were recorded are all classified as known hatchling painted turtle predators (Ernst and Lovich 2009). The one predation attempt by the opossum was on the hatchling replica augmented with turtle scent. Opossums depend predominately on their keen sense of smell to locate prey (Krause and Krause 2006). These tactile marsupials also rely heavily on touch for foraging in the dark (Krause and Krause 2006). Research suggests that vision is not their predominate sense used while foraging (Krause and Krause 2006). The observed predation event, and this background information, suggests that the opossum was attracted to the hatchling replica as a result of its scent. Images show the opossum sniffing out the area with its nose held low to the ground.

Both the definitive and potential predation events involved olfactory organisms. Striped skunks are also olfactory predators (Conover and Borgo 2009). The two potential predation attempts involved one striped skunk in the fall and one in the spring. This suggests that some local predators may rely heavily on their sense of smell for detecting hatchling turtles. Using water from a hatchling turtle tank has the potential to mimic turtle scent and attract predator species (Marchand et al. 2002).

Raccoons, another olfactory predator, are considered the most devastating painted turtle nest and hatchling predators (Ernst and Lovich 2009). One raccoon was detected during the spring, but it was not observed interacting with the hatchling replica. The seemingly low intensity of raccoon predation in this area may be a result of hunting and trapping that takes place on the property. Culling mammalian predator species has the potential to positively impact freshwater turtle populations (Christiansen and Gallaway 1984). Therefore, the local raccoon hunting may benefit the general biodiversity at the study site.

The distance the cameras and replicas were placed from the pond could also contribute to low rates of predation. Prior research has shown that the predation of freshwater turtle nests decreases as the distance from the water increases (as the area surrounding the edges of ponds and waterways is the most highly predated) (Marchand et al. 2010). Predator species may be more prevalent directly surrounding the Huntsdale pond compared to along the railroad tracks, and their ranges could fluctuate with seasonal changes. Cameras could be set directly around the perimeter of the pond in future studies. This would allow us to observe any relationship or correlation between distance from the pond and predation intensity.

Human activity was moderate in the study region during the fall as a consequence of the white-tailed deer bow hunting season. Hunters were photographed on various occasions throughout the day and night (during the period in which nocturnal predators would be active). This human disturbance has the potential to deter skittish predator species from predating hatchling turtles (Leighton et al. 2010). Predator species in the spring may have been more active near the game camera sites as a result of decreased human activity at night. Recent research by Leighton et al. (2010) describes the “scarecrow effect,” in which human activity could dissuade predators from targeting specific areas at a given time. Thus, predator displacement at this site could aide in the conservation of turtle species by limiting predation (Leighton et al. 2010). A follow-up study could be done to classify the level of human activity at this site.

Each of the previous factors provides insight into local predation intensity. The classification of potential hatchling turtle predators is important in understanding mortality rates and thus overall population dynamics at his study site. Understanding the complexity of predator-prey dynamics is a significant factor in conserving threatened species and understanding local ecosystem ecology.

CONCLUSIONS

Currently our results suggest that hatchling predation levels do not differ between the fall and the spring. Therefore, our hypothesis that hatchling painted turtles delay emergence into the spring as a way to minimize predation pressure cannot be supported. The hypothesis could be tested further with more data collection over a few consecutive years. Repetition of this study is important as a result of yearly variation in predation pressures. Other factors may be influencing overwintering behavior if the levels of predation remain unchanged after continued research. Overwintering may have evolved as a result of other environmental factors unrelated to predation, or there is the possibility that this study site is unique. This study could be replicated at the Wildwood Park in Harrisburg, Pennsylvania. Repeating the study at another site would enable us to examine the seasonal predation trends on another painted turtle population within the same state. Various factors could be considered when expanding the camera trap study. Replicas could be set at a variety of sites (such as directly next to the pond, on walking paths, and along the railroad tracks) to connect both the study of hatchling predation levels and the observations regarding female nest site selection. This would enable us to look at correlations between predation intensity and nest location. Cameras could also be set on real nests and live hatchlings. This would allow us to consider hatchling movement as another cue for predators.

Our current analyses evaluate mortality (via predation) at a single life stage for this species. Young painted turtles are considered extremely vulnerable within the first few years of life. We could thus examine young mortality in other ways to evaluate the varying predation intensities at different stages and in different environments. Using game cameras, nest predation could be examined to investigate nest predation intensity versus hatchling predation intensity. Game cameras could be set on artificial nests throughout the study site. Hatchling predation could also be investigated in an aquatic environment. Hatchling replicas could be placed on aquatic vegetation on the surface of the pond. Game cameras could then be set to record warm blooded aquatic predators (avian species). An alternative study could be designed to investigate the levels of fish predation on hatchlings. This would enable us to investigate the predation intensity from the time that the hatchling is developing within the egg to the time that it enters the pond. Predation attempts, and therefore mortality rates, may be greater after the hatchling has entered the pond. Both the nesting ecology and our hatchling predation study contribute to the overall understanding of the painted turtles at the Huntsdale Fish Hatchery pond. These data, along with the mark-recapture results will allow us to evaluate the stability of the population over time and its response to environmental fluctuations. Studying the turtles inhabiting this anthropogenic pond provides valuable data that can be compared to that of other populations across North America.

ACKNOWLEDGEMENTS

Thank you to Kevin Wood '11, Elise Rodriguez '11, Nicole Davidson '13, Matt Miller '13, Mary McClintock '11, Bailey Frankel '11, Aaron Gittleman '11, Brendan Gallagher '10, Matt DeStefano '10, and Atandi Anyona '10 field assistance. I would also like to thank Jim Wetherhill at the Huntsdale Fish Hatchery for providing us with site access. Special thanks to Gene Wingert, Mike Potthoff, Greg Howard, and Katie McCann '11 for support. Finally I would like to thank the Dickinson College Center for Sustainability Education and the Department of Biology for funding.

LITERATURE CITED

BAKER, P.J., J.B. IVERSON, R.E.LEE, AND J.L. COSTANZO. 2010. Winter severity and phenology of spring emergence from the nest in freshwater turtles. Naturwissenschaften 97: 607-615.

CHRISTIANSEN, J.L. AND B.J. GALLAWAY. 1984. Raccoon removal, nesting success, and hatchling emergence in Iowa turtles with special reference to *Kinosternon flavescens (Sinosternidae)*. The Southwestern Naturalist 29: 343-348.

CONOVER, M.R., AND J.S. BORGO. 2009. Do sharp-tailed grouse select loafing sites to avoid visual or olfactory predators. Journal of Wildlife Management 73: 242-247.

COSTANZO, J.P., S.A. DINKELACKER, J.B. IVERSON, AND R.E. LEE. 2003. Physiological ecology of overwintering in the hatchling painted turtle: multiple-scale variation in response to environmental stress. Physiological and Biochemical Zoology 77: 74-99.

COSTANZO, J.P., R.L. LEE, G.R.ULTSCH. 2008. Physiological ecology of overwintering in hatchling turtles. Journal of Experimental Zoology 309: 297-379.

ERNST, C.H. 1971. Population dynamics and activity cycles of *Chrysemys picta* in Southeastern Pennsylvania. Journal of Herpetology 5: 151-160.

ERNST, C.H., AND J.E. LOVICH. 2009. Turtles of the United States and Canada (2nd Edition). Baltimore: Hopkins Press.

ETCHBERGER, C.R., M.A, EWERT, B.A. RAPER., AND C.E. NELSON. 1992. Do low incubation temperatures yield females in painted turtles. Canadian Journal of Zoology 70: 391-394.

FRAZER, N.B., J.W. GIBBONS, AND J.L. GREENE. 1990. Growth, survivorship, and longevity of painted turtles *Chrysemys picta* in a southwestern Michigan marsh. American Midland Naturalist 125: 245-258.

GIBBONS, J.W., AND D.H. NELSON. 1978. The evolutionary significance of delayed emergence from the nest site by hatchling turtles. Evolution 32: 297-303.

HULIN, V.,V. DELMAS, M.H. GODFREY, AND J.M. GUILLON. 2009. Temperature-dependent sex determination and global change: are some species at greater risk? Oecologia 160:493-506.

JANZEN, F.J. 1994. Vegetational cover predicts the sex ratio of hatchling turtles in natural nests. Ecology 75:1593-1599.

JANZEN, F.J., AND C.L. MORJAN. 2001. Repeatability of microenvironment-specific nesting behavior in a turtle with environmental sex determination. Animal Behavior 62: 73-82.

KRAUSE, W.J., AND W.A. KRAUSE. 2006. The opossum: its amazing story. Michigan: Self-published.

LEIGHTON, P.A., J.A. HORROCKS, AND D.L. KRAMER. 2010. Conservation and the scarecrow effect: can human activity benefit threatened species by displacing predators? Biological Conservation 143: 2156-2163.

MARCHAND, M.N., J.A. LITVAITIS, T.J. MAIER, AND R.M. DEGRAFF. 2002. Use of artificial nests to investigate predation on freshwater turtle nests. Wildlife Society Bulletin 30:1092-1098.

MARCHAND, NIELSON, C.K., AND M.A. MCCOLLOUGH. 2009. Considerations on the use of remote cameras to detect Canada lynx in northern Maine. Northeastern Naturalist 16: 153 157.

MCGAUGH, S.E., L.E. SCHWANZ, R.M. BOWDEN, J.E. GONZALEZ, AND F.J. JANZEN. 2010. Inheritance of nesting behaviour across natural environmental variation in a turtle with temperature-dependent sex determination. Proceedings of the Royal Society B 277: 1219-1226.

MORJAN, C.L. 2003. Variation in nesting patterns affecting nest temperature in two populations of painted turtles (*Chrysemys picta*) with temperature-dependent sex determination. Behav. Ecol. Sociobiol. 53: 254-261.

ROWCLIFFE, J.M., J. FIELD, S.T. TURVEY, AND C. CARBONE. 2008. Estimating animal density using camera traps without the need for individual recognition. Journal of Applied Ecology 45: 1228-1236.

SCHWANZ, L.E., R.J. SPENCER, R.M. BOWDEN, AND F.J. JANZEN. 2010. Climate and predation dominate juvenile and adult recruitment in a turtle with temperature-dependent sex determination. Ecology 91: 3016-3026.

SPENCER, R.J. 2002. Experimentally testing nest site selection: fitness trade-offs and predation risk in turtles. Ecology 83: 2136-2144.

STEFFEN, J.E. 2009. Perch-height specific predation on tropical lizard clay models: implications for habitat selection in mainland neotropical lizards. Rev. Biol. Trop. 57: 859-864.

STRICKLAND, J., P. COLBERT, AND F.J. JANZEN. 2010. Experimental analysis of the effects of markers and habitat structure on predation of turtle nests. Journal of Herpetology 44: 467-470.

STUART-FOX, D.V., A. MOUSSALLI, N.J. MARSHALL, AND I.P. OWENS. 2003. Conspicuous males suffer higher predation risk: visual modeling and experimental evidence from lizards. Animal Behavior 66: 541-550.

VALENZUELA, N., D.C. ADAMS, R.M BOWDEN, AND A.C. GAUGER. 2004. Geometric morphometric sex estimation for hatchling turtles: A powerful alternative for detecting subtle sexual shape dimorphism. Copeia 2004: 735-742.

WILBUR, H.M. 1975. The evolutionary and mathematical demography of the turtle *Chrysemys picta*. Ecology 56: 64-77.

Table 1. Possible explanations for *Chrysemys picta* overwintering behavior. List developed by Gibbons and Nelson, 1978.

Reason	**Explanation**
Rainfall	"Ground softening" may be necessary for hatchling turtles to emerge from the nest.
Temperature	It is possibly more thermally stable underground. A certain number of warm days are required for development.
Food resources	Food resources may be limited in the fall.
Shelter	Ponds and waterways could potentially be dry in the fall (thus limiting the amount of shelter available).
Multiple clutches	Females lay multiple clutches, and clutches laid late in the nesting season need more time to develop. Overwintering allows all clutches to emerge at the same time (spring).
Predator avoidance	Increased risk of predation if hatchling emerges from the nest.

Table 2. Ten female Painted turtles observed out of the pond between June 12 and August 12. Females were primarily observed during daily visual encounter surveys at the study site. All but one of the females was observed along the railroad tracks.

Date	**Time**	**Site**	**Behavior**	**Processed Nest**	**Idenfified**
12-Jun	1140	Railroad Tracks	Excavating nest	Yes	No
13-Jun	1130	Railroad Tracks	Heading towards pond	No	Yes-985121019899716
22-Jun	0845	Railroad Tracks	Digging nest, abandoned site without laying	No	Marked, tag not read
22-Jun	0855	Railroad Tracks	Digging nest, abandoned site without laying	No	Yes-985121019846306
23-Jun	0828	Pine Road	Crossing road heading towards field	No	Yes-985121019846306
23-Jun	0840	Railroad Tracks	Digging nest, abandoned site without laying	No	Yes-985121019901043
23-Jun	0922	Railroad Tracks	Resting, no nesting behavior observed	No	No
24-Jun	0823	Railroad Tracks	Not digging	No	No
24-Jun	0824	Railroad Tracks	Resting, no nesting behavior observed	No	Yes-985121019846683
6-Jul	1010	Railroad Tracks	Digging nest, abandoned site without laying	No	No
10-Aug	0915	Pond Path	Crossing path from South to North	No	Yes-985121021185137
12-Aug	AM	Pond Parking Lot	Resting, no nesting behavior observed	No	Marked, tag not read

Table 3. Characteristics of Painted turtle nests at the Huntsdale Fish Hatchery Pond study site compared to values found in previous research done in New Mexico, Illinois, Indiana, and Pennsylvania. The mean, standard deviation, range, and sample size is included for each.

	Morjan 2003	**Morjan 2003**	**Baker & Iverson 2010**	**Ernst & Lovich 2009**	**Our Study**
Location	New Mexico	Illinois	Indiana	Pennsylvania	Pennsylvania
Clutch Size	9.9±2.1(6-14)21	10.5±2.0(6-16)106	N/A	4.73(4-6)15	5.3 ±1.3(4-7)4
Nest Depth (cm)	11.2±1.0(9-13)21	8.7±0.8(6-11)106	8.1(4.5-12.5)207	10.4(9.9-11.1)14	5.9±0.97(4.5-6.7)4
Nest Width (cm)	N/A	N/A	N/A	4.5(4.1-5.1)14	4.0±1.71(2.4-5.6)4
Distance to Pond (m)	2.3±2.1(0.7-11.4)34	32.1±24.1(0-86.3)364	N/A	N/A	26.9±29.35(0.4-68.9)4
Canopy Coverage	N/A	N/A	N/A	N/A	79.5±10.8(70-92)4
Slope	N/A	N/A	N/A	N/A	3.9±0.66(3.1-4.7)4

Table 4. Images recorded by game cameras during predator detection periods in fall 2010 and spring 2011 at hatchling replicas. Each observed species is listed along with the total number of images obtained for that species. The proportion of the total number of images of each species is shown. Only images taken of identifiable organisms are included in this analysis.

	Fall		**Spring**	
Species	**Total #**	**Proportion (%)**	**Total #**	**Proportion (%)**
Predator species				
American crow (*Corvus brachyrhynchos*)	0	0	24	4
Northern raccooon (*Procyon lotor*)	0	0	5	1
Red fox (*Vulpes vulpes*)	3	0.2	1	0.2
Striped skunk (*Mephitis mephitis*)	15	1	5	1
Virginia opossum (*Didelphis virginiana*)	33	2	17	3
Total	51	3	52	9
Non-Predator Species				
Black capped-chickadee *(Poecile atricapillus*)	19	1	0	0
Eastern chipmunk (*Tamias striatus*)	61	3	0	0
Eastern cottontail (*Sylvilagus floridanus*)	741	38	22	4
Eastern gray squirrel (*Sciurus carolinensis)*	46	2	0	0
Groundhog (*Marmota monax*)	0	0	217	36
Northern cardinal (*Cardinalis cardinalis*)	103	5	0	0
Tufted titmouse (*Baeolophus atricapillus*)	95	5	0	0
White-footed mouse (*Peromyscus leucopus*)	807	41	0	0
White-tailed deer (*Odocoileus virginianus*)	42	2	306	51
Total	1914	97	545	91

FIGURE LEGENDS

Figure 1. Photographs taken of the summer 2010 nesting ecology study. A. Female Painted turtle laying her clutch, B. Photograph of Lily Bieber-Ham processing a painted turtle nest next to the pond. Five eggs were removed from the nest, massed and measured, C. Example of nest exclosures that were placed on each nest to deter predators and to capture hatchlings upon emergence, C. Hatchling painted turtle that emerged from its nest on September 28, 2010. This was one hatchling from a clutch of seven.

Figure 2. Camera trap study design. A. Comparative photograph of live painted turtle hatchling and hatchling replicas that was used for the predation study, B. Photograph of hatchling replica secured to the ground at the study site with set game camera.

Figure 3. Painted Turtle Nest Sites and Nest Attempts and the Huntsdale Fish Hatchery Pond. Map of study site and all recorded painted turtle nests and nest attempts. Map was made by Lily Bieber-Ham and Elise Rodriquez.

Figure 4. Sample game camera images of predator species taken throughout the fall and spring experimental periods. All photographs were taken at hatchling replicas with scent. A. Predation event showing Virginia opossum with hatchling replica in its mouth (fall), B. Virginia opossum spitting out replica, C. Red fox not observed interacting with replica (fall), D. Potential predation event involving striped skunk (spring).

Figure 5. Sample game camera images of non-predator species taken throughout the fall and spring experimental periods. A. Eastern cottontail (fall), B. Eastern chipmunk (fall), C. Northern cardinal (fall), D. white-tailed deer (spring). The text surrounding each image includes: date, time, photo number compared to total number taken for that trigger, lunar phase, and temperature.

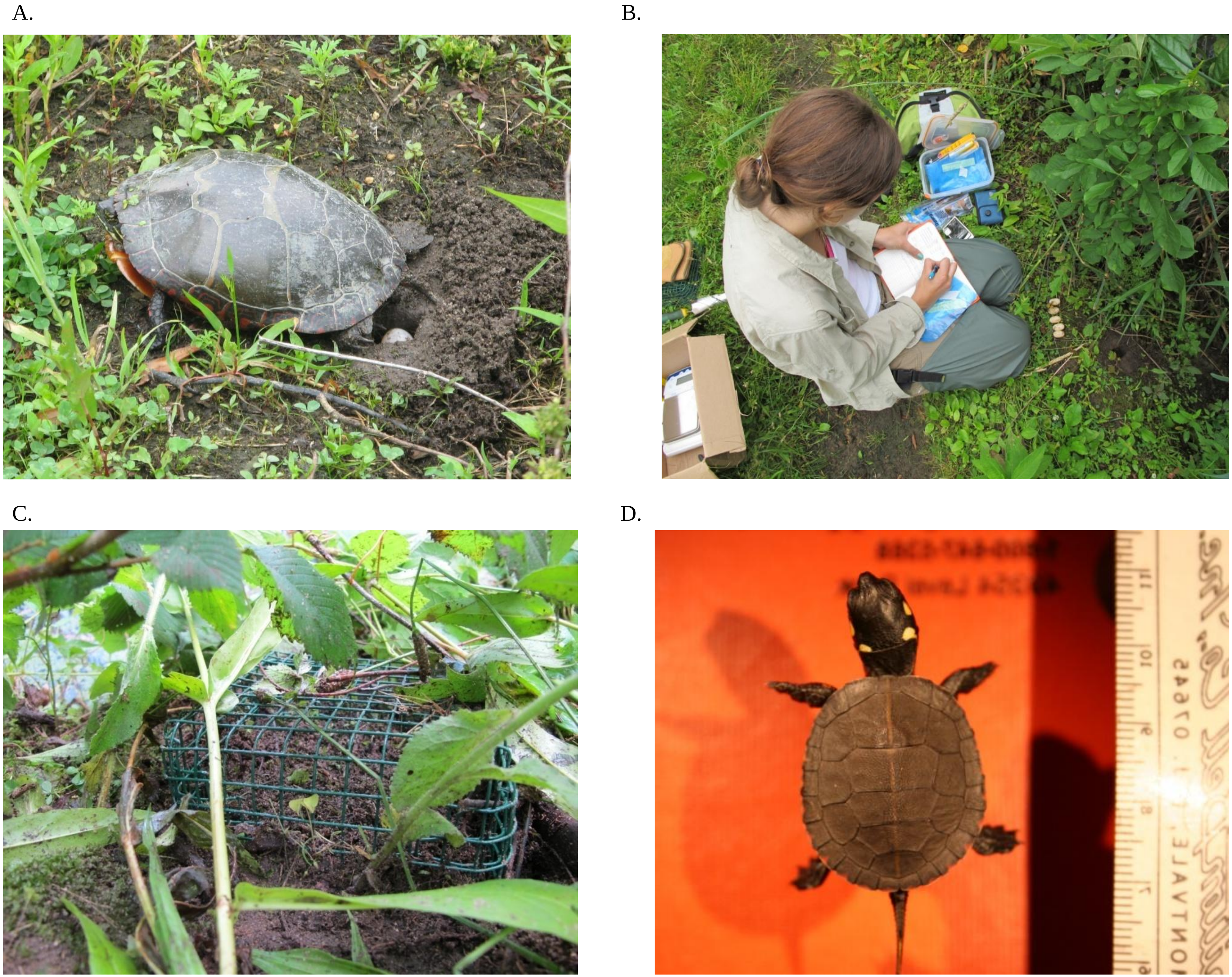
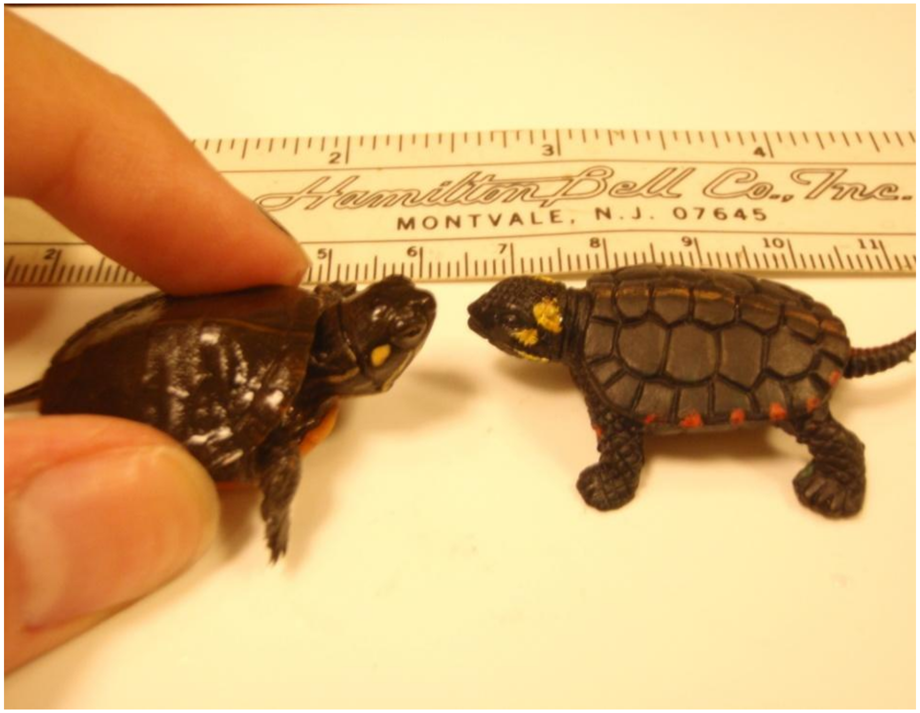


Figure 1.

A.



B.

Figure 2.

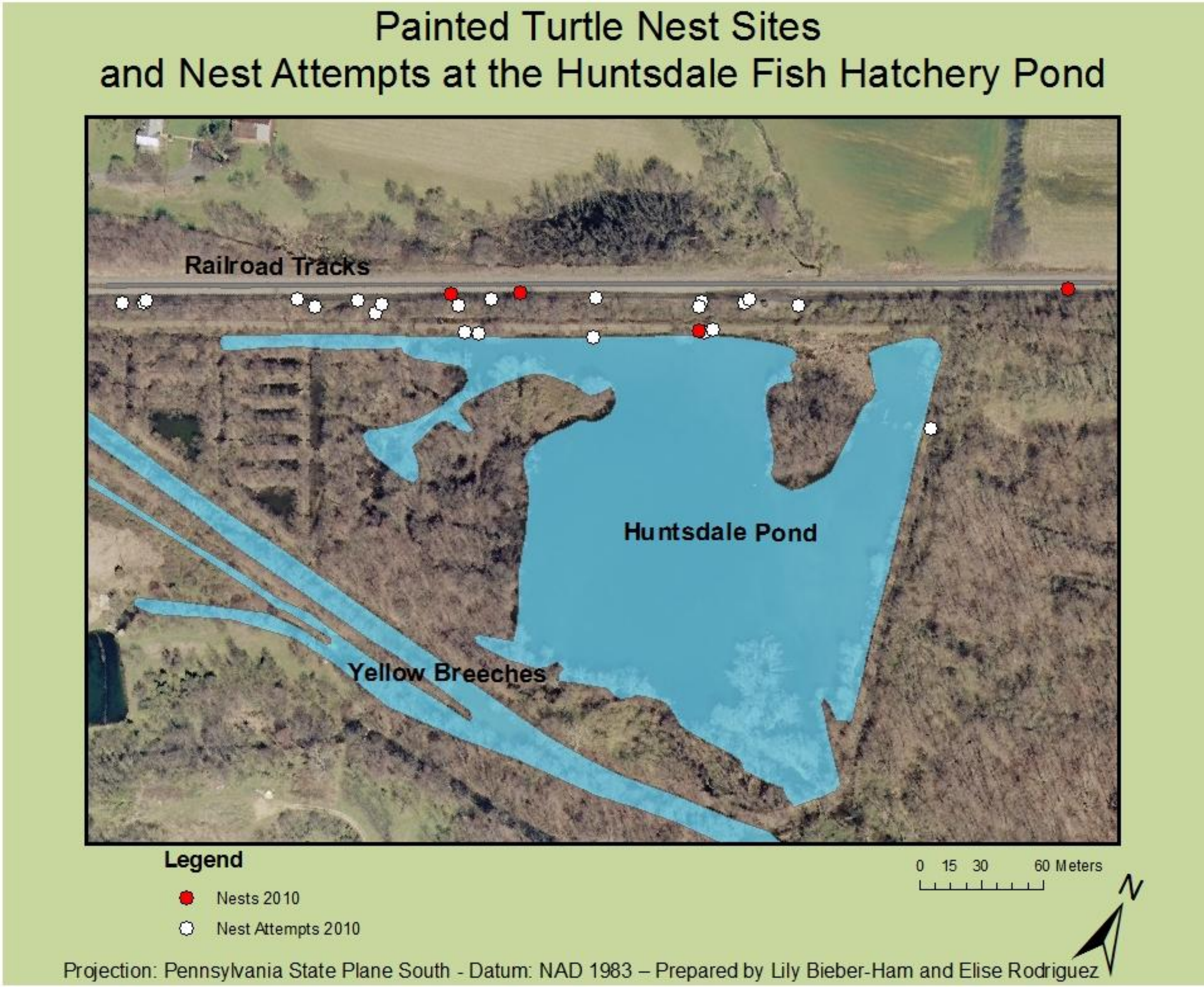

Painted Turtle Nest Sites
and Nest Attempts at the Huntsdale Fish Hatchery Pond
Railroad Tracks
Huntsdale Pond
Yellow Breeches
Legend
Nests 2010
Nest Attempts 2010
0 15 30 60 Meters
N
Projection: Pennsylvania State Plane South - Datum: NAD 1983 – Prepared by Lily Bieber-Ham and Elise Rodriguez

Figure 3.


A.
2010-11-05 3:49:58 AM M 1/5
3°C
PC800 HYPERFIRE PRO
RECONYX
B.
2010-11-05 3:50:00 AM M 4/5
3°C
PC800 HYPERFIRE PRO
RECONYX
C.
2010-11-05 3:25:31 AM M 2/5
3°C
PC800 HYPERFIRE PRO
RECONYX
D.
2011-04-08 12:10:44 AM M 5/5
6°C
PC800 HYPERFIRE PRO
RECONYX

Figure 4.


A.
2010-10-28 6:22:19 AM M 5/5 8°C
PC800 HYPERFIRE PRO
B.
2010-11-01 1:09:49 PM M 5/5 7°C
PC800 HYPERFIRE PRO
C.
2010-11-02 9:52:34 AM M 2/5 -3°C
PC800 HYPERFIRE PRO
D.
2011-04-24 3:20:00 AM M 3/5 14°C
PC800 HYPERFIRE PRO

Figure 5.