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Assessment of Painted Turtle Size and Age from Long-term Pond Study

Ellen Dolph
University of Nebraska Lincoln

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Assessment of Painted Turtle Size and Age from Long-term Pond Study

by

Ellen Dolph

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Ellen Dolph, B.S.

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Advisor: Dr. Larkin Powell

Abstract:

In recent years there has been a decline in herpetofauna, sparking an interest in inventory and monitoring of these declining species. Climate and drought affect population dynamics. Growth rates can be indicators of healthy individuals, populations or habitats (Moldowan et. al, 2015; Armstrong and Brooks, 2013), however, growth rates can be hard to obtain for long-lived species with individuals of unknown age, and long-term studies are rare. Mark-recapture sampling techniques can provide growth information from individuals of unknown age (Armstrong and Brooks, 2013). Though painted turtles (*Chrysemys picta*) are not currently a species of conservation concern and have stable populations, they are a good study species because they are common throughout the United States. The study area for this research is located at a single pond near Cedar Point Biological Station in Ogallala, Nebraska. My goal was to predict growth rates of painted turtles based on their ages through a long-term, single pond study using unknown age, mark re-capture methods. The study period included drought and non-drought periods. My objectives were to 1) fit a nonlinear growth model to my data to estimate growth rates, 2) use sex-specific models to compare growth rates of male and female painted turtle, and 3) determine impacts of climate on turtle growth rates by looking at drought years versus normal years. Turtles have been caught during the summer months for 12 years and measurements taken were

used to predict turtle growth rates. Results indicated that female painted turtles have a growth rate double that of males (male k=0.1122, female k=0.2269). Drought conditions also affected growth rate, causing a decrease in growth rate (male D=-0.0226, female D=-0.0393). This has implications for scientists studying climate change impacts on wildlife if drought conditions become the new normal summer conditions.

Introduction:

In recent years there has been a decline in herpetofauna species and popluations. This decline has sparked an interest in inventory and monitoring of these declining species because conservation efforts often rely on distribution and abundance information (Barela and Olson, 2014). According to Tingley et. al (2016), reptiles are the most species-rich group of terrestrial vertebrates, but there is a lack of research and understanding of their extinction risk. The IUCN has only assessed 45% of known reptile species (4648 out of 10,400). Out of the assessed species, 20% (945) are threatened with extinction and 19% (867) are data deficient (Tingley et. al, 2016). There are around 328 recognized turtle species in the world. Out of these 47.6% have been identified as threatened, with 27.4% of these identified as endangered or critically endangered (Barela and Olson, 2014). This is a higher percent than all other higher vertebrate groups, with amphibians at 41%, mammals at 25%, and birds at 13% (Barela and Olson, 2014) and highlights the need for further research and assessment of reptilian species (Tingley et. al, 2016).

One factor, among many, affecting herpetofauna population dynamics is climate and drought impact. Environmental factors have a large impact on both turtle populations and individual turtles by influencing reproduction and survival (Riley et. Al, 2014). The environmental conditions and amount of resources available at the time of reproduction, incubation, and then at the time of hatching can have a great impact on the vitality and growth of hatchlings. These factors can also create variability in adult growth (Packard and Packard, 2001).

Growth rates of animals are important because they provide insight into the health of a population and relate to habitat quality (Moldowan et. al, 2015; Armstrong and Brooks, 2013). Growth rates, however, can be hard to obtain for long-lived species with individuals of unknown age, and long-term studies are rare. Therefore, it could be useful to approach this problem using mark-recapture sampling techniques (Armstrong and Brooks, 2013), which can provide useful information from animals of unknown age that are caught repeatedly over time.

Though painted turtles (*Chrysemys picta*) are not currently a species of conservation concern and have stable populations, similar turtle species are declining and climate change could have implications on many different species (Barela and Olson, 2014). Improved growth information can be used to help with turtle management efforts in the United States. Painted turtles are a good study species because they are common throughout the United States and are a species of least concern.

Painted turtles are small pond turtles characterized by red markings on the outer edges of their marginal scutes and a notched upper jaw. They have a smooth olive to black colored carapace with yellow or red borders along the seams and a yellow plastron often with black, brown or red blotches. The carapace is the portion of the shell covering the back, while the plastron is the portion covering the belly. Coloring, especially darkness, may vary depending on habitat characteristics such as darkness of substrate. Their skin is olive to black colored with the neck, legs, and tail being striped with yellow and red. They have a very large native range, spanning across the United States and have been an interesting topic of study due to their wide range and ability to withstand freezing temperatures. Their distribution that naturally ranges across North America. The turtle can be found across southern Canada from New Brunswick to British Columbia and south to the United States, as far south as western Texas, New Mexico and Mexico in scattered populations (Ernst and Lovich, 2009).

Painted turtles are a semi-aquatic species requiring an aquatic environment, but with the ability to cross inhospitable land in order to move between bodies of water (Caldwell and Nams, 2006). Painted turtles prefer areas with slow-moving, shallow water with soft bottoms and aquatic vegetation. These areas include lakes, ponds, swamps, marshes, drainage ditches, creeks and other areas. The species can tolerate annual drying and polluted waters fairly well. As turtles transition from hatchlings to adults, they occupy different water depths. Hatchlings and small juveniles occupy shallow waters, but as they become adults, they move to deeper waters. Juveniles also tend to stay away from moving water habitats such as rivers and live in pond or marsh areas. Along with water, painted turtles also require plenty of basking sites (Ernst and Lovich, 2009).

Painted turtle male and females can be distinguished based on physical characteristics. Male painted turtles possess much longer foreclaws and have longer tails with the cloacal opening positioned outside of the shell. Female painted turtles have shorter claws and tails with the cloacal opening located at the base of the tail underneath the shell (Ernst and Lovich, 2009). Male painted turtles have lower growth rates than female painted turtles. In general, as a turtle ages, its growth rate slows. When an adult reaches full sexual maturity, growth rate changes depending on the sex of the turtle. An adult female painted turtle now must put more energy into egg production and development once or twice per year. This requires a female turtle to be larger, so growth rates of females will be faster than male turtles (Moldowan et. al, 2015).

For long-lived, long-growing species like the painted turtle, reproductive maturity is more dependent on size than age. Female painted turtles reach sexual maturity at around 13 cm plastron and 13.1 cm carapace length, which correlates to around 14 years according to a study of female painted turtles in Ontario (Moldowan et. al, 2015). Ernst and Lovich (2009) found female painted turtles reach sexual maturity between 6 and 10 years with plastron lengths between 9.7 and 12.8 centimeters, but can be much larger (16.5-17.7 cm) in some populations. Male painted turtles reach sexual maturity in 2 to 4 years at plastron lengths of 7 to 9.5 centimeters. Both female and male size and age of maturity are variable depending on environmental conditions, as well as latitude (Ernst and Lovich, 2009).

Climate is an important environmental factor that has potential to affect painted turtle reproduction, survival, and growth. Climate impact is especially important to consider as climate changes become more prevalent (Richard et. al, 2014). Turtles have temperature-dependent sex determination (TSD), which means that as embryos develop, the temperature of the embryo determines what sex it will be. Environmental conditions and nest placement have effects on TSD, but climate plays a role as well. It has been predicted that with projected directional environmental change, the production of extreme offspring sex ratios could occur and would lead to either reducing the temperature-dependence, altering maternal behavior in nesting, or changing of geographic ranges or extinction (Schwanz, 2010).

Climate also directly impacts yearly growth. Scute rings on turtles are formed each year, and although they are not accurate for determining age (Armstrong and Ronald, 2012), they can be useful in studying climate impacts on growth (Richard et. al, 2014). Richard et. al (2014) studied annual plastron growth increments on Blanding's turtles and found the width of growth increments varies each year and can provide information on ecological factors that drive overall growth.

Understanding this relationship between annual growth and climate can increase understanding of limiting and driving growth factors as well as predict effects of climate change on a species using projected climate models (Richard et. al, 2014). Drought can be used to assess climate effects in the short term. If drought conditions become the normal summer conditions with climate change (Dai, 2012), it could be beneficial to use drought years now, while they are more infrequent, to study effects on painted turtles that can be applied to climate change projection models.

This research is unique in that it is a long-term study with over ten years of painted turtle data from a single pond. Long term studies are rare, so not only is it a unique opportunity for me to work with this type of data, but it can provide different scientific information than short-term studies can provide. The long-term nature of the study, with uniquely-marked individuals that are recaptured through time, allows it to be used for finding growth relationships. My research is important to other scientists because it will provide new insight into painted turtle growth patterns and apply methods used in other areas, such as fisheries growth curves, to a new application. The data from the long term study will allow me to see how the population has changed and how individuals have responded over the years in the same, but fluctuating environment. For example, I can see how painted turtles respond to drought or wet conditions by studying the population size and structure. This could provide useful insight to how turtles respond to changes in weather and climate conditions and is especially important as our climates continue to change.

Until recently, the most commonly used method of age determination for painted turtles and turtles as a whole, was using scute rings. It was thought that the number of a turtle's growth rings on each individual scute of its shell indicated the turtle's age. This method of aging assumes that a turtle has one growth period every year, which is not necessarily the case. If resources are abundant, a turtle may have a lot of new growth, whereas if resources are lacking, a turtle may not grow as much (Armstrong and Ronald, 2012). Another problem with using growth increments as indicators of age is that the rings wear off of the scutes due to normal movements with age, making them hard to see (Richard et. al, 2014).

Instead of this scute aging method, a new method using linear regression to calculate age based on size is becoming more common (Armstrong and Ronald, 2012). This method is commonly used in fisheries science, but is now being applied to turtles. However, the fish are often of known age. Fish can be easily aged using different physical characteristics, such as their operculum. In fisheries science, the von Bertalanffy model is commonly used and modified to determine fish growth. This is a non-linear model based on a growth rate that decreases with age used to predict a future size based on current size (Shackell et. al, 1997). In another study a likelihood-based model was used and based on a non-Bayesian hierarchical model (Ruben, 2010). It is not as easy to determine a turtle's age, unless it has been known from birth, and in many research scenarios, it is not likely to have only known-age turtles. For this reason, it is important to develop another method that can be used for turtles of unknown age.

Mark – recapture is a common way to collect data on both individuals and populations of animals. In this method, an animal is caught, marked in some way and returned to the wild. The amount of marked animals that are captured again can help determine population size and survival statistics. Marked turtles are used to estimate a population size; age regressions are being developed for use with this method. Armstrong and Brooks (2012) used a similar regression to that used in fisheries science on both known and unknown age snapping turtles, but the original fisheries equation was modified to allow for individual and sexual variation. They found that a biphasic model, or model with two different parts, fit best for female turtles because female turtles change their growth pattern after becoming reproductively mature to a greater extent than males. As a turtle grows, it starts out growing linearly and as it reaches reproductive size, its growth slows (Armstrong and Brooks, 2012). Sung et. Al (2015) used a similar equation, as well as other logistic equations, on data from a nine year mark-recapture study on the endangered big-headed turtle in order to find the best age-predicting model. They found the von Bertalanffy model best described the growth rate and that counting rings on the turtles' scutes is not reliable or accurate (Sung et. al, 2015).

My goal was to predict growth rates of painted turtles based on their ages through a long-term, single pond study using unknown age, mark re-capture methods. The study period included drought and non-drought periods. My objectives were to 1) fit a nonlinear growth model to my data to estimate growth rates, 2) use sex-specific models to compare growth rates of male and female painted turtle, and 3) determine impacts of climate on turtle growth rates by looking at drought years versus normal years.

Methods:

In order to work with live animals, first a certification course to perform research on animal subjects had to be completed. I took IACUC training, which is training for safe and ethical treatment of animals in research. Once the training was completed, I could begin the study. We also obtained an IACUC permit for the project (IACUC permit number 1074).

Study Area:

The study area for this research is located at a single pond near Cedar Point Biological Station in Ogallala, Nebraska in western Nebraska, near Lake McConaughy and Lake Ogallala. This area is located in western high plains where the tall grass and short grass prairies meet, and is on the south edge of the Sandhills and the North Platte River valley. Cedar Point contains over 900 acres of land with prairie and redcedar-filled canyon habitats. The study pond was located on rangeland dominated by prairie habitat and used for cattle grazing. The pond is located next to a gravel road, which serves as a dam that keeps the pond full year-round. The water level in the center of the pond is two to three meters deep and is shallower along the edges.

Yellow Pond near Cedar Point Biological Station

Field Methods:

Painted turtle data has been collected at the pond by the school of Natural Resources at the University of Nebraska-Lincoln every summer since 2005, with the exception of 2009. In 2016, I spent five days collecting data at the study pond in the middle of July. Turtles were collected using two types of traps: basket traps and hoop nets. Ten mesh wire basket traps were suspended along the perimeter of the pond at regular space intervals. The traps work by floating in the water with a tether holding them to shore. The baskets have ramps up from the water that the turtles use for basking and then the turtles simply jump into the water in the middle of the trap and are caught in the basket (Singleton et. al, 2013). Three hoop nets were also used (Richard et. al, 2014). Hoop nets consist of netting strung over metal rings to create a cylindrical shape. The hoop net has a narrow entrance that turtles can enter through, but cannot exit. The nets were placed three quarters of the way under water to allow space for the turtles to surface and were baited with sardines. The traps were set every day and collected the next morning.

Once the traps were collected, the turtles were either identified from previous markings or marked using two methods. The first method used in marking was scute drilling. Each turtle received a unique combination of holes drilled into their marginal scutes (McGuire et. al, 2011). The second method was passive integrated transponder (PIT) tagging. The previously unmarked turtles were PIT tagged, which consists of being injected with a small metal tag containing a unique identification number into the skin on their anterior shoulder. The turtles were then measured for size and aged if they were two to four years old, and placed back into the pond. Seven measurements were taken on each turtle: carapace length, carapace width, plastron length, plastron width, bridge lengths, mass, and sex. Lengths and widths were measured using adjustable calipers. Hanging scales and mesh bags were used to measure mass. Sex of individuals was determined using physical characteristics including nail length, tail length and cloaca placement. Here, I use carapace length to model turtle growth between years.

Data Analysis:

After the data was collected, it was combined with all of the previous years' data into a database to make it easier to analyze. The turtle data was sorted by individual turtles and the turtles that were caught multiple years were chosen for the analysis. These turtles were sorted by date so that each capture was a separate event. One capture for each year of capture was chosen (repeated captures within the year were ignored). Critical data were: gender, size at beginning of capture interval, size at end of capture interval, and length of time between captures. We used a non-linear regression model to estimate coefficients in a growth equation that could be used to estimate size of a turtle in a subsequent time period, based on current size. We used Von

Bertalanffy growth models to estimate growth rate (k), asymptotic size (L_∞), and a drought effect, D (adjustment to k, if capture interval period included a drought) for males and females separately:

$$L_R = [L_\infty - L_C] \times \left[1 - e^{-(k-D)t}\right]$$

Separate analyses were then performed for males and females. As research by Shakell et. al (1997) indicated, male and female turtles have different postmaturational growth rates and turtle growth could be more accurately explained by incorporating female variability into the von Bertalanffy equation (Shackell et. al 1997). Instead of altering the equation to make up for differences between male and female postmaturational growth, I used two separate sex-specific analyses to determine differences in growth rate. However, I did modify the Von Bertalanffy Model was to include a drought effect (D) and used it to find asymptotic size (L_∞), L_c, and drought effect (D) for male and female turtles.

Finally, I performed an analysis of known annual growth as a second assessment of climate's effects on growth. The Palmer Hydrologic Drought Severity Index was used to determine years with drought conditions between 2005 and 2016: 2006, 2012, 2013, and 2014. Turtles that were captured both during and after each year were considered drought turtles and turtles captured and recaptured in years spanning non-drought years were considered non-drought turtles. The capture years were noted for each turtle. Male and female turtles were considered separately. I estimated G, the average length of annual growth, for drought and non-drought years. I also used a linear regression to determine if the year-specific PHDI was related to the length of growth (G) for turtles in that year.

I then used my growth model's estimates to create growth curves for male and female turtles. Empirical data from the study showed that two-year-old turtles had a carapace length of

50 cm, which I used to start the growth curve. I then applied my equation to predict the length of males and females after 1 year, from 2 to 40 years. I then used drought-specific (D) parameters to create growth curves for turtles living their entire life under drought or non-drought conditions.

I validated my growth model using individual turtles set aside from the analyses. I calculated the error for each turtle's predicted size from its actual size and compared these errors to the turtle's actual size to find how accurately my model predicted size.

Results:

We used data from 98 female turtles with 312 capture intervals and 64 males with 194 capture intervals in our growth model. Female size ranged from 68 to 190 mm and males ranged in size from 85 to 186 mm.

The Von Bertalanffy Model predicted females (L_{∞}=178.3 mm) grow to larger size than males (L_{∞} 166.9 mm). Male and female painted turtles also had significant differences in growth rate (male k=0.1122, female k=0.2269) (see Table 1).

Our analyses show the impact of drought on growth of males and females using classifications of drought years from the continuous Palmer Hydrologic Drought Index. Our results suggest that drought conditions significantly impact growth rates for both male and female painted turtles (male D=-0.0226, female D=-0.0393).

However, the effect on females, when assessing annual growth (G) appears more severe. Annual growth (G) for males was less than females and drought lowered annual growth (G) of females more than for males.

The validation test showed a larger margin of error for female size prediction than for male size prediction (Figure 3).

	Male	SE	CI		Female	SE	CI	
k	0.1278	0.0115	0.1051	0.1505	0.1977	0.0331	0.1321	0.2633
L∞	166.9	2.2835	162.4	171.5	193.9	9.3994	175.3	212.5
D	-0.0226	0.0108	-0.0438	-0.00134	-0.0393	0.0169	-0.0727	-0.00585
G (drought)	5.25	0.403	4.25	6.25	11.5	1.03	10.3	12.7
G (nondrought)	6.61	0.65	5.21	8.01	15	0.61	13.4	16.6

Table 1: results of Von Bertalanffy model of male and female painted turtles in drought and nondrought years where k is growth rate, L∞ is asymptotic size, D is drought effect and G is annual growth increment. Also shown are standard errors and 95% confidence intervals for each parameter.

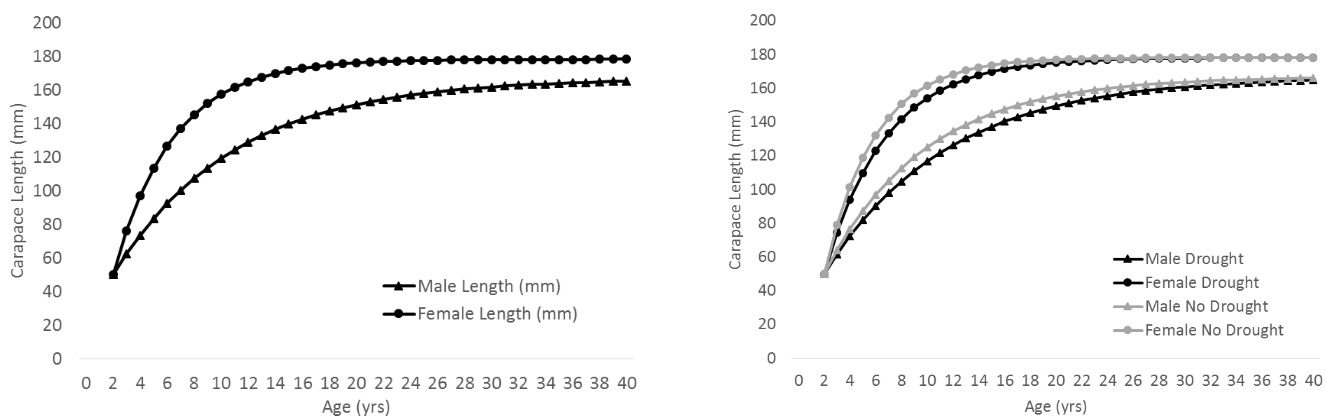

Figure 1. Left: overall growth of male and female painted turtles during 2005-2016 Ogallala, NE, given known size-at-age of 2-year-old turtles (50mm) using parameters estimated from captured and recaptured individuals. Right, growth of male and female painted turtles when incorporating the drought effect (D) assuming ‘drought’ turtles grew in constant drought conditions.

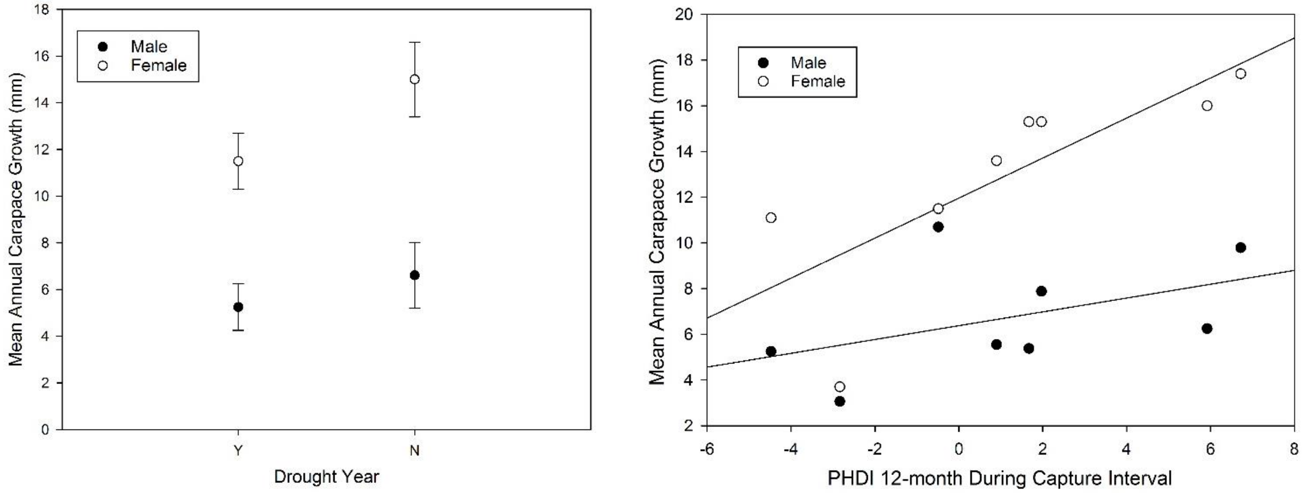

Figure 2. Left: comparison of annual carapace growth, G, for male and female painted turtles when growth occurred during a drought or non-drought year. Right: relationship, for males and females, between the Palmer Hydrologic Drought Index during annual capture intervals and mean annual carapace growth, G.

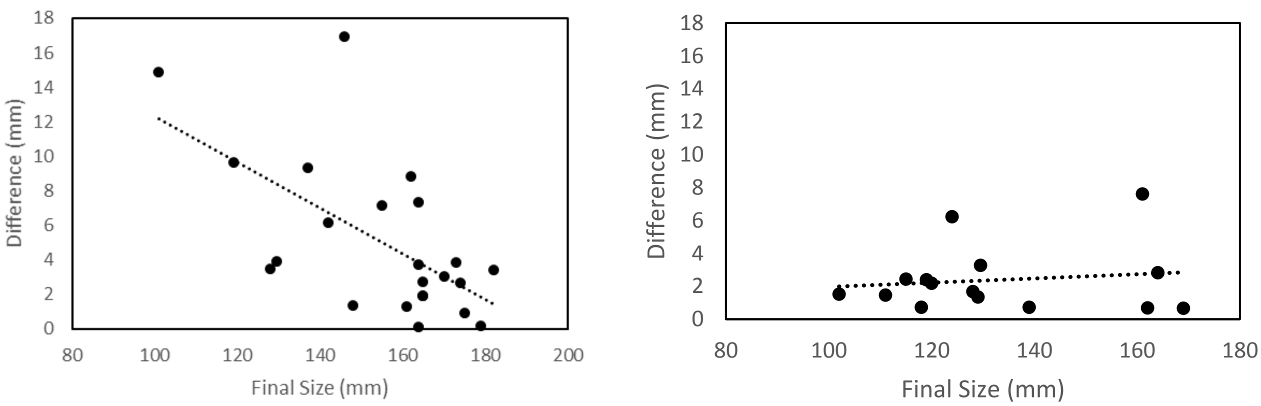

Figure 3. The comparison of the difference between actual final size and predicted final size from the Von Bertalanffy model of painted turtles compared to the final size for females (left) and males (right) using 15 females and 14 males. Male turtles show no difference in actual and predicted final size. Female turtles have more variability in error size and show more error when small because of their faster growth rate.

Discussion:

Our model was successfully able to predict the age of unknown-age painted turtles at our study site. The Von Bertalanffy Model used is commonly used to age fish, but has not been applied to many other species, especially ones of unknown-age. Mark-recapture methods are commonly used in wildlife science and having models that can predict based on this could be useful (Armstrong and Brooks, 2013).

The Von Bertalanffy Model predicted females grow to larger size than males. Our predictions of asymptotic carapace size for females (178.3 mm) and males (166.9 mm) were averages and not maxima. According to literature, midland painted turtles reach a maximum size of 19.5 cm (195 mm) straight carapace length (Ernst and Lovich, 2009). Although for the species as a whole, females can reach a straight carapace length of 254 mm, while males have a max of 153 mm (Ernst and Lovich, 2009). Our largest female turtle had a carapace length of 190 mm, while our largest male had a length of 186 mm. These are close to both the predicted asymptotic carapace sizes and the sizes given in the literature for midland painted turtles.

My analysis conferred that male and female painted turtles have significant differences in growth rate. Female turtles grow faster than males. Female turtles put energy into growth early, to arrive at the size needed for sexual maturity, and growth rate then slows as adult females put their energy into egg production and development (Moldowan et. al, 2015). Therefore my analyses show that female painted turtles have both a larger asymptotic size, and double the growth rate of males.

Our model is consistent with other findings of painted turtle size at sexual maturity. We can predict that a female between the sizes of 120 and 140 mm (Figure 1), the size of sexual maturity determined by Ernst and Lovich (2009), would be within the ages of 5 and 7 years, which also coincides with the age range (6-12 years) given by Ernst and Lovich. Our model predicts male painted turtles within the size range of 70 and 95 mm would be 3 to 5 years old, also consistent with previous findings (Ernst and Lovich, 2009). The drought model predicted both female (6-8 years) and male painted turtles (3-6 years) to reach sexual maturity at older ages. This could have implications for painted turtle populations if drought becomes a more common occurrence. We speculate that hotter and drier conditions of drought years cause shortages of food resources for turtles. In addition, warmer temperatures are known to increase metabolic rates of animals and this coupled with food shortage can cause slower growth rates. Our model, however, assumes that every year is a drought year, which is not usually the case for the entirety of a turtle's life cycle, but is still useful in understanding potential impacts of drought on painted turtle growth and related factors.

Our analysis of one-year growth intervals during drought and non-drought years validated the results from our mark-recapture model. Our analyses also show the impact of drought on growth of males and females using classifications of drought years from the continuous Palmer Hydrologic Drought Index. Our results suggest that drought conditions impact growth rates for both male and female painted turtles. However, the effect on females, when assessing annual growth (G) appears more severe (Figure 2).

According to climate change predictions, many areas within the Great Plains Region are expected to receive more total rainfall, however, rain during the summer months is expected to decrease, causing an increase in severe droughts (Dai, 2012), (Kunkel, 2013). This means droughts could become the new normal summer conditions. If food resources were limited by changing climate conditions our results indicate that extensive drought would cause decreases in painted turtle growth rate, as well as push back the age of sexual maturity. This could cause turtles to reproduce later in their lives, which would have effects on their population size and growth.

Alternatively, climate change is predicted to move habitat zones northward. If sufficient food resources exist, it could cause more northern turtles to begin maturing more rapidly and at smaller sizes because of a longer growing season, which could also have population implications, increasing population size and growth more rapidly. Since painted turtles have temperature sex determination, hotter and drier summer months, which is the reproductive season, could have impacts on the population sex ratios as well (Schwanz, 2010).

Our study is representative for painted turtles in our study pond. It would have been useful to study turtles in multiple areas to compare growth rates and asymptotic sizes. In addition, we only used one measurement (carapace length) and it could be interesting to use other measurements, such as plastron length or width, in our growth model to compare results.

Conclusion and Implications:

Painted turtles are slow-growing species, and it is difficult to age individuals at time of capture. Our growth curves allow predictions of age from known-size turtles, which will be useful to analyses of population structure.

Growth rates differed significantly between male and female turtles, as well as between years with drought conditions. Our results may help biologists understand the effects of changing climate on turtles.

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