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RECRUITMENT, COST INDEXES, AND MANAGEMENT OF BOX-NESTING WOOD DUCKS IN SOUTH CAROLINA AND NORTH CAROLINA

A Thesis Presented to The Graduate School of Clemson University

In Partial Fulfillment of the Requirements for the Degree Master of Science Wildlife and Fisheries Biology

> by Emily M. Miller August 2022

Submitted to: Dr. Greg Yarrow, Committee Chair Dr. Kyle Barrett Dr. Jim Anderson Dr. Richard M. Kaminski

ABSTRACT

The wood duck (*Aix sponsa*) has experienced one of the most significant declines and recoveries among species of North American waterfowl (Anatidae). With enactment of the Migratory Bird Treaty Act (1918) and installation of hundreds of thousands of artificial nest structures for this cavity-nesting species in North America, wood duck populations have recovered and remain a sustainable harvested resource. However, longterm research on box-nesting wood ducks conducted at the Savannah River Ecology Laboratory, Aiken, South Carolina, has revealed uncertainty as to whether recruitment rates of yearling females from natal box-nesting populations are self-sustaining without immigration of hens from other complexes of boxes and natural cavities.

North Carolina and South Carolina have intensive wood duck nest box programs on public and private lands, numbering in the tens of thousands of boxes. Therefore, I conducted a study in 2020 and 2021 at Lake Moultrie in South Carolina and in North Carolina at Mattamuskeet and Roanoke River National Wildlife Refuges (NWR) and Heron Bay, a private property adjoining Mattamuskeet NWR. I monitored nearly 400 boxes examining wood duck reproductive and recruitment data and evaluating strategies to deter rat snakes (*Pantherophis alleghaniensis*) and woodpeckers, both major predators of wood duck eggs at my study sites. I also calculated cost indexes of yearling female recruits over an assumed 20-year longevity of nest boxes.

Box use was high at all sites and both years but was greater at Lake Moultrie (98%) than the pooled average for North Carolina sites (86%). Across all sites and years, nest success was 54.6%, and frequencies of successful and unsuccessful nests did not

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differ between sites in 2020 but did in 2021. The difference between years can be attributed to increased snake predation (9%) from 2020 to 2021. An average of 2.31 more ducklings exited successful nests in boxes at Lake Moultrie than from the North Carolina sites in 2020, but the 1.84 more ducklings exited North Carolina boxes in 2021. Despite the between-year differences, no difference in number of ducklings exiting boxes occurred between sites when yearly data were pooled (i.e., 11.3 ducklings/successful nest). I calculated an index of the cost of wood duck box construction and maintenance over 20 years in relation yearling female recruits, using a published recruitment rate and the average recruitment rate from Lake Moultrie from 2020–2021. The average cost per recruit was 1–4 times less than the cost of the box and its management over 20 years, suggesting cost-efficiency based on this approach.

To deter rat snakes from entering nest boxes, I attached a black cotton sock containing one cup of Snake-A-Way[®] pellets on nest boxes above the predator guard at boxes with previously recorded or current snake predation of wood duck eggs. An empty black sock was deployed likewise as a control in this experiment. In addition, I tagged snakes with a passive integrated transponder (PIT tag) and took morphometric measurements of each rat snake captured in nest boxes. Using a Lincoln-Petersen estimator, I estimated 138 (SE = 25) rat snakes using nest boxes at Lake Moultrie, where snake depredation of eggs was greatest. I PIT tagged 106 snakes among all North Carolina sites and Lake Moultrie. Average total length of male and female snakes combined was 143.1 (SE = 2.01) cm, with captured males being larger than females at all sites. A total of 456 eggs were consumed by captured snakes, with peak snake predation

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occurring in April. Snake pellets were deployed 39 times across field sites where snakes were encountered; there were 14 snake encounters after pellets were deployed compared to no encounters after the other 25 pellets deployments. However, I did not detect a significant difference in the number of snakes accessing boxes treated or not treated with pellets. I deployed trail cameras in each state to determine how snakes were circumventing functional predator guards. Snakes wrap their body around the post and use predator guards to leverage their ascent upward to access the box entrance.

To deter woodpeckers from entering nest boxes, I attached a Bird-B-Gone[®] plastic hawk decoy to boxes with previously recorded woodpecker depredations of wood duck eggs. Decoys were deployed after 12 twelve days of incubation to discourage hen abandonment of nests from the hawk effigy. Across both years and all sites, woodpeckers were responsible for 65.6% of nest depredations. A total of 26 woodpecker encounters were recorded for nest boxes with and without the hawk decoy. There was a significant difference in the number of successful nests in boxes containing the hawk decoy in 2021. My study indicated there is a need for continued recruitment data and sensitivity analysis to determine if rat snakes, woodpeckers, and other agents of nest loss are decreasing recruitment rates from boxes in my study sites, in our regional study across the Southeast, and throughout the range of North American wood ducks.

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ACKNOWLEDGEMENTS

I am truly grateful to all who have supported me during my time at Clemson University and throughout my time as a student and technician in this field. Everyone in South and North Carolina has helped me become a better scientist and was always ready to lend a helping hand if needed. I had the honor of working with Dr. Richard Kaminski who took me under his wing as a technician during the 2019 pilot study for the regional wood duck study. Without Dr. Kaminski taking me on as a graduate student, I would not have had the opportunity to obtain my degree from Clemson University, for that I am truly grateful. I would not be the person I am today without him. Rick and Loretta, thank you for endless meals of various waterfowl species, and sage advice. Rick, thank you for always providing me with steadfast support and guidance. It has been an absolute honor working with you and I'm honored to say I was your final graduate student. Enjoy your retirement, it is more than well deserved. Thank you to Dr. Mike Losito of SUNY Cobleskill who originally spiked my passion of waterfowl through his courses. Thank you, Doc for taking students banding through the student chapter of Ducks Unlimited, this is where I held my first wood duck, it honestly changed my life. Thank you to my other committee members, Drs. Greg Yarrow, Kyle Barrett, and Jim Anderson for assisting in revisions of my thesis. I thank Dr. Ernie Wiggers and Beau Bauer from Nemours Wildlife Foundation. Without their guidance and collaboration and support from Nemours, this incredibly large and impactful study spanning two flyways and eight states would not have been possible. You both have provided me with many wonderful opportunities and endless knowledge and wisdom. Thank you for always going to bat for your students and technicians, and always lending a helping hand. Beau, thank you for selecting me from the pool of applicants for the pilot study, I would not have this wonderful opportunity otherwise. Thank you to Ms. Alreda Grate, you were always so kind and helpful with everything, especially when I missed time punches on KRONOS. Thank you to Andrea Kesler, Vickie Byko, and Alexis Jennings, you all are the glue that has held together the Forestry and Environmental Conservation Department at Clemson University.

Thank you to Clemson University's James C. Kennedy Waterfowl and Wetlands Conservation Center, Nemours Wildlife Foundation, and the South Carolina Department of Natural Resources (SCDNR) for their financial and logistical support. Additionally, I thank Mr. Billy Dukes of the SCDNR, for without him, the pilot study would never have occurred on Lake Moultrie in South Carolina. Thank you to SCDNR biologists Zadok Moss, Will Charlise, Lynda Holseberg, Molly Kneece, and Alicia Farrell. Thank you Zadok, you were a wonderful boss before the pilot study began. I will never forget Remi jumping off the back of your truck to demonstrate her zeal for chasing wild hogs. Thank you, Will, for assisting us throughout the duration of the wood duck project, Lynda for always being a friendly face and answering my endless questions, and Molly and Alicia for assisting us in obtaining wood duck boxes and bands and providing guidance. I thank the U.S. Fish and Wildlife Service for providing study locations, housing, and support while working at Mattamuskeet and Roanoke River National Wildlife Refuges. Thank you to Dr. Heath Hagy, Wendy Stanton, Kendall Smith, and Jean Richter of USFWS for

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all your kindness and support of this study. Additionally, at Mattamuskeet NWR, thank you to Sanders Overman for all your help and assistance in installing boxes, and Ken Andrews for your endless technological assistance. Sanders and Ken, I'll come back one Wednesday to get my club sandwich! Thank you to everyone at Heron Bay in North Carolina, Brinkley Melvin and Jake Hodges, and John Tucker. Thank you, Brinkley for allowing us access to your property and nest boxes. Jake, thank you for your endless hospitality in addition to the countless opportunities and meals you have provided, and Mr. John, thank you for your wonderful meals and endless smiles that made our days brighter. I thank my fellow graduate student on the project, Jacob Shurba, and my technicians over the years (Samantha Fishman, Susannah Halligan, Haley Keff, Jake Merendino, Collin Mulcahey, John Powell, Cindy Von Haugg, and Laura Wallace); our collective work was essential for the success of this project. Thank you to Jason Andrews, Stephanie Braswell, and Jon Mackey for assisting myself and Rick in clearing brush and vegetation around nest boxes on Lake Moultrie in 2019 and 2020.

Last but surely not least, I thank my family whose unwavering support guide me every day. It can't be put into words how grateful and appreciative I am of you all. Thank you, mom and dad for supporting me in applying and accepting the technician position for the pilot study in 2019. I understand it was hard to let me "jump from the nest box", but in doing so I have had an opportunity of a lifetime. I especially thank my father, Michael Sr., for imparting his love of the outdoors to me, without which I would not be

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where I am today. I will never forget when we were walking though the swamp at the property and I got the phone call offering me the position for the pilot study, we were both terrified and excited. Thank you to my mother, Patricia, for helping me find my undergraduate alma mater SUNY Cobleskill, it is here where I learned this field would become my professional niche, and for always helping me look for the positive side in every situation. Thank you to my brother, Michael Jr., and sister, Alyssa, for your infinite support, answering my endless phone calls and being voices of reason. Michael, thank you for assisting me in figuring out my R code. Alyssa, thank you for going over my thesis with me to make sure all my t's were crossed, and i's dotted. And finally thank you to my grandpa, Denis Farone. Our scheduled 5 pm phone calls were something I looked forward to everyday and made being away from family easier. I hope to see some wood ducks using the nest boxes you built on your pond. Ryan, thank you for all the support you have given; I look forward to all our adventures to come with Otis and Castor by our sides.

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CHAPTER ONE

Recruitment and Cost-indexes of Box-nesting Wood Ducks

in South Carolina and North Carolina

Wood ducks (Aix sponsa) have experienced one of the most significant population declines and recoveries among species of North American waterfowl (Anatidae). Due to market hunting, absence of bag limits, and extensive forest and natural cavity loss, wood duck populations declined in the late 19th and early 20th centuries (Bellrose and Holm 1994). With enactment of the Migratory Bird Treaty Act (1918), wood ducks were designated for complete protection (Lawyer 1919). In addition to protection provided by the Migratory Bird Treaty Act, installation of artificial nest boxes aided the species' recovery (Bellrose and Holm 1994). Subsequently, wood ducks consistently have been the second-most harvested duck species in the Atlantic and Mississippi Flyways (Fronczak 2019), where wood ducks comprise $\geq 10\%$ of annual waterfowl harvests in the United States (Davis 2018). In the Atlantic Flyway, wood duck harvest estimates exceeded 370,000 birds during the 2017 and 2018 hunting seasons (Raftovich et al. 2019). Additionally, wood ducks and other Nearctic waterfowl contribute significant economic values and ecosystem services (e.g., Grado et al. 2011, Green and Elmberg 2014, Croft 2018). Moreover, the wood duck was named the state duck of South Carolina, where part of my study was conducted (South Carolina General Assembly 2009).

Wood ducks are year-round residents in the southeastern United States, wherein South Carolina has one of the most intensive nest-box programs in the country (Prevost et al. 1990). The South Carolina Department of Natural Resources and the South Carolina Waterfowl Association have provided thousands of nest boxes to public and privatesector cooperators. The South Carolina Department of Natural resources published its 7th annual statewide wood duck box production report in 1988. In that year, an estimated 7,721 boxes were installed yielding an estimated 20,000 ducklings (Prevost 1988). By 1992, estimates indicated >23,000 boxes existed in South Carolina (Otis and Dukes 1995, Croft 2018), with an additional ~35,000 boxes in the Atlantic Flyway and hundreds of thousands continentally (Bellrose and Holm 1994).

In 2018, the Nemours Wildlife Foundation (Yemassee, South Carolina; https://nemourswildlifefoundation.org) hosted a workshop involving over 30 waterfowl biologists from state agencies, the U.S. Fish and Wildlife Service, Ducks Unlimited, Inc., and several universities to identify waterfowl and wetlands research needs and priorities for the Southeast. Identifying factors affecting nest-site selection and recruitment of boxnesting wood ducks (Schmidt et al. 2018, Bauer et al. 2020, Bauer et al. 2021) emerged as an important need given the prevalence of and prior investment in nest boxes in the Southeast. A pilot study was initiated at Lake Moultrie, South Carolina in 2019 to test methodologies and feasibility of a regional study across the Southeast. Research methods

were refined, and the region-wide study was initiated in 2020 in eight states (Maryland, Delaware, North Carolina, South Carolina, Georgia, Florida, Mississippi, and Louisiana).

Wood duck recruitment is defined as the percentage of yearling female wood ducks produced in nest boxes and marked with unique tags that return to nest in their natal box or colony of boxes in the year following hatching (Hepp et al. 1989). Hepp et al. (1989, 2020) conducted studies of wood duck recruitment in South Carolina, but there have been no regional studies in the continental range of wood ducks. Because Hepp et al. (2020) questioned whether recruitment by box-nesting wood ducks was adequate to sustain local populations without immigration of females from other box colonies or natural cavities, our regional study was initiated in 2020 with planned continuance for five years. I conducted my study in 2020 and 2021 at Lake Moultrie in South Carolina and in North Carolina at Mattamuskeet and Roanoke River National Wildlife Refuges (NWR) and a private property adjoining Mattamuskeet NWR. Given this background and justifications, my objectives were to 1) estimate female wood duck percent use of nest boxes, nest success, and average number of ducklings that exited boxes and compare results between my study sites; 2) develop an index of yearling female recruitment based on re-encounter rates of these females web-tagged as ducklings in 2019 and 2020 and returning to nest in boxes in 2020 and 2021 at Lake Moultrie, South Carolina; 3) compare my results with like data from our regional study and other previous similar studies (e.g., Hepp et al. 1989, 2020; Croft et al. 2022); 4) use a procedure published by Croft et al.

(2022) to calculate the cost-indexes of nest boxes in my study using my wood duck
reproductive data and a published recruitment rate estimate for wood ducks (Hepp et al.
1989, 2020) and the mean 2020–2021 recruitment index from my study at Lake Moultrie,
South Carolina; and 5) address management implications pertinent to my results.

Study Area

My study was conducted at Lake Moultrie (33.319334, -80.059367) in Berkley County, South Carolina and in North Carolina at Mattamuskeet NWR (35.451383, -76.178463), Heron Bay Farms (35.458366, -76.270811), a private property adjacent to Mattamuskeet NWR, both in Hyde County, and Roanoke River NWR (35.912948, -76.724382) in Bertie and Washington Counties (Figure 1.1). Lake Moultrie is located in east-central South Carolina and the North Carolina sites are in the eastern region of the state (Figure 1.1).

Lake Moultrie is part of the Santee-Cooper Lake system, which are reservoirs for hydroelectric power, human recreation, water supply, and fish and wildlife habitat. Lake Moultrie is the third largest lake in South Carolina covering about 24,444 ha. I monitored nest boxes at seven sites in 2020 and 2021 around Lake Moultrie, where boxes were previously erected and managed by the South Carolina Department of Natural Resources (Figure 1.1, Appendix A). Therefore, selection of sites and boxes was not random. Boxes were mainly located along shorelines within bays of the lake, where water depth rarely exceeded 1.5 m.

Dominant vegetation at Lake Moultrie included giant cutgrass (*Zizaniopsis miliacea*), giant salvinia (*Salvinia molesta*), cattail (*Typha* spp.), bald cypress (*Taxodium distichum*), slash pine (*Pinus elliottii*), pond pine (*P. serotina*), red maple (*Acer rubrum*), water and willow oaks (*Quercus nigra*, *Q. phellos*, respectively), willows (*Salix* spp.), white water lily (*Nymphaea odorata*), watershield (*Brasenia schreberi*), water primrose (*Ludwigia peploides*), scouring rush (*Equisetum hyemale*), and duckweed (*Lemna minor*). Submerged aquatic vegetation generally was absent in areas where boxes were located due to shallow water and lily pads shading the water column. Observed submerged vegetation mainly consisted of hydrilla (*Hydrilla verticillata*) and Eurasian watermilfoil (*Myriophyllum spicatum*).

Precipitation and temperature at Lake Moultrie during my study period of January–July 2020 and 2021 averaged 11 cm (range of 7.5–15.7 cm) and 19.2 °C (range of 11.4–27.9 °C), respectively (weatherwx.com 2021*a*). There were several severe storms that occurred during my study, but none had a significant impact on monitored nest boxes. From January to early February 2021, the water level at Lake Moultrie was lowered ~1 m by the U.S. Army Corps of Engineers. Box monitoring was limited due to inaccessibility during this brief period. Long-term precipitation and temperature at Lake Moultrie from January–July 2012–2022 averaged 16 cm, (range of 9.4–23.6 cm) and 18.7 °C (range of 5.6–32.2 °C), respectively.

In North Carolina, Mattamuskeet NWR currently encompasses over 20,000 ha including Lake Mattamuskeet, the largest natural lake in North Carolina. Lake Mattamuskeet had a long history of anthropogenic influences, especially from agriculture, such as being drained three times to expose fertile soil and create profitable farmland. Eventually the pump system draining the lake was no longer cost-effective and was terminated. Currently, the lake is less than half of its historic area of >404,000 ha. I monitored nest boxes at five sites surrounding Lake Mattamuskeet containing boxes (Figure 1.1, Appendix B); therefore, selection of sites and boxes was not random. Heron Bay Farms is a private property bordering Mattamuskeet NWR on the southwest side of the lake; all boxes there were monitored in 2021 (Figure 1.1, Appendix B).

Roanoke River NWR includes about 210 km of the 660-km Roanoke River. The river begins in Virginia in the Blue Ridge Mountains and flows into Albemarle Sound in North Carolina. The Roanoke, Middle, and Cashie Rivers all converge and branch apart near Plymouth, North Carolina, near where the wood duck boxes were located along the river edges, as well as on Grennell and Broad Creeks. I monitored nest boxes at two separate sites on the Roanoke River (Figure 1.1, Appendix C).

Nest boxes in North Carolina were spread across managed impoundments, fields, drainage ditches, creeks, and rivers. Dominant vegetation included giant cutgrass, bald cypress, slash pine, pickerelweed (*Pontederia cordata*), alligator weed (*Alternanthera philoxeroides*), common reed (*Phragmites australis*), sweetgum (*Liquidambar styraciflua*), water tupelo (*Nyssa aquatica*), red maple, water and willow oaks, willows, and mountain maple (*Acer spicatum*).

Precipitation and temperature at my North Carolina sites during my study period of January–July 2020 and 2021 averaged 13.7 cm (range of 9.8–19.3 cm) and 17.8 °C (range of 9.8–28.2 °C), respectively (weatherwx.com, 2021*b*). Few severe storms

occurred in North Carolina; thus, monitoring was rarely disrupted, and no boxes were damaged by severe weather. Long-term precipitation and temperature at my North Carolina sites from January–July 2012–2022 averaged 15.5 cm (range of 12.4–20.3 cm) and 16 °C (range of 1.1–31.7 °C), respectively.

Methods

Nest Boxes, Box Maintenance, and Nest Monitoring

Nest boxes at my study sites in South Carolina and North Carolina were constructed of cypress or pine and mounted on posts or trees, and 93% (n = 373) of boxes had a predator guard. Internal volume of boxes monitored averaged 29,416 cm³ and 37,881 cm³ in South Carolina and North Carolina, respectively, similar to conventional boxes described by Bellrose and Holm (1994). I did not address variation in box construction and dimensions in my study; that was recently conducted and published by Croft et al. (2022) and will be an investigation of the regional study.

For all study areas in both states combined, I monitored 305 and 373 nest boxes in 2020 and 2021, respectively (Table 1.1). In January 2020, I opened 12 nest boxes that were closed during the 2019 pilot study at Lake Moultrie. In March 2020, I added two boxes at Lake Moultrie closer to the shoreline away from possible boating disturbed areas, totaling 181 boxes monitored that season at that site (Table 1.1). Prior to the 2021 field season, South Carolina DNR installed two additional boxes on Lake Moultrie. One box detached from the post between the 2020 and 2021 nesting seasons and was never found. Thus, I monitored 182 boxes at Lake Moultrie in 2021. In 2019–2020 at Lake Moultrie, not all boxes on the lake (n = 1)

~215) were monitored due to inaccessibility and poor box condition; thus, I deemed those monitored a nearly complete sample (~84%) of the box population at Lake Moultrie.

In October 2019 and 2020, prior to each spring field season, I removed encroaching emergent and woody vegetation around nest boxes at Lake Moultrie to retard new growth in spring and possibly deter access to boxes by rat snakes (*Pantherophis alleghaniensis*) that we observed depredating eggs in the boxes during the pilot study. From January to February 2020 and 2021, I examined all nest boxes and predator guards to determine if repairs were needed. Any gaps between predator guards and posts were sealed with expanding foam. I repaired any dilapidated boxes and placed fresh cedar or pine shavings into each nest box to a depth of approximately 6-10 cm. I monitored each box weekly for use by wood ducks from Januarymid-July 2020–2021. During weekly monitoring of boxes, I recorded hen wood duck presence or absence and species-specific number of eggs present in a box. When a clutch hatched successfully or was depredated or abandoned, its fate was recorded, noting the number of ducklings that exited successful nests, residual unhatched eggs, egg-shell membranes indexing departing ducklings (Davis et al. 1999), and dead or alive ducklings. Before ducklings departed nest boxes, I marked them with web tags to enable subsequent estimation of recruitment rates (Hepp et al. 1989, Haramis and Nice 1980; see duckling tagging described below).

In March 2020, I installed seven additional boxes at Mattamuskeet NWR to increase box sample size in an area where numerous wood ducks were observed in early January 2020. I monitored 124 boxes at Mattamuskeet during 2020 (Table 1.1). In February 2021, I included the additional 67 boxes at Heron Bay Farms and monitored a total of 191 boxes at these

contiguous sites during the 2021 field season (Table 1.1). All boxes were located on impoundments or canals within the refuge boundary; those at Heron Bay were 0.5 km from boxes at Mattamuskeet NWR. Not all boxes available at Mattamuskeet and Roanoke River NWRs were monitored due to inaccessibility and missing boxes. However, I monitored 79% (n = 241) boxes at these three sites.

Duckling Web Tagging

To calculate a recruitment index, wood duck ducklings were captured and tagged in the inner web of their right foot web with an alphanumeric tag (i.e., monel #1005-1, National Band and Tag Company, Cincinnati, Ohio); each tag had a unique code for each state (e.g., SC0001, NC0001). Male and female ducklings were tagged to reduce error in sex determination of day-old ducklings. Following Haramis and Nice (1980), I modified needlenose pliers by placing a small, tapered cypress board on the flat sides of the pliers' jaws. In one cypress board, I measured and drilled holes through the board the width of a web tag. I cut the tips off two scalpel blades and inserted and glued the blades into the holes in the cypress board to cut slots in ducklings' webbing (Figure 1.2 A, B). Having one blank board was ideal for spreading the webbing to create a flat surface for a clear view to cut holes in the webbing (Haramis and Nice 1980). When a tag was successfully inserted into the web (Figure 1.2 C), I used another set of needle-nose pliers to crimp the tag closed (Figure 1.2 D). Once closed, the tag was located in the middle of the web (Figure 1.2 E, F). Web tagging was authorized as subpermittes under federal and state bird banding permits (North Carolina, Dr. Heath Hagy, U.S. Fish and Wildlife Service Waterfowl Ecologist [24239]; South Carolina, Mr. Billy Dukes, South Carolina Department of Natural Resources Chief of Wildlife [06658])

and through Clemson University's Institutional Animal Care and Use Committee (IACUC Protocol 2018-069).

Statistical Analysis

My analytical units for statistical analyses (a priori $\alpha = 0.05$) were total boxes monitored for within-year percent use (≥ 1 egg present), total nests for percent successful nests (≥ 1 egg hatched), and total successful nests for average number of ducklings exiting boxes. I determined within-year statistics to explore any trends between 2020 and 2021 but did not test for year effects lacking empirical data to explain such effects. I calculated the percentages of boxes used and not used by wood ducks and their associated standard error (SE) for each state in 2020 and 2021, because I monitored a sample and not all boxes (Microsoft Excel 2021). I tested for the difference in frequencies of boxes used and not used between sites within years using chi-square analysis (Microsoft Excel 2021). I calculated percentages of successful and unsuccessful nests and their respective SE (Microsoft Excel 2021), and the differences between North Carolina and South Carolina sites within years using chi-square analysis (Microsoft Excel 2021). I conducted a Mann-Whitney U-test to determine if number of ducklings exiting boxes differed between Mattamuskeet and Roanoke River NWRs in North Carolina in 2020 (R Core Team 2021). In 2021, when I monitored boxes at three sites in North Carolina, I performed a Kruskal-Wallis test to determine if any differences existed in number of ducklings exiting boxes among Mattamuskeet NWR, Roanoke River NWR, and Heron Bay in North Carolina (R Core Team 2021). Finally, I used a Mann-Whitney U-test to test if there was a difference in number of ducklings departing nest boxes between Lake Moultrie and the pooled North Carolina sites within and across years (R

Core Team 2021). I performed tests among sites to provide comparative data and inform sponsors of our research.

I calculated an index of recruitment by yearling female wood ducks that were webtagged in 2019 or 2020 and discovered nesting as a yearling in boxes at Lake Moultrie in 2020 or 2021. I only used Lake Moultrie recruitment data from my study, because two years of data existed, whereas only one year of data existed for the North Carolina sites. The index was a re-encounter rate in the year following initial web-tagging and thus not an actual recruitment rate adjusted for duckling capture probability as performed by Hepp et al. (1989). To calculate the recruitment index, I divided the number of recaptured web-tagged individuals in year i + 1by the total number of females web-tagged in year i.

Additionally, I computed an index of cost per yearling female returning to nest in a box at Lake Moultrie for 2020–2021 combined. The process included reproductive data from my study, a published index of yearling female recruitment rate (Hepp et al. 2020), calculated costs to fabricate and annually maintain boxes, and an assumed longevity of 20 years for an annually maintained bald-cypress wooden boxes (i.e., Barry 1992, Croft 2018, Croft et al. 2022). I also calculated the same cost analysis using my average 2020–2021 recruitment index for marked females from Lake Moultrie 2019–2020.

Results

Box Use

Box use was 97.7% and 97.8% in Lake Moultrie in 2020 and 2021, respectively. Pooling the North Carolina sites, box use was 86.2% and 84.8% in 2020 and 2021, respectively. Box use was great at all sites and both years but was greater at Lake Moultrie than the pooled North Carolina sites each year (2020, $\chi^2 = 15.18$, df = 1, $P \le 0.001$; 2021, $\chi^2 = 19.49$, df = 1, $P \le 0.001$; Table 1.2). No difference was detected between pooled North Carolina sites and Lake Moultrie in the number of boxes used twice ($\chi^2 = 0.312$, df = 1. P = 0.576) or three times ($\chi^2 = 0.075$, df = 1, P = 0.784) in 2020 and 2021. At Lake Moultrie and the North Carolina sites, the same 37 and 12 boxes, respectively, were used twice in both years.

Nest Success and Failures

Across all sites and years, there were 583 successful nests from 373 boxes (1.56 nests/box) and 1,067 nesting attempts (overall nest success = 54.6%). Frequencies of successful and unsuccessful nests did not differ between sites in 2020 ($\chi_1^2 = 0.631$, P = 0.427) but did in 2021 ($\chi_1^2 = 4.007$, P = 0.045, Table 1.3). There was an 11.2% decrease in nest success at Lake Moultrie between 2020 and 2021 ($\chi_1^2 = 8.354$, P = 0.004), but no such difference between years was detected for the pooled North Carolina sites ($\chi_1^2 = 0.037$, P = 0.847). For all sites and years, rat snakes and woodpeckers were responsible for 222 (46.3%) and 93 (19.4%) of nest failures, respectively (Table 1.4).

Ducklings

A Mann-Whitney U-test failed to reveal a difference in the number of ducklings exiting boxes at Mattamuskeet and Roanoke River NWRs in 2020 (P < 0.001, Table 1.5). A Kruskal-Wallis test also failed to reveal a difference in number of ducklings leaving boxes at Mattamuskeet NWR, Roanoke River NWR, and Heron Bay in 2021 ($\chi_1^2 = 2.44$, df = 2, P =0.295). Thus, I performed a Mann-Whitney U-test and found that 1.22 more ducklings exited boxes at Lake Moultrie than at the pooled North Carolina sites in 2020 (Table 1.5). In 2021, 1.18 more ducklings exited boxes from North Carolina sites (Table 1.5). However, no significant difference was detected between sites when data for 2020 and 2021 were combined (Table 1.5). Of 583 successful nests across both years, an average of 11.31 and 11.28 ducklings exited nest boxes at Lake Moultrie and the pooled North Carolina sites, respectively.

Recruitment Index

In 2019 and 2020, a total of 577 and 1,136 ducklings were tagged at Lake Moultrie, respectively. Although I did sex ducklings in both years, I assumed a sex ratio of 1:1 (Hepp. et al. 1989, Bauer et al. 2021), yielding 288 and 568 tagged wood duck females in 2019 and 2020, respectively. A total of 12 and 19 web-tagged yearling females were encountered as returning yearling nesting hens tagged in 2020 and 2021, respectively. Therefore, the recruitment index for web-tagged yearling female wood ducks on Lake Moultrie was 4.16 % and 3.35% in 2020 and 2021, respectively, and an average of 3.76% (SE = 3.5, n = 2 years).

Cost Index Analysis

Estimated cost of a nest box (including installation and annual maintenance) over 20 years was \$158.15 (Croft et al. 2022). For Lake Moultrie, using the published recruitment rate by Hepp et al. (2020), I calculated 0.20 yearling female recruit/box/year for 2020 and 2021 combined (i.e., 97.7% box use \times 54.3% nest success \times 11.3 ducklings per successful nest \times 50% females [Hepp et al. 2020] \times 6.8% yearling female recruitment rate per year [Hepp et al. 2020]). Hence, calculated cost per yearling female wood duck recruit over 20 years was \$39.54 (\$158.15 per box / [0.20 return/box/year \times 20 years = 4.0 recruits/box]). I applied the same formula for Lake Moultrie and my average recruitment index of 3.76% for 2020–2021

and calculated 0.11 yearling female /box/year (i.e., 97.7% box use \times 54.3% nest success \times 11.3 ducklings per successful nest \times 50% females [Hepp et al. 2020] \times 3.76% yearling female recruitment index per year) indicating a calculated cost per yearling female wood duck over 20 years was \$70.29 (\$158.15 per box / [0.11 return/box/year \times 20 years = 3.0 recruits/box]). The 25% lower recruitment index at Lake Moultrie resulted in an increased cost per yearling female at my study site; however, the average of both costs per recruit was \$87-118 less than the cost of the box and its management over 20 years.

Discussion

This research was initiated in 2019 as a pilot study to establish methodologies for a regional study to examine use, productivity, and recruitment of female wood ducks nesting in artificial nesting structures. The study has since expanded to become an inaugural regional examination of wood duck and other species nesting in boxes in eight states in the Mississippi and Atlantic flyways.

Within my study sites, the wood duck was the dominant species nesting in boxes, with only two nesting attempts by hooded mergansers (*Lophodytes cucullatus*) in North Carolina. Bellrose and Holm (1994) examined available studies on nest box occupancy at the national and regional flyway levels. Regional flyway level studies indicated that box use was generally greater in southern regions, and the authors indicated an average use rate of 48.3% in the southern Atlantic flyway. This phenomenon may be due in part to longer nesting seasons which allow for multiple nesting attempts, as reported recently by Croft et al. (2020) for wood ducks nesting in coastal South Carolina. I observed a greater box use rate, generally above 95% and 80% at Lake Moultrie and pooled North Carolina sites, respectively. Box use in

nearby Georgia and Florida averaged 59.2% and 89.1%, respectively in 2021 (Bauer et al. 2021). Combining data from boxes in North Carolina, South Carolina, Georgia, and Florida, the overall use rate was 84.6% in 2021 (Table 1.6). Observed box use rate in North Carolina in 2021 was within the standard error estimate of the observed regional average of the study for all eight states, suggesting its regional similarity (Table 1.6). My observed high box use rates at Lake Moultrie may be related to the abundant availability of nest boxes in the state (Croft et al. 2022, Otis and Dukes 1995), and regular annual maintenance of boxes on managed areas at all my field sites in both states (Utsey and Hepp 1997, Croft et al. 2022). I cannot explain the lower use of boxes in Georgia and Delaware (<60%), but these will be discussed by cooperating investigators. Additionally, high box use rate may indicate a low availability of suitable natural cavities in southern forests, as reported for Mississippi (Lowney and Hill 1989), unlike the availability of natural cavities for wood duck nesting in northern states (Soulliere 1985, Nielsen and Gates 2007). No contemporary data on availability of suitable nesting cavities exist for South Carolina and North Carolina or other southern states but sampling is currently underway at Francis Marion National Forest and Hobcaw Barony at the Baruch Institute of Coastal Ecology and Forest Science in South Carolina in 2022 (J. T. Anderson, Clemson University, personal communication).

Utsey and Hepp (1997) examined effects of different frequencies of routine box maintenance and monitoring on box use and production by wood ducks in South Carolina. Boxes that were maintained during the nesting season (i.e., removal of old nesting material, addition of new wood shavings, etc.) produced 35% more ducklings compared to boxes that were cleaned once annually. In my study, boxes were regularly maintained within and

between nesting seasons, with each box being cleaned a minimum of two times each season. Nonetheless, nest success at Lake Moultrie decreased by 11.2% between 2020 and 2021 in association with an increase in predation events by rat snakes and woodpeckers. Within the North Carolina sites, I observed approximately a 2% increase in nest success between years, perhaps partly attributable to an addition of 67 boxes and additional 62 nesting attempts in 2021 at Heron Bay, which entered the study that year. Possible reasons for my observed combined 54.3% nest success rate at Lake Moultrie for 2020 and 2021 included increased snake predation rates of about 9% from 2020 to 2021. Nonetheless, nest success was 1.1 and 1.3 times greater at Lake Moultrie and the pooled North Carolina sites, respectively, than the regional average (Table 1.6).

Stephens et al. (1998) studied differences in number of ducklings successfully exiting large conventional and small experimental boxes in Mississippi. The authors observed an average of 8.3 and 7.3 ducklings leaving large and small boxes, respectively. I observed a larger average number of ducklings exiting nest boxes across years at all my study sites ($\bar{x} = 11.3$). My estimate is about 1.5 times greater than that observed by Stephens et al. (1998) for both small and large boxes. Of the 373 boxes sampled in my study, 59.8% and 40.2% were larger than the large and small boxes observed in Stephens et al. (1998), respectively, allowing more space for eggs and potential for increased duckling production.

Between the North Carolina and South Carolina sites, there was a significant difference in number of ducklings exiting boxes each year; however, when data were pooled across years, no difference was detected and the average number of ducklings exiting boxes from my study sites in both states was 11.3 ducklings. My estimate of ducklings exiting boxes

at Lake Moultrie is 3.7 times greater than that observed by Luckett's (1977) estimate of three wood duck ducklings exiting boxes in the upstate Piedmont region of South Carolina. All my study sites experienced a duckling exodus rate similar to that observed by Utsey and Hepp (1997; 12–13 ducklings [SE \leq 0.9]). At Lake Moultrie, an average of 2.55 fewer ducklings exited boxes in 2021 than 2020. This decrease was influenced by increased predation rates by snakes and woodpeckers in 2021. My North Carolina sites experienced a 1.6 increase in the average number of ducklings exiting boxes from 2020 to 2021. This observation was associated with the addition of boxes monitored on Heron Bay in 2021, where the average number of ducklings exiting boxes at Lake Moultrie in 2021 were within the standard error of the overall regional estimate, whereas the estimate for my study sites in North Carolina was 1.5 greater than that of the regional average (Table 1.6).

Using the previously published recruitment estimate of 6.8% from Hepp et al. (2020), my calculated cost per yearling female wood duck recruit over 20 years was 30% less than the value (\$56.48) reported by Croft et al. (2022) for coastal South Carolina. When using my recruitment index, calculated cost per yearling female wood duck recruit over 20 years was \$13.81 more than Croft et al.'s (2022) estimate. My cost per yearly female recruit indexes were 1–4 times less than the estimated cost of the box and its maintenance over 20 years. Possible reasons for my lower calculated cost, when using the Hepp et al. (2020) recruitment estimate, were that I had a 36.5% greater box use rate on average and one more duckling successfully exiting nest boxes in South Carolina compared to the Croft et al. (2022). However, my low recruitment index (3.76%) is possibly attributable to high numbers of

missed ducklings in the 2019 (n = 598) and 2020 (n = 1,088) nesting seasons in addition to predation events.

Management and Research Implications

Nest box use, success, and duckling production can be readily affected by maintenance and egg depredation. Boxes on Lake Moultrie in South Carolina were relatively conspicuous and actively maintained for three nesting seasons (2019-2021), as were boxes in my North Carolina sites during 2020–2021. Natural cavity availability around Lake Moultrie currently is not known but could be a proximate and ultimate factor (Kaminski and Elmberg 2014) related to greater use of boxes by wood ducks. Additionally, for nest box programs to remain effective, predator deterrents (i.e., metal cones, and stovepipe baffles) should be checked every year for rust and weathering and replaced or repaired when necessary. To determine effectiveness of nest box programs, continued observation should be maintained with annual cleaning of boxes during, and after the nesting season. Annual removal of vegetation surrounding boxes also is recommended to prevent visual obstruction of boxes decreasing use by nesting hens. At Lake Moultrie, Santee Cooper, who is responsible for management of the lake, volunteered to herbicide emergent and woody vegetation around boxes; this cooperation between the Santee Cooper and South Carolina Department of Natural Resources should be continued. Boxes should not be placed directly next to high boating traffic areas as this may decrease box use.

Sensitivity analyses are necessary to determine if wood duck recruitment and populations are sustainable amid snake and woodpecker depredation without immigration of female wood ducks from other nest box colonies or natural cavities (Hepp et al. 2020). These data are being collected and analyses will be conducted as a primary goal of this and ongoing studies in all states involved in the wood duck project (Bauer et al. 2020, 2021). Predator abundance, especially rat snakes at Lake Moultrie, should be quantified and deterrence strategies further assessed (Chapter 2) concurrent with continuance of the nest box programs to avoid an "ecological trap" for nesting wood ducks (Gates and Gysel 1978, Ratti and Reese 1988). Notwithstanding high predation pressure on wood duck eggs by predators, my study and Croft et al. (2022) concluded that the wood duck nest box program in South Carolina was cost-effective over a 20-year period, based on our reproductive metrics and recruitment indexes from my study and Hepp et al. (1989, 2020). I suggest current and future investigators in our regional study and elsewhere conduct sensitivity analyses (sensu Hepp et al. 2020) to determine if recruitment rates are adequate to sustain box-nesting populations ($\lambda \ge 1$) without immigration of yearling females and assess costs of these programs.

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State	Site	2020	2021
South Carolina	Lake Moultrie	181	182
North Carolina	Mattamuskeet NWR	79	79
	Roanoke River NWR	45	45
	Heron Bay Farms	0	67
	Total	305	373

Table 1.1 Number of boxes monitored in 2020 and 2021 at study sites in North Carolina (Mattamuskeet NWR, Roanoke River NWR, and Heron Bay Farms) and South Carolina (Lake Moultrie).

Table 1.2 Frequency (*n*), percentage (%), standard error (% SE^a), and chi-square test (χ^2) of the null hypothesis^b that frequencies of nest boxes used and not used by wood ducks (*Aix sponsa*) would not differ between South Carolina and North Carolina sites in 2020 and 2021.

		South Carolina site ^c		North	h Carolina	sites ^c	
Boxes	Year	n	%	% SE ^a	n	%	% SE
Used	2020	177	97.7	1.1	107	86.3	3.1
	2021	178	97.8	1.1	162	84.8	2.6
Not used	2020	4	2.2	1.1	17	13.7	3.1
	2021	4	2.2	1.1	29	15.2	2.6

^aStandard error of a proportion: $SE = \sqrt{\hat{p}(1-\hat{p})/n}$, where $\hat{p} = \text{proportion}$ and n = frequency (https://www.statology.org/standard-error-of-proportion/).

^bRejected the null hypothesis for 2020 ($\chi_1^2 = 15.18$, $P \le 0.001$) and 2021 ($\chi_1^2 = 19.49$, $P \le 0.001$), concluding that nest box use depended on state sites in each year.

^c South Carolina site is Lake Moultrie; data pooled for North Carolina sites (Mattamuskeet and Roanoke Rivers National Wildlife Refuges and Heron Bay).

Table 1.3 Frequency (*n*), percentage (%), standard error (% SE^a), and chi-square test (χ^2) of the null hypothesis^b that frequencies of successful and unsuccessful nests of wood ducks (*Aix sponsa*) would not differ between South Carolina and North Carolina sites in 2020 and 2021.

		South Carolina site ^c		North Carolina sites ^c			
Nest	Year	n	%	% SE ^a	n	%	%SE
Successful	2020	172	59.9	2.9	92	56.1	3.9
	2021	190	48.7	2.5	129	57.1	3.3
Not successful	2020	115	40.1	2.9	72	43.9	3.9
	2021	200	51.3	2.5	97	42.9	3.3

^aStandard error of a proportion: $SE = \sqrt{\hat{p}(1-\hat{p})/n}$, where \hat{p} = proportion and n = frequency (https://www.statology.org/standard-error-of-proportion/).

^bFailed to reject the null hypothesis for 2020 ($\chi_1^2 = 0.631$, P = 0.427) but not for 2021 ($\chi_1^2 = 4.007$, P = 0.045).

^c South Carolina site is Lake Moultrie; data pooled for North Carolina sites (Mattamuskeet and Roanoke Rivers National Wildlife Refuges and Heron Bay).

				Nest failures			
State	Year ^a	n	Successful	Abandoned	Snake	Woodpecker	Mammal/Other
South Carolina site ^b	2020	287	59.9	15.0	18.5	5.9	<1
	2021	390	48.7	13.8	27.4	9.5	<1
North Carolina sites ^b	2020	164	56.1	15.2	20.1	4.9	3.7
51105	2021	226	57.1	16.4	12.8	13.7	<1

Table 1.4 Percentages (*n*) of successful nests and cause-specific failures at South Carolina and North Carolina sites in 2020 and 2021.

^aNest fates differed between years for South Carolina ($\chi_4^2 = 12.39$, P = 0.015) and North Carolina ($\chi_4^2 = 16.18$, P = 0.003).

^b South Carolina site is Lake Moultrie; data pooled for North Carolina sites (Mattamuskeet and Roanoke Rivers National Wildlife Refuges and Heron Bay).

Table 1.5 Mean (\bar{x}) number and standard error (SE) of wood duck (*Aix sponsa*) ducklings per successful nest (n) at South Carolina and North Carolina sites in 2020 and 2021. Statistics are for Mann-Whitney U tests (W, P) to test the null hypothesis of no difference in number of ducklings departing nest boxes between states within and across years.

	South Carolina site ^a		So		North Carolina sites ^a			
Year	n	\bar{x}	SE	n	\overline{x}	SE	W	Р
2020	172	12.65	0.39	92	10.34	0.42	9,718	< 0.001
2021	190	10.10	0.37	129	11.94	0.39	9,709	< 0.001
Combined	362	11.31	0.27	221	11.28	0.29	39,642	0.812

^a South Carolina site is Lake Moultrie; data pooled for North Carolina sites (Mattamuskeet and Roanoke Rivers National Wildlife Refuges and Heron Bay).

Table 1.6 Frequency (*n*) and percentage (%) of boxes used by wood duck females containing $\geq 1 \text{ egg}$, *n* and % successful nests with ≥ 1 hatched egg and mean (\bar{x}) number of ducklings that exited boxes per successful nest from states involved in the wood duck recruitment project in 2021 (Note: Values in this table were computed from those in Bauer et al. 2021 or those reported in previous tables herein for North Carolina, and South Carolina).

	Boxes used		Successf	Successful nests		
State	п	%	n	%	\bar{x}^{a} ducklings	
Delaware	110	53.9	51	40.8	10.73	
Florida	123	89.1	89	42.4	7.28	
Georgia	71	59.2	47	48.0	12.06	
Louisiana	320	96.7	173	40.5	9.58 ^b	
Maryland	86	88.7	48	51.1	12.25	
Mississippi	149	85.6	51	25.0	8.31	
North Carolina	162	84.8	129	57.1	11.94	
South Carolina	178	97.8	190	48.7	10.10	
$\bar{x} \pm SE^{c}$	81.98	8 ± 5.8	44.20	± 3.4	10.28 ± 0.7	

^a Includes marked and unmarked ducklings exiting nest boxes minus any dead ducklings found in nest boxes.

^b Includes 505 web- and 643 PIT-tagged ducklings (Bauer et al. 2021).

^c Standard error (SE) of mean percentages of boxes used and successful nests and \bar{x} number of ducklings exiting boxes across the eight states.

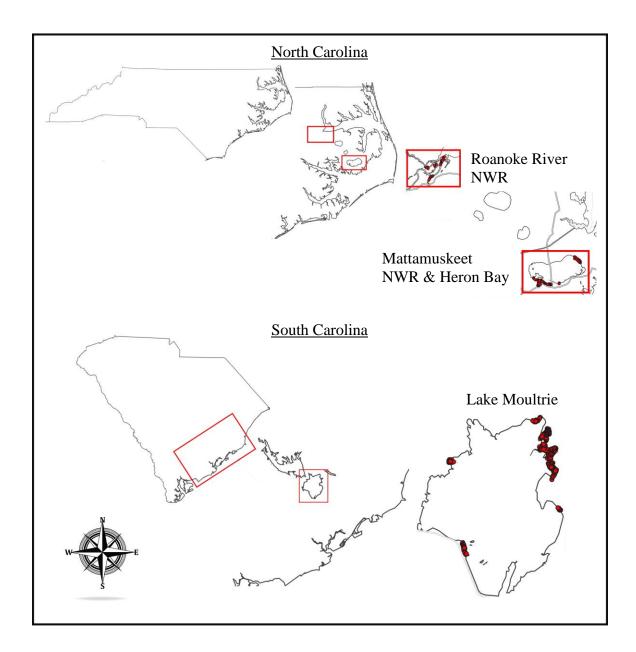


Figure 1.1 Locations of study areas (red boxes) and GPS coordinates of nest boxes (red dots) in South Carolina (Lake Moultrie) and North Carolina (Mattamuskeet and Roanoke River National Wildlife Refuges [NWR] and Heron Bay) in 2020–2021.

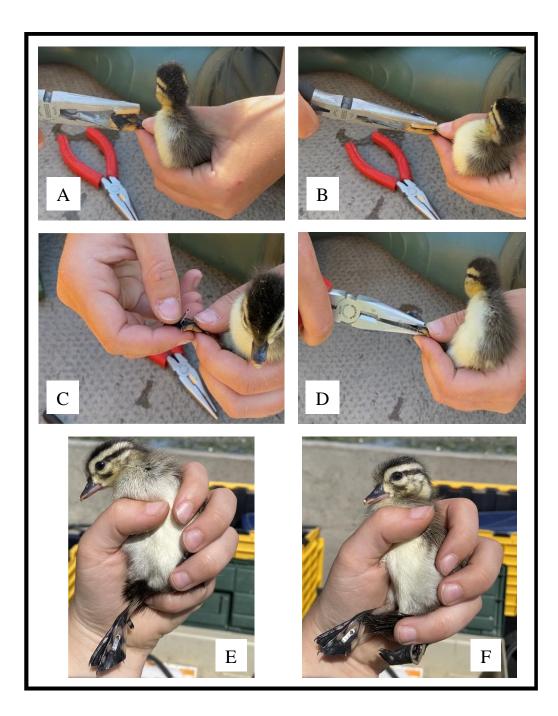


Figure 1.2 Web tagging a wood duck duckling by using modified needle-nose pliers (A, B), inserting the tag through the holes (C), pierced through the webbing with a razor blade affixed to one of the pliers' jaws (B), and closing and crimping the tag shut (D). The result should place the tag in the middle of the proximal webbing between the first two toes of the right foot, as close to the ankle as possible (E, F).

CHAPTER TWO

Rat Snake Abundance and Strategies to Deter Snake and Woodpecker Depredation of Wood Duck Eggs

A diversity of avian, mammalian, and reptilian predators exploit eggs of cavitynesting ducks, such as wood ducks (*Aix sponsa*; Bellrose and Holm 1994). Forbush (1912) was first to suggest using artificial structures to increase nesting sites for wood ducks and lessen egg predation. Hawkins and Bellrose (1940) pioneered monitoring nest structures and concluded boxes were a useful tool for wood ducks and other cavitynesting ducks. However, responsibility falls on managers to maintain structures and provide effective predator guards. Currently, the best-known method to repel predators is a metal cone placed directly under the nest box (Bellrose and Holm 1994). When installed properly, the cone should flare outward and fit snugly to the post to which the box is mounted. However, predator guards can loosen, rust, and become ineffective. For example, Strange et al. (1971) observed raccoons (*Procyon lotor*) were able to access boxes in their Mississippi and Louisiana sites due to poor installation or maintenance of predator guards.

In the southeastern United States, tree climbing snakes are a common predator of wood duck eggs in nest boxes and natural cavities (Bellrose and Holm 1994). Smith (1961) reported gray rat snakes (*Pantherophis spiloides*) in Louisiana were responsible for an average 21% of all unsuccessful nests. Hansen and Fredrickson (1990) reported black rat snakes (*P. aleghaniensis*) depredated 15–36% of wood duck clutches in nest boxes at Mingo National Wildlife Refuge in southeast Missouri. Previous studies and

Chapter 1 indicate that snakes are significant predators of wood duck eggs and can decrease nest success (Chapter 1).

Conner et al. (2001) observed nest site competition between a nesting pair of pileated woodpeckers (Dryocopus pileatus) and multiple pairs of wood ducks. Wood duck females attempted to enter active woodpecker nest cavities, sometimes remaining in the cavity for an hour before expelled by the male woodpecker. In addition to nest site competition, woodpeckers also depredate wood duck eggs. Cunningham (1968) conducted a three-year study of wood duck nest boxes at Yazoo National Wildlife Refuge in Mississippi, where numbers of nest boxes nearly quadrupled between 1966 (n = 55) and 1968 (n = 202). Between 1967–1968, a total of 65 nests (38%) were destroyed by northern flickers (Colaptes auratus), red-headed woodpeckers (Melanerpes erythrocephalus), and red-bellied woodpeckers (*M. carolinus*). Bellrose and Holm (1994) reported woodpeckers were responsible for 20-30% of nest failures from 1983-1985 at Nauvoo Slough in Illinois, where 20 and 80 nest boxes were available in 1983 and 1984– 1985, respectively. Woodpeckers puncture holes to consume egg contents or cause nest desertion by hens (Bellrose and Holm 1994, Cunningham 1968). Woodpecker depredation is detected when small holes are pecked in eggs, ranging in size from a pinprick to a standard pencil eraser (Bellrose and Holm 1994). Woodpecker species depredating nests are often difficult to identify based on holes observed in wood duck eggs. Researchers rely on observing and listening to woodpecker species in the area; however, research suggests that red-headed and red-bellied woodpeckers and northern flickers are the main predators (Clawson 1975, Cunningham 1968, Wilkins et al. 1990)

Over 23,000 boxes were erected in South Carolina (Otis and Dukes 1995, Croft 2018, Chapter 1) by the early 1990s, and an additional ≥35,000 boxes exist in the Atlantic Flyway with hundreds of thousands of units deployed continentally (Bellrose and Holm 1994). In South Carolina and North Carolina, rat snakes and woodpeckers were primary predators of wood duck eggs during my study (Chapter 1). Thus, my objectives were to 1) estimate the number of rat snakes using nest boxes at Lake Moultrie in South Carolina, 2) report recapture frequencies of snakes, 3) quantify gender ratios and lengths of captured snakes, 4) compare demographic and morphological characteristics between snakes in my study and available literature, 5) describe snakes' process of accessing a nest box with a functional predator guard, 6) describe spatial distribution of encountered snakes at study sites in South Carolina and North Carolina, 7) evaluate strategies to deter rat snakes and woodpeckers from entering boxes, and 8) address management and research implications pertinent to my results.

Study Area

My study was conducted at Lake Moultrie, South Carolina and in North Carolina at Mattamuskeet National Wildlife Refuge (NWR), adjacent Heron Bay Farms, and Roanoke River NWR (Figure 1.1). Lake Moultrie is located in east-central South Carolina and North Carolina sites are in the eastern region of the state (Figure 1.1).

For both states, selection of sites and boxes was not random but was designated by cooperating and sponsoring agencies. Boxes were mainly located along shoreline of lakes and bays, managed impoundments, fields, drainage ditches, creeks, and rivers. Site descriptions are detailed in Chapter 1.

Methods

Rat Snake Processing

To estimate the number of rat snakes using nest boxes in 2020 and 2021 at Lake Moultrie, all snakes captured in nest boxes were tagged with a passive integrated transponder (i.e., PIT tag, CYNTAG 2X12 mm glass tags with unique IDs [CATALOG #601201]). When a snake was captured, I recorded the following information: 1) date of capture, 2) state, 3) site within state, 4) box code, 5) PIT tag number, 6) recapture (yes/no), 7) sex, and 8) total length (cm). Prior to tagging a snake, I scanned captured individuals to ensure they were not previously PIT tagged. In addition, each PIT tag was read prior to insertion with a RFID 134.2Khz microchip reader (Sonew, UPC: 792117399640) to ensure its functionality. I placed a single tag into the hollow needle of the syringe (N125 PIT tag implanter needle and MK10 PIT tag implanter [Biomark, USA]), then inserted the tag between the 10th and 20th scale anterior of the cloaca. I inserted the tag where the dorsal and ventral scales meet, pinching the skin so that the tag entered the muscle and did not enter under the rib cage. Tags and needles were stored in a chlorhexidine solution (Nolvasan). After a snake was tagged, it was scanned again with the microchip reader to ensure the tag was functioning. After tagging, total length (i.e., from the snout to tip of tail) of snakes was measured to the nearest cm and individuals were sexed by visually examining the tail below the cloaca for hemipenes (Powell et al. 2016:358). Rat snakes were not tagged until July 2020 at the North Carolina sites; all snakes encountered in boxes in 2021 were tagged. Tagging snakes was permitted through

Clemson University's Institutional Animal Care and Use Committee (IACUC Protocol Number: 2018-069).

Snake Deterrent Experiment

For Lake Moultrie and the North Carolina sites in 2021, I examined a sample of boxes (n = 27) with recorded snake depredations or presence during the 2019–2021 nesting seasons. Depredation data existed only for Lake Moultrie in 2019 because the pilot study was conducted there only. Boxes with previous snake occurrences or depredations were entered into a random generator in program R (R Core Team 2021) to determine boxes that would serve as treatment and control (see treatment and control descriptions below). There were seven treated and seven control boxes and eight and nine boxes at Lake Moultrie and the North Carolina sites in 2021, respectively (Table 2.1). In addition, boxes were treated opportunistically from May–July 2021 to increase sample size at Lake Moultrie (Table 2.1). When a snake was found in a box and eggs remained, commercially available snake repellent pellets were deployed (see Methods below).

In October 2019 and 2020, I removed emergent and woody vegetation in a 2-m radius around all vegetatively obstructed boxes at Lake Moultrie in an attempt to deter snakes from entering nest boxes. In 2020, I also sprayed an expanding foam sealant in any gaps between the predator guards and posts on boxes at Lake Moultrie. In the North Carolina sites, boxes were not placed in or near areas of dense emergent or woody vegetation, and predator guards were new, nonexistent, or previously sealed with caulk. I discovered that vegetation removal alone did not prevent snakes from entering boxes in 2020–2021. Thus, I tested a commercial snake repellant, Snake-A-Way[®] (Victor, USA)

pellets (7% naphthalene and 28% sulfur). In Lake Moultrie and the North Carolina sites, I placed 1 cup of pellets in a black cotton sock attached directly under the nest box above the predator guard, on the wooden post holding the box, or to the lower back panel of the nest box. Socks were zip-tied to metal posts in the same location as described above. For control boxes, only an empty sock was applied likewise. Socks were deployed at the first sign of eggs in a nest box that previously or currently contained a snake(s). Boxes can be used multiple times in a season by nesting hens, so some boxes were treated twice. If a box never contained eggs throughout the season, it was not included in the analysis. Pellets were changed every two weeks to maintain their potency. After a nest was complete, socks were removed.

Woodpecker Deterrent Experiment

At all sites in 2021, I examined a sample of boxes with previously recorded woodpecker egg depredations during the 2019–2020 nesting seasons. Boxes with previously recorded depredations were entered into a random generator in program R to determine treatment and control boxes (R Core Team 2021). I attached a Bird-B-Gone[®] (Bird-B-Gone, USA) plastic hawk decoy to treatment boxes. There were nine treated and eight control boxes at Lake Moultrie, and eight treated and nine control boxes at the North Carolina sites.

Decoys were attached with screws to the top of the post supporting a nest box. Decoys were deployed after 12 days of incubation to discourage hen abandonment of nests. After 12 days of incubation, wood duck hens are less likely to abandon nests

considering the great time and resources invested in the nest (Bellrose and Holm 1994). After a nest was complete, decoys were removed.

Statistical Analysis

I used a Lincoln-Petersen estimator to calculate the abundance of rat snakes that were encountered using nest boxes at Lake Moultrie, South Carolina (Powell and Gale 2015). The estimator is $N = \frac{n_1 n_2}{m_2}$, where N = abundance, n_1 = number of snakes marked in 2020, n_2 = number of snakes marked in 2021, and m_2 = number of recaptures in 2021 from 2020. Assumptions of this estimator include: 1) capture probability is equal for all individuals, 2) marks do not affect capture probability, 3) marks are not lost between capture periods, 4) all marks are reported in second and subsequent capture events, and 5) each capture period is a random sample of the population (Powell and Gale 2015). I assumed all assumptions were met. I used the following equation, SE =

 $\sqrt{\frac{n_1(n_2+1)(n_2-m_2)}{(m_2+1)^2(m_2+2)}}$, to calculate the standard error of this estimate.

I conducted a chi-square analysis to test for a difference in number of male and female rat snakes captured at Lake Moultrie and the North Carolina sites (R Core Team 2021). I also conducted a *t*-test to determine if mean total length of male and female snakes differed within North Carolina and South Carolina sites (Microsoft Excel 2022). I conducted a chi-square analysis, with Yate's correction for continuity and small sample size (e.g., some expected values < 5; Ott and Longnecker 2010), to test for differences in the number of snakes entering boxes treated and not treated with pellets or with or without a raptor decoy to deter woodpeckers (Microsoft Excel 2022). I pooled data from

all sites to increase sample size and statistical power. Snake and woodpecker deterrent experiments are continuing in 2022 to increase sample size and rigor of analyses.

Results

Rat Snake Abundance and Recapture Frequencies

I marked 44 and 34 rat snakes in 2020 and 2021, respectively, at Lake Moultrie, South Carolina, and recaptured 16 (36.4%) snakes in 2021. The estimated number of snakes using nest boxes was 138 individuals (SE = 25, CV = 18% [25/138 x {100}]), based on snakes captured, marked, released from a total of 182 monitored nest boxes at Lake Moultrie (i.e., 138/182 = 0.76 snake/box). However, snakes were encountered in a cluster of 57 (31.3%) of boxes; thus, snake encounters were spatially clustered around the lake where box concentration was the highest (Appendix D).

A total of 6 snakes at Lake Moultrie had recapture frequencies of 4–6 times between 2020 and 2021. In 2020 and 2021, 47.7% and 41.6% of snakes, respectively, were recaptured in the same year as they were tagged (Table 2.2). A total of 159 initial and subsequent snake encounters were recorded, with 47.8% and 52.2% encounters occurring in 2020 and 2021, respectively.

In all North Carolina sites, snakes were captured and marked in a cluster (14.1%) of 191 boxes monitored; again, suggesting snake encounters were spatially clustered (Appendix D). A total of 28 snakes were tagged (Table 2.2) with twelve recapture events from eight individuals in 2021. Only one snake was tagged late season in 2020 and was not recaptured in 2021; thus, I was not able to estimate snake abundance for North Carolina sites. In 2021, 7 snakes were recaptured in the same year they were tagged

(Table 2.2). Only one snake encounter was recorded at Heron Bay, where boxes were erected from January 2020–April 2021. Here, I encountered a snake that was climbing the post and was able to intercept it prior to it gaining access to the box. This individual was included in the analysis.

Snake Sex Ratios and Lengths

At Lake Moultrie, most snakes captured were males (65.3%; $\chi^2 = 7.38$, df = 1, P = 0.006). Mean total length of male and female snakes was 150.2 cm (SE = 21.1, n = 50) and 136.9 cm (SE = 18.5, n = 26), respectively (2 snakes were not measured), and males were longer than females ($t_{57} = 2.84$, P = 0.006). At the North Carolina sites, no difference was detected in the number of captured male (53.6%) and female (46.4%) snakes ($\chi^2 = 0.143$, df = 1, P = 0.705). Mean total length of male and female snakes was 145.9 cm (SE = 18.5, n = 15) and 124.3 cm (SE = 12.7, n = 13), respectively, with lengths differing between genders ($t_{24} = 3.64$, P = 0.001).

For all sites, average total length of male and female snakes combined was 143.1 cm (SE = 2.01, n = 104; Figure 2.1). No difference was detected in the size of males between sites ($t_{26} = 0.769$, P = 0.22); however, females at Lake Moultrie ($\bar{x} = 136.9$ cm, SE = 18.5, n = 26) were, on average, 12.6 cm longer ($t_{33} = 2.51$, P = 0.009) than females at the North Carolina sites ($\bar{x} = 124.3$ cm, SE = 12.7, n = 13). Captured snakes generally exceeded 1 m in length, indicating snakes were relatively large and able to surmount predator guards and gain access to nest boxes (Figure 2.2).

Rat Snake and Woodpecker Egg Depredation

Combining data for both years and sites, snakes and woodpeckers were responsible for 69% and 29 % (n = 321) of nest losses from depredations, respectively. At Lake Moultrie and the North Carolina sites, 382 and 74 eggs, respectively, were known to be depredated by rat snakes (i.e., visible protrusions within the stomach of snakes). Peak snake predation and encounters occurred in April 2020–2021, with at least 180 known egg depredations from just over 80 snake encounters (Figure 2.3). Peak woodpecker depredations (i.e., eggs observed with punctures) occurred in May (Figure 2.4). Across 2020 and 2021, snakes and woodpeckers combined were responsible for 98% (n = 321) of nest depredation events and 65.6% (n = 480) of nest failures. Additionally, these predators were responsible for the loss of 10% (n = 10,382) and 14% (n = 5,595) of all eggs at Lake Moultrie and the North Carolina sites, respectively.

A total of 1,735 eggs disappeared from boxes during the 2021 nesting season alone, possibly due to snakes, woodpeckers, or other predators, or removed from the box by the nesting hen. All missing eggs in 2020 were quantified as "non-viable" so discerning between "missing" and "non-viable" eggs was not possible that year.

Rat Snake Distribution and Access to Nest Boxes

On Lake Moultrie and Mattamuskeet NWR, most captures occurred in the northeast section of each lake (Figure 2.5). I deployed trail cameras on 4 boxes in each state to determine how snakes circumvented predator guards and entered nest boxes. Captured snakes generally were 1–1.5 m in total length; seemingly, they wrap their body around the post and use predator guards to leverage their ascent upward (Figure 2.2). Two dead hens were found in two separate nest boxes (~2.7 km apart) at Lake Moultrie

during the 2021 nesting season, where most rat snake encounters occurred (Figure 2.5). Possible cause of death was rat snake constriction and suffocation, with another hen found dead in March 2022 (Figure 2.6).

Snake Deterrent Experiment

Pellet treatments were deployed 39 times across all field sites. Fourteen snake encounters were recorded for nest boxes with socks containing pellets; snakes were never encountered in a box after the other 25 pellet deployments. The empty sock control was deployed 15 times across both states on 11 boxes, with 6 snake encounters after deployment (Table 2.3). No significant difference was detected in the number of snakes accessing boxes treated or not treated with pellets ($\chi^2 = 0.063$, df = 1, *P* = 0.801). This experiment is being repeated in 2022 to increase sample size and power of statistical analysis.

Woodpecker Deterrent Experiment

A total of 26 woodpecker encounters were recorded for nest boxes at all field sites with and without the hawk decoy, with an observed 11 successful nests with the decoy and 8 failures without the decoy (Table 2.4). I detected a difference in the number of observed successful nests in boxes with the hawk decoy in 2021 ($\chi^2 = 6.80$, df = 1, P = 0.009). This experiment also is being repeated in 2022 to increase sample size and power of statistical analysis.

Discussion

Rat Snake Abundance, Sex Ratios, and Body Length

Rat snake populations generally increase in abundance from north to south in North America (Bellrose and Holm 1994). Within my study locations, we only observed eastern rat snakes accessing wood duck nest boxes (Powell et al. 2016). Despite the increase in snakes south of the 37-degree parallel (Bellrose and Holm 1994), no recent or historical estimates of rat snakes exist for South Carolina on North Carolina. In Ontario, the known northern range of eastern rat snakes, there are two populations, both of which are considered threatened (Weatherhead et al. 2002, Prior and Weatherhead 1998). Population estimates from Queen's University Biological Station and Hill Island in Thousand Island National Park in Ontario indicated an average of 35 (19.4 snakes/km²) and 50 individuals (8.93 snakes/km²), respectively (Weatherhead et al. 2002). I calculated a 600 m buffer around the shoreline of Lake Moultrie (91.57 km²), quantifying possible snake habitat given that most individual rat snakes do not exceed a 600 m range (Stickel et al. 1980). My estimate for snakes in nest boxes at Lake Moultrie (n = 138) was >3 times greater than those observed in Ontario, although at a lower density 1.5 snakes/km².

DeGregorio et al. (2018:1199) sampled rat snake populations latitudinally and longitudinally across 9 states (Maryland, Virginia, North Carolina, South Carolina, Florida, Tennessee, Illinois, Kansas, and Texas) and a population in Ontario, Canada. However, the authors do not report the area of their study areas to enable me to directly compare snake densities from my study with theirs. My estimates are conservative because they were derived from encounters with snakes only at wood duck nest boxes and not across the entire landscapes of our study areas.

Stickel et al. (1980) investigated black rat snakes in a natural setting at Patuxent Wildlife Research Center in Maryland for 35 years (1942–1976). They indicated that males were captured more frequently than females, as I experienced at Lake Moultrie. Previously marked females were recaptured, on average, every 3.2 years, and males every 2.1 years. Males were larger than females similar to my sexually dimorphic data for captured snakes from nest boxes. The largest male and female in Maryland was 177.4 cm and 151.6 cm long, respectively (Stickel et al. 1980). The largest male and female in my study was 193.0 cm and 168.0 cm, respectively.

However, I measured snakes from the tip of the snout to the tip of the tail. Stickel et al. (1980) and others measured snakes from the tip of the snout to the vent, which is the standard measurement for snakes because total length can be biased by partial tail loss or injury (Blouin-Demers 2003, DeGregorio et al. 2018). To correct my measurements, I have obtained precise regression equations ($r^2 > 0.90$) from Dr. Gabriel Blouin-Demers (University of Ottawa, Ontario, Canada) to estimate snout-vent length from total length for male and female rat snakes, based on data presented by Blouin-Demers and Weatherhead (2021). I plan to generate estimates from snout-vent length using these equations and incorporate these data for comparisons in publications from my thesis.

Ashton and Feldman (2003:1156) documented inconsistencies in Bergmann's Rule for squamates, and DeGregorio et al. (2018) found rat snake length also did not increase with latitude to follow this rule. They reported that some of the largest snakes were found at the center of the geographic range for the species (i.e., Maryland, Tennessee, Virginia, South Carolina) while some of the smallest occurred at the edges of

the species' geographic range (i.e., Ontario, western Kansas, southern Florida, and Texas). In Kansas, Fitch (1963) documented relatively large male and female snakes, the largest individual male and female were 153 cm and 140 cm from snout to vent, respectively, and they observed a 4:1 hatching ratio of males to females. Stickel et al. (1980) observed a 1:1 ratio among young snakes (4-8 years old). Alternatively, among older snakes (10+ years), males outnumbered females 50:1 (Stickel et al. 1980). In my study, I observed a 3:1 male:female sex ratio at Lake Moultrie similar to Fitch (1963) in Kansas, while sex ratios observed at the North Carolina sites were nearly 1:1.

Stickel et al. (1980:9) observed that females were shorter than males. These authors also suggested that snakes continually grow, with males and females' growth rates slowing, on average, at about 6.8 and 6.6 years of age, respectively. Stickel et al. (1980) suggested some of the largest snakes in their study could be as old as 22 years. Using the aforementioned regression equations to estimate snout-vent lengths of male and female snakes in my study, I will attempt to investigate age structure of captured snakes at the North Carolina sites and Lake Moultrie to compare age and body size demographics among these populations.

Rat Snake Deterrent Experiment

Although nearly twice fewer rat snakes accessed nest boxes with Snake-A-Way[®] pellets, preliminary results indicated pellets did not significantly deter rat snakes from entering nest boxes in the method applied. In another study of the effects of the same pellets on gopher snakes (*Pituophis catenifer*), Marsh (1993) applied the pellets in a 30.5 cm band (per instructions on the packaging) on the ground. All 11 gopher snakes passed

the band at least once, and one snake crossed the band 5 times within one hour. Additionally, researchers observed two small western rattlesnakes (*Crotalus atrox*) did not cross the band, while two larger western rattlesnakes did cross (Marsh 1993). Ferraro (1995) used the two active ingredients in Snake-A-Way[®] (naphthalene and sulfur) separately, but neither ingredient alone deterred plains garter snakes (*Thamnophis radix*). Increased sample size from continued experimentation in 2022 will provide greater statistical power to test deterrence capacity of pellets at my study sites.

We found 3 dead hens on Lake Moultrie, the most recent found in March 2022. Hens killed by black rat snakes have been documented in the literature (Fendley 1980, Hansen 1971, Stewart 1981, Hester and Dermid 1973), but snakes generally do not pose a threat to incubating hens. Characteristics of hen mortality from rat snakes include wet matted feathers from snake saliva around the head and twisted necks (Stewart 1981, Hester and Dermid 1973).

Snake Capture Distributions

Snake capture frequencies were greatest in the northeast sections of Lake Mattamuskeet in North Carolina and on Lake Moultrie in South Carolina (Appendix D). At Lake Moultrie, this phenomenon may have been due to the fact that the northeast section of the lake has the greatest density of boxes, hence the likelihood of encountering snakes where boxes were most dense. This is consistent with Hansen's (1971) finding that some snakes learn to search for nest boxes as a reliable source of eggs. At Lake Mattamuskeet, the northeast section of the lake is dominated by bald cypress (*Taxodium distichum*) which grow closely together and may facilitate snake access to nest boxes.

Woodpecker Deterrent Experiment

Preliminary results indicated that placing a hawk decoy on wood duck nest boxes after 12 days of clutch incubation may be successful in deterring woodpeckers from depredating wood duck eggs. Lukas et al. (2020) indicated that visual bird deterrents, such as hawk decoys, are a useful method for deterring certain birds in agricultural areas. There is no example in the literature of hawk decoys being used to deter woodpeckers from entering wood duck nest boxes. However, because an incubating hen has invested large amounts of time and energy into her nest ≥12 days, she may not abandon her clutch (Bellrose and Holm 1994). Decoys in my study were placed atop boxes for 15-20 days during incubation and removed once nest fate was recorded (1-5 days after tagging ducklings).

Management and Research Implications

Rat snakes encountered at Lake Moultrie in South Carolina and Mattamuskeet and Roanoke River NWRs in North Carolina averaged 1.45 m in length; these individuals can surmount predator guards to gain access to nest boxes. Removal of vegetation is a good practice to prevent vegetation from encroaching and concealing boxes. However, vegetation removal alone was inadequate to deter snakes from accessing boxes. As mentioned in Chapter 1, predator guards should be checked every year for rust and weathering and replaced or repaired when necessary. Sealing gaps between the predator guard and post is a temporary fix until predator guards need to be totally replaced.

Another possible strategy to deter snakes from entering boxes may be to widen the base of predator guards and flaring out the cone to increase the distance from the post to the

base of the predator guard to a minimum of 1.5–2 m to exceed length of most snakes encountered in my study. Additionally, further study is needed to determine the effectiveness of Snake-A-Way[®] granules in repelling rat snakes from wood duck nest boxes. Distance between shorelines and nest boxes with and without snake presence should be examined to determine a possible optimal distance away from shorelines to place boxes. I have these data for each field site and will be conducting analyses apart from this thesis to investigate relationships between successful and unsuccessful nest fates and nest-box and microhabitat variables. Knowing if large snakes are in an area could assist managers in proper box placement and determining effective predator guard size and type (i.e., metal cone, stovepipe baffles).

If woodpecker depredation is resulting in significant (e.g., $\geq 10\%$) loss of eggs as recorded in my study, possible management strategies may include placing a hawk or other raptor decoy atop the box/post after ≥ 12 days of incubation to reduce the chance of woodpeckers entering nest boxes and depredating eggs. Additionally, I placed the hawk decoy on three nest boxes with current woodpecker depredation of eggs but with <12 days of incubation and found that the hen did not abandon, and two of three clutches hatched. Given the small sample sizes in my study, further study is needed to examine the effectiveness of hawk decoys deterring woodpeckers from entering wood duck nest boxes before and after 12 days of incubation.

Snakes and woodpeckers depredate eggs at different rates at varying locations. There is a need for continued recruitment data and sensitivity analysis to determine if rat snakes, woodpeckers, and other agents of nest loss are decreasing recruitment from boxes in South

Carolina, North Carolina, and throughout the range of North American wood ducks (Hepp et al. 2020). Hundreds of thousands of wood duck boxes exist in North America (Bellrose and Holm 1994). Although our regional study of box-nesting wood ducks and other cavity-nesting ducks in the Southeast is unique and will inform scientists and managers whether these populations are sustaining themselves without immigration of conspecifics (sensu Hepp et al. 2020), there is need for a coordinated cross-flyway investigation of populations of wood ducks and other waterfowl species using artificial nest structures.

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Table 2.1 Number of wood duck nest boxes selected for Snake-A-Way[®] pellets or Bird-B-Gone[®] raptor treatments or none (control) for snake and woodpecker deterrent experiments at Lake Moultrie, South Carolina, and the pooled North Carolina sites from January–July 2021.

	Pel	llets	Rapto	r decoy
	Lake Moultrie	North Carolina sites ^b	Lake Moultrie	North Carolina sites ^b
Treatment	7 ^a	8 ^a	9	8
None	7	9	7	9

^aAn additional 6 boxes were added to the treatment sample size for the snake deterrent experiment in each state during 2021.

^b Data pooled for North Carolina sites (Mattamuskeet and Roanoke Rivers National Wildlife Refuges and Heron Bay).

	South C	Carolina ^a	North C	arolina ^a
	2020	2021	2020	2021
N captured	44	50	1	28
N marked	44	34	1	28
N marked previously	0	16	0	0
% recaptured	0	36.4	0	0
N recaptured same year	21	14	0	7

Table 2.2. Number of rat snakes captured, marked, and recaptured from nest boxes at Lake Moultrie, South Carolina, and the pooled North Carolina sites from January–July 2020–2021.

^a South Carolina site is Lake Moultrie; data pooled for North Carolina sites (Mattamuskeet and Roanoke Rivers National Wildlife Refuges and Heron Bay).

Table 2.3 Number of nest boxes treated with socks containing Snake-A-Way® pellets, empty				
socks (control), and rat snake encounters or not at Lake Moultrie, South Carolina, and the pooled				
North Carolina sites from January–June 2021.				

	Treated	Not treated	Total
Snake encounter	14	6	20
None	25	9	34
Total	39	15	54

	Treated	Not treated	Total
Success	11	6	17
Failure	1	8	9
Total	12	14	26

Table 2.4. Number of successful and failed wood duck nests from boxes treated and not with the raptor decoy at Lake Moultrie, South Carolina, and the pooled North Carolina sites from January–July 2021.

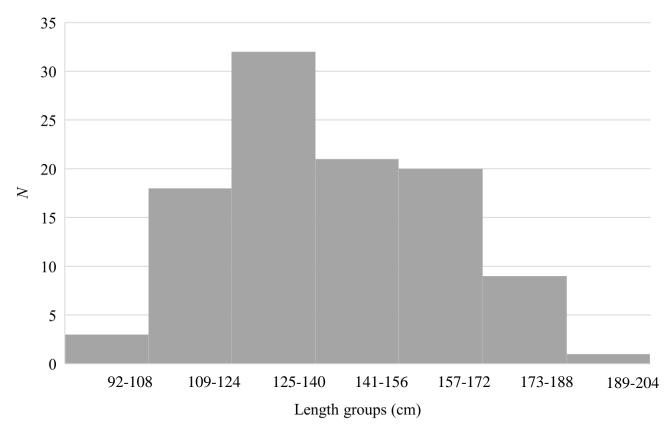


Figure 2.1. Histogram of lengths of snakes captured in wood duck nest boxes from January–June at Lake Moultrie, South Carolina (2020–2021), and the pooled North Carolina sites (2021).

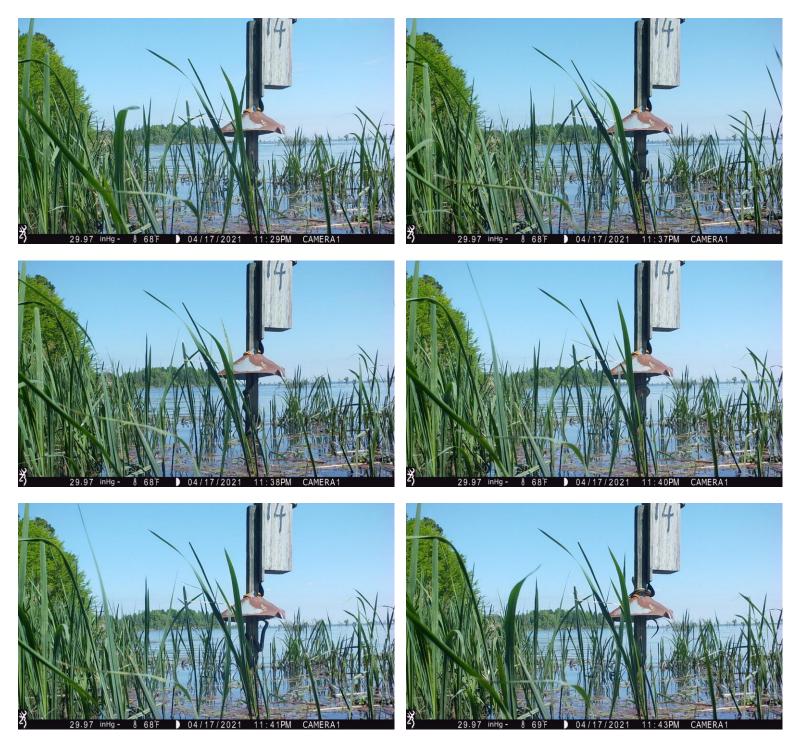
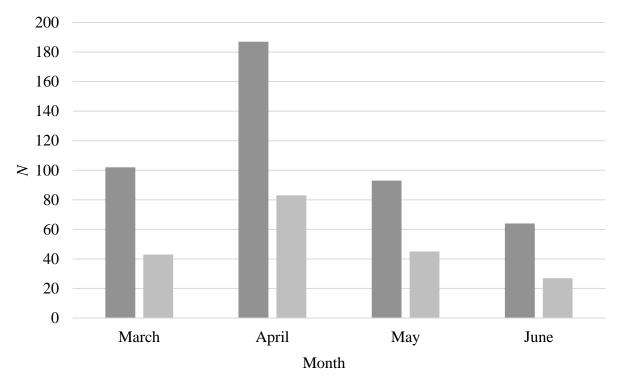
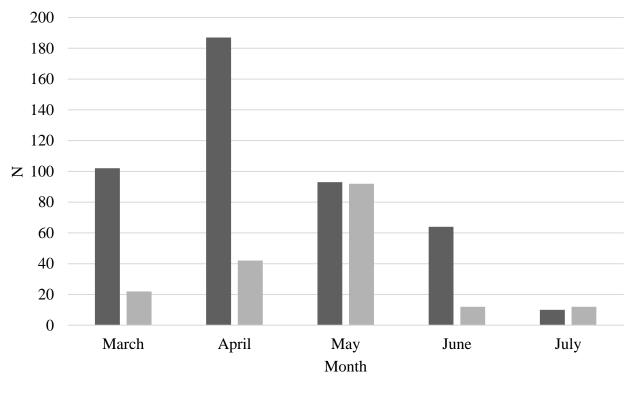


Figure 2.2. Selected trail camera photos of a rat snake entering a wood duck box on Lake Moultrie, South Carolina, 2021. This trail camera captured over 200 photos of this particular snake circumventing the predator guard and climbing the post, where it took the snake 23 minutes to reach the top of the nest box. This snake was captured on 4/19/21 with 6 eggs in its belly. It was originally tagged on 3/5/20 at this box with another subsequent recapture on 5/8/20.



■ N eggs depredated ■ N captures

Figure 2.3. Summary of snake capture events and egg predation frequency from all captured snakes during January–June at Lake Moultrie, South Carolina (2020–2021), and the pooled North Carolina sites (2021).



■ N depredated by snake ■ N depredated by woodpecker

Figure 2.4. Observed number of eggs depredated by black rat snakes (i.e., protrusions in stomach of captured snakes) and woodpeckers (i.e., holes pecked in eggs) January–July 2020 and 2021 at Lake Moultrie, South Carolina, and the pooled North Carolina sites.

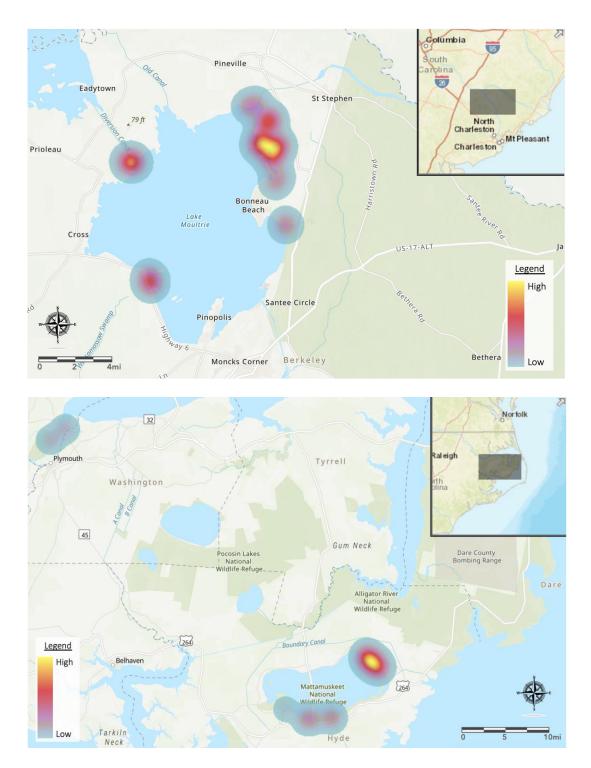


Figure 2.5. Heat map (concentrations) from GPS coordinates of rat snake captures and recaptures in wood duck nest boxes on Lake Moultrie, South Carolina (top) and in North Carolina (bottom) on Roanoke River (bottom image, top left) and Mattamuskeet NWRs and Heron Bay from January–June 2020–2021.



Figure 2.6. Deceased wood duck hen found in an artificial nest box on Lake Moultrie, South Carolina, 14 March 2022. This hen likely died as a result of rat snake constriction and suffocation, as her feathers on her head and neck are displaced and appear to be covered in dried saliva, likely from a rat snake putting its mouth over the hen's head to try and eat it.

APPENDICES

Appendix A

Wood Duck Box Locations on Lake Moultrie, South Carolina



Appendix B

Wood Duck Box Locations on Mattamuskeet National Wildlife Refuge and Heron Bay, <u>North Carolina</u>



Appendix C

Wood Duck Box Locations on Roanoke River National Wildlife Refuge, North Carolina



Appendix D

