FINAL REPORT

Distribution, Abundance, and Seasonal Habitat Use Patterns in Ornate Box Turtles (*Terrapene ornata*) in South Dakota

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EXECUTIVE SUMMARY

Ornate box turtles (*Terrapene ornata*) range from Wisconsin and northern Indiana through the central Great Plains from southern South Dakota to Arizona, northern Mexico, and the Gulf Coast in Texas. Concern over population levels of all North American box turtle species led to their 1994 listing in Appendix II of the Convention of International Trade of Endangered Species of Wild Fauna and Flora (CITES). The ornate box turtle is also listed as "*Near Threatened*" by the International Union for Conservation of Nature (IUCN). Considered a "*Species of Greatest Conservation Need*" within South Dakota, ornate box turtles were known from only 19 records prior to this study. Such federal and state designations indicate urgency in establishing conservation measures for this species. In 2009, ornate box turtles were sighted on the Pine Ridge Reservation during an Oglala Lakota College (OLC) field ecology course. Since little is known about ornate box turtles in our state, these sightings created an ideal opportunity to initiate further study on this species. Subsequently, the following objectives were proposed: 1) document the macro- and microhabitat use throughout the active season (May through September); 2) estimate home range size and describe movements; 3) document daily and seasonal activity periods; 4) estimate population size; 5) estimate the geographic range of ornate box turtles in South Dakota through the use of species distribution model (MaxEnt); and 6) provide training in ecological field research to OLC students. In addition, aspects on reproduction and growth *T. ornata* are also reported.

The 320 ha study area is partially located on privately owned land within the Pine Ridge Reservation, Shannon County in south-central South Dakota. The exact location is omitted due to the risk of illegal collection for the commercial pet, but can be made available on request. Elevation of the study area is approximately 2820 feet. Average annual precipitation is 15.52 inches at nearby Scenic, South Dakota over a 31-year period, with monthly precipitation ranges from a low of 0.29 inches in December to a high of 3.10 inches in June. Average monthly temperatures varied from 8^0 F in January to a high of 89.4^0 F in July. The study area receives light grazing by livestock during the summer. Well-developed, windblown sands that form dunes that extend into the Sand Hills Region of Nebraska are dominant. Two macrohabitat types, with transition zones in between, were identified within the study area. We termed these uplands and lowlands, but they are described in the literature as Sands and Clayey. Upland habitat was characterized by deep sandy soils and the lowland habitat by clay soils. Dominant grasses of the Sands portion of the study area include, prairie sandreed (*Calamovilfa longifolia*), needle and thread (*Hesperostipa* sp), blue grama (*Bouteloua gracilis*), and hairy grama (*Bouteloua hirsuta*). Common forbs included cuman ragweed (*Ambrosia psilostachya*) and slimflower scurfpea (*Psoralidium tenuiflorum*). The common and dominant shrub was the aromatic sand sagebrush (*Artemisia filifolia*). Plants on the Clayey portion of the study area include common grasses, western wheatgrass (*Pascopyrum smithii*), green needlegrass (*Nassella viridula*), and blue grama (*Bouteloua gracilis*). Common forbs were scarlet globemallow (*Sphaeralcea coccinea*), and cuman ragweed (*Ambrosia psilostachya*). Shrubs present were fringed sagewort (*Artemisia frigida*), and cactus (*Opuntia* sp).

Dogs specifically trained to hunt box turtles were used during survey periods. Turtles were also found by researchers walking in throughout the habitat during the turtle's active season, from the end of April through mid-October in both years. A few turtles were captured as

they crossed roads, or were brought to us by citizens in response to "wanted posters" placed in key areas.

A total of 170 living turtles (67 males, 69 females, and 34 juveniles) and nine dead turtles were found during the course of the two-year study (2010 - 2011). While most of these specimens (87%) were found at our study area, a few (13%) were located in scattered locations in Shannon, Jackson, Pennington, and Todd Counties. Once a turtle was located, standard measurements were taken, sex was determined, and age estimated. Turtles were individually marked utilizing unique combinations of shell notches. Thirteen females were x-rayed to detect oviducal eggs. A total of 25 ornate box turtles (10 males, 14 females, and 1 juvenile) in 2010 and 24 turtles (12 females, 12 males) in 2011 were fitted with radio transmitters and temperature data loggers. These turtles were located weekly with a minimum interval of three days during the active season for home range size estimations. At two-week intervals, microhabitat measurements such as the visual obstruction readings (VOR) and the canopy cover were collected at the turtle location. For comparative purposes, these measures were also taken at designated distances from the turtle utilizing a stratified random design. Ornate box turtles highly selected for greater VORs during both years compared to the availability. Preferences by the turtles were generally for shrubs and sand sagebrush, with less preference of grass and forbs in more open areas. This extremely high preference for sand sagebrush would indicate that if habitat change did occur with the shrub component, negative impacts with these turtles are expected, as the sagebrush facilitates thermoregulation by providing cool areas during the summer and favorable hibernation sites during the winter.

Most of the radio-tracked turtles burrowed underground for the winter by the middle to the end of October in both years. The mean direction of most of the hibernation sites was south-facing, where winter temperatures presumably are warmer. All of the hibernation sites were located on upland habitat, characterized by loose sandy soils, where *T. ornata* burrow into the sand below the frost line or occasionally use mammal burrows as an underground retreats. Temperature data loggers readings estimate the maximum depth of hibernation for ornate box turtles in South Dakota as 105 cm (± 7 cm). The hibernation depth varied with soil temperature, and turtles moved deeper as the winter progressed and soil temperature decreased. Most of the turtles did not experience freezing temperatures, and spring emergence was estimated around early May. Precise hibernation dates were not determined, but the hibernation period for *T. ornata* in South Dakota, was projected to range from 161 to 182 days. Most of the turtles hibernated in clusters, and site fidelity was observed with turtles hibernating in the same or nearby locations from the previous year.

Annual home ranges for both years were calculated for adult box turtles using the Minimum Convex Polygons (MCP) for total area, and the local convex hull (LoCoH) nonparametric Kernel method for utilization distribution areas. Home range calculations utilizing both methods varied greatly among turtles, raging from 0.35 - 89 ha. However, home range means were not statistically significant different either between sexes or between years. Box plotted home range medians detected three extreme outliers, possibly transient individuals, that when removed from the analyses showed males having bigger home ranges than females in 2010 but not in 2011. Home ranges overlapped both between and within sexes, consequently no territoriality was observed. When comparing MCP and LoCoH home ranges for both sexes, similar home range sizes were observed although females and males use their areas differently. Females had a more centralized utilization distribution area, or smaller core areas than males. The cause/s of the differences in utilization distribution between sexes is/are not clear, although

nesting behavior may be an important factor resulting in a smaller utilization distribution area for females.

Based on this study and historical box turtles records, and using local environmental variables, the maximum entropy species distribution model (MaxEnt) predicted potential suitable habitats for *T. ornata* in South Dakota as the south west-central portion of the state. Highest probability of box turtle presence is Shannon, Bennet, and Todd counties, followed by Jackson, Pennington, Mellette, Tripp, and Gregory. The lowest probability is on the west corner of the state in Fall River and on the central part, Haakon. The Isothermality variable was the most important predictor (41.8%) of *T. ornata* habitat distribution. Although this prediction seems to be very useful in helping plan management programs around the existing populations, discovering new populations, or setting priorities to restore natural habitat for more effective species conservation, it should be used with caution. This was the first range prediction for this species in South Dakota, and more refined analysis are needed.

Copulation was observed on six occasions, representing every month from April through September. Of 11 females x-rayed on 7 June 2010, eggs were observed in seven (64%). Clutch sizes ranged from 2 - 5, with an average of 4.3. Based on growth-ring size data, estimated hatchling plastron length averaged 31.7 mm, with males larger than females, but not statistically significant. Growth was rapid during the first few years of life, but slowed thereafter. Growth rates were 28.3, 22.4, and 15.7% during the first three years, but varied from 8.7 to 9.8% to age nine. Beyond that age, samples were too small to estimate growth patterns. Adult females were larger than adult males. Male and female turtles can mature by age nine, and the smallest adult male we observed had carapace and plastron lengths of 103.3 and 104.0 mm, respectively. The smallest carapace and plastron lengths measured for adult females were larger, at 110.2 and 113.4 mm, respectively. Comparisons of reproductive and growth parameters of the South Dakota population were made with those published from studies in other parts of the species' range.

OLC undergraduate students were exposed to different aspects of research related to the ecology of the box turtle, including field and laboratory work, and were able to bring this information back to the scientific community, the college, and tribal communities. Ornate box turtle project results were also featured at the Dakota Amphibian and Reptile News, and disseminated on local and national scientific conferences.

A comprehensive list of conservation concerns, including threats and issues, and management recommendations for ornate box turtles are presented.

INTRODUCTION

The following report details new findings related to the biology and ecology of the ornate box turtle (*Terrapene ornata*) in South Dakota. This study was conducted between May 2010 to May 2012 and was funded in part by federal funding through State Wildlife Grant T-44-R-1, Federal Aid Study # 2452, administered through the US Fish and Wildlife Service, South Dakota Department of Game, Fish, and Parks (SDGFP) Wildlife Action Plan Competitive Grant, National Science Foundation (NSF) - Tribal Colleges and Universities Program (TCUP), and Oglala Lakota College. Description, distribution, threats and issues in South Dakota sections were obtained from Smith and Quinn (2012).

Description

The ornate box turtle (*Terrapene ornata*) is a 4 - 5 inches terrestrial turtle with a hinged, completely closeable shell. It is highly ornamented above and below with yellow radiating lines on the otherwise brownish shell (Conant and Collins 1991, Ernst and Lovich 2009).

Distribution

The ornate box turtle ranges from Wisconsin and northern Indiana through the central Great Plains from southern South Dakota to Arizona, northern Mexico, and the Gulf Coast in Texas (Dodd 2001, Ernst and Lovich 2009, Redder et al. 2006).

South Dakota:

- Historical: Over (1923) reported that this species was found in the Sandhills region in the southwestern corner of the state. He also mentioned that it did not occur in the Black Hills, north of Pennington County, or east of Washabaugh County (a former county, now merged with Jackson County). Later, Carr (1952) showed this species ranging across the entire southern portion of the state from Iowa to Wyoming.
- Current: Ornate box turtles are found in the Sandhills (sand prairie) region in the south-central part of the state (Backlund 2004, Ballinger et al. 2000, Kiesow 2006). This species appears to be commonly transported and released, as specimens have been found on LaFramboise Island by Pierre (Doug Backlund; Retired Wildlife Biologist; South Dakota Department of Game, Fish and Parks; personal communication).

Need for Conservation

For threats, issues, and protection of ornate box turtles in South Dakota see the conservation concern section at the end of the report. Concern over population levels of all North American box turtle species led to their 1994 listing in Appendix II of the Convention of International Trade of Endangered Species of Wild Fauna and Flora (CITES). The ornate box turtle is also listed as "*Near Threatened*" by the International Union for Conservation of Nature (IUCN). Considered a "*Species of Greatest Conservation Need*" within the state of South Dakota (South Dakota Department of Game, Fish, and Parks 2006), box turtles were known from only 19 specimens, with many of these records decades old, prior to the onset of this study (South Dakota Game, Fish and Parks Natural Heritage Database 2010). Such federal and state designations indicate urgency in establishing conservation measures for this species. However, since so little

is known about ornate box turtles in our state, further investigations linked directly to defining effective conservation programs are critically needed at this time.

About this Study

In 2009, while teaching a field ecology course in north-central Shannon County for Oglala Lakota College (OLC), the principle investigators, their field assistant, and students sighted four ornate box turtles along the BIA Highway 27 on the Pine Ridge Reservation. These sightings indicate a potentially robust population there, which creates an ideal opportunity to further study this species in our state. As such, we initially proposed the following objectives:

1. Document the macro- and microhabitat use throughout the active season (May through September);
2. Estimate home range size and describe movements;
3. Document daily and seasonal activity periods;
4. Estimate population size;
5. Estimate the geographic range of ornate box turtles (*Terrapene ornata*) in South Dakota through the use of a species distribution model (MaxEnt);
6. Provide training in ecological field research to OLC students.

In addition to these initial objectives we also report some other biological aspects of the ornate box turtle in South Dakota, such as reproduction and growth.

Habitat

The ornate box turtle (*Terrapene ornata*) prefers open grasslands although it utilizes other adjacent habitat types such as mesic forest and woodlands (Dodd 2001, Redder et al. 2006). Habitat loss mainly through agricultural development and fragmentation has been the greatest threat to the ornate box turtle throughout its range (Bowen et al. 2004, Redder et al. 2006). Most available literature reports on the demography of the box turtle with limited information on quantitative habitat assessments (Redder et al. 2006). The relationship between habitat usage and available space allows the identification of essential habitats for the species (Thomas and Taylor 1990). The objectives of this section were to examine the habitat characteristics of ornate box turtles in South Dakota based on locations of radio-tracked turtles and at a stratified random distance of 10 m from the turtle for: 1) visual obstructions readings (height density of vegetation) base on a modified Robel pole, and 2) canopy cover and physical attributes of key variables. An additional objective was to develop management guidelines, as an understanding of a species' habitat use is crucial in conservation biology.

Home Range

Critical habitats can be identified through the study of an animal's home range and movements. The home range concept has been a source of debate among ecologists (Osborn 2004) although the most common home range definition is the area that an animal or group of animals utilizes to perform daily activities such as feeding, resting, mating, and caring for young (Burt 1943). Shelter, nesting, suitable thermal conditions, and overwintering sites are other animal's requirements also included in their home range area (Harestad and Bunnel 1979, Perry and Theodore 2002, Redder et al. 2006). The size of the home range is believed to reflect behavioral and physiological requirements (e.g., access to mates and food) in relation to their availability in

the environment. In other words, individuals who reside in better quality habitats will have smaller home range sizes (Bernstein et al. 2007). Home range size may vary with sex, age, and season. The goal of this section was to determine home range size and daily movements of ornate box turtles (*Terrapene ornata*) in South Dakota.

Hibernation

Ornate box turtles as well as all reptiles and amphibians are ectothermic organisms, relying on their environment for thermoregulation. Therefore, regardless of presence of favorable foraging habitat in summer, winter survival through hibernation is crucial for the continued existence of the species. Several factors influence hibernation. However, the availability of suitable hibernacula that protects the animals from freezing and predation may be one of the most important factors limiting distribution and abundance of temperate-zone reptiles (Gregory 1982). Box turtles reach the northern extend of their range in South Dakota, and are exposed to very cold winters. For that reason, turtles may spend more than half of their lives underground. Thus, it is important to investigate their behavioral adaptations and the ecology during their hibernation period.

SD Geographic Range Estimation

Prediction and mapping of potential suitable habitat for threatened species is critical for monitoring, conservation of their natural habitats, and management decisions. However, little is known about the distribution of *T. ornata* in South Dakota. Therefore, in this section our objectives were to: 1) estimate suitable habitat distribution for ornate box turtles (*Terrapene ornata*) in South Dakota using historical and current records obtained during this study, and 2) identify the environmental factors associated with box turtle distribution in the state.

Reproduction and Growth

Information regarding various aspects of reproduction and/or growth of this species has been reported for turtles from Colorado, Illinois, Indiana, Kansas, New Mexico, Oklahoma, and Wisconsin (Brumwell 1940, Marr 1944, Legler 1960, Minton 1972, Blair 1976, Bull 1980, Vogt 1980, Caldwell and Collins 1981, Metcalf and Metcalf 1985, Packard et al. 1985, Packard and Packard 1986, Temple 1987, Doroff and Keith 1990, Collins 1993, Nieuwolt-Dacanay 1997, St. Clair 1998, Converse 1999, Hammerson 1999, Converse et al. 2002, Bowen et al. 2004). Nevertheless, no life history information has been reported about ornate box turtles from South Dakota. Such information is critical in developing effective conservation and management plans. However, geographic variation in reproductive and growth traits has been reported in this species. For example, two clutches per year are common in Kansas (Legler 1960), but in Wisconsin and New Mexico not all females reproduce annually (Vogt 1981, Doroff and Keith 1990, Nieuwolt-Dacannay 1997); clutch sizes are smaller in New Mexico than in Kansas or Wisconsin (Legler 1960, Doroff and Keith 1990, Nieuwolt-Dacanay 1997); and males and females mature at an earlier age in Texas than in Kansas (Legler 1960, Blair 1976). Therefore, in designing conservation programs for ornate box turtles, life history traits of populations specific to geographic areas where such programs are to be implemented need to be utilized. It is the purpose of this section to provide such information on South Dakota populations by reporting observations on reproduction and growth of the *T. ornata*.

MATERIALS AND METHODS

Study Area

The 320 ha study area is partially located on of private owned land within the Pine Ridge Reservation, Shannon County in south-central South Dakota. The exact location is omitted due to the risk of illegal collection for the commercial pet trade (see Converse 1999, Redder at al. 2006) but can be made available on request. Well-developed windblown sands that form dunes that extend into the Sand Hills Region of Nebraska are dominant in the study area (USDA-NRCS 2001). Soil features of the greater study area are sandy textured with slopes of 3 to 24 percent and is described as Sands Ecological Site (USDA-NRCS 2001, Figure 1). Soils are well drained with the surface texture 51 to 229 mm (2 - 9 inches) deep. Sub-surface texture is loamy fine sand to fine sand. Some areas have patterns of rill and gully from limited water flow from rain and snow. Elevation of the study area is approximately 860 m (2820 feet). Average annual precipitation is 394 mm (15.52 inches) at Scenic SD over a 31-year period (High Plains Regional Climate Center 2012). Monthly precipitation ranges from a low of 7.37 mm (0.29 inches) in December to a high of 78.74 mm (3.10 inches) in June. Average monthly temperatures varied from -13.3^0 C (8^0 F) in January to a high of 31.9^0 C (89.4^0 F) in July. The study area receives light grazing by livestock during the summer.

Locating Turtles

Dogs specifically trained to hunt box turtles were used during survey periods: the first two weeks of June in 2010 and 2011. Turtle-sniffing dogs hunt and trail turtles by scent and sight, and are extremely efficient in locating and retrieving turtles. They detect more turtles than humans, as turtles frequently are hidden under shrubs or are underground and invisible to human eyes (Converse 1999, Schwartz and Schwartz 1974, Schwartz et al. 1984). Turtles were also found by researchers walking in habitat during their active season, from the end of April through mid-October in both years. A few turtles were captured as they crossed roads, or were brought to us by citizens in response to wanted posters (Appendix 1) placed in key areas, such as stores, schools, and a park visitor center near our study site. These posters were also distributed to individuals encountered in the field and elsewhere.

Collecting Data

Once the turtle was located, standard methods were used for measuring carapace length, plastron length, interabdominal seam length, abdominal lamina growth-ring length (scute growth zones), and body mass (e.g., Legler 1960). An estimate of age was made by counting growth-rings (Legler 1960, Zug 1991, Germano and Bury 1998); however, the plastrons of many turtles were too worn to accurately detect such rings. If some growth-rings could be counted, but not all, the age was indicated as being greater than the number of rings observed. Turtles were assumed to be juveniles if they were smaller than approximately 110 mm (4.33 inches) carapace length, and had no obvious secondary sex characteristics (Legler 1960, Converse 1999). Sex was determined by looking at the colors of the eyes, heads, and antebrachial scales, the plastron shape, and the orientation of the first toe of the hindfoot as described in Legler (1960). Captured turtles were individually marked by filing a unique combination of shell notches, made with a Dremel tool, into marginal scutes of the carapace (Cagle 1939).

In June 2010, 25 ornate box turtles (10 males, 14 females, and 1 juvenile) were fitted with 3.6 gr radio transmitters (model # R1680, Advanced Telemetry System, Isanti, MN) attached between the 3rd and 4th right pleural scutes, with the antenna placed toward the posterior of the specimen (Figure 2). In 2011, same procedure was used on 24 ornate box turtles (12 females, 12 males). The same turtles were also fitted with temperature data loggers (Thermochron iButtons, model # DS1922L-F5, -40°C through +85°C, Embedded Data Systems, Lawrenceburg, KY) attached between the 3rd and 4th left pleural scutes (Figure 2). Data loggers were waterproofed with a double rubber coat of Plasti Dip (Performix by Plasti Dip International, Blaine, MN) and programmed to record temperature at 2 hours intervals (maximum of 96 weeks). Both radio transmitters and data loggers were glued on turtles using quick-dry epoxy, smoothly contoured with plumber's putty, and covered with dirt for camouflage. Transmitters and data loggers plus glue did not exceed 6% of the turtle's body weight. Radio transmitters were replaced after one year (in 2011) due to battery life limitation. At the same time, temperature data loggers were removed from turtles for data collection. They were then reset and replaced. From 28 May through 12 June 2011, we recovered 16 ornate box turtles' temperature data loggers. All temperature data collected from these data loggers were averaged by week for data analyses during the 2010-2011 overwinter period. Sampling weeks ranged from week 20 (starting 22 October 2010) in the Fall 2010 to week 50 (ending 25 May 2011) in the Spring 2011.

Turtles were located using a hand-held receiver (model R410, Advanced Telemetry Systems, Isanti, MN) and a three-pronged antenna (3 Element Folding Yagi antenna, Advanced Telemetry Systems, Isanti, MN). Locations were collected weekly with a minimum interval of three days to assure independent sightings during home range estimations.

Macrohabitat types in our study area were identified based on soil composition and plant diversity. Microhabitat measurements, including visual obstruction readings (VOR) and canopy cover estimates, were made approximately at two-week intervals for each year beginning late spring for a minimum of three females and three males. A stratified random design was utilized, where microhabitat measures were made at each turtle, and at distances of 10 m from each turtle in four cardinal directions, thus establishing five stations (Figure 3). For VORs we used a modified Robel pole with alternating 1.27 cm white and gray bands (Figure 4). Bands were numbered beginning with 0 (white band) at the bottom of the pole and placed at the soil surface (Uresk and Benzon 2007, Uresk and Mergen 2012). Readings were taken at a distance of 4 m from the pole in four cardinal directions at each of the five stations. For each VOR, the lowest visible band was recorded. Canopy cover (Daubenmire 1959) were estimated for:

- Total grass
- Total forbs
- Total shrubs
- Total vegetation
- Sand sagebrush (*Artemisia filifolia*)
- Fringed sagewort (*Artemisia frigida*)
- Cactus (*Opuntia* sp.)
- Yucca (*Yucca* sp.)
- Litter
- Bare ground

Canopy cover estimates were made at the location of the turtle and also at four cardinal directions in 10 m stratified random distance from the turtle (Figures 3 and 5). For analyses, all data were averaged as two site means, one for the turtle location and one for the 10 m distance from the turtle. We reduced data collected from each turtle location and at a 10 m distance from the turtle to 1 mean for each attribute of the Robel pole and canopy cover. A total of 11 habitat variables were evaluated.

Hibernation site slope was measured in degrees using a clinometer, and aspect (direction the hibernation site faces) was estimated in degrees utilizing a compass. Direct body temperature and hibernation depth were not measured, as we did not disturb turtles during the hibernation period, which started on 31 October 2010. Instead, both measurements were inferred using temperature data loggers information. To estimate the turtle's hibernation depth, duration, and temperature variation, on 30 October 2010 we buried seven temperature data loggers (Thermochron iButtons, model # DS1922L-F5, -40°C thru +85°C, Embedded Data Systems, Lawrenceburg, KY) spaced and labeled at 0 (surface), 15, 30, 50, 75, 105, and 140 cm (soil depth) attached to a wooden dowel (pole). As control, we also buried extra individual temperature data loggers at 15, 30 and 50 cm below the surface, with a very thin string attached to them and tied to a permanent structure above ground, such as sand sagebrush. Temperature data loggers were programmed to record temperatures at 2 hours intervals, were buried perpendicular into the slope of the ground, and were placed within one meter of a cluster of five hibernating turtles. Data collected included GPS coordinates, elevation, aspect, time of burying, and date. Each site was marked with waterproof flags. All the buried temperature data loggers were removed from the ground on 28 May 2011. Temperatures from each buried individual data loggers were averaged by week. To estimate turtles body temperature we compare the data loggers temperature information from the turtle's carapace and with the data loggers temperature information from the pole. Legler (1960) mentioned that when turtles are underground, body temperatures approximate soil temperatures. In addition, past studies demonstrated no differences between turtle cloacal temperature, ambient temperature, and carapace data logger temperature (Peterson 1987, Bernstein and Black 2005). Sampling weeks ranged from week 20 (starting 22 October 2010) in the Fall 2010 to week 50 (ending 25 May 2011) in the Spring 2011. Hibernation data from 2011-2012 were not analyzed yet, as temperature data loggers were only retrieved this past spring-summer.

On 7 June 2010, 13 turtles were collected and x-rayed to detect oviducal eggs following the method described by Gibbons and Greene (1979). These females were then released at their site of capture. Egg-laying dates of a subsample of those x-rayed were determined by weighing gravid females daily to detect weight loss approximately equivalent to 10 g per egg.

<u>*Data Analysis*</u>

Microhabitat. Examination of data for homogeneous variances was evaluated by using Levene's test. VOR and canopy cover data were analyzed at turtle locations and at 10 m with SPSS (2003). One-way analyses of variance were used to test attributes for both years. The Least Significant difference (LSD) was used for multiple comparisons. We used a paired T-test to compare differences between turtle and 10 m distances by sample period for each attribute and a two sample T-test for comparisons between years. Statistical inferences were made at $p = 0.10$ for all comparisons unless actual p-values are presented.

Home range. To facilitate comparisons with previous box turtle home range studies we applied two home range indices: the Minimum Convex Polygons (MCP) (Mohr 1974) and the local convex hull (LoCoH) nonparametric Kernel method with K values of 20, 15, 10, and 5 (Getz and Wilmers 2004, Getz et al. 2007). MCP uses a 100% confidence interval in which the furthest outside points are connected into a polygon, and the polygon area makes up the home range. Although this method incorporates all points into analysis, it does not construct utilization distribution (Burgman and Fox 2003). In LoCoH a convex hull is constructed for each point and the associated k-1 nearest neighbors. For each k value the convex hulls can be united to form a home range. K values of 5, 10, 15, and 20 were used in determining utilized distribution. Unlike MCP, LoCoH can construct utilization distribution and capture hard boundaries while incorporating all points into analysis. MCP and LoCoH individual annual home ranges were calculated using ArcGIS 9.3 (ESRI 2008) with extension tools, the home range extension tool (HRT, version 1.1, Rodgers et al. 2007, http://flash.lakeheadu.ca/~arodgers/hre/) and LoCoH (Local Convex Hull) Homerange Generator for ArcGIS 9 (http://nature.berkeley.edu/~ajlyons/locoh/arcgis9/index.html), respectively. Home range sizes were analyzed separately for males and females for both years. No seasonal comparison and individual daily distances calculations were possible due to the small sample size per period year. Statistical tests for home range were calculated using IBM SPSS (2011). Student's T-Test was used to compare home range size between gender (females and males) and between both years 2010 and 2011. MCP and LoCoH home ranges were compared with a paired *t*-test. Statistical inferences were determined at $p = 0.10$ for all comparisons unless actual *p*-values are presented.

Hibernation. Quadratic regression (SPSS 2003) was used to quantify the relationship between mean turtle temperature and weeks. Cluster analyses with ISODATA (Ball and Hall 1967, del Morell 1975) and KMEANS (SPSS 2003) was used to establish weekly and soil depth groupings with temperature data loggers on dowels (poles) with turtle temperatures. Discriminant analyses determined the significance between and among groupings. EXPLORE (SPSS 2003) provided basic means and variance terms. Statistical inferences were determined at $p = 0.10$ for all comparisons unless actual *p*-values are presented.

SD Geographic Range Estimation. Due to the small number of box turtle distribution records prior to this study, the species distribution model selected was the maximum entropy distribution or MaxEnt (Phillips et al. 2006), as it may remain effective despite small sample size. The model establishes relationships between occurrences of species (presence-only) and biophysical and environmental conditions in the study area. Historical and current box turtle occurrence records, Geographic Information System (GIS) bioclimatic layers, and a soil data layer that measured the percent composition of sand for all soils throughout the state of South Dakota were used in the analysis. The soil data layer was created by calculating the percent sand composition for each soil within each county separately, then adding this attribute to their spatial data table, and merging all South Dakota counties together to make one single layer. The soil data layer was then changed to a raster and ASCII and used into MaxEnt modeling. Initially, we considered 21 environmental variables, 19 bioclimatic variables from WorldClim dataset (Hijmans et al. 2005; http://www.worldclim.org/bioclim.htm), as potential predictors of *T. ornata* habitat distribution in South Dakota (Table 1). Analyses were performed using ARC GIS

version 9.2 (ESRI, Redlands, CA) and the freely available MaxEnt software, version 3.3 (http://www.cs.princeton.edu/~schapire/maxent/). After the first analysis, some of the bioclimatic variables (Bio4, Bio5, Bio7, Bio10, Bio12, and Bio13) did not contribute to box turtle distribution and consequently were removed from the final analysis.

Reproduction and Growth. The lengths of growth-rings on the abdominal lamina were used to estimate the approximate lengths of the plastron at the time each was formed. This process is based on a formula derived by Legler (1960), and can estimate plastron length at given ages. The formula is $AB/PL = AB^1/X$, where AB is the abdominal length, PL the plastron length, AB^1 the length of any growth-ring, and X the plastron length at the time the growth-ring AB^1 was formed. Data were examined for homogeneous variances using Levene's test. We used Student's t-tests to compare mean estimated plastron sizes between sexes for each age class, as well as to compare carapace and plastron lengths between sexes of adults encountered in the field. Statistical inferences were made at $p = 0.05$ for all comparisons unless otherwise stated in the text. We performed statistical analyses with IBM SPSS (2011).

RESULTS

A total of 170 living turtles (67 males, 69 females, and 34 juveniles) and nine dead turtles were found during the course of the two-year study. While most of these specimens (87%) were found at our study area, a few (13%) were located in scattered locations in Shannon, Jackson, Pennington, and Todd Counties.

Habitat

Macrohabitat

Two macrohabitat types were identified within the study area, which we termed uplands and lowlands, but described in the literature as Sands and Clayey, respectively (USDA-NRCS 2001, 2008). Upland habitat was characterized by deep sandy soils and the lowland habitat by clay soils. Transition zones also existed between the two macrohabitat types. Dominant grasses of the Sands habitat include, prairie sandreed (*Calamovilfa longifolia*), needle and thread (*Hesperostipa* sp), blue grama (*Bouteloua gracilis*), and hairy grama (*Bouteloua hirsuta*). Common forbs included cuman ragweed (*Ambrosia psilostachya*) and slimflower scurfpea (*Psoralidium tenuiflorum*). The common and dominant shrub was the aromatic sand sagebrush (*Artemisia filifolia*) a freely branching, rounded crown, growing up to 66 cm tall, and generally considered an indicator of sandy soil (Johnson and Larson 1999). Plants on the Clayey site included common grasses, western wheatgrass (*Pascopyrum smithii*), green needlegrass (*Nassella viridula*), and blue grama (*Bouteloua gracilis*). Common forbs were scarlet globemallow (*Sphaeralcea coccinea*), and cuman ragweed (*Ambrosia psilostachya*). Shrubs present on the area were fringed sagewort (*Artemisia frigida*), and cactus (*Opuntia* sp).

Microhabitat

Visual Obstruction Readings

The mean visual obstruction reading (VOR) of vegetation over the two-year period was greater ($p < 0.10$) at the turtle sites than at a 10 m distance by 6.8 bands (8.64 cm) in 2010 and 5 bands (6.35 cm) in 2011 (Table 2). VOR at the turtle locations showed no differences between years with mean bands of 17.7 (22.5 cm) and 17.2 (21.8 cm) for 2010 and 2011, respectively.

VOR varied over sampling periods ($p = 0.10$) for turtle sites and at distances of 10 m during 2010. Turtle locations had a VOR range from 0 to 48 bands with a mean of 17.7 ± 0.8 SE, while at 10 m, the range was 1 to 26 bands and a mean of 10.9 ± 0.4 SE. The ornate box turtle clearly selected for greater VORs throughout the summer sampling periods ($p < 0.10$) than what was available at a 10 m distance (Figure 6). Only two sampling periods did not show significant differences.

VORs were greater at turtle sites than at 10 m away ($p < 0.10$) in 2011, 17.2 ± 0.9 and 12.2 ± 0.5 bands, respectively. Turtle locations had a range of VORs from 1 to 50 bands while at a 10 m distance, the range was from 3 to 26 bands with less variability. Ornate box turtles clearly selected for greater VOR's throughout the summer (Figure 7). No significant differences were observed between turtle location and available habitat at 10m.

Canopy Cover: Ornate Box Turtle

Canopy cover for ornate box turtles varied between years for several variables (Table 3). They showed a greater increased preference of cover ($p < 0.10$) for total grass (18%), total forbs (17%), total vegetation (8%) and litter (24%). However, turtles showed a significant decrease in 2011 compared to 2010 for total shrubs (13%) and sand sagebrush (15%). Overall, sand sagebrush is highly preferred by turtles. Ornate box turtles showed no differences ($p < 0.10$) between years but were consistent in their use of bare ground, 15.5 ± 1.9% and 12.5 ± 2.1% SE in 2010 and 2011, respectively. Other variables, such as fringed sagewort, cactus, and Yucca were limited for evaluation.

Canopy Cover: Ornate Box Turtles vs. Available Habitat

Canopy cover for ornate box turtles combined over years (Table 4), showed an extremely high preference over existing habitat ($p < 0.10$) for total shrubs (32%), sand sagebrush (30%), and total vegetation (7%). Total grass (13%), total forbs (4%), and bare ground (5%) were not preferred ($p < 0.10$) by turtles but avoided compared to availability.

Turtles in 2010 selected for a lower mean grass cover ($p < 0.10$) throughout the sampling season than available habitat on the grasslands (Figure 8). Grass canopy cover selected by turtles was 15% lower than available habitat. Similar results were observed for turtles in 2011 with grass canopy being 10% lower than existing habitat (Figure 9).

Sand sagebrush canopy cover was highly preferred by box turtles ($p < 0.10$) over existing cover both years. A discriminating mean difference between turtle selection and availably of sand sagebrush was 36 ± 3% and 20 ± 3% SE for 2010 and 2011, respectively. Figure 10 clearly shows the selectivity pattern of turtles for sand sagebrush throughout the sampling season

compared to existing sand sagebrush. Turtles showed similar preference in 2011 (Figure 11). The two year average of sand sagebrush canopy cover selected by ornate box turtles was 37.5 ± 2.3% SE.

Ornate box turtles selected sites with a lower forb cover for both years ($p < 0.10$). Forb cover ranged from 13 to 32% during the two-year study. Litter cover was not an important variable for box turtles during the sampling season in 2010, 50% cover compared to availability with 47% cover ($p = 0.146$). However, in 2011, litter was selected by turtles at 74% cover compared to existing liter at 67% cover ($p = 0.013$). Other plant variables limited in the study with no biological meaning were cactus and silver sagebrush.

Ornate box turtles did not show differences in use of bare ground between years, however when compared to availability, they selected habitat with less bare ground ($p < 0.10$) both years. Turtle avoidance varied throughout 2010 as related to availability (Figure 12). However, bare ground used by turtles throughout the season is presented in Figure 13. The mean bare ground used by the turtles was 15% and 13% during 2010 and 2011, respectively.

Home Range

Annual home ranges were calculated for 21 adult box turtles (11 females and 10 males) in 2010 and for 24 adults (12 females, 12 males) in 2011 using the Minimum Convex Polygons (MCP) and the local convex hull (LoCoH) nonparametric Kernel method with K values of 20, 15, 10, and 5. We had 8 females and 6 males both years. Turtles included in the analysis had an average of 20 locations per year during the active season (late April to early November), with a minimum of 13 and a maximum of 25 recaptures. Only one juvenile (BT#20) was radio tracked showing a considerable difference in home range size between years. In 2010, with 16 locations, its home range was 3.01 ha and in 2011, with 19 locations, 25.79 ha.

Home range size calculated as MCP varied greatly among adult box turtles, raging from 0.35 - 89 ha (Table 5). However, the home range means were not statistically significant different either between sexes ($t(19) = -1.37$, $p = 0.187$ in 2010; $t(22) = 0.987$, $p = 0.334$ in 2011) or between years ($t(9.13) = 1.61$, $p = 0.141$ for males; $t(21) = 0.069$, $p = 0.946$ for females). For males and females combined, the 2010 minimum home range was 0.58 ha and the maximum 89 ha; the mean was 10.50 ha and the median 4.10 ha. In 2011 the minimum home range was 0.35 ha and the maximum 25.50 ha; the mean was 5.13 ha and the median was 2.97 ha.

Although not statistically different we observed that males had particularly bigger mean home ranges than females in 2010, but the opposite was observed in 2011 (Table 5). In order to visually explore these observations, we box plotted the home range medians and detected three extreme outliers (individuals that have values more than three times the height of the boxes) both females and males in both years. As observed on Table 5, the means were greater than the medians (particularly for 2010 males), possibly caused by these outliers (Figure 14). To generate more refined home range estimations, we removed the three extreme outliers that could possibly be transient individuals, and recalculated the home ranges. The new results (Table 6 and Figure 15) demonstrated that males have statistically significant bigger home ranges than females in

2010 (t (8.5) = -1.99, $p = 0.080$) but no statistically significant differences where observed in 2011 (t (21) = 1.80, $p = 0.859$). Home ranges overlapped both between and within sexes.

Home range size calculated as LoCoH with K values of 20, 15, 10, and 5 also varied greatly among adult box turtles, raging from 0.35-89 ha (Table 7). However, the home range means were not statistically significant different either between sexes or between years (Table 8).

The MCP and LoCoH-20 are similar home range calculations, as we had an average of 20 locations per turtle in both years. Therefore, the home range minimum and maximum values (Tables 5 and 7), as wells as the mean home range values are analogous (Tables 5 and 8). Conversely, this statement is not valid for LoCoH-25, LoCoH-10, and LoCoH-5.

Females and males MCP and LoCoH home ranges were compared with a Paired *t*-test. For females both years combined (n=23), MCP was not statistically significant different than LoCoH-20 ($p < 0.05$), as expected and explained above, however MCP was statistically significant larger than LoCoH with K values of 15, 10, and 5 ($p < 0.05$). LoCoH-20 was statistically significant larger than LoCoH-15, LoCoH-10, and LoCoH-5 ($p < 0.05$). For males both years combined (n=22), MCP was not statistically significant different than LoCoH with K values 20 (as expected and explained above), 15 and 10, although it was statistically significant larger than LoCoH-5 ($p = 0.02$). LoCoH-20 was statistically significant larger than LoCoH-15, LoCoH-10, and LoCoH-5 ($p < 0.06$).

Hibernation

Most of the radio-tracked turtles burrowed underground for the winter by the middle to the end of October in both years. Mean hibernation site slope for the combined winters of 2010 - 2011 and 2011 - 2012 was $19.5^0 \pm 4.2$ SD (n = 42), with measures ranging from 8^0 - 29^0. Mean aspect for the combined winters was $188^0 \pm 48.3$ SD (n = 42), or roughly south, with measures ranging from 80^0 - 270^0, or roughly from east to west. Ornate box turtle overwinter hibernation temperatures ranged from 34^0 F to 69^0 F (1.1^0 C to 20.5^0 C) from 22 October 2010 to 25 May 2011 (week 20 through week 50 – Table 9). Soil temperatures in different depths (0 - 140 cm) varied from 22^0 F to 64^0 F (-5.5^0 C to 17.8^0 C) for this same period. The quadratic equation that describes the relationship of mean temperatures of turtles with week is expressed as Y (Turtle Soil Degrees F) = 179.09 -8.19 week + 0.12 week^2. $R^2 = 0.91$, SE = 2.4 (Figure 16). The equation and constants are significant at $p = 0.001$.

The maximum depth of hibernation estimated for box turtles in South Dakota was 105 cm (± 7 cm) based on our quadratic equation and cluster analyses of temperature with soil depth. The hibernation depth varied with soil temperature, and turtles moved deeper as the winter progressed and soil temperature decreased (Figure 16). Most of the turtles did not experience freezing temperatures. In three events two turtles were exposed to freezing temperatures, but only for a short periods of time. On 2 April 2011, one female (BT#9) was exposed to freezing temperatures (0^0 C) for less than 2 hours. Another female (BT#7) was exposed to freezing temperatures (0^0 C) for less than 6 hours in two events: (1) on 16 April 2011 and (2) on 20 April 2011.

Based on the quadratic equation (Figure 16) we estimate that box turtles emerged from hibernation around early May 2011 (week 48) with average surface soil temperature at 66^0 F (19^0 C) (min. 63^0 F and max. 69^0 F; SD = 1.8). Precise dates are not available for this study; however, based on our analysis we estimated that the mean hibernation period is in between 161 to 182 days.

Comparing overwinter hibernation sites between 2010-2011 and 2011-2012, we observed site fidelity, where 75% of the turtles (n = 9) were hibernating at or near (within 2.5 m) the previous year's location. Twenty-five percent of the turtles (n = 3) hibernated 5, 10, and 116.5 m away from their previous year sites.

In 2010-2011 and 2011-2012 winters, turtles were observed to hibernate in clusters (communal sites) of an average of 4.5 turtles per cluster (range 2 - 6 turtles). Within the cluster, turtles were buried within a couple of meters to no more than 15 m apart from each other. Clusters were an average of 567 m apart from each other (range 350 - 883 m) with the exact same location in both winters.

SD Geographic Range Estimation

A total of 170 living and nine dead box turtles were recorded during the course of the two-year study. While most of these specimens (87%) were found at our study area, a few (13%) were located in scattered locations in Shannon, Jackson, Bennett, Pennington, and Todd Counties. An additional 19 historical records were available from Shannon, Todd, Bennett, Jackson, and Tripp Counties. Based on these records, both this study and historical, and local environmental variables, the MaxEnt model predicted potential suitable habitats for *T. ornata* in South Dakota. Figure 17 represents the MaxEnt species distribution model for ornate box turtles, where the highest probability species presence are represented in warmer colors of orange, yellow, green, and light blue, predicting the most suitable habitats as the south west-central portion of the state. Highest probability of box turtle presence is Shannon (orange and yellow), Bennet (yellow and green), and Todd (yellow and green) counties, followed by Jackson (a bit of yellow, green, and light blue), Pennington (green and light blue), Mellette (green and light blue), Tripp (green and light blue), and Gregory (green and light blue). The lowest probability is on the west corner of the state in Fall River (light and dark blue) and on the central part, Haakon (light and dark blue). The MaxEnt model's internal jackknife test of variable importance showed that Isothermality (BIO3) variable was the most important predictor (41.8%) of *T. ornata* habitat distribution. This variable was calculated as follow: (BIO2/BIO7) (* 100), where BIO2 is defined as "mean diurnal range (mean of monthly (max temp minus min temp)) and BIO7 is defined as temperature annual range (the maximum temperature of warmest month minus the minimum temperature of coldest month) (Table 1).

Reproduction and Growth

Of the live turtles, 67 were males, 69 females, and 34 juveniles, creating a ratio of 0.97:1:0.49 for males, females and juveniles, respectively. Sex could not be determined on the dead turtles, as they were either too mutilated, were juveniles, or were represented by only bone fragments.

Copulation was observed on six occasions, one in every month from April through September, all during morning hours, except one in early afternoon (Table 10). One male was observed

copulating with two different females, but all other animals were observed copulating only once. Minimum and maximum carapace lengths of males observed copulating were 111.4 and 120.6 mm, respectively. Those measures for females were 114.4 and 125.3 mm. Minimum and maximum plastron lengths of males were 110.7 and 126.5 mm, respectively, while those of females were 116.8 and 129.1 mm. Two females, one observed copulating on 26 July 2010 and one on 9 August 2010, had been x-rayed earlier in the year (June), and contained oviducal eggs. With one exception, the plastrons of all animals observed copulating were too smooth to accurately count growth-rings, but one male had more than eight, and one female more than 12. The exception was a male with 15 growth-rings.

Eleven adult females were x-rayed on 7 June 2010, and seven contained eggs (64%) (Tables 11 and 12). Additionally, two juveniles of unknown sex were x-rayed, and neither contained eggs. Both were eight years old, and had carapace lengths of 104.6 and 105.9 mm. Their plastron lengths were 110.5 and 106.3 mm. Clutch sizes ranged from 2 - 5, with an average of 4.3 (Table 11). The three females weighed daily to detect egg deposition laid their eggs during the night of 15 – 16 June, presumably in upland habitat. No nests were found in spite of intensive search efforts, but the female pre- and post- weight loss locations were all within a few meters, and the closest lowland habitat was approximately 0.5 km from the site. Based on weight loss of females, average egg mass was 12.0 g. Clutches averaged 13.6% of female mass. With two exceptions, the plastrons of all females x-rayed with eggs were too smooth to accurately count growth rings. The two exceptions had 9 and 14 growth rings. Three females had more than 12 growth rings, one had more than 13, and no growth-rings could be counted on the remaining animal, as the plastron was too worn. Minimum and maximum carapace lengths for females with eggs were 113.2 and 125.3 mm, respectively. The smallest clutch size (2) was from the smallest female.

The four x-rayed females that contained no eggs ranged in carapace length from 112.1 to 124.3 mm (Table 12). One of these females had nine growth-rings, and one had more than 19, and two had plastrons too worn to count rings.

Complete sets of growth-rings were visible enough to count on 28 (16%) turtles, with ages ranging from 1 - 15 years. While 15 was the maximum number of growth-rings we counted in the above sample, we estimate that many turtles older and younger than 15 had growth rings too worn to count. The first growth-ring is formed during the first winter, which begins a month or so after hatching, and turtles may or may not grow during this period (Legler 1960). Based on growth-ring size data, mean male, female and combined (male, female, juvenile) plastron lengths at their first winter were 33.9, 29.6, and 31.7 mm, respectively (Table 13). Although males were larger than females until age eight, they were not significantly so ($p > 0.05$). At this age female size surpassed that of males until age 11. However, beyond age nine sample sizes were too small to make reliable growth assessments. Samples were small, as plastrons became worn in older animals, making age estimation difficult.

Based on growth-ring measurement data, the annual percent increase in plastron length was greatest in young animals, and gradually decreased as animals aged (Table 14). Animals continued to grow through age nine, and may have continued to do so at later ages, but, as noted above, samples were too small to make reliable growth assessments for older animals.

When comparing carapace lengths of all adults measured in the field, females were significantly larger than males, t (124) = 3.31, p = 0.001, with mean carapace lengths 117.8 and 114.5 mm, respectively (Table 15). Minimum and maximum carapace length of males was 103.3 and 137.5 mm, respectively, while those of females were 110.2 and 137.0 mm. When comparing plastron lengths, females were significantly larger than males, t (107.7) = 5.87, $p < 0.001$, with mean plastron lengths 123.9 and 118.1 mm, respectively (Table 15). Minimum and maximum plastron lengths of males were 104.0 and 134.0 mm, respectively, while those of females were 113.4 and 135.6 mm.

Of known age turtles, the youngest males exhibiting secondary sex characteristics were 9 years old, and had mean carapace and plastron lengths of 110.8 and 111.7 mm, respectively (Table 16). Of these, the smallest individual had carapace and plastron lengths of 107.9 and 110.4 mm, respectively. The youngest adult females had mean carapace and plastron lengths of 112.6 and 116.7 mm, respectively, and were 9 years old. The smallest of these two females had carapace and plastron lengths of 112.1 and 115.5 mm, respectively.

DISCUSSION

Habitat

Macrohabitat

The ornate box turtles in this study are located in a choppy sands and sands ecological sites that extends from Nebraska into South Dakota (USDA-NRCS 2001, Uresk 2012). These turtles generally prefer open grasslands and shrubby sites within the sand hills (Converse and Savidge 2003). The habitat at this study area in the sands ecological site is considered excellent for the ornate box turtle.

Microhabitat

Historically, our study area has been under heavy grazing by livestock as indicated by the dominant plant species. Sand sagebrush and cuman ragweed are common and expand under heavy grazing (USDA-NRCS 2001). Prairie sandreed with needle and thread, blue grama and hairy grama are common on the study area. Other plants, sand bluestem, little bluestem are rare on the study area but are generally considered abundant for this ecological site (USDA-NRCS 2001). Currently, the area is receiving limited livestock grazing. Based on the average of two years of VOR data collected for available ornate box turtle habitat, the area is classified as light to no grazing (Uresk 2012).

Livestock grazing has been used as a tool for wildlife habitat management for many years (Severson 1990). Ornate box turtle habitat can be manipulated with a high degree of control by livestock grazing. Fire, mowing, and spraying are additional options but are very expensive and may not be practical. Sand sagebrush and cuman ragweed are common on the area, and probably

required heavy grazing for several years to promote their prevalence (USDA-NRCS 2001). Once established, management for this habitat (sand sagebrush, cuman ragweed) at light to no grazing will maintain ornate box turtle habitat in South Dakota. Converse and Savidge (2003) in the Nebraska Sand Hills reported that ornate box turtles also had a strong relationship to the amount of shrub cover used. Ornate box turtles were common on our study area and highly preferred microhabitats of sand sagebrush for canopy cover.

Ornate box turtles highly selected for greater VORs during both years compared to the availability. Preferences by the turtles were generally for shrubs and sand sagebrush, with less preference of grass and forbs in more open areas. Microsite selections for VOR were similar for both years at 17.5 ± 0.6 bands (1.27 cm bands). This extremely high preference for sand sagebrush would indicate that if habitat change did occur with the shrub component, negative impacts with these turtles are expected.

Converse and Savidge (2003) reported inactive turtles (not moving) used areas of more shrub and litter cover but these observations were not consistent over years on the Nebraska Sandhills. In New Mexico, ornate box turtles were observed preferring shrubs (Converse and Savidge 2003). Shrubs are an important component of ornate box turtle microhabitat selection, as they facilitate thermoregulation by providing cool areas during the summer and favorable hibernation sites during the winter (Dodd 2001, Reeder et al. 2006).

Microhabitat selection of canopy cover by ornate box turtles only varied between years. Turtles preferred a greater amount of cover in 2011 for grass, forbs, total vegetation and litter. The decrease in shrub and sand sagebrush cover preference was augmented by these habitat variables. However, use of bare ground by turtles remained relatively low and constant both years. This indicates that ornate box turtles may adapt to some environmental changes to meet microhabitat requirements.

In comparing turtle microhabitat locations with habitat availability for canopy cover, sand sagebrush was highly preferred both years. Grass and forb cover was used less by the ornate box turtles both years than available in the habitat. Turtles were seeking areas with a lower grass and forb cover on this study area both years. Litter was not really an important habitat component for the turtle but showed a 7% increase in use over habitat availability.

In summary, the most important microhabitat variable for the ornate box turtle was sand sagebrush, with an average canopy cover of 38% and requiring 17 - 18 bands (22 - 23 cm) of visual obstruction readings of the vegetation, primarily sand sagebrush. Bare ground was used less by the turtles than availability by 5 - 7%. Greater amount of litter use by turtles was only evident during the second year of the study.

Home range

Comparing home range (HR) results with other box turtles populations is not an easy task. Home range studies applied different computational methods, results were reported in many different ways, and sample sizes and field methods were not comparable. Dodd (2001) presents a table contrasting home range size of box turtles. In Texas (Blair 1976), Wisconsin (n = 47, two years study, HR observed for males was 8.2 ha and 3.4 ha and for females 12 ha and 6.9 ha)

(Doroff and Keith 1990), and Nebraska (n = 22, two years study) (Holly 1995) no differences where found between sexes whereas in Kansas (Metcalf and Metcalf 1970) females have bigger home range than males. In our study no differences in home range size were found between sexes or between years with the total sample. When extreme outliers were removes, males had bigger home ranges than females in 2010 but no differences where observed in 2011. The extreme outliers may be transient turtles or turtles that went to explore other areas but still have the core area of their home ranges in our study site. No territoriality was observed as home ranges overlapped both between and within sexes. Our home range size calculations (0.35-89 ha and 0.35 – 22.2 ha without the extreme outliers) are congruent to what has been found in Wisconsin, 0.2 - 58.1 ha (Doroff and Keith 1990). Several factors such as habitat quality and diversity, and individual preferences affect home range. Additionally, home range size is expected to increase with increasing latitude (Harestad and Bunnell 1979). Northern habitats can be less productive than the southern ones thus generating larger home ranges, as turtles may need to travel longer distances searching for food, appropriate nest sites, and shelter areas. However, this was not observed in our study, meaning that our study site provided sufficient foraging, nest sites, and protection for box turtles.

The fact that MCP did not differ from LoCoH-20 for both sexes was expected as explained in the results section. On the other hand, the differences observed between the MCP and the LoCoH with K values of 15 and 10 by gender are especially interesting. For males, MCP was not different than their LoCoH-15 and LoCoH-10. Conversely, MCP for females was larger than their LoCoH-15 and LoCoH-10. Therefore, we could interpret these results as both sexes having similar home range size although using the area differently. Males had a larger utilization distribution area than females; in other words, females had a more centralized utilization distribution area, or smaller core areas than males. The cause/s of the differences in utilization distribution between sexes is/are not clear, although nesting behavior may be an important factor resulting in a smaller utilization distribution area for females. To date, no comparisons with other studies are possible, as the local convex hull (LoCoH) nonparametric Kernel method was not applied to other box turtle populations.

Hibernation

In our study, a mean direction of most of the hibernation sites was a south-facing slope and as highlighted by Dodd (2001), in northern habitats south- or west-facing slopes may be where winter temperatures presumably are warmer than in other locations. All of the hibernation sites were located on upland habitat, characterized by loose sandy soils, where *T. ornata* burrow into the sand, generally under or near sand sagebrush but open areas were also utilized. Occasionally mammal burrows (e.g. badger, skunk, fox, coyote, raccoon, or kangaroo rat) could be used as an underground retreat as observed a couple of times in this study and on Nebraska prairies (Trail 1995). For most northern box turtle populations, sandy soils are extremely important for winter survival, as turtles can burrow below the frost line (Doroff and Keith 1990, Dodd 2001). Dodd (2011) stated, "turtles would not survive the winters if only shallower soils were available, despite the presence of favorable foraging habitat in summer." In southern states such as New Mexico and Arizona, due to the compaction of the dry soil or as an opportunistic behavior, the desert box turtle, *T. o. luteola*, used mainly burrows of kangaroo rats (*Dipodomys spectabilis*) to hibernate (Nieuwolt 1996, Plummer 2004). If sandy habitats were available, southern turtle populations (e.g. Oklahoma) would also favor this type of habitat (Carpenter 1957). Deep sandy

soil is one of the limiting factors that allow ornate box turtles to maintain or even extend their range to the north (Dodd 2011).

Turtles in our study were not excavated to directly verify hibernation depth, as reported in other studies (e.g. Doroff and Keith 1990, Converse et al. 2002). Instead, we used our temperature data logger data to estimate the maximum depth of hibernation for ornate box turtles in South Dakota as 105 cm (± 7 cm). In a 10-year study of a small and disjunct population of box turtles in south-central Wisconsin (comparable latitude in relation to our study site), burrow depths were in between 50 - 180 cm (Doroff and Keith 1990). In Nebraska, the mean depth was 54 cm (Conserve et at. 2002), and in Iowa, the minimum depth was in between 5 - 75cm (Bernstein and Black 2005). Figure 16 shows that turtles in our study moved vertically along a thermal gradient below the surface. Legler (1960) stated that hibernating turtles would gradually deepen their burrows during the winter. Our results are consistent with other studies related to the genus *Terrapene* (e.g. Norris and Zweifel 1950, Carpenter 1957, Legler 1960, Dolbeer 1971).

The quadratic equation model may be used for predicting turtle temperatures from week 20 (October 22nd) for future years through week 50 (May 19th). However, this model is base only on one year (2010-2011) of data. Assumptions of the model are that the turtles will behave similarly throughout the 30 weeks from fall through spring. An example for use of the model Y (Turtle Soil Degrees F) = 179.09 - 8.19 week + 0.12 week^2 follows. We will assume that our interest is for week 35.

Turtle Soil Degrees = 179.09 - 8.19 (35th week) + 0.12 (35 week)^2.
Turtle Soil Degrees = 179.09 - 286.7 + 0.12 (1225).
Turtle Soil Degrees = 179.09 - 286.7 + 147.
Turtle Soil Degrees = 39.4^0 F.

Therefore in the 35 week we can expect the turtle temperature to be at 39.4^0 F.

Precise hibernation dates were not determined, but the hibernation period for ornate box turtles in South Dakota, ranging from 161-182 days, were shorter than in Wisconsin (209 - 216 days), where turtles hibernate from early September through the first week of October and emerged from hibernacula beginning the second week of April (Doroff and Keith 1990). In Nebraska, no precise hibernation dates were available, but Converse et al. (2002) mentioned that more likely turtles began hibernation sometime after 1 October and turtles were still underground after the beginning of April. In Iowa turtles hibernate from late September to April, ranging from 166 - 201 days (Bernstein and Black 2005). In Kansas, even though precise dates were not available, the hibernation period observed was approximately 152 - 171 days (Legler 1960, Metcald and Metcalf 1970). The shortest hibernation period for ornate box turtles was observed in Texas, as turtles were not active only in January and February and few turtles were active during November and December (Blair 1976). Interestingly our results were more similar to the desert box turtle (*T. o. luteola*) in central New Mexico, that hibernate by the middle to the end of October up to May (Nieuwolt 1996). This may be just an equivalence in hibernation duration as different environmental factors may be influencing hibernation in southern populations as summer monsoon rains are an important predictor for turtles activity (Nieuwolt 1996, Plummer 2003).

No clear factors influencing box turtle emergence from hibernation are defined, but are likely related to soil temperature and perhaps moisture (Dodd 2011). In eastern Missouri, Grobman (1990) stated that soil moisture has not been observed to be a factor affecting the time of emergence for turtles. Conversely, moisture appears to be an important emergence-controlling factor for *T. ornata* in Kansas (Fitch 1959, Metcalf and Metcalf 1970).

Dodd (2001) stated that box turtles generally overwinter safely, but may be killed during severe winters by extreme cold temperatures. If turtles have a premature emergence from hibernation, and experience freezing temperatures, sometimes they return underground, or simply perish. In Missouri, during two different years and with no explanations, turtles had early emergence around mid-March with minimum temperature at 20 cm below the surface around 2.2^0 C, and they didn't survive (Grobman 1990).

During the winter, besides protection from the cold, turtles also need protection from predators, as they are more vulnerable due to the dormancy. Metcalf and Metcalf (1979) attributed deaths of 163 turtles to exhumation by predators. In our study are, turtle's shells were not found after consecutive months of intensive fieldwork following hibernation emergence.

Utilization of the same hibernation site in consecutive years was also observed in 57% of the turtles monitored in Wisconsin (Doroff and Keith 1990). In Kansas eight of 14 turtles hibernated within 1 m of their previous hibernation sites, and five of them were within 0.5 m of each other (Metcalf and Metcalf 1979), as was the case in one of the three monitored turtles in Nebraska (Conserve et al. 2002). In Oklahoma, six three-toed box turtles (*Terrapene carolina triunguis*) were found hibernating within the same hole and 53% of the turtles were within 0.3 to 0.6 m of one another (Carpenter 1957).

In our study site, most of the ornate box turtles hibernated in clusters. A similar pattern was observed in Wisconsin, where five *T. ornata* hibernated within 0.5 m from each other (Doroff and Keith 1990), and in Kansas where three areas of overwintering concentration sites were found (Metcalf and Metcalf 1979). Conversely, all turtles overwintered individually and no communal sites were observed in the Nebraska Sand Hills (Converse et al. 2002).

SD Geographic Range Estimation

The habitat distribution map produced by the MaxEnt model for *T. ornata*, even with a low probability, revealed new areas of potentially appropriate box turtle habitat where surveys could be performed, such as Mellette and Gregory counties. Although this prediction seems to be very useful in helping plan management around the existing populations, discovering new populations, or setting priorities to restore its natural habitat for more effective species conservation, it should be used with caution. This was the first prediction for the state of South Dakota and we still refining our analysis and results.

Reproduction and Growth

While we found nearly an equal number of male and female *T. ornata* (0.97 males: 1 female), sex ratios in other areas suggest that females predominate, with 0.58 males:1female in Kansas (Legler 1960), 0.7:1 in Illinois (Bowen et al. 2004), 0.64:1 in Wisconsin (Doroff and Keith

1990), and 0.58:1 (Converse 1999), and 0.51:1 in western Nebraska (Converse et al. 2002). However, recent data from Wisconsin suggest that sex ratios there are approximately equal (Redder et al. 2006). Variation in observed sex ratios across the range of this species could be due to variation in differential survival between sexes. Additionally, sex ratios may vary among years within and among any of the above sites due to the interacting effects of weather, environmental temperature and humidity, nest site location and depth, and oviposition dates (Vogt and Bull 1982). Temperature may play a key role in sex ratio variation, as *T. ornata* demonstrates temperature dependent sex determination, with females usually resulting from incubation temperatures above 29 C (Bull 1980, Packard et al. 1985, Packard and Packard 1986), but the temperature at which differentiation occurs is unknown.

Legler (1960) reported that courtship and mating in Kansas are most common in the spring, soon after emergence from hibernation, but occasionally take place in the summer or fall. Brumwell (1940) also noted mating in May from Kansas. Copulation was observed in Wisconsin from 28 May until 6 September (Temple 1987, Doroff and Keith 1990, Vogt 1991), but the earliest and latest matings for the species were observed in Texas on 5 April and 13 October (Blair 1976). We observed mating six times, once during every month from 28 April to 20 September (Table 1). Ernst and Lovich (2009) reported that both males and females may copulate several times a season, particularly during the spring, with more than one partner. We did not observe any individuals mating more than once a year, but did see a male copulating with two different females, one on 26 July 2010 and the other 28 April 2011.

Nesting has been reported from early May to mid-July in Kansas, but was most frequent in mid-June (Legler 1960). In Wisconsin, nesting has been reported from May through June (Temple 1987, Doroff and Keith 1990, Vogt 1991). Nieuwolt-Dacanay (1997) found gravid female *T. ornata luteola* from May through August in New Mexico. Based on weight loss, three females we followed laid eggs during the night of 15 – 16 June (Table 2). Although we never found the nests, they were assumed to be constructed in upland habitat, which consists of deep sandy soils with scattered grasses, forbs and shrubs. Ernst and Lovich (2009) reported that preferred nesting sites are similar to ours, with open, well drained areas that provide soft substrates.

In Kansas all adult females apparently lay eggs each year, with about 33% producing a second clutch in late June or early July (Legler 1960). In contrast, over a three-year period in New Mexico, percentages of gravid female *T. ornata luteola* ranged from 41.9 and 61.3% for two wet years, to only 10% during a dry year (Nieuwolt-Dacany 1997). Additionally, only 58% of females laid eggs in consecutive years. Doroff and Keith (1990) did not observe double-clutching in their Wisconsin population, and noted that during a two-year period, only 57% of the females laid eggs. Of the 11 females we x-rayed on 7 June 2010, seven (64%) contained eggs (Tables 2 and 3). This is roughly in line with the New Mexico population figures during wet years, as well as those for Wisconsin populations, but is well below the 100% figure cited for Kansas. Variation in these figures could be due to spring rainfall amounts, as proposed by Nieuwolt-Dacany (1997), with more females nesting in wet years than in dry. Since we only x-rayed females one year, we have no information to judge how our population would respond to rainfall variation. Redder et al. (2006) postulated that since sperm storage is possible in this species, the disparity between localities in the number of breeding females and the number of clutches per year per female is likely due to energy intake and/or thermoregulatory limitations.

Further, it should be noted that possible reasons some of the females we x-rayed did not contain eggs are: 1) eggs were already laid, 2) eggs were not developed enough to visualize on x-ray, but may develop and be laid later in the year, and 3) not all females reproduce every year.

Clutch sizes vary across the range of the species from 1 – 8, with mean clutch sizes of 3.5 for Wisconsin (Doroff and Keith 1990), 4.7 for Kansas (Legler 1960), and 2.7 in New Mexico for *T. o. luetola* (Nieuwolt-Dacanay 1997). Our clutch sizes ranged from 2 – 5, with an average of 4.3 (Table 2), which surpasses the averages for Wisconsin and New Mexico, but is slightly less than that reported for Kansas. Ernst and Lovich (2009) reported average egg mass for 65 eggs as 10.5 (8.0 – 14.3) g. Based on weight loss of females pre- and post- egg laying, our eggs averaged 12.0 g (Table 2), within the range of the above figures by Ernst and Lovich (2009). In addition to losing weight from egg mass during oviposition, females also lose weight by placing fluids into the nest cavity. Therefore, our average egg mass figure will be slightly above actual egg mass. Ernst and Lovich (2009) calculated relative clutch mass (clutch mass as a percent of female mass) from Legler's (1960) data as 12%. This figure is slightly smaller than ours, at 13.6%. Therefore, in terms of overall reproductive strategy, comparing the South Dakota to Kansas populations, South Dakota females appear to be producing fewer clutches (64% of females laying eggs annually vs. 100% annually plus 33% double clutching) and slightly fewer eggs per clutch (4.3 vs. 4.7) than those in Kansas, but producing slightly larger relative clutch masses in the process (12% vs. 13.6%).

As noted above, three of our females laid eggs during the night of 15 - 16 June. Incubation periods are temperature dependent (Legler 1960), but average duration in naturally incubated clutches in Wisconsin was 80 (79 - 84) days (Doroff and Keith 1990). If we use this incubation period, our eggs mentioned above would have hatched around 3 September. Legler (1960) reported that young box turtles in Kansas become active and alert as soon as they hatch, and remained so until low temperatures induced quiescence. However, Converse et al. (2002) and Costanzo et al. (1995) reported that the young usually hatch and emerge from the nest in August and September, but some dig beneath the nest cavity and overwinter there, emerging the next spring. Caldwell and Collins (1981) reported that hatching in Kansas usually occurs in the fall, but it may be delayed until spring if nest conditions are dry. It is unknown whether South Dakota turtles emerge upon hatching in the fall, bury beneath their nests upon hatching to spend the winter, hatch in the spring, or do all the above. However, the adult turtles we observed did not enter hibernation until late September to mid-October, which indicates conditions are appropriate for adult activity in September and part of October. This may or may not translate to appropriateness of conditions for hatchling activity. If South Dakota hatchlings do not emerge in the fall, they may not grow prior to formation of their first growth-ring. However, if they do emerge, some growth may occur. Thus, our data presented in Table 4 may overestimate the size of hatchlings, since those figures are based on the relative size of the first growth-ring.

Legler (1960) found that sexual maturity seemed more closely correlated with size than age. He found that males matured when smaller and younger than females, with 76% of the males maturing at a plastron length of 100 – 109 mm and at an age of 8 – 9 years. For females, 66% were mature at a plastron length of 110 – 119 mm and at an age of 10 – 11 years. For Texas populations, Blair (1976) reported that both sexes matured at a carapace length of nearly 100 mm, with males achieving that length in 7 years, and females in 8. In Colorado, Hammerson

(1999) reported females first reproduced when 11 years old. Captive males held under semi-natural conditions in Oklahoma matured in 5 years and 100 mm (curved carapace length), and captive females matured in 8 years and 128 mm (curved carapace length) (St. Clair 1998). Our smallest male exhibiting secondary sex characteristics had carapace and plastron lengths of 103.3 and 104.0 mm, respectively, and this same male was also among the youngest to exhibit these characteristics, at age 9 years (Table 16). Therefore, it seems males in our population mature at roughly the same size and age as those in the Kansas, at roughly the same size, but an older age than those in Texas and at an older age than the captives from Oklahoma. Carapace length comparisons cannot be made with the Oklahoma animals, as they were measured along the curvature of the shell rather than straight line measurements. We and all other investigators cited utilized straight-line measures.

Adult females are defined as individuals > 110 mm carapace length that do not demonstrate male secondary sex characteristics (Legler 1960, Converse 1999). Our smallest animal fulfilling these requirements had carapace and plastron lengths of 110.2 and 121.3 mm, respectively. The age of this animal could not be determined, but she had more than nine growth rings. Another female had a slightly longer carapace (111.6 mm), but had a shorter plastron (113.4 mm). The youngest known age animals fulfilling the above requirements were 9 years old (Table 16). The youngest known-age female we x-rayed that contained eggs was also 9 years old, and she was the smallest female with eggs, having carapace and plastron lengths of 113.2 and 117.9 mm, respectively (Table 11). The smallest female observed copulating was larger, with carapace and plastron lengths of 114.4 and 121.7, respectively. Age could not be determined on any copulating females, as their shells were too worn, but one was > 12 years. Therefore, South Dakota female box turtles can mature by age 9, with carapace and plastron lengths of 110.2 and 113.4 mm, respectively. Thus, our female turtles mature at roughly the same size as those in Kansas, but at a slightly younger age. They also mature at a younger age than those in Colorado, but at an older age and larger size than those in Texas. Oklahoma captive females also matured at an earlier age than our animals.

In Indiana, hatchlings ranged in size from 28.8 – 37 mm (Marr 1944, Minton 1972). Our hatchling size as calculated from growth-ring data averaged 31.7 mm (Table 13), which falls within the range cited above. Legler (1960) reported mean increments in plastron growth of 68, 29, and 18% for the first three years of life. From the fourth to the fourteenth year the growth rate slowed gradually from 13.3 to about 3%. The growth rates observed by us in South Dakota were slower, with the first three years averaging 28.3, 22.4, and 15.7% (Table 14). Our growth rates then varied from 8.7 to 9.8% to age nine, beyond which time sample sizes were too small to estimate growth. The growth variation observed from animals ages 4 - 9 may be due to variations within factors contributing to growth. For example, high precipitation and correspondingly high grasshopper numbers have been associated with rapid growth of hatchlings (Legler 1960). Such factors also may explain the slower growth of South Dakota turtles compared to those in Kansas. Additionally, the more southerly Kansas may provide a greater number of growing days per year. Overall, our females grew more rapidly than males (Table 14), but our sample sizes were too small to estimate growth beyond age 9, the age of the youngest adults of both sexes we observed in our population. However, St. Clair (1998) found that captive Oklahoma males grew more when adult than did females.

Training in Ecological Field Research to Oglala Lakota College (OLC) Students and Outreach to the Pine Ridge Reservation Communities

OLC undergraduate students were exposed to different aspects of research related to the ecology of the box turtle, including field and laboratory work, and were able to bring this information back to the scientific community, the college, and tribal communities.

The box turtle project provided a unique hands-on and place-based scientific method instruction to several OLC undergraduate students during the Field Ecology course (Figure 18) and to intern students (named below and supported by the National Science Foundation, Tribal Colleges and Universities Program) that have consistently been involved in the field and/or laboratory work during 2010 and/or 2011:

- Rebecca Bingham
- Requaw Pavy
- Michelle LeBeau
- Wendy Green
- Sheronne White
- Osceola BlueHorse
- Delaine Peterson
- Rhonda Kihega
- Lester Richards
- Camille Griffith
- Theodora Charging Crow

The students listed above devoted many hours collecting data, often under somewhat harsh environmental conditions. They truly demonstrated their enthusiasm to gain experience in field biology. A few have also been highly instrumental in organizing and analyzing the mass amount of data accumulated.

Michelle LeBeau

2012 American Indian Higher Education Consortium (AIHEC) – Rapid City, SD (poster presentation about ornate box turtle habitat selection in SD).

Rebecca Bingham

2012 American Indian Higher Education Consortium (AIHEC) – Rapid City, SD (oral presentation about ornate box turtle home range and ecological niche modeling; awarded 1st place).

2011 American Indian Higher Education Consortium (AIHEC) - Bismarck, ND (oral presentation about ornate box turtle home range in SD; awarded 3rd place).

2011 Bingham, R., Higa, A., and Quinn, H. Home range of ornate box turtle (*Terrapene ornata*) in South Dakota. The Wildlife Society's 18th Annual Conference. Abstracts (poster). Waikoloa, HI.

Requaw Pavy

2011 Pavy, R., Higa, A., and Quinn, H. Hibernation conditions and duration of an ornate box turtle (*Terrapene ornata*) population on the Pine Ridge Indian Reservation in South

Dakota. The Wildlife Society's 18th Annual Conference. Abstracts (poster). Waikoloa, HI.

Outreach (K-12)

Several presentations were given to OLC undergraduates and faculties and to the Pine Ridge Reservation (PRR) students and teachers (K-12 grade). PRR middle and high school students were also brought to the field for a hands-on experience during collaborations with OLC outreach programs such as the "*Native Science Field Center*" funded by the Hopa Mountain Foundation and the National Science Foundation - Tribal College and Universities Program (NSF-TCUP) and "*From the Field to the Fair*" funded by the National Science Foundation - Opportunities for Enhancing Diversity in the Geosciences Program (NSF-OEGD). We also participated in two Reservation Career Fairs. In 2011 the career fair had 119 participants from Little Wound High School, Shannon County Virtual High Schools, and Kadoka High School.

Dissemination

Higa. A. & Quinn, H. 2012. Ornate box turtle (*Terrapene ornata*) research update for South Dakota. South Dakota Game, Fish, and Parks Annual Wildlife Staff Meeting. Custer, SD.

Quinn, H., Quinn, H. & Higa. A. 2012. Notes on reproduction and growth of South Dakota ornate box turtle (*Terrapene ornata*). Dakota Amphibian and Reptile Network (DARN) Annual Meeting. Spearfish, SD.

Quinn, H., Quinn, H. & Higa, A. 2012. Overview of the South Dakota box turtle ecology project with emphasis on reproduction and growth. Black Hills Area Botanist & Ecologist Workshop (BHABEW X). Rapid City, SD.

Quinn, H., Quinn, H. & Higa, A. 2011. Overview of the South Dakota ornate box turtle ecology project. Dakota Amphibian and Reptile Network (DARN) Annual meeting, Minot, ND.

Quinn, H., Quinn, H. & Higa. A. 2011. South Dakota Ornate Box Turtle Ecology Project. Dakota Amphibian and Reptile News 4 (1): 12-14.

CONSERVATION CONCERNS

Threats and Issues in South Dakota

Road mortality: Many authors over the years have commented on deaths of large numbers of ornate box turtles on roads (Blair, 1976, Collins 1993, Doroff and Keith 1990, Ernst and Barbour 1972, Legler 1960, Rodeck 1949), indicating that roadkill is a significant cause of mortality. In fact, more adults seem to be killed by automobiles than all predators combined (Ernst and Lovich 2009).

It is critical to mitigate these losses given the life history traits of ornate box turtles. Such traits include a long, somewhat sedentary life; a low rate of recruitment; high adult survivorship; high egg and hatchling mortality; relatively late maturity; and low fecundity (Dodd 2001). As a result of these traits, the loss of even one or two reproductive females per year from small populations can result in the eventual extinction of those populations (Doroff and Keith 1990, Congdon et al. 1993, Belzer 2000, Gibbs and Shriver 2002).

Habitat alteration: Another leading cause of population declines across the geographic range of ornate box turtles is habitat loss due to human activities (Bowen et al. 2004). In South Dakota, conversion of native prairie to agricultural land is probably the leading cause of habitat alteration negatively affecting this species. Additionally, it appears that management practices that result in: 1) succession of grasslands to a more wooded stage, 2) destruction of shrubs essential for thermoregulation, or 3) elimination of rodents whose burrows are used for thermoregulation or hibernation could be harmful to ornate box turtle populations (Nieuwolt 1996, Curtin 1997 1998, Converse and Savidge 2003, Fitch 2006).

Farm machinery and mowers: Roadside mowing and using farm machinery can be fatal to box turtles (Metcalf and Metcalf 1985, Doroff and Keith 1990, Dodd 2001).

Commercial pet trade: Substantial numbers of box turtles have been collected in the past for the domestic and international pet trade, with potentially significant population impacts (van Dijk and Hammerson 2011). Instances are known where entire truckloads of ornate box turtles have been collected in a small area in a single day (Redder et al. 2006). To protect box turtles from international trade, the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) listed North American box turtles on Appendix II in 1994. This international treaty regulates and monitors trade in species that either are or might become endangered by trade. Box turtles receive an additional measure of protection from the U.S. Food and Drug Administration. Commercial sale of turtles less than four inches in carapace length is prohibited in order to prevent the spread of salmonella (see Levell 1997). However, even with these protective measures from various state and federal agencies, as well as an international treaty, a large black market for commercial trade of box turtles still persists in Japan and Europe, with individual animals being sold for hundreds of dollars (Dodd 2001). A recent check of the internet found ornate box turtles for sale for $219.95 (www.turtlesale.com), $249 (baby), $269 (juvenile), and $219 (adult) (www.turtleshack.com).

As mentioned in the Road Mortality section above, removal of even small numbers of adult box turtles is detrimental to population sustainability. Populations can take decades to recover (Hall et al. 1999), and simply cannot withstand the pressures of commercial collection.

Disease (*Ranaviris*): We have not observed any apparent box turtle mass mortality events in South Dakota due to diseases. However, as a cautionary note, such mortality events have occurred as a result of the pathogen *Ranaviris* in eastern and Florida box turtles (*Terrapene carolina carolina* and *T. carolina bauri*) from Florida, Georgia, New York, and Pennsylvania. These mortalities occurred between 2003 – 2005, but were preceded by die-offs in Texas during 1998 (Johnson et al. 2007). More recently (2008 – 2011), an outbreak in Maryland decimated populations there (Farnsworth and Seigel 2012). While these mortalities impacted the closely related box turtles to the east and south, ornate box turtles can also be affected by this disease (Johnson et al. 2007). This disease has been detected in South Dakota, and can impact not only turtles, but also frogs, salamanders, fishes, and insects (Jacob Kirby, University of South Dakota, personal communication; Farnsworth and Seigel 2012).

Climate change: Predictions of climate change in South Dakota point to wetter springs and hotter, drier summers (Cochrane and Moran 2011). It is unknown how box turtle

populations will respond to such a changing environment; however, Converse et al. (2005) found that survival rates of hibernating ornate box turtles in Nebraska are highest in years with low minimum winter temperatures. This suggests that warmer winters might produce higher metabolic rates during hibernation, resulting in depletion of lipid reserves, which would adversely affect subsequent survival and reproduction.

Another concern about climate change is that box turtles experience temperature dependent sex determination (Packard et al. 1985). This means that the temperature at which eggs incubate determines sex of the resulting offspring. At low incubation temperatures, males are produced, and at high temperatures, females (Vogt and Bull 1982). Intermediate temperatures produce varying ratios of both sexes. As a result of climate change, it is predicted that South Dakota temperatures for June, July, and August (which, coincidentally, is the period when box turtle eggs are incubating) will increase up to 2^0 C (3.6^0 F) above current temperatures (Cochrane and Moran 2011). This increase in environmental temperature as a result of climate change could easily result in the production of a skewed sex ratio toward females, or perhaps even all female populations, which would clearly be deleterious to ornate box turtles. In support of this, some scientists have recently suggested that climate change has the potential to eliminate the production of male turtle offspring if average global temperatures increase 4^0 C (7.2^0 F), and that increases of less than 2^0 C (3.6^0 F) may dramatically skew sex ratios (Janzen 1994).

Protection Status is South Dakota

Ornate box turtle may be collected under the authority of a state fishing license, and have a bag limit of two daily, with a maximum possession of four. No commercial activity is permitted regarding ornate box turtles. This species may not be shot with a firearm.

MANAGEMENT RECOMMENDATIONS

Items one through seven were extracted from Smith and Quinn (2012).

1. Identify areas of high road mortality, and design methods to ameliorate such losses. A proven method to mitigate turtle losses is to provide drift fences that lead to culverts, so the animals may safely move under roads (Aresco 2005).

2. Encourage an appropriate level of cattle grazing where box turtle populations are known to exist. Grazing helps maintain a fairly open environment for box turtle basking, while retaining certain shrubs (sage, yucca) for shelter from heat. While Caldwell and Collins (1981) reported trampling by cattle to be an important cause of death in ornate box turtles, Fitch (2006) found the presence of cattle to be an important factor in maintaining box turtle populations. At his Kansas site, succession from grassland to a more wooded stage occurred after cattle were removed, resulting in a collapse of ornate box turtle and other reptile populations. Controlled burns may also be a method to limit tree growth, but could be disastrous, killing many turtles, unless done during the winter when the turtles are hibernating underground.

3. Encourage construction of wetlands where box turtle populations occur. Box turtle densities in sand prairies are highest where there are nearby wetlands (including ephemeral ponds and windmill overflows) (Converse et al. 2002). These microhabitats are used during times of water stress. Hence, constructing ponds or installing wildlife/cattle watering windmills will increase ornate box turtle populations (assuming that cattle densities aren't too high) (John Iverson, Earlham College, personal communication).

4. In areas of known turtle populations, encourage county, federal, and tribal agencies to set roadside mower blades at eight inches, thus avoiding contact of blades with turtles (Redder et al. 2006). To minimize the chances of such mortality, mowing should be done during the heat of the day when turtles will be inactive and under cover, or when it is too cold for turtles to be active on the surface.

5. Assure South Dakota wildlife officers are aware that a lucrative, illegal trade in ornate box turtles exists, and that no turtles can be collected in our state for commercial purposes.

6. Continue surveys for populations of ornate box turtles in the state, especially on protected land (i.e., Buffalo Gap National Grassland, Badlands National Park, Lacreek National Wildlife Refuge, The Nature Conservancy land and easements), and quantify population densities as well as area of continuous suitable habitat within each protected area. This will identify strong populations on protected land for which conservation management plans can be developed. Doroff and Keith (1990) tentatively suggested that areas of at least 100 ha (247 acres) are required to sustain viable box turtle populations, also noting that these areas should be roadless.

7. Encourage enactment of tribal law to provide protection of ornate box turtles on the Pine Ridge and Rosebud Reservations, especially in light of the commercial trade value of this species. The Oglala Sioux Tribe currently has no wildlife protection laws. Additionally, encourage development of conservation management plans on tribal lands where box turtle populations occur.

8. Ornate box turtles preferred dense shrub vegetation, as indicating by the visual obstruction readings (VOR) for optimal microhabitat at 17 bands (22 cm). Sand sagebrush provided most of the visual obstruction at turtle locations. Optimum target conditions for box turtle management for canopy cover would require the following: total shrubs (40%), sand sagebrush (38%), total vegetation (79%), and litter (59%). Turtles preferred lower amounts of canopy cover than available in the habitat for total grass (37%), total forbs (19%), and bare ground (14%). These guidelines should be beneficial and effective in conservation of the ornate box turtle in these sand hills of South Dakota.

9. Due to the turtles long lifespan it will be important to monitor and estimate box turtles annual home ranges in a long term, as home range size could be influenced by the resources available, predation avoidance, and human disturbance. In addition,

differentiating home range estimations and identification of core areas will help to set priorities for more effective species conservation and management plans.

FUTURE RESEARCH

2012 - 2014 "*Biology, Ecology, Conservation Genetics, and Management of the Ornate Box Turtle (Terrapene ornata) in South Dakota"*. Awarded by the National Science Foundation (NSF), Tribal Colleges and Universities Undergraduate Program (TCUP), Research Initiation Awards (RIA). Collaborative grant between the Oglala Lakota College and Black Hills State University.

Abstract: Landscape genetics, conservation, and management of the ornate box turtle in SD. There is a rising need for conservation studies and resource managers to estimate the amount of population connectivity in threatened species in order to maximize the impact of present and future management efforts (Segelbacher et al. 2010). Habitat loss and fragmentation are considered two of the chief contributors to global biodiversity loss (Fischer and Lindenmeyer 2007, Lindenmeyer et al. 2008, Heller and Zavaleta 2009, Segelbacher et al. 2010). Rare or endangered species, such as the ornate box turtle, are especially susceptible to population fragmentation because the small population sizes found in these species already limit population connectivity. If gene flow is limited among local populations, genetic drift can effectively increase the frequency of deleterious alleles (Lynch et al. 1995). Conversely, frequent gene flow among local populations will maintain genetic variation and increase population viability effectively counteracting the impacts of genetic drift. Thus an essential component of conservation biology is the need to estimate population connectivity across landscapes by directly measuring gene flow among local populations (Van Dyck and Baguette 2005). Landscape genetics offers an increasingly powerful set of tools that integrate population genetics, landscape features, and spatial statistics to examine genetic variation in space and time and the relationship of genetic variation to environmental variables of import to a given species (for review see Segelbacher et al. 2010). Using a landscape genetics approach to study population connectivity in the *T. ornata* in South Dakota will be the most efficient and effective way to develop informed management and conservation planning for the species. Our specific aim is to use landscape genetics to rigorously identify and evaluate the factors that drive gene flow in the ornate box turtle in the complex and dynamical landscape of the South Dakota sandhills. Understanding of the regional corridors that connect populations of box turtles in South Dakota may also contribute to our understanding of corridors important to other species endemic to the sandhills habitats. We will work closely with managers and officers of the Oglala Sioux Parks and Recreation Authority and the South Dakota Game, Fish, and Parks to ensure that the results of the landscape genetics study are used to develop management and conservation plans for the ornate box turtle.

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LITERATURE CITED

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TABLES & FIGURES

Table 1. Environmental variables, including the bioclimatic variables (BIO#) from WorldClim dataset (Hijmans et al. 2005; http://www.worldclim.org/bioclim.htm), used to predict suitable habitat distribution for ornate box turtle (*Terrapene ornata*) in South Dakota.

Environmental variable #	Environmental variable description	Percent of contribution
BIO1	Annual Mean Temperature	6.4[4]
BIO2	Mean Diurnal Range (Mean of monthly (max temp - min temp))	0.4
BIO3	Isothermality (BIO2/BIO7) (* 100)	41.8[1]
BIO4*	Temperature Seasonality (standard deviation *100)	--
BIO5*	Maximum Temperature of Warmest Month	--
BIO6	Minimum Temperature of Coldest Month	0.2
BIO7*	Temperature Annual Range (BIO5-BIO6)	--
BIO8	Mean Temperature of Wettest Quarter	0.9
BIO9	Mean Temperature of Driest Quarter	3.5
BIO10*	Mean Temperature of Warmest Quarter	--
BIO11	Mean Temperature of Coldest Quarter	19.6[2]
BIO12*	Annual Precipitation	--
BIO13*	Precipitation of Wettest Month	--
BIO14	Precipitation of Driest Month	1.7
BIO15	Precipitation Seasonality (Coefficient of Variation)	7.6[3]
BIO16	Precipitation of Wettest Quarter	4.3
BIO17	Precipitation of Driest Quarter	1.7
BIO18	Precipitation of Warmest Quarter	1.1
BIO19	Precipitation of Coldest Quarter	0.1
Sdsoila**	Soil data layer: percent sand composition	6.0[5]
Sdalt	Altitude	4.7

Note: Bioclimatic variables marked with * where removed from the final analysis as they did not contribute to the Maxent modeling. ** Soil data layer created by calculating the percent sand composition for each soil within each county separately, then adding this attribute to their spatial data table, and merging all South Dakota counties together to make one single layer. The soil data layer was then changed to a raster and ASCII and used in MaxEnt modeling. The superscript numbers on percent column are the contribution ascendant order of the first five environmental variables.

Table 2. Visual obstruction readings (VOR) by Robel pole band number (1.27cm) over a two-year period at the site of the turtle, 10 m distance and yearly average.

Year	Sample Size	Turtle Mean ± SE Bands	10m Distance Mean ± SE Bands
2010	185	17.7 ± 0.8*	10.9 ± 0.4
2011	112	17.2 ±0.9*	12.2 ± 0.5

* Significantly different at $p < 0.10$.

Table 3. Comparison of ornate box turtle preferences between years by habitat category.

Category	Canopy Cover (%) Mean ± SE 2010 (n = 184)	Canopy Cover (%) Mean ± SE 2011 (n = 114)
Total Grass	29.8 ± 2.1*	48.0 ± 3.0
Total Forbs	12.8 ± 1.3*	29.5 ± 2.8
Total Shrubs	45.4 ± 3.0*	32.0 ± 3.6
Cactus	0.2 ± 0.1	1.4 ± 0.9
Yucca	1.4 ± 0.8	0.1 ± 0.1
Sand Sagebrush	43.3 ± 3.1*	28.3 ± 3.5
Silver Sagebrush	0.7 ± 0.5	0.9 ± 0.4
Total Vegetation	76.0 ± 2.2*	84.1 ± 1.8
Litter	50.2 ± 2.6*	73.8 ± 3.0
Bare Ground	15.5 ± 1.9	12.5 ± 2.1

* Significant differences between years $p < 0.10$.

Table 4. Comparison of ornate box turtle canopy cover preferences from available habitat (10 m distance from turtle). Mean of 2010 and 2011 data combined, n = 298.

Category	Canopy cover (%) Mean ± SE Turtle	Canopy cover (%) Mean ± SE 10 m
Total Grass	36.8 ± 1.8 *	49.8 ± 1.3
Total Forbs	19.2 ± 1.4 *	23.6 ± 1.0
Total Shrubs	40.3 ± 2.3 *	8.6 ± 0.7
Cactus	0.6 ± 0.4	0.8 ± 0.2
Yucca	0.9 ± 0.5	0.3 ± 0.1
Sand Sagebrush	37.5 ± 2.3 *	7.6 ± 0.7
Silver Sagebrush	0.8 ± 0.3	0.8 ± 0.2
Total Vegetation	79.1 ± 1.6 *	71.2 ± 1.1
Litter	59.2 ± 2.1 *	54.6 ± 1.5
Bare Ground	14.3 ± 1.4 *	19.9 ± 1.0

* Significant differences at $p < 0.10$.

Table 5. Home range area (ha) calculated as Minimum Convex Polygons (MCP) of adult ornate box turtles in South Dakota with an average of 20 locations annually, showing comparisons between gender and years.

MCP	**2010**		**2011**	
	Females (n=11)	Males (n=10)	Females (n=12)	Males (n=12)
Mean	5.46 ± 7.18	16.80 ± 26.48	5.27 ± 6.56	3.27 ± 2.47
Median (ha)	3.05	6.58	3.54	2.85
Minimum (ha)	0.58	2.10	0.96	0.35
Maximum (ha)	26.67	89.00	25.50	9.36

* Significant differences between sexes $p = 0.10$

Table 6. Home range area (ha) calculated as Minimum Convex Polygons (MCP) of adult box turtles in South Dakota with an average of 20 locations annually, showing comparisons between gender and years. Three extreme outliers were removed after the box plot graph.

MCP	**2010**		**2011**	
	Females (n=10)	Males (n=9)	Females (n=11)	Males (n=12)
Mean (ha) ± SD	3.34 ± 1.53*	8.78 ± 8.07*	3.42 ± 1.65	3.27 ± 2.47
Median (ha)	3.05	4.37	3.31	2.85
Minimum (ha)	0.58	2.10	0.96	0.35
Maximum (ha)	5.34	22.28	6.67	9.36

* Significant differences between sexes $p = 0.10$

Table 7. Minimum and maximum home range areas (ha) calculated as local convex hull (LoCoH) nonparametric Kernel method of adult box turtles in South Dakota with an average of 20 locations annually, showing comparisons between gender and years.

LoCoH (K values)	**2010**		**2011**	
	Females (n=11) Min. / Max. (ha)	Males (n=10) Min. / Max. (ha)	Females (n=12) Min. / Max. (ha)	Males (n=12) Min / Max. (ha)
20	0.58 / 26.47	2.10 / 89.00	0.96 / 25.50	0.35 / 19.70
15	0.58 / 22.00	2.07 / 89.00	0.96 / 25.50	0.35 / 19.70
10	0.53 / 21.00	2.01 / 79.82	0.93 / 24.06	0.35 / 19.70
5	0.40 / 16.31	1.27 / 70.15	0.50 / 15.42	0.35 / 8.71

* Significant differences between sexes $p = 0.10$.

Table 8. Average home range areas (ha) calculated as local convex hull (LoCoH) nonparametric Kernel method of adult box turtles in South Dakota with an average of 20 locations annually, showing comparisons between gender and years.

LoCoH (K values)	**2010**		**2011**	
	Females (n=11) Mean (ha) ± SD	Males (n=10) Mean (ha) ± SD	Females (n=12) Mean (ha) ± SD	Males (n=12) Mean (ha) ± SD
20	5.38 ± 7.13	16.76 ± 26.50	5.26 ± 6.56	4.74 ± 5.30
15	4.81 ± 5.86	16.74 ± 26.52	5.02 ± 6.65	4.73 ± 5.30
10	4.51 ± 5.59	14.63 ± 24.08	4.46 ± 6.35	4.52 ± 5.35
5	3.32 ± 4.37	12.92 ± 21.38	2.945± 4.15	2.53 ± 2.20

* Significant differences between sexes $p = 0.10$

Table 9. Sample weeks from 2010-2011 hibernation, minimum, maximum, and average (± SD) temperatures (in ^{0}F), collected from temperature data loggers attached to adult ornate box turtles (*Terrapene ornata*) in South Dakota.

Week #	Start date	End date	N	Minimum temp (^{0}F)	Maximum temp (^{0}F)	Mean temp (^{0}F) ± SD
Week 20	10/22/10	10/28/10	16	57.42	66.34	61.27 ± 2.68
Week 21	10/29/10	11/04/10	16	54.58	65.18	58.49 ± 3.04
Week 22	11/05/10	11/11/10	16	54.04	63.95	58.16 ± 2.94
Week 23	11/12/10	11/18/10	16	49.78	62.88	54.26 ± 3.90
Week 24	11/19/10	11/25/10	16	45.97	61.2	49.84 ± 4.77
Week 25	11/26/10	12/01/10	16	42.83	59.36	46.85 ± 5.12
Week 26	12/02/10	12/08/10	16	40.42	57.38	44.84 ± 5.13
Week 27	12/09/10	12/15/10	16	39.39	55.31	43.37 ± 4.95
Week 28	12/16/10	12/22/10	16	37.67	53.76	41.92 ± 4.89
Week 29	12/23/10	12/29/10	16	36.02	51.88	40.63 ± 4.69
Week 30	12/30/10	01/05/11	16	36.53	50.88	40.19 ± 4.41
Week 31	01/06/11	01/12/11	16	35.81	49.95	39.56 ± 4.28
Week 32	01/13/11	01/19/11	16	35.7	49.24	39.20 ± 4.45
Week 33	01/20/11	01/26/11	16	34.69	49.56	38.38 ± 4.30
Week 34	01/27/11	02/02/11	16	35.02	49.01	38.21 ± 4.06
Week 35	02/03/11	02/09/11	16	34.03	48.21	37.37 ± 4.11
Week 36	02/10/11	02/16/11	16	34.46	47.3	37.22 ± 3.80
Week 37	02/17/11	02/23/11	16	35.09	46.71	38.70 ± 3.33
Week 38	02/24/11	03/02/11	16	35.48	47.27	38.13 ± 3.67
Week 39	03/03/11	03/09/11	16	34.35	45.8	37.36 ± 3.25
Week 40	03/10/11	03/16/11	16	34.9	44.97	37.54 ± 2.88
Week 41	03/17/11	03/23/11	16	38.07	47.78	43.22 ± 2.63
Week 42	03/24/11	03/30/11	16	41.13	45.57	43.87 ± 1.39
Week 43	03/31/11	04/06/11	16	43.38	53.74	47.72 ± 2.96
Week 44	04/07/11	04/13/11	16	47.17	59.29	54.45 ± 2.92
Week 45	04/14/11	04/20/11	16	42.08	48.18	44.59 ± 1.46
Week 46	04/21/11	04/27/11	16	51.95	55.44	53.67 ± 1.25
Week 47	04/28/11	05/04/11	16	49.33	59.66	56.74 ± 2.58
Week 48	05/05/11	05/11/11	16	62.88	68.61	66.27 ± 1.75
Week 49	05/12/11	05/18/11	16	53.83	59.99	57.11 ± 1.37
Week 50	05/19/11	05/25/11	16	60.08	64.31	62.33 ± 1.19

Table 10. Observed copulations in South Dakota box turtles (*Terrapene o. ornata*) during 2010-2011. Dashed lines in the age columns indicate plastrons were too worn to count growth-rings. Such lines in the carapace and plastron length columns indicate no such measures were made.

Date	Time (h)	Male age (years)	Female age (years)	Male carapace length (mm)	Female carapace length (mm)	Male plastron length (mm)	Female plastron length (mm)
20 September 2010	0820	----	----	----	115.7	----	122.5
26 July 2010	0730	----	----	----	114.4	----	121.7
9 August 2010	0850	> 8	----	120.6	122.2	123.3	129.1
28 April 2011	1100	----	> 12	111.4	125.3	110.7	127.4
3 May 2011	1315	15	----	111.7	116.5	115.3	119.6
20 June 2011	0919	----	----	119.0	115.3	126.5	116.8

Table 11. Clutch sizes of seven South Dakota female box turtles (*Terrapene o. ornata*) as visualized on x-ray 2 June 2010. Clutch mass represents changes in weight of females before and after egg-laying. Percent of female weight loss from laying eggs was calculated as [(female pre egg-laying weight) – (female post egg-laying weight)/pre egg-laying weight)] [100]. Dashed lines indicate no data are available.

Age (years)	Carapace length (mm)	Plastron length (mm)	Number of eggs	Clutch mass (g)	Mean egg mass (g)	Female weight loss (%)
> 12	116.4	121.5	5	65	13	13.8
> 12	125.3	127.4	4	60	12	11.8
> 12	----	----	5	55	11	15.3
9	113.2	117.9	2	----	----	----
----	122.1	129.1	5	----	----	----
> 13	119.3	123.2	5	----	----	----
14	114.4	121.7	4	----	----	----

Table 12. Female South Dakota box turtles (*Terrapene o. ornata*) found not to contain eggs upon x-ray. Dashed lines in the age column indicate plastrons were too worn to count growth-rings.

Age (years)	Carapace length (mm)	Plastron length (mm)
> 19	118.4	121.5
9	112.1	115.1
----	124.3	130.4
----	116.7	125.0

Table 13. Plastron lengths in mm as calculated for various ages from annual growth-rings measured in adult males and females. Combined figures are those calculated from males, females and juveniles collectively. H represents plastron length during the first winter.

Annual growth-rings	Male mean plastron length ± SD (n)	Female mean plastron length ± SD (n)	Combined mean plastron length ± SD (n)
H	33.9 ± 5.4 (13)	29.6 ± 5.0 (9)	31.7 ± 5.9 (39)
1	43.8 ± 6.0 (13)	39.2 ± 4.6 (9)	40.6 ± 6.3 (37)
2	52.2 ± 6.1 (12)	49.3 ± 5.1 (8)	49.8 ± 6.6 (35)
3	59.9 ± 7.8 (12)	57.5 ± 2.8 (7)	57.5 ± 7.8 (32)
4	67.1 ± 7.2 (12)	65.2 ± 6.5 (7)	63.7 ± 8.7 (29)
5	75.2 ± 8.3 (11)	71.3 ± 4.4 (7)	72.2 ± 9.8 (27)
6	81.7 ± 9.8 (10)	78.7 ± 4.9 (8)	78.4 ± 10.6 (26)
7	89.5 ± 9.5 (10)	88.8 ± 6.8 (8)	86.1 ± 10.8 (24)
8	95.3 ± 8.9 (8)	97.1 ± 5.7 (8)	92.8 ± 10.6 (22)
9	100.8 ± 8.1 (6)	107.7 ± 6.4 (7)	101.9 ± 9.8 (17)
10	99.6 ± 11.5 (2)	103.4 (1)	99.8 ± 7.2 (4)
11	107.3 (1)	104.7 (1)	106.0 ± 1.8 (2)
12	----	120.5 (1)	120.5 (1)

Table 14. Percent increase in plastron length as calculated from annual growth-rings of adult males and females. Combined figures are those from males, females, and juveniles collectively. Annual growth-ring 1 represents growth from the first to the second winter.

Annual growth-rings	Males (n)	Females (n)	Combined (n)
1	29.3 (13)	39.2 (9)	28.3 (37)
2	19.1 (12)	25.6 (8)	22.4 (35)
3	14.8 (12)	16.6 (7)	15.7 (32)
4	12.0 (12)	13.4 (7)	10.8 (29)
5	12.1 (11)	9.5 (7)	13.2 (27)
6	8.8 (10)	10.3 (8)	8.7 (26)
7	9.5 (10)	12.8 (8)	9.8 (24)
8	6.5 (8)	9.4 (8)	7.8 (22)
9	5.7 (6)	10.9 (7)	9.8 (17)
10	-1.2 (2)	-4.0 (1)	-2.0 (4)
11	7.8 (1)	1.3 (1)	6.2 (2)
12	----	15.0 (1)	13.6 (1)

Table 15. Comparison of mean carapace and plastron lengths of adult male and female South Dakota box turtles (*Terrapene o. ornata*).

Sex	**Carapace**	**Plastron**
	Mean (mm) ± SD (*n*)	Mean (mm) ± SD (*n*)
Male	114.5 ± 6.1 (59)*	118.1 ± 6.1 (59)
Female	117.8 ± 5.2 (67)*	123.9 ± 4.6 (67)

* Significant differences between sexes $p = 0.05$.

Table 16. Carapace and plastron lengths of known age turtles. M, F, and J are male, female, and juvenile turtles, respectively.

Age (years)	Sex	n	Mean carapace length (mm) ± SD	Mean plastron length (mm) ± SD
1	J	2	44.5 ± 3.0	44.5 ± 6.5
2	J	2	62.7 ± 10.8	63.8 ± 8.4
3	J	1	67.2	71.6
4	J	3	67.5 ± 2.2	68.9 ± 3.2
7	J	2	93.2 ± 3.1	98.3 ± 8.1
8	J	2	96.3 ± 11.8	98.7 ± 16.6
9	M	3	110.8 ± 3.1	111.7 ± 2.0
	F	2	112.6 ± 0.8	116.7 ± 1.8
	J	2	103.6 ± 5.6	108/.0 ± 7.8
10	M	2	111.5 ± 3.0	115.8 ± 1.8
	F	1	111.6	113.4
	J	1	103.2	97.3
11	M	1	135.0	130.0
	F	2	116.7 ± 0.9	121.9 ± 1.6
12	F	1	116.2	124.1
15	M	1	111.7	115.3

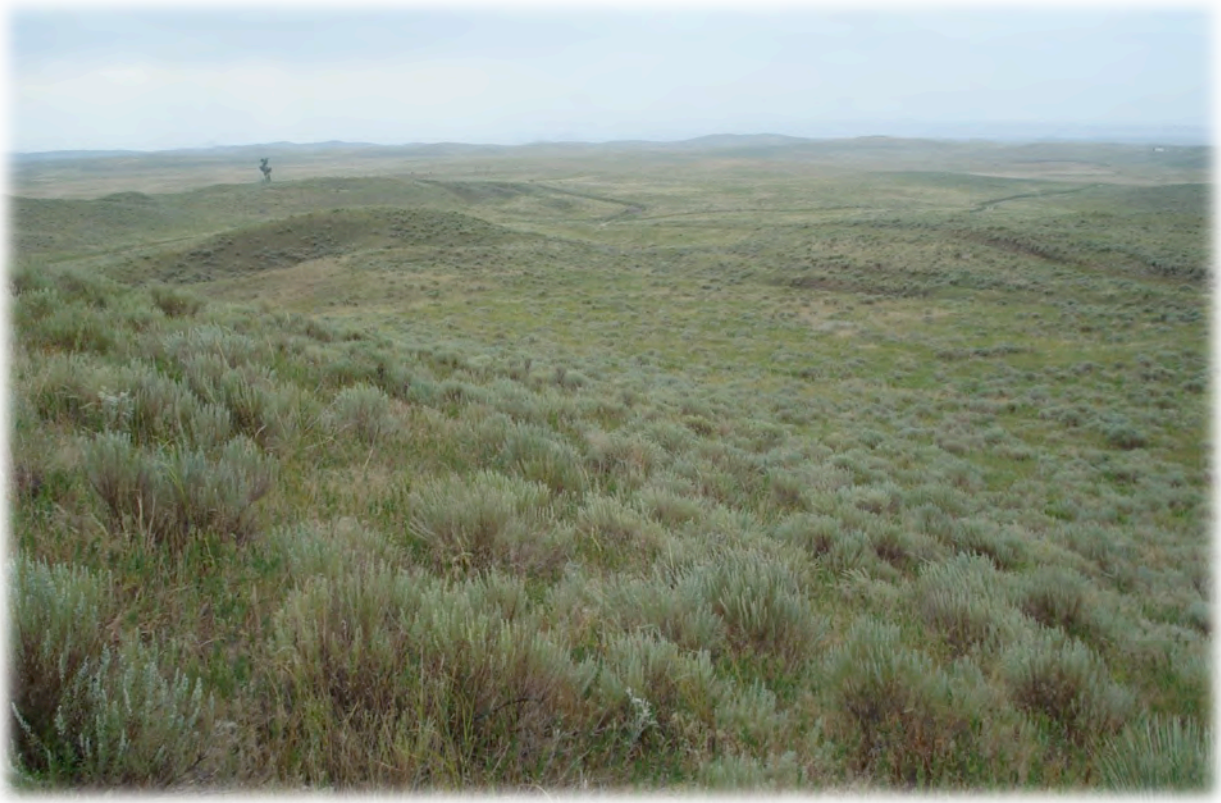

Figure 1. Ornate box turtle study area in a sands ecological site dominated with sand sagebrush located on the Pine Ridge Reservation, Shannon County in southern central South Dakota.

Figure 2. Ornate box turtle fitted with radio transmitter and temperature data logger.

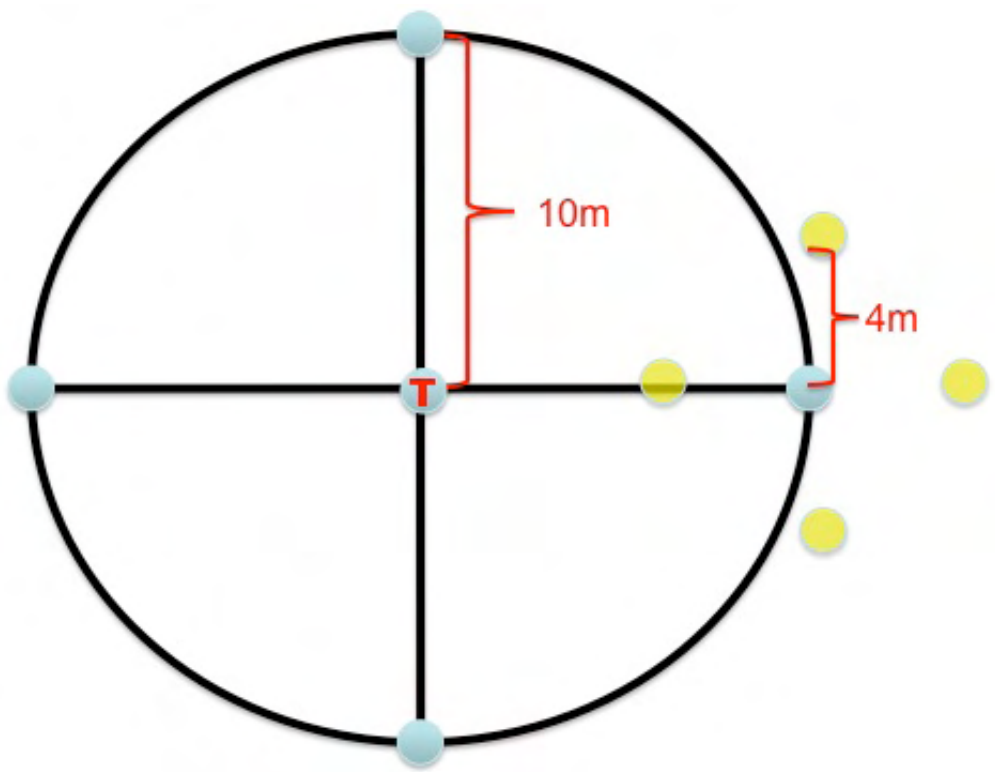

Figure 3. Diagram showing locations where visual obstruction readings (VOR) and canopy cover measures were collected (blue dots), including the ornate box turtle location (blue dot marked as "T" in the center). Four VOR readings (yellow dots) were made at each station (blue dots), totaling 20 readings per turtle.

Figure 4. Robel pole with white and gray alternating bands (1,27 cm) for collecting visual obstruction readings (VORs).

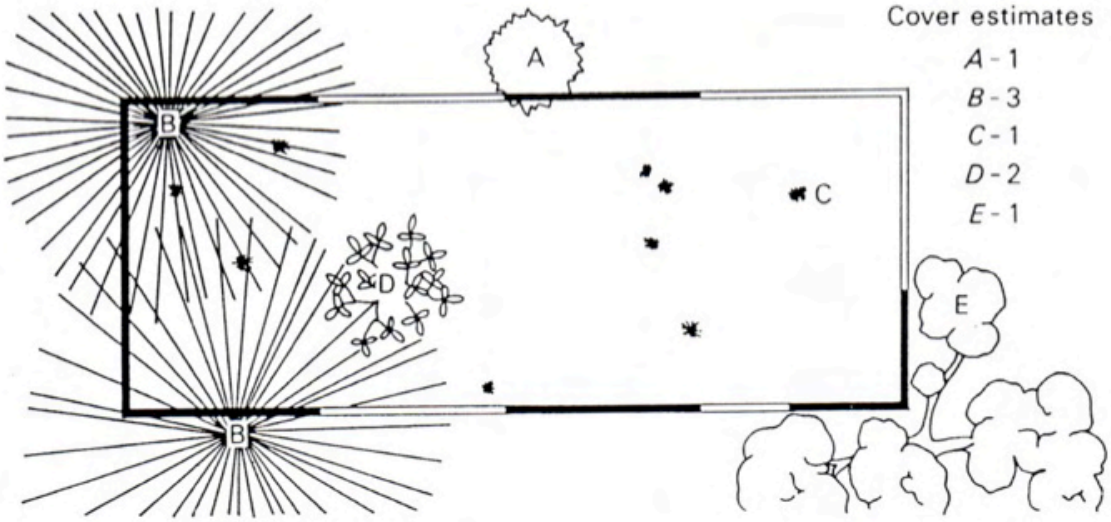

Figure 5. Daubenmire frame for estimating canopy cover of habitat variables for ornate box turtles (*Terrapene ornata*) in South Dakota. Diagram adapted from Daubenmire (1959).

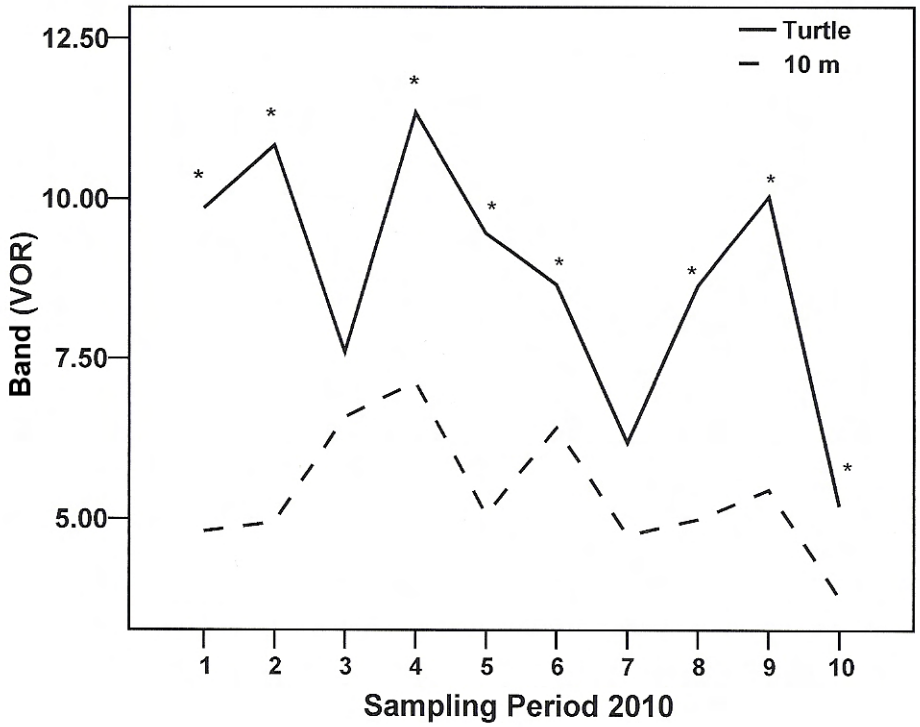

Figure 6. Visual obstruction readings (VOR; band = 1.27 cm) comparing ornate box turtle (*Terrapene ornata*) preferences with habitat availability (10 m distance from turtle) during 2010. *Significant statistical differences between turtle locations and habitat availability.

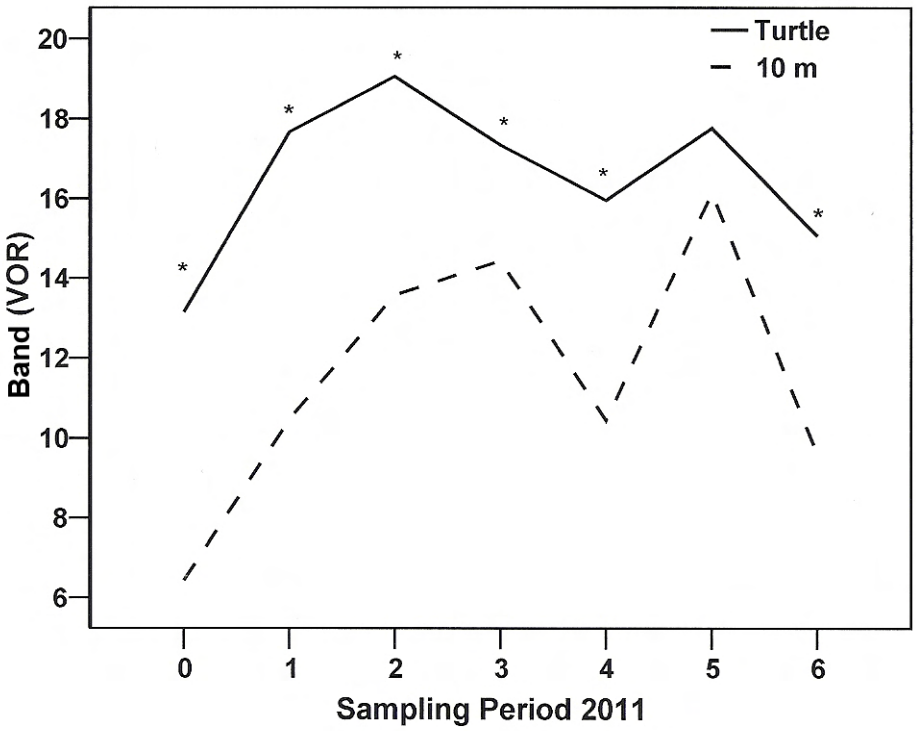

Figure 7. Visual obstruction readings (VOR; band = 1.27 cm) comparing ornate box turtle (*Terrapene ornata*) preferences with habitat availability (10 m distance from turtle) during 2011. *Significant statistical differences between turtle locations and habitat availability.

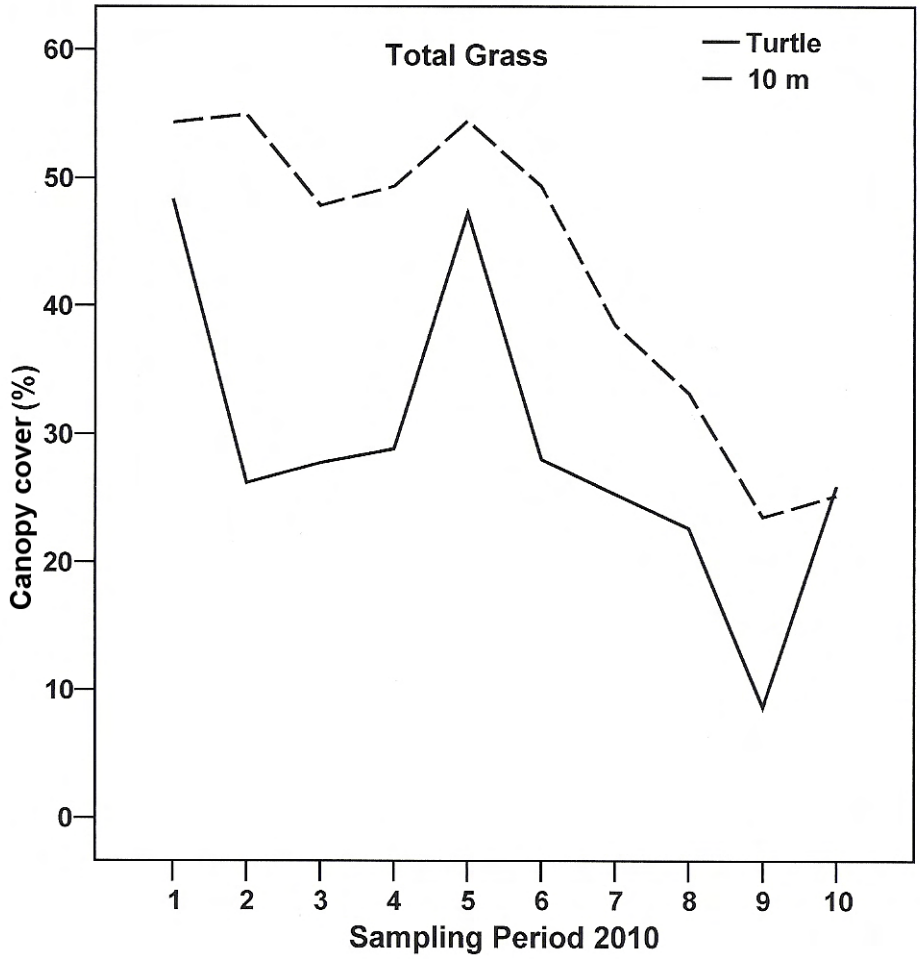

Figure 8. Canopy cover (%) for total grass comparing ornate box turtle (*Terrapene ornata*) preferences with habitat availability (10 m distance from turtle) during 2010.

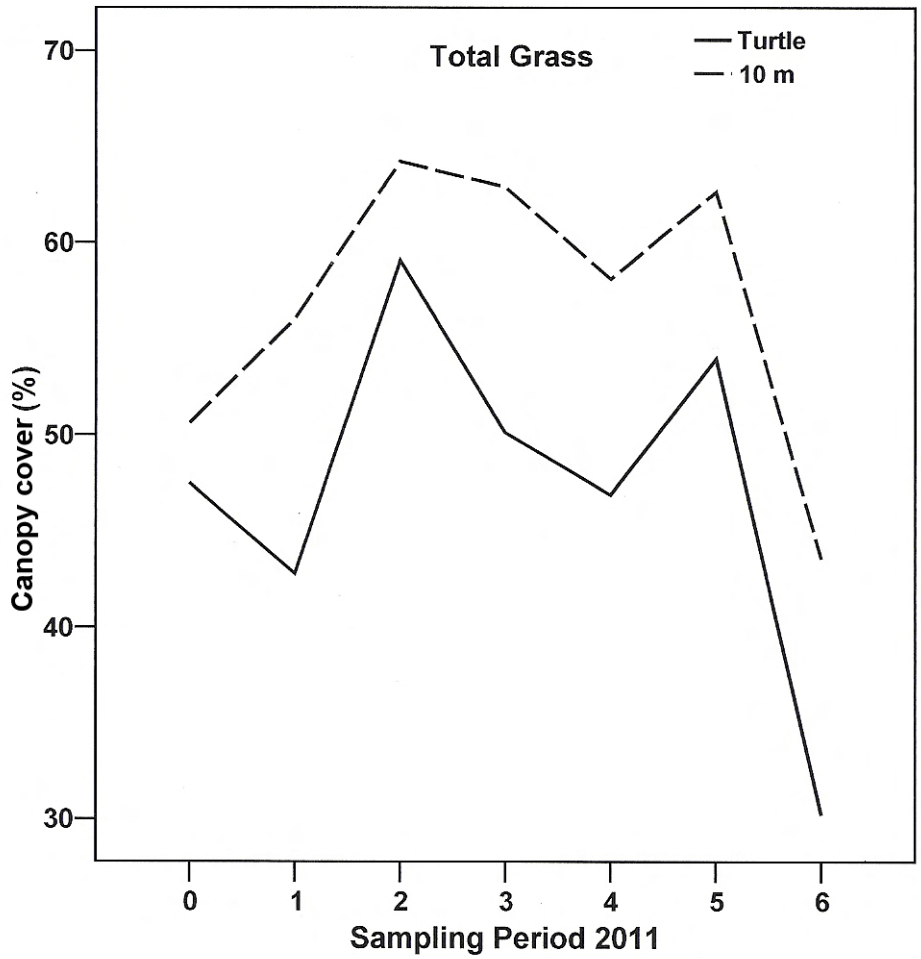

Figure 9. Canopy cover (%) for total grass comparing ornate box turtle (*Terrapene ornata*) preferences with habitat availability (10 m distance from turtle) during 2011.

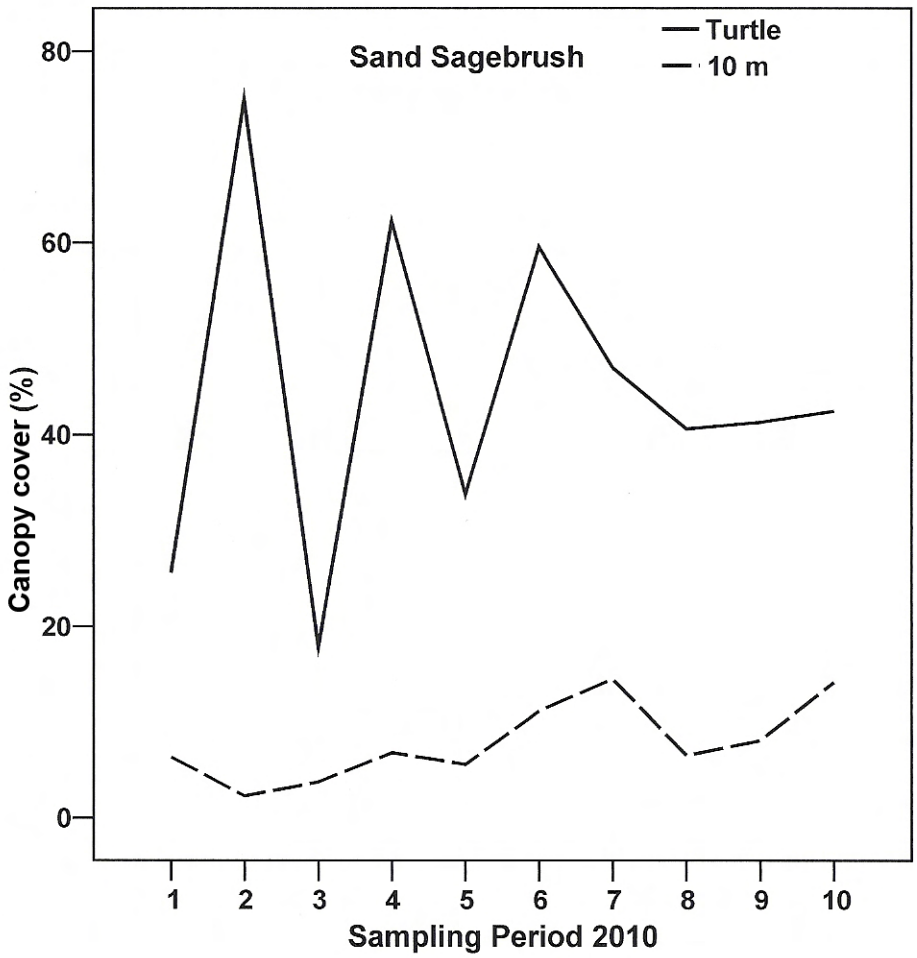

Figure 10. Canopy cover (%) for sand sagebrush comparing ornate box turtle (*Terrapene ornata*) preferences with habitat availability (10 m distance from turtle) during 2010.

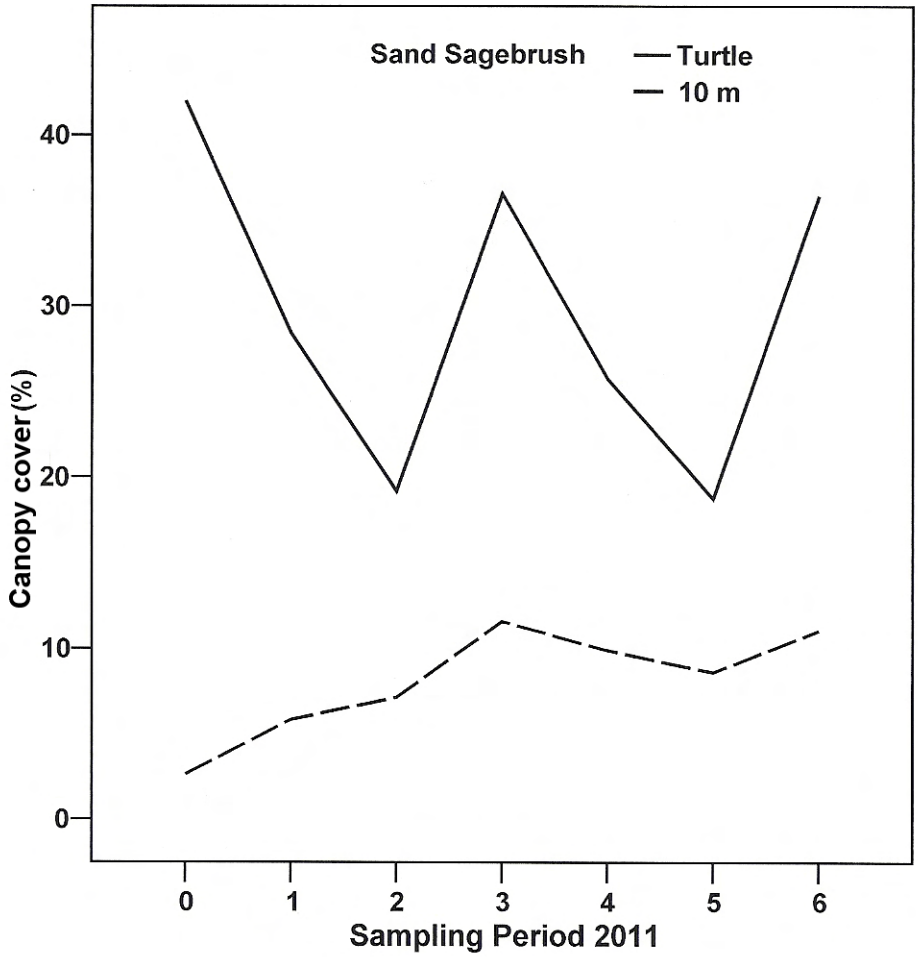

Figure 11. Canopy cover (%) for sand sagebrush comparing ornate box turtle (*Terrapene ornata*) preferences with habitat availability (10 m distance from turtle) during 2011.

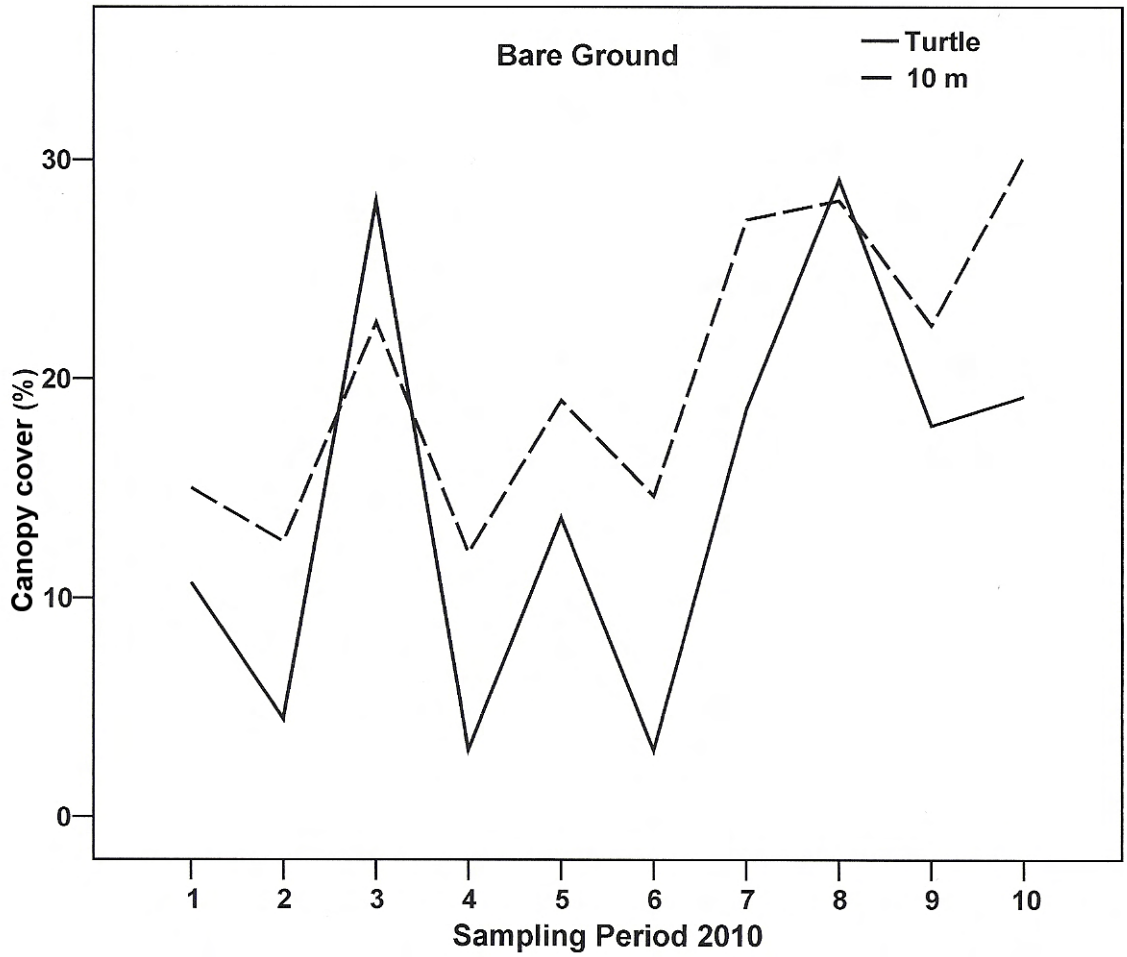

Figure 12. Canopy cover (%) for bare ground comparing ornate box turtle (*Terrapene ornata*) preferences with habitat availability (10 m distance from turtle) during 2010.

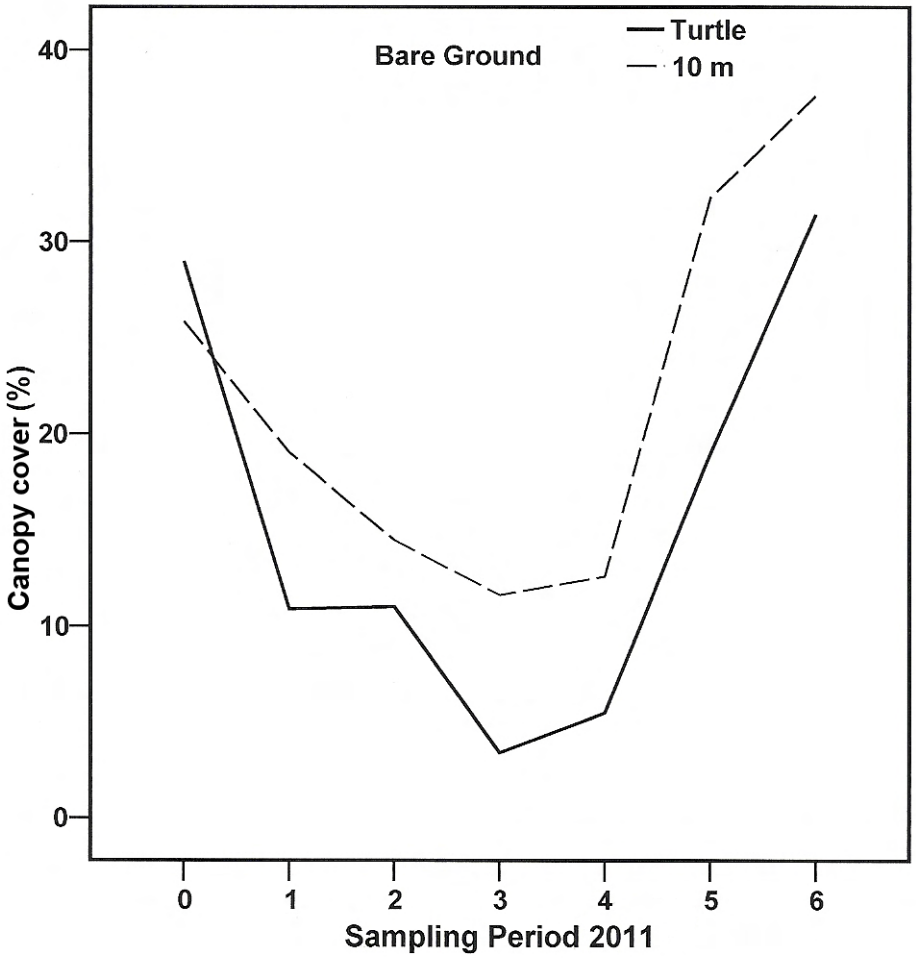

Figure 13. Canopy cover (%) for bare ground comparing ornate box turtle (*Terrapene ornata*) preferences with habitat availability (10 m distance from turtle) during 2011.

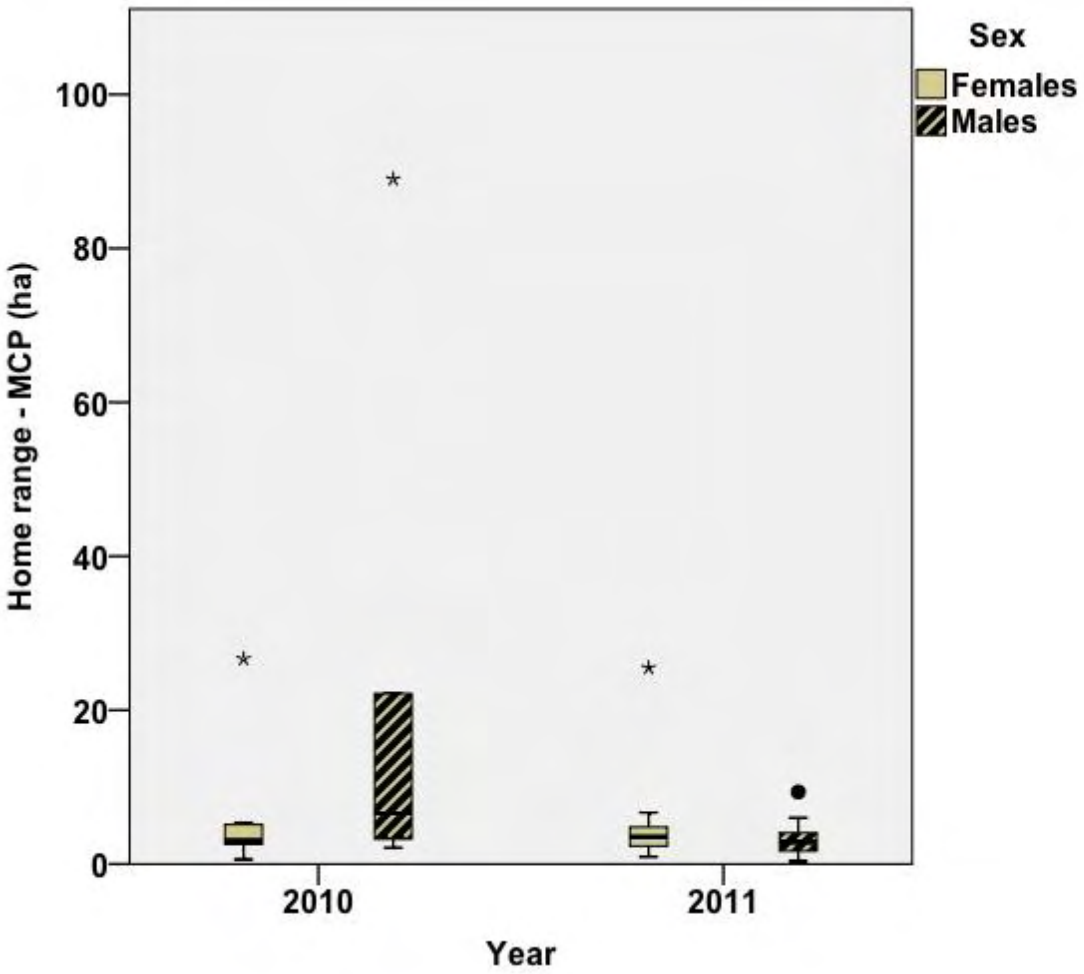

Figure 14. Box plot of the ornate box turtle (*Terrapene ornata*) home range medians calculated as Minimum Convex Polygons (MCP) in hectares. * Represents three extreme outliers both females and males in both years.

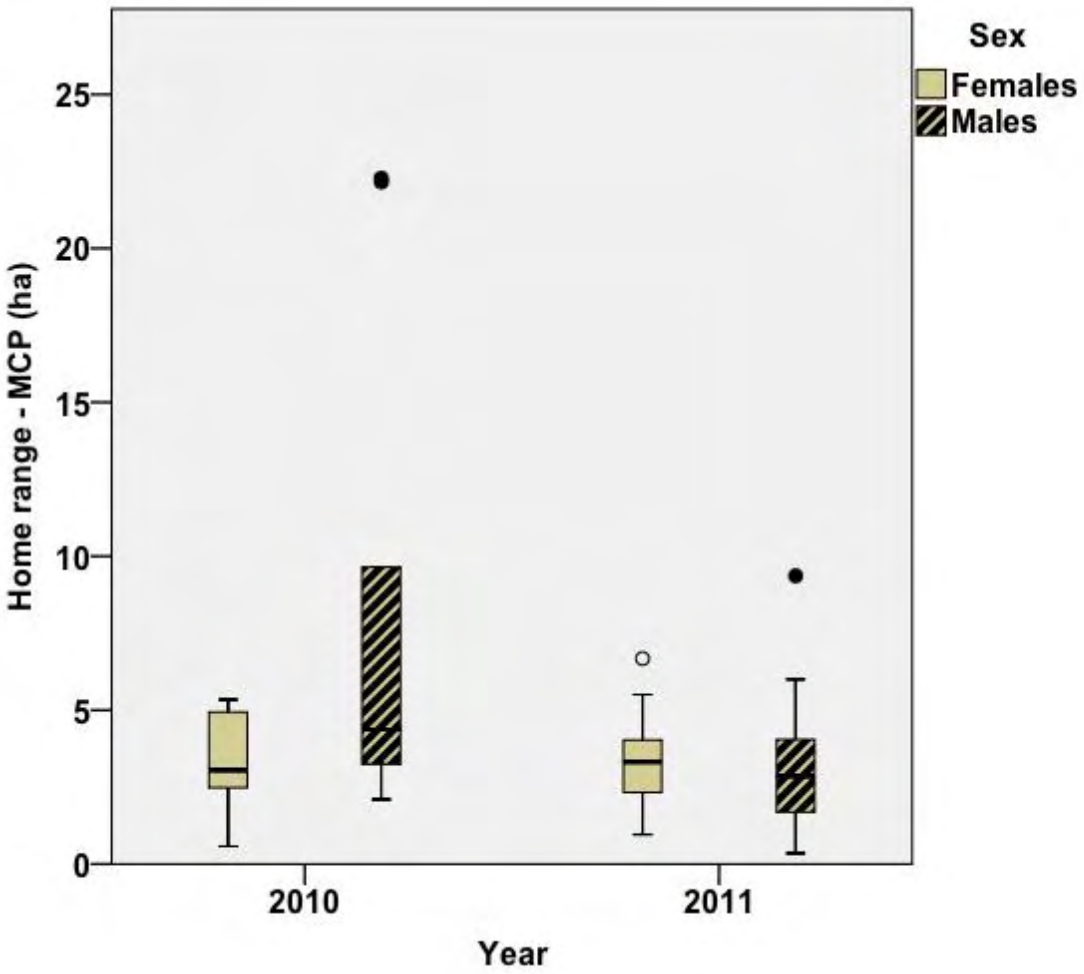

Figure 15. Box plot of the ornate box turtle (*Terrapene ornata*) home range medians calculated as Minimum Convex Polygons (MCP) in hectares without the three extreme outliers.

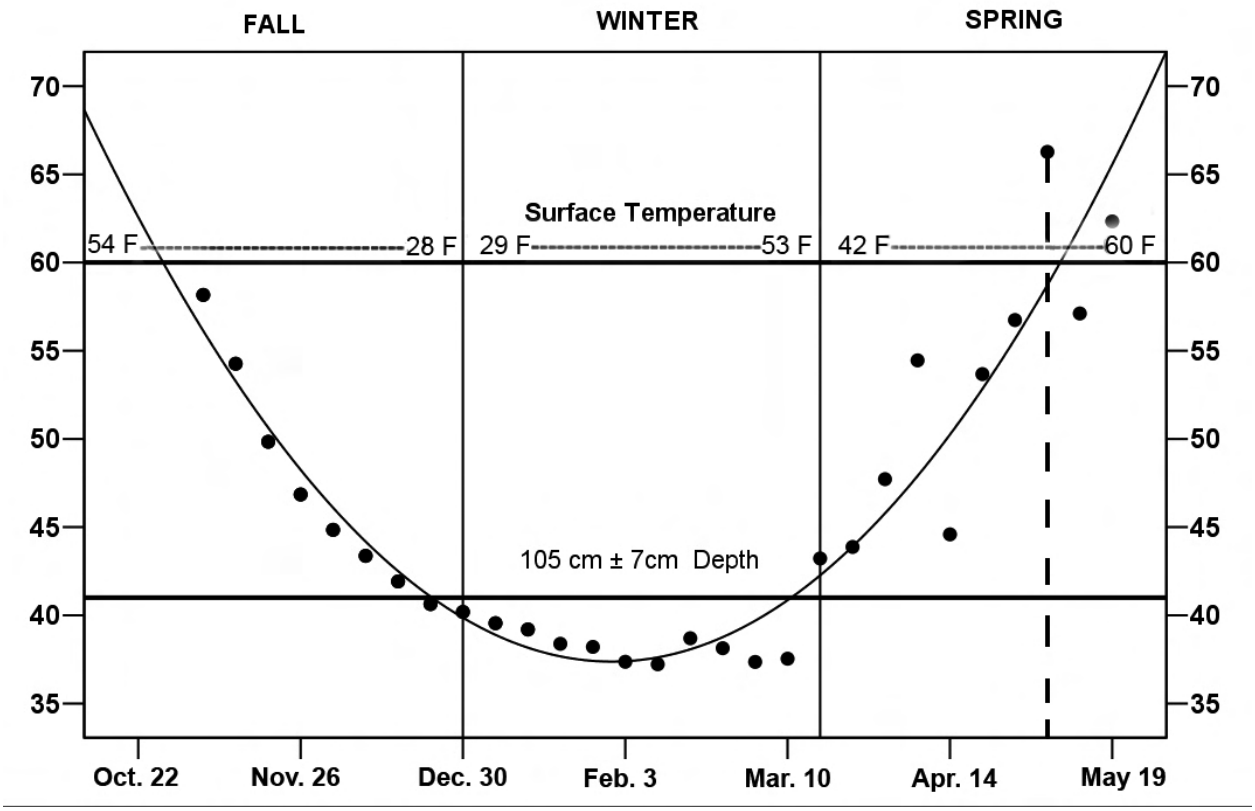

Figure 16. Quadratic curve describing the relationship of mean temperatures of ornate box turtles (*Terrapene ornata*) with week during hibernation. X-axis represents sample weeks (week 20 to week 50 in increments of 5) from 22 October 2010 to 19 May 2011. Y-axis represents temperature in ^{0}F. Dashed vertical line represents week 48 (early May), the estimated turtles emergence from hibernation in South Dakota.

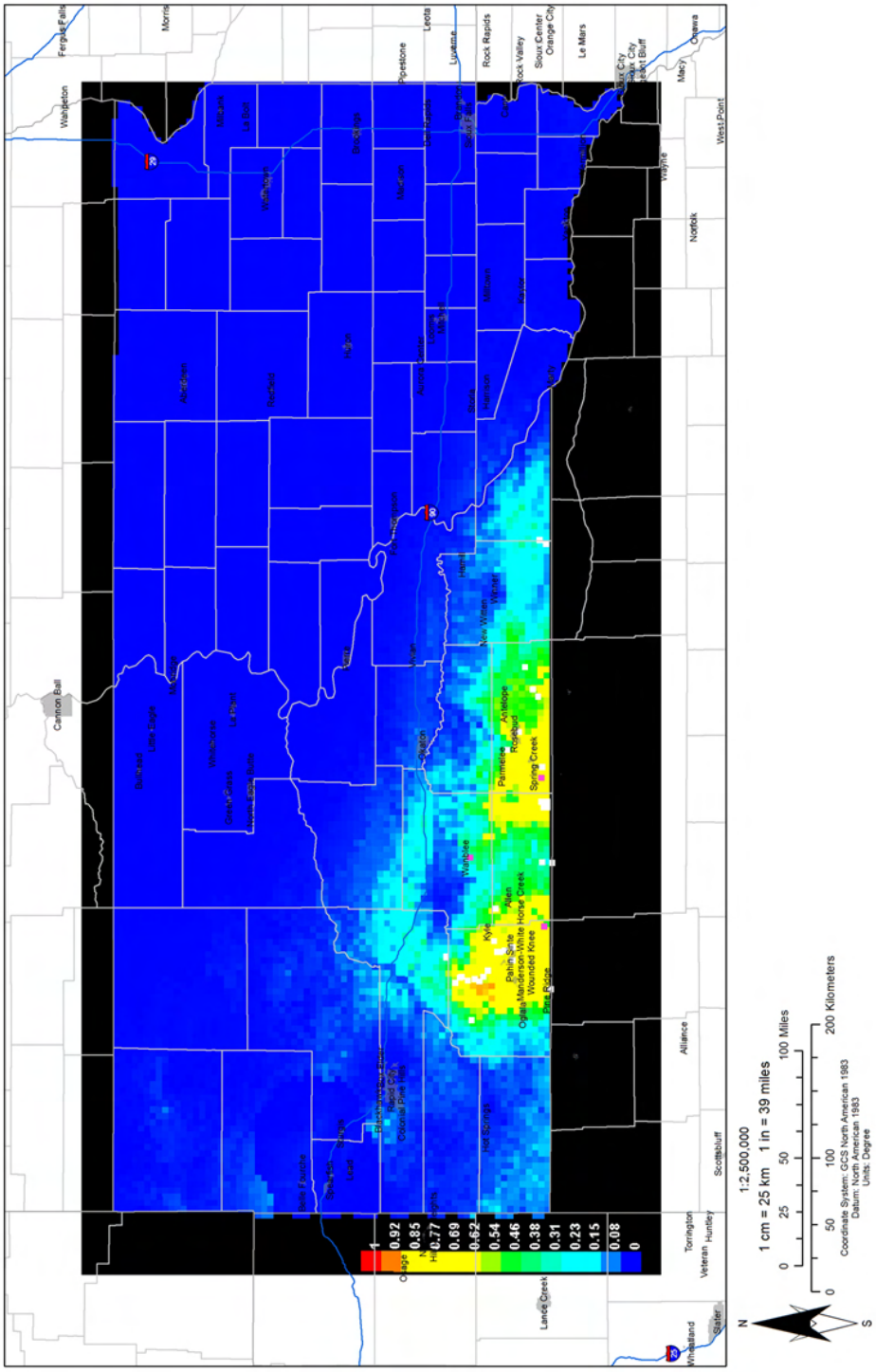

Figure 17. The maximum entropy distribution (MaxEnt) model for ornate box turtle (*Terrapene* ornata) in South Dakota. Warmer colors (0.54 - 1) represent the highest probability of species presence, predicting the most suitable habitats as the south west-central portion of the state.

Figure 18. Oglala Lakota College undergraduate students and instructor during the 2010 Field Ecology course. The box turtle project provided a unique hands-on and place-based scientific method instruction.

APPENDIX I

HAVE YOU SEEN ME?

Bottom View Top View

ORNATE BOX TURTLE

These turtles are found from Wisconsin into Mexico, but their whereabouts in South Dakota are not well known. If you've seen any, please let us know where and when, by e-mail or phone. If you happen to have a camera with you, please take a photograph. Thank you!

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