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Four-Toed Salamander (hemidactylium Scutatum) Nest Site Characteristics In Natural And Constructed Wetlands In Eastern Kentucky

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FOUR-TOED SALAMANDER (*HEMIDACTYLIUM SCUTATUM*) NEST SITE CHARACTERISTICS IN NATURAL AND CONSTRUCTED WETLANDS IN EASTERN KENTUCKY

By

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By

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DEDICATION

This thesis is dedicated to my dear friend, Sina Barney. Her unwavering support and encouragement through difficult times kept me focused on my goals. And to Hal, who re-sparked my interest in herpetology and inspired me to keep learning!

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Kentucky Fly Fisher's Association: Red Barrington Scholarship, The Kentucky Society of Natural History: Research Grant, and the Department of Biological Sciences at Eastern Kentucky University.

ABSTRACT

Forested freshwater wetlands have undergone loss and alteration more than other types of wetlands. Wetland creation has slowed wetland losses, but many created wetlands do not functionally replace natural wetlands. Plant and animal communities and wetland drying cycles often differ between natural and constructed wetlands. It is important to understand what specific habitat characteristics differ between natural and constructed wetlands and what impact these differences might have on the animal assemblages. Having restrictive habitat requirements makes the four-toed salamander a good candidate for study. The objectives of this study were to understand four-toed salamander (*Hemidactylium scutatum*) nesting ecology and nest-site characteristics and to determine if these differ between natural and constructed wetlands. Another objective was to add to our knowledge of the natural history of the species in Kentucky. Six natural and six constructed wetlands were studied in the Daniel Boone National Forest in Kentucky during 2011. Several nest- and wetland-level variables were measured in each wetland and at each nest site. Data were collected at 207 nests (133 nests in natural wetlands, and 74 nests in constructed wetlands). Multiple regression analyses indicated that four-toed salamander eggs were more abundant in natural wetlands ($P = 0.03$), although there were more eggs per nest in constructed wetlands ($P < 0.001$). There were more nests in wetlands with more moss ($P < 0.001$), and amount

of moss available for nesting was more limited in constructed wetlands. Constructed wetlands were similar in many measured characteristics to those in natural wetlands, and the results underscore the importance of abundant moss and moisture for nesting substrate. However, this study was unable to address embryonic and larval survival in natural and constructed wetlands. In the absence of such data, long-term population monitoring with nest surveys is recommended to determine if this species is impacted by greater predation in constructed wetlands.

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CHAPTER 1

INTRODUCTION

The concern over the draining of wetlands on amphibian populations dates back almost 100 years (Wright 1918). In the United States, forested freshwater wetlands have lost more area than any other type of wetland from 1974–2009 (Dahl 2000, 2011). Wetlands are constructed for wildlife enhancement and to mitigate loss as per the Clean Water Act. However, many ephemeral wetlands are isolated from navigable waters and not jurisdictional, thus regulatory responsibility falls on the states for protection and mitigation (Downing et al. 2003). In Kentucky there is currently no protection for wetlands isolated from jurisdictional waters (Environmental Law Institute 2008).

Because wetlands are created for mitigation or to enhance wildlife populations, it is imperative to understand how well constructed wetlands replicate natural systems. Dahl (2000) reports that between 1986 and 1997 freshwater wetland area increased, and the majority of this increase was due to aquaculture, urban development, and farming. These gains do not necessarily function as ecological equivalents to wetlands being lost (Moreno-Mateos et al. 2012). Dahl (2011), however, reported that there was no net loss (nor any gains) of freshwater wetlands in the U.S. from 2004–2009. Forested freshwater wetlands represent almost 50% of all freshwater wetlands in the U.S., and they are being lost at a faster pace than has been seen in decades; farm ponds and urban wetlands continue to replace acreage of destroyed natural wetlands (Dahl 2011). This method of analyzing wetland loss simply addresses total area and wetland classification; it does not address the biodiversity present. If created wetlands do not function equivalently to natural systems, we will continue to lose critical wildlife habitat and ecosystem services.

Research has shown that the floral and faunal community of created wetlands can differ from that of natural wetlands (Monello and Wright 1999, Zedler and Callaway 1999, Pechmann et al. 2001, Denton 2011, Drayer 2011). Bird and plant communities have been found to fall short of conservation goals in a marsh constructed to comply with mitigation criteria (Zedler and Callaway 1999). Studies have found that created wetlands differ in amphibian community structure from natural wetlands; reasons for this difference include habitat connectivity, hydrology, floral assemblages, and the ability of species to exploit new wetlands (Monello and Wright 1999, Pechmann et al. 2001, Denton 2011, Drayer 2011). Furthermore, pond colonization does not imply reproductive success; some species using wetlands do not reproduce, and other species' embryos and larvae are heavily depredated (Vasconcelos and Calhoun 2006, S. Richter and A. Drayer, unpubl. data).

Focal species should be selected that have more restrictive habitat requirements. This allows the comparison of specific habitat characteristics between different locations. The four-toed salamander (*Hemidactylium scutatum*) is one such species because they have fairly specific habitat needs (Petranka 1998, Beazley and Cardinal 2004, Harris 2005). Ponds similar in habitat characteristics exhibit large differences in four-toed salamander population size; this might be explained if females are selecting certain microhabitat characteristics (Harris 2005). Female four-toed salamanders tend to select steep, north-facing slopes with thick mats of moss for nesting (Chalmers and Loftin 2006, Wahl et al. 2008).

Hydroperiod influences habitat selection in many amphibians. For example, large ranid frogs tend to use permanent bodies of water and have a longer larval period and larger size at metamorphosis than do other amphibians (Wells 2007). Some species use ponds that dry completely, thus avoiding fish predators. However, these species are at risk of desiccation if they are unable to alter the rate of metamorphosis in response to pond drying (Wells 2007). Four-toed salamanders have the shortest larval period of any plethodontid that undergoes metamorphosis (Bruce 2005). Furthermore, the timing of metamorphosis is fixed, suggesting that risk of predation has pushed the timing of metamorphosis to begin as early in development as possible; otherwise metamorphosis would be expected to correlate to food availability and growth rate (O'Laughlin and Harris 2000).

Environmental variables influence amphibian distribution and abundance at wetlands. For example, Eagan and Paton (2004) found that spotted salamanders (*Ambystoma maculatum*) laid more eggs in ponds with greater canopy cover, shrub cover, and more vegetation within the pond. Closed-canopy ponds tend to have lower water temperatures (Werner and Glennemeier 1999, Skelly et al. 2002, Schiesari 2006). Four-toed salamander nest success might be influenced by canopy closure due to its impact on water temperature and other microclimate variables (Werner and Glennemeier 1999, Skelly et al. 2002, Schiesari 2006). Increased acidity can slow embryonic and larval development in amphibians (Dunson and Connell 1982, Sadinski and Dunson 1992). Wahl et al. (2008) found that female four-toed salamanders selected nest sites with a lower pH than unused sites and that embryonic survival was correlated with lower pH at nest sites; however, these pH levels were higher than those reported to slow development in other species (Dunson and Connell 1982, Sadinski and Dunson 1992).

Even though four-toed salamanders have a broad distribution, the species is not without need for management. Closer examination of this species' patchy distribution reveals specific microhabitat needs that vary over their lifetime. Hydrology, cover, and substrate influence the distribution and survival of four-toed salamander embryos and larvae (Chalmers and Loftin 2006, Wahl et al. 2008). Oviposition sites can affect reproductive fitness in amphibians by influencing both embryonic and larval success (Resetarits and Wilbur 1989).

Created wetlands often have different hydrological characteristics and different amphibian communities than do natural wetlands (Pechmann et al. 2001, Denton 2011, Drayer 2011). Managing for habitat requirements for all life stages is essential to successful management of amphibian species. Because wetlands are created to offset habitat loss, it is critical to understand how characteristics of the terrestrial and aquatic habitat differ between natural and constructed wetlands, and how this impacts four-toed salamanders' nest site characteristics and nesting ecology.

Study Objectives:

The objectives of this study were to compare nesting ecology and nest site characteristics of four-toed salamanders inhabiting natural and constructed wetlands in the Daniel Boone National Forest (DBNF), KY. Another objective was to determine which habitat characteristics were important in explaining the number of eggs in a nest, and the number and placement of eggs in a wetland. Natural history data are lacking for four-toed salamanders in Kentucky; and this study will document the nesting period and record frequency of communal nesting and nest attendance.

CHAPTER 2

METHODS

In the Daniel Boone National Forest, KY, wetlands have been created on ridge-tops for wildlife management and conservation (Brown and Richter 2012). Study sites were selected from a system of natural and constructed ridge-top wetlands including all wetlands where four-toed salamanders had previously been documented (Drayer 2012, Denton 2012). Wetlands were searched to locate all four-toed salamander nests present. An array of nest-site and wetland characteristics were measured and analyzed in order to compare four-toed salamander nest site characteristics between natural and constructed wetlands and determine which characteristics were important to the nesting ecology of the species.

Study Species:

Four-toed salamanders belong to the largest family of salamanders (Plethodontidae) but are taxonomically isolated because their sub-family (Hemidactylinae) is monotypic (Vitt and Caldwell 2009). The overall conservation status of four-toed salamanders is unclear, but populations range from locally rare to abundant throughout their range in the eastern United States (Harris 2005). In Kentucky, the distribution of these salamanders is discontinuous, and some populations are disjunct. Although there are no recent data on the distribution and status of four-toed salamanders in Kentucky, populations are considered stable (KCWCS 2010). The IUCN Red List status for four-toed salamanders is Least Concern (IUCN 2010).

Four-toed salamander courtship occurs in the fall or winter and primarily at night, and involves the tail-straddle walk (Branin 1935). Females lay eggs in a solitary or communal nest during the following spring and oviposit only once per breeding season (Harris and Gill 1980, Harris and Ludwig 2004). Female four-toed salamanders may tend their nests, but if nesting communally only one female typically remains to brood the nest (Blanchard 1923, Harris and Ludwig 2004, Harris 2005). Four-toed salamanders remain philopatric to a pond once nesting has commenced (Harris and Ludwig 2004). When the embryos hatch, larvae enter the pond to complete their development, and upon metamorphosis become forest dwelling (Harris 2005).

Four-toed salamander migrations to nesting ponds begin in mid-February in lowland Virginia and in April in montane Virginia (Harris 2005). In the Daniel Boone National Forest, KY, four-toed salamander nests have been observed in early March (A. Drayer, pers. comm.). The embryonic period generally lasts 5–6 weeks (Blanchard 1923, Harris and Ludwig 2004, Chalmers and Loftin 2006) with a 3–6 week larval period (Blanchard 1923, Harris et al. 1995, Harris 2005).

Study Sites:

Selection of study sites began with nine natural and nine human-constructed wetlands in the Morehead Ranger District of the Daniel Boone National Forest, KY, where four-toed salamanders have been detected previously (Denton 2011, Drayer 2011). All wetlands were fishless and hydrologically isolated on forested ridge tops. The area encompassing all the wetlands exists on the unglaciated portions of the Western Allegheny Plateau Ecoregion and is dominated by mixed mesophytic forest (Woods et al. 2002). Further description of the study wetlands can be found in Drayer (2011) and Denton (2011).

Starting with the nine natural and nine constructed wetlands, wetlands were excluded if no nests were found, were considerably smaller than the other wetlands, or were recently disturbed. This resulted in six natural and six constructed wetlands being selected as study sites (Table 1, Figure 1).

Table 1. Natural and constructed wetlands in the Daniel Boone National Forest, KY, where four-toed salamander (*Hemidactylium scutatum*) nest site characteristics were studied during 2011.

Natural Wetlands	**Abbreviation**	**Constructed Wetlands**	**Abbreviation**
Booth Pond	Booth	35-97	35-97
Dark Cave 2	DC 2	Elk Lick Artificial	ELA
Dark Cave 6	DC 6	High Energy Artificial	HEA
Elk Lick Natural	ELN	Jones Ridge Artificial	JRA
High Energy Natural	HEN	Long Ridge Artificial	LRA
Jones Ridge Natural	JRN	Pond 5	P5

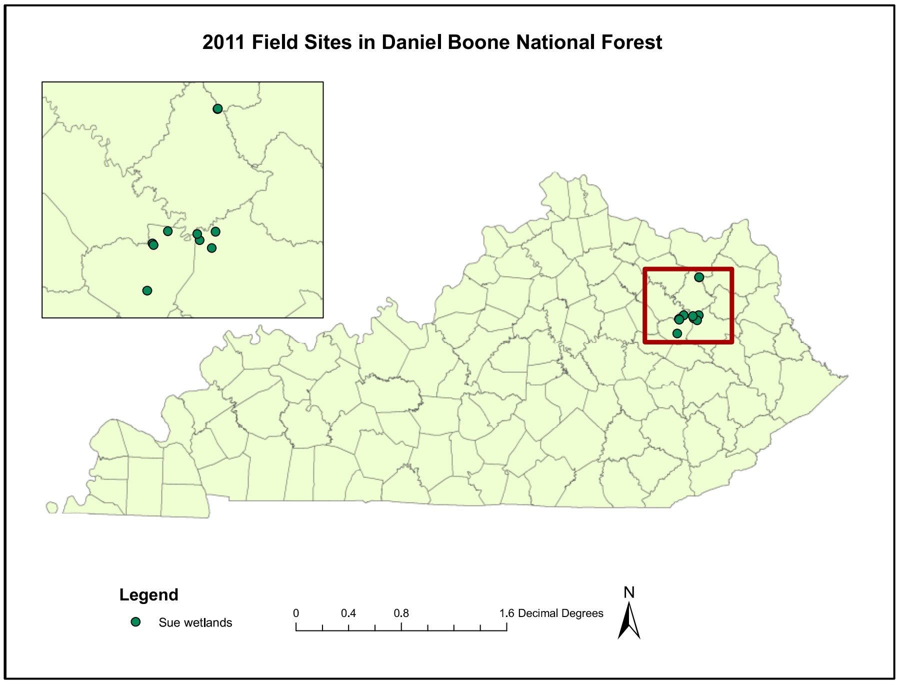

Figure 1. Map of wetlands in the Daniel Boone National Forest, KY, where four-toed salamander (*Hemidactylium scutatum*) nest sites were studied. Inset is close-up of area encompassing study sites. Four pairs of wetlands (one natural pair and three pairs of both wetland types) are overlapping on the map because of geographic proximity, but actually represent eight wetlands (DC2/DC6, ELN/ELA, HEN/HEA, JRN/JRA; Table 1).

Data Collection:

The shoreline of each wetland was searched from the edge to 1 m past the high-water line to locate four-toed salamander nests. This distance was selected because most nests in previous studies were found within 20 cm of the water (Blanchard 1923, Chalmers and Loftin 2006, Wahl et al. 2008). Each wetland was searched every two weeks during the nesting period beginning the first week in March and continuing until no viable embryos remained (12 June). Each nest was marked and numbered with a flag placed on vegetation several feet behind the nest to prevent double counting and attraction of predators. To be certain the same nest was monitored on return visits the distance and direction of the nest from the marker was recorded.

During each visit, the number of four-toed salamander eggs present was counted. Because distinguishing separate clutches was not possible, a liberal and conservative estimate was made of the number of clutches per nest. The liberal estimate was calculated post-hoc using a maximum of 45 eggs per clutch because there was a break in the data at this point, and it was very close to the 40 eggs per clutch limit used by Gilbert (1941), Harris and Gill (1980), and Chalmers (2004). The conservative estimate was made using 65 eggs per clutch as per Harris et al. (1995), Chalmers (2004), and Corser and Dodd (2004). So, for the liberal estimate, if a nest had 1–45 eggs it was considered 1 clutch, 46–90 eggs was 2 clutches, 91–135 eggs was 3 clutches, etc.; and for the conservative estimate, 1–65 eggs was considered 1 clutch, 66–130 eggs was 2 clutches, etc. Other authors (Harris and Gill 1980, Banning et al. 2008) were able to differentiate clutches in an egg mass based on stage of development, but this could only be discerned for one nest in this study.

The number of female four-toed salamanders present on the nest was noted at each visit. Because some females lay eggs and leave the nest while others remain on the nest, nest attendance is defined as the presence of a female on the nest after 28 March. This date represents the day after the last occurrence of multiple females on the same nest; there was a break point in the data before which more than 50% of the attended nests had ≥ 2 females present, but after which none of the attended nests had more than one female present. Sixty percent of the nests had been established at this point. Female presence was defined as the maximum number of females observed on a given nest.

When each four-toed salamander nest was found, aspect (the direction toward the water the slope faced where a nest was located) was measured with a compass to the nearest degree. A map of the nest's location in the wetland was hand drawn. To define nest placement for each nest, the map was divided into twelve 30° sections centered on north, and the number of nests in each section was counted. A small plot (10 cm^2) centered on the nest site was used to determine the percent ground cover by plant type (moss, grass, sedge, and herbaceous plants) and non-living material (decaying wood, leaf litter, pine needles, soil, gravel, dead vegetation). The slope from the nest to the water line was measured (using a clinometer, accuracy to 1°). Soil pH (Kelway soil acidity and moisture tester, model HB-2; Kel Instruments Company, Inc., Wyckoff, NJ) was measured to the nearest 0.1 when each nest was initially found. When no soil was present at a nest, soil pH was measured in the nearest soil within 1 m in any direction; if there was no soil within that distance a measurement was not taken.

During each visit to a nest, soil moisture was measured as volumetric water content (Fieldscout TDR 100 Soil Moisture Meter; Spectrum Technologies, Inc., Plainfield, IL) in standard mode; this is the ratio of water volume to total soil volume. Sampling locations were determined following the procedure outlined for soil pH. Moisture measurements from weeks 6 and 7 (14–24 April) were used in the analyses because this represented the shortest span of time that all wetlands were visited. Straight-line distance from the center of the nest to the nearest water was measured at each visit (to nearest mm). The original distances to water were used in the analyses because subsequent distances were correlated to the original distance.

After hatching, larvae were captured by dipnetting using a standardized effort of five sweeps per 25 m^2 of wetland area with a minimum of five sweeps per wetland, sampling all areas equally (Jung et al. 2002, Werner et al. 2007, Shulse et al. 2010). Dipnet sampling was continued until a minimum of ten consecutive sweeps yielded no additional larvae captured. At most wetlands fewer than 3 larvae were captured, so dipnetting was discontinued. A trial run of bottle traps baited with glowsticks (Grayson and Roe 2007) resulted in no larvae trapped, so the traps were not deployed in the remaining wetlands. Because few larval four-toed salamanders were captured, determining larval survival in natural and constructed wetlands was not pursued.

After full leaf-out, beginning 15 May, canopy closure at each study site was determined using a spherical densiometer; measurements were taken in the center of each wetland and at the perimeter of the wetland in the cardinal and ordinal directions (Skelly et al. 2002, Schiesari 2006). The nine canopy closure readings taken at each wetland were averaged to determine a single canopy closure measurement per wetland. The length and width of each wetland was measured to estimate surface area, and the perimeter was recorded (to the nearest cm). The total amount of moss in each wetland was determined by measuring the length of any moss clump intersecting the perimeter of the wetland, measuring the longest line transecting every clump of moss in the interior of a wetland, and then combining these measurements. Linear moss density was calculated by dividing the total moss in a wetland by the perimeter of the wetland. Wetlands were scored using the Ohio Rapid Assessment Method (ORAM)(Mack 2001) and Kentucky Wetland Rapid Assessment Method (KYWRAM)(D. Brown and S. Richter, pers. comm.).

Equipment and waders were disinfected with Nolvasan between wetlands following recommendations of Green et al. (2010). Approval from the Eastern Kentucky University Institutional Animal Care and Use Committee (IACUC) was obtained prior to beginning this project (protocol number 04-2011).

Statistical Analyses:

Multiple stepwise regressions were performed in SAS version 9.2 (SAS Institute Inc., Cary, NC) using a negative binomial distribution to determine which factors influence the number of eggs and nests in a wetland and to determine the relationship between the number of eggs in a nest and environmental factors. The first model included the number of eggs in a nest as the response variable and type of wetland, amount of moss in a wetland, and aspect, substrate, slope, moisture, distance to water, pH, and number of females present as explanatory variables. A second model had number of eggs in a wetland as the response variable and wetland type, total moss in a wetland, canopy closure in a wetland, and distance to nearest wetland (< 200 meters) as explanatory variables. The final model had number of nests in a wetland as the response variable and type of wetland, total moss in a wetland, distance to nearest wetland, and canopy closure in a wetland as explanatory variables.

Program R, version 2.14.2, package 'vegan' (The R Foundation for Statistical Computing, Vienna, Austria) was used to perform multivariate analyses to determine how nest characteristics vary among wetlands and by type. Several Principle Component Analyses (PCAs) were conducted using nest-level data because initial analyses with all data indicated the results were being driven by wetland-level variables. The first PCA plot revealed one wetland was quite distinct from the remaining wetlands, so nests from this wetland (Booth Pond) were removed and the analysis repeated. As stated previously, not every nest has pH and soil moisture data, but PCA requires there to be no missing data. Because adding moisture and pH data did not add much to the explanatory ability of the analysis, these variables were excluded, rather than excluding nests with missing data (n = 19). Neither pH nor moisture had a major influence on the first two axes. Only the PCA of nest-level variables, before and after removing nests from Booth Pond are presented and discussed.

Program Orianna (version 3.21, Kovach Computing Services, Anglesey, Wales, UK) was used to analyze aspect and nest placement in a wetland. Because aspect and nest placement was measured in degrees, circular statistics are needed for analyses of these data (Zar 2010). Rayleigh's Test was used to determine if the distribution of nest placement was uniform around each wetland, and if the aspect of each nest was uniform. To determine if the number of wetlands with a clumped or random distribution of nest aspect and nest placement varied between wetland types the results of Rayleigh's test were used. First, if Rayleigh's test indicated a clumped distribution (i.e. $P < 0.05$) the wetland was assigned a score of '1', if Rayleigh's test indicated a random distribution of nests ($P > 0.05$) the wetland was assigned a score of '0'. Then, Chi-square contingency table analyses were used to compare the number of wetlands with a random distribution of nest aspect and nest placement in the wetland.

To determine if use of 100% moss for nesting substrate varied between wetland types, Chi-square contingency table analyses were used. An independent samples t-test was used to determine if the variables measured in each wetland differed between wetland types. If the variance of the data was unequal, then test statistics were reported based on Welch's adjusted degrees of freedom. Nested ANOVAs with individual wetlands nested within wetland type were run on the nest-level variables. Data for distance, slope, pH, and moisture were log-transformed; and egg and clutch count data were square root transformed because the data failed Levene's test of equal variance. The transformed data for slope were the only data to pass Levene's test of equal variance. The remaining ANOVA results are reported even though the variances of the data are unequal. Mean values ± 1 standard error are reported. Descriptive statistics, analyses of variance (ANOVA), t-tests, and chi-square tests were run using SPSS 18.0 (IBM Statistical Package for the Social Sciences, Armonk, NY). Only results indicating significant differences are reported here.

CHAPTER 3

RESULTS

A total of 207 four-toed salamander nests were discovered and monitored in 12 wetlands: 133 nests in natural wetlands, and 74 nests in constructed wetlands. Of the 207 nests monitored, females attended 56 nests (27.1 ± 0.03%). In natural wetlands 39 of 133 (29 ± 0.04%) nests were attended, whereas 17 of 74 (23 ± 0.05%) nests were attended in constructed wetlands. Egg laying began during the week of 7 March. The greatest number of new nests detected in a week (n = 74) was during the week of 21 March (week 3). By the end of week 6 (ending 17 April) the vast majority (85%) of eggs had been deposited. Many nest-site characteristics were similar between wetland types, but there were more eggs found in natural wetlands and more clutches in the same nest in constructed wetlands.

Factors Explaining the Number of Eggs in a Nest:

The overall model addressing numbers of four-toed salamander eggs in a nest was significant ($F_{6,130} = 8.3$, $P < 0.001$, $R^2 = 0.276$) and included wetland type ($F_{1,130} = 13.1$, $P < 0.001$), moisture ($F_{1,130} = 9.5$, $P = 0.003$), and number of females present ($F_{4,130} = 7.5$, P = < 0.001). There were more eggs per nest in constructed wetlands (77.2 ± 7.0) than in natural wetlands (52.8 ± 3.0)(Figure 2). The conservative estimate for the number of clutches per nest ranged from 1–6 clutches, and the liberal estimate was 1–8 clutches per nest. Using the conservative estimate, there was a mean of 1.4 ± 0.1 clutches per nest in constructed wetlands and a mean of 1.2 ± 0.1 clutches per nest in natural wetlands (Table 2). There was a positive relationship between the number of eggs per nest and both the number of females present (Figure 3) and moisture (Figure 4). The number of females present per nest ranged from 0 to 4: 0.71 ± 0.07 females present at nests in natural wetlands and 0.70 ± 0.10 females present at nests in constructed wetlands. The mean percent moisture at nests was 27.55 ± 0.50%: 28.88 ± 0.76% in natural wetlands and 25.90 ± 0.53% in constructed wetlands (Figure 5). Also, there was an inverse relationship between the number of eggs per nest and the total moss in a wetland (Figure 6).

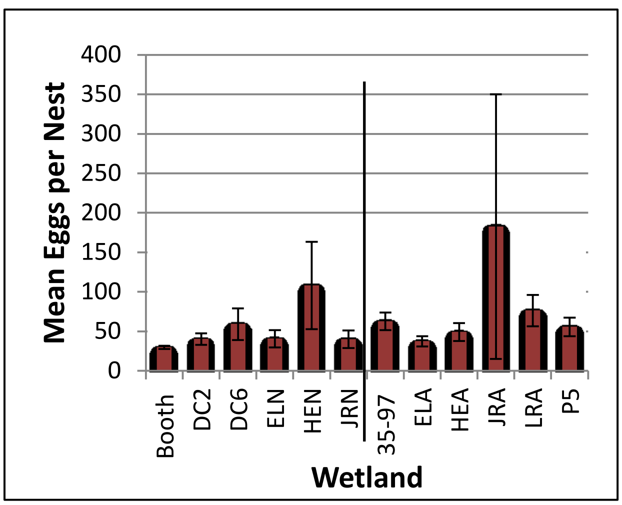

Figure 2. Mean (± 1 SE) number of four-toed salamander (*Hemidactylium scutatum*) eggs per nest in 12 wetlands in the Daniel Boone National Forest, KY. The vertical line separates the first six wetlands, which are natural, from the next six, which are constructed.

Table 2. Counts of eggs, clutches, and nests of four-toed salamanders (*Hemidactylium scutatum*) at natural and constructed wetlands in the Daniel Boone National Forest, KY.

Wetland	**Type**	**Nests (n)**	**Mean eggs per nest ± 1 SE**	**Mean clutches per nest ± 1 SE**
Booth	Natural	89	29.5 ± 2.0	1.1 ± 0.0
DC 2	Natural	15	40.0 ± 7.3	1.1 ± 0.1
DC 6	Natural	10	59.0 ± 20.2	1.6 ± 0.3
ELN	Natural	8	40.6 ± 11.1	1.3 ± 0.2
HEN	Natural	4	108.0 ± 55.4	2.3 ± 0.8
JRN	Natural	7	39.9 ± 11.1	1.3 ± 0.2
35-97	Constructed	24	62.5 ± 11.1	1.5 ± 0.2
ELA	Constructed	5	37.2 ± 6.5	1.0 ± 0.0
HEA	Constructed	17	48.9 ± 11.3	1.2 ± 0.2
JRA	Constructed	2	182.5 ± 167.5	3.5 ± 2.5
LRA	Constructed	10	76.2 ± 19.9	1.5 ± 0.3
P5	Constructed	16	55.7 ± 11.8	1.4 ± 0.2

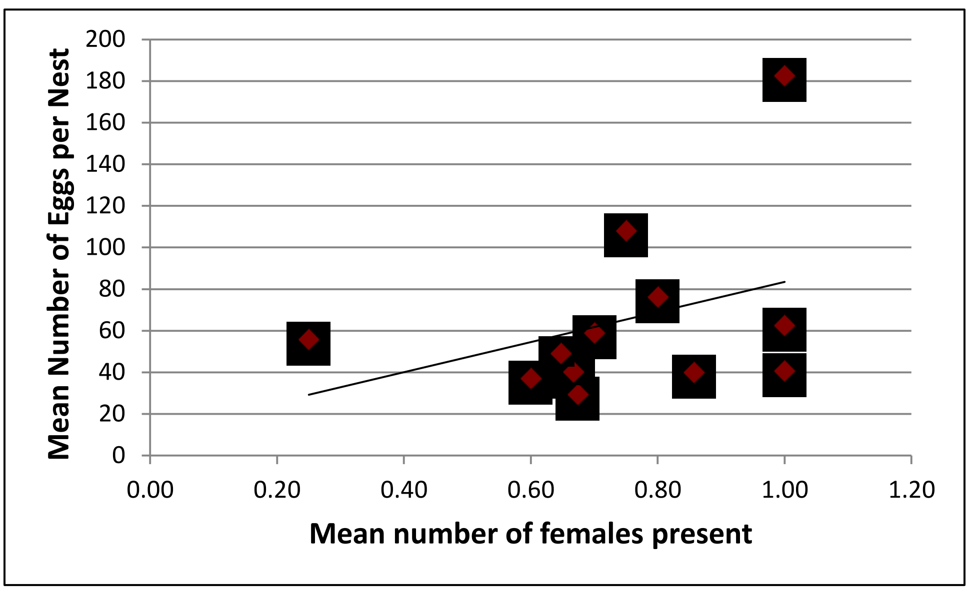

Figure 3. Relationship between the mean number of four-toed salamander (*Hemidactylium scutatum*) eggs in a nest and the mean number of females present in a nest at 12 wetlands in the Daniel Boone National Forest, KY.

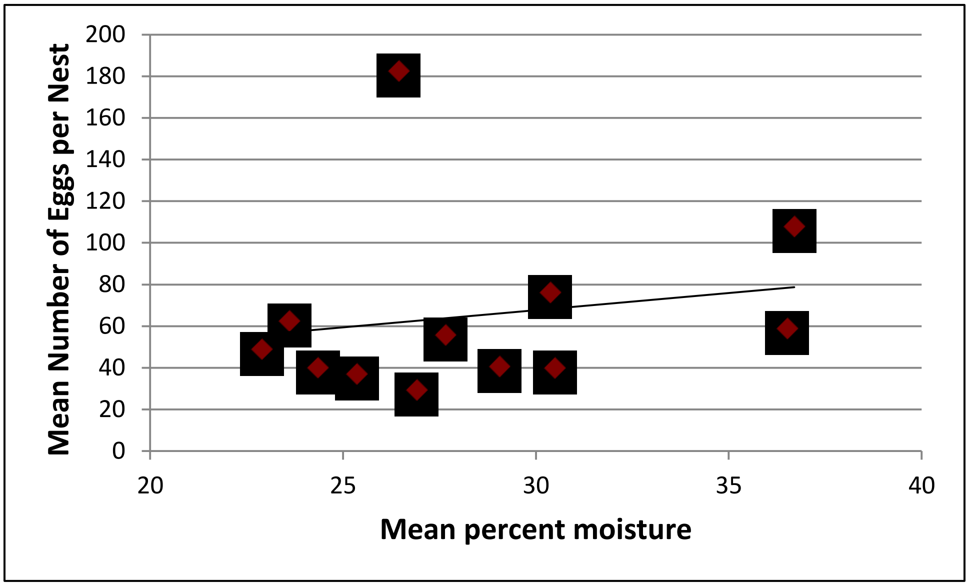

Figure 4. Relationship between the mean number of four-toed salamander (*Hemidactylium scutatum*) eggs in a nest and the mean moisture in a nest at 12 wetlands in the Daniel Boone National Forest, KY.

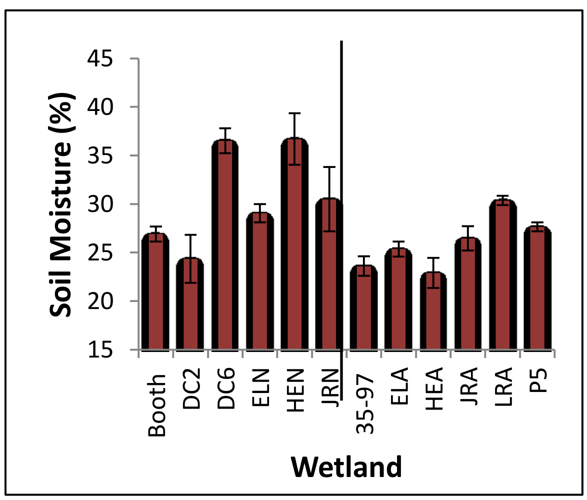

Figure 5. Mean (± 1 SE) percent moisture at four-toed salamander (*Hemidactylium scutatum*) nests sites in 12 wetlands in the Daniel Boone National Forest, KY. The vertical line separates the first six wetlands, which are natural, from the next six, which are constructed.

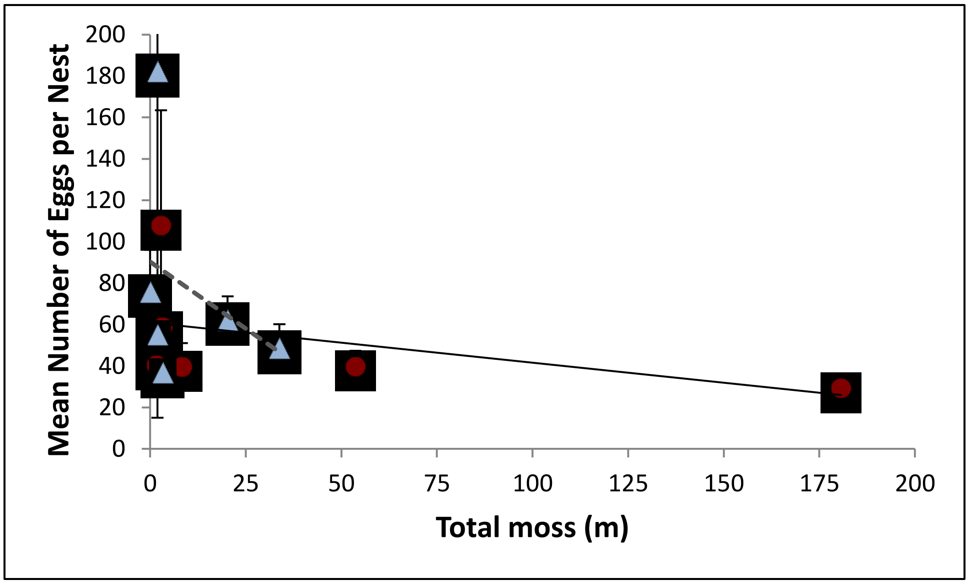

Figure 6. Relationship between the mean (± 1 SE) number of four-toed salamander (*Hemidactylium scutatum*) eggs per nest and the total amount of moss in a wetland at 12 wetlands in the Daniel Boone National Forest, KY.
Natural = Circle, solid line   Constructed = Triangle, dashed line

Factors Explaining the Number of Eggs and Nests in a Wetland:

The overall model explaining the number of four-toed salamander eggs in a wetland was significant ($F_{3,8}$ = 16.9, P < 0.001, R^2 = 0.864) and included wetland type ($F_{1,8}$ = 7.4, P = 0.026), canopy closure ($F_{1,8}$ = 4.1, P = 0.08), and total moss ($F_{1,8}$ = 48.4, P < 0.001). Natural (808.3 ± 367.1) wetlands had more total eggs than constructed (756.2 ± 187.7) wetlands. The mean density of eggs was greater in constructed (16.7 ± 3.7 eggs/meter) than natural (7.7 ± 1.1 eggs/meter) wetlands. There was a positive relationship between total number of eggs in a wetland and both total moss (Figure 7) and canopy closure (Figure 8). Canopy closure was greater in natural wetlands (96.1 ± 1.5%) than in constructed wetlands (84.7 ± 4.6%). The total amount of moss was 41.68 ± 28.97 m in natural wetlands, and 10.20 ± 13.76 m in constructed wetlands. The linear moss density was similar between natural (26.3 ± 13.4%) and constructed (17.2 ± 8.3%) wetlands. The mean perimeter of natural wetlands (94.0 ± 26.6 m) was greater than constructed wetlands (46.1 ± 5.9 m), and the mean area of natural wetlands (383.0 ± 194.0 m^2) was greater than constructed wetlands (138.6 ± 49.3 m^2)(Table 3).

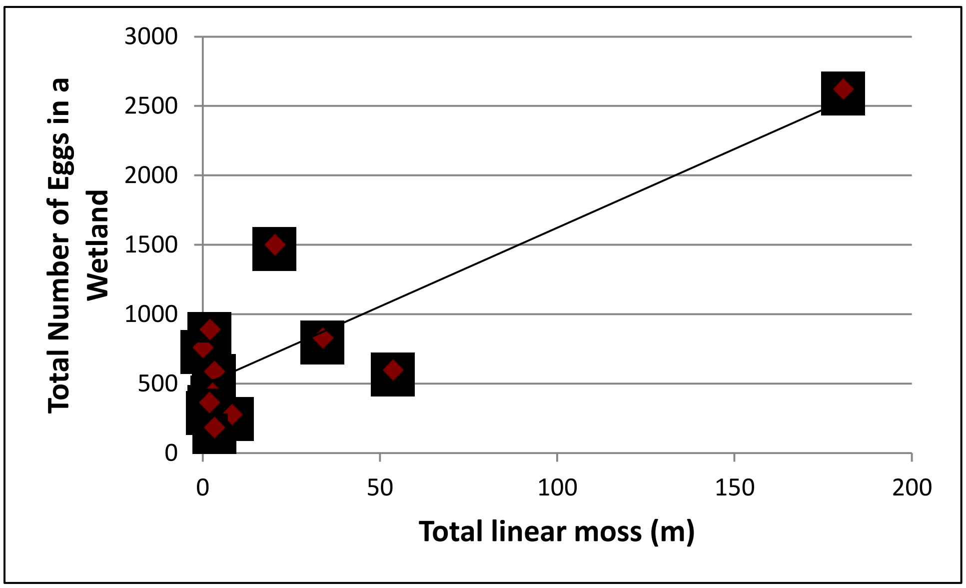

Figure 7. Relationship between the total number of four-toed salamander (*Hemidactylium scutatum*) eggs in a wetland and the total linear moss in 12 wetlands in the Daniel Boone National Forest, KY.

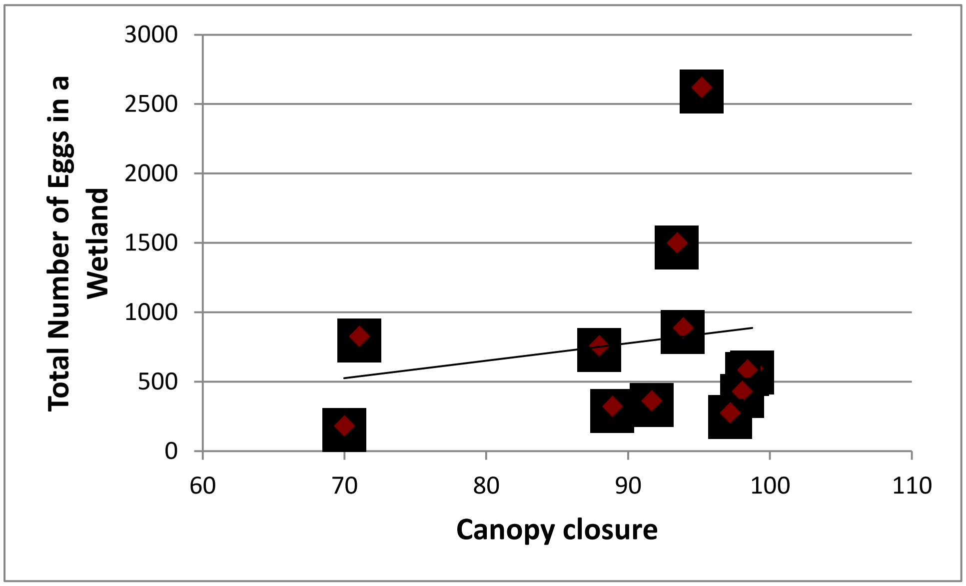

Figure 8. Relationship between the total number of four-toed salamander (*Hemidactylium scutatum*) eggs in a wetland and the canopy closure of 12 wetlands in the Daniel Boone National Forest, KY.

Table 3. Size of wetlands and amount of moss present in 12 wetlands in the Daniel Boone National Forest, KY where four-toed salamander (*Hemidactylium scutatum*) nest site characteristics were studied. Percent moss was calculated by dividing total moss by perimeter.

Wetland	Type	Area (m^2)	Perimeter (m)	Total moss (m)	% moss
Booth	Natural	1281.70	208.10	180.65	86.81
DC2	Natural	547.93	135.40	53.65	39.62
DC6	Natural	198.64	72.30	3.20	4.43
ELN	Natural	87.92	47.00	1.50	3.19
HEN	Natural	126.45	58.00	2.80	4.83
JRN	Natural	55.54	43.45	8.30	19.10
35-97	Constructed	194.04	54.35	20.25	37.26
ELA	Constructed	90.43	47.10	3.30	7.01
HEA	Constructed	360.68	70.00	33.80	48.29
JRA	Constructed	66.98	31.70	1.90	5.99
LRA	Constructed	41.23	33.10	0.00	0.00
P5	Constructed	70.05	40.50	1.95	4.81

The number of nests in a wetland was best explained by the total amount of moss in a wetland ($F_{1,10}$ = 116.7, P < 0.001, R^2 = 0.921)(Tables 2 and 3). Natural wetlands had more nests (22.2 ± 13.5) than constructed wetlands (12.3 ± 3.4). There was a positive relationship between the number of nests in a wetland and the amount of moss in that wetland (Figure 9), when the extreme value was removed the relationship was not as strong (Figure 10).

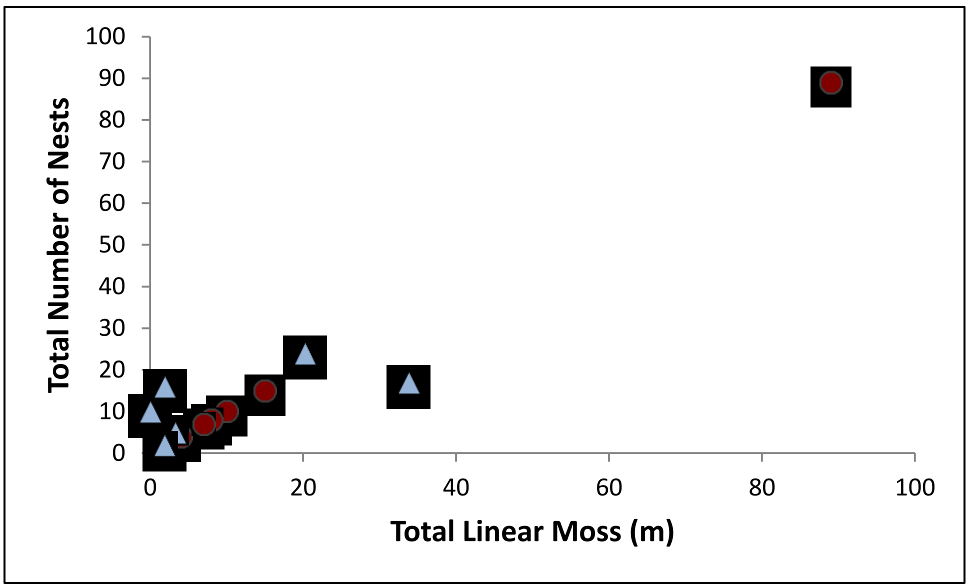

Figure 9. Relationship between the number of four-toed salamander (*Hemidactylium scutatum*) nests and the total amount of moss in 12 wetlands in the Daniel Boone National Forest, KY.
Natural = Circle Constructed = Triangle

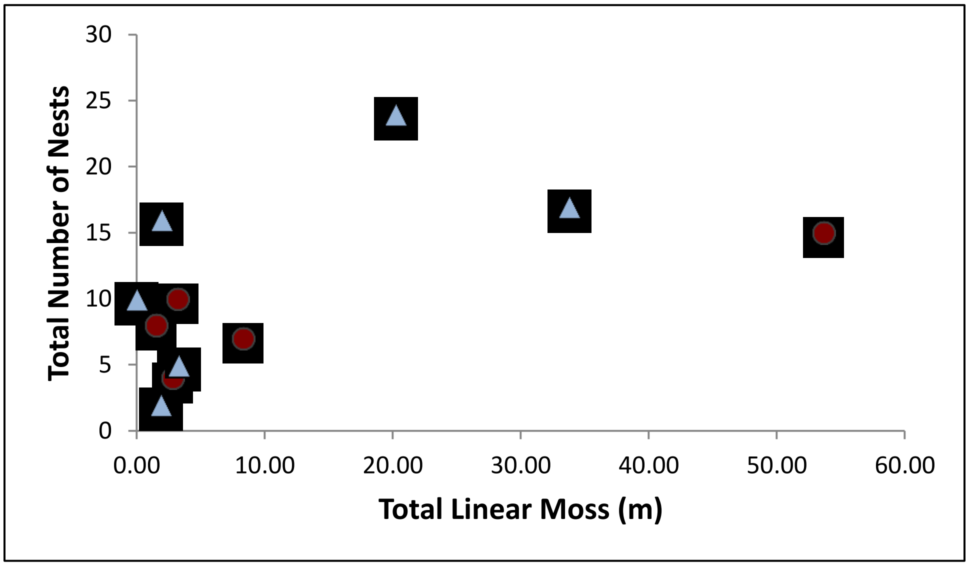

Figure 10. Relationship between the number of four-toed salamander (*Hemidactylium scutatum*) nests and the total amount of moss after extreme value (Booth Pond) is removed, in 11 wetlands in the Daniel Boone National Forest, KY. Natural = Circle   Constructed = Triangle

Other Nest Site Characteristics:

The PCA of four-toed salamander nest site characteristics in 12 wetlands in the DBNF captured 33.1% of the variation on the first axis. The distance of eggs to the water, slope, and percent moss substrate were the most important variables on the first axis (Table 4). Axis two captured 21.6% of the variation; the number of females present, number of eggs per nest, and percent moss substrate were the most important variables (Table 4). When the nests from Booth Pond were removed the analysis captured slightly less variation on the first two axes, but the most important variables explaining variation were the same as in the first analysis (Table 4, Figures 11 and 12).

Table 4. Principle Component Analysis (PCA) of nest-level variables at four-toed salamander (*Hemidactylium scutatum*) nest sites in 12 wetlands in the Daniel Boone National Forest, KY (top) and after removing nests from Booth Pond (bottom). The site scores are listed for the first three axes (proportion of variation explained).

Variable	**PC1 (0.331)**	**PC2 (0.216)**	**PC3 (0.166)**
Eggs per nest	-1.112	1.433	-0.757
Aspect	-0.464	0.380	2.285
Moss substrate	1.749	0.923	0.082
Slope	1.794	-0.073	-0.062
Distance	-1.940	-0.449	-0.103
Females present	-0.283	2.070	0.043
Variable	**PC1 (0.283)**	**PC2 (0.242)**	**PC3 (0.167)**
Eggs per nest	-0.591	1.302	-1.154
Aspect	-0.834	-0.357	1.647
Moss substrate	1.364	1.033	0.441
Slope	1.417	-0.220	-0.225
Distance	-1.506	-0.587	-0.316
Females present	-0.521	1.759	0.127

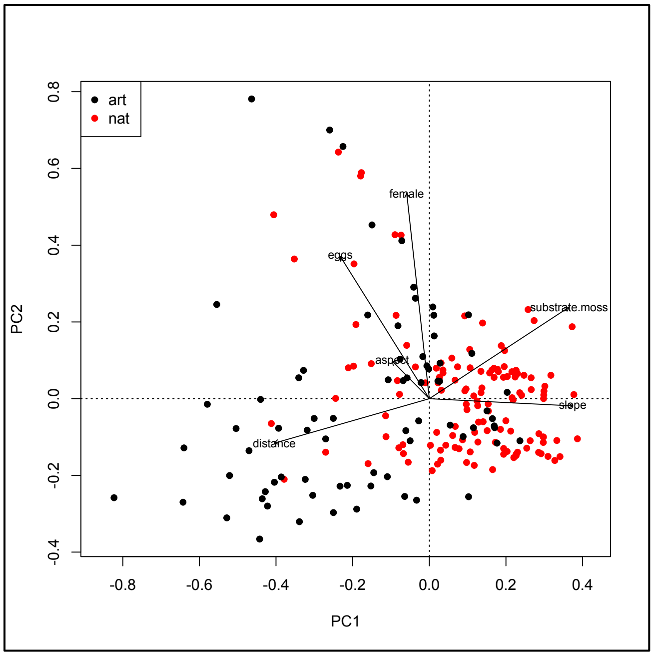

Figure 11. PCA plot of four-toed salamander (*Hemidactylium scutatum*) nest-level variables for 205 nest sites in 12 wetlands in the Daniel Boone National Forest, KY.
Natural wetlands = nat Constructed wetlands = art

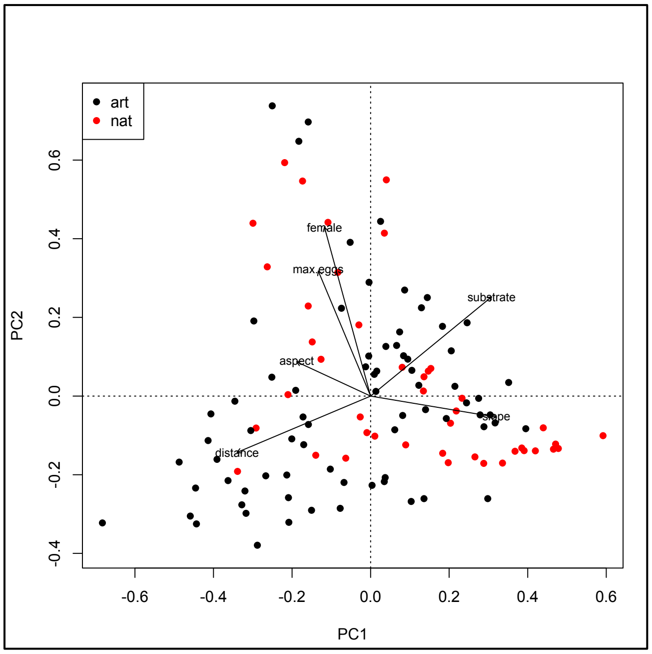

Figure 12. PCA plots of four-toed salamander (*Hemidactylium scutatum*) nest-level variables for 117 nest sites in 11 wetlands in the Daniel Boone National Forest, KY, after removing nest sites from Booth Pond.
Natural wetlands = nat Constructed wetlands = art

The mean distance of eggs to water was 11.48 ± 0.60 cm; with shorter distances in natural wetlands (9.17 ± 0.52 cm) than in constructed wetlands (15.59 ± 1.26)(Figure 13). The range of slopes used for nesting was 0–90°, and the mean slope was 46.7 ± 1.5°: 52 ± 2.0° in natural wetlands and 36.5 ± 2.4° in constructed wetlands (Figure 14). Moss was the most commonly used nesting substrate: 78% of nests were found in areas with 100% moss and 82% were found in areas of > 50% moss. There was one constructed wetland (Long Ridge), which had no moss, and nests were found primarily in leaf litter. Use of 100% moss for nesting was higher in natural wetlands ($X^2 = 41.9$, df = 1, $P < 0.001$): 122 of 133 (92%) nests in natural wetlands and 39 of 74 (53%) in constructed wetlands (Figure 15).

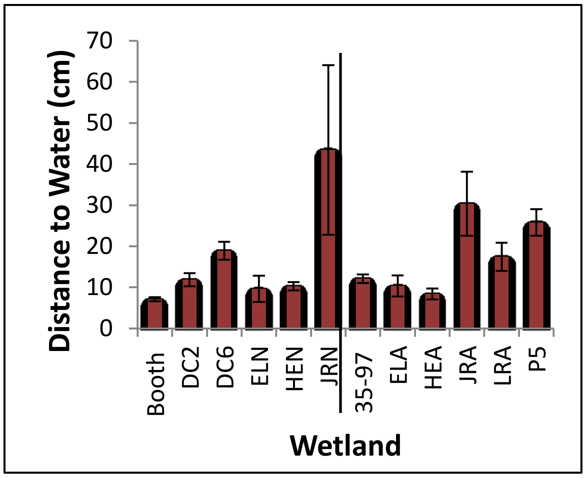

Figure 13. Mean (± 1 SE) distance of four-toed salamander (*Hemidactylium scutatum*) nests from water in 12 wetlands in the Daniel Boone National Forest, KY. The vertical line separates the first six wetlands, which are natural, from the next six, which are constructed.

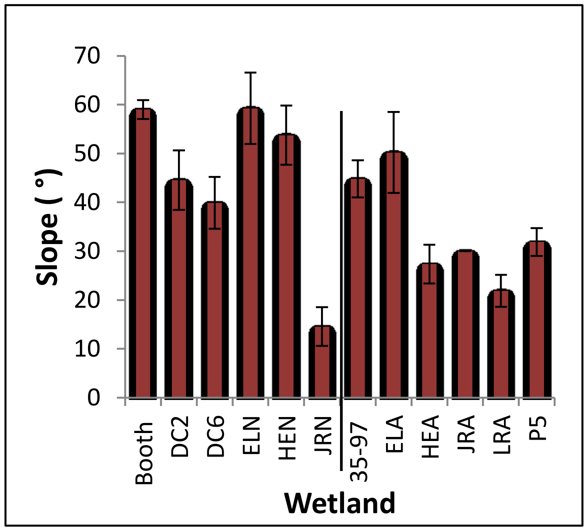

Figure 14. Mean (± 1 SE) slope under four-toed salamander (*Hemidactylium scutatum*) nests sites in 12 wetlands in the Daniel Boone National Forest, KY. The vertical line separates the first six wetlands, which are natural, from the next six, which are constructed.

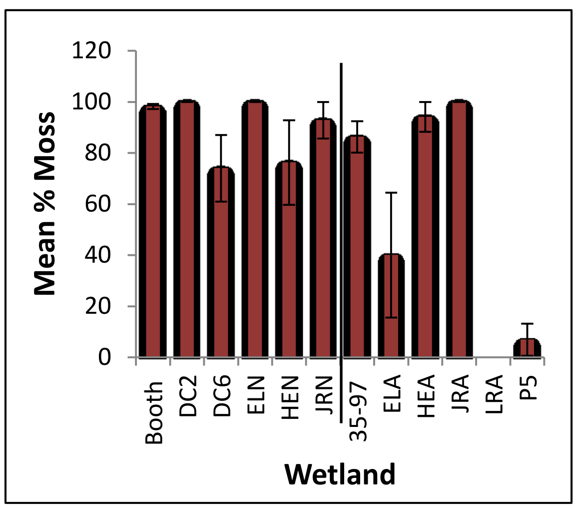

Figure 15. Mean (± 1 SE) amount of moss substrate at four-toed salamander (*Hemidactylium scutatum*) nests sites in 12 wetlands in the Daniel Boone National Forest, KY. The vertical line separates the first six wetlands, which are natural, from the next six, which are constructed.

Soil pH at the nest site was statistically different among wetlands ($F_{10,163}$ = 2.5, P= 0.008) and between wetland types ($F_{1,15.8}$ = 8.7, P= 0.01)(Figure 10). The soil pH averaged 5.98 ± 0.04: 5.78 ± 0.05 in natural wetlands and 6.27 ± 0.03 in constructed wetlands (Figure 16).

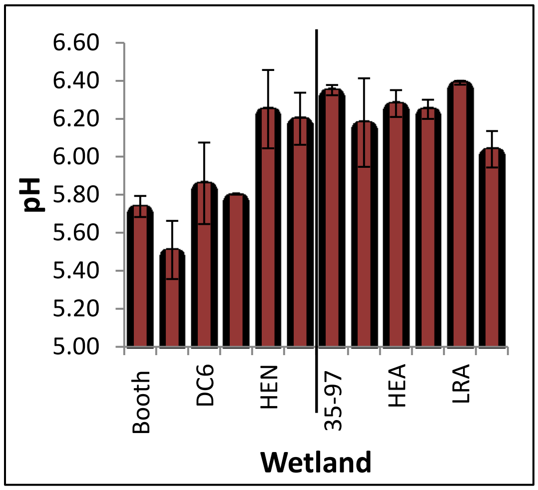

Figure 16. Mean (± 1 SE) pH at four-toed salamander (*Hemidactylium scutatum*) nests sites in 12 wetlands in the Daniel Boone National Forest, KY. The vertical line separates the first six wetlands, which are natural, from the next six, which are constructed. Natural wetlands had a lower pH than constructed wetlands (P = 0.01).

Some wetlands had a clumped distribution and some had a random distribution of nest aspect and nest placement in 30° sections (Tables 5 and 6). Many nests in natural wetlands faced north but this pattern was not apparent in constructed wetlands (Figure 17). The placement of nests in a wetland was widely scattered in both types of wetlands (Figure 18). The number of wetlands with a clumped or random distribution of nest aspect did not vary by wetland type (X_1^2 =

0.3, P=0.56). There also was no difference between wetland type in clumped or random distribution of nest placement in 30° sections (X_1^2 = 0.8, P=0.38).

Table 5. Analysis of aspect of nests of four-toed salamanders (*Hemidactylium scutatum*) around wetlands in the Daniel Boone National Forest, KY.

Wetland	**Type**	**Rayleigh Test (Z)**	**Rayleigh Test (p)**	**Mean Vector (μ)**	**Circular Standard Deviation**
Booth	Natural	7.85	<0.001	348.45	89.27
DC2	Natural	2.48	0.082	16.67	76.90
DC6	Natural	1.89	0.152	31.88	73.97
ELN	Natural	3.26	0.033	14.87	54.30
HEN	Natural	0.78	0.488	317.89	73.21
JRN	Natural	3.36	0.028	334.80	49.08
35-97	Constructed	4.42	0.011	337.09	74.53
ELA	Constructed	0.06	0.948	344.43	120.69
HEA	Constructed	2.60	0.073	173.35	78.55
JRA	Constructed	0.13	0.901	80.00	94.20
LRA	Constructed	1.62	0.202	215.03	77.32
P5	Constructed	3.57	0.025	263.92	70.16

Table 6. Analysis of four-toed salamander (*Hemidactylium scutatum*) nest placement in 30° sections of around wetlands in the Daniel Boone National Forest, KY. Values for JRN could not be calculated.

Wetland	Type	Rayleigh Test (Z)	Rayleigh Test (p)	Mean Vector (μ)	Circular Standard Deviation
Booth	Natural	4.20	0.010	74.64	99.93
DC2	Natural	0.93	0.402	350.10	95.58
DC6	Natural	1.42	0.247	218.17	80.03
ELN	Natural	4.92	0.004	212.48	39.96
HEN	Natural	0.88	0.442	322.91	70.40
JRN	Natural	1.30	0.284	155.10	74.41
35-97	Constructed	7.34	<0.001	125.39	62.37
ELA	Constructed	0.31	0.755	263.79	95.70
HEA	Constructed	5.40	0.003	13.85	61.36
JRA	Constructed	---	---	---	---
LRA	Constructed	2.52	0.077	118.45	67.24
P5	Constructed	4.49	0.009	48.07	64.60

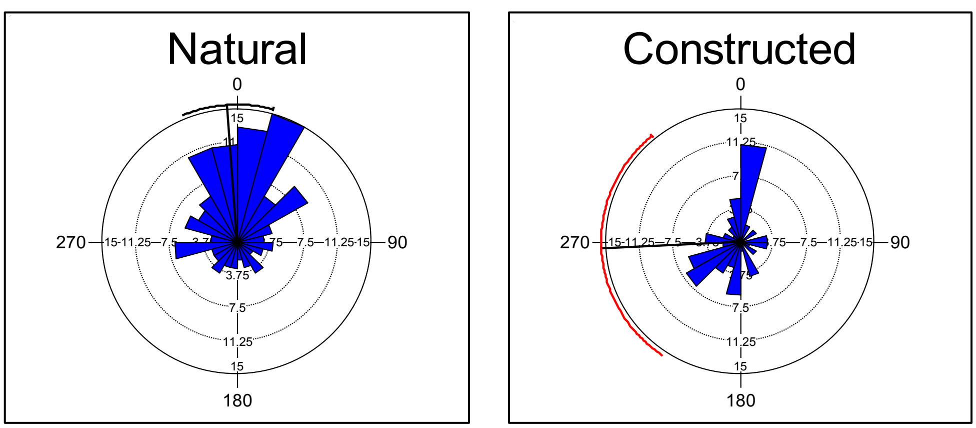

Figure 17. Circular histogram of four-toed salamander (*Hemidactylium scutatum*) nest aspect from 207 nests in 12 wetlands in the Daniel Boone National Forest, KY.

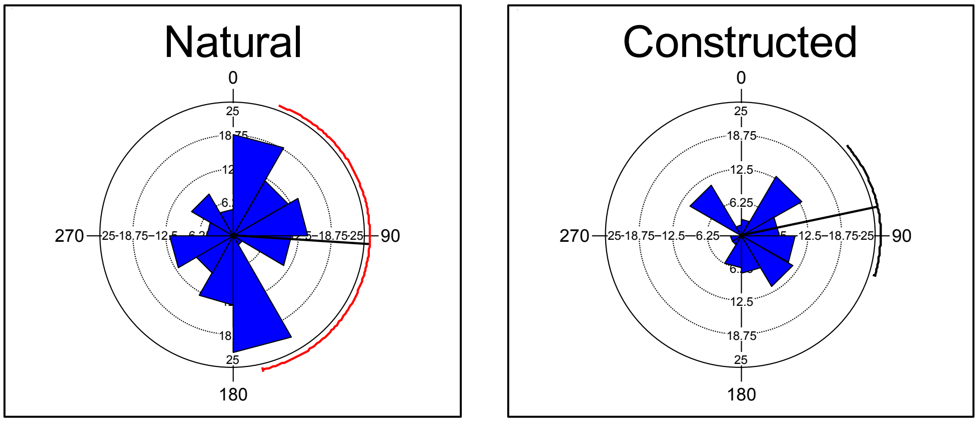

Figure 18. Circular histogram of four-toed salamander (*Hemidactylium scutatum*) nest placement in 30° sections around 12 wetlands in the Daniel Boone National Forest, KY.

Natural wetlands scored higher than constructed wetlands using KYWRAM (T_{10} = 4.5, P = 0.001) and ORAM (T_{10} = 5.1, P < 0.001). Natural wetlands scored an average of 72.5 ± 3.6 with KYWRAM and 70.7 ± 1.5 using ORAM. Constructed wetlands scored an average of 47.7 ± 4.3 using KYWRAM and 59.4 ± 1.6 using ORAM.

CHAPTER 4

DISCUSSION

This study indicated that natural wetlands provide better nesting opportunities for four-toed salamanders, as indicated by the trend toward more eggs and nests in natural wetlands. The greater number of clutches and eggs per nest in constructed wetlands, as indicated by multiple regression analysis, could be related to the decreased availability of moss for nesting in these wetlands. Also, more eggs were found in wetlands with greater canopy closure and more total moss, characteristics that were found primarily in natural wetlands. Constructed wetlands provide nest sites for four-toed salamanders that are similar to natural wetlands in many characteristics; however, quality nest sites with thick beds of moss, on steep slopes, and with greater moisture are limited in quantity in constructed wetlands. An effort should be made to construct wetlands with more trees and downed woody debris in the wetland, and to upturn clumps of soil to increase microhabitat heterogeneity. Embryonic and larval survival was unable to be determined, and in the absence of these data, long-term population monitoring using nest surveys is recommended.

Factors Explaining the Number of Eggs in a Nest:

The multiple regression analyses indicated that wetland type was an important factor explaining the number of four-toed salamander eggs in a nest, with more eggs (i.e., more clutches) per nest in constructed wetlands. There was an inverse relationship between the number of eggs per nest and the total amount of moss in a wetland (Figure 6). The total amount of moss and density of moss was greater in natural wetlands (Table 3). The number of nests in a wetland was positively and significantly associated with total moss (Figure 9). This indicates there was a greater need for joint nesting in wetlands with less total moss. Females had to choose between sharing a nest in moss or nesting in a different substrate. Chalmers and Loftin (2006) concluded that four-toed salamander distribution is limited by the availability of suitable nesting habitat, and all the nests in their study were found in moss.

Habitat saturation has been postulated as a reason for joint nesting seen with four-toed salamanders (Harris et al. 1995). By manipulating the density of gravid females in constructed wetlands and mesocosms, Harris et al. (1995) concluded that joint nesting was not due to habitat saturation because it occurred even at low densities. However, as in this study, there were more clutches per nest at higher densities (Harris et al. 1995). Harris et al. (1995) did not address density and nesting behavior between different types of wetlands. In this study, four-toed salamanders nest in 100% moss more often in natural wetlands than constructed wetlands. Nests in constructed wetlands often were found in smaller clumps of moss and spread onto other substrates. Twenty-two of 28 nests found in four natural wetlands with < 10 m total moss (three of which had < 5 m moss) were found in 100% moss; but in four constructed wetlands with < 5 m moss only 5 of 33 nests were found in 100% moss (Tables 2 and 3). The lack of large clumps of moss in constructed wetlands required females to nest jointly.

Other research indicated joint nesting was related to body condition (Harris 2008). Females in poorer body condition were more likely to dump eggs in a nest to be attended by a female in better body condition, and females in intermediate to high body condition were more likely to nest alone and remain with their eggs (Harris 2008). It has also been shown that frequency of reproduction depended on foraging opportunities (after brooding and before winter), which were influenced by rainfall (Harris and Ludwig 2004). I did not measure body size or weight or upland habitat variables, so I cannot address the possibility that the number of clutches per nest might be related to foraging opportunities or the body condition of the females. The possibility that quality nest sites and quality foraging opportunities are correlated could not be addressed either, but this raises an interesting question about the ability of a constructed wetland to provide conditions similar to those of natural wetlands. Joint nesting occurred at greater frequency in constructed wetlands, and the data indicated this was due to a limited amount of moss for nesting. Increased rate of joint nesting in constructed wetlands could be due to poorer body condition of the females, but this study did not address body condition.

Female presence (Figure 3) and moisture (Figure 4) were also positively and significantly associated with number of four-toed salamander eggs in a nest. When more females lay eggs in a nest there is a greater chance that a female will be seen on the nest. Other studies indicated moisture was important to nest sites (Chalmers and Loftin 2006, Wahl et al. 2008). Chalmers and Loftin (2006) and Wahl et al. (2008) stated that more moss at nest sites might help maintain moisture at nest sites and provide structure that allows the salamander to easily enter the site and remain concealed.

Factors Explaining the Number of Eggs and Nests in a Wetland:

The multiple regression analyses indicated wetland type was one factor explaining the number of four-toed salamander eggs in a wetland. Total moss explained the number of four-toed salamander eggs (Figure 7) and nests (Figure 9) in a wetland. Chalmers and Loftin (2006) found that the presence of woody debris was a predictor of nesting within a wetland, and proposed this was due to the likelihood of such debris to be colonized by thick mats of moss, as nests were located on stumps, logs, root balls, and natural and artificial earthen banks covered in thick mats of moss. In this study, there were more eggs and nests in wetlands with more total moss. There was also a trend toward more nests and eggs in natural wetlands.

This indicates that natural wetlands provide better nesting habitat than constructed wetlands, due to the increased availability of moss for nesting. It is also possible that four-toed salamander abundance in constructed wetlands is lower due to some unmeasured habitat variables.

Harris and Ludwig (2004) reported that female four-toed salamanders skip reproduction after years with poor foraging opportunities. As previously mentioned, my study did not address female body condition or foraging opportunities. However, the wetland assessment methods employed in this study do incorporate upland habitat characteristics into the scoring metrics, and natural wetlands scored higher than constructed wetlands examined in this study. Denton (2011) also reported higher ORAM scores for natural wetlands compared to constructed wetlands. While individual metrics were not compared, it seems that the differences in wetland assessment scores are due to the greater habitat interspersion and microtopographic features and the amount of quality buffer surrounding natural wetlands (see Denton 2011), and the greater presence of invasive plant species found in constructed wetlands. As mentioned previously, the quality of the upland habitat might influence foraging opportunities, and thus the nesting behavior of females the following season (Harris and Ludwig 2004).

In this study, canopy closure also explained the number of eggs in a wetland. There were more eggs in wetlands with greater canopy closure (Figure 8). Canopy closure and moss amount and density was greater in natural wetlands. Canopy closure has previously been shown to be lower in constructed wetlands in this system (Denton 2011, Drayer 2011). This reflects construction activities, as construction typically requires removal of trees or building in openings, and soil compaction due to construction inhibits the colonization of plants (Biebighauser 2011, Drayer 2011, Brown and Richter 2012). Canopy closure in a wetland influences the amphibian community and microhabitat characteristics (Werner and Glennemeier 1999, Skelly et al. 2002, Eagan and Paton 2004, Schiesari 2006). Increased canopy closure also increases rate of evapotranspiration, which can influence a wetland's drying cycle (Lott and Hunt 2001).

Other Nest Site Characteristics:

The PCA on four-toed salamander nest-level characteristics indicated that differences in nest-site characteristics were driven primarily by the amount of moss substrate, steepness of slopes, and distance to the water (Figure 10). Booth Pond, which contained two-thirds of all the natural nests, had many trees in the interior of the wetland that were covered by thick mats of moss or forming small islands, as well as many moss clump 'islets' in the interior of the wetland. Because these moss mats grew into and below the level of the water there were a large number of nests in Booth Pond that were very close to the water. Nests were found on average 13 cm from the shore, similar to what has been reported by other researchers (Blanchard 1923, Gilbert 1941). Nests were closer to the water in natural wetlands.

Chalmers and Loftin (2006) found steep slope was one of the components in the best model predicting presence of four-toed salamander nests at the microhabitat scale. Similar to the findings of Chalmers (2004), nests in this study also had very steep slopes due to the common use of moss-covered trees and moss clump 'islets' for nesting in natural wetlands. The nests on moss clump 'islets' had water directly beneath them and thus many slopes were near 90°. In constructed wetlands, many nests were located directly on the bank of the wetland. If the slopes of the bank of the two types of wetlands were compared, the natural wetlands have a lower angle of slope. In Virginia, Wahl et al. (2008) found nests on slopes that were steeper than unused sites. Steep slopes may aid larvae in entering the wetland after hatching (Chalmers and Loftin 2006). I found many embryos in nests on moss 'islets' in natural wetlands so hatched larvae were able to drop directly into the water.

Many nest-site characteristics varied among nests, but did not differ between wetlands or wetland types. The number of females present, number of eggs per nest, and percent moss substrate explained most of the variation in nest data (Table 4). Female four-toed salamanders attended roughly 25% of the nests in this study, with no difference in either female presence or attendance between wetlands types. Chalmers (2004) reported females in Maine attended over 80% of nests, and Breitenbach (1982) reports 56% of nests in Michigan were attended. In these studies most wetlands were only visited once, so there was no way of knowing how long the females remained on the nest or if geographic variation in attendance behavior exists. I found females on 47% of the nests over the field season, so my attendance estimates might be lower based on my definition of attendance; i.e. females staying on the nest after the 'egg dumpers' had left the wetland. Harris and Gill (1980) reported that brooding eggs increased embryonic survival in four-toed salamanders, but there was no correlation between the length of brooding and embryonic survival.

Female four-toed salamanders have been documented to nest in acidic wetlands (Chalmers and Loftin 2006, Wahl et al. 2008) and in this study, the pH was lower at four-toed salamander nest sites in natural wetlands than at nest sites in constructed wetlands. In Maine, nests have been found in swamps and less acidic fens, but not fens or bogs that were more acidic (Chalmers and Loftin 2006); however, the wetlands in Maine were more acidic overall than the wetlands in Kentucky. In the Wahl et al. (2008) study, females nested in sites with a lower pH (5.3 at nest sites vs. 5.6 in unused sites). Drayer (2011) reported pH in natural wetlands in the DBNF was lower than in constructed wetlands.

Aspect and nest placement within a wetland were clumped in some wetlands and not in others and this was similar in both natural and constructed wetlands (Tables 5 and 6). There was a trend in this study toward more northerly facing nests in natural wetlands versus constructed wetlands (Figure 15), however this relationship was not examined with statistical analyses. Wahl et al. (2008) found nests primarily face north to northeast; these nests remained cooler and tended to be found in more moss than more southerly facing nests.

Other Considerations:

One objective of this study was to compare four-toed salamander embryonic and larval survival between wetland types, but these were not able to be determined. Due to unusually heavy rainfall during April of this study, many nests were inundated and some eggs were washed away and were unable to be located. Some embryos survived inundation, while others did not. Due to high levels of uncertainty, embryonic survival data were not analyzed and length of embryonic period was not determined. Although survival could not be accurately determined in this study, Petranka (1998) reports approximately 10–20% of embryos remain viable after inundation (Petranka 1998). Larval survival was unable to be addressed due to the low success rate of capture.

The ability to avoid predators is important to the success of four-toed salamanders. Even though this study was unable to quantify embryonic and larval survival, some assumptions can be made based on previous work with four-toed salamanders at other sites and other species within my study system. Four-toed salamander brooding females do not defend their nests from predators (Carreño and Harris 1998), but eggs are considered unpalatable to carabid beetles, centipedes, and eastern newts (*Notophthalamus viridescens*) due to the low rate of predation compared to predation rates on eggs of other species of plethodontid salamanders (Carreño and Harris 1998, Hess and Harris 2000). On just one occasion, I witnessed a centipede (taxon unknown) on a nest (no embryos were missing), and I never detected beetles at a nest. However, two nests were located immediately next to an entrance to a single ant colony, and both nests later disappeared, presumably due to the ants. Representative ants were collected and identified to the highest taxonomic resolution possible by the School of Ants in the Department of Biological Sciences at the North Carolina State University. One sample was identified to the genus *Myrmica*; the other was identified as *Tapinoma sessile*, the odorous house ant. Five nests were assumed depredated by raccoons, coyotes, or opossums as nests were discovered missing and the moss substrate had been torn with claw marks noted on the underlying tree roots.

Previous research documented that four-toed salamander larvae decreased activity levels, hid in the substrate, and grew more slowly in the presence of a predator, eastern newts, which readily consume four-toed salamander larvae (Carreno et al. 1996, Wells and Harris 2001, Harris et al. 2003). Because four-toed salamander larvae never grow to be larger than predators can consume, selection has favored a short larval period. Four-toed salamander larvae can only escape predation by metamorphosing out of the wetland (O'Laughlin and Harris 2000, Wells and Harris 2001). Harris (2005) postulated that increased newt abundance could be responsible for declines of four-toed salamander abundance in George Washington

National Forest in Virginia. Denton (2011) and Drayer (2011) report a greater abundance of newts in constructed wetlands in the DBNF, but the effect on four-toed salamander larval survival is unclear.

Management Implications in the Daniel Boone National Forest:

This study was unable to address four-toed salamander embryonic or larval survival. However, the greater abundance of nests and eggs in natural wetlands indicates the habitat is better for the four-toed salamander population than constructed wetlands, even though many of the nest site characteristics were very similar between wetland types. For constructed wetlands to provide the best possible nesting sites for four-toed salamanders an effort needs to be made to provide greater canopy closure and microhabitat complexity to encourage the colonization and growth of moss. Creating small areas of upturned soil will increase the microhabitat complexity and provide sites for moss to colonize (Petranka 1998). Maintaining downed woody debris and trees in the interior of the wetland will also provide steep slopes for nesting if the tree or log becomes covered with moss (Chalmers and Loftin 2006). One characteristic that natural wetlands have is a shallow slope to the bank. However, steep slopes have been shown to be important to four-toed salamander nesting ecology. Creating wetlands with shallow bank slopes and providing steep slopes in the form of standing or downed trees would more closely replicate natural wetland condition. Maintaining trees, downed woody debris, and hummocks in the interior of a wetland will encourage the growth of moss and provide steep slopes for nesting (Petranka 1998, Biebighauser 2011).

General Management Implications:

Little is known about the habitat requirements of larval and adult four-toed salamanders, and studies of four-toed salamander larval development are lacking. Larval surveys provide evidence of mating and more accurate estimations of breeding success than egg mass surveys (Skelly and Richardson 2010). However, due to the difficulty in capturing four-toed salamander larvae and newly metamorphosed juveniles, nest surveys are the best available method for monitoring populations (this study, Chalmers 2004, Chalmers and Loftin 2006, Drayer 2011). Corser and Dodd (2004) report that even with low annual variability of population size, several years (beyond four) of monitoring would be needed to detect four-toed salamander population size changes of 10% per year. Questions remain concerning the numbers of larvae that survive to metamorphosis in constructed wetlands compared to natural wetlands, and long-term population monitoring using nest surveys is recommended.

Four-toed salamanders and other amphibians that typically breed in ephemeral wetlands face a difficulty if constructed wetlands do not provide suitable habitat (Brown and Richter 2012). Efforts to restore forested freshwater wetlands have been limited, and hydrologically isolated wetlands are at risk of loss due to climatic change (Dahl 2011). Creating a wetland with the proper hydroperiod has proven to be difficult (Denton 2011, Brown and Richter 2012). Protecting the few remaining natural wetlands, and constructing wetlands that dry to better mimic natural wetlands is imperative to conserve amphibian populations within the Daniel Boone National Forest.

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VITA

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