

SPATIAL ECOLOGY OF EASTERN BOX TURTLES (TERRAPENE C. CAROLINA) IN  
THE OAK OPENINGS REGION OF NORTHWESTERN OHIO

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A Dissertation

Submitted to the Graduate College of Bowling Green  
State University in partial fulfillment of  
the requirements for the degree of

DOCTOR OF PHILOSOPHY

August 2016

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## ABSTRACT

Karen V. Root, Advisor

Eastern Box Turtles (*Terrapene c. carolina*) have experienced range-wide declines as the result of extensive habitat loss, fragmentation, and alteration. The Oak Openings Region of northwestern Ohio is a biodiversity hotspot that exists in a highly fragmented landscape and provides a unique case study from which to examine the effects of anthropogenic disturbance on Eastern Box Turtles. In an effort to inform management and conservation efforts in the Oak Openings Region, I initiated a radio-telemetry project with the following objectives: 1) examine the spatial ecology of Eastern Box Turtles over several years to understand how they interact with their habitat in an area as unique as the Oak Openings Region, 2) develop predictive models depicting the temporal distributions of Eastern Box Turtles, 3) examine the impacts of one of the most common management tools in the Oak Openings Region, prescribed fire, on Eastern Box Turtles, and 4) evaluate pattern-recognition software as a low-cost alternative of identifying individual Eastern Box Turtles. Turtles at my study site exhibited larger home ranges than previously reported for this species as well as hierarchical habitat selection at multiple scales. Predicted distributions followed phenological shifts in habitat use and were influenced primarily by habitat type and canopy cover. Prescribed fires have the potential to have a devastating effect on box turtle populations, but management activities that take box turtle ecology into account will minimize these impacts while maintaining a critical disturbance regime. Computer-assisted photo-recognition has a great deal of potential as a supplemental method of identifying box turtles and provides a low-cost means of incorporating citizen science data into mark-recapture studies. My work suggests that conservation for Eastern Box Turtles in the Oak Openings Region should focus on maintenance and restoration of remaining box turtle habitat, connectivity

between critical habitat patches, and altering burn regimes to minimize negative effects on box turtles while still meeting ecosystem-level management objectives.

I dedicate this work to Jess, Ian, and Gizmo. Your love and support made all of this possible.

## ACKNOWLEDGMENTS

I would like to begin by thanking my graduate committee for all of their assistance. Specifically, I'd like to thank Dr. Karen Root, for her guidance over the course of this project. Additional thanks are owed to Dr. Shannon Pelini for her invaluable discussions and contributions to my dissertation.

Thank you to Haley Scheidler, Paul Lund, Angela Klosinski, Heather Clendenin, Rebecca Briedenbach, Caitlin Cunningham, and Cari Ritzenthaler for their hours of field work and putting up with me. I am especially grateful to Cari for her dedication and giving me someone to play cards with during nesting season.

This was a collaborative effort and would not have been possible without support from Kent Bekker, Pete Tolson, Tim Schetter, Karen Menard, LaRae Sprow, Tim Gallaher, Greg Lipps, and Val Hornyak. I would be remiss if I didn't thanks to the numerous individuals who called in turtles sightings, stayed with turtles until I arrived, or took the time to tell their favorite box turtle story.

I would like to thank fellow Root Lab members, past and present, for reviewing manuscripts and dissertation drafts. Special thanks are owed to Kat Baczynski and Sara Zaleski for keeping me in check and giving me someone to pester when I needed a distraction. I am grateful to Bryce Adams for our in-depth, and often intense, statistical discussions, and getting me into R. I am especially grateful to my colleague, Pat Cain, for acting as a support system, keeping me on track, and for indulging my love of gizmos and gadgets.

I would like to recognize my family for their support and encouragement during, what turned into, a lengthy academic career. I am especially grateful to my wife, Jessica, for the sacrifices she made while I pursued my education. I could not have done this without your support.

Funding and support for this project were provided by Bowling Green State University, the Toledo Zoo, Metroparks of the Toledo Area, The Toledo Naturalist's Association, and Ohio Biological Survey. Finally, I wish to acknowledge the support of Bowling Green State University in producing this work.

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## INTRODUCTION

Global biodiversity is closely linked to human well-being and ecosystem health (Duffy 2009) and recent, unprecedented declines in biodiversity are cause for great concern. Research indicates that biodiversity within ecosystems is necessary for proper ecosystem function and provision of ecosystem services (Schulze and Mooney 1994, Rapport et al. 1998, Sala et al. 2000, Chapin et al. 2007, Duffy 2009). As such, the loss of biodiversity may have just as profound of an impact on ecosystem functioning as other causes of anthropogenic change (Tilman et al. 2012). Biodiversity is the foundation of healthy, functioning ecosystems, so conserving species is vital to preserving the ecosystems they support (Goldstein 2011, Tilman et al. 2012).

Habitat loss, fragmentation and their consequences have been identified as some of the main factors contributing to the wide-scale decline of biodiversity (Lovejoy et al. 1986, Kucera and Barrett 1995, Hilty et al. 2006). Land conversion decreases the amount of original habitat, alters the ability of organisms to move between patches of habitat and can influence the persistence of species if movement rates are insufficient to prevent inbreeding, rescue declining populations or recolonize habitats (Fahrig and Merriam 1994, Calabrese and Fagan 2004, Bowne et al. 2006). Species with low mobility and behavioral impediments to moving are unable to move freely within their landscape and may be restricted to small, fragmented patches with low connectivity which can lead to local extirpations (Laurance 1991, Gascon et al. 1999). Conserving biodiversity will, therefore, depend largely on the ability of organisms to persist in fragmented, human-dominated landscapes.

The Oak Openings Region of northwestern Ohio contains one of a few landscape-scale oak savanna systems in North America and, within its borders, it harbors a disproportionate

number of threatened and endangered species compared to the total area of the state. In spite of boasting high biodiversity, the region is in an area that has a high number of reptilian species at risk due to habitat loss and alteration. Of particular concern is the only truly terrestrial turtle species, the Eastern Box Turtle (*Terrapene c. carolina*), a mesic forest specialist that has experienced range-wide declines as a result of habitat loss. In order to ensure the long-term survival of this species, it is necessary to obtain information regarding the ecology of these turtles at various geographic locations throughout their remaining range, especially in an area as unique as the Oak Openings Region where available habitat differs greatly from what is found elsewhere.

The goal of this work is to address the paucity of research regarding the ecology of Eastern Box Turtles in the Oak Openings Region and to inform conservation and management of this species in such a unique area. Previous research has identified micro- and macrohabitat features important to box turtles, but a holistic assessment will include examining spatial ecology at a landscape scale that is relevant to land managers. This dissertation is presented as four stand-alone research chapters, each prepared as a manuscript following the specific guidelines of the target journal. These chapters are written with various coauthors and therefore use plural pronouns, however, I take sole responsibility for the content. The final chapter provides a general summary and conclusions.

Chapter I was written with coauthor Karen Root and was submitted to *Herpetologica*. This chapter details the four-year radio-telemetry and mark-recapture study that provides the foundation upon which all of the other chapters are built by providing baseline data regarding home range sizes, movement, and multi-scale habitat utilization.

Chapter II was written with coauthors Greg Lipps, Jr., and Karen V. Root for submission to *Herpetological Conservation and Biology*. Here, I use my telemetry and encounter data to explore the temporal distributions of Eastern Box Turtles on a regional scale and examine the landscape features influencing these patterns.

Chapter III was written with coauthors Patrick W. Cain and Karen V. Root for submission to *The Journal of Wildlife Management*. In this chapter we examine the direct and indirect effects of the most common management technique used to maintain oak savannas, prescribed fires, on box turtle spatial ecology. This includes fire-related mortality and injuries, and changes in body condition, area use, daily movements, and overwintering site fidelity.

Chapter IV was written with coauthors Eric J. Tobin, Gregory J. Lipps, Jr., Janice Sapak, and Karen V. Root. In this chapter, we pioneer the use of photo-recognition software as a method of identifying individual box turtles. Using more than 1500 images from three sites in two states, we test a new software program's ability to identify individual turtles based on carapace and plastron patterns. This chapter was submitted to *Herpetological Review* and published in their December 2014 issue (Vol. 45, pgs. 584 - 586).

## LITERATURE CITED

- Bowne, D.R., M.A. Bowers, and J.E. Hines. 2006. Connectivity in an agricultural landscape as reflected by interpond movements of a freshwater turtle. *Conservation Biology* 20: 780-791.
- Calabrese, J.M., and W.F. Fagan. 2004. A comparison-shopper's guide to connectivity metrics. *Frontiers in Ecology and the Environment* 2: 529-536.
- Chapin, F.S., E.S. Zaveleta, V.T. Eviner, R.L. Naylor, P.M. Vitousek, H.L. Reynolds, D.U. Hooper, S. Lavorel, O. E. Sala, S.E. Hobbie, M.C. Mack, and S. Diaz. 2000. Consequences of changing biodiversity. *Nature* 405: 232-242.
- Duffy, J.E. 2009. Why biodiversity is important to the functioning of real-world ecosystems. *Frontiers in Ecology and the Environment* 2009: 437-444.
- Fahrig, L., and G. Merriam. 1994. Conservation of fragmented populations. *Conservation Biology* 8: 50-59.
- Gascon, C. T.E. Lovejoy, R.O. Bierregaard, Jr., J.R. Malcolm, P.C. Stouffer, H.L. Vasconcleos, W.F. Laurance, B. Zimmerman, M. Tocher, and S. Borges. 1999. Matrix habitat and species richness in tropical forest remnants. *Biological Conservation* 91: 223-229.
- Goldstein, N. 2011. *Global Issues: Biodiversity*. Facts on file, Inc., New York, New York, USA.
- Hilty, J.A, W.Z. Lidicker, Jr., and A.M. Merenlender. 2006. *Corridor Ecology: The Science and Practice of Linking Landscapes for Biodiversity Conservation*. Island Press, Washington D.C., USA.
- Kucera, T.E., and R.H. Barrett. 1995. California wildlife faces uncertain future. *California Agriculture* 49: 23-27.
- Laurance, W.F. 1991. Ecological correlates of extinction proneness in Australian tropical

rainforest mammals. *Conservation Biology* 5: 79-89.

Lovejoy, T.E., R.O. Bierregaard, Jr., A.B. Rylands, J.R. Malcolm, C.E. Quintela, L.H. Harper, K.S. Brown Jr., G.V.N. Powell, H.O.R. Schubart, and M.D. Hays. 1986. Edge effects and other effects of isolation on Amazon forest fragments. *In*: Soule, M.E. (Ed.), *Conservation Biology: The Science of Scarcity and Diversity*. Sinauer Associates Inc., Sutherland, MA, pp. 257-285.

Rapport, D.J., R. Costanzo, and A.J. McMichael. 1998. Assessing ecosystem health. *Trends in Ecology and Evolution* 13: 397-402.

Sala, O.E., F.S. Chapin, J.J. Arnesto, E. Berlow, J. Bloomfield, R. Dirzo, E. Huber-Sanwald, L.F. Hunneke, R.B. Jackson, A. Kinzig, R. Leemans, D.M. Lodge, H.A. Mooney, M. Oosterheld, N.L. Poff, M.T. Sykes, B.H. Walker, M. Walker, and D.H. Hall. 2000. Global biodiversity scenarios for the year 2100. *Science* 287: 118-130.

Schulze, E.D., and H.A. Mooney, eds. 1994. *Biodiversity and Ecosystem Functioning*. Springer, New York, USA.

Tilman, D., P.B. Reich, and F. Isbell. 2012. Biodiversity impacts on ecosystem productivity as much as resources, disturbance or herbivory. *PNAS* 109: 10394-10397.

CHAPTER I: SPATIAL ECOLOGY OF EASTERN BOX TURTLES (*TERRAPENE C. CAROLINA*) IN A BIODIVERSITY HOTSPOT IN NORTHWESTERN OHIO

**ABSTRACT**

We conducted a radio-telemetry study in a biodiversity hotspot of northwestern Ohio to investigate the spatial ecology of Eastern Box Turtles (*Terrapene c. carolina*) in this unique region. We radio-tracked 22 turtles to assess daily movements, home range sizes, and habitat selection during the 2012-2014 active seasons. Home range sizes and movement rates radio-telemetered turtles in our study site were higher than reported in most studies. Turtles traveled extensively during our study and showed frequent use of wet forests, upland hardwoods, and prairies while avoiding most anthropogenic habitats. Within their habitats, turtles selected sites with higher surface humidity, deeper leaf litter, closer proximity to logs and greater vegetative complexity than conditions that would be found at random locations. Conservation strategies for this species that account for habitat preferences at multiple scales will likely be the most effective. The results of our study can be used to incorporate box turtle spatial ecology into existing management and conservation activities within the region.

**INTRODUCTION**

Habitat loss, fragmentation, and their consequences have been identified as some of the main factors contributing to the wide-scale decline of biodiversity (Lovejoy et al. 1986; Kucera and Barrett 1995; Hilty et al. 2006). Land conversion decreases the amount of original habitat available, alters the ability of organisms to move between patches, and can influence the persistence of species if movement rates are insufficient to prevent inbreeding, rescue declining populations, or recolonize habitats (Farigh and Merriam 1994; Calabrese and Fagan 2004; Bowne et al. 2006). As such, a detailed understanding of an organism's movements, home range

size and habitat selection are crucial prerequisites for any conservation or management plan. This is especially true for species on the decline. Within a given area, animal distributions and patterns of habitat selection can provide insight into the quality and availability of resources (Johnson 2005). For many species, activity patterns vary spatially and temporally (Powell and Mitchell 2012), necessitating studies at various localities throughout their range.

Many turtle species have complex habitat requirements that vary temporally and regionally (Seigel and Dodd 2000; Dodd 2001). For turtles, a holistic conservation strategy will include understanding movement patterns, habitat use, demographic responses to perturbations, and regional distribution of populations and habitats (Mitchell and Klemens 2000; Donaldson and Echternacht 2005; Redder et al. 2006) as well as protecting habitats (Dodd 2001). Life history characteristics such as delayed reproduction, low annual fecundity, low recruitment, and high adult survivorship make adult turtles particularly vulnerable to disturbance and subsequent mortality (Congdon et al. 1993; Congdon et al. 1994; Dodd et al. 2016). It is therefore important to understand the regional spatial ecology of target species in order to develop sound conservation strategies.

While geographically widespread, Eastern Box Turtles (*Terrapene c. carolina*) have experienced range-wide declines (Stickel 1978; Williams and Parker 1987; Schwartz and Schwartz 1991; Hall et al. 1999; Dodd 2001) and are protected in some states (Ernst and Lovich 2009). This species predominantly inhabits mesic open woodlands, but regional habitat variation is common and with frequent use of pastures, marshy meadows, riparian zones, and aquatic habitats (Dodd 2001; Donaldson and Echternacht 2005; Ernst and Lovich 2009). Habitat preferences are believed to be driven by microhabitat selection which tends to favor litter, humidity, temperature, and ground cover (Reagan 1974; Dodd 2001; Penick et al. 2001; Rossell

et al. 2006). Eastern box turtle home ranges are relatively small, typically 1-5 ha in size (Dodd 2001; Ernst and Lovich 2009), but recent studies have reported home ranges much larger (Wiley 2010; Laarman 2013; Greenspan et al. 2015). This variation makes it difficult to generalize space requirements or develop conservation strategies that work effectively throughout their range.

Given the regional variation in Eastern Box Turtle home range sizes, movements and habitat use (Dodd 2001; Ernst and Lovich 2009), it is important to obtain data from a wide range of geographic locations, especially in areas where previous work has been limited. Our study focuses on the spatial ecology of Eastern Box Turtles in a region of Ohio for which we have little ecological information. The Oak Openings Region of northwestern Ohio presents a unique case in which to study box turtles spatial ecology because of the amount of fragmentation the region has experienced combined with the biodiversity it contains, which includes several globally rare plant communities (Schetter and Root 2011; EPA 2016). These fire dependent plant communities are maintained by a combination of prescribed fire and mechanical removal of woody vegetation and invasive species (Peterson and Reich 2001; Abella et al. 2007). Box turtles are of particular conservation interest because of frequency and amount of land management that occurs and the paucity of information regarding Eastern Box Turtle spatial ecology within the Oak Openings Region. Our objective was to elucidate the activity patterns, home range requirements, and habitat selection for box turtles in a unique study area. Overall, the goal of this study was to improve conservation efforts for Eastern Box Turtles in a heavily-managed area by directing region specific maintenance of habitat patch size, juxtaposition of requisite habitats, and protection of critical areas (e.g., nesting and overwintering sites).

## METHODS

### *Study Site*

This study took place within the Oak Openings Preserve Metropark (Lucas County, Ohio), at 1,528 ha, is the largest park in the Oak Openings Region; a 48,000 ha stretch of habitat that contains one of a few landscape-scale oak savanna systems in the Midwestern United States (Figure 1; Schetter and Root 2011). This region is a biodiversity hotspot (Abella et al. 2001; Abella et al. 2004; Schetter and Root 2011) and contains disproportionately more organisms of conservation interest than any other similar-sized area in the state. It is estimated that of the total land area that makes up the Oak Openings Region, approximately 27% of this is thought to exist in a natural or semi-natural state (Schetter and Root 2011). Much of the preserve is dominated by deciduous forests, but oak savanna, Eurasian meadow, and prairie habitats contribute to the heterogeneity of the landscape. These early successional plant communities are maintained through the use of periodic prescribed fires. The preserve has approximately 99.1 km of trails (biking/hiking: 50.6 km, horse: 35.7 km and all paved: 12.8 km) and is intersected by approximately 14.9 km of roads that range in speed from 24 to 88 km/h. Soils in this area are post-glaciated beach sand deposited over clay till (Hawkins and Weintraub 2011).

### *Capture and Telemetry*

Beginning in the spring of 2012, box turtles were located via visual encounter surveys. Upon capture we recorded weight, straight-line carapace length and width, determined their sex using secondary sexual characteristics (Ernst and Lovich 2009) and implanted them with a passive integrated transponder (PIT) tag (AVID® MicroChip ID Systems, Folsom, Louisiana, USA) for individual identification. A subset of captured turtles had transmitters (models RI-2B

or SI-2F; Holohil Systems, Ltd., Ontario, Canada) affixed to their carapaces. The PIT tag and transmitter made up no more than 5% (max 15 g) of the turtle's total body weight.

Following morphometric measurements, turtles were released at their initial capture location. Turtles that were part of the telemetry portion of the project were tracked using an R1000 radio-telemetry receiver (Communications Specialists, Inc., Orange, California, USA) and a folding Yagi 3-element antenna (Advanced Telemetry Systems, Isanti, Minnesota, USA). Turtles were radio-tracked two to three times per week throughout their active season (March through November) with order and time of day randomized to reduce autocorrelation (Martin 2009; Millspough et al. 2012). For each tracked location, we recorded GPS coordinates, date, time, activity, litter depth, temperature (ground and ambient) and humidity (ground and ambient).

Due to small sample sizes (3 male, 19 female), statistical comparisons between sexes were not made for any of our analyses. All results are reported  $\pm$  SD, unless otherwise noted.

#### *Daily Movements and Home Ranges*

Daily movements were estimated from telemetry points by measuring the straight-line distance between successive locations. The Animal Movement Extension ArcView© GIS 3.2a (Hooge and Eichenlaub 1997) was used to conduct asymptote analysis to determine the minimum number of relocations required to adequately describe a home range (Odum and Kuenzler 1955). The Geospatial Modeling Environment Version 0.7.2.1 (Beyer 2012) was used to estimate 100% minimum convex polygon (MCP) and 95% kernel home (KDE) ranges. We calculated 95% kernel isopleths of utilization distributions using the fixed kernel method with least squares cross validation. We chose MCP and KDE home ranges because they are some of

the most commonly used methods in the literature, therefore allowing for ease of comparison with other studies.

Home range sizes and daily movements were compared between the three years of this study using Kruskal-Wallis tests followed by post-hoc Tukey tests.

### *Microhabitat Utilization*

To characterize the microhabitat at the turtle and random point locations, we used a modified version of the methods employed by Blouin-Demers and Weatherhead (2001). Once a week, we measured 18 structural and climatic variables within circular plots of different radii around the turtles and the random points (Table 1). We selected variables that were similar to other microhabitat features reported in previous studies (Nieuwolt 1996; Penick et al. 2002; Converse and Savidge 2003; Rossell et al. 2006). Distances were measured to the nearest tenth of a centimeter using a 30 m measuring tape. Log decay class and percent on the ground were estimated visually (Brown 1974; Pyle and Brown 1998). Percent ground cover (green vegetation, log, leaf litter, twigs and other) was estimated using a sighting tube with a crosshair at one end (modified from Winkworth and Goodall 1962). We recorded ground cover data at points within the 2 m circle along the cardinal and intercardinal directions for a total of 25 points within the plot (one center, four along each cardinal direction and two along each intercardinal direction). The same suite of data was gathered at an associated random location based on a random azimuth and distance (10-100 m) from the turtle location.

To analyze box turtle microhabitat use, we used conditional logistic regression (PROC LOGISTIC; SAS 9.3) for 1:1 matched pairs with a stepwise selection procedure and “STRATA” command to account for repeated measures. This method compares each turtle location to its associated random point by subtracting the random point values from the turtle location

(Compton et al. 2002). The results are interpreted as the differences in habitat variables instead of absolute measured values as in traditional logistic regression (Compton et al. 2002; Moore and Gillingham 2006). To reduce multicollinearity we minimized the number of candidate variables for our models by removing highly correlated variables (correlation coefficient  $\geq 0.70$ ) and biologically similar variables. All possible models were tested using Akaike's Information Criterion corrected for small sample size ( $AIC_c$ ) to select and rank the most parsimonious model (Burnham and Anderson 2002). The model(s) with the lowest  $AIC_c$  and all models within two units of the lowest, containing only significant variables, were considered to be supported (Burnham and Anderson 2002).

Due to logistical constraints, microhabitat analyses were only conducted during the 2012 active season.

#### *Macrohabitat and Landscape-scale Utilization*

We used a land cover map specific to our study area (Schetter and Root 2011; cell size: 30 m) to examine habitat utilization at the macrohabitat and landscape scales. Of the original 15 habitat classes, the following were aggregated based on ecological similarities: open (prairie, Eurasian meadow, and barrens), residential (residential and asphalt) and wet forest (swamp forest and floodplain forest). Macrohabitat, or third order selection (Johnson 1980), was defined as the area within the kernel home range. Using the land cover dataset, all turtle points were classified based on the land cover type in which they occurred (ArcGIS 10.1, Environmental Systems Research Institute, Inc., Redlands, CA). Compositional analysis was used to analyze habitat selection within the kernel home ranges and to generate rankings among habitat types (Aeibischer et al. 1993; Garshelis 2000). This method considers the individual animal as the sampling unit instead of individual radiolocations (Aebischer et al. 1993, Pendleton et al. 1998).

Proportion of habitat used for each turtle was determined by dividing the number of locations within each habitat type by the total number of locations for that turtle. Available habitat at this level was defined as the proportion of habitat use within the kernel home range. Compositional Analysis was performed in SAS 9.3 (BYCOMP.SAS Version 1.0, <http://nhsbig.inhs.uiuc.edu/wes/habitat.html>, accessed 24 August 2012) to examine turtle locations for disproportionate habitat use. Habitat types that were available but not used by the turtles were replaced with small, non-zero proportions (0.0001) as per Aebischer et al. (1993). We removed the agricultural, scrub/shrub and wet prairie habitat types to avoid potential confounding effects of rare habitat types in compositional analysis (Aebischer et al. 1993). These habitat types were removed because they were rarely available and were not used by the turtles where these habitat types were available.

We also used compositional analysis to analyze habitat use at the landscape-scale, or second order selection (Johnson 1980), at the level of the study site. The methods for this were identical to those used for the macrohabitat analysis with the exception of differing habitat availability. We defined available habitat at the landscape-scale by creating a buffer around each turtle GPS point with radius equal to the greatest length of any 100% MCP home range (1,708 m).

## RESULTS

### *Daily Movements and Home Ranges*

A total of 168 Eastern Box Turtles were captured during the course of this study, 42 of which were recaptures (Appendix A). A total of 22 turtles were selected for transmitter attachment and tracked during their active seasons (March – November). Of these, 16 were tracked for more than one season. The number of locations per individual turtle ranged from 32

to 53, and all turtles were tracked until they overwintered. Box turtles at our study site followed the general movement pattern of emergence from overwintering sites mid-March, then moved to different habitats based on sex: males tended to remain in closed-canopy, upland forests and made a mid-summer shift to low, wet forests; gravid and non-gravid females moved into savanna and prairie habitats from May-July then moved back into upland and wet forests. Use of wet forest remained relatively consistent throughout the active season, while the proportion of use of upland deciduous, conifer, and open habitats varied (Figure 2). Nesting occurred from early-May to mid-June in sandy dune areas and along bike trails within the park (Appendix II). Both sexes began moving back to their overwintering locations in September, but ingress did not occur until mid-late October. Daily movements for the 2012-2014 seasons averaged 22.55 m/d, 24.42 m/d, and 18.29 m/d, respectively and ranged from < 1.0 m to 742 m/d. Many of the pronounced movements were of females traveling to nesting sites. Minimum convex polygon home ranges for the 2012-2014 seasons averaged  $13.95 \pm 8.01$  ha,  $22.29 \pm 22.70$  ha, and  $26.86 \pm 26.71$  ha, respectively and ranged in size from 1.50 to 98.85 ha (Figure 3 A-C). There were no difference in daily movement or home range sizes between years ( $P = 0.4287$  and  $P = 0.3615$ , respectively).

#### Microhabitat Utilization

The best model selected by the conditional logistic regression to explain the differences between turtle and random locations contained the variables litter depth, percent litter covering the ground, surface humidity and distance to nearest log ( $y = 0.5116 ld + -0.0252 pl + 0.1235 sh + -0.0552 dl$ ). This model indicates that at the microhabitat scale, turtles tended to select areas with deeper litter, lower percentage of litter covering the ground (i.e., more vegetative complexity), higher surface humidity, and closer proximity to logs. There were no competing

models (i.e., those within 2 AIC<sub>c</sub> of the most likely model), however we show the top five models for comparison (Table 2).

### *Macrohabitat and Landscape-scale Utilization*

Results of the compositional analyses detected non-random habitat use in all years at the second-order scale, but only in one year at the third-order scale (Table 3). Habitat use varied annually, but wet forests, upland deciduous, savanna, and open were the most preferred habitat types across years, whereas conifer and residential habitats were generally avoided (i.e., ranked last; Table 3).

## **DISCUSSION**

Our data indicate that turtles within the Oak Openings Preserve select their habitat preferentially at multiple spatial scales. Box turtles at our study site established home ranges that were larger than many other reported studies. Daily movements were characterized by relatively shorter movements followed by occasional long-distance bursts, with gravid females traveling long distances in short amounts of time to reach nesting sites. Box turtles were generally associated with deciduous forested and grassland-type habitats which offered microhabitats that favored increased surface humidity and vegetation complexity, deeper litter and closer proximity to logs.

The daily movements of box turtles in our study were similar to some recent studies (Donaldson and Echternacht 2005: 26 m/d; Iglay et al, 2007: 26 m/d; Baker 2009: 31.5 m/d), but were much larger than reported by Hester et al. (2008: 8.6 m/d). Home ranges of Eastern Box Turtles in the Oak Openings Preserve were larger than the species' average (4.43 ha; Ernst and Lovich 2009) and other studies not included in the average (Hester et al. 2008: 6.45 ha; Quinn 2008: 4.97 ha; Baker 2009: 7.4 ha; Currylow 2011: 7.45 ha). The observed home ranges in our

study were most similar to those reported by Wiley (2010: 12.9 ha), Laarman (2013: 18.8 ha), and Greenspan et al. (2015: 10.3 ha). Variation in home range sizes across studies can be attributed to regional variation, availability or distribution of resources (Nieuwolt 1996; Iglay et al. 2007), quality of habitat, and habitat structure (Stickel 1989; Dodd 2001). In general, home range sizes are expected to increase where life requisites impacting abundance and distribution cannot be fulfilled within a small area and, therefore, require large movements (McDonald et al. 2012). The observed differences in our study may also be driven by the number of gravid females in our sample who often traveled long distances, up to 1 km, to find suitable nesting sites. In general, females tend to have larger home ranges than males (Ernst and Lovich 2009). Patterns of movement and area use for females and males could be related to nesting and mate-searching (Stickel 1950; Iglay et al. 2007), therefore, in areas where nesting sites are limited and females have to travel long distances to find suitable habitat, we would expect to see larger home ranges in both sexes. Nesting sites in the park appear to be limited or near roads and females must therefore travel to find the resources they need. It is important to note that many of these studies have varying durations, sample sizes, monitoring methods and method of calculating a home range. Comparisons between studies can be difficult if researchers have differing sample sizes and home range estimation methods (Seaman et al. 1990), which may account for the differences in home range size between our study and others.

Box turtles in our study primarily selected wet forest, upland deciduous forest, and savanna, and to a lesser extent, prairies and barrens. The association with these habitats is well-documented (reviewed in Dodd 2001; Ernst and Lovich 2009), with the exception of wet forests (swamp and floodplain). Donaldson and Echternacht (2005) and Baker (2009) reported extensive use of aquatic habitats by box turtles at their study and Laarman (2013) found a close association

with riparian areas in Northern Michigan. Reports of box turtles using hydric habitats are less common in the literature and these findings suggest these sites are of regional importance and supports obtaining detailed information at various geographic locations. The importance of wet forests and their juxtaposition within our study site to other preferred habitats (e.g. prairie and savanna) could be another possible explanation for our larger home ranges. While the Oak Openings Preserve boasts high biodiversity and associated habitat heterogeneity (Schetter and Root 2011; Schetter et al. 2013), the spatial arrangement of preferred habitats may be a factor influencing movement patterns and area use of turtles in this region.

Similarly, the availability and distribution of suitable microhabitats will contribute to patterns of movement and areas use at larger scales. Within their preferred habitats, box turtles selected areas based on humidity, leaf litter depth, ground cover, and proximity to logs. These factors are consistent with what other studies have found (Reagan 1974; Converse and Savidge 2003; Rossell et al. 2006) and are presumably related to optimizing thermoregulation, reducing water loss (Penick et al. 2001; Rossell et al. 2006; Nazdrowicz et al. 2008) and prey acquisition (McKnight 2011). These findings can be useful to managers as management activities (e.g., prescribed burns) can influence the availability of suitable microhabitats.

Our study provides information that can be used to aid box turtle conservation within the Oak Openings Region and contributes new information to the spatial ecology of this species along the northern reaches of its range. Being near the periphery of their range may make Eastern Box Turtles more sensitive to fragmentation and might also contribute to geographic variation in activity patterns and habitat use (Swihart 2003). Habitat loss, fragmentation, degradation, and alteration can cause changes in microhabitat and natural vegetation (Hilty et al. 2006), which could be detrimental to box turtles that rely on appropriate microhabitats within

their home range for thermoregulation and foraging. Management to maintain and restore early-successional plant communities (e.g., oak savanna) may create a patchy landscape which may necessitate more travel for turtles to find the biotic and abiotic components for their reproduction and survival. In turn, this can lead to increased energy expenditure, inability to find resources, and increase encounters with hazards (e.g. roads), which may result in decreased survival (Hester et al. 2008).

By understanding the requirements of box turtles at unique locations, managers will be better suited to make decisions that benefit this species and can incorporate the ecological requirements of Eastern Box Turtles into existing management plans. Reports of box turtles using hydric habitats are less common in the literature and these findings suggest these sites are of regional importance and supports obtaining detailed information at various geographic locations. Future research should attempt to assess population demographics and determine whether the patterns reported in this study vary temporally. Conservation plans would benefit from increasing the amount of nesting and foraging habitats within close proximity to each other to reduce the distance traveled and subsequent risk of encountering roads.

#### **ACKNOWLEDGMENTS**

This project was supported by the Toledo Metroparks with funding provided by the Toledo Zoo and Bowling Green State University. We thank our field assistants R. Breidenbach, H. Clendenin, J. Cross, C. Cunningham, A. Klosinski, P. Lund, C. Ritzenthaler, and H. Shiedler for their time and dedication. Thanks are owed to P. Tolson, G. Lipps, C. Ellsworth, K. Menard and A. Selden for logistic support. Additional thanks are owed to N. Boudreau and the Bowling Green State University Center for Business Analytics. We are grateful to B. Adams and P. Cain for providing insight throughout the writing process. All animals were handled in compliance

with Bowling Green State University's Institutional Animal Care and Use Committee guidelines (IACUC Approval # 12-005). These procedures followed the regulations outlined by the Ohio Department of Natural Resources Wild Animal Permit (#15-48) and the Metroparks of the Toledo Area (#042512).

### LITERATURE CITED

- Abella, S.R., J.F. Jaeger, D.H. Gehring, R.G. Jacksy, K.S. Menard, and K.A. High. 2001. Restoring historic plant communities in the Oak Openings Region of Northwest Ohio. *Ecological Restoration* 19: 155-160.
- Abella, S.R., J.F. Jaeger, and L.W. Brewer. 2004. Fifteen years of plant community dynamics during a Northwest Ohio oak savanna restoration. *The Michigan Botanist* 43: 117-127.
- Abella, S.R., J.F. Jaeger, and T.A. Schetter. 2007. Public land acquisition and ecological restoration: an example from Northwest Ohio's Oak Openings Region. *Natural Areas Journal* 27: 92-97.
- Aebischer, N.J., P.A. Robertson, and R.E. Kenward. 1993. Compositional analysis of habitat use from animal radio-tracking data. *Ecology* 74: 1313-1325.
- Baker, J.E. 2009. Home range and movement of the Eastern Box Turtle (*Terrapene carolina*) in East Central Illinois. M.S. thesis, University of Illinois at Urbana-Champaign, USA.
- Beyer, H.L. 2012. Geospatial Modeling Environment (Version 0.7.2.1). (software). URL: <http://www.spataleecology.com/gme>.
- Blouin-Demers, G., and P.J. Weatherhead. 2001. Habitat use by black rat snakes (*Elaphe obsoleta obsoleta*) in fragmented forests. *Ecology* 82: 2882-2896.
- Bowne, D.R., M.A. Bowers, and J.E. Hines. 2006. Connectivity in an agricultural landscape as

- reflected by the interpond movements of a freshwater turtle. *Conservation Biology* 20: 780-791.
- Brown, J.K. 1974. Handbook for inventorying downed woody material. USDA For. Serv. Gen. Tech. Rep. INT-16, Intermt. For. And Range Exp. Stn., Ogden, Utah, USA.
- Burnham, K.P., and D.R. Anderson. 2002. Model selection and inference: a practical information-theoretic approach. Springer-Verlag, New York, New York, USA.
- Calabrese, J.M., and W.F. Fagan. 2004. A comparison-shopper's guide to connectivity metrics. *Frontiers in Ecology and the Environment* 2: 529-536.
- Compton, B.W., J.W. Rhymer and M. McCollough. 2002. Habitat selection by wood turtles (*Clemmys insculpta*): an application of paired logistic regression. *Ecology* 83: 833-843.
- Congdon, J.D., A.E. Dunham, and R.C. Van Loben Sels. 1993. Delayed sexual maturity and demographics of Blanding's Turtles (*Emydoidea blandingii*): implications for conservation and management of long-lived organisms. *Conservation Biology* 7: 826-833.
- Congdon, J.D., A.E. Dunham, and R.C. Van Loben Sels. 1993. Demographics of Common snapping Turtles (*Chelydra serpentina*): implications for conservation and management of long-lived organisms. *American Zoologist* 34: 397-408.
- Converse, S.J., and J.A. Savidge. Ambient temperature, activity, and microhabitat use by ornate box turtles (*Terrapene ornata ornata*). *Journal of Herpetology* 37: 665 – 670.
- Currylow, A.F. 2011. Effects of forest management on the ecology and behavior of Eastern Box Turtles. M.S. thesis, Purdue University, Indiana, USA.
- Dodd, C.K., Jr. 2001. North American box turtles: a natural history. Norman, Oklahoma, USA: University of Oklahoma Press. 231 p.

- Dodd, C.K., V. Rolland, and M.K. Oli. 2016. Consequences of individual removal on persistence of a protected population of long-lived turtles. *Animal Conservation* ??: 1-11.
- Donaldson, B.M., and A.C. Echternacht. 2005. Aquatic habitat use relative to home range and seasonal movement of eastern box turtles (*Terrapene carolina carolina*: Emydidae) in eastern Tennessee. *Journal of Herpetology* 39: 284-287.
- Environmental Protection Agency (EPA) 2016. Oak Openings Site Conservation Plan. Available at <http://www.epa.gov/ecopage/upland/oak/oakopen.html> . Accessed 23 March 2016.
- Ernst, C.H., and J.E. Lovich. 2009. Turtles of the United States and Canada, 2<sup>nd</sup> edition. The Johns Hopkins University Press, Baltimore, Maryland, USA.
- Farigh, L., and G. Merriam. 1994. Conservation of fragmented populations. *Conservation Biology* 8: 50-59.
- Faber-Langendoen, D (ed.). 2001. Plant communities of the Midwest: classification in an ecological context. Association for Biodiversity Information, Arlington, VA.
- Greenspan, S.E., E.P. Condon, and L.L. Smith. 2015. Home range and habitat selection in the Eastern Box Turtle (*Terrapene carolina carolina*) in a Longleaf Pine (*Pinus palustris*) reserve. *Herpetological Conservation and Biology* 10: 99-111.
- Hawkins, J, and M.N. Weintraub. 2011. The effect of trails on soil in the Oak Openings of Northwestern Ohio. *Natural Areas Journal* 31: 391-399.
- Hester, J.M., S.J. Price, and M.E. Dorcas. 2008. Effects of relocation on movements and home ranges of Eastern Box Turtles. *The Journal of Wildlife Management* 72: 772-777.
- Hilty, J.A., W.Z. Lidicker Jr., and A.M. Merenlender. 2006. Corridor Ecology: The Science and Practice of Linking Landscapes for Biodiversity Conservation. Island Press, Washington D.C., USA.

- Hooge, P.N., and W.M. Eichenlaub. 1997. Animal movement extension to ArcView version. 1.1. Alaska Science Center, Biological Science Office, USGS, Anchorage, Alaska, USA.
- Igley, R.B., J.L. Bowman, and N.H. Nazdrowicz. 2007. Eastern Box Turtle (*Terrapene carolina carolina*) movements in a fragmented landscape. *Journal of Herpetology* 41: 102-106.
- Johnson, D.H. 1980. The comparison of usage and availability measurements for evaluating resource preference. *Ecology* 61: 65-71.
- Johnson, M.D. 2005. Habitat quality: a brief review for wildlife biologists. *Transactions of the Western Section of the Wildlife Society* 41: 31-41.
- Kucera, T.E., and R.H. Barrett. 1995. California wildlife faces uncertain future. *California Agriculture* 49: 23-27.
- Laarman, P.L. 2013. Monitoring eastern box turtles in the Manistee National Forest: progress report and preliminary summary of field data. Unpublished technical report to the U.S. Forest Service. 68 p.
- Lovejoy, T.E., R.O. Bierregaard, Jr., A.B. Rylands, J.R. Malcolm, C.E. Quintela, L.H. Harper, K.S. Brown, Jr., G.V.N. Powell, H.O.R. Schubart, and M.D. Hays. 1986. Edge effects and other effects of isolation on Amazon forest fragments. Pp. 257-258 in *Conservation Biology: The Science of Scarcity and Diversity* (M.E. Soule, ed). Sinauer Associates Inc., Sutherland, Massachusetts, USA.
- Martin, J., V. Tolon, B. Van Moorter, M. Basille, and C. Calenge. 2009. On the use of telemetry in habitat selection studies. Pp. 37-55 in *Telemetry: Research, Technology and Applications* (D. Barculo and J. Daniels, eds). Nova Science Publishers Inc., New York, USA.
- McDonald, L.L., W.P. Erickson, M.S. Boyce, and J.R. Alldredge. 2012. Modeling vertebrate use

- of terrestrial resources. Pp. 410-428 in *The Wildlife Techniques Manual: Research*, 7<sup>th</sup> edition (N.J. Silvy, ed). The Johns Hopkins University Press, Baltimore, Maryland, USA.
- Millsbaugh, J.J., D.C. Kesler, R.W. Kays, R.A. Gitzen, J.H. Schulz, C.T. Rota, C.M. Bodinof, J.L. Belant, and B.J. Keller. 2012. Wildlife radiotelemetry and remote monitoring. Pp. 258-283 in *The Wildlife Techniques Manual: Research*, 7<sup>th</sup> edition (N.J. Silvy, ed). The Johns Hopkins University Press, Baltimore, Maryland, USA.
- Mitchell, J.C., and M.W. Klemens. 2000. Primary and secondary effects of habitat alteration. *In: Turtle Conservation*. M.W. Klemens (ed.). Smithsonian Institution Press, Washington, D.C., USA.
- Moore, J.A., and J.C. Gillingham. 2006. Spatial ecology and multi-scale habitat selection by a threatened rattlesnake: the eastern massasauga (*Sistrurus catenatus catenatus*). *Copeia* 4: 742-751.
- Nieuwolt, P.M. 1996. Movement, activity, and microhabitat selection in the western box turtle, *Terrapene ornata luteola*, in New Mexico. *Herpetologica* 52: 487-495.
- Nazdrowicz, N.H., J.L. Bowman, and R.R. Roth. 2008. Population ecology of the eastern box turtle in a fragmented landscape. *The Journal of Wildlife Management* 72: 745-753.
- Odum, E.P., and E.J. Kuenzler. 1955. Measurement of territory and home range size in birds. *Auk* 72: 128-137.
- Ohio Department of Natural Resources: Division of Natural Areas and Preserves. [ODNR Division of Natural Areas and Preserves] 2012. Rare native Ohio plants: 2012-2013 Status List. Available online < <http://ohiodnr.com/Portals/3/heritage/2012-13-plant-list.pdf>>. Accessed 24 April 2013.
- Ohio Department of Natural Resources: Division of Wildlife [ODNR Division of Wildlife].

2012. Wildlife that are considered to be endangered, threatened, species of concern, special interest, extirpated, or extinct in Ohio. Available online <<http://ohiodnr.com/Portals/9/pdf/pub356.pdf>>. Accessed 24 April 2013.
- Penick, D.N., J. Congdon, J.R. Spotila, and J.B. Williams. Microclimates and energetics of free-living box turtles, *Terrapene carolina*, in south Carolina. *Physiological and Biochemical Zoology* 75: 57-65.
- Powell, R.A., and S. Mitchell. 2012. What is a home range? *Journal of Mammalogy* 93: 948-958.
- Pyle, C., and M.M. Brown. 1998. A rapid system of decay classification for hardwood logs of the eastern deciduous forest floor. *Journal of the Torrey Botanical Society* 125: 237-245.
- Quinn, D.P. 2008. A radio-telemetric study of the eastern box turtle (*Terrapene carolina carolina*), home-range, habitat use, and hibernacula selection in Connecticut. Biology. Central Connecticut State University.
- Reagan, D.P. 1974. Habitat selection in the three-toed box turtle, *Terrapene carolina triunguis*. *Copeia* 1974: 512-527.
- Redder, A.J., C.K. Dodd Jr., D. Keinath, D. McDonald, and T. Ise. 2006. Ornate Box Turtle (*Terrapene ornata*): a technical conservation assessment. U.S.D.A. Forest Service, Rocky Mountain Region, USA. Available at [http://www.fs.usda.gov/Internet/FSE\\_DOCUMENTS/stelprdb5182076.pdf](http://www.fs.usda.gov/Internet/FSE_DOCUMENTS/stelprdb5182076.pdf). Archived by WebCite at <http://www.webcitation.org/6ghJGG2Rd> on 11 April 11, 2016.
- Robel, R.J., J.N. Briggs, A.D. Dayton, and L.C. Hulbert. 1970. Relationships between visual obstruction measurements and weight of grassland vegetation. *Journal of Rangeland Management* 23: 295-297.
- Rossell, R., J.R., I.M. Rossell, and S. Patch. 2006. Microhabitat selection by eastern box turtles

- (*Terrapene c. carolina*) in a North Carolina mountain wetland. *Journal of Herpetology* 40: 280-284.
- Schetter, T.A. and, K.V. Root. 2011. Assessing an imperiled oak savanna landscape in northwestern Ohio using Landsat data. *Natural Areas Journal* 31: 118-130.
- Schetter, T.A., T.L. Walters, and K.V. Root. 2013. A multi-scale spatial analysis of native and exotic plant species richness within a mixed-disturbance oak savanna landscape. *Environmental Management* 52: 581-594.
- Schwartz, C.W., and E.R. Schwartz. 1974. The three-toed box turtle in central Missouri: its population, home range, and movements. *Terrestrial Series*. Missouri Department of Conservation, Jefferson City.
- Schwartz, E.R., C.W. Schwartz, and A.R. Keister. 1984. The three-toed box turtle in central Missouri, Part II: a nineteen-year study on home range, movements and population. *Terrestrial Series*. Missouri Department of Conservation, Jefferson City.
- Seaman, D.E., J.J. Millsbaugh, B.J. Kernohan, and C. Brundige. 1990. Effects of sample size on kernel home range estimates. *The Journal of Wildlife Management* 63: 739-747.
- Seigel, R.A., and C.K. Dodd. 2000. Manipulation of turtle populations for conservation. Pp. 218-238 in *Turtle Conservation* (M.W. Klemens, ed). Smithsonian Institution Press, Washington D.C., USA.
- Stickel, L.F. 1950. Populations and home range relationships of the box turtle, *Terrapene c. carolina* (Linnaeus). *Ecological Monographs* 20: 351-378.
- Stickel, L.F. 1989. Home range behavior among box turtles (*Terrapene c. carolina*) of a bottomland forest in Maryland. *Journal of Herpetology* 23: 40-41.
- Swihart, R.K., T.M. Gehring, M.B. Kolozsvary, and T.E. Nupp. 2003. Responses of 'resistant'

vertebrates to habitat loss and fragmentation: the importance of niche breadth and range boundaries. *Diversity and Distributions* 9: 1-18.

Wiley, L.L. 2010. Spatial ecology of the Eastern Box Turtle (*Terrapene c. carolina*) in Central Massachusetts. Ph.D. dissertation, University of Massachusetts Amherst, USA.

Wilson, S.D. 2012. Movement and ecology of the eastern box turtle (*Terrapene carolina carolina*) in a heterogeneous landscape. M.S. thesis, Bowling Green State University, Bowling Green, OH.

Winkworth, R.E., and D.W. Goodall. 1962. A crosswise sighting tube for point quadrat analysis. *Ecology* 43: 342-343.

Table 1. Climatic and structural variables used in the microhabitat analysis of box turtles in the Oak Openings Preserve with sampling radii (if applicable) and associated abbreviations.

Variable	Radius (m)	Description
st*	N.A.	Surface temperature (°C)
at	N.A.	Ambient temperature (°C, ~1.5m above surface)
sh*	N.A.	Relative humidity (%) on the surface
ah	N.A.	Ambient relative humidity (% ,~1.5m above surface)
el*	N.A.	Elevation (m)
ld*	N.A.	Litter depth (cm)
dl*	10	Distance (m) to nearest log ( $\geq 10$ cm maximum diameter)
md	10	Maximum diameter (cm) of nearest log
dc	10	Log decay class (1-5)
pt	10	Percent of the main trunk on the ground (1-5)
dt	10	Distance (m) to nearest overstory tree ( $\geq 7.5$ cm DBH)
dbh*	10	Diameter at breast height (cm) of overstory tree at breast height
du*	10	Distance (m) to nearest understory tree ( $\leq 7.5$ cm DBH, $> 2$ m tall)
pg	2	Percent green vegetation (%)
plo	2	Percent logs (%)
pl*	2	Percent leaf litter (%)
cwd*	2	Percent twigs and brush (%)
po	2	Percent other (%)

\*Indicated variables retained for candidate models following removal of correlated covariates.

Table 2. Akaike's Information Criterion corrected for small sample sizes ( $AIC_c$ ), difference of  $AIC_c$  between the model with the lowest  $AIC_c$  ( $\Delta AIC_c$ ), and model weights ( $w_i$ ) for the best conditional logistic regression models for environmental data from Eastern Box Turtle microhabitat selection within the Oak Openings Preserve during 2012. The most parsimonious model is emboldened; models beyond the two-point  $\Delta AIC_c$  deviation cutoff are shown for comparison.

Model <sup>a</sup>	$AIC_c$	$\Delta AIC_c$	$w_i$
<b><i>sh + ld + dl + pl</i></b>	425.272	0.000	0.758
<i>el + st + ld + dl + pl</i>	428.509	3.237	0.150
<i>ld + pl + cwd</i>	431.400	6.127	0.035
<i>el + sh + ld + pl + cwd</i>	431.682	6.410	0.031
<i>ld + dl + pl</i>	431.997	6.724	0.026

<sup>a</sup>sh = surface humidity, st = surface temperature, ld = litter depth, dl = distance to log, pl = percent litter, el = elevation, cwd = coarse woody debris.

Table 3. Habitat use, availability, and preference based on results from compositional analysis for Eastern Box Turtles in the Oak Openings Preserve of northwestern Ohio from 2012-2014. Habitats that share an underline were equally preferred or avoided based on pairwise t-tests ( $P < 0.05$ ).

Category	<i>N</i>	%Use/%Available						Preferred ↔ Avoided <sup>a</sup>	Statistics		
		Residential (R)	Savanna (S)	Wet Forest (W)	Conifer (C)	Upland Deciduous (U)	Open (O)		<i>A</i> <sup>b</sup>	<i>F</i>	<i>P</i>
2012 2 <sup>nd</sup> order	13	2.3/5.2	6.5/5.6	46.3/33.5	9.0/14.0	23.0/27.5	12.8/11.4	W <u>U</u> <u>S</u> <u>O</u> <u>C</u> <u>R</u>	0.1238	12.74	0.0007
2012 3 <sup>rd</sup> order	13	5.2/9.4	5.6/2.6	33.5/34.8	14.0/18.0	27.5/26.3	11.4/8.2	<u>U</u> <u>W</u> <u>R</u> <u>O</u> <u>S</u> <u>C</u>	0.5336	1.57	0.2611
2013 2 <sup>nd</sup> order	14	0.0/6.6	13.7/4.3	29.3/23.7	17.4/18.8	24.3/21.3	15.3/25.4	<u>S</u> <u>W</u> <u>U</u> <u>C</u> <u>O</u> <u>R</u>	0.0463	37.08	<0.0001
2013 3 <sup>rd</sup> order	14	6.6/12.1	4.3/2.5	23.7/27.6	18.8/18.7	21.3/23.1	25.4/16.0	<u>O</u> <u>W</u> <u>U</u> <u>S</u> <u>R</u> <u>C</u>	0.3207	3.81	0.0393
2014 2 <sup>nd</sup> order	12	0.0/6.6	10.3/4.0	35.5/28.9	13.4/17.6	21.4/24.5	19.5/17.5	<u>W</u> <u>S</u> <u>O</u> <u>C</u> <u>U</u> <u>R</u>	0.0262	52.13	<0.0001
2014 3 <sup>rd</sup> order	12	6.6/16.8	4.0/1.9	28.9/26.1	17.6/13.3	24.5/20.0	17.5/21.9	<u>W</u> <u>U</u> <u>S</u> <u>R</u> <u>C</u> <u>O</u>	0.4066	2.04	0.1889

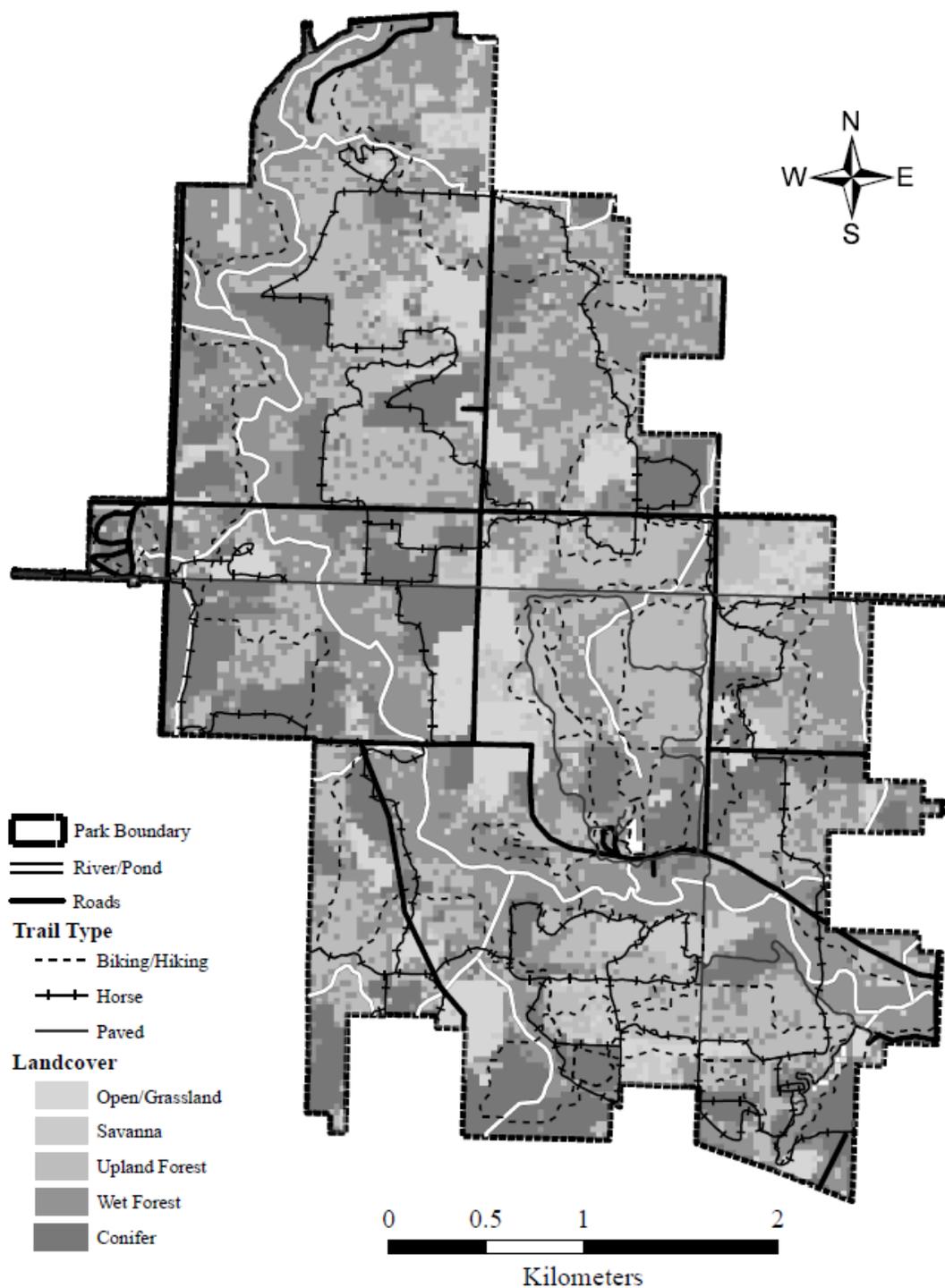


Figure 1. Study area for daily movements, home range, and, multi-scale habitat selection study of Eastern Box Turtles in the Oak Openings Preserve Metropark, Lucas County, Ohio from 2012-2014.

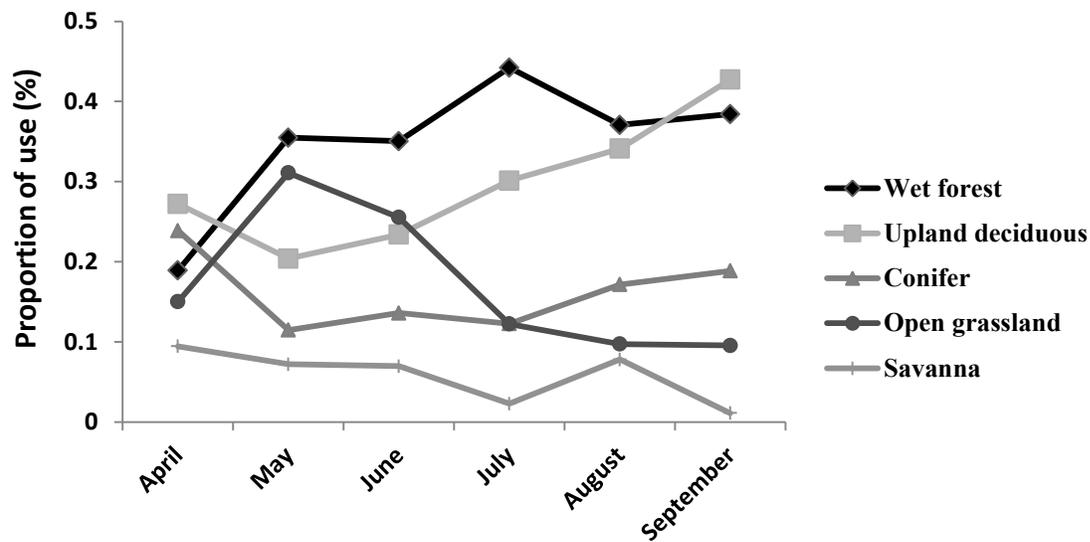


Figure 2. Proportion of main habitat types used per month by Eastern Box Turtles in the Oak Openings Preserve Metropark from 2012-2014.



Figure 3 A-C. Distribution and overlap of all 100% MCP home ranges for Eastern Box Turtles tracked at the Oak Openings Preserve from 2012-2013 including turtle ID, sex (M = male, F = female), and home range size.

CHAPTER II: HABITAT SUITABILITY MODELING FOR EASTERN BOX TURTLES  
(*TERRAPENE C. CAROLINA*) IN THE OAK OPENINGS REGION OF NORTHWESTERN  
OHIO

**ABSTRACT**

Eastern Box Turtles (*Terrapene c. carolina*) are a species on the decline throughout their range and are one of nine species of conservation interest in the Oak Openings Region of northwestern Ohio. To aid local conservation efforts, we used Eastern Box Turtle presence data from 2006-2014 and the program Maxent to build general, yearly and monthly models predicting probability of occurrence for this species in the Oak Openings Region. We then examined the amount of overlap between models to identify areas where shifts are likely to occur. Variables contributing to model performance included canopy cover, landcover, distance to forest edge, percent impervious surface, NDVI, soil composition, and slope. Amount of range overlap between models varied greatly, but there was the least amount of overlap among higher probability of occurrence sites, suggesting more pronounced temporal shifts in core areas. Analysis of the distribution of occurrence probability quantiles (0-100) revealed there to be a disproportionate amount of highly-ranked habitat within protected areas, particularly the Oak Openings Preserve. Our results highlight critical areas for box turtles in this region and can be used by land managers to designate priority areas or to guide timing of management activities (e.g., mowing or prescribed burning) that might have a negative impact on box turtles. Future conservation planning efforts for Eastern Box Turtles in the Oak Openings Region will need to address management issues on public and private lands.

## INTRODUCTION

As a taxa, reptiles are globally imperiled with one in five species threatened with extinction (Böhm et al. 2013). Major threats to reptiles include habitat loss and degradation, invasive species, pollution, disease, unsustainable use, and global climate change (Gibbons et al. 2000). Life history characteristics such as delayed sexual maturity, high juvenile mortality and subsequent low recruitment, low fecundity and the need for high adult survivorship (Wilbur and Morin 1988; Congdon et al. 1993) make chelonians particularly vulnerable. Additionally, many turtle species have complex habitat requirements, thus making habitat loss and degradation especially problematic (Mitchell and Kelmens 2000). North America, which contains approximately 20% of the world's population of turtle species, has experienced loss of 95 to 98% of the virgin forests, 99% of which includes the turtle-rich eastern deciduous forest biome (Postel and Ryan 1991; Whitney 1994; Mitchell and Klemens 2000).

Eastern box turtles (*Terrapene carolina carolina*, hereafter EBT) are a terrestrial turtle that has experienced range-wide declines over the last half century (Stickel 1978; Williams and Parker 1987; Hall et al. 1999; Dodd 2001). In Ohio, EBT are a species of special concern (Wynn and Moody 2006; ODNR Division of Wildlife 2012) and they are one of nine species selected as model organisms within the Oak Openings Region (hereafter, OOR) to guide conservation and land management efforts (T. Schetter, Metroparks of the Toledo Area, *personal communication*). A conservation goal for this region is to increase the amount of protected area so that 20% of the OOR is in preserves (Abella et al. 2007). However, there is a paucity of information regarding the status, distribution and ecology of box turtles within the region, one of the only locations in northwest Ohio where EBT are found (Wynn and Moody 2006).

A holistic conservation strategy for turtles will include understanding movement patterns, habitat use, demographic responses to perturbations and regional distribution of populations and habitats (Mitchell and Klemens 2000). Often the first step in species conservation planning is setting aside critical habitat for the species in question (Possingham et al. 2006). Protecting natural habitats has been suggested as the only real long-term solution to box turtle conservation (Dodd 2001). A number of different methods are available to delineate a species' habitat including multiple and generalized linear regression, neural networks, ordination, Bayesian models, locally weighted approaches (e.g. GAM), environmental envelopes, niche factor modeling, etc. (Guisan and Zimmermann 2000). One of the many tools available to aid in predicting species' distributions are species distribution models (SDMs), which have become an increasingly popular method to test ecological hypotheses about the distribution of organisms (Guisan and Thuiller 2003).

The goal of this study was to use SDMs to identify areas of ecological importance for Eastern Box Turtles that could be prioritized for conservation-planning efforts in the Oak Openings Region. Specifically, we sought to use turtle presence data to: 1) predict their geographic distribution in the region; 2) identify habitat associations and landscape features influencing these distributions; 3) relate the resulting predicted distribution patterns to patterns of current land ownership; and 4) examine predicted distribution patterns and their influencing features temporally.

## **METHODS**

### *Study Area*

This study took place within the OOR located in Fulton, Henry and Lucas counties of northwestern Ohio (Figure 1). The region is a 40,000 ha area that contains one of a few

landscape-scale oak savanna systems in the Midwestern United States (Schetter and Root 2011). The Oak Openings Region is a biodiversity hotspot (EPA 2016; Schetter and Root 2011) and supports five globally vulnerable or imperiled plant communities (Faber-Langendoen 2001). This region is home to 143 state endangered, threatened or potentially threatened plant species (ODNR Division of Natural Areas and Preserves 2008), 24 state endangered, threatened or of concern animal species (ODNR Division of Wildlife 2012), including the federally listed Karner blue butterfly (*Lycaeides melissa samuelis*). In total, the region accounts for less than 0.5% of Ohio's total land area with approximately 27% of it thought to exist in a natural or semi-natural state (Schetter and Root 2011).

Protected land in the OOR primarily falls under the ownership of the Ohio Department of Natural Resources, The Nature Conservancy, and Metroparks of the Toledo Area. These protected lands make up approximately 8.1% of the OOR, 3.2% of which is attributed to the Oak Openings Preserve Metropark, the largest protected area in the region.

#### *Habitat Suitability Modeling*

For our study we used the program, Maxent (version 3.3.1; Phillips et al. 2006), to investigate the distribution of suitable box turtle habitat within the OOR. We chose Maxent because it has demonstrated efficacy for distribution modeling when compared to more traditional statistical approaches (e.g. generalized linear models; Olden et al. 2008), particularly when presence-only data are used (Phillips et al. 2006; Dudik et al. 2007; Phillips and Dudik 2008) or when using small datasets (Hernandez et al. 2006; Papes and Gaubert 2007; Wisz et al. 2008). The method of using presence-only data is particularly useful when dealing with cryptic species, such as box turtles, where detectability is often low (Refsnider 2011; Kapfer et al. 2012) and failing to detect the presence of an organism does not necessarily mean the organism is not

present. Maxent uses the concept of maximum entropy, in which the distribution of a species will tend toward a uniform distribution after all ecologically important parameters are taken into account (Phillips et al. 2004). Maxent uses a machine learning approach to predict the probability of occurrence of a species given equal sampling across location data (Phillips et al. 2006). In addition to generating probability distribution maps, Maxent makes it possible to assess which habitat variables have the most predictive power.

### *Presence Data*

Turtle presence data came from an ongoing telemetry study (2012-2014) at the Oak Openings Preserve (OOP) Metropark (M. Cross, Bowling Green State University, unpublished data) and from reported sightings throughout the OOR (G. Lipps, The Ohio State, unpublished data) dating back to 2006. Using geographically clustered data, such as that from radio-telemetry, can cause overfitting (i.e., when models fit too tightly to calibration data) and inflate model performance estimates (Veloz 2009; Hijmans et al. 2012; Boria et al. 2014). To reduce the amount of spatial autocorrelation among our dataset, we applied a spatial filter (Boria et al. 2013) to our data points using the “Spatial Rarefy” tool in the SDMtoolbox (Brown 2014). We specified a distance between points of 75 m, more than three times the largest average distance moved between telemetry locations across years (Wiley 2010). The resulting dataset was greatly reduced and accounted for 16.7% of the original presence data.

### *Background Points*

When using SDMs, many algorithms require absence, pseudo-absence, or background points. In cases, where absence or pseudo-absence data are unnecessary (e.g., Maxent) or unavailable (e.g., for cryptic species), background points can be substituted. Background points do not attempt to determine absence locations, but instead characterize the available

environments within a region of interest (Phillips et al. 2009). Typical selection methods of background points involving user-defined areas of interest often contain habitats that may be environmentally suitable, but were never colonized by the target species. This selection method tends to use less informative background points and can lead to increases in commission errors (e.g. “false positives”) and overfitting (Anderson Raza 2010; Barbet-Massin et al. 2012). To avoid these problems, it is recommended to limit the focal region to areas the study organism might inhabit by buffering the data points and selecting background points from the organism-defined area. We created a buffer of 2,430 m, the longest home range length of any turtle in our study, around all of our presence points and sampled background points from within the new area. However, as we wished to examine the potential distribution of box turtles within a larger region, we allowed our models to predict EBT occurrence probability outside of the sampled range.

#### *Habitat Suitability Variables*

All remote-sensing data used for the habitat suitability model were obtained from the USDA NRCS (<http://datagateway.nrcs.usda.gov/>) and were at a 30 m resolution, unless otherwise noted. Editing and manipulation of these data layers took place in ArcGIS version 10.1 (ESRI Inc., Redlands, CA, USA). Because the study area encompassed multiple counties, all raster and vector datasets had to be mosaicked or merged (raster vs. vector, respectively) prior to editing.

For our SDM we incorporated publicly-available landscape variables that we thought were likely to influence the distribution of box turtles within our study area. Variables included aspect (sin and cosine), canopy density, compound topographic index (“wetness index”; CTI), canopy height, elevation, distance to forest edge, distance to water, normalized difference

vegetative index (NDVI), organic content of the soil, percent impervious surface, Shannon's diversity index of habitat (SHDI), solar insolation, slope, and surface curvature.

Data for elevation (digital elevation model; DEM), landcover (Schetter and Root 2011) and soil type required no manipulation. All topographic and surface variables (slope, aspect, solar insolation, CTI, and surface curvature) used in this study were derived from a base DEM layer using the respective tools in the Spatial Analyst or Geomorphometric and Gradient Metrics toolboxes (CTI; Evans et al. 2014). Slope and aspect are used in the calculation of solar insolation and CTI, which are surrogates for surface temperature and humidity, respectively.

Canopy height and density were derived from Lidar data (OGRIP: <http://ogrip.oit.ohio.gov/Home.aspx>; 2.1 m post spacing). We created digital surface (DSM) and digital terrain models (DTM) from first-return and ground return points, respectively. Canopy height was calculated by subtracting the DTM from the DSM. Canopy density was calculated by dividing the number of above ground returns per cell (pixel) by the total returns per cell to get above ground density. Both height and density rasters were then resampled to a 30 m resolution to be consistent with the rest of the data.

Distance to water, roads and edge rasters were created using the Euclidean Distance tool which uses the respective shapefile and the DEM to create an intermittent "dummy raster" with the same extent as the DEM. The pixels of the dummy raster are populated with the straight-line distance values from each individual pixel to the shapefile, resulting in a raster depicting the distance to the input feature. We defined water as any open or moving body of water; road as any paved road; and edge as the boundary between "open" habitat (wet prairie, savanna, scrub/shrub, barrens, Eurasian meadow, prairie and cropland) and "closed" habitat (swamp forest, coniferous forest, deciduous forest and floodplain forest (as per Blouin-Demers and Weatherhead 2001)).

We included two measures of biodiversity: NDVI and SHDI. Normalized difference vegetative index was calculated in ArcGIS using the equation:

$$NDVI = \frac{(NIR - VIS)}{(NIR + VIS)}$$

Where “NIR” is reflectance in the near infrared band and “VIS” is the reflectance in the visible red band. This measure gives an estimate of vegetative productivity, and can also provide information about the temporal distribution of vegetative communities, vegetation biomass, food quality for herbivores, extent of habitat degradation, biodiversity, species distributions, and movement patterns of animals (*reviewed in* Pettorelli et al. 2005; Pettorelli et al. 2011). Images for the NDVI calculation were obtained from the Ohio Geographically Referenced Information Program’s website (OGRIP 2011-2014; <http://ogrip.oit.ohio.gov/Home.aspx>). Shannon’s diversity index was calculated for the landuse/landcover map using Fragstats v.4.2 (McGarigal et al. 2012). This metric provides an estimate of landscape diversity based on landcover types where values are larger where more landcover types are present and smaller where fewer exist (Güthlin et al. 2013).

All vector data were converted to raster format. Because there were some discrepancies between the rasters created from the vector data, all vector-based rasters were resampled so the pixels would align with the DEM-derived rasters. Finally, all of the raster data were converted into American Standard Code for Information Interchange (ASCII) format for use in the SDM.

#### *Selection of Environmental Variables, Regularization Multiplier, and Model Evaluation*

Using the default settings of Maxent can lead to inappropriate model complexity, overfitting, reduced ability to infer relative importance of variables, and result in poor predictive power (Warren and Seifert 2011, Radosavljevic and Anderson 2014). Merow et al. (2013) identified six key decisions when selecting input data and Maxent settings that should be made

based on the ecology of the species in question: background data, environmental features, regularization, sampling bias, types of output and evaluation methods. We addressed potential factors contributing to overfitting among background data and sampling bias in previous section (see “Presence Data” and “Background Points”). To reduce overfitting our Maxent models, we used the R package “MaxentVariableSelection” (Jueterbock 2015; Jueterbock et al. 2016) to determine the regularization multiplier ( $\beta$ ), choose environmental variables, and evaluate the models. The MaxentVariableSelection package (Jueterbock 2015; Jueterbock et al. 2016) is designed to reduce model complexity by identifying the least correlated set of environmental predictor variables, fine-tune the regularization multiplier, and assessing model performance through a combination of AIC (Akaike 1974) and AUC (Fielding and Bell 1997). Because models performance estimated by AUC can lead to overfitting (Jiménez-Valverde 2012), we selected the model with the lowest AIC<sub>c</sub> as the method of selecting the best model parameters. To further assess model overfitting, we estimated the difference between AUC values from test and training data (Warren and Seifert 2011). The variable selection process was performed for a range  $\beta$  values ranging from 1-4 in increments of 0.5 (Jueterbock et al. 2016). This range was suggested by Radosavljevic and Anderson (2014) to reduce model overfitting.

Following selection of model parameters and settings based on the results of the MaxentVariableSelection, we created the following Maxent models: a generalized model encompassing all survey years (2006-2012; hereafter, general model), yearly models for 2012-2013, and monthly models for the 2012-2014 active seasons (April-September). We ran Jackknife procedures for each variable on regularized training gain, test gain, and area under the curve (AUC) of a receiver-operating characteristic graph of sensitivity versus 1-specificity. Response curves were graphed to assess how each variable affected the model. Plotting

sensitivity by 1-specificity, the AUC indicated the ability to the model to predict the species' distribution. Models with AUC values greater than 0.5 indicated a better-than-random prediction, with increased efficiency as AUC values approached 1.0 (Swets 1988). All models were created using cross-validation to generate training points.

#### *Distribution Across Probability Classes*

Following completion of the Maxent runs, we further analyzed the proportion of probability distribution classes within the study area. Within ArcGIS we converted the Maxent's probability of occurrence distribution and created 10% probability classes (0-100%). This was done for the OOR, the protected lands within the OOR, and the Oak Openings Preserve Metropark. We also created a binary map from the general model depicting the predicted distribution of box turtles using the minimum sensitivity plus specificity logistic threshold from the Maxent output. This threshold minimizes the mean error rate for positive and negative observations; roughly equivalent to finding the point on ROC curves whose tangent has a slope of one (Cantor et al. 1999; Freeman and Moisen 2008) and provides a minimum occurrence of probability value.

#### *Monthly Shifts*

To explore month-to-month shifts in box turtle occurrence probability we used ENMTools (Warren et al. 2008; Warren et al. 2010) to measure range overlap from the models produced by Maxent. ENMTools interfaces with Maxent to perform measurements on range overlap between two or more distribution models. This allows the user to select probability of occurrence thresholds against which to test the overlap between two or more predicted distributions. We specified three levels of occurrence probability from Maxent's average distribution model for all of our models: low ( $\geq 0.29$ ), medium ( $\geq 0.50$ ), and high ( $\geq 0.75$ ). The

threshold for the lowest level was set using the minimum sensitivity plus specificity logistic threshold value from the Maxent output

## RESULTS

### *Model Performance and Importance of Environmental Variables*

The general model with the lowest  $AIC_c$  was built with a beta-multiplier of 1.5, and four uncorrelated variables with a model contribution of  $>5.0\%$  (Tables 1 & 2): canopy cover, elevation, distance to forest edge, NDVI, landcover, organic content of the soil, and percent sand in the soil. Landcover contained the most information by itself whereas elevation had the most information not contained in other variables. The average AUC of the general model (based on 10 replicate runs), was 0.74, suggesting that the model could adequately discriminate between presence and background locations. As indicated by the low AUC.Diff value (0.03, Table 1), the model was not overfit to the presence locations (Warren and Seifert 2011).

The model parameters, important variables, and their contribution to the models for yearly and monthly models varied (Tables 1 & 3). While the number of variables within models ranged from 2–17, AUC.Diff values were low for all models and ranged from 0.01–0.09 (Table 1). Variables contributing highly to models included canopy cover, distance to forest edge, NDVI, landcover, and percent sand, and to a lesser extent, aspect, percent impervious surface, percent soil organic content, and slope (Table 3). Landcover, NDVI, and canopy cover were consistently selected as the variables that contained the most information by themselves or contained the most information not present in other variables (Table 3). Landcover types most associated with increases in model performance were savanna, upland deciduous, wet forests, and open grasslands.

### *Distribution Across Probability Classes*

Total predicted area for Eastern Box Turtles, based on a minimum probability of occurrence value, made up 20% of the OOR (Figure 2). The general Maxent model (Figure 3) indicated that a large proportion of the available landscape fell into the 0-10% quantile range while approximately 3.57% of the OOR fell into the  $\geq 50\%$  categories (Table 2). Within the protected areas there was a less skewed distribution of probability of occurrence values (Figure 4 A—C) and 23.26% of these areas fell into the  $\geq 50\%$  categories. Areas ranked as medium- to high-probability of occurrence made up 50% of the Oak Openings Preserve. The average probability of occurrence value for turtles within our study area was  $57.04 \pm 21.33$  (SD).

### *Monthly Shifts*

Maxent SDM maps of month-to-month occurrence probability showed similar core areas to the full model, but differed visually in distribution of intermediate occurrence probability (due to the potential of box turtles being illegally collected, we do not present month-to-month distribution maps). Monthly overlap for sites with low ( $>0.29$ ) probability of occurrence averaged 0.76 and ranged from 0.46 to 1.00 (Table 5). Monthly overlap for sites with medium probability of occurrence ( $> 0.50$ ) averaged 0.59 and ranged from 0.36 to 1.00 (Table 6). Monthly overlap for sites with high probability of occurrence ( $> 0.75$ ) averaged 0.20 and ranged from 0.00 to 1.00 (Table 7). In general, the amount of overlap decreased as the probability of occurrence threshold increased with high probability sites overlapping the least, indicating more of a shift in physical area. Intra-year overlap (within) was greatest for medium- and high-ranked sites between the months of May-July (Tables 6 & 7).

## DISCUSSION

Our results show that high-probability sites for Eastern Box Turtle habitat were relatively rare within our study region, but the proportion of high-probability of occurrence locations was higher within protected areas, particularly the Oak Openings Preserve Metropark. Hotspots for this species appear to be confined to the larger sections of contiguous land. Given the limited amount of high-ranked habitat, our results suggest that focusing conservation efforts on these areas could be an effective preliminary method of managing the OOR for Eastern Box Turtles.

High AUC and low AUC.Diff values verified that our Maxent models effectively characterized suitable areas for Eastern Box Turtles across years and that our models were not overfit (Swets 1988; Warren and Seifert 2011). We were unable to test our models with an independent dataset, but for a cryptic species like box turtles, where detection probabilities are low (Refsnider et al. 2011; Kapfer et al. 2012), this sort of dataset would be difficult to come by. We considered using sightings from park-goers as an independent dataset from which to test our model assumptions, but access in the parks is limited to roads and trails and would likely be biased to these features. The SDMToolbox and MaxentVariableSelection package allowed us to improve model accuracy by selecting parameters that would further reduce overfitting. Using this method, we were able to gain insight into the temporal distributions of Eastern Box Turtles in the Oak Openings Region and the landscape variables associated with shifts in area use.

The results of our study show that turtles within the Oak Openings Region predominantly select habitats based on landcover and canopy cover, but also NDVI, soil type, elevation, and distance to forest edge. Elevation, slope, substrate type, habitat type, surface humidity, surface temperature have been shown to be important variables for predicting box turtle movements and presence (Claussen et al. 2002; Chapter I). Box turtles in general prefer mesic woodlands with

relatively closed canopy, well-defined leaf litter, moisture-retaining soils and moderate understory vegetation (Dodd 2001; Ernst and Lovich 2009). The peaks in model performance at savanna, swamp forest, conifer, upland forest, floodplain forest and prairie are typical of this species (Dodd 2001) and corroborate the habitat-selection findings of box turtles in the OOR (Chapter I; Wilson 2012). While landcover was a dominant variable across all our models, it is important to note that the landcover map we used (Schetter and Root 2011) was designed specifically for the OOR. In areas outside of our study region, other landscape variables may play a more important role in determining Eastern Box Turtle distributions.

It was not unexpected to find that a majority of the OOR landscape was classified as having a low probability of occurrence. Much of the landscape has been altered by agricultural conversion, draining of prairies, fire suppression, and urban sprawl (Schetter and Root 2011). It is important to note that protected areas account for 8.1% of the OOR yet they contain a disproportionate amount of habitat considered by our general model to have a high occurrence probability. Perhaps even more interesting is the Oak Openings Preserve accounts for 3.2% of the OOR and it appears to contain some of the highest-ranked habitat. Within protected areas that contain high-probability of occurrence areas, it becomes imperative to manage habitat in a manner that reduces impacts on box turtles. This is particularly true within the Oak Openings Preserve, which appears to be the epicenter of high-probability box turtle habitat within the OOR. The preserve, and the protected areas within the region, are managed through prescribed fire, mowing, herbicide application and manual removal (Peterson and Reich 2001; Abella et al. 2007). While the full effects of various management practices on box turtles are equivocal (Dodd and Dreslik 2008; Platt et al. 2010, Currylow et al. 2012; M. Cross Chapter IV), these activities have the potential to disrupt habitat contiguity and alter movement patterns.

Monthly Maxent models and ENMTool analysis showed visible shifts in monthly occurrence probability as well as differences between months in range overlap. Intra-year shifts reflected the changes in habitat selection as the turtles conduct their seasonal movements through their home ranges. Seasonal shifts in habitat associations are well-documented for box turtles (Dodd 2001; Frederickson 2014; Wilson 2012; M. Cross *Chapter 1*) and in general, Eastern Box Turtles at our study site tended to use upland deciduous and wet forests consistently throughout the active season with use of wet forests increasing during warmer months; use of open grasslands peaked during the months of May—June and then decreased as the season progressed. Shifts in habitat selection are likely governed by resource availability including prey-acquisition, location of suitable nesting sites, mate-searching, and availability of microhabitat features promoting form construction (Penick et al. 2001; Rossell et al. 2006; Iglay et al. 2007; Nazdrowicz et al. 2008; McKnight 2011).

Our inter- and intra-yearly distribution models can be used by land managers in the Oak Openings Region to predict where box turtles are likely to occur at a given time of year and plan management practices accordingly. Timing of disturbance is important when considering the long and short-term effects on box turtle populations; disturbance effects are more likely to have a pronounced impact on populations if it occurs prior to oviposition (Dodd et al. 2016). One of the more popular suggestions to reduce management-related mortality of reptiles and amphibians is to conduct management activities during periods of ectothermic dormancy (Dodd 2001; Platt et al. 2010; Kingsbury and Gibson 2012). However, the ecological benefits from activities, such as prescribed fire, are greatly reduced when burning during these suggested periods (Knapp et al. 2009). Using our monthly models managers in the OOR could identify box turtle hotspots to

avoid or target at times of year when probability of occurrence is high or changing to minimize the impact of management activities on the population.

The results of this study represent an initial effort to examine the distribution and habitat associations of Eastern Box Turtles in the Oak Openings Region. It is important to note that a majority of the points used (~80%) came from the Oak Openings Preserve, which is unique in size and habitat when compared to the other Metroparks of the Toledo area. As such, it is possible our results are biased towards this site. However, our results are consistent with those of Lipps (2010), and improve upon this effort by providing maps depicting sites of variable probability of occurrence which can be used by managers to target specific areas instead of large tracts of land. Additionally, this method provides a means of which to explore box turtle distributions under projected climate change scenarios. Combining the landscape-scale distribution and habitat associations presented here with micro- and macro-habitat associations of Eastern Box Turtles (M. Cross, Chapter I) should provide managers in the region with a multi-scale, holistic guide from which to base conservation and management decisions. Future research should seek to test and validate the distribution patterns documented herein.

The amount of high-ranked area contained within protected habitats indicates that current measures to conserve prime habitat for Eastern Box Turtles are succeeding. However, a large amount of potential habitat remains outside of protected areas and conservation efforts will need to include public and private lands. Additionally, low probability of occurrence sites should not be immediately discounted as they may play an important role in dispersal between highly-ranked habitats. Thus, development on private lands may play a significant role in future management of box turtle habitat in this region. Our models can be used to identify priority areas within these habitats or target high-ranked habitat for future land acquisition. With the goal to

increase the amount of protected land in the region, box turtle conservation efforts would be best suited if priority was given to medium or high probability of occurrence sites or to areas that might contribute to landscape connectivity. Given the amount of fragmentation and obstacles between highly-ranked sites, road mortality and landscape connectivity are likely to be issues governing long-term conservation of Eastern Box Turtles in the Oak Openings Region.

### **ACKNOWLEDGMENTS**

Support and funding for this project was provided by Bowling Green State University, The Metroparks of the Toledo Area, The Toledo Zoo, The Ohio Biological Survey and The Toledo Naturalist's Association. We thank our field assistants H. Clendenin, J. Cross, A. Klosinski, P. Lund, H. Shiedler, C. Cunningham, and C. Ritzenthaler for their time in the field. Additional thanks are owed to P. Tolson, C. L. Ellsworth, E. Gomezdelcampo, K. Menard and A. Selden for logistic support. Animals in this study handled in compliance with Bowling Green State University's Institutional Animal Care and Use Committee guidelines (IACUC Approval # 12-005 and the regulations outlined by the Ohio Department of Natural Resources Wild Animal Permit (#15-48) and the Metroparks of the Toledo Area (#042512).

### **LITERATURE CITED**

- Abella, S.R., J.F. Jaeger, and T.A. Schetter. 2007. Public land acquisition and ecological restoration: an example from northwest Ohio's Oak Openings Region. *Natural Areas Journal* 27:92-97.
- Akiake, H. 1974. A new look at the statistical modeling identification. *IEEE Transactions on Automatic Control* 19:716-723.
- Blouin-Demers, G., and P.J. Weatherhead. 2001. Habitat use by black rat snakes (*Elaphe obsoleta obsoleta*) in fragmented forests. *Ecology* 82:2882 – 2896.

M. Böhm, B. Collen, J.E.M. Baillie, P. Bowles, J. Chanson, N. Cox, G. Hammerson, M. Hoffmann, S.R. Livingstone, M. Rama, A.G.J. Rhodin, S. N. Stuart, P. P. van Dijk, B.E. Young, L.E. Afuang, A. Aghasyan, A. Garcia, C. Aguilar, R. Ajtic, F. Akarsu, L.R.V. Alencar, A. Allison, N. Ananjeva, S. Anderson, C. Andren, D. Ariano-Sanchez, J.C. Arredondo, M. Auliya, C.C. Austin, A. Avci, P. J. Baker, A. F. Barreto-Lima, C. L. Barrio-Amoros, D. Basu, M.F. Bates, A. Batistella, A. Bauer, D. Bennett, W. Bohme, D. Broadley, R. Brown, J. Burgess, A. Captain, S. Carreira, M. del Rosario Castaneda, F. Castro, A. Catenazzi, J.R. Cedeno-Vazquez, D.G. Chappl, M. Cheylan, D. F. Cisneros-Heredia, D. Cogalniceanu, H. Cogger, C. Corti, G. C. Costa, P. J. Couper, T. Courtney, J. Crnobrnja-Isailovic, P. Crochet, B. Crother, F. Cruz, J. C. Daltry, R.J. R. Daniels, I. Das, A. de Silva, A. C. Diesmos, L. Dirksen, T. M. Doan, C. K. Dodd Jr., J.S. Doody, M. E. Dorcas, J. D. de Barros Filho, V. T. Egan, E. H. El Mouden, D. Embert, R. E. Espinoza, A. Fallabrino, X. Feng, Z. Feng, L. Fitzgerald, O. Flores-Villela, F. G.R. Franca, D. Frost, H. Gadsden, T. Gamble, S.R. Ganesh, M. A. Garcia, J. E. Garcia-Perez, J. Gatus, M. Gaulke, P. Geniez, A. Georges, J. Gerlach, S. Goldberg, J. T. Gonzalez, D.J. Gower, T. Grant, E. Greenbaum, C. Grieco, P. Guo, A. M. Hamilton, K. Hare, S. B. Hedges, N. Heideman, C. Hilton-Taylor, R. Hitchmough, B. Hollingsworth, M. Hutchinson, I. Ineich, J. Iverson, F. M. Jaksic, R. Jenkins, U. Joger, R. Jose, Y. Kaska, U. Kaya, J. S. Keogh, G. Kohler, G. Kuchling, Y. Kumlutas, A. Kwet, E. La Marca, W. Lamar, A. Lane, B. Lardner, C. Latta, G. Latta, M. Lau, P. Lavin, D. Lawson, M. LeBreton, E. Lehr, D. Limpus, N. Lipczynski, A. S. Lobo, M. A. Lopez-Luna, L. Luiselli, V. Lukoschek, M. Lundberg, P. Lymberakis, R. Macey, W. E. Magnusson, D. L. Mahler, A. Malhotra, J. Mariaux, Bryan Maritz, Otavio A.V. Marques, R. Marquez, M. Martins, G.

- Masterson, J. A. Mateo, R. Mathew, N. Mathews, G. Mayer, J. R. McCranie, G. J. Measey, F. Mendoza-Quijano, M. Menegon, S. Metrailler, D. A. Milton, C. Montgomery, S. A.A. Morato, T. Mott, A. Munoz-Alonso, J. Murphy, T. Q. Nguyen, G. Nilson, C. Nogueira, H. Nunez, N. Orlov, H. Ota, J. Ottenwalder, T. Papenfuss, S. Pasachnik, P. Passos, O.S.G. Pauwels, N. Perez-Buitrago, V. Perez-Mellado, E. R. Pianka, J. Pleguezuelos, C. Pollock, P. Ponce-Campos, R. Powell, F. Pupin, G. E. Quintero Diaz, R. Radder, J. Ramer, A. R. Rasmussen, C. Raxworthy, R. Reynolds, N. Richman, E. L. Rico, E. Riservato, G. Rivas, P. L.B. da Rocha, M. Rodel, L. Rodriguez Schettino, W. M. Roosenburg, J. P. Ross, R. Sadek, K. Sanders, G. Santos-Barrera, H. H. Schleich, B. R. Schmidt, A. Schmitz, M. Sharifi, G. Shea, H. Shi, R. Shine, R. Sindaco, T. Slimani, R. Somaweera, S. Spawls, P. Stafford, R. Stuebing, S. Sweet, E. Sy, H. J. Temple, M. F. Tognelli, K. Tolley, P. J. Tolson, B. Tuniyev, S. Tuniyev, N. Uzumae, G. van Buurt, M. Van Sluys, A. Velasco, M. Vences, M. Vesely', S. Vinke, T. Vinke, G. Vogel, M. Vogrin, R. C. Vogt, O. R. Wearn, Y. L. Werner, M. J. Whiting, T. Wiewandt, J. Wilkinson, B. Wilson, S. Wren, T. Zamin, K. Zhou, and G. Zug. 2013. The conservation status of the world's reptiles. *Biological Conservation* 157:372-385.
- Boria, R.A., L.E. Olson, S.M. Goodman, R.P. Anderson. 2014. Spatial filtering to reduce sampling bias can improve the performance of ecological niche models. *Ecological Modeling* 275: 73-77.
- Brown, J.L. 2014. SDMtoolbox: a python-based GIS toolkit for landscape genetic, biogeographic and species distribution model analyses. *Methods in Ecology and Evolution* 5:694-700.
- Cantor, S.B, C.C. Sun, G. Tortler-Luna, R. Richards-Kortum, and M. Follen. 1999. A

- comparison of C/B ratios from studies using receiver operating characteristic curve analysis. *Journal of Clinical Epidemiology* 52:885-892.
- Claussen, D.L., R. Lim, M. Kurz, and K. Wren. 2002. Effects of slope, substrate, and temperature on the locomotion of the ornate box turtle, *Terrapene ornata*. *Copeia* 2002:411-418.
- Congdon, J.D., A.E. Dunham, and R.C. Van Loben Sels. 1993. Delayed sexual maturity and demographics of Blanding's turtles (*Emydoidea blandingii*): implications for conservation and management of long-lived organisms. *Conservation Biology* 7:826-833.
- Dodd, C.K., Jr. 2001. North American box turtles: a natural history. University of Oklahoma Press, Norman, Oklahoma, USA.
- Dudik, M., S.J. Phillips, and R.E. Schapire. 2007. Maximum entropy density estimation with 500 generalized regularization and an application to species distribution modeling. *Journal of Machine Learning Research* 8:1217-1260.
- Environmental Protection Agency (EPA). 2016. Oak Openings Site Conservation Plan. <https://archive.epa.gov/ecopage/web/html/oak-openings.html>. Accessed June 7, 2016.
- Ernst, C.H., and J.E. Lovich. 2009. Turtles of the United States and Canada, 2<sup>nd</sup> edition. The Johns Hopkins University Press, Baltimore, Maryland, USA.
- Evans, J.S., J. Oakleaf, S.A. Cushman, and D. Theobald. 2014. An ArcGIS Toolbox for Surface Gradient and Geomorphometric Modeling, version 2.0-0. Available: <http://evansmurphy.wix.com/evansspatial>. Accessed: February 3, 2013.
- Faber-Langendoen, D (ed.). 2001. Plant communities of the Midwest: classification in an ecological context. Association for Biodiversity Information, Arlington, Virginia, USA.
- Fielding, A.H., and J.F. Bell. 1997. A review of methods for the assessments of prediction errors

- in conservation presence/absence models. *Environmental Conservation* 24: 38-49.
- Forman, R.T. 2000. Estimate of the area affected ecologically by the road system in the United States. *Conservation Biology* 14:31-35.
- Frederiksen, T.S. 2014. Thermal regulation and habitat use of the Eastern Box Turtle in Southwestern Virginia. *Northeastern Naturalist* 21: 554-564.
- Freeman, E.A., and G.G. Moisen, 2008. A comparison of the performance of threshold criteria for binary classification in terms of predictive prevalence. *Ecological Modelling* 217:48-58.
- Gibbons, J.W., D.E. Scott, T.J. Travis, K.A. Buhlmann, T.D. Tuberville, B.S. Metts, J.L. Greene, T. Mills, Y. Leiden, S. Poppy, and C.T. Winne. 2000. *Bioscience* 50: 653-666.
- Gibbs, J.P., and W. G. Shriver. 2002. Estimating the effects of road mortality on turtle populations. *Conservation Biology* 16:1647-1652.
- Guisan, A., and W. Thuiller. 2005. Predicting species distribution: offering more than simple habitat models. *Ecology Letters* 8:993-1009.
- Guisan, A., and N.E. Zimmermann. 2000. Predictive habitat distribution models in ecology. *Ecological Modelling* 135:147-186.
- Güthlin, D., I. Storch, and H. Küchenhoff. 2013. Landscape variables associated with relative abundance of generalist mesopredators. *Landscape Ecology* 28:1687-1696.
- Hall, R.J., P. F.P. Henry, and C.M. Bunck. 1999. Fifty-year trends in a box turtle population in Maryland. *Biological Conservation* 88:165-172.
- Hernandez, P.A., C.H. Graham, L.L. Master, and D.L. Albert. 2006. The effect of sample size and species characteristics on performance of different species distribution modeling methods. *Ecography* 29:773-785.

- Hijmans, R.J. 2012. Cross-validation of species distribution models: removing spatial sorting bias and calibration with a null model. *Ecology* 93: 697-688.
- Jiménez-Valverde, A. 2012. Insights into the area under receiver operating characteristic curve (AUC) as a discrimination measure in species. *Ecology* 5:498-507.
- Jueterbock, A. 2015. R package MaxentVariableSelection: selecting the best set of relevant environmental variables along with the optimal regularization multiplier for Maxent niche modeling. <https://cran.r-project.org/web/packages/MaxentVariableSelection/index.html>.
- Jueterbock, A., I. Smolina, J.A. Coyer, and G. Hoarau. 2016. The fate of the Arctic seaweed *Fucus distichus* under climate change: an ecological niche modeling approach. *Ecology and Evolution* 6:1712-1724.
- Kapfer, J.M., D.J. Muñoz, and T. Tomasek. 2012. Use of wildlife detector dogs to study eastern box turtle (*Terrapene carolina carolina*) populations. *Herpetological Conservation and Biology* 7:169-175.
- Knapp, E.E., B.L. Estes, and C.N. Skinner. 2009. Ecological effects of prescribed fire season: A literature review and synthesis for land managers. U.S. Department of Agriculture, Forest Service General Technical Report no. PSW-GTR-224., Pacific Southwest Research Station. Albany, California, USA.
- Lipps, G.J. 2010. Landscape characteristics of eastern box turtle locations in the Oak Openings Region of northwest Ohio. Technical report to the Ohio Division of Wildlife, Columbus, Ohio.
- McGarigal, K., S.A. Cushman, and E. Ene. 2012. FRAGSTATS v4: Spatial Pattern Analysis

Program for Categorical and Continuous Maps.

<http://www.umass.edu/landeco/research/fragstats/fragstats.html>. Accessed 14 January 2014.

Mitchell, J.C., and M.W. Klemens. 2000. Primary and secondary effects of habitat alteration. Pp. 5 – 32 *In* Turtle Conservation. M.W. Klemens (ed.). Smithsonian Institution Press, Washington, D.C., USA.

[ODNR Division of Natural Areas and Preserves] Ohio Department of Natural Resources: Division of Natural Areas and Preserves. 2012. Rare native Ohio plants: 2012-2013 Status List. Available online < <http://ohiodnr.com/Portals/3/heritage/2012-13-plant-list.pdf>>. Accessed 24 April 2013.

[ODNR Division of Wildlife] Ohio Department of Natural Resources: Division of Wildlife. 2012. Wildlife that are considered to be endangered, threatened, species of concern, special interest, extirpated, or extinct in Ohio. Available online <<http://ohiodnr.com/Portals/9/pdf/pub356.pdf>>. Accessed 24 April 2013.

Olden, J.D., J.J. Lawler, and N.L. Poff. 2008. Machine learning methods without tears: A primer for ecologists. *The Quarterly Review of Biology* 83: 171-193.

Papes, M., and P. Gaubert. 2007. Modelling ecological niches from low numbers of occurrences: assessment of the conservation status of poorly known viverrids (Mammalia, Carnivora) across two continents. *Diversity and Distributions* 13: 890-902.

Phillips, S.J., R.P. Anderson, and R.E. Schapire. 2006. Maximum entropy modeling of species geographic distributions. *Ecological Modelling* 190:231 – 259.

Phillips, S.J., and M.K. Dudik. 2008. Modeling of species distributions with Maxent: new extensions and comprehensive evaluation. *Ecography* 31:161-175.

- Phillips, S.J., M.K. Dudik, and R.E. Schapire. 2004. A maximum entropy approach to species distribution modeling. Pp. 655-662 *in* Proceedings of the twenty-first international conference on machine learning. ACM. Banff, Alberta, Canada.
- Phillips, S.J., M. Dudik, J. Elith, C.H. Graham, A. Lehmann, J. Leathwick, and S. Ferrier. 2009. Sample selection bias and presence-only distribution models: implications for background and pseudo-absence data. *Ecological Applications* 19:181-197.
- Possingham, H.P., K.A. Wilson, S.J. Andelman, and C.H. Vynne. 2006. Protected areas: goals limitations, and design. Pp 509-551 *In* Principles of Conservation Biology, Third Edition. M.J. Groom, G.K. Meffe, and C.R. Carroll (eds.). Sinauer Associates, Inc., Sunderland, Massachusetts, USA.
- Refsnider, J.M., T.S. Mitchell, H.M. Streby, J.T. Strickland, D.A. Warner, and F.J. Janzen. 2011. A generalized method to determine detectability of rare and cryptic species using the ornate box turtle as a model. *Wildlife Society Bulletin* 35:93-100.
- Rodasavljevic, A., and R.P. Anderson. 2014. Making better Maxent models of species distributions: complexity, overfitting and evaluation. *Journal of Biogeography* 41:629-643.
- Stickel, L.F. 1978. Changes in a box turtle population during three decades. *Copeia* 1978:221-225.
- Swets, J.A.. 1988. Measuring the accuracy of diagnostic systems. *Science* 240:1285-1293.
- Veloz, S.D. 2009. Spatially correlated sampling falsely inflates measures of accuracy for presence-only niche models. *Journal of Biogeography* 36: 2290-2299.
- Warren, D.L., and S.N. Seifert. 2011. Ecological niche modeling in Maxent: the importance of

- model complexity and the performance of model selection criteria. *Ecological Applications* 21:335-342.
- Warren, D.L., R.E. Glor, and M. Turelli. 2008. Environmental niche equivalency versus conservatism: quantitative approaches to niche evolution. *Evolution* 62:2868-2883.
- Warren, D.L., R.E. Glor, and M. Turelli. 2010. ENMTools: a toolbox for comparative studies of environmental niche studies. *Ecography* 33:607-611.
- Wilbur, H.M., and J.P. Morin. 1988. Life history and evolution in turtles. Pp. 396-447 *In* *Biology of the Reptilia v.16 Ecology* B. C. Gans and R. Huey (eds.). Alan R. Liss, Inc., New York, USA.
- Willey, L.L. 2010. Spatial ecology of eastern box turtles (*Terrapene c. carolina*) in central Massachusetts. Ph.D. Dissertation, University of Massachusetts, Amherst, Massachusetts, USA. 222 p.
- Williams, E.C., Jr., and W.S. Parker. 1987. A long-term study of a box turtle (*Terrapene carolina*) population at Allee Memorial Woods, Indiana, with emphasis on survivorship. *Herpetologica* 43:328-335.
- Wilson, S.D. 2012. Movement and ecology of the eastern box turtle (*Terrapene carolina carolina*) in a heterogeneous landscape. M.S. Thesis. Bowling Green State University, Bowling Green, Ohio, USA. 54 p.
- Wisz, M.S., R.J. Hijmans, J. Li, A.T. Peterson, C.H. Graham, and A. Gusian. 2008. Effects of sample size on the performance of species distribution models. *Diversity and Distributions* 14:763-773.
- Wynn, D.E., and S.M. Moody. 2006. Ohio turtle, lizard and snake atlas. Ohio Biological Survey: Columbus, Ohio.

Table 1. Results of the variable selection process for the monthly Maxent models showing the beta multiplier, number of variables, number of sample locations, AIC<sub>c</sub>, and difference between the AUC of the test and training points (AUC.Diff).

Month-Year	Beta Multiplier	Variables	Samples	AIC <sub>c</sub>	AUC.Diff
General	1.5	17	343	8530.3	0.03
2012	1.5	17	106	2547.9	0.06
2013	2.5	17	128	3054.8	0.05
2014	2.5	17	130	3143.9	0.05
May 2012	2.0	17	39	927.71	0.05
June 2012	2.0	17	32	1783.29	0.05
July 2012	3.0	17	31	1466.15	0.03
August 2012	4.0	17	24	1292.42	0.06
May 2013	1.0	17	32	1851.95	0.09
June 2013	4.0	17	51	2562.73	0.03
July 2013	2.5	3	30	1344.26	0.01
August 2013	2.5	4	37	855.13	0.03
September 2013	2.0	3	25	588.32	0.04
May 2014	1.5	2	32	1652.28	0.03
June 2014	3.5	17	68	3183.17	0.01
July 2014	2.0	17	39	2956.49	0.04
August 2014	1.0	17	22	1716.40	0.08
September 2014	2.5	3	29	695.01	0.03

Table 2. Percent contribution and influence on model performance of landscape variables for the general and yearly Maxent models. Emboldened numbers are the variable that contains the most useful information by itself; underlined numbers are the variable which contains the most information not present in other variables. Note: only variables with a contribution higher than 5% are reported here.

Variable	General	2012	2013	2014
	Contribution (%) / Influence (+/-)			
% Canopy Cover	11.6 (+)	40.5 (+)	<b><u>42.5 (+)</u></b>	27.3 (+)
Dist. Forest Edge	6.0 (-)	---	---	---
Elevation	<u>11.6 (+)</u>	---	---	---
% Impervious Surface	---	---	10.3 (-)	11.3 (-)
NDVI	35.1 (+)	---	---	10.0 (+)
Landcover*	<b>14.4</b>	22.4	9.6	<b><u>21.6</u></b>
% Organic	9.2 (-)	8.4 (-)	8.3 (-)	6.6 (-)
% Sand	6.7 (+/-)	5.9 (+)	9.0 (+)	8.6 (+)

\*Contribution for landcover is not noted as it is a categorical variable and each cover type influences model performance in a different manner.

Table 3. Percent contribution and influence on model performance of landscape variables for the 2012-2014 monthly Maxent models. Emboldened numbers are the variable that contains the most useful information by itself; underlined numbers are the variable which contains the most information not present in other variables. Note: only variables with a contribution higher than 5% are reported here.

Model	Aspect	Variable Contribution(%) / Influence(+/-)							
		Canopy Cover	Dist. Forest Edge	Impervious Surface	NDVI	Landcover*	% Organic	% Sand	Slope
May 2012	---	---	5.4 (-)	---	16.0 (+)	<b>31.2</b>	10.8 (-)	26.9 (+)	---
June 2012	---	---	22.9 (-)	---	12.5 (+)	<b>43.7</b>	---	9.0 (+)	---
July 2012	---	<b>64.9 (+)</b>	---	---	6.7 (+)	5.1	---	---	---
Aug. 2012	---	47.8 (+)	---	---	<b>16.1 (+)</b>	22.4	---	---	---
May 2013	---	19.0 (+)	15.4 (-)	---	---	<b>19.8</b>	6.4 (-)	19.0 (+)	6.6 (-)
June 2013	---	20.0 (+)	---	---	11.4 (+)	<b>26.5</b>	9.7 (-/+)	14.7 (+)	---
July 2013	---	<b>71.6 (+)</b>	---	---	---	17.7	---	---	10.3 (+)
Aug. 2013	9.7 (+)	<b>54.3 (+)</b>	---	---	---	<b>27.4</b>	---	8.6 (+)	---
Sept. 2013	10.6 (+)	<b>54.7 (+)</b>	---	---	---	10.9	---	23.8 (+)	---
May 2014	---	---	---	30.9 (-)	---	<b>69.1</b>	---	---	---
June 2014	---	---	---	---	---	<b>64.9</b>	---	---	---
July 2014	---	24.0 (+)	---	6.3 (-)	---	<b>49.8</b>	---	---	9.6 (+)
Aug. 2014	---	5.2 (+)	---	5.6 (-)	---	<b>58.5</b>	---	---	10.4 (+/-)
Sept. 2014	---	12.2 (+)	---	6.0 (-)	<b>20.4 (+)</b>	39.9	---	11.5 (+)	---

\*Contribution for landcover is not noted as it is a categorical variable and each cover type influences model performance in a different manner.







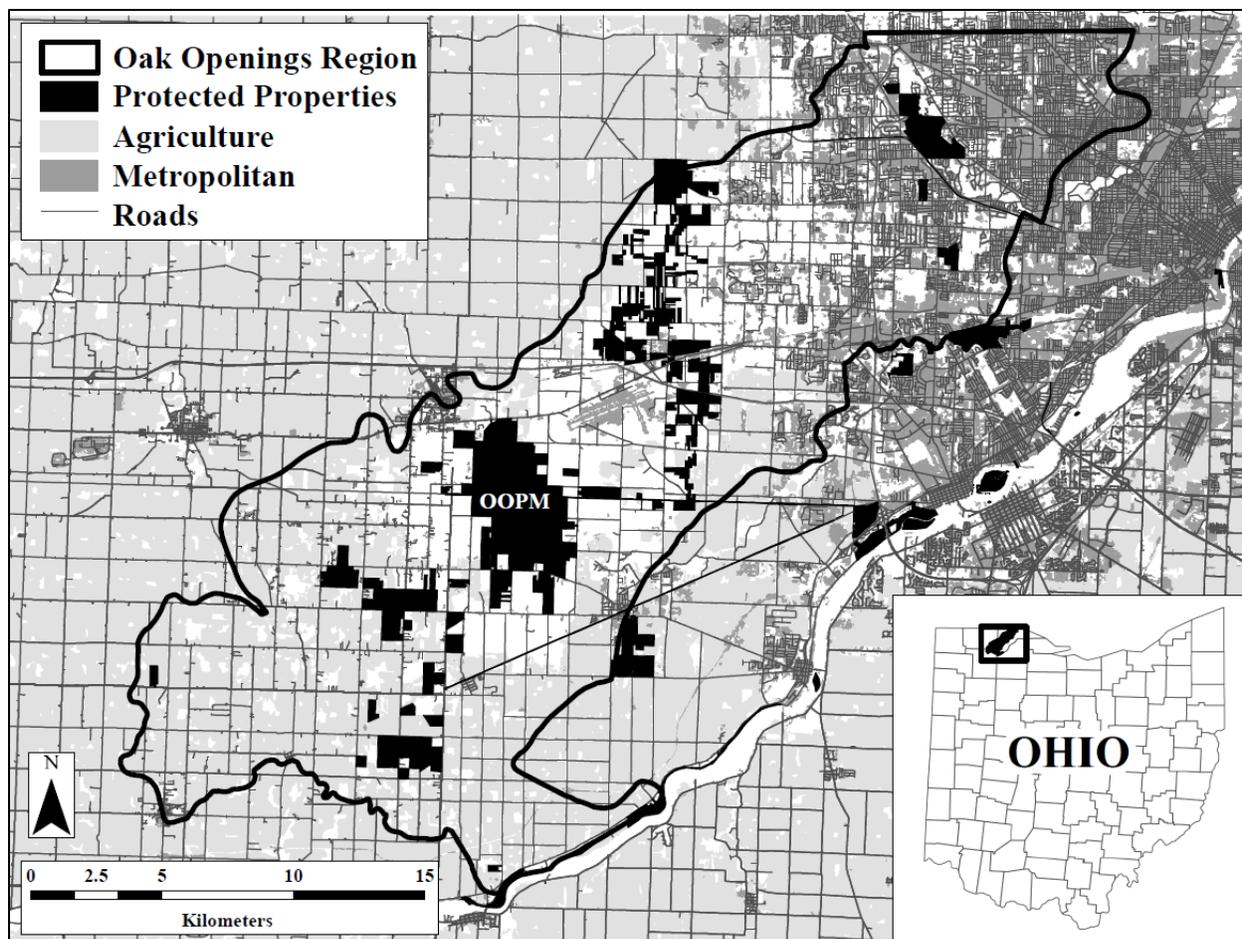


Figure 1. The Oak Opening Region of Northwestern Ohio, including roads, protected properties, metropolitan and agricultural areas, and the Oak Openings Preserve Metropark (OOPM).

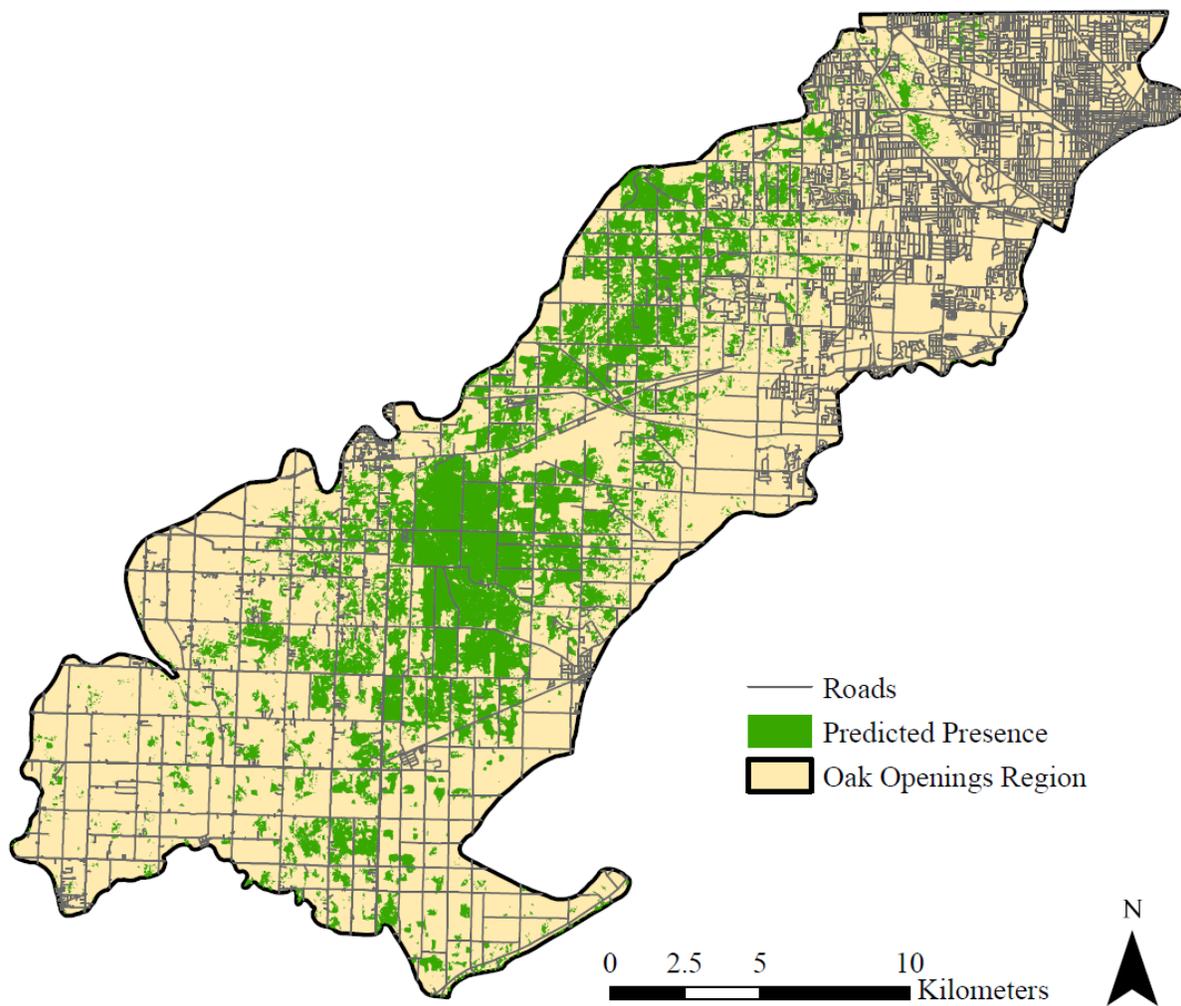


Figure 2. Binary map of predicted presence locations for Eastern Box Turtles from the general model for the Oak Openings Region based on a probability of occurrence threshold of 0.29.

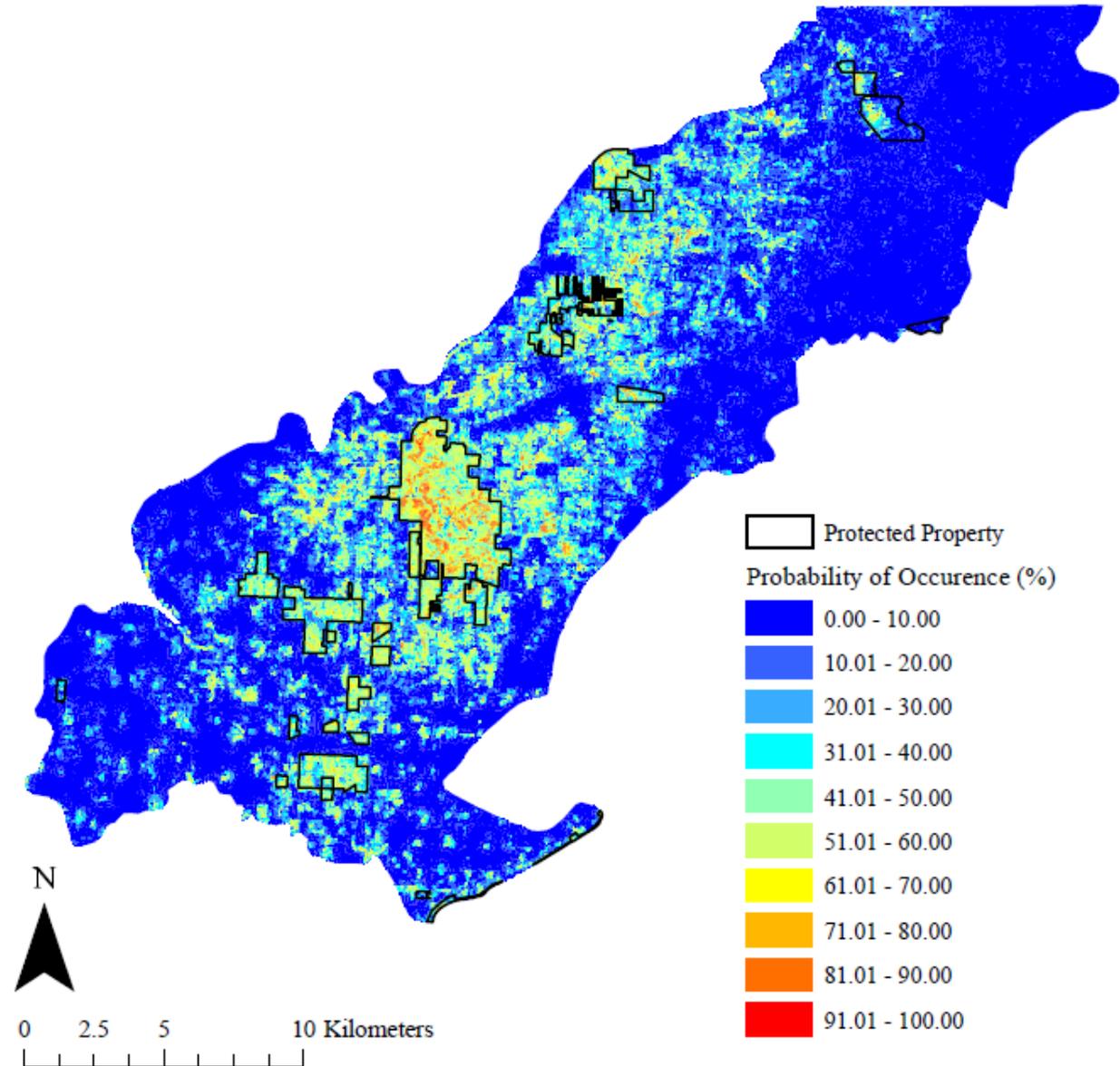


Figure 3. Ten percent probability of occurrence quantiles from the general model for Eastern Box Turtle habitat within the Oak Openings Region of Northwestern Ohio. Also shown are the protected areas within the region (black outlines).

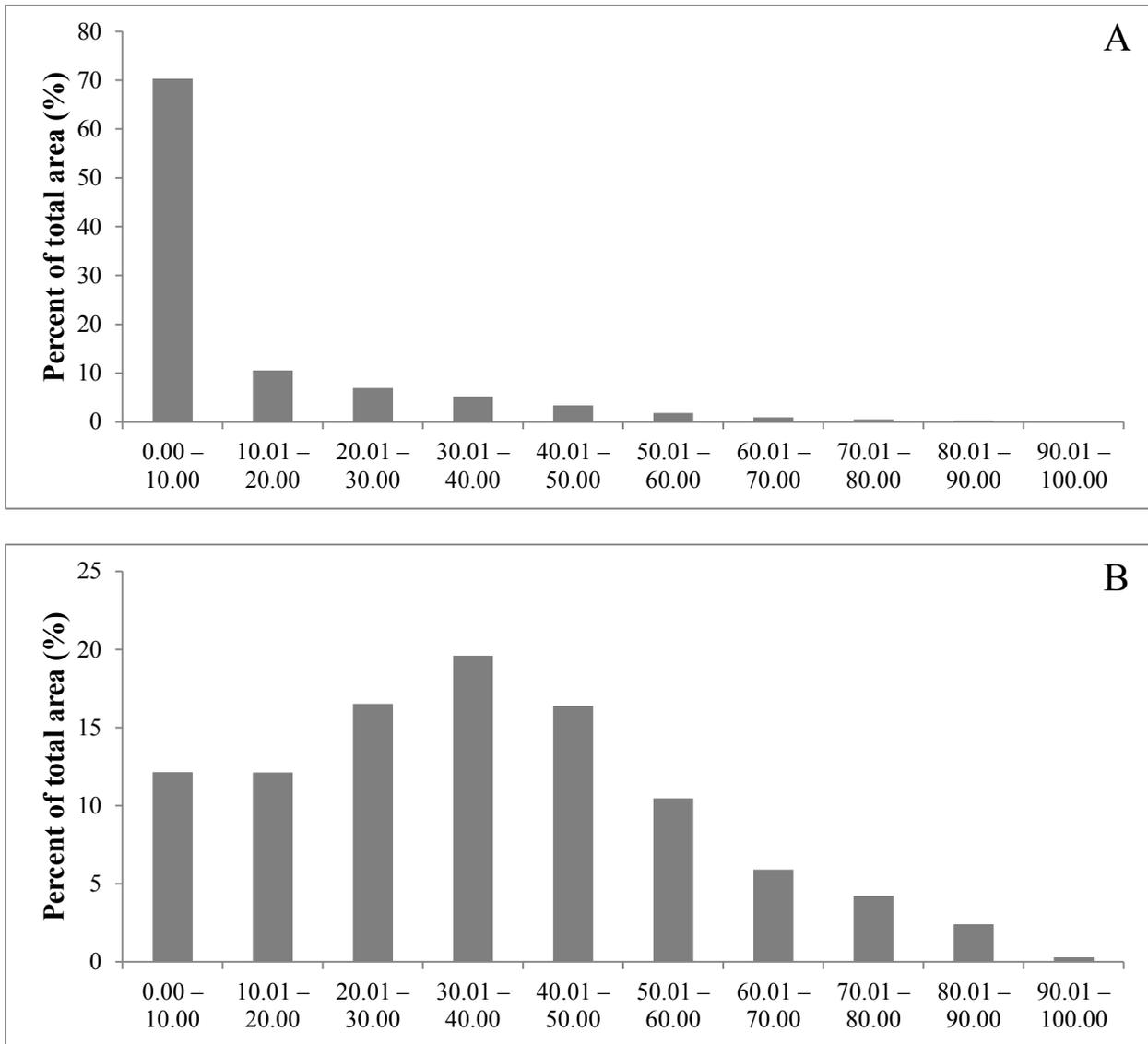


Figure 4 A-C. Distribution across probability of occurrence quantiles for Eastern Box Turtles from the general model for the Oak Openings Region (A), protected areas within the region (B), and the Oak Openings Preserve Metropark (C).

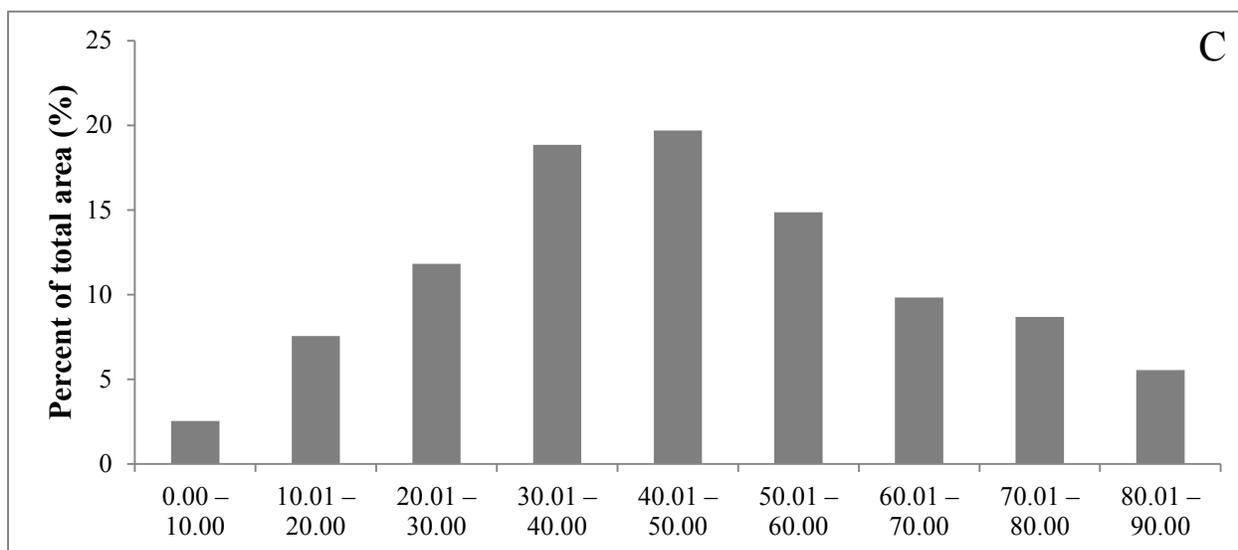


Figure 4. (continued)

## CHAPTER III: EFFECTS OF EARLY-SEASON PRESCRIBED FIRES ON EASTERN BOX TURTLE SPATIAL ECOLOGY

### ABSTRACT

Eastern box turtles (*Terrapene c. carolina*) inhabit many fire-prone habitats throughout their range, however, the effects of fire on this species are poorly understood. We investigated the effects of early-season prescribed fires on movement, home range sizes, habitat use, body condition, and overwintering site fidelity of box turtles in northwestern Ohio. We observed no immediate fire-related mortality, but noted burn injuries and delayed burn mortality for up to four months post-fire. Individuals that experienced a burn did not exhibit different movement patterns or home range sizes than those in unburned areas. There were, however, differences in habitat selection, with burned turtles generally exhibiting weaker habitat preferences than turtles that did not experience a burn, which disproportionately preferred upland and wet forest habitats. Turtles on the burn units exhibited less overwintering site fidelity than those not on burn units and also lost more weight during the active season. Our data indicate that while prescribed fires do not appear to affect the activity patterns of box turtles in our study area, fires may influence habitat selection, overwintering site fidelity, and body condition, at least in the short term. We recommend long-term monitoring of box turtles following exposure to prescribed burns to further elucidate the potential effects fires may have on box turtle populations.

### INTRODUCTION

Fire is a dominant force affecting plant and animal communities, but is especially important in fire-dependent systems, where prescribed fire is the primary method of maintaining habitat (Russell et al. 1999; Van Lear and Harlow 2000; Peterson and Reich 2001). Such fire-dependent plant systems include oak savanna, oak woodlands, and wet prairie; restoration of

these rare plant communities is primarily achieved through the use of prescribed fire (Peterson and Reich 2001; Abella et al. 2007), and, to a lesser extent, mechanical removal of woody vegetation and invasive species. Fire is necessary in maintaining the vegetative structure and diversity of these communities by suppressing woody encroachment, reducing competition from more fire-sensitive saplings and seedlings in the understory, and removing litter to release herbaceous vegetation (Abella et al. 2004; Hutchinson et al. 2005). Burn frequencies of every two to five years are recommended to maintain oak savannas (Grigore and Tramer 1996; Abella et al. 2004; Hutchinson et al. 2005), but factors such as such as other management objectives, ease and safety of burning, and minimization of management costs also play a role (Peterson and Reich 2001). Many of the recommendations and strategies target plant communities and their restoration.

Although it is well known that fire exerts a strong influence on savanna and woodland stand dynamics and structure, the relationship between prescribed fire and herpetofaunal communities is poorly understood (Russell et al. 1999). Given the well-documented global decline of reptiles and amphibians (Gibbons et al. 2000; Böhm et al. 2013), it is necessary to understand how they respond to a frequently used disturbance, like prescribed fire (Driscoll et al. 2010). Seasonality of prescribed fires, for example, can have both a positive and negative direct influence on reptiles (Griffiths and Christian 1996, reviewed in Russell et al. 1999, Keyser et al. 2004). In the long term, appropriate fire regimes can result in higher species diversity, especially in fire-dependent communities like California chaparral (Lillywhite 1977a,b) and African savannas (Barbault 1976), possibly by offering thermoregulatory opportunities via increased surface temperature and radiation, or by improving habitat quality by impeding succession. In general, any detrimental direct or indirect effects from fires are presumably outweighed by the

benefits of having a natural disturbance in the ecosystem (Means and Campbell 1981, Brennan et al. 1998, Russell et al. 1999, Knapp et al. 2009). However, disturbance-related mortality can be more substantial with longer-lived species, like eastern box turtles, where population viability depends on high adult survivorship (Congdon et al. 1993, Dodd et al. 2006, Dodd and Dreslik 2008, Dodd et al. 2016).

The Oak Openings Region of northwestern Ohio is a biodiversity hotspot (Schetter and Root 2011; EPA 2015), containing a number of globally rare, fire-dependent, plant communities (Abella et al. 2001; Abella et al. 2007; Schetter and Root 2011). Managers in this region make extensive use of prescribed fires to maintain these communities (Abella et al. 2007, Schetter and Root 2011). However, the Oak Opening Region is one of the only places in northwestern Ohio where Eastern Box Turtles (*Terrapene c. carolina*) are found (Wynn and Moody 2006) and the specific effects of prescribed fires on this vulnerable species have not been thoroughly investigated.

Long-term studies indicate that many box turtle populations are declining (Stickel 1978; Williams and Parker 1987; Schwartz and Schwartz 1991; Hall et al. 1999). Eastern box turtles have declined throughout their range (Klemens 2000; Dodd 2001), but are widely distributed East of the Rocky Mountains, ranging from southern Maine, south to the Florida Keys, and west to Michigan, eastern Kansas, Oklahoma and Texas (Ernst and Lovich 2009). Like many other long-lived organisms, they possess a suite of life history characteristics such as delayed reproduction, low annual fecundity, low recruitment, and high adult survivorship that make populations vulnerable to perturbations or increases in adult mortality (Congdon et al. 1993; Congdon et al. 1994; Dodd et al. 2016); thus, information regarding their responses to prescribed fire and associated mortality is critically needed (Maret 2004). Additionally, eastern box turtles

prefer microhabitats that include combustible leaf litter and woody debris, making them particularly vulnerable to fires (Reagan 1974; Dodd 2001; Converse and Savidge 2003; Russell et al. 2006; M.Cross *unpublished data*). In many areas of its range, eastern box turtles occur in habitats that are subjected to periodic prescribed fires (Russell et al. 1999; Platt et al. 2010). In the Oak Openings Region, eastern box turtles can frequently be found in unique, fire-dependent habitats like oak savannas or vegetated dunes (Lipps 2010; M.Cross, Bowling Green State University, *unpublished data*).

There are numerous reports of fire-related injuries and mortality in eastern box turtles (Babbitt and Babbitt 1951; Bigham et al. 1965; Ernst et al. 1995; Dodd et al. 1997; Frese 2003; Gibson 2009; Platt et al. 2010; Howey and Roosenberg 2013; Cross et al. 2015), however, much of this is anecdotal with few studies quantitatively assessing the direct and indirect effects of prescribed fire on box turtle populations (Gibson 2009; Platt et al. 2011). Variation in available resources, such as vegetative structure and invertebrate prey, is likely to occur following a fire, thus daily movements and home range sizes may be altered in response.

As part of a larger project examining the spatial ecology of eastern box turtles in northwestern Ohio, we studied the effects of early-season prescribed burns on a subset of our radio-tracked turtles. The objectives of this research were to determine 1) what mortality, if any, resulted from prescribed burns, and 2) the behavioral responses of the turtles to prescribed burns. We recorded direct and indirect sources of mortality for turtles injured by burns. We examined the responses of turtles to prescribed burns by comparing movement rates, home range sizes, habitat use, weight and overwintering site fidelity between turtles that experienced a burn and those that did not.

## STUDY AREA

Our study was conducted in the Toledo Metroparks-owned Oak Openings Preserve Metropark (hereafter, OOPM) located in northwestern Ohio's Oak Openings Region. This region is known for its biodiversity (Abella et al. 2001, Abella et al. 2007) and harbors more plants and animals of conservation interest than any area of similar size in the state, including the federally endangered Karner blue butterfly (*Lycaeides Melissa samuelis*), and five globally rare plant communities: Great Lakes Twig-rush Wet Meadow, Great Lakes Swamp White Oak – Pin Oak Flatwoods, Mesic Sand Prairie, Midwest Sand Barrens, and Black Oak/Lupine Barrens (Farber-Langendoen 2001, EPA 2015). Prior to European settlement, this area consisted of a mixture of fire-maintained oak savanna, oak woodland, and wet prairie habitats (Abella et al. 2001). Soils in the Oak Openings Region are dominated by post-glacial beach sand (Stone et al. 1980). Agricultural clearing, draining of prairies, and fire suppression altered the historic plant communities such that only 27% of the region exists in a natural or semi-natural state (Schetter and Root 2011).

The OOPM, at 1520 ha, is the largest, semi-contiguous, tract of protected natural habitat in Oak Openings Region. The park is made up of a heterogeneous mix of habitats, but is primarily composed of upland, swamp, and floodplain forests, conifers, and, to a lesser extent, oak savanna, Eurasian meadow, and prairie. Within the OOPM, prescribed burns are the highest priority management tool to control for non-native plants prevent woody succession, remove accumulated litter, encourage the spread of prairie grasses and forbs, and cycle nutrients (EPA 2015). The park is divided into 40 management units and burns are implemented on a rotational basis such that treatments are at irregular intervals with at least two years between burns (Grigore and Tramer 1996, Abella et al. 2004).

In 2014 and 2015, three burns (24 ha, 23 ha, and 22 ha) were conducted in mid-March to the end of April in areas where we had turtles with transmitters affixed to their carapaces. Burn prescriptions called for backing fires that encircled the units and moved towards the center, and were restricted to days when ambient relative humidity was 27-33%, ambient temperatures were 13.3– 16.1°C, and wind speeds were 8 – 27 km/h. Ignition for each burn took place between 1200 and 1430 with flameout usually occurring 5-6 hours post-ignition. Primary habitat types in the units were oak and mixed hardwood forests, swamp forest, prairie and oak savanna.

## METHODS

### *Capture and Telemetry*

Turtles were located on potential management units via visual encounter surveys. We affixed radio-transmitters (RI-2B; Holohil System, Ltd., Ontario, Canada) with epoxy to 31 adult turtles on potential burn units in the park. All turtles were weighed upon emergence and just prior to ingress during the 2015 active season using a Pesola® spring scale. We located turtles twice per week, randomizing the order and time of day, and recorded GPS coordinates at each location. On the morning of a burn (0700-0800), however, individuals were located again as soon as it was safe for researchers to enter the burn unit (~10-20 min). This study was conducted in compliance with state (18-101) and local (043015) permits, and all interactions with animals were approved by Bowling Green State University's IACUC (737417-2).

### *Daily Movements and Home Ranges*

Distance moved per day was calculated by dividing the total straight-line distance moved between successive locations by the number of tracking days. We estimated turtle home range sizes by calculating 100% minimum convex polygons (MCPs; White and Garrott 1990). We used the kernel density estimate (KDE; Worton 1989) method with smoothing factor determined

by MCP area (Row and Blouin-Demers) to estimate season ranges (95% probability isopleths). Geospatial Modeling Environment (Beyer 2012) was used to calculate daily movement and to generate the two area estimators.

### *Habitat Use*

Because many habitat types within our study area were rare or seldom used, we condensed the 15 landcover categories (Schetter and Root 2011) into eight. Residential, asphalt, and dense urban habitats were combined into a generic “anthropogenic” category. We classified swamp forest and floodplain forest collectively as “wet forest”, and Eurasian meadow and prairie as “grassland”.

We examined habitat use using compositional analysis (Aeibschler et al. 1993) for burn and non-burn turtles at both second- and third-order scales (Johnson 1980). This technique considers the individual animal as the sampling unit and compares proportional habitat use with proportional habitat availability (Aeibschler et al. 1993; Pendleton 1998). For second-order analyses, usage was defined by the home range (KDE), while available habitat was defined by buffering all of the telemetry locations with radius equal to the greatest length of any 100% MCP home range (982 m). For third-order selection, all individual points were classified based on the cover type in which they occurred. Proportion of habitat used for each turtle was determined by dividing the number of locations within each habitat type by the total number of locations for that turtle. Proportions of each habitat type within the 95% KDE home range represented availability at this level of selection. Compositional analyses were performed in SAS v. 9.3 (BYCOMP.SAS Version 1.0, <http://nhsbig.inhs.uiuc.edu/wes/habitat.html>, accessed 18 January 2016) to examine turtle locations for disproportionate habitat use. Habitat types that were available but not used by the turtles were replaced with small, non-zero proportions (0.0001) as

per Aebischer et al. (1993). To avoid potential confounding effects of rare habitat types in compositional analysis, we removed wet prairie, pond, and scrub/shrub habitats because they were rarely available throughout our study area and were not used by the turtles in our study (Aebischer et al. 1993).

#### *Overwintering Site Fidelity*

To locate overwintering sites, turtles were tracked until they were consistently found underground (ca. mid-November). Unseasonably warm weather can cause late-season movements (M. Cross, Bowling Green State University, personal observation), therefore we considered final overwintering locations to be the location where turtles were underground and had not moved for two weeks following initial ingress or ambient temperature spike. Following fall ingress, we measured the distance between that point and the location of the previous year's overwintering site.

#### *Statistical Analyses*

With the exception of habitat use (see above), we compared daily movement, home range area, weight change and overwintering site fidelity for turtles that experienced a spring burn and those that did not (hereafter, burn and reference, respectively) turtles using a Wilcoxon rank-sum test. Platt et al. (2010) demonstrated that fires appear to affect male and female turtles equally; therefore comparisons were not made between sexes. Unless otherwise noted, all statistical analyses were carried out using an  $\alpha = 0.05$ , and all means are reported  $\pm$  SD.

## **RESULTS**

During the course of our study, 11 turtles with transmitters were in three areas that underwent prescribed burns (Table 1). The exact behaviors of turtles during the fire are unknown because logistical constraints prevented researchers from directly observing the turtles

during the fires. However, it was apparent after the burns that turtles avoided fires by seeking shelter in moist, refugia, including low, wet areas in water and/or under mud ( $n = 4$ ), a pile of wet leaves in a shallow depression ( $n = 1$ ), or a hollow, rotting log ( $n = 1$ ). Two turtles were found mating less than 20 min after the fire had passed through their area.

Four telemetered turtles were injured during fires. Fire related injuries included mild and severe burns to digits and extremities, blistering and subsequent loss of the carapacial scutes, and bleeding from the nose (presumably from damage due to inhalation; Table 1). Three of the injured turtles eventually died, but time to death ranged from one to four months post-burn. Necropsies of dead turtles, conducted by a veterinarian, ruled out non-fire related health complications as a cause of death. These examinations further revealed that two of the females killed by fires were gravid (indicated by the presence of well-developed yolk follicles). Of the non-fatally injured turtles, one experienced open wounds, development of necrotic tissue, loss of burned digits, weight loss, and other complications (e.g. aural abscesses and oral plaque). This turtle and the seven other turtles on burn units, all overwintered at the end of the 2015 field season.

#### *Daily Movements, Home Ranges, and Weights*

We observed no differences between burn and non-burn turtles (Figure 1 A & B) in either daily movement ( $8.99 \pm 4.66$  m, and  $11.69 \pm 6.74$  m, respectively;  $P > 0.40$ ) or home range sizes ( $27.48 \pm 30.21$  ha and  $53.52 \pm 93.57$  ha respectively;  $P > 0.27$ ). Proportion of burn turtle home ranges made up of the burn units ranged from 15 to 100% and averaged 77.5%.

Burn turtles lost more weight at the end of the season than reference turtles ( $P < 0.01$ ). All turtles on the burn units lost weight at the end of the season for an average weight loss of

48.0 ± 33.5g , whereas only three of the 21 reference turtles lost weight for an overall average weight gain of 19.7 ± 32.4g (Figure 1 C).

#### *Habitat Use*

Results of the compositional analyses indicate that burn turtles were not strongly selective in their use of habitats at the second- and third-order scales (Table 2). Reference turtles were selective in their use of habitat at both scales and tended to select upland deciduous forest, wet forest, savanna, and conifer over all others (Table 2).

#### *Overwintering Site Fidelity*

Turtles subjected to a burn exhibited less site fidelity than reference turtles ( $P = 0.031$ ; Figure 1 D). Distances between successive overwintering sites for burn and reference turtles were 247.21 ± 136.08 m and 54.00 ± 14.25 m, respectively.

## **DISCUSSION**

Eastern box turtles face three possible outcomes when exposed to a fire: they can move to undisturbed habitat, they could survive and stay in one place, or they can be killed (Dodd 2006). We observed no direct mortality from prescribed fires, but documented delayed fire-related mortality in three turtles, two of which were gravid females. Following exposure to a prescribed burn, turtles on burn units had weaker habitat preferences, lost weight, and exhibited less overwintering site fidelity than reference turtles.

Turtles in burned landscapes are confronted with several challenges including altered macro- and microhabitat, thermoregulatory opportunities, moisture levels, and prey availability (Russell et al. 1999; Iverson and Hutchinson 2002; Howey 2014). These alterations are not necessarily detrimental as more thermoregulatory opportunities and an increase in prey abundance could be beneficial. However, open areas on burned landscape may become too warm

(Iverson and Hutchinson 2002), and in order to take advantage of thermal opportunities, thermal refugia must be present in sufficient amounts for turtles to escape unfavorable temperatures. Alteration of the thermal landscape can lead to shifts on movement rates, thermoregulatory behavior, and energy expenditure (Belzer 1997; Howey 2014).

Habitat quality, proximity of undisturbed habitat, and availability of resources have been suggested as possible factors influencing area use and movements following a disturbance (Dodd 2001, Dodd et al. 2006, Slavenko et al. 2015). Similar to what Gibson (2009) found, turtles in our study did not change daily movements or home range sizes in response to a burn and did not immediately move off of the burn units. Since our turtles did not change movement or area use patterns following prescribed fires, it is likely that there were still suitable resources available within burn units, but they might have been limited or had a patchy distribution? Contrary to Gibson's (2009) findings that prescribed fires did not influence landscape-scale habitat selection, our burned turtles did not exhibit hierarchical habitat selection. While lack of habitat selection at this scale is not uncommon at our site (M. Cross, Chapter I), the tendency of burn turtles to be more general in their habitat selection might be a reflection of altered microhabitat features.

Habitat selection and activity patterns in eastern box turtles is thought to be heavily influenced by the availability of microhabitat features that promote thermoregulation, prey acquisition, preventing desiccation, and creation of resting forms (Penick et al. 2001; Rossell et al. 2006; Nazdrowicz et al. 2008; McKnight 2011). Temperature, humidity, ground cover, and litter have been found to be key components governing microhabitat selection (Reagan 1974; Converse and Savidge 2003; Rossell et al. 2006; M. Cross, Chapter I), all of which are expected to be altered after a fire. Animals in altered habitats may need to move more frequently to find

resources (Debinski and Holt 2000; Hansbauer et al. 2008). In the case of box turtles, these resources include thermoregulation sites and areas that promote form construction.

Box turtles are known to exhibit overwintering site fidelity, often returning to the same areas in consecutive years. Wiley (2010) reported an average distance of 118.3 m between consecutive overwintering sites over a four-year period. The average distance between overwintering sites for our reference turtles was less than this, but as with other activity patterns, this is expected vary geographically. The decrease in site fidelity for burn turtles was most likely related to availability of suitable microhabitat features. While overwintering site-selection involves dealing with different stressors than during the active season (e.g., freezing and desiccation), selection of these sites involves many of the same microhabitat features (Dolbeer 1970, Claussen et al. 1991, Ultsch 2008, Wiley 2010). Because turtles along the northern edge of their range may spend up to half their life overwintering (Gregory 1982; Ultsch 2008), it is expected that they will be very selective of these sites and exhibit a higher degree of site fidelity. Alterations to the overwintering habitat could cause temporary abandonment and force turtles to make long-distance movements across unfavorable matrix (e.g., roads) into unknown areas where the quality and availability of suitable overwintering sites are limited or absent.

Altered habitat, microhabitat, and resources availability are also possible explanations for the differences in habitat use, weight and overwintering site fidelity between burn and reference turtles in our study. Since movement patterns and area use did not change, it is likely that some or all of these resources were still available within burn units, but might have been limited in their availability, thus causing turtles to spend more time moving from habitat to habitat in search of patches supporting suitable microclimates. The combination of altered prey and microhabitat availability might subsequently cause changes in movement rates, thermoregulatory

behavior, and energy expenditure (Belzer 2002; Dodd and Dreslik 2008; Howey 2014), which could explain the overall loss in weight for burn turtles. Measuring microhabitat selection for burned turtles was beyond the scope of this project, but it is likely they exhibited similar patterns to those of reference turtles, but had to be more discerning or make sacrifices given availability of suitable sites.

We did not observe the direct mortality reported by some studies, but did detect delayed mortality in 27% of our telemetered turtles. Platt et al. (2010) found 14 fire-killed box turtles after four burns and estimated between 10.2% and 21.6% of turtles/ha were killed during these burns. This is similar to the 10.2 and 21.6% of turtles per ha killed after four burns in Florida (Platt et al. 2010). Gibson (2009) also reported fire-related delayed mortality for up to four months post-burn in 10-13% of telemetered box turtles at her study site in southwestern Michigan. Following loss of carapacial scutes, turtles tend to move into wet areas, presumably because they had difficulty regulating water loss (Rose 1969; Gibson 2009), where they might die if they were unable to maintain water balance. We do emphasize caution when interpreting fire-related mortality across studies as different burn prescriptions might influence these estimates. For instance, the Gibson (2009) study presents data from an area that was burned consecutively two years in a row, a fire return interval that is not recommended and not used at our study site. Additionally, not knowing population size or demographics of turtles in our study area makes formulating these types of estimates difficult.

It has been posited that fires affect male and female turtles equally with a moderately female-biased sex ratio of fire-killed turtles (Ernst et al. 1995; Dodd et al. 1997; Platt et al. 2010). Our limited sample of fire-related mortality (1 male: 2 females) supports these observations, but the two females that were gravid raises serious concerns. Turtle populations are

sensitive to removal of individuals, particularly of gravid females (Citations). Population-level effects may be more pronounced if gravid female mortality is high during prescribed fires. As long as initial are large or the population growth rate is increasing or stable, box turtles may be resilient to mortality from rare disturbances (Dodd et al. 2016). However, box turtle populations have little resilience to chronic disturbances, such as frequent prescribed fires, especially in fragmented habitats and may not be able to recover following removal of individuals (Kuo and Janzen 2004; Dodd et al. 2006; Dodd et al. 2016).

Delayed mortality presents a unique challenge for post-fire monitoring efforts. Results from this and Gibson's (2009) study suggest that many most-fire assessments might underestimate box turtle mortality, and should incorporate a temporal component. The cryptic nature of box turtles limits their detectability (Refsnider et al. 2011, Kapfer et al. 2012), and the tendency for fire-injured turtles to spend time in hydric habitats makes surveying for them more difficult. Therefore, post-fire surveys might not be sufficient to account for delayed mortality and significant population losses could go undetected.

### **MANAGEMENT IMPLICATIONS**

We found evidence that eastern box turtles are capable of avoiding prescribed fires, but suggest cautionary management when implementing burns in areas where box turtles might be present. Detailed population estimates are needed prior to a burn as estimates of mortality mean little unless they can be extrapolated to the whole population. In areas where box turtle mortality is a concern for land managers, the current recommendation of reducing mortality by burning when box turtles are inactive (Dodd 2001, Kingsbury and Gibson 2012) should be sufficient. However, burning during these times might generate less favorable fire effects (citations) and the needs of the ecological community should be considered.

If prescribed fires need to be conducted the active season, mortality can be minimized by taking box turtle ecology into account. For instance, in our study, adult box turtles were rarely in open-canopied areas between the months of October and April (M. Cross, Chapter I), meaning these habitats could be burned during these times and mortality should be low. This recommendation is strictly for mitigating adult mortality as the turtles nest in these open areas and neonate mortality during burns is expected to be high (Ernst et al. 1995). Our observations of box turtles avoiding fire by seeking shelter support recommendations of slow-moving, low-intensity backing fires (Platt et al. 2010, Kingsbury and Gibson 2012). Habitat heterogeneity and availability of refugia will contribute to box turtle survival and should be part of box turtle-compatible burn plans. Current recommendations of burning small areas on a rotational basis are sound, but future research should seek to determine if a fire-return-interval of 2-5 years is compatible with time necessary for populations to recover from any fire-related mortality that may occur. Long-term monitoring associated with radio-telemetry is the best ways to address delayed mortality and the indirect, legacy effects of prescribed fires. Fire-related gravid female mortality should be a concern to long-term population viability in areas that are frequently burned. Disturbances affecting female turtles prior to egg deposition are expected to have a greater impact on populations (Dodd 2016), we therefore recommend considering late-season burns to avoid mortality in gravid females.

#### **ACKNOWLEDGMENTS**

Funding for this project was provided by the Toledo Zoo and the Metroparks of the Toledo Area. We are grateful to the Metroparks burn crew for their cooperation and for implementing the prescribed burns. K. Bekker, P. Tolson, and R. Berlinski from the Toledo Zoo offered logistic

support and performed necropsies for us. Special thanks are owed to John Rucker for the use of his trained Boykin Spaniels.

### LITERATURE CITED

- Abella, S.R., J.F. Jaeger, D.H. Gehring, R.G. Jacksy, K.S. Menard, and K.A. High. 2001. Restoring historic plant communities in the Oak Openings Region of Northwest Ohio. *Ecological Restoration* 19: 155-160.
- Abella, S.R., J.F. Jaeger, and L.G. Brewer. 2004. Fifteen years of plant community dynamics during a northwest Ohio oak savanna restoration. *The Michigan Botanist* 43: 117-127.
- Abella, S.R., J.F. Jaeger, and T.A. Schetter. 2007. Public land acquisition and ecological restoration: an example from northwest Ohio's Oak Openings Region. *Natural Areas Journal* 27: 92-97.
- Aebischer, N.J., P.A. Robertson, and R.E. Kenwood. 1993. Compositional analysis of habitat use from animal radio-tracking data. *Ecology* 74: 1313-1325.
- Auffenberg, W., and R. Franz. 1982. The status and distribution of *Gopherus polyphemus*. In R.B. Bury (ed.) *North American Tortoises: Conservation and Ecology*. U.S. Fish and Wildlife Service Research Report 12. Pp. 95-126.
- Babbitt, L.H., and C.H. Babbitt. 1951. A herpetological study of burned-over areas in Dade County, Florida. *Copeia* 1951:79.
- Belzer, B. 2002. A nine year study of eastern box turtle courtship with implications for reproductive success and conservation in a translocated population. *Turtle and Tortoise Newsletter* 6:17-26.
- Beyer, H.L. 2012. Geospatial modelling environment (Versoin 0.7.3.0). (software). URL: <http://www.spatalecology.com/gme>.

- Bigham, S.R., J.L. Hepworth, and R.P. Martin. 1965. A casualty count of wildlife following a fire. *Proceedings of the Oklahoma Academy of Sciences* 45: 47-50.
- Cooper, J.E. 2006. Dermatology. Pages 196-216 *in* D.R. Mader, editor. *Reptile Medicine and Surgery*. Second edition. Elsevier Inc., St. Louis, Missouri, USA.
- Debinski, D.M., and R.D. Holt. 2000. A survey and overview of habitat fragmentation experiments. *Conservation Biology* 14:342-355.
- Dodd, C.K. 2001. *North American Box Turtles: A Natural History*. University of Oklahoma Press, Norman, OK.
- Dodd, C.K., Jr., and M.J. Dreslik. 2008. Habitat disturbances differentially affect individual growth rates in a long-lived turtle. *Journal of Zoology* 275:18-25.
- Dodd, C.K., Jr., A. Ozgul, and M.K. Oli. 2006. The influence of disturbance events on survival and dispersal rates of Florida Box Turtles. *Ecological Applications* 16:1936-1944.
- Dodd, C.K., V. Rolland, and M.K. Oli. 2016. Consequences of individual removal on persistence of a protected population of long-lived turtles. *Animal Conservation*  
DOI: 10.1111/acv.12253.
- Driscoll, D.A., and M.K. Henderson. 2008. How many common reptile species are fire specialists? A replicated natural experiment highlights the predictive weakness of a fire succession model. *Biological Conservation* 141: 460-471.
- Driscoll, D.A., D.B. Lindenmayer, A.F. Bennett, M. Bode, R.A. Bradstock, G.J. Cary, M.F. Clarke, N. Dexter, R. Fensham, G. Friend, M. Gill, S. James, G. Kay, D.A. Keith, C. MacGregor, J. Russell-Smith, D. Salt, J.E.M. Watson, R.J. Williams, A. York. 2010. Fire management fore biodiversity conservation: key research questions and our capacity to answer them. *Biological Conservation* 143: 1928-1939.

- Environmental Protection Agency (EPA). 2015 September 9. Oak Openings Site Conservation Plan. < <http://archive.epa.gov/ecopage/web/html/oak-openings.html>>. Accessed 12 January 2016.
- Ernst, C.H., T.P. Boucher, S.W. Sekscienci, and J.C. Wilgenbusch. 1995. Fire ecology of the Florida box turtle, *Terrapene carolina bauri*. Herpetological Review 26: 185-187.
- Ernst, C.H., and J.E. Lovich. 2009. Turtles of the United States and Canada. The Johns Hopkins University Press, Baltimore, MD.
- Gregory, P.T. 1982. Reptilian hibernation. Pages 53-154 in C. Gans, editor. Biology of the Reptilia. Vol.13. Academic Press, Inc., New York, USA.
- Griffiths, A.D, and K.A. Christian. 1996. The effect of fire on the frillneck lizard (*Chlamydosaurus kingii*) in northern Australia. Australian Journal of Ecology 21: 386-398.
- Grigore, M.T., and E.J. Tramer. 1996. The short-term effects of fire on *Lupis perennis* (L). Natural Areas Journal 16: 41-48.
- Hansbauer, M.M., I. Storch, R.G. Pimentel, and J.P. Metzger. 2008. Comparative range use by three Atlantic Forest understorey bird species in relation to forest fragmentation. Journal of Tropical Ecology 24:291-299.
- Iverson, L.R., and T.F. Hutchinson. 2002. Soil temperatures and moisture fluctuations during and after prescribed fire in mixed-oak forests, USA. Natural Areas Journal 22: 296-304.
- Johnson, D.H. 1980. The comparison of usage and availability measurements for evaluating resource preference. Ecology 61: 65-71.
- Kapfer, J.M., D.J. Munoz, and T. Tomasek. 2012. Use of wildlife detection dogs to study eastern

- box turtle (*Terrepeene caorolina carolina*) populations. *Herpetological Conservation and Biology* 7: 169-175.
- Keyser, P.D., D.J. Sausville, W.M. Ford, D.J. Schwab, and P.H. Brose. 2004. Prescribed fire impacts to amphibians and reptiles in Shelterwood-harvested oak-dominated forests. *Virginia Journal of Science* 55: 159-168.
- Kingsbury, B.A., and J. Gibson (editors). 2012. *Habitat Management Guidelines for Amphibians and Reptiles of the Midwestern United States. Partners in Amphibian and Reptile Conservation Technical Publication HMG-1, 2<sup>nd</sup> Edition. 155p.*
- Knapp, E.E., B.L. Estes, and C.N. Skinner. 2009. Ecological effects of prescribed fire season: A literature review and synthesis for land managers. U.S. Department of Agriculture, Forest Service General Technical Report no. PSW-GTR-224., Pacific Southwest Research Station. Albany, California, USA.
- Kuo, C.H., and F.J. Janzen. 2004. Genetic effects of persistent bottleneck on a natural population of Ornate Box Turtles (*Terrapene ornata*). *Conservation Genetics* 5:425-437.
- Lee, M.A., K.L. Snyder, P. Valentine-Darby, S.J. Miller, and K.J. Ponzio. 2005. Dormant season prescribed fire as a management tool for control of *Salix caroliniana* Michx. in a floodplain marsh. *Wetlands Ecology and Management* 13: 479-487.
- Lillywhite, H.B. 1977a. Animal responses to fire and fuel management in chaparral. *In* H.A. Mooney and E.C. Conrad (eds.) *Proceedings of the Symposium on Environmental Consequences of Fire and Fuel Management in Mediterranean Ecosystems*. U.S. Department of Agriculture Forest Service GTR-WO3. Pp. 368-372.
- Lillywhite, H.B. 1977b. Effects of chaparral conservation on small vertebrates in southern California chaparral. *Biological Conservation* 11: 171-184.

- Lindenmayer, D.B., J.T. Wood, C. MacGregor, D.R. Michael, R.B. Cunningham, M. Crane, R. Montague-Drake, D. Brown, R. Muntz, and D.A. Driscoll. 2008. How predictable are reptile responses to wildfire? *Oikos* 117: 1086-1097.
- Mader, D.R. 2006. Thermal Burns. Pages 916-923 *in* D.R. Mader, editor. *Reptile Medicine and Surgery*. Second edition. Elsevier Inc., St. Louis, Missouri, USA.
- Maret, T., J. Mitchell, Jr., and I.L. Brisbin. 2004. Summary of Breakout Sessions: Research Needs. *In*: C. Swarth and S. Hagoood (*Eds.*). Summary of the Eastern Box Turtle Regional Conservation Workshop. The Humane Society of the United States, pp. 13-14.
- Moore, J.A., and J.C. Gillingham. 2006. Spatial ecology and multi-scale habitat selection by a threatened rattlesnake: the eastern massasauga (*Sistrurus catenatus catenatus*). *Copeia* 2006: 742-751.
- Pendleton, K.T, E. DeGayner, C.J. Flatten, and R.E. Lowell. 1998. Compositional analysis and GIS for study of habitat selection by goshawks in Southeast Alaska. *Journal of Agricultural, Biological, and Environmental Statistics* 3: 280-295.
- Platt, S.G., H. Liu, and C.K. Borg. 2010. Fire ecology of the Florida box turtle (*Terrapene carolina bauri Taylor*) in pine rockland forests of the Lower Florida Keys. *Natural Areas Journal* 30: 254-260.
- Refsnider, J.M., T.S. Mitchell, H.M. Streby, J.T. Strickland, D.A. Warner, and F.J. Janzen. 2011. A generalized method to determine detectability of rare and cryptic species using the Ornate Box Turtle as a model. *Wildlife Society Bulletin* 35: 93-100.
- Rose, F.L. 1969. Desiccation rates and temperature relationships of *Terrapene ornata* following scute removal. *Southwestern Naturalist* 14: 67-72.
- Row, J.R. and G. Blouin-Demers. 2006. Kernels are not accurate estimators of home-range size

- for herpetofauna. *Copeia* 2006: 797-802.
- Schetter, T.A., and K.V. Root. 2011. Assessing an imperiled oak savanna landscape in northwestern Ohio using Landsat data. *Natural Areas Journal* 31: 118-130.
- Seaman, D.E., and R.A. Powell. 1996. An evaluation of the accuracy of kernel density estimators for home range analysis. *Ecology* 77: 2075-2085.
- Slavenko, A., Y. Itescu, F. Ihlow, and Shai Meiri. 2015. Home is where the shell is: predicting turtle home range sizes. *Journal of Animal Ecology* 85: 106-114.
- Stone, K.L., E.H. McConoughey, G.D. Bottrell, and D.J. Crowner. 1980. Soil survey of Lucas County, Ohio. U.S. Department of Agriculture, Soil Conservation Service, Washington D.C.
- VanLear, D.H., and R.F. Harlow. 2000. Fire in the Eastern United States: influence on wildlife habitat. *In* W.M. Ford, K.R. Russell, and C.E. Moorman (eds.) *The Role on Nongame Wildlife Management and Community Restoration: Traditional Uses and New Directions*. Proceedings of a Special Workshop. U.S. Forest Service General Technical Report NE-288. U.S. Department of Agriculture, Northeastern Research Station, Newtown Square, Pennsylvania, USA. pp. 2—10.
- Worton, B.J. 1989. Kernel methods for estimating the utilization distribution in home range studies. *Ecology* 70: 164-168.

Table 1. Date (*yymmdd*) of burn, Turtle ID, post-fire behavior, list of fire-related injuries, and fate of the turtles in the Oak Openings Preserve Metropark that experienced a prescribed fire between 2014-2015.

Burn Date	Turtle	Behavior	Injuries*	Fate
140424	15	Unknown	Loss of scute Rp1, loss of front digits, forearm injuries became necrotic	Died four months post-burn
150415	98	Fled to water; under mud	None	Survived
150415	99	Fled to water; under mud	None	Survived
150424	11	Fled to water; under mud	None	Survived
150424	13	Fled to water; under mud	Burns to front legs	Survived; Burn injuries became necrotic, evidence of secondary infection
150424	19	Unknown	None	Survived; mating with 115
150424	94	Hid under wet leaves	None	Survived
150424	95	Hid in a decaying log	None	Survived
150424	114	Exposed near ignition site	Blistering of scutes Lp3, Lp4, v5, Rp4 and all marginals; exposed bone on Lp4 and Rm4; burned digits; bloody discharge from mouth and nose	Died one month post-burn
150424	115	Unknown	None	Survived
150424	118	Unknown	Blistering of scutes Rp1 and Rp2	Died one month post-burn

\*Carapacial scutes are named following the figure in Ernst and Lovich 2009 and are read as

“Rp1” = first right pleural scute

Table 2. Habitat use, availability, and preference by eastern box turtles during 2015 in the Oak Openings Preserve Metropark as determined by compositional analysis.

Category	<i>N</i>	%Use/%Available								Preferred ↔ Avoided <sup>a</sup>	Statistics		
		Agriculture (A)	Residential (R)	Savanna (S)	Wet Forest (W)	Conifer (C)	Upland Deciduous (U)	Barrens (B)	Grassland (G)		<i>A</i> <sup>b</sup>	<i>F</i>	<i>P</i>
Burn 2 <sup>nd</sup> order	8	1.5/1.3	2.3/4.3	11.7/9.8	30.4/32.1	2.9/5.1	25.0/26.5	2.0/1.6	24.3/19.3	<u>B S U W G A R C</u>	0.205	0.55	0.779
Burn 3 <sup>rd</sup> order	8	1.3/1.7	4.3/8.8	9.8/3.0	32.1/28.1	5.1/16.5	26.5/26.1	1.6/1.2	19.3/14.6	<u>S G W U C R A B</u>	0.068	1.95	0.503
Reference 2 <sup>nd</sup> order	21	0.0/0.6	0.0/6.5	4.4/4.5	23.3/22.7	17.2/18.8	38.1/30.4	0.8/1.1	16.1/15.2	<u>U G W C S B A R</u>	0.066	28.39	<0.001
Reference 3 <sup>rd</sup> order	21	0.6/5.7	6.5/16.8	4.5/1.9	22.7/26.0	18.8/13.2	30.7/20.7	1.1/1.6	15.2/14.1	<u>U W C S R G B A</u>	0.210	7.50	0.001

<sup>a</sup>Habitat classifications: Agriculture (A), Barrens (B), Conifer (C), Grassland (G), Residential (R), Savanna (S), Upland Deciduous (U), Wet Forest (W).

<sup>b</sup>Wilk's lambda.

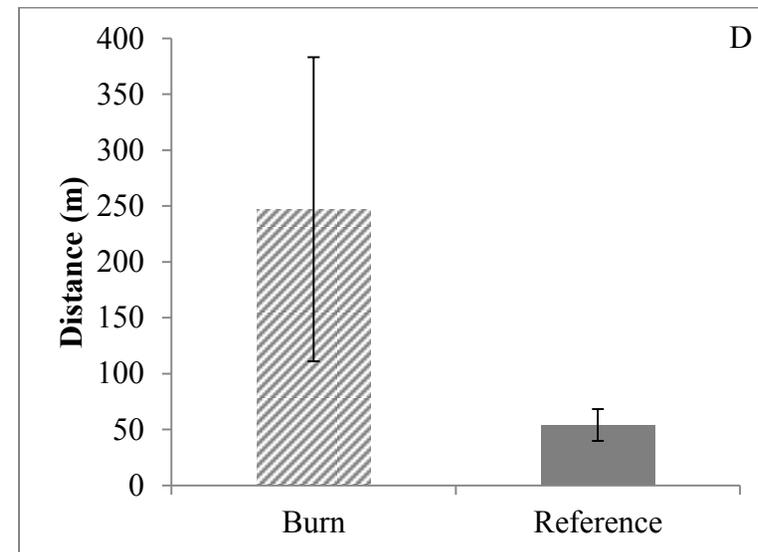
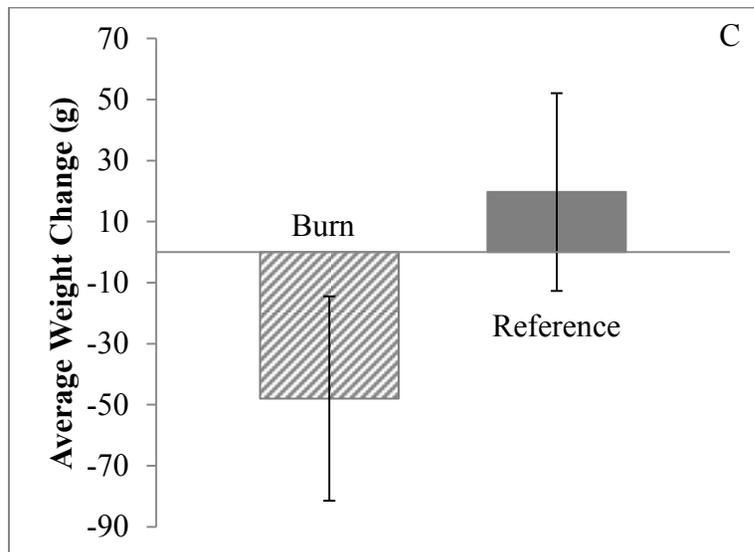
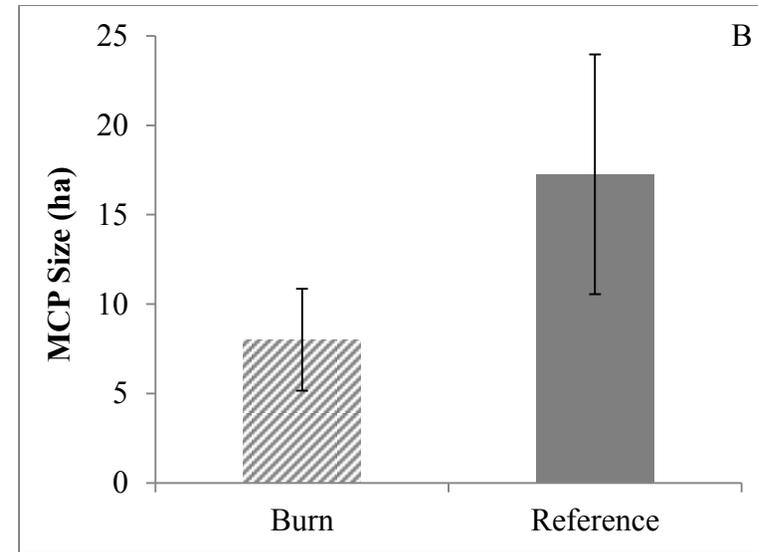
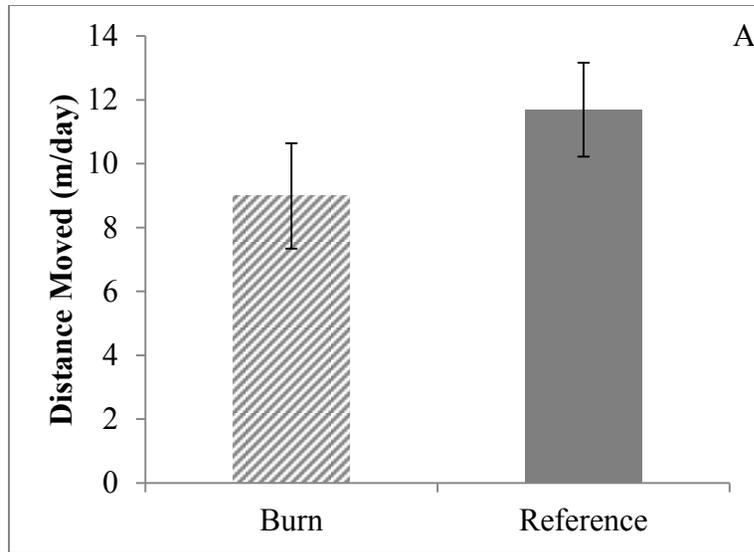


Figure 1(A-D). Average distance moved per day (A), 100% MCP home range size (B), change in weight (C) and distance between successive overwintering sites (D) for burn and reference turtles in the Oak Openings Preserve Metropark during 2015.

CHAPTER IV: PATTERN RECOGNITION SOFTWARE AS A SUPPLEMENTAL METHOD OF IDENTIFYING INDIVIDUAL EASTERN BOX TURTLES (*TERRAPENE C. CAROLINA*)

**ABSTRACT**

Identifying and monitoring individuals is essential in behavioral and ecological studies. As technology improves, researchers have shifted from traditional marking techniques and have started photographing conspicuous marks to identify individuals. Eastern box turtles (*Terrapene c. carolina*) are ideal candidates for photographic identification of unique markings on their shell. The objectives of this study are: test pattern recognition as a viable method of identifying individual eastern box turtles; determining if top-down/off-center carapace or plastron photos were more diagnostic; and to test a pattern recognition program, WildID, in identifying individuals from different populations. We collected 1200 photos of box turtles from four locations in two states. Using both original and cropped, distorted or altered images, the program never mismatched individuals, making WildID highly sufficient in identifying individual box turtles. Additionally, we found carapace and plastron photos to be more accurate than off-center images. We encourage researchers and naturalists who have taken photographs of eastern box turtles to utilize this software to rapidly analyze historical photo libraries for identifying recaptured individuals. This sort of recognition software, combined with citizen-science programs, could provide the means for mark-recapture studies of eastern box turtles over a large geographic range.

**INTRODUCTION**

Identifying and monitoring individuals is an essential part of behavioral and ecological studies of wild animals. Traditional methods for permanently marking turtles include shell notching, tagging with aluminum bands or insertion of a passive integrated transponder (PIT) tag

(reviewed in Ferner 2007; Plummer and Ferner 2012). These methods have been shown to be effective, but applying such marking techniques may result in increased stress, altered behavior, or opportunities for infection (McGregor and Peake 1998; Markowitz et al. 2003; Fisher 2007). Therefore, it is desirable to use methods of long-term identification that minimize the potential impact on the organism.

As technology improves, researchers have taken advantage of photographing naturally occurring or conspicuous marks to identify individuals (reviewed in Reisser et al. 2008 and Bolger et al. 2012). Traditionally, pattern-recognition involved comparing hard copies of photographs against catalogues of photographs to identify recaptures. In the case of large image catalogues, the number of images makes pattern-matching a time-consuming process and increases the probability of visual errors (Hammond et al. 1990; Katonas and Beard 1990; Sears et al. 1990; Gamble et al. 2008).

Shell patterns have been suggested to be a viable means of identifying individual box turtles (Budischak et al. 2006; Wynn and Moody 2006; Weiss 2009; Hoss et al. 2013). Photo-recognition techniques offer several advantages such as reduced handling time of the animal, an increase in the efficiency of identifying individuals, and elimination of the problems associated with loss of tags or other artificial marks (Reisser et al. 2008). In addition, pattern recognition is inexpensive, less invasive than most of the traditional methods of permanently marking turtles, and lessens the chance of identity failure due to wear or malfunction. As such, pattern-recognition software has the potential to be a cost-effective alternative method of identifying box turtles.

At our study site, in northwest Ohio, for example, the required method of marking Eastern Box Turtles (*Terrapene carolina carolina*) is use of PIT tags. The expense of using PIT

tags and the readers required to confirm the presence of a PIT tag can quickly eat away at the small grants available to fund local mark-recapture studies. Additionally, this method of marking carries the extra complication of making it impossible to identify recaptures without a PIT tag reader. To aid in future mark-recapture studies and to involve the public in local Eastern Box Turtle conservation efforts, we sought to use digital photography to supplement our use of PIT tags. The goals of our project were to: (1) test pattern recognition as a viable method of identifying individual Eastern Box Turtles; (2) to determining if top-down carapace, off-center carapace, or plastron photos were more diagnostic; and (3) to test the ability of a pattern recognition program to identify individuals from spatially different populations.

## **METHODS**

We collected photographs of Eastern Box Turtles from four areas in two states: the Oak Openings Region (OOR) of northwestern Ohio (41.556°N, 83.854°W) from 2004 – 2013, Ft. Custer Training Center (FCTC) and the adjacent Ft. Custer Recreation Area (FCRA) in Michigan's southwestern Lower Peninsula (42.324°N, 85.298°W) in 2005 and from 2011-2013, and from the Manistee National Forest (MNF) from 2009 – 2013 (43.875°N, 85.914°W). The impetus for obtaining images from multiple states was to determine whether or not images from different locations would result in false matches. Upon capture, top-down pictures were taken of the turtle's carapace and plastron (N = 610 of each; Ohio = 170; FCTC/FCRA = 307; MNF = 133). Additionally, we used a subset of images (194) taken without standardizing (i.e. pictures taken from any angle; hereafter "off-center") to compare the efficacy of these two methods. A minimum bounding rectangle that reduced the amount of visible background present was used to crop each image. The pictures from all locations were combined into carapace/off-center and

plastron categories within an image database. Turtles with a plastron length less than 7.0 cm were not used because their shell patterns do not appear to be fully developed (Hoss et al. 2013).

To identify individual Eastern Box Turtles in our study, we employed the relatively new pattern-recognition software Wild-ID (<http://www.dartmouth.edu/~envs/faculty/bolger.html>; Bolger et al. 2012). Wild-ID is stand-alone, open-source, multi-platform software that uses Java to implement pattern-recognition. The Wild-ID software uses a Scale Invariant Feature Transform operator (SIFT; Lowe 2004) to find and extract distinctive features invariant to the scale, rotation, viewpoint, local distortion and illumination of the image. The geometric arrangement of these SIFT features for each pair of images in the dataset are compared to one another. The program then calculates the goodness-of-fit between the images and assigns a matching score (values range from 0.0000 to 1.0000, where values closer to 1.0000 indicate a stronger match). A full description of these steps can be found in Bolger et al. (2012).

Once Wild-ID has completed the above steps, the user interface displays each focal image along with 20 of the top-ranked images (Figures. 1 and 2). This allows the user to assess the images visually and based on matching scores to conclude whether or not the focal image has a match. Once matching scores were assigned, we used the Kruskal-Wallis procedure and post hoc Mann-Whitney U-tests in R (R Development Core Team 2011) to compare carapace, off-center, and plastron matching scores. For this project, a correct response from Wild-ID was recorded if: 1) a successful recapture was identified, and 2) if no mismatches between individuals or sites occurred. The individual marking techniques used at the study sites, PIT tags in the OOR; shell notches at FCTC/FCRA and MNF, were used to verify the matches reported by Wild-ID.

## RESULTS

Mean recapture matching scores for carapace, off-center and plastron images were  $0.1918 \pm 0.0266$  (SE),  $0.0396 \pm 0.0111$ (SE), and  $0.1813 \pm 0.0373$  (SE), respectively (Figure 3 A-C). In no case was the highest-ranked image a different turtle if it was a recapture. Additionally, Wild-ID did not erroneously match images between sampling locations or states and verified recaptures, as confirmed by shell-notching and PIT tagging, at each location. Results of the Kruskal-Wallis tests indicated that there was a significant difference between matching scores of the three image categories ( $\chi^2 = 59.91$ ,  $df = 2$ ,  $P < 0.0001$ ); post-hoc tests indicated that there were significant differences in matching scores between carapace and off-center ( $P < 0.0001$ ), plastron and off-center ( $P < 0.0001$ ), but not between carapace and plastron ( $P = 0.6492$ ).

## DISCUSSION

Overall, Wild-ID performed ideally for identifying Eastern Box Turtle recaptures from carapace and plastron images, but exhibited lower efficacy when using off-center images. We initially expected plastron images to be less accurate because the plastron occasionally lacks distinguishing patterns (*MDC, pers. obs.*), but there was no statistical difference between the two. Plastron images were correctly matched just as often as carapace images and what little pattern may be present was sufficient for the program to identify a match. Because this method has not been tested before with Eastern Box Turtles, we wanted to make sure the program would not mismatch turtles from Ohio with turtles from Michigan; we found no mismatches, suggesting that Wild-ID is in fact suitable for identifying individual adult Eastern Box Turtles from their shell patterns.

Wild-ID represents a cost-and time-effective method of identifying individual Eastern Box Turtles for mark-recapture studies. However, this is not to say that this should be the only means of identifying turtles. In terms of long-lived species, like Eastern Box Turtles, whose patterns do not fully develop until later on in life, photo-recognition software alone may not be sufficient for life-long identification and should still be coupled with an additional marking technique until the long-term validity of computer-assisted pattern-recognition has been verified. Additionally, shell damage is a potential source of error, especially in the case of burn scars where a large portion of the shell may be rendered pattern-less. Photographic mark recapture relies on three conditions: 1) individuals can be photographed, 2) individuals bear some phenotypic pattern variation that easily identifies them from other individuals, 3) an individual's pattern does not vary through time (Bolger et al. 2012). The unique patterns of Eastern Box Turtle shells lend themselves to this method with the exception of the criterion that the pattern remains unchanged. Many turtles in our study had visible carapace imperfections that appeared to be the result of fires, mower blades, cars or general wear-and-tear. Shell damage may influence the results of programs such as Wild-ID, but as we saw with several of our turtles, as long as a majority of the pattern remained, Wild-ID could identify the image as a match. While carapace injuries and subsequent pattern alterations are relatively common, we observed that the plastron pattern remained relatively unchanged in most individuals. Whereas, carapace images have been suggested as the minimum requirement to identify a turtle (Wynn and Moody 2006), based on our results, we strongly suggest photographing the plastron as a secondary form of identification, particularly in the case of individuals with damaged carapaces.

While Wild-ID surpassed our expectations for pattern-recognition with Eastern Box Turtles, we noted that there are a number of ways future studies can improve the probability of

the program identifying a positive match. We identified soil and glare on the turtle's shell and amount of background as potential factors contributing to lower matching scores. We recommend turtles be photographed on a uniform background (e.g. blank piece of paper) in an area that reduces the amount of glare and shadow on the shell and cleaning soil from the shell prior to image-capture. Similarly, for studies specifically seeking to accurately record recaptures, we recommend using cameras with the same resolution. Glare, patterns marred with soil, and images with a disparity in resolution can all lead to reduced matching scores. The SIFT operator, however, will still work with whatever pattern is present in the image (D. Bolger, Dartmouth College, *pers. comm.*). Standardizing the method of image-capture in the manner we suggest should lead to increased matching scores, but is not necessary and will depend on the goals of the study. For instance, if the goal of the project is to reduce the amount of handling, off-center photos could be used as long as photos are taken from approximately the same angle. Again, as there is currently no way to confirm a correct identification prior to the development of a pattern on the shell, this method should be considered a supplement to traditional marking techniques.

We encourage researchers and naturalists who have taken photographs of Eastern Box Turtles in the past to utilize this software as a means of rapidly analyzing historical photographs to identify recaptured individuals. This sort of recognition software combined with citizen science programs (e.g. Davidson College Herpetology Laboratory's Box Turtle Mark-Recapture Program; Hester et al. 2008), could provide the means for mark-recapture studies of Eastern Box Turtles over a large geographic range.

#### **ACKNOWLEDGEMENTS**

Funding and support for this project were provided by Bowling Green State University, the Toledo Zoo, the Metroparks of the Toledo Area and the Michigan DNR. We would like to

thank A. Ihnken, T. Swem, S. Bristow, E. Cannarile, H. Tripp, O. Stacey, K. Klein, J. Forward, C. Schuttler, K. Akright, N. Birmingham, E. Cipolla, L. Liehr, L. Wisnieski, M. Archer, and C. Lucas, who gathered data in the field as well as the numerous people who called-in turtle sightings and sent us pictures, especially the staff of the Metroparks of the Toledo Area and members of the Toledo Naturalist's Association. Special thanks go to D. Bolger and B. Adams for their assistance and advice. All turtles in this study were handled in compliance with guidelines set forth by Bowling Green State University's IACUC and the Michigan and Ohio Departments of Natural Resources.

#### LITERATURE CITED

- Bolger, D.T., T.A. Morrison, B. Vance, D. Lee, and H. Farid. 2012. A computer-assisted system for photographic mark-recapture analysis. *Methods Ecol. Evol.* 3: 813–822.
- Budischak, S.A., J.M. Hester, S.J. Price, and M.E. Dorcas. 2006. Natural history of *Terrapene carolina* (box turtles) in an urbanized landscape. *Southeast. Nat.* 5: 191–204.
- Ferner, J.W. 2007. A review of marking and individual recognition techniques for amphibians and reptiles. *Herpet. Circ. No. 35*, Society for the Study of Amphibians and Reptiles. 72 pp.
- Fisher, A.R. 2007. Turtle assemblages in the Eastern Panhandle of West Virginia with and emphasis on *Pseudemys rubriventris* (LeConte). M.S. Thesis, Marshall University, Huntington, West Virginia, USA. 75pp.
- Gamble, L., S. Ravela, and K. McGargial. 2008. Multi-scale features for identifying individuals in large biological databases: an application of pattern recognition technology to the marbled salamander, *Ambystoma opacum*. *J. Appl. Ecol.* 45: 170–180.
- Hammond, P.S., S.A. Mizroch, and G.P. Donovan. 1990. Individual recognition of

- cetaceans: use of photo-identification and other techniques to estimate population parameters. *Rep. Int. Whal. Commn.*, Special Issue 12, 3–17.
- Hester, J.M., S.A. Budischak, and M.E. Dorcas. 2008. The Davidson College box turtle mark-recapture program: urban herpetological research made possible by citizen scientists. *In* J.C. Mitchell, R.E. Jung, and B. Bartholomew (eds.), *Urban Herpetology: Conservation and Management of Amphibians and Reptiles in Urban and Suburban Environments*. Volume 3, pp. 549–555. Society for the Study of Amphibians and Reptiles, Salt Lake City, Utah.
- Hoss, D.E., C.R. Hoss, and A.M. Gorgone. 2013. Photography as a means of identifying individual eastern box turtles, *Terrapene carolina carolina*. Poster session presented at: 2013 Box Turtle conservation Workshop; 2013 Mar 22-23, North Carolina Zoological Park, Asheboro, NC, USA.
- Katonas, K., and J.A. Beard. 1990. Population size, migrations and feeding aggregations of the humpback whale (*Megaptera novaeangliae*) in the Western North Atlantic Ocean. *Rep. Int. Whal. Commn.*, Special Issue 12: 295–305.
- Lowe, D. 2004. Distinctive image features from scale-invariant keypoints. *Int. J. Comput. Vis.* 60: 91–110.
- Markowitz, T.M., A.D. Harlin, and B. Würsig. 2003. Digital photography improves efficiency of individual dolphin identification. *Mar. Mamm. Sci* 19: 217–223.
- McGregor, P., and T. Peake. 1998. The role of individual identification in conservation biology. *In* T.M. Caro, (ed.), *Behavioral Ecology and Conservation Biology*, pp. 31–55. Oxford University Press, New York, USA.
- Plummer, M.V., and J.W. Ferner. 2012. Marking Reptiles. *In* R.W. McDiarmid, M.S. Foster,

- C. Guyer, J.W. Gibbons, and N. Chernoff (eds.), *Reptile Biodiversity: Standard Methods for Inventorying and Monitoring*, pp. 143-150. University of California Press, Berkeley, California.
- R Development Core Team. 2011. *R: A Language Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0, URL <http://www.R-project.org/>.
- Reisser, J., M. Proietti, P. Kinas, and I. Sazima. 2008. Photographic identification of sea turtles: method description and validation, with an estimation of tag loss. *Endangered Species Research* 5: 73–82.
- Sears, R.J., J.M. Williamson, F.W. Wenzel, M. Berube, D. Gendron, and P. Jones. 1990. Photographic identification of the Blue Whale (*Balenoptera musculus*) in the Gulf of St. Lawrence, Canada. *Rep. Int. Whal. Commn., Special Issue* 12: 335–342.
- Weiss, J.A. 2009. Demographics, activity, and habitat selection of the eastern box turtle (*Terrapene c. carolina*) in West Virginia. M.S. Thesis, Marshall University, Huntington, West Virginia, USA. 96 pp.
- Wynn, D.E., and S.M. Moody. 2006. *Ohio Turtle, Lizard, and Snake Atlas*. Ohio Biological Survey, Columbus, Ohio. 81 pp.

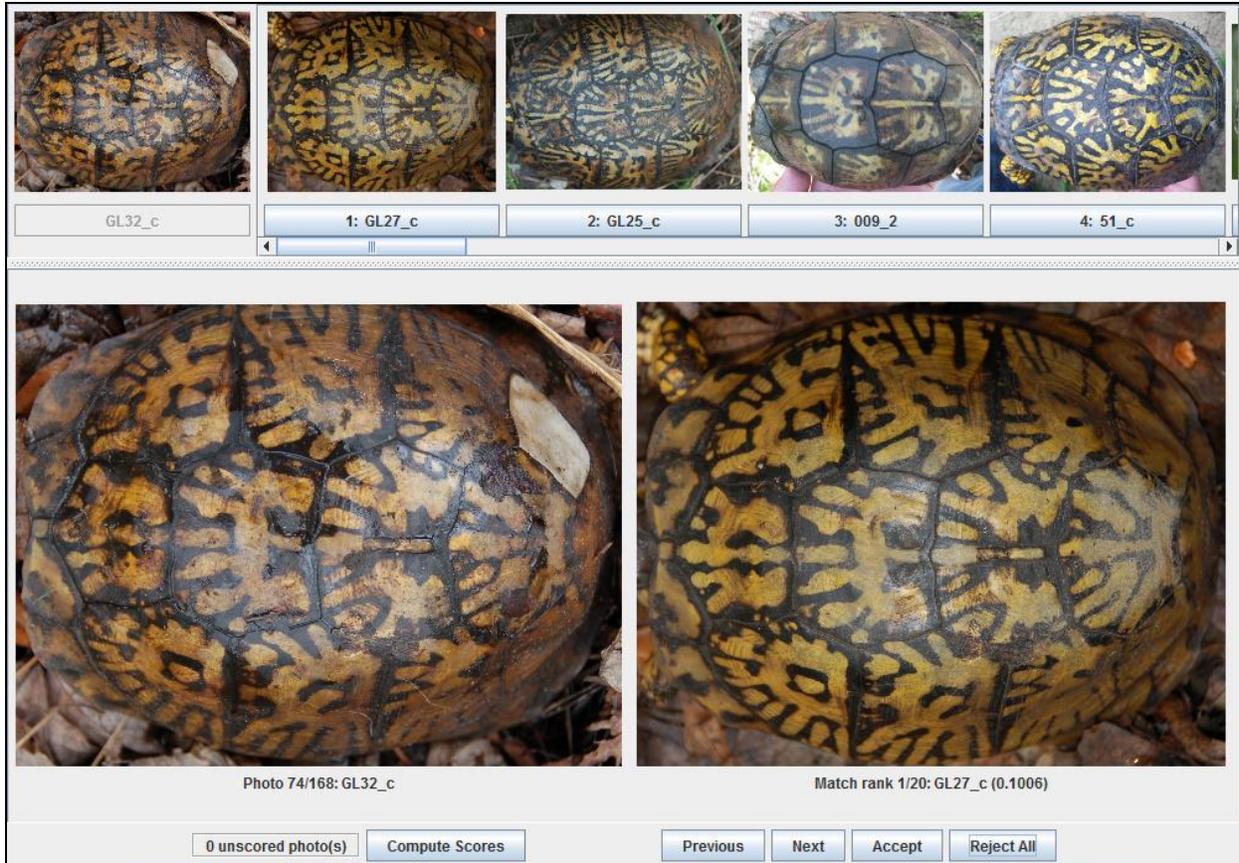


Figure 1.—User interface of WildID showing the focal image (bottom left and first in the row along the top), the active comparison window of the top-ranked matching score (bottom right), and the ranked potential images (top row). Note: the loss of pleural scute 4 in the recapture image (active comparison window).

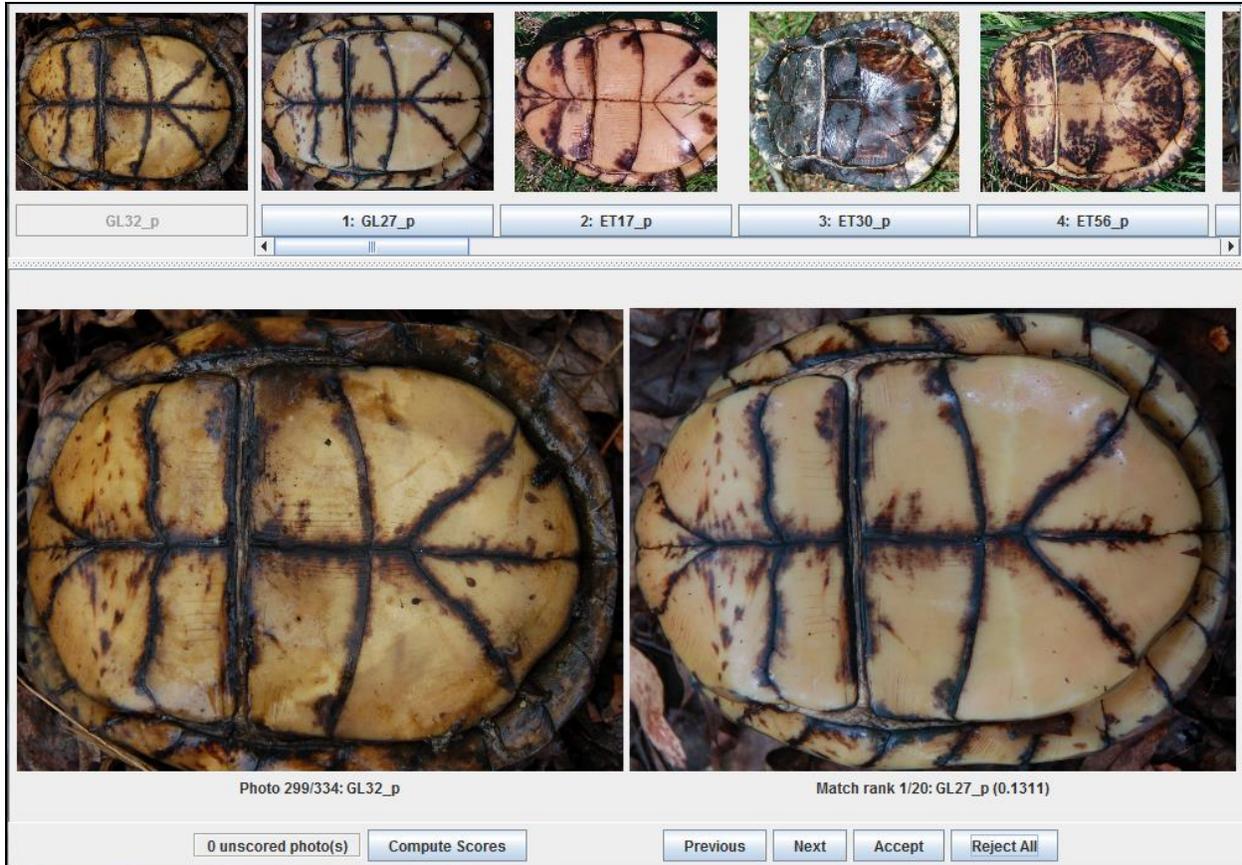


Figure 2. –User interface of WildID showing the focal image (bottom left and first in the row along the top), the active comparison window of the top-ranked matching score (bottom right), and the ranked potential images (top row). Note: these are the plastron images from the same turtle in Figure 1.

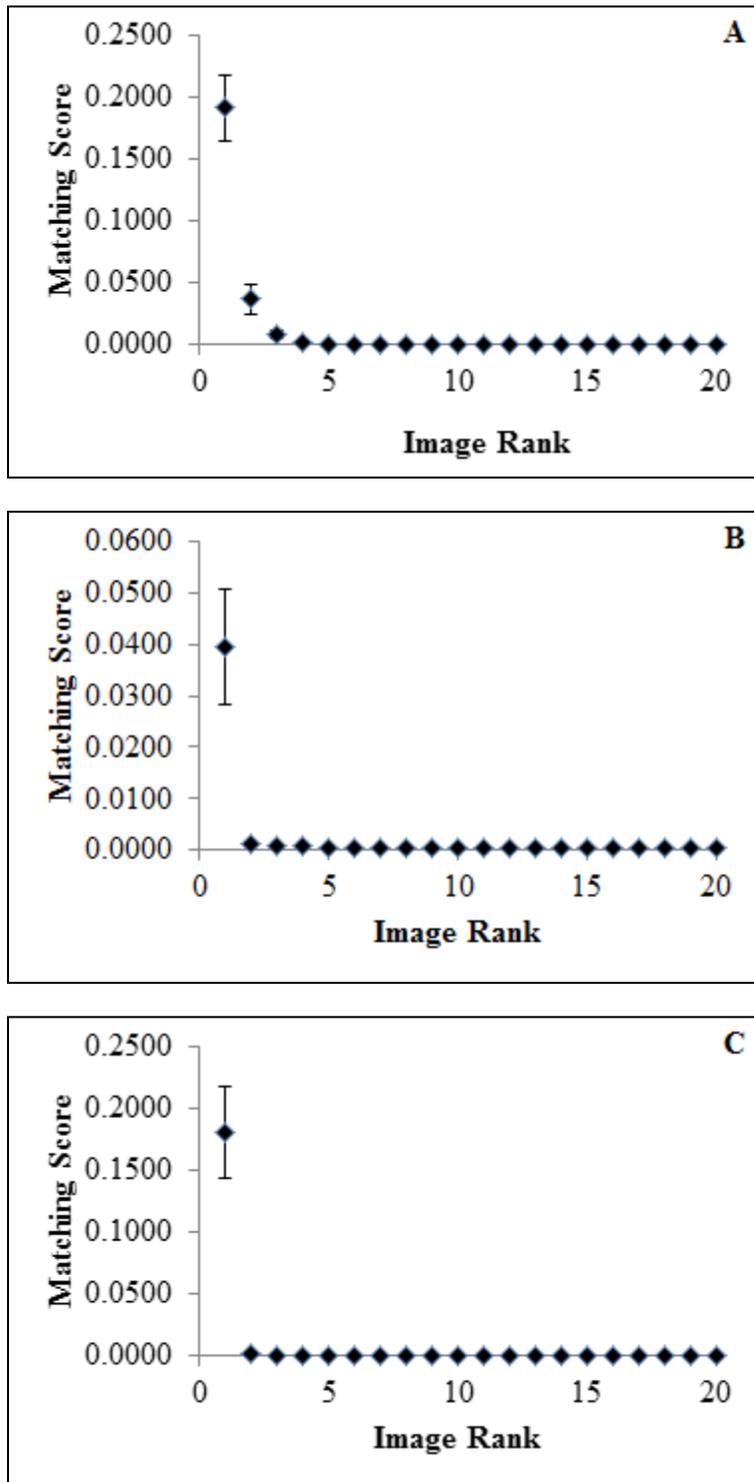


Figure 3 (A-C).—Top 20 average Eastern Box Turtle matching scores for: A) Carapace, B) off-center, and C) plastron images.

## CHAPTER V: CONCLUSIONS

*“...all models are approximations. Essentially, all models are wrong, but some are useful. However, the approximate nature of the model must always be borne in mind.” George E.P. Box (1987)*

In this dissertation, I explored the spatial ecology of Eastern Box Turtles across multiple scales within a unique, biodiversity hotspot, the Oak Openings Region. In the first chapter I examined temporal and multi-scale movements, and habitat and area use. I found that in general, daily movements and home range sizes of Eastern Box Turtles at my study site were larger than previously reported by most other studies. Additionally, turtles in my study tended to use more wet forests than what is typically reported for this species. These findings support the need for studies such as this across a wide geographic range in order to further elucidate how local variation in habitat and landscape features might influence conservation of this species.

In the second chapter, I predicted the distribution of Eastern Box Turtles within the Oak Openings Region in two ways: 1) a general model including data points from all four years of this study, and 2) temporally by creating monthly models and comparing the amount of variation within and between years. I employed relatively new methods to improve model accuracy, reduce the amount of overfitting, and increase predictive accuracy. The general model suggested that a large portion of highly-ranked suitable areas lie within the boundaries of protected properties. Temporal models depicted seasonal shifts in distribution and associated landscape variables, with the most drastic shifts occurring among medium and high-ranked sites. The location of box turtle habitat hotspots and areas where they are likely to be moving through can be used by managers to plan management activities in a manner that will reduce their impact on box turtles.

In the third chapter, I examined the early-season effects of prescribed fires on Eastern Box Turtles. Three of the 11 turtles with transmitters on burn units died as a result of the fires. When compared to reference turtles, turtles on the burn units did not alter their daily movements or home ranges following a fire, but did exhibit slightly altered habitat utilization, lost more weight, and displayed less overwintering site fidelity. Behavioral responses of the turtles that survived the fires suggest that this species is capable of avoiding prescribed fires conducted at this time of year. While the mortality, particularly among gravid females, is concerning, it is difficult to assess the impact of the burns without a detailed population estimate. It is important to note that these findings are short-term responses and it is possible for this species to recover from losses and altered habitat if the disturbance is not chronic. In the Oak Openings Preserve Metropark, the current method of burning small patches on a rotational basis, with slow backing fires seems to be the best method of conducting burns during the box turtle active season. This may not be true in other parks within the region and future studies should seek to estimate population sizes within the various managed properties.

In the fourth chapter, I pioneered the use of computer-assisted photo-recognition as a method of identifying individual Eastern Box Turtles. I found no difference in diagnostic ability between carapace and plastron photos, but find that off-center photos performed poorly in comparison. These results highlight the usefulness of the low-cost method as an alternative to traditional, invasive marking techniques such as shell-notches or PIT tags. Photo-recognition provides a method by which citizen scientists can get involved and help provide an independent dataset with which to test the predictions made by distribution and connectivity models.

These chapters provide insight into the spatial ecology of Eastern Box Turtles in a unique region from multiple scales. This work contributes to a growing body of knowledge regarding

box turtles and highlights important factors regarding their conservation. Conservation efforts for this species should focus on promoting connectivity in a highly-fragmented landscape and attempt to find ways to reduce sources of adult mortality (e.g., roads or prescribed fires).

## APPENDIX A: EASTERN BOX TURTLES ENCOUNTERED DURING THE 2015-2015

## FIELD SEASONS

Date (yyymmdd)	Recapture	ID#	Age Class	Annuli	Sex	Carapace Length (mm)	Carapace Width (mm)	Weight (g)	PIT
120322	Y	1	A		M	148.09	105.39	620	
120322	Y	2	A	15	F	140.24	107.89	630	016110600
120329	Y	3	A	15	M	154.04	110.31	620	061259281
120329	Y	4	A	18	M	160.11	118.98	725	016122312
120322	Y	6	A	16	F	157.21	113.88	760	016262549
120408	N	7	A	14-15	F	129.86	98.29		
120524	N	8	A	15	M	146.72	105.73	545	095116618
120427	N	9	A	16-17	F	136.81	101.38	550	016055816
120511	N	10	A	14-15	F	151.03	111.48	735	095584312
120521	N	11	A	20+	F	141.85	104.53	560	095117088
120522	N	12	A	15-16	F	139.31	94.8	540	095118878
120524	N	13	A	13	F	132.1	103.41	580	016259636
120525	N	14	A	16	F			520	095584326
120610	N	15		25+	F	158.47	115.39	710	016098039
120615	N	16	A	15	F	139.35	100.07	520	016051815
120616	N	17	A	20+	F	148.51	115.8	685	016091256
120618	N	18	A	15	M	144.46	105.94	490	016065271
120629	N	19	A	18-20	M			642	016097067
120630	N	20	A	10	M	132.31	103.83	415	016069125
120705	N	21	A	~19	F	131	113	427	016048044
120730	N	22	A	21+	F	139.99	104.88	905	016088861
120731	N	23	A	<10	F	118.61	90.92	470	467 67E 0C47
120804	N	24	A	17	F	142.49	103.35	595	
120808	Y	19	A	18-20	M	190.47	143.36	665	016097067
120809	N	25	A		M				016053055
120809	N	26	A		M				
120813	N	27	A	~15	F	138.58	99.53	510	016053352
120828	N	28	SA?	14	F	123.08	93.17	330	016086376
120815	N	29	A	22+	M	135.6	105.8	540	016092084
120925	N	30	A	14-15	M	146.53	102.95	530	016078287
130418	Y	2	A		F	141.45	91.3	640	016110600
130506	Y	4	A	18+	M			690	016122319
130422	Y	6	A		F	114.6	82.1	720	016262549
130417	Y	7	A		F			580	016079097
130422	Y	8	A		M	118.1	91.51	590	095116618
130417	Y	9	A	18	F			570	016055816
130418	Y	10	A		F	118.23	81.83	770	095584312

Date (yyymmdd)	Recapture	ID#	Age Class	Annuli	Sex	Carapace Length (mm)	Carapace Width (mm)	Weight (g)	PIT
130418	Y	11	A	21+	F	118.6	87.52	630	095117088
130418	Y	12	A		F	119.47	94.32	550	095118878
130405	Y	13	A	15+	F	110.37	87.56		016259636
130418	Y	14	A	18+	F	114.19	83.4	520	095584326
130407	N	31	A		M	151			
130426	N	32	A	Old	M	115.56	84.1	545	016049377
130426	N	33	A	16+	M	118.14	84.74	680	016051800
130502	N	34	A	16+	M	114.13	82.12	420	013313127
130502	Y	35	A	11	M	149.12	80.45	600	011271605
130506	N	36	A	11	F	114.38	80.25	550	013310801
130507	N	37	A		M	86.8	151	490	013359631
130507	N	38	SA	8	F	116	86.5	239	013325832
130514	N	39	SA	7	F	109.15	82.59	219	013520528
130515	N	40	A	20+	M	145.52	103.41	510	013311893
130520	Y	22	A	21+	F				016088861
130521	N	41	A	15	F	142.92	101.46	635	013361624
130521	N	42	A	26	M	101.09	85.76	475	013377768
130521	N	43	A	20	F	140.47	110.46	590	013334290
130521	N	44	A	16	F	127	91.46	690	013339082
130521	N	45	A	23	F	127	93.08	660	013530790
130522	N	46	A	17	F	140.16	101.28	625	013303591
130522	N	47	A	21	F	107.4	153.46	710	013334369
130527	N	48	SA	10	F	118.03	81.43	260	013518812
130524	Y	43	A	26	F				013334290
130529	N	49	A	13	F	144.85	107.79	750	013371584
130530	N	50	A	20	F	132.59	103.56	585	013526866
130609	N	51	A	20+	F				013365016
130613	N	52	A	19	M	146.17	102.29	560	013527309
130613	N	53	A	20	M	154	108.87	620	013309015
130617	N	54	A	19	F	130.9	100.4	500	015030534
130626	Y	43	A		F				013334290
130705	Y	25	A	15-17	M	158.34	108.27	575	016053055
130708	N	56	A	17	M	151.49	118.16	660	015050609
130711	N	57	A	18	F	150.43	111.08	690	014872282
130715	N	58	A	18-20	M	165.1	118.35	740	015008056
130717	Y	42	A		M				013377768
	N	59	A	16	M	150.91	111.9	610	014859027
130808	N	60	A	13-15	M	154.82	116.46	700	
130820	N	61	A	18+	F	154.58	114.85	690	014873107
130927	N	62	A	22+	M	159.07	108.87	650	014878622

Date (yyymmdd)	Recapture	ID#	Age Class	Annuli	Sex	Carapace Length (mm)	Carapace Width (mm)	Weight (g)	PIT
130902	N	63	A	18-19	F	150.1	107.9	675	014890003
130911	N	64	A	15-17	M	152.71	110.09	650	015033089
131011	N	65	A	23+	M	154.14	117.07	655	015048829
131003	Y	30	A		M	146.49	102.7	515	016078287
140426	N	66	A	18	F	141.78	95.47	690	015006272
140426	N	67	A	15-17	M	138.79	109.1	510	015051362
140426	N	68	A	17-18	M	141.32	93.55	520	014882256
140503	N	69	A	15	F	197.4	95.34	780	014858839
150506	N	70	SA	9		119.47	87.4	320	014891598
140424	Y	15	A	25+	F	142.3	116.93	820	
140424	Y	46	A	?	F	141.85	103.07	610	
140426	Y	61	A	20+	F	151.34	114.29	710	015052009
140424	Y	50	A		F			570	
140424	Y	57	A	20+	F	150.37	112.82	745	
140424	Y	49	A	14	F	143.01	108	775	
140424	Y	55	A	17	F			610	
140429	Y	2	A		F	141.31	108.04	635	016110600
140515	Y	41	A	16	F	141.46	102.32	630	013361624
140516	Y	13	A	51+	F	130.2	101.91	590	016259636
140522	Y	35	A	15	M	149.93	93.95	625	011271605
140523	Y	33	A	21+	M	149.88	111.72	680	016051800
140528	Y	68	A	19	M	141.86	90.13	510	014882256
140529	N	71	J	4	F	88.75	59.49	120	014873007
140601	Y	27	A	15	F	139.04	100.47	540	016053352
140602	N	72	A	22	F	151.3	109.19	790	015055797
140602	N	73	A	20+	F	154.27	111.04	790	014867771
140602	N	74	A	20+	F	130.72	87.4	640	015038799
140603	N	75	A	18	F	143.48	110.03	720	014870040
140604	N	76	A	16	F	144.43	110.48	640	015001322
140615	N	77	A		F	143.1	109.27	680	014875858
140617	N	78	A	24+	F	153.55	109.95	755	004889827
140627	N	79	A	25	F	145	113	705	015006887
140714	N	80	A		F	145.1	102.5	600	013326537
140714	N	81	A		F	141.5	110	680	013364860
140715	Y	35	A	~21	M	154	107	580	011271605
140722	Y	32	A	14+	M	143	101.5	530	016049377
140729	N	82	A	20+	M	123.68	109.98	500	031036585
140801	Y	52	A	17	M	150.47	103.69	535	013527309
140808	Y	35	A	15+	M	154.04	108.64	575	011271605
140808	N	83	A	23	F	144.69	104.41	630	030866827

Date (yyymmdd)	Recapture	ID#	Age Class	Annuli	Sex	Carapace Length (mm)	Carapace Width (mm)	Weight (g)	PIT
140813	N	WW2	SA	12	M	121.95	95.56	305	030843770
140822	N	84	A	23	F	142.68	104.61	590	031032047
140827	N	85	A	20+	M	154.05	112.52		031074552
140901	N	86	A	~30	M	149.53	107.8	625	031018381
140909	N	106	A			158.72	113.99	750	031073573
140909	Y	108	A			150.45	107.82	630	099381025
140909	Y	41	A		F	140	101.96	640	013361624
140909	N	99	A	21	M	148.65	114.55	745	031088833
140909	N	98	A	13	M	152.65	112.33	690	030894782
140911	N	93	A		F	131.42	102.28	500	030895613
140911	N	94	SA	13	M	129.22	97.36	465	030881090
140911	N	102	A		M	139.13	101.34	445	031100103
140911	Y	27	A		F	139.03	99.57	530	016053352
140911	Y	20	A		M	143.96	114.6	590	016069125
140910	N	97	A	15	M	146.56	111.89	630	030846599
140910	N	100	A	19	F	141.65	113.77	650	031005316
140910	N	112	A	17	F	149.3	107.94	630	031075569
150412	N	113	A	18-20	M			630	031005035
150415	N	114	SA	10	M				
150416	Y	44	A		F				013339082
150417	N	115	A		F				013019001
150424	Y	19	A		M				016097067
150501	Y	44	A		F			640	013339082
150518	N	116	A	22-24	F			555	031083628
150520	Y	10	A	~20	F	150.76	111.55	765	
150520	N	117	A	~17	F	152.65	111.24	850	031090820
150522	N	118	A	?	F	138.04	103.35	540	
150525	N	119	A	18-20	F	138.06	101.38	620	030844260
150526	N	WG1	A	20+	F	151.28	112.36	670	031065776
150613	N	120	A	24	F	134.65	96.47	480	030884878
150616	N	WG3	A	21+	F	140.48	102.88	640	030845067
		County							
150618	N	Rd K	A	15-18	F	140.05	104.83	680	031016037
150622	N	121	A		M	154.35	113.04	715	030866266
150622	N	122	A		M	146.09	111.58	455	031018854
150630	Y	82	A	19	M	123.69	110.55	485	031036585
150713	N	123	A	18+	F	155.32	115.59	735	030876853
150714	N	124	SA	10	F	115.37	81.45	340	031040045
150718	N	125	A		M	142.94	108.86	600	030894536
150726	Y	33	A		M	150.16	113.7	685	016051800
150726	Y	125	A		M				030894536

Date ( <i>yymmdd</i> )	Recapture	ID#	Age Class	Annuli	Sex	Carapace Length (mm)	Carapace Width (mm)	Weight (g)	PIT
150726	Y		A	18	F	139.58	97.16	605	041060334
150726	N	126	A	18-20	F	133.66	103.01	570	030870818
150806	Y	25	A	21+	M		107.94	530	016053055
150806	Y								
150808	N		A	23+	M	155.39	112.43	740	
150925	N		A	20+	M	152.43	107.13	600	
150925	Y	52	A		M				013527309
150930	N		A	22+	M	148.56	110.39	540	
151109	Y	65	A	23+	M			665	

## APPENDIX B: EASTERN BOX TURTLE NESTING DATA

Year	Turtle	Egg #	Length (mm)	Width (mm)	Weight/turtle (g)	Fate
2012	12	1	N.A.	N.A.	N.A.	Depredated (ants)
		2	N.A.	N.A.	N.A.	Hatched
		3	N.A.	N.A.	N.A.	Hatched
		4	N.A.	N.A.	N.A.	Hatched
		5	N.A.	N.A.	N.A.	DUD (ants)
		6	N.A.	N.A.	N.A.	DUD
		7	N.A.	N.A.	N.A.	DUD
2012	Unknown	1	N.A.	N.A.	N.A.	Depredated (ants)
		2	N.A.	N.A.	N.A.	Depredated (ants)
		3	N.A.	N.A.	N.A.	Depredated (ants)
		4	N.A.	N.A.	N.A.	Squished
		5	N.A.	N.A.	N.A.	Hatched
2013	72	1	29.44	33.02	8.69	Hatched
		2	30.17	33.55	8.69	Hatched
		3	29.19	32.81	8.69	Hatched
		4	29.69	34.35	8.69	Hatched
		5	29.67	34.75	8.69	Hatched
		6	31.02	33.80	8.69	Hatched
		7	29.48	32.96	8.69	Hatched
		8	30.45	34.49	8.69	Hatched
2013	49	1	N.A.	N.A.	N.A.	DUD
		2	N.A.	N.A.	N.A.	DUD
		3	26.83	30.13	7.57	Hatched
		4	29.31	32.24	7.57	Hatched
		5	27.83	29.34	7.57	Hatched
		6	26.27	29.79	7.57	Hatched
		7	28.99	32.22	7.57	Hatched
		8	29.34	31.55	7.57	Hatched
		9	21.91	28.96	7.57	Hatched

Year	Turtle	Egg #	Length (mm)	Width (mm)	Weight/turtle (g)	Fate
2013	50	1	32.85	29.06	N.A.	Hatched
		2	31.00	28.65	N.A.	Hatched
		3	31.54	27.99	N.A.	Hatched
		4	31.47	28.70	N.A.	Hatched
		5	32.85	28.60	N.A.	Hatched
		6	N.A.	N.A.	N.A.	DUD
2013	12	1	N.A.	N.A.	N.A.	DUD
		2	N.A.	N.A.	N.A.	DUD
		3	N.A.	N.A.	N.A.	DUD
		4	N.A.	N.A.	N.A.	DUD
		5	N.A.	N.A.	N.A.	DUD
		6	N.A.	N.A.	N.A.	DUD
		7	N.A.	N.A.	N.A.	DUD
		8	N.A.	N.A.	N.A.	DUD
		9	N.A.	N.A.	N.A.	DUD
2013	6	1	33.13	29.59	~5-6	Hatched
2013	13	1	31.30	27.01	~5-7	Hatched
		2	N.A.	N.A.	N.A.	DUD
		3	N.A.	N.A.	N.A.	DUD
		6	N.A.	N.A.	N.A.	DUD
		5	N.A.	N.A.	N.A.	DUD
		6	N.A.	N.A.	N.A.	DUD

## APPENDIX C: INSTITUTIONAL ANIMAL CARE AND USE COMMITTEE APPROVAL



Office of Research Compliance  
309A University Hall  
Bowling Green, OH 43403-0183  
Phone: (419) 372-7716  
Fax: (419) 372-6916  
E-mail: hsrb@bgnet.bgsu.edu

April 18, 2012

Dr. Karen Root  
Biological Sciences  
Bowling Green State University

Re: IACUC Protocol 12-005

Title:

*Examining the Impacts of Land Management on Eastern Box Turtles (*Terrapene c. carolina*) in a Fragmented Landscape with Emphasis on Characterization and Predication of Nesting and Overwintering Sites*

Dear Dr. Root:

On **April 14, 2012** the above referenced protocol received **final approval** after review of the requested modifications by Designated Member Review. The modifications have been incorporated into the official copy of your protocol (see modifications below).

This approval expires on April 13, 2013, by which time renewal must be requested if you wish to continue work on the protocol. The Office of Research Compliance will send notification reminding you of the need for renewal in advance of that date.

Please have all members of your research team read the approved version of the protocol. Please also remember to keep a copy of the approved protocol in the animal facility room(s) in which your animals are housed and in any associated procedure rooms (contact the UAF staff for assistance in this regard).

Please consult with the staff of the Animal Facility about your requirements to get started on this project. Good luck with your project.

Sincerely,

Hillary Harms, Ph.D.  
IACUC Administrator

**Incorporated Modifications:**

1. In item 3, "injection" was selected.
2. In item 8, "PIT" tag was defined.
3. In item 12:
  - More information was provided to clarify the method and site of PIT tagging.
  - Information about PIT tagging guidelines was provided.
  - Indicated that animals are handled each time they are located, which varies with the time of year.
  - Clarified that PIT tags can identify a turtle at close range and transmitters are used to track a turtle.
4. A copy of the appropriate permit(s) were provided.



Office of Research Compliance  
309A University Hall  
Bowling Green, OH 43403-0183  
Phone: (419) 372-7716  
Fax: (419) 372-6916  
E-mail: [hsrb@bgnet.bgsu.edu](mailto:hsrb@bgnet.bgsu.edu)

February 25, 2013

Dr. Karen Root  
Biological Sciences  
Bowling Green State University

Re: Annual Renewal of IACUC Protocol 12-005

Title:

*Examining the Impacts of Land Management on Eastern Box Turtles (*Terrapene c. carolina*) in a Fragmented Landscape with Emphasis on Characterization and Predication of Nesting and Overwintering Sites*

Dear Dr. Root:

On February 20, 2013 the annual renewal for the above referenced protocol was **approved** after review by the IACUC. This renewal is in effect for one calendar year and expires on February 19, 2014. Please consult with the staff of the Animal Facility about any special needs you might have to continue with this project.

**Comment(s):**

You have used 26 turtles and have been approved for 30. You may need to request an increase in the number of turtles approved for use.

Sincerely,

Hillary Harms, Ph.D.  
IACUC Administrator



Office of Research Compliance  
309A University Hall  
Bowling Green, OH 43403-0183  
Phone: (419) 372-7716  
Fax: (419) 372-6916  
E-mail: hsrb@bgsu.edu

February 24, 2014

Dr. Karen Root  
Biological Sciences  
Bowling Green State University

Re: Annual Renewal of IACUC Protocol 12-005

Title:

*Examining the Impacts of Land Management on Eastern Box Turtles (*Terrapene c. carolina*) in a Fragmented Landscape with Emphasis on Characterization and Predication of Nesting and Overwintering Sites*

Dear Dr. Root:

On February 21, 2014 the annual renewal for the above referenced protocol received **final approval** after review of the requested modifications by the IACUC. This renewal is in effect for one calendar year and expires on February 20, 2015. Please see the next page for a summary of the approved modifications.

Please consult with the staff of the Animal Facility about your requirements to continue with this project.

Sincerely,

Hillary Snyder, Ph.D.  
IACUC Administrator



BOWLING GREEN STATE UNIVERSITY

Office of Research Compliance

DATE: February 16, 2015

TO: Karen Root, Ph.D.

FROM: Bowling Green State University Institutional Animal Care and Use Committee

PROJECT TITLE: [687071-2] Examining the impacts of land management on eastern box turtles (*Terrapene c. carolina*) in a fragmented landscape with emphasis on characterization and prediction of nesting and overwintering sites

IACUC REFERENCE #: 12-005

SUBMISSION TYPE: Continuing Review/Progress Report

ACTION: ACKNOWLEDGED/APPROVED

APPROVAL DATE: February 16, 2015

EXPIRATION DATE: April 13, 2015

REVIEW TYPE: Full Committee Review

Thank you for your submission of Continuing Review/Progress Report materials for the above referenced research project. The Bowling Green State University Institutional Animal Care and Use Committee has acknowledged/approved your Progress Report. This was the final renewal for this project. Note that the project expires on April 13, 2015.

If you have any questions, please contact the Office of Research Compliance at 419-372-7716 or [hsrb@bgsu.edu](mailto:hsrb@bgsu.edu). Please include your project title and reference number in all correspondence with this committee.

This letter has been electronically signed in accordance with all applicable regulations, and a copy is retained within Bowling Green State University Institutional Animal Care and Use Committee's records.