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# Dorsal and Ventral Color Patterns in a South Georgia Population of Agkistrodon piscivorus contanti, the Florida Cottonmouth

#### **Cover Page Footnote**

As the editor-in-chief of the Georgia Journal of Science, upon submission of this manuscript, I, David L. Bechler, have recused myself of all aspects of the review process and assigned the associate editor all aspects of the review process to include acceptance or rejection.

#### DORSAL AND VENTRAL COLOR PATTERNS IN A SOUTH GEORGIA POPULATION OF Agkistrodon piscivorus conanti, THE FLORIDA COTTONMOUTH

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#### ABSTRACT

We examined dorsal pigments and ventral patterns in the Florida Cottonmouth, Agkistrodon piscivorus conanti, in the Alapahoochee watershed, Lowndes County, Georgia. Cottonmouths darken as they age; but the process has not been quantified in the literature. Thus, we examined both graphically and statistically changes in dorsal color pattern that occurs when snout vent length (SVL) increases as well as discrete patterns involving splotching and block-like patterns, and cream to white coloration on the ventral surface, which indicate underlying genetic factors. Snakes with SVLs between approximately 26.8 and 120.3 cm possessed an array of dorsal colors involving white, tan, dark brown and black. Snakes greater than 60 cm SVL had fewer dorsal white and tan colors with dark brown and black being the primary remaining colors in snakes up to 120 cm. Nonparametric regression analysis provided graphic representation of the process, which is confirmed by correlation analyses. Ventral color patterns show discrete relationships involving the occurrence of all white coloration and splotch and block patterns involving dark pigments. If a block pattern was present, then a splotch pattern was less likely to be present and vice versa regardless of SVL. Correlation analysis supports the observed ventral patterns. Possible genetic explanations would be a single locus with incomplete dominance expressed by one allele resulting in all white or no dark blocked pattern, another allele resulting in incomplete dark bars, and heterozygotes showing only partial bars or blotches primarily on the rear location anterior to the vent.

*Keywords:* water moccasin, color pattern, dorsal and ventral, genetics, age related

#### **INTRODUCTION**

The Alapahoochee River basin lies within the coastal plain of Georgia and is composed primarily of flatwoods habitats and other associated subhabitats (Edwards et al. 2013). The basin has been the location of seven recent studies on fish, crayfish, and a management survey (Bechler 2006; Chaney and Bechler 2006; Barnett et al. 2007; Hightower and Bechler 2012; Bechler and Salter 2013; Bechler et al. 2014) with this study being an extension of the work of Kirkley (2014). Variations in snake color patterns are well documented in the literature and have been examined from a variety of aspects such as a function of age or snout vent length (SVL), subspecies differences, genetics, and behavioral correlations. Wright and Wright (1957), Bechtel (1978), and Conant and Collins (1998) identify multiple species that experience color changes as their snout vent length (SVL) increases (Appendix 1). Genetically related studies on color and patterning are fairly numerous. Bechtel (1978) reviewed colors and patterns of multiple species and found strong relationships involving genetic control. Studies on genetic factors influencing individual species and populations provide valuable information and cover a period of 32 years. A review of these studies is provided in appendix 2.

Other studies involving color variation in snakes provide useful information on the roles of pattern and color variation. Bittner et al. (2002) examined the relationship of color pattern and body size, and concluded that large melanistic females may gain an advantage over males and smaller cohorts in higher body temperatures for breeding. Hayakawa et al. (2011) studied how color (gray image versus color image) influences on how a child is able to detect a snake in a matrix of images. Dunn (1954) and Greene and McDiarmid (1981) confirmed the hypothesis that other species of snakes mimic venomous species thus potentially providing them protection. Creer (2005) observed variations in antipredator behavioral patterns as they related to the color patterns of individual racers, *Coluber constrictor*.

An examination of the literature and web locations on color and color pattern change in *Agkistrodon piscivorus* as a function of aging or SVL resulted in only references to the relationship of increasing darkness of the dorsum with respect to SVL. Gloyd and Conant (1990), Conant and Collins (1998, 401), and the USGS (2017) web location, which references the aforementioned authors, notes colors associated with *A. piscivorus* involve olive to brown, black, and white. Cross band patterns range from distinct to obscure to absent. The same authors describe three subspecies noting color differences associated with each of them. Behler and King (1979, 684) and Conant and Collins (1998) in their description of *A. piscivorus* only describe the general pattern as SVL increases. Zaidan (2001) notes that variation in the Western Cottonmouth, *A. piscivorus leucostoma*, in northwestern Arkansas involves both sexual dimorphism and dichromatism (assessed statistically), and specifically states that melanism increases with age and that females darken at smaller SVLs. He attributes earlier changes to darker colors in females to possible thermal regulation with the possible need for gravid females to elevate body temperature.

We examined dorsal and ventral coloration to establish patterns in the Florida Cottonmouth, *A. piscivorus conanti*. Specific objectives of the study were to develop tabular, graphic, and statistical analyses that describe in detail color patterns and color pattern changes, and assess possible genetic structure related to ventral color patterns.

#### Study Area

#### **MATERIALS & METHODS**

Specimens of *Agkistrodon piscivorus conanti* were collected in the Alapahoochee watershed, which is composed of Grand Bay Creek (GBC), Mud Creek (MC), and Knight's Creek, a tributary of Mud Creek. The creeks converge to form the Alapahoochee River, which drains portions of Lanier, Echols, and Lowndes Counties, Georgia (Barnett et al. 2007). Near the confluence of Grand Bay and Mud Creeks, the habitat shifts from

flatwoods (Edwards et al. 2013, 424) to a mix of woodlands dissected by narrow ravines cutting into Pleistocene limestone rock. In this and other studies of the basin, which covered a 15-year period, no Florida Cottonmouths were seen or collected south of the confluence of the two creeks (Bechler 2006; Chaney and Bechler 2006; Barnett et al. 2007; Hightower and Bechler 2013; Kirkley 2014; Bechler et al. 2014).

#### Protocol

Snakes were collected in 2008 and 2009 under Georgia DNR collecting permit number 1934. Euthanizing was carried out based on the American Society of Ichthyologists and Herpetologists's guidelines (ASIH Guidelines 2004) in which, upon capture, the head of the snake was crushed. As such, snout vent length measurements are not exact due to damage to the head of the snake, but are considered reasonably accurate since only the head was impacted.

In the laboratory, visual observations and photographs of intact skins were taken at the following fixed dorsal and ventral locations: anterior (A), body posterior to the neck, middle (M) or center of the body based on snout vent length (SVL), and posterior (P) position anterior to the vent with each picture covering five ventral scales in length. Dorsal images (Figure 1) were equal in length to five scales lengths involved in ventral images (Figure 2). Sex and SVL were recorded with gender determined by dissection.

Statistical tests (Smirnov test, Friedman test, Kruskal-Wallis pairwise comparisons test, and nonparametric regression) were computed with StatsDirect (2014). Each dorsal and ventral location was assessed as to the presence of the colors tan (T), brown (R), black (B), and white (W). Ventral patterns of blocked (B) and splotched (S) were assessed as well as the absence of a pattern. Dorsal banding pattern, which is common in *A. piscivorus*, was not assessed.



*Figure 1.* Dorsal colors. White boxes with letters identify each picture or images. White and tans (A, B) to browns (B, C) and black (D).



*Figure 2.* Ventral patterning. White boxes with letters identify each picture or images. A. No dark pattern except for injury marks. B. Slight pattern. C. Splotched. D. Splotched. E and F. Blocked pattern. Yellow to orange colors were associated with apparent aged ectoderm skins.

#### RESULTS

A total of 28 snakes were used for the analyses, 14 females and 13 males, with the gender of one snake not determinable. Since genetic analysis (Kirkley 2014) implied possible differences between Grand Bay Creek and Mud Creek, specimens were statically compared. Snakes from each creek were ranked by SVL and the occurrence of dorsal colors described below (Table I) were tested to determine if snakes from each creek possessed statistically different dorsal patterns (Figure 1). A 2-sample Smirnov test (a version of Kolmogorov-Smirnov test, StatsDirect, 2014) for distribution differences was not significant for dorsal color (D = 0.426, P = 0.116). To analyze ventral patterns, a Kruskal-Wallis pairwise comparison (Dwass-Steel-Chritchlow-Fligner method, StatsDirect, 2014) test was conducted to compare blocked, splotched, and all white patterns between Grand Bay and Mud Creek snakes. All three comparisons were nonsignificant (black blocked pattern, P = 0.9232; white only, P = 0.9232; posterior splotched, P = 0.9867) indicating that ventral patterns were equivalent between the snakes in each creek. A Mann-Whitney U test, which compares the distributions of median differences, was used to test differences between males' and females' SVL to determine potential influences on coloration patterns between males and females. Female median length was 76.65 cm and male median length was 78.40 cm with a lower side P =0.453 (indicates differences between medians), which was not significant, but showed that females involved in the study tended to have shorter SVL than males. Since dorsal and ventral colors and patterns and median SVL differences were not significant, males and females were combined for all analyses.

**Table I.** Dorsal color analysis. Cells in which color was present are highlighted in gray. SVL, as measured for each snake, is arranged from shortest to longest. Abbreviations are as follows: A = anterior, M = middle, P = posterior, B = black, R = brown, T = tan, W = white, 1 = present, o = absent. A question mark (?) indicates that gender was not identified.

SVL (cm)	Sex	A B	M B	P B	A R	M R	P R	A T	M T	P T	A W	M W	P W
26.8	F	1	1	1	1	1	1	1	1	1	1	1	1
35.9	F	1	0	1	1	1	1	1	1	1	1	1	1
40.0	F	1	1	1	1	1	1	1	1	1	0	0	0
43.2	F	1	1	1	1	1	1	0	0	1	1	0	0
46.7	?	1	1	1	1	1	1	1	1	0	1	1	1
51.3	Μ	1	1	1	1	1	1	1	1	1	1	1	0
58.3	Μ	1	1	1	1	1	1	1	1	1	1	0	0
58.4	M	1	1	1	1	1	1	1	1	0	0	0	0
60.8	Μ	1	1	1	1	1	0	0	0	0	0	0	0
61.8	Μ	1	1	1	1	1	1	1	1	1	0	0	0
62.4	Μ	1	1	1	1	1	0	0	0	0	0	0	0
65.0	F	1	1	1	1	1	0	1	1	0	0	0	0
69.5	F	1	1	1	0	1	0	0	0	0	0	0	0
75.3	F	1	1	1	1	1	1	0	0	0	0	0	0
78.0	F	1	1	1	1	1	1	0	0	0	0	0	0
78.1	F	1	1	1	1	1	0	1	1	1	0	0	0
78.4	Μ	1	1	1	1	1	1	0	0	0	0	0	0
79.2	Μ	1	1	1	1	1	0	1	0	0	0	0	0
79.6	M	1	1	1	1	1	0	0	1	1	0	0	0
79.8	Μ	1	1	1	1	1	0	0	0	0	0	0	0
81.7	Μ	1	1	1	1	1	1	0	0	0	0	0	0
84.2	F	1	1	1	1	1	1	1	0	0	0	0	0
84.4	Μ	1	1	1	1	1	0	0	0	0	0	0	0
85.0	F	1	1	1	0	1	1	0	0	0	0	0	0
85.3	F	1	1	1	1	1	0	0	0	0	0	0	0
85.5	F	1	1	1	1	0	0	0	0	0	0	0	0
86.8	F	1	1	1	1	1	0	0	0	0	0	1	0
120.3	Μ	1	1	1	1	0	0	0	0	0	0	0	0

Dorsal colors (Figures 1 and 2) were whites (W), tans (T), browns (R), and variations of dark gray to black (B) with all grays to black treated as only black. An influencing factor on the expression of colors observed (grays to black) was time since the last shedding or ecdysis of the skin with older skins appearing duller and grayer. Otherwise, newer skins expressed black. Each color was scored as present or absent at each body location. Scores were added together for each snake at each location and compared to SVL in tabular and graphic formats (Table I, Figure 3). Snakes with SVL > 60 cm had dark brown and black colors more prominent and white or tan colors less commonly present. A nonparametric linear regression (Figure 3, total number of colors present at all locations versus SVL) was significant (two-sided P < 0.0001). The total number of colors significantly decreased as SVL increased.



*Figure 3.* Nonparametric linear regression. Total number or sum of dorsal colors at each location (anterior, middle, posterior) are regressed against snout vent length.

The venter consisted of black and white colors only with grays and orange to yellow as a function of approaching ecdysis (Figure 2B and E). Ventral color was analyzed based on the presence or absence of two basic patterns and no pattern or only white scales present (Table I). The blocked pattern (B) was surrounded by white and often with distinct sharp-edged square-like to oblong images in close proximity to each other (adjoining ventral scales) so they formed a complex figure (Figure 2E and F). Splotched patterns (S) were surround by white and consisted of irregular marks of dark pigment interspersed with white and often in close proximity to each other (adjoining ventral scales) forming a complex figure (Figure 2C and D). Blocked and splotched patterns would occasionally show some mixing at anterior and middle locations, but the blocked pattern dominated and was thus analyzed as blocked pattern. Blocked patterns were most common (Figure 1A and B) and occurred at all locations with an occurrence rate of 50 of the total 84 body locations (28 snakes X 3 locations/snake) on the 28 snakes. Splotched pattern, not counting those mixed into the block pattern, was the next most common occurring at 19 locations and occurred at only posterior locations. Least common was all white, which occurred at 15 of the 84 locations. As such, blocked and splotched pattern were the most prevalent (Table III) and white only at locations was least prevalent.

Blocked and splotched patterning at all three locations occurred on 20 snakes accounting for 71.43% of all snakes with the remaining eight snakes (28.57%) lacking a dark pattern on at least one of the three locations. Blocked, splotched, and white occurred at posterior locations (Table II). All white coloration was observed at all three locations on two snakes. White only at the anterior and middle locations with splotched pattern at the posterior location was present on three specimens.

**Table II.** Ventral pattern analysis. Cells in which a pattern was present are highlighted in gray. SVL is arranged from shortest to longest. Abbreviations are as follows: A= anterior, M = middle, P = posterior, B = blocked, W = white, S = splotched, 1 = present, o = absent. A question mark (?) indicates that gender was not identified. Sum = number of locations with color.

<u> </u>											
SVL (cm)	Sex	AB	MB	PB	AW	MW	PW	AS	MS	PS	Sum
26.8	F	1	1	1	0	0	0	0	0	0	3
35.9	F	1	1	0	0	0	0	0	0	1	3
40.0	F	1	1	0	0	0	0	0	0	1	3
43.2	F	0	0	0	1	1	0	0	0	1	1
46.7	?	0	0	0	1	1	0	0	0	1	1
51.3	Μ	1	1	1	0	0	0	0	0	0	3
58.3	Μ	1	0	0	0	1	0	0	0	1	2
58.4	Μ	1	1	0	0	0	0	0	0	1	3
60.8	Μ	1	1	0	0	0	0	0	0	1	3
61.8	Μ	1	1	1	0	0	0	0	0	0	3
62.4	Μ	1	1	0	0	0	0	0	0	1	3
65.0	F	1	1	1	0	0	0	0	0	0	3
69.5	F	1	1	0	0	0	0	0	0	1	3
75.3	F	1	1	1	0	0	0	0	0	0	3
78.0	F	1	1	0	0	0	0	0	0	1	3
78.1	F	1	1	0	0	0	0	0	0	1	3
78.4	Μ	1	1	0	0	0	0	0	0	1	3
79.2	Μ	1	0	0	0	1	0	0	0	1	2
79.6	Μ	0	0	0	1	1	1	0	0	0	0
79.8	Μ	0	0	0	1	1	1	0	0	0	0
81.7	Μ	1	1	0	0	0	0	0	0	1	3
84.2	F	1	1	0	0	0	0	0	0	1	3
84.4	Μ	1	1	1	0	0	0	0	0	0	3
85.0	F	1	1	0	0	0	0	0	0	1	3
85.3	F	1	1	1	0	0	0	0	0	0	3
85.5	F	1	1	0	0	0	0	0	0	1	3
86.8	F	1	0	0	0	1	0	0	0	1	2
120.3	Μ	0	0	0	1	1	0	0	0	1	1

A comparison of total blocked pattern rates at all body locations to SVL using a nonparametric regression analysis was nonsignificant (tau b = -0.107, P = 0.485). A comparison of combined blocked plus splotched patterns summed across all body locations was nonsignificant (tau b = -0.096, P = 0.549), with both analyses indicating no correlation of ventral pattern to SVL.

However, Pearson product moment correlation (Table IV) analyses confirm the description of the patterns above. Analyses of significant positive correlation coefficients indicates the following: (1) correlation of anterior blocked pattern to middle blocked pattern, (2) correlation between middle blocked and posterior white, (3) correlation of anterior white with middle white, and (4) correlation of middle white with posterior white. Analyses of significant negative correlation coefficients indicates the following: (1) anterior blocked pattern is rarely correlated with middle white, (2) anterior blocked pattern is rarely correlated with middle white, (2) anterior blocked pattern is rarely correlated with posterior white, and (3) middle blocked pattern is rarely associated with posterior white. All other correlations show moderate to weak, nonsignificant correlations (positive = 0.077 to 0.365, negative = -0.097 to -0.365) indicating possible random occurrences.

**Table III.** Combined ventral patterns of blocked, splotched, and white across all body locations (anterior, middle, and posterior). *Pattern* represents the combination of pattern types for all three locations. *Total* indicates the number of specimens in each pattern category. Percentages are based on sums of anterior, middle, and posterior locations as depicted in Table II and divided by the total of 28 snakes used in the study.

Pattern	Total	Percentages
Block only	7	25.00%
Block + Splotched	13	46.43%
Block + Splotched+ White	3	10.71%
White only	2	7.14%
White + Splotched	3	10.71%

**Table IV.** Pearson product moment correlation analysis. All anterior to posterior ventral locations where relationships, whether significant or not, existed are listed. The top value in each cell shows the correlation coefficient. The bottom values in each cell are *P*-values. Abbreviations are the same as those in Table II. Abbreviations are as follows: A = anterior, M = middle, P = posterior, B = blocked, W = white, S = splotched. Negative values indicate negative relationships. An asterisk (\*) indicates significance.

Locations	MB	PB	AW	MW	PW	PS
AB	0.737	0.269		-0.737	-0.595	0.078
	< 0.001*	0.166	0 =0=	< 0.001*	< 0.001*	0.692
<u>MB</u>		0.305	-0.737		-0.439	-0.097
		0.050	< 0.001	-0.265	0.020*	0.024
<u>PB</u>				-0.305		
				0.030	0 505	-0.078
<u>AW</u>				< 0.001*	< 0.001*	0.692
<b>N 67</b> 4 7					0.439	0.097
<u>MW</u>					0.020*	0.624

#### DISCUSSION

The overall frequency of dorsal colors across all three locations (Figure 3, Table I) supports general statements that A. piscivorus darkens with age or increasing SVL (Conant and Collins 1998, 401). Behler and King (1979, 684) and Conant and Collins (1998, 401) in their description of A. piscivorus only describe the general pattern as SVL increases. Our data shows that snakes with SVL greater than 60 cm have increased substantially in darker colors (black and brown) and lose many lighter colors producing a significant negative correlation with SVL. This general pattern is well documented in the literature. Wright and Wright (1957), Bechtel (1978), and Conant and Collins (1998, 401) have referenced it for multiple species (Appendix 1) as well as A. piscivorus. To our knowledge, a quantified approach to ontogenetic color change in A. piscivorus or other snakes has not been reported. A search of the literature did not find any references that quantified the process as done in this study showing loss of lighter colors as SVL increased. Figure 3 and Table I demonstrate that the total number of lighter colors begins to decrease in snakes whose SVL begins to exceed about 35.9 cm and continues to decline thereafter. Nonparametric regression analysis provides a graphic depiction of the decrease in lighter colors and an increase in darker colors across the anterior, middle, and posterior locations examined on each snake as shown in Table I.

A gender difference analysis of SVL showed that females had a lower median SVL value than males, but the difference was not significant, and all snakes were combined for analyses. However, Zaidan (2001), working with snakes 45+ cm total length, notes that variation in the Western Cottonmouth, *A. piscivorus leucostoma*, in northwestern Arkansas involves both sexual dimorphism and dichromatism and specifically states that melanism increases with age and that females darken at lower SVLs. Table I is supportive of his findings in that the four snakes with the shortest SVL are all females and snake 40.0 and 43.2 cm (SVL) show loss of tan and white colors. However, larger samples sizes would be needed to show that snakes from the Alapahoochee basin follow the same patterns of female verses male darkening that Zaidan (2001) observed. He attributed earlier changes to darker colors in females to possible thermal regulation such that females need to elevate body temperature when carrying developing embryos by absorbing more sunlight.

When examining ventral color patterns, a compounding factor was that white occurred at all three locations regardless of whether blocked and splotched patterns were present. The presence of blocked and splotched patterns was assessed for the study with the presence of white indicating no blocked or splotched pattern being present at a location. Ventral pattern analyses show that splotched pattern was the only pattern in the posterior location of these snakes (Table II and IV). Blocked pattern is most common at both anterior and middle locations with some splotched-like pattern intermixed, but white is also exhibited.

The prevalence of white at all locations with some locations also possessing blocked or splotched raises the question of genetic causation. One plausible genetic cause would be a single locus showing incomplete dominance having one allele resulting in no dark blocked pattern, another allele resulting in incomplete dark bars, and heterozygotes showing only partial bars or splotches. Modifiers may also be influencing observed patterns and may account for the perceived differences between blocked and splotched patterns.

The fact that no specific trend of patterns relates to increasing SVLs further supports the argument that genetic components are the cause of ventral pattern, and age or SVL is not a likely factor. Zweifel (1981) studied California King Snakes, *L. getulus californiae*, and found simple Mendelian patterns with the ringed pattern being recessive to the striped pattern with modifiers influencing the patterns observed. King (2003) also studied melanism and striped patterning in the Garter Snake, *T. sirtalis*, and found melanism to be a simple Mendelian trait with the pattern of striped patterning dominant. Kuriyama et al. (2013) studied Japanese Four-Lined Snakes, *Elaphe quadrivirgataused*, and looked at genetic mechanisms using histological techniques. They developed genetic models comparing striped and nonstriped patterns and variations in melanism that involved complete and incomplete dominance with pigment cell composition being a key factor.

The pattern of increased darkening of the dorsum in *A. piscivorus* in the Alapahoochee River basin without change in the ventral patterns has also been demonstrated in the Northern Water Snake, *N. sipedon*. Bowen (2003) looking at ground color changes in a study of island populations found a pattern similar to that found in this study.

#### CONCLUSIONS

This study used a statistical approach to quantify with a higher level of precision what has been more generally stated in the literature with respect to the darkening of the dorsal colors of *A. piscivorus* as SVL increases. It has also shown that ventral patterning does not follow a similar pattern, but that ventral color pattern is independent of SVL. Potential genetic relationships that may control ventral patterns were discussed. Support for this study and the studies involving general statements referenced in the literature can be further supported by analyses of dorsal and ventral color patterning in other populations of *A. piscivorus* as well as other species of snakes. This study also provides a method for teaching the value of using statistical methods to describe biological processes such as those involving color change and patterning or the lack of them.

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#### REFERENCES

- ASIH Guidelines. 2004. American Society of Ichthyology and Herpetology. http://www.asih.org/locations/default/files/documents/Resources/guidelineshe rpsresearch2004. pdf.
- Barnett, J., D.L. Bechler, C. Denizman, J. Grable, J. Nienow, J. Turco, W. Tietjen, and G.L. Wood. 2007. Watershed restoration action strategy development in the Alapahoochee River watershed. Nonpoint Source Management Program, Section 319. Report Submitted to Environmental Protection Division, Department of Natural Resources, Georgia, USA. 92 pp.
- Bechler, D.L. 2006. A survey and analysis of fish diversity in the Alapahoochee River of South Georgia. Georgia Journal of Science, 64(1), 28–29, abstract.
- Bechler D.L., P. Hightower, J. Rousey, and M.E. Smith. 2014. The use of nest-traps to study behavior, population structure, and life history of *Procambarus spiculifer*. Freshwater Crayfish, 20(1), 7–16.

- Behler, J.L. and F.W. King. 1979. The Audubon Society Field Guide to North American Reptiles and Amphibians. Alfred A. Knopf: Random House.
- Bechler, D.L. and J.S. Salter. 2013. The status of the blackbanded sunfish and other species of concern in the state of Georgia. Final report, Non Game Division, Georgia Department of Natural Resources, Social Circle, Georgia.
- Bechtel, H.B. 1978. Color and pattern in snakes (Reptilia, Serpentes). Journal of Herpetology, 12(4), 521–532. <u>http://www.jstor.org/stable/1563357</u>.
- Bittner, T.D., R.B. King, and J.M. Kerfin. 2002. Effects of body size and melanism on the thermal biology of garter snakes (*Thamnophis sirtalis*). Copeia, 2002(2), 477–482. doi:10.1643/0045-8511(2002)002[0477:EOBSAM]2.0.CO;2.
- Brodie, E.D. 1992. Correlational selection for color pattern and antipredator behavior in the garter snake *Thamnophis ordinoides*. Evolution, 46(5), 1284–1298. doi:10.1111/j.1558-5646.1992.tb01124.x.
- Bowen, K.D. 2003. Ontogenetic changes in the coloration of the northern watersnake, *Nerodia sipedon sipedon*. Journal of Herpetology, 37(4), 729–731.
- Chaney, J.C. and D.L. Bechler. 2006. The occurrence and distribution of *Heterandria formosa* (Teleostei, Poeciliidae) in Lowndes County, Georgia. Georgia Journal of Science, 64 (2), 67–75.<u>http://digitalcommons.gaacademy.org/gjs/vol64/iss2/9/</u>.
- Conant, R. and J.T. Collins. 1998. A Guide to Reptiles and Amphibians: Eastern and Central North America. Volume 12. Houghton Mifflin Harcourt.
- Creer, D.A. 2005. Correlations between ontogenetic change in color pattern and antipredator behavior in the racer, *Coluber constrictor*. Ethology, 111(3), 287–300. doi:10.1111/j.1439-0310.2004.01062.x.
- Dunn, E.R. 1954. The coral snake "mimic" problem in Panama. Evolution, 8(2), 97–102. doi:/10.1111/j.1558-5646.1954.tb00116.x.
- Edwards, L., J. Ambrose, and L.K. Kirkman, eds. 2013. The Natural Communities of Georgia. University of Georgia Press.
- Greene, H.W. and R.W. McDiarmid. 1981. Coral snake mimicry: does it occur? Science, 213 (4513), 1207–1212. <u>http://www.jstor.org/stable/1687324</u>.
- Gloyd, H.K. and R. Conant. 1990. Snakes of the Agkistrodon Complex: a Monographic Review. Society for the Study of Amphibians and Reptiles, Contributions to Herpetology 6.
- Hayakawa, S., N. Kawai, and N. Masataka. 2011. The influence of color on snake detection in visual search in human children. Scientific Reports, 1, 80. doi:<u>10.1038/srep00</u> <u>080</u>.
- Hightower, P.W. and D.L. Bechler. 2013. The life history of the crayfish *Procambarus spiculifer* in the Alapahoochee River. Freshwater Crayfish, 19(1), 77–89.
- King, R.B. 1988. Polymorphic populations of the garter snake, *Thamnophis sirtalis*, near Lake Erie. Herpetologica, 44(4), 451–458. <u>https://www.jstor.org/stable/3892411?seq=1#page\_scan\_tab\_contents</u>.
- King, R.B. 1993. Color-pattern variation in Lake Erie water snakes: prediction and measurement of natural selection. Evolution, 47(6), 1819–1833. doi:10.1111 /j.1558-5646.1993.tb01272.x.
- King, R.B. 2003. Mendelian inheritance of melanism in the garter snake *Thamnophis sirtalis*. Herpetologica, 59(4), 484–489. doi:<u>10.1655/02-93</u>.
- King, R.B. and R. Lawson. 1997. Microevolution in island water snakes. BioScience, 47(5), 279–286. doi:<u>http://dx.doi.org/10.2307/1313189</u>.

- Kirkley, J.A. 2014. Biogeography and population genetic structure of the cottonmouth, *Agkistrodon piscivorus*, in the Alapahoochee watershed. Master's Thesis, Valdosta State University.
- Kuriyama, T., H. Misawa, K. Miyaji, M. Sugimoto, and M. Hasegawa. 2013. Pigment cell mechanisms underlying dorsal color- pattern polymorphism in the Japanese four- lined snake. Journal of Morphology, 274(12), 1353–1364. doi:10.1002/jmor.20182.
- Microsoft. 2014. Microsoft Corporation, <u>https://www.microsoft.com/en-us/</u>.
- StatsDirect. 2014. StatsDirect Ltd, 9 Bonville Chase, Altrincham, CHESHIRE WA14 4QA, UK. <u>http://www.statsdirect.com/</u>.
- USGS. 2017. *Agkistrodon piscivorus* (Lacepède, 1789). <u>https://nas.er.usgs.gov/queries</u> /FactSheet.aspx?speciesID=1197.
- Westphal, M.F., J.L. Massie, J.M. Bronkema, B.E. Smith, and T.J. Morgan. 2011. Heritable variation in garter snake color patterns in postglacial populations. *PloS one*, *6*(9), e24199. doi:10.1371/journal.pone.0024199.
- Westphal, M.F. and T.J. Morgan. 2010. Quantitative genetics of pigmentation development in 2 populations of the common garter snake, *Thamnophis sirtalis*. Journal of Heredity, 101 (5), 573–580. doi:<u>10.1093/jhered/esq044</u>.
- Wright, A.H. and A.A. Wright. 1957. Handbook of Snakes of the United States and Canada, Volume I and II. Comstock Publishing Associates, Cornell University.
- Zaidan, F.I. 2001. Western cottonmouth (*Agkistrodon piscivorus leucostoma*) sexual dimorphism and dichromatism in northwestern Arkansas. Herpetological Natural History, 8, 79–82.
- Zweifel, R.G. 1981. Genetics of color pattern polymorphism in the California kingsnake. Journal of Heredity, 72(4), 238–244. doi:/10.1093/oxfordjournals.jhered.a10 9487.

#### APPENDICES

*Appendix 1.* Examples of species and subspecies that change dorsal colors or patterns as SVL increases. The species listed are taken from Conant and Collins (1998).

Scientific Name of Species and Subspecies

Common Names

Shift to Darker Colors as SVL Increases

Redbelly Water Snake Northern Water Snake Banded Water Snake Fox Snake Black Rat Snake Yellow Rat Snake Baird's Rat Snake Northern Pine Snake Black Pine Snake Desert King Snake

#### Shift to Lighter Colors as SVL Increases

Virginia striatula Carphophis amoenus amoenus Opheodrys aestivus Rough Earth Snake Eastern Worm Snake Rough Earth Snake

#### Lose Pattern as SVL Increases

Nerodia erythrogaster flavigaster Lampropeltis getula nigra Lampropeltis getula holbrooki Leptodeira septentrionalis septentrionalis Yellow Belly Water Snake Black King Snake Speckled King Snake Northern Cat-Eyed Snake

#### Change Pattern as SVL Increases

Lampropeltis triangulum triangulum Rhinocheilus lecontei tessellatus Eastern Milk Snake Texas Longnose Snake

## Appendix 2. Genetic studies on snake species

<u>Common and</u> <u>Scientific Names</u> California king snake, <i>Lampropeltis getulus</i> <i>californiae</i>	<u>Genetic Traits and Characteristics</u> Mendelian patterns with ringed pattern being recessive to the striped pattern with influencing modifiers.	<u>Reference</u> Zweifel 1981
Common garter snake, <i>Thamnophis sirtalis</i>	Genetic drift and founder effect may be key factors influencing variations in striped and melanistic morphs.	King 1988
Northwestern garter snake, <i>Thamnophis</i> ordinoides	Demonstrated variances and covariances between color patterns and behavior patterns.	Brodie 1992
Northern water snake, <i>Nerodia sipedon</i>	Concluded natural selection acted upon color morphs and gene flow between populations of mainland and Lake Erie island snakes.	King and Lawson 1997
Common garter snake, <i>Thamnophis sirtalis</i>	Studied age specific pigmentation phenotypes in Northern California and Manitoba, Canada, populations with geographic isolation and phenotypically being distinct.	Westphal and Morgan 2010
Common garter snake, <i>Thamnophis sirtalis</i>	Studies post glacial migrations of populations and found more recently migrated northern populations exhibited equal or greater pattern and genetic diversity than more southern populations.	Westphal et al. 2011
Lake Erie water snake, Nerodia sipedon insularum	Found predation on juveniles and associated natural selection factors influenced gene flow between islands in Lake Erie and the mainland.	King 1993
Common garter snake, Thamnophis sirtalis	Concluded that melanism is a simple Mendelian trait and stripe patterning is dominant to melanism.	King 2003
Japanese four-lined snake, <i>Elaphe</i> quadrivirgata	Heredity models involving complete and incomplete dominance related to melanism, nonstriped and striped patterns as well as striped, pale-striped, and nonstriped polymorphisms based on differences in spatial structure of pigment cell composition.	Kuriyama et al. 2013