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The Trophic and Spatial Ecology of the Southern Flying Squirrel (*Glaucomys Volans*) and Non-Lethal Deterrent Methods

Robert Timothy Meyer

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The trophic and spatial ecology of the southern flying squirrel (*Glaucomys volans*)
and non-lethal deterrent methods

By

Robert Timothy Meyer

A Thesis
Submitted to the Faculty of
Mississippi State University
in Partial Fulfillment of the Requirements
for the Degree of Master of Science
in Wildlife, Fisheries, and Aquaculture
in the College of Forest Resources

Mississippi State, Mississippi

May 2018

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2018

The trophic and spatial ecology of the southern flying squirrel (*Glaucomys volans*)
and non-lethal deterrent methods

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Southern flying squirrels (SFS; *Glaucomys volans*) are known kleptoparasites on the endangered red-cockaded woodpecker's (RCW; *Picoides borealis*) cavities and cost land managers time and money to control, and the ecology of SFS in habitats managed for RCWs is poorly understood. This study is designed to obtain a better understanding of the general ecology of SFSs surrounding RCW habitat and provide possible non-lethal deterrent methods to prevent harmful interactions between SFSs and RCWs. Spatially-explicit capture-recapture showed generally higher SFS densities and habitat associations outside of RCW cluster partitions. Stable isotope analysis of SFS diets across Mississippi and Alabama revealed a narrow dietary breath relative to food items, thus habitat management may still be viable option to control SFSs. Lastly, investigation of odor deterrents resulted in the greatest avoidance of cavities containing gray rat snake (*Pantherophis spiloides*) feces and warrants further investigation.

DEDICATION

I dedicate my research to my family whose love and support never went unnoticed and in memory of my mother. I love you more.

ACKNOWLEDGEMENTS

This project would not have been possible if it wasn't for the encouragement of a select few. I would like to express my gratitude to my advisor Dr. Scott Rush who saw potential in me and granted me the opportunity to pursue a degree at MSU. I would like to thank the U.S. Fish and Wildlife Service at the Sam D. Hamilton Noxubee National Wildlife Refuge for my housing as well as their financial and logistical support through the years. Technical support from USDA APHIS as well as the advice of Dr. Brian Dorr was greatly appreciated. Additionally, the passion and interest in this project shown by Dr. Guiming Wang and wildlife biologists Steven E. Lewis and Eric C. Soehren have attributed greatly to the quality and scope of the research. The collective minds of these people have significantly improved this project and I aspire to hold the same passion and dedication to wildlife science as they do.

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CHAPTER I

INTRODUCTION TO RED-COCKADED WOODPECKER (*PICOIDES BOREALIS*) AND SOUTHERN FLYING SQUIRREL (*GLAUCOMYS VOLANS*) CONFLICT

Red-cockaded Woodpecker Biology and Conservation

The red-cockaded woodpecker (RCW; *Picoides borealis*) is an endemic species to coastal pine habitat across the southeastern United States (U.S. Fish and Wildlife Service [USFWS] 2003). The RCW is a cooperative breeder, typically living in groups of two or more individuals consisting of a breeding pair and one or more ‘helpers’ that will incubate the eggs or feed the young of the breeding pair (Ligon 1970, Conner and Rudolph 1995). An RCW group will often forage together during the day and roost close to each other at night with each RCW occupying a separate tree. The area encompassing the group of trees is referred to as the ‘cluster’ and is used by the RCW group for many years (Walters 1990). Unique among woodpeckers, the RCW creates cavities in living pine trees (*Pinus sp.*) typically weakened by the red heart-rot fungus (*Phellinus pini* [Thore ex. Fr.]). Cavity trees are often ≥ 60 years old and possess a core that has no active sap flow (Conner and Locke 1982; Conner and O’Halloran 1987; Nebeker et al. 1995). It can take an RCW group several months to more than a year to create a cavity (Jackson et al. 1979; Conner and Rudolph 1995).

The RCW is currently listed as endangered and has been since 1970 (USFWS 2003). There are many reasons for their decline but chief among them is the lack of

suitable habitat. The RCW is historically associated with old growth pine savannahs of which, few remain (Ware et al. 1993). The open system comprising of a low basal area of trees and herbaceous understory promotes foraging habitat and the older trees provide characteristics suitable for the RCWs to excavate cavities. Past and current timber harvest practices limit the availability of these more suitable trees to RCWs. These include the timber harvest following early settlement of North America, current short timber harvest rotations promoting trees less than 30yrs in age, replanting land with even-aged pine stands such as loblolly pine (*Pinus taeda*), and fire suppression (Ware et al. 1993; Conner et al. 2001). Fire is required to suppress hardwood midstory growth, a habitat feature known to cause cluster abandonment (Conner and Rudolph 1989). For these reasons, the RCW is now typically restricted to small and isolated areas across their former range in the southeast (James 1995).

Conflicts with Southern Flying Squirrels

Southern flying squirrels (SFS; *Glaucomys volans*) commonly occupy RCW cavities (Rudolph et al. 1990; Loeb 1993). The SFS may displace RCWs from their cavities and may negatively affect RCW nest success (Dennis 1971; Loeb and Hooper 1997; Laves and Loeb 1999). SFSs are typically the most common occupant of RCW cavities besides RCWs themselves (Rudolph et al. 1990). The SFS can be aggressive (Weigl 1978) and there is no published record documenting an RCW evicting a SFS from its cavity. Thus, once occupied by a SFS, the cavity is unavailable until the SFS vacates. In this way, SFSs may take the most suitable cavities (but not necessarily always) from the RCW and may occupy multiple cavities in a cluster. The RCWs are then forced to roost in less suitable cavities, or exposed on tree limbs. Additionally, if a

SFS occupies an RCW during the breeding season, they may destroy the eggs of the RCW that were in that cavity. When a nest is destroyed, an RCW pair can re-nest and thus may compensate for the loss of the previous nesting attempt(s). However, successive nests may be destroyed as well, increase the energetic cost to the RCW colony (the breeders and helpers), and delay the time the RCWs fledge which could put them at a disadvantage (Ingold 1996).

The SFS completely overlaps the RCWs range, thus these negative interactions occur wherever there are RCWs. Some evidence suggests that smaller populations may suffer larger impacts from SFS kleptoparasitism (Laves and Loeb 1999; Kappes 2008). It is likely that these species existed together historically and there was enough pine savannah habitat and available cavities that SFSs posed a less significant threat. Past and current land practices however have left many RCW populations small and isolated (James 1995), thus SFSs pose a threat to the recovery of the species across much of their range.

Dealing with Kleptoparasites

Once an RCW cavity is usurped by a SFS, it becomes unavailable to the RCW until the SFS vacates. As to date, there has been no published record of an RCW evicting a SFS from a cavity (Dennis 1971; Montague et al. 1995). Thus, managers forcefully eject or euthanize SFSs found in RCW cavities, which can be both costly and time-consuming (USFWS 2003). The efficacy of these removal efforts is uncertain but if a SFS is euthanized it is reasonable to assume that specific SFS will not reoccupy the cavity.

Effective management strategies against nuisance species should take advantage of fundamental aspects of their behavior and ecology. Management actions that are not properly informed may result in unintended consequences. In the case of the SFS, without knowing basic aspects of their biology, i.e. reproductive rates, movements, social structure etc. removal efforts may not impact population sizes around RCW clusters in a meaningful way. With additional knowledge of SFS life history characteristics, RCW managers may capitalize on effective control methods for this common kleptoparasite.

References

- Dennis, J. V. 1971. Species using red-cockaded woodpecker holes in northeastern South Carolina. *Bird-banding* 42:79–87.
- Conner, R. N., B. A. Locke. 1982. Fungi and red-cockaded woodpecker cavity trees. *The Wilson Bulletin* 94:64–70.
- K. A. O'Halloran. 1987. Cavity-tree selection by red-cockaded woodpeckers as related to growth dynamics of southern pine. *The Wilson Bulletin* 99:398–412.
- D. C. Rudolph. 1995. Excavation dynamics and use patterns of red-cockaded woodpecker cavities: relationships with cooperative breeding. Pp. 343–352 in *Red-cockaded woodpecker: recovery, ecology, and management* (D. L. Kulhavy, R. G. Hooper, and R. Costa, eds.). Center for Applied Studies, College of Forestry, Stephen F. Austin State University. Nacogdoches, Texas, USA.
- D. C. Rudolph. 1989. Red-cockaded woodpecker colony status and trends on the Angelina, Davey Crockett, and Sabine National Forests. USDA Forest Service Research Paper SO-250, New Orleans, Louisiana.
- D. C. Rudolph, and J.R. Walters. 2001. *The red-cockaded woodpecker: surviving in a fire maintained ecosystem*. University of Texas Press, Austin, TX.
- Ingold, D. J. 1996. Delayed nesting decreases reproductive success in northern flickers: implications for competition with European starlings. *Journal of Field Ornithology* 67:321-326.
- James, F. C. 1995. The status of the red-cockaded woodpecker in 1990 and the prospect for recovery. Pp. 439-451 in *Red-cockaded woodpecker recovery, ecology, and management* (D.L. Kulhavy, R. G. Hooper, and R. Costa, eds.). College of Forestry, Stephen F. Austin State University, Nacogdoches, Texas.
- Kappes, J.J. Jr. 2008. Cavity number and use by other species as correlates of group size in red-cockaded woodpeckers. *Wilson Ornithological Society*. 120: 181–189.
- Ligon, D. J. 1970. Behavior and breeding biology of the red-cockaded woodpecker. *The Auk* 87:255–278.
- Loeb, S. C. 1993. Use and selection of red-cockaded woodpecker cavities by southern flying squirrels. *Journal of Wildlife Management* 57:329–335.

- Montague, W.G., J.C. Neal and J.E. Johnson. 1995. Techniques for excluding southern flying squirrels from cavities of Red-cockaded Woodpeckers. Pages 401–409 in D. L. Kulhavy, R. G. Hooper, and R. Costa, editors. Red-cockaded Woodpecker: recovery, ecology, and management. Center for Applied Studies, College of Forestry, Stephen F. Austin State University. Nacogdoches, Texas, USA.
- Nebeker, T. E., M. Pelligrine, R. A. Tisdale, and J. D. Hodges. 1995. Site/stand factors associated with red-cockaded woodpecker colonies on the Noxubee National Wildlife Refuge, Mississippi. Pages 431–435 in D. L. Kulhavy, R. G. Hooper, and R. Costa, editors. Red-cockaded woodpecker: Recovery, ecology, and management. Center for Applied Studies, College of Forestry, Stephen F. Austin State University. Nacogdoches, Texas, USA.
- Rudolph, D. C., R. N. Conner, and J. Turner. 1990. Competition for red-cockaded woodpecker roost and nest cavities: effects of resin age and entrance diameter. *Wilson Bulletin* 102:23–26.
- U.S. Fish and Wildlife Service. 2003. Recovery plan for the red-cockaded woodpecker (*Picoides borealis*): second revision. U.S. Fish and Wildlife Service, Atlanta, GA.
- Walters, J. R. 1990. The red-cockaded woodpecker: a “primitive” cooperative breeder. Pgs. 67-101 in Cooperative breeding in birds: long term studies of ecology and behavior (P.B. Stacey and W. D. Koenig, Editors). Cambridge University Press, Cambridge, United Kingdom.
- Ware, S., C. Frost, and P. D. Doerr. 1993. Southern mixed hardwood forest: the former longleaf pine forest. (W.H. Martin, S.G. Boyce, and A. C. Echternacht, Editors). Biodiversity of the southeastern United States: lowland terrestrial communities. John Wiley and Sons. Inc., New York, NY.
- Weigl, P. D. 1978. Resource overlap, interspecific interactions and the distribution of the flying squirrels *Glaucomys volans* and *G. sabrinus*. *American Midland Naturalist* 100:83-96.

CHAPTER II

SOUTHERN FLYING SQUIRREL (*GLAUCOMYS VOLANS*) SPATIAL ECOLOGY
WITHIN RED-COCKADED WOODPECKER (*PICOIDES BOREALIS*)
CLUSTERS IN EAST-CENTRAL MISSISSIPPI

Abstract

Southern flying squirrels (SFS; *Glaucomys volans*) displace and destroy the nests of the endangered red-cockaded woodpecker (RCW; *Picoides borealis*) yet few studies have assessed how SFS use habitats occupied by RCWs. The objective of this study was to obtain seasonal density, home range estimates, and vegetation associations for SFSs within ('cluster partition') and outside ('control') RCW cluster partitions at the Sam D. Hamilton Noxubee National Wildlife Refuge. Seasonal spatially explicit capture-recapture models revealed higher densities of SFSs in control than in cluster partition trap grids with a significant difference in winter. Kernel density estimates (95% KDE) of average home range size for each season varied but were not statistically different (ANOVA, $P = 0.19$) nor were they different between cluster partition and control grids (t-test, $P = 0.98$). A linear mixed-effects model of the core (50% KDE) relative to the non-core (50–95% KDE) home range for the relative importance value for oaks (*Quercus* spp)

showed no significant differences between seasons nor between control and cluster partition grids ($P > 0.05$). These results indicate that SFSs may select cluster cavities based on more than just habitat and cavity characteristics alone, suggesting other factors such as food availability or predator avoidance may also be involved.

Introduction

The red-cockaded woodpecker (RCW; *Picoides borealis* [Vieillot, 1809]), is a federally listed endangered species endemic to pine forests of the southeast United States (U.S Fish and Wildlife Service, 2003). RCWs create cavities in living pine trees, close to one another, in areas referred to as ‘clusters’ (Walters 1990). RCWs typically create these cavities in pines inoculated with red heart-rot fungus (*Phellinus pini* [Thore ex. Fr.]). Cavity trees are often ≥ 60 years old (Conner and Locke 1982; Conner and O’Halloran 1987; Nebeker et al. 1995) and it can take an RCW several months to more than a year to create a cavity (Jackson et al. 1979; Conner and Rudolph 1995). Past and current timber harvest practices limit the availability of these more suitable trees to RCWs, due to shorter 30 yr harvest rotations of commonly planted loblolly pine (*Pinus taeda*), and even-aged stand management which does not support RCWs in a long-term, sustainable manner (Ware et al. 1993; Conner et al. 2001). Thus, establishing and maintaining nest and roost cavities is an important part of recovery efforts for RCWs over much of their range.

Southern flying squirrels (SFS; *Glaucomys volans*) are known kleptoparasites of RCW cavities (Loeb 1993; Conner et al. 1996; Borgo et al. 2010). The SFS may displace RCWs from their cavities and may negatively affect RCW nest success (Dennis 1971; Loeb and Hooper 1997; Laves and Loeb 1999). When a nest is destroyed, a RCW pair can re-nest and thus may compensate for the loss of the previous nesting attempts. However, successive nests may still be destroyed, increase the energetic cost to the RCW colony, and delay the time the RCWs fledge which could put them at a disadvantage. Once a RCW cavity is usurped by a SFS, it becomes unavailable to the RCW until the SFS vacates. As to date, there has been no published record of an RCW evicting a SFS from a cavity (Dennis 1971; Montague et al. 1995). Thus, there is need for managers to forcefully eject or euthanize squirrels found in RCW cavities (U.S. Fish and Wildlife Service [USFWS] 2003), a practice that is both costly and time-consuming.

Movement and behavior of SFSs around RCW cluster partitions (the area maintained around the cluster trees) is poorly understood. SFS home-ranges have been measured in a variety of habitat types and geographic locations (Bendel and Gates 1987; Fridell and Litvaitis 1991; Stone et al. 1997; Garroway et al. 2013) but little is known about SFS ecology within RCW cluster partitions. Even fewer studies have examined the ecology of SFSs in areas outside these clusters, especially in loblolly pine dominated RCW habitat.

Studies of SFS ecology have largely been limited in focus with reference to features of RCW cluster trees and the area around cluster centers (Loeb 1993; Conner and Rudolph 1996; Loeb et al. 2012). If SFSs regularly use features of habitats outside of RCW clusters then the results of existing studies are limited by the spatial scope of these assessments. Consequently, effective management of SFS relative to RCW should likely incorporate an understanding of SFS response to conditions within and outside of RCW clusters. Properties of these ecosystems can be managed to best support RCW populations by identifying habitat features associated with SFS abundance and movement, and lowering harmful interactions between the two species. To this end, I obtained information on SFS density, nest selection, home range characteristics, and habitat associations across seasons within and outside of cluster forest partitions. All work with SFS was carried out under MSU IACUC protocol #s 15-072, 16-037, and 16-490, as well as state and federal permits.

Methods

Study Site

I conducted this study on the USFWS Sam D. Hamilton Noxubee National Wildlife Refuge (hereafter; Noxubee) in east-central Mississippi. The Noxubee Refuge is 19,513 ha and consists of 44% upland pine, 40% bottomland hardwood. Approximately ~16% of the landcover within Noxubee is open fields, upland hardwoods and water

bodies. Bottomland hardwood species are mainly white oak (*Quercus alba*), southern red oak (*Q. falcata*), water oak (*Q. nigra*), mockernut hickory (*Carya tomentosa*), shagbark hickory (*C. ovata*), sweetgum (*Liquidambar styraciflua*), black gum (*Nyssa sylvatica*), and flowering dogwood (*Cornus florida*). The majority of upland pine habitat is dominated by loblolly, with occasional short leaf pine (*P. echinata*) intermixed with some hardwoods (USFWS 2014).

Noxubee is managed for RCW through forest thinning, stem injection of herbicides, and prescribed fire with the intent of creating Good Quality Foraging Habitat (GQFH; USFWS 2003). The GQFH is characterized by a predetermined basal area (BA) of pine of separate size classes (see USFWS 2003 for details), with little or no hardwood midstory, and < 30% of the canopy hardwood. Prescribed fire is performed throughout the year on the Noxubee Refuge in a mosaic pattern, with three-year cycles for each burn area. Fires are targeted low intensity, aimed at setting back succession without scorching pyrogenic members of the plant community. Noxubee manages for GQFH around RCW clusters within a forest partition, an area delineated by firebreaks and managed as a unit (hereafter; 'cluster partition'). Noxubee has not fully achieved GQFH but continues to manage towards this for forest partitions containing RCWs.

Squirrel Capture and Telemetry

I set grids for sampling SFS within and outside (control) of RCW cluster partitions, enabling comparison of SFS density estimates between these two area types. I randomly selected eight of the 29 active clusters at Noxubee within which to set trap grids.

Each trap grid was comprised of 25 bluebird boxes in a 5 x 5 grid arrangement with ~35m spacing between boxes (Fig. 1.1). Trap boxes were constructed of pinewood and had internal dimensions 10 x 10 x 25cm with a 3.8cm diameter entrance hole positioned on the right side of the box close to the top of the box and near the bole of the tree. Trap boxes were affixed to trees, 4 – 5m above the ground. Trap grids set in cluster partitions encompassed some, or all, active RCW trees.

Boxes within clusters partitions were not affixed directly to RCW cavity trees, an effort taken to prevent direct interactions between SFSs and RCWs. Increasing the number of cavities in active RCW clusters has not been shown to detrimentally affect RCW reproductive success (Borgo et al. 2006) thus I did not anticipate any negative interactions of placing boxes within active clusters. Trees selected to receive boxes were those closest to the proximate grid point (Fig. 1.1). Control grids were set in a random cardinal direction from the cluster grid with 200m between the borders of each grid.

Trap grids were deployed at least one week prior to trapping. Boxes were left open to allow SFSs to acclimate to their presence and boxes were kept open during the

duration of the study. Trap grids were checked once per season for five consecutive days, with all checks conducted during daytime. I delineated seasons as spring (March–May), summer (June–August), fall (September–November) and winter (December–February), starting with fall of 2015 and continuing until the fall of 2017. For trapping, I focused efforts on the middle month of each season. For checking traps, I used a Hastings tree pole (Hastings, Michigan) with a camera mounted to the top (IBWO, Little Rock, Arkansas), and a wireless monitor to check for the presence of SFSs in boxes.

When a SFS was observed within a box, I plugged the entrance hole of that box using a cloth, thus preventing the squirrel from escaping. The box was then removed from the tree and lowered to the ground for processing. While trapping, any pups found in boxes were aged to estimate date of birth using characteristics described by Sollberger (1943). Captured SFSs over 30g were injected with a passive implantable transponder (PIT; HPT12, Biomark, Boise, Idaho), weighed, sexed, and aged. Each captured SFS was aged based on hind-foot length and body weight, as described in Laves and Loeb (2006). Once measured and marked, squirrels were returned to the box from which they were captured, and the box returned to its position on the tree.

I selected one adult SFS among those captured within a grid to receive a 1g radio-transmitter collar (M1420, Advanced Telemetry Systems, Isanti, Minnesota). I applied approximately ten radio-transmitters per season to SFSs in an even ratio of males to females for each of the selected cluster partition and control grids (five collars each for

cluster and control grids) providing an examination of differences in SFS movements among seasons, between grid types, and between sexes. Individual SFSs were not collared twice during the course of the study nor were two SFSs tracked on the same grid at the same time. At least one SFS was tracked at each grid except one. Transmitters were custom fit to each SFS using methods similar to those employed by Adams and Campbell (1996).

Radio-collared SFSs were tracked no earlier than three days after initial capture to ensure that fixes were not biased by atypical movements brought on through the stress of capture/collaring. Each SFS's location was determined once per day, during daytime, to obtain cavity selections, and again, once every three hours at night, to obtain nighttime movements. Nighttime telemetry took place at least a half hour after sundown and a half hour before sunrise to ensure fixes are taken when SFSs are most likely to be active. I rotated the time of night when taking fixes such that I did not sample the SFS during the same time of night for consecutive nights. Night-time telemetry was conducted until ≥ 30 points total were collected or as long as the battery within each collar allowed (typically, 60–90 days). I collected location data for SFS over an approximately three-week period, usually during the later portion of the season. Telemetry was conducted in all weather conditions except for conditions considered hazardous to personal safety.

To obtain locations for each SFS I employed a Biotracker receiver (Lotek, Ontario, Canada) equipped with a three-element yagi antenna using a homing method

(Mech 1983). A SFS's location was determined by moving in a spiral motion inwards towards the SFS until a predetermined signal strength was obtained that indicated a distance of 30m away (unique gain for each collar). In this way, I was able to use bearing, distance, and current location (GPSmap 60CSx, Garmin, Olathe, Kansas) to estimate the true location of the SFS so as to reduce potential bias of their movements through my presence. Only nighttime fixes were used in home range estimation to include mainly locations where the SFS were active and avoid the high degree of autocorrelation associated with using the same daytime rest site that includes boxes, cavities, or drays (hereafter; 'nests').

All statistical analyses were carried out in program R (R Core Development Team, 2016). I calculated SFS density for each of nine trap sessions (composed of five consecutive trap occasions) using spatially explicit capture-recapture modeling (SECR; *secr* R package; Efford 2016) to determine population density for clusters ($n = 8$) and non-cluster controls ($n = 8$). To improve accuracy of density estimates, trap boxes that were inoperable due to mechanical damage or wasp occupancy were temporarily censored from trap histories during periods when they were inoperable. Seasonal home range sizes were averaged over the duration of the study and used to inform the sigma parameter of each SECR model. I chose the detection function for the SECR model by comparing half-normal, exponential, and hazard rate detection functions within the null

model (fixed model parameters; Efford 2016) selecting the detection function with the lowest ΔAIC (Akaike 1973).

Increasing the number of cavities in the environment can artificially increase the probability of capture if the environment is cavity deficient. If there is a difference between cluster partition and control grids then density estimates may be biased high towards the cavity deficient grids. Since it is unknown how many available cavities exist in either cluster partition or control grids, I compared the probability of capture of grid pairs as derived from the top performing model provided by SECR.

I created fixed-kernel density estimates (KDE) for each SFS's home range using the R package *adehabitatHR* (Calenge 2006). I used least squares cross validation to determine the smoothing parameter to be used for each home range. Based on the KDE for each SFS, I sampled vegetation in the core home range (50% KDE) and in the peripheral home range (95% KDE, excluding core home range) so that I could compare habitat metrics that correlate with SFS activity.

Vegetation Analysis

When finding vegetation characteristics associated with animal use, many studies are forced to define what may or may not be accessible to the animal. To reduce potential bias in what may be an arbitrary delineation, I only sampled vegetation within areas used by each SFS, with use intensity used to determine important vegetation characteristics.

Here, I generated 5 – 20 random points using ArcMAP (Version 10.2; ESRI Arc Products) within each SFSs' core and non-core use-areas based on the 50%KDE and 95%KDE of their home range, respectively (~40 points/SFS). Within these areas, using GIS (ArcMAP), I included as many vegetation points as the size of the home range could allow while also keeping a 30m buffer between points (Fig. 1.2).

Within each vegetation point I measured basal area (BA), total tree density, and the relative frequency of oaks (*Quercus* spp.), hickory (*Carya* spp.), and pine (*Pinus* spp.) using the point-centered quarter method (Smith and Smith 2001). Relative importance value (RVI) was estimated for oak, hickory, and pine by taking the average of the sum of relative dominance, relative frequency and relative density for each tree genus for each SFS. Midstory density was assessed by counting the number of stems between 10–25cm in diameter at breast height (DBH) within a 15m radius circular plot at each point. This range generally includes trees larger than saplings and lower than 10m tall that contain many limbs between the understory and canopy (Lockhart et al. 2010). Canopy cover was estimated at each point using a concave spherical crown densitometer (Forestry Suppliers Inc.).

Statistical Analysis

I compared the sizes of SFS home range among seasons using analysis of variance (ANOVA), with season as the independent variable and SFS home range size as

the dependent variable. I performed a student's t-test to determine if choice of cavity in the cluster or control grid affected SFS home range size. I also used a student's t-test to determine if SFS home range related to sex of the SFS. Prior to statistical assessment, I tested for violations to the requirements of normality of the data for application in t-tests and ANOVA, using Shapiro-Wilks tests and by observing Q-Q (quantile-quantile) plot for linear mixed effects models. I used an accumulation curve to determine the number of nests used by male and female squirrels over several days of tracking. I used a linear mixed effects model with the individual SFS as the random effect and the cumulative number of nests as the dependent variable with sex and the log-transformed number of observations as the fixed effects. In this way, I could project over a one month period to find the average number of nests used despite the discrepancy of the number of times each individual is able to be located. Resulting R^2 for mixed effects models were calculated based on methods described by Nakagawa and Schielzeth (2013). I used a proportion test to determine if use of artificial cavities (i.e. boxes) differed significantly between control and cluster SFSs.

Of the habitat characteristics sampled I chose to use the relative importance index (RIV) of oaks (*Quercus* spp.), as nuts from these trees are dominant, relative to other food items, incorporated in their diet (Harlow and Doyle 1990; Thomas and Weigl 1998). I also used midstory density as it is often cited as a reason for declines in RCW populations (USFWS 2003) and it may either facilitate or hinder SFS movements. I used

a mixed-effects generalized linear model (*lmer*, through package *lme4* in R [R Core Development Team, 2016]) with RIV for oaks or midstory density as the dependent variables and season, core/non-core home range, cluster/control grid as independent variables. Random-effects included the individual SFS and the specific trap grid where the SFS was caught. Error structure was individual SFS nested within trap grid, and with each model having a random intercept. Tukey's honest significant difference (Tukey's HSD) test was used post hoc for each linear model as a means to evaluate differences in RIV within SFS home range among seasons. All statistical tests were considered significant at $\alpha = 0.05$.

Results

Cluster partition and control areas differed in the basal area (BA) of pine and hardwood (paired t-test, $t_{2,36} = 3.89$, $P < 0.01$ and $t_{2,36} = -3.44$, $P = 0.01$, respectively). The average BA of pine and hardwoods of the cluster partition grids are 17.71 ± 2.51 m²/ha (95% CI and 5.44 ± 1.52 m²/ha, respectively). By contrast, the average BA of pine and hardwoods of the control grids are 10.78 ± 3.92 m²/ha and 12.64 ± 3.57 m²/ha, respectively.

I captured 334 unique SFSs over 14,346 trap nights during this study. Only six out of the sixteen grids were operational for the first fall (2015) season, but by the first winter (2015–2016) all trap grids were operational. Total detections ranged from 21–197 for 23–

136 unique individuals per trap session. All SECR models used exponential detection functions as they showed the lowest ΔAIC value when compared to hazard rate and half-normal detection functions in the null model (0, 3.21, and 35.94 respectively). The best model (lowest AIC) was the occasion model which represents a different probability of capture at each capture event (Table 1.3). I chose to use only the winter of 2016–2017 to compare capture probabilities between grid pairs as it had the most captures and thus could derive accurate estimates. The resulting capture probabilities did not differ significantly between cluster partition and control grid pairs (Fig. 1.4). SECR estimates of SFS densities showed that density is generally higher in control grids than in cluster grids, generally fluctuating around 0.15 SFSs/ha in control grids and 0.80 SFSs/ha in cluster partition grids across all seasons (Fig. 1.5). The SFS densities are likely biased low for the first two trap sessions since all trap grids were set up by the first winter (2015–2016). It is likely that fewer SFSs could be caught due to the unfamiliarity of the boxes to pregnant/lactating females that have restricted movement during the early stages of the young's development.

In the first winter I recorded no juvenile squirrels (56 captures total) however, the second winter resulted in 11.76%, 13.24%, and 75% of total captures for juveniles, subadults, and adults respectively (204 total captures). I found a total of 33 unique litters, of which 22 were estimated to be born from mid-September to mid-December and 11 were estimated to be born in early March to late April. I observed two females with two

litters within one year and observed one instance of a female born in the spring, reproducing the following fall.

I tracked a total of 54 SFSs but only 36 SFSs ($n = 20$ males, $n = 16$ females) had useable data for home range analysis (smoothing parameter converged and ≥ 30 locations were obtained within the season). To bolster the sample size for summer I tracked an additional three squirrels from other clusters that did not contain trap grids. Shapiro-Wilks test resulted in an approximately normal distribution for home range size across all SFSs ($P > 0.05$). These home range sizes did not differ to those of grid-caught cluster-using squirrels ($t_{2.97} = -0.28$, $P = 0.80$), thus I included them in the analysis. Sample sizes differed between spring, summer, fall, and winter ($n = 15$, $n = 7$, $n = 8$, $n = 6$, SFS tracked per season, respectively). Home range size did not differ between seasons (Fig. 1.3; ANOVA, $F_{3,31} = 1.67$, $P = 0.19$). Home range size did not differ between control or treatment grids ($n = 15$, $n = 21$ respectively; $t_{23.01} = 0.02$, $P = 0.98$) but was significantly different between males and females ($n = 20$, $n = 16$ respectively; $t_{33.78} = 3.05$, $P < 0.01$), with the male home range size ($9.43 \pm 1.84\text{ha}$ [$\bar{x} \pm 95\% \text{ CI}$]) 64% larger than female's ($5.74 \pm 0.15\text{ha}$). Home ranges ranged from 2.26–17.41ha with one male SFS which had a 95% home range size of 65.20ha. The same male SFS also made nightly movements of at least 1.65km and 2.00km (straight line) on separate nights. This SFS was considered an outlier and removed from analysis. One other, non-collared male was caught 1.64km away from the original grid it was captured an hour later.

The number of nests used by SFS was shown to not significantly differ between males and females (marginal $R^2 = 0.59$, conditional $R^2 = 0.84$, $P = 0.22$, Fig. 1.6). Thus, I obtained the equation for both sexes combined as $y = 0.35 + 2.03 \cdot \ln(x)$ where y is the number of nests and x is the number of observations. On average SFSs were seen to use 6 – 8 nests over a 27day period (27 was the maximum number of observations obtained for any SFS). On average, SFSs in control grids used 25% more artificial cavities than cluster partition squirrels (proportion test, $Z_1 = -2.71$, $P = 0.01$) and SFSs in cluster partitions used 18% more RCW cavities than control-using SFSs (proportion test, $Z_1 = 2.73$, $P = 0.01$; Table 1.4).

The linear mixed-effects model (marginal $R^2 = 0.13$, conditional $R^2 = 0.56$) showed no significant differences in the RIV value of oaks between any season tested (Tukey HSD; $P \geq 0.65$), between core and non-core ($Z_1 = -0.68$, $P = 0.56$), nor between control and cluster partition ($Z_1 = -1.52$, $P = 0.13$). Estimates of the RIV of oak within SFS home ranges revealed a trend of increasing value of oaks from non-core to core home range and from treatment to control across most seasons (Tables 1.1 and 1.2), although these differences were not statistically significant. For all squirrels, average canopy closure within home range was 95.32% (95% CI = 87.89–100%), varying little between seasons or grid type. Midstory tree density (trees/ha; marginal $R^2 = 0.21$, conditional $R^2 = 0.89$) did not differ by season (Tukey HSD, $P = 0.86$) nor by core vs.

non-core ($Z_1 = -1.42$, $P = 0.16$) but was significant between cluster partition and control-using SFSs ($Z_1 = -2.24$, $P = 0.03$).

Discussion

SFSs appear to display a neutral to weakly negative affinity to RCW cluster partition habitat. The mark-recapture conducted in this study was designed to determine whether SFSs prefer to use nests in RCW cluster partitions. My trap design allowed equal cavity/nest space and cavity characteristics to allow this comparison. Capture probability for the most productive season (winter 2016–2017) showed less variation between cluster partition and control grids and more variation between pairs of grids (Fig. 1.4) which makes this comparison more equitable. In eight out of nine trap sessions, control grids were estimated to contain a greater density of nesting SFSs compared to cluster partition grids with most seasons overlapping in confidence intervals (Fig. 1.5). Control-nesting SFSs also showed a 25% higher proportional use of artificial boxes than cluster partition SFSs (z-test, $P = 0.01$; Table 1.4) which may indicate that cavity space may, to some degree, limit the overall capacity of control grids to house SFSs. I caught a large number of SFSs in winter for both cluster partition and control grids due to the breeding season the previous fall. I was able to observe a distinct difference between cluster partition and control grids during this time. Also during this time, home range sizes were the smallest compared to other seasons, although statistically insignificant. The previous breeding

season in fall resulting in a higher density of SFSs and smaller home ranges is typical of other sciurid species reacting to a pulsed resource event from hard mast tree species.

Given equal nesting opportunity, SFSs generally occur in higher densities in non-cluster habitats. This intuitively makes sense due to the greater abundance of hardwood trees that would offer many food items for SFSs. Comparatively, hardwoods occur far less often in managed RCW clusters (USFWS 2003). Given the opportunity, an optimal forager would decrease the travel distance to these foods to increase the ratio of the energy gained to energy lost due to traveling (MacArthur and Pianka 1966). If SFSs act optimally, they should nest outside of cluster partitions, closer to hardwoods with presumably more abundant food. The nightly home range size estimated may indicate whether SFSs spend more time and space searching for foods and if so, there would be a difference in home range size between cluster partition SFSs and control SFSs. In this study, I did not find a difference in the area used between cluster-partition and control SFSs. The home range sizes created from night-time movements include searches for food, mates, and time spent at their nest so it is uncertain whether cluster partition-using SFSs are behaving differently than control-using SFSs but from a general amount of use-area the SFSs appear to be using the same amount of space.

Home range size did not differ between the two grid types and neither was the composition of those home ranges. The relative importance value for oak was used because it includes the trees with the highest density, frequency, and BA within one

estimate as it relates to other tree species, and it is a dominant tree at Noxubee and a tree that SFSs are known to consume (Thomas and Weigl 1998). I observed a decrease in RIV for oak when comparing control to cluster partition-using SFSs for both their core and non-core home range (Table 1.2). Similarly, the mean RIV of oaks ranged between 18.06–30.13% for core and non-core home ranges between all seasons with either equal or greater RIV of oaks in the core areas. However, neither grid type nor season was shown to be statistically significant. There was also considerable variation around these means (Table 1.1), which suggests that SFSs do not need a large component of oaks within their home range. This is either due to the abundance of other foods or the large amount of acorns dropped by each individual oak tree, which may be sufficient for an individual SFS.

I chose to investigate midstory density due to the RCWs propensity to abandon clusters that contain dense hardwood midstory. In SFS home ranges, I found no difference in midstory density between seasons nor in the core vs. non-core use areas, but I did find a difference between control and cluster partition grids. Since there was insufficient evidence to indicate a difference in the sizes of core and non-core use areas, it is likely that the difference seen between control and cluster partition-using SFSs is an artifact of the habitat types themselves. Cluster partitions are actively managed against hardwood midstory and thus a cluster partition SFS will have a lower midstory density.

Furthermore, density estimates in these areas are largely the same throughout the year, thus it would seem that SFSs are indifferent to midstory density.

The density of nesting SFSs could increase in clusters during the time of year when hard mast is limited (i.e. late spring or early summer) and the SFSs may switch to consuming pine seed (Harlow and Doyle 1990). This effect of food availability was not specifically examined here, but in summer months when squirrels may switch to consuming pine seed there did not appear to be an appreciable difference in cluster and control grid cavity/nest use densities nor was there a significant increase of the RIV of pine during spring and summer (Fig. 1.5, Table 1.1). Interestingly, this absence of strong habitat associations applies to both a cluster-centric aspect of SFS occupancy (Mitchell et al. 2005, Loeb et al. 2012) as well as the current study's SFS-centric use-area aspect. Perhaps the caching behavior may complicate the association of SFSs to oaks and other major diet components on the landscape if SFSs collect food items but store them elsewhere (Weigl 1978). Assuming SFSs rely on acorn mast as a primary food source throughout the year, the absence of correlation in this resource may be complicated by variation in mast production. This variation can be driven by weather (Sork and Bramble, 1993), tree-to-tree variation (Tryon and Carvell 1962), ≥ 3 year cycles in mast production (Olson, 1974; Sork and Bramble, 1993), or pest species (ex. *Curculio* weevil; Winston 1956).

Throughout the year, SFSs showed differences in home range size based on the sex of the individual with females having smaller home ranges than males. Female SFSs are known to raise the young exclusively and may nurse young pups every two hours (Sollberger 1943). The smaller home range seen for female SFS throughout the year is likely due to this intensive care for the young. Nests seen in boxes while trapping indicate that female SFSs are likely building nests or caring for the young for much of the year. Of the female SFSs observed, 14 out of 16 were lactating or post lactating, thus reproduction might account for the smaller home range of female SFSs throughout the year.

Overall SFSs did not appear to use boxes in greater densities in clusters if given equal cavity space. Yet, biologists and land managers routinely find SFSs in RCW cavities (Dennis 1971; Loeb and Hooper 1997; Laves and Loeb 1999). I propose three possibilities for why SFS occur within cluster partitions despite not appearing to select heavily for them: 1) SFSs may choose a cavity/nest with cluster partitions despite the lack of food available there because the distance to food is low enough that it does not significantly impact survivorship or energetic returns on investment, 2) perhaps only a few oaks are necessary for squirrel survivorship thus the relatively few oaks that do occur in and around cluster partitions may be sufficient to warrant resting in a cluster partition cavity or, 3) SFSs may occur in RCW cavities because SFSs prefer them over other natural cavities. In regards to the latter, this study was able to equalize cavity

characteristics in both areas and the SFSs still occupied boxes within the partition. Additionally, I never had a control grid completely saturated with SFSs during any trap occasion thus their choice of a cluster partition box was not due to the absence of available boxes either. Therefore, it is likely squirrels choose boxes within cluster partitions for reasons other than cavity characteristics alone.

Management Implications

The SFS presents a challenging management issue to deal with. Land managers must first assess to what degree SFSs may impact their RCW population. Data provided by the U.S. Fish and Wildlife Service at Noxubee (unpublished data) shows an average number of fledges per breeding group ranging from 2.00–2.58 fledges from 2012–2017 despite removing 50–300 SFSs from active RCW clusters during the breeding season during the same time frame. Thus, removal efforts may not necessarily equate to a significant increase in RCW fledges. This may be the case at Noxubee for several reasons. When an RCW nest is destroyed due to a SFS they may re-nest. The second attempt is no more protected from SFSs than the first nest attempt but it provides a second chance for a successful fledge event. Additionally, there may not be a correlation between removal of SFSs and RCW fledge success if SFSs are not the limiting factor. Perhaps SFS nest depredation is compensatory to other environmental factors. This may not be the case in all locations containing RCWs and SFSs, thus the effort to exclude and

remove SFSs should be decided on a case by case basis. If the exclusion of SFSs is a priority the data provided in this study suggest that SFSs may trend with hard mast tree species but only weakly so. Radio-collared SFSs have sizable home ranges (5.74–9.43ha, depending on sex) and can travel up to two or more kilometers per night. These large movements make habitat management questionable yet it would stand to reason that limiting possible food items would decrease suitability of RCW cluster partitions to SFS occupancy. If removal of SFSs is conducted it should be noted that this species was shown in this study to reproduce in two pulses per year with infrequent nesting throughout and there is potential for an individual to breed twice per year as well as reproduce within the first year of life. Further studies should describe how SFSs respond to removal of hardwoods at to what extent you would have to manage hardwoods relative to cluster centers to obtain a significant decrease in SFS occupancy.

Tables and Figures

Table 2.1 Results of vegetation analysis by season.

| KDE | Midstory Tree Density ¹ | | Relative Importance Value for Oak* | | Relative Importance Value for Hickory* | | Relative Importance Value for Pine* | | Relative Importance Value for Other Trees* | |
|--------------------|------------------------------------|---------------------|------------------------------------|---------------------|--|--------------------|-------------------------------------|----------------------------|--|---------------------------|
| | 50% | 95% | 50% | 95% | 50% | 95% | 50% | 95% | 50% | 95% |
| Spring (n = 14) | 101.97 (± 44.45) | 106.98 (± 46.10) | 18.06 (0– 37.53) | 20.01 (0– 40.25) | 10.74 (0– 26.41) | 5.99 (0– 18) | 55.66 (30.52– 80.8) | 57.69 (32.69– 82.69) | 15.54 (0– 36.4) | 16.31 (1.31– 38.71) |
| Summer (n = 3) | 122.33 (± 37.89) | 134.16 (± 81.76) | 29.75 (0– 81.49) | 30.22 (0– 82.19) | 14.9 (0– 55.19) | 6.86 (0– 35.45) | 40.81 (0– 96.43) | 43.29 (0– 99.36) | 14.54 (0– 69.64) | 19.63 (0– 75.17) |
| Fall (n = 8) | 147.31 (± 67.80) | 119.84 (± 42.22) | 30.13 (0–61.93) | 20.6 (0–48.62) | 11.65 (0–33.88) | 7.43 (0–25.6) | 42.71 (8.43– 76.99) | 50.3 (15.65– 84.95) | 15.51 (5.04– 55.22) | 21.68 (0–49.15) |
| Winter (n = 6) | 128.86 (± 59.08) | 93.24 (± 47.98) | 25.32 (0–60.12) | 25.33 (0– 60.13) | 3.42 (0–17.95) | 3.38 (0– 17.83) | 51.18 (11.18– 91.17) | 51.31 (11.32– 91.3) | 20.09 (0–57.38) | 19.98 (0– 57.33) |

*Percentage, ¹trees/hectare

Results of vegetation analysis on seasonal southern flying squirrel (*Glaucomys volans*) home ranges for both core and peripheral areas (50% and 95% kernel density estimate respectively) reported with 95% confidence intervals in parenthesis. Assessment made for the U.S. Fish and Wildlife Service Sam D. Hamilton Noxubee National Wildlife Refuge in Mississippi.

Table 2.2 Results of vegetation analysis by cluster partition or control squirrel.

| KDE | Midstory Tree Density ¹ | | Relative Importance Value for Oak* | | Relative Importance Value for Hickory* | | Relative Importance Value for Pine* | | Relative Importance Value for Other Trees* | |
|----------------|------------------------------------|---------------------|------------------------------------|----------------------|--|-------------------|-------------------------------------|------------------------|--|----------------------|
| | 50% | 95% | 50% | 95% | 50% | 95% | 50% | 95% | 50% | 95% |
| Control (N=14) | 159.89 (± 46.79) | 147.76 (± 45.76) | 28.13 (5.38–50.88) | 28.03 (5.3–50.76) | 15.29 (0–33.5) | 7.93 (0–21.61) | 36.89 (12.47–61.31) | 41.53 (16.59–66.46) | 19.7 (8–48.26) | 22.51 (6.9–49.17) |
| Cluster (N=16) | 85.28 (± 27.04) | 77.56 (± 17.58) | 19.48 (0.66–38.31) | 16.89 (0–34.69) | 5.3 (0–15.96) | 4.18 (0–13.7) | 61.92 (38.84–85.01) | 63.68 (40.82–86.54) | 13.29 (3.35–35.62) | 15.25 (0–33.98) |

*Percentage, ¹trees/hectare

Results of vegetation analysis between cluster partition and non-cluster (control) home range areas used by southern flying squirrel (*Glaucomys volans*) for both core and peripheral areas (50% and 95% kernel density estimate respectively) reported with 95% confidence intervals in parenthesis. Assessment made for the U.S. Fish and Wildlife Service Sam D. Hamilton Noxubee National Wildlife Refuge in Mississippi.

Table 2.3 SECR model results.

| Model Name | Model Parameters | K | AIC | AIC _c | ΔAIC |
|---------------------------|------------------------------------|----|---------|------------------|---------|
| Occasion | D~session, g0~t, sigma~season | 23 | 6674.52 | 6676.82 | 0.00 |
| Behavior | D~session, g0~b, sigma~season | 20 | 6715.81 | 6717.55 | 41.30 |
| Session | D~session, g0~1, sigma~season | 19 | 6999.77 | 7001.34 | 325.25 |
| Site-specific response | D~session, g0~bk, sigma~season | 20 | 7020.38 | 7022.12 | 345.86 |
| null | D~1, g0~1, sigma~1 | 3 | 7081.34 | 7081.39 | 406.83 |
| Grid | D~session, g0~grid,sigma~season | 20 | 7689.13 | 7690.87 | 1014.61 |
| Sex | D~session, g0~sex, sigma~season | 21 | 7944.57 | 7946.47 | 1270.06 |

Results of SECR model selection for habitat use by southern flying squirrel (*Glaucomys volans*) at the U.S. Fish and Wildlife Service Sam D. Hamilton Noxubee National Wildlife Refuge in Mississippi. Three parameters were modified; D (density), g0 (probability of capture), and sigma (spatial extent). K represents the number of parameters estimated for each model. Symbols are as follows: b: behavioral response, bk: site-specific behavioral response, t: per trapping occasion effect, session: a per trap session effect.

Table 2.4 Nest site selections.

| | Control (n = 17) | Cluster Partition (n = 18) |
|--------------|-----------------------------|---|
| Box | 47.08 (24.02 – 70.14) | 22.24 (3.03 – 41.46) |
| Dray | 2.74 (0 – 10.27) | 2.58 (0 – 9.91) |
| Snag | 11.23 (0 – 25.82) | 17.05 (0 – 34.42) |
| RCW | 0.59 (0 – 4.12) | 19.03 (0.89 – 37.16) |
| Other | 38.36 (0 – 60.83) | 39.10 (16.55 – 61.64) |

Southern flying squirrel (SFS; *Glaucomys volans*) nest selection derived from telemetered SFSs given as a percentage with confidence intervals in parenthesis.

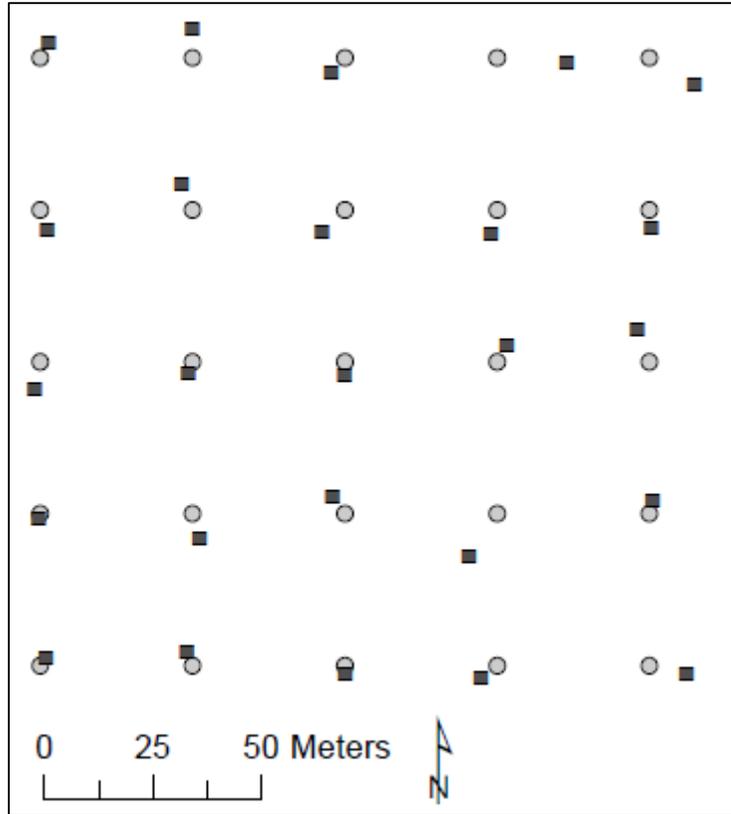


Figure 2.1 Trap grid configuration

Illustration of a grid setup for trapping southern flying squirrels at the U.S. Fish and Wildlife Service Sam D. Hamilton Noxubee National Wildlife Refuge in Mississippi. Circles represent the theoretical grid point locations and the boxes represent the locations of nest boxes placed on the nearest tree to the grid point covering an area approximately 2ha in size.

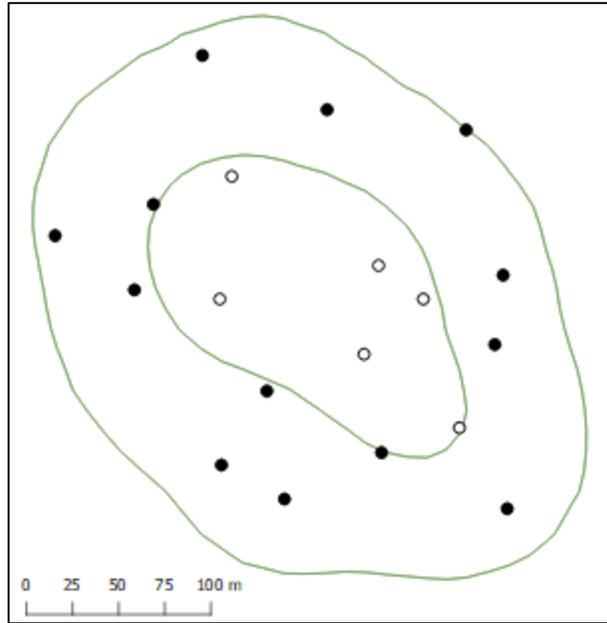


Figure 2.2 Example home range for vegetation analysis

The KDE home range of a female southern flying squirrel (*Glaucomys volans*) that occupied a cluster partition at the U.S. Fish and Wildlife Service Sam D. Hamilton Noxubee National Wildlife Refuge in Mississippi. Inner polygon represents the 50% KDE and the outer polygon represents the 95% KDE. Points represent the randomly generated vegetation sampling points for the 50% KDE (open circle) and 95% KDE (closed circles).

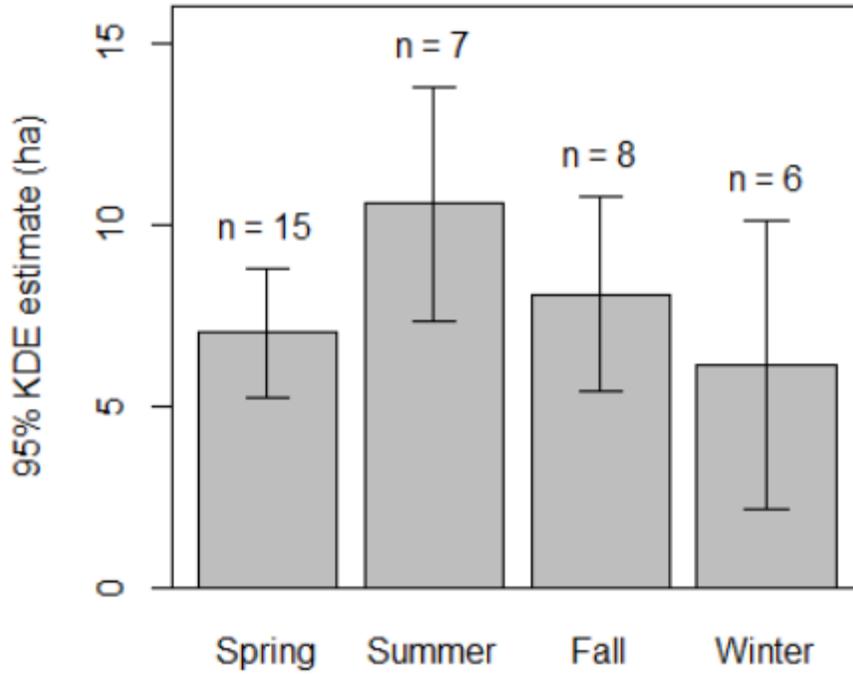


Figure 2.3 Seasonal home range size

Kernel density estimates of 95% KDE home range size (ha, $\pm 95\%$ CI) of southern flying squirrels (*Glaucomys volans*) at the U.S. Fish and Wildlife Service Sam D. Hamilton Noxubee National Wildlife Refuge in Mississippi, reported with sample sizes for each season.

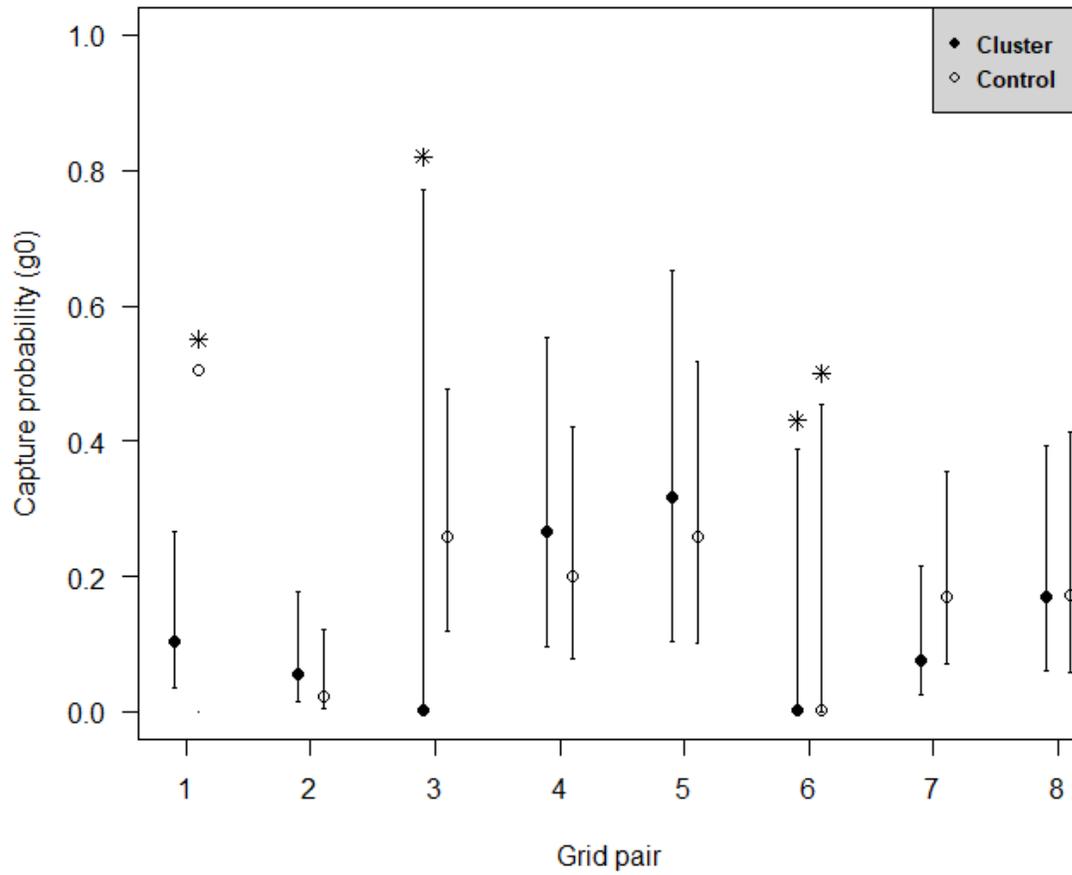


Figure 2.4 Capture probability for winter

Southern flying squirrel capture probabilities for each grid pair derived from the top performing model for winter 2016–2017. Several grids caught too few southern flying squirrels to accurately estimate a capture probability (represented by the ‘*’ symbol).

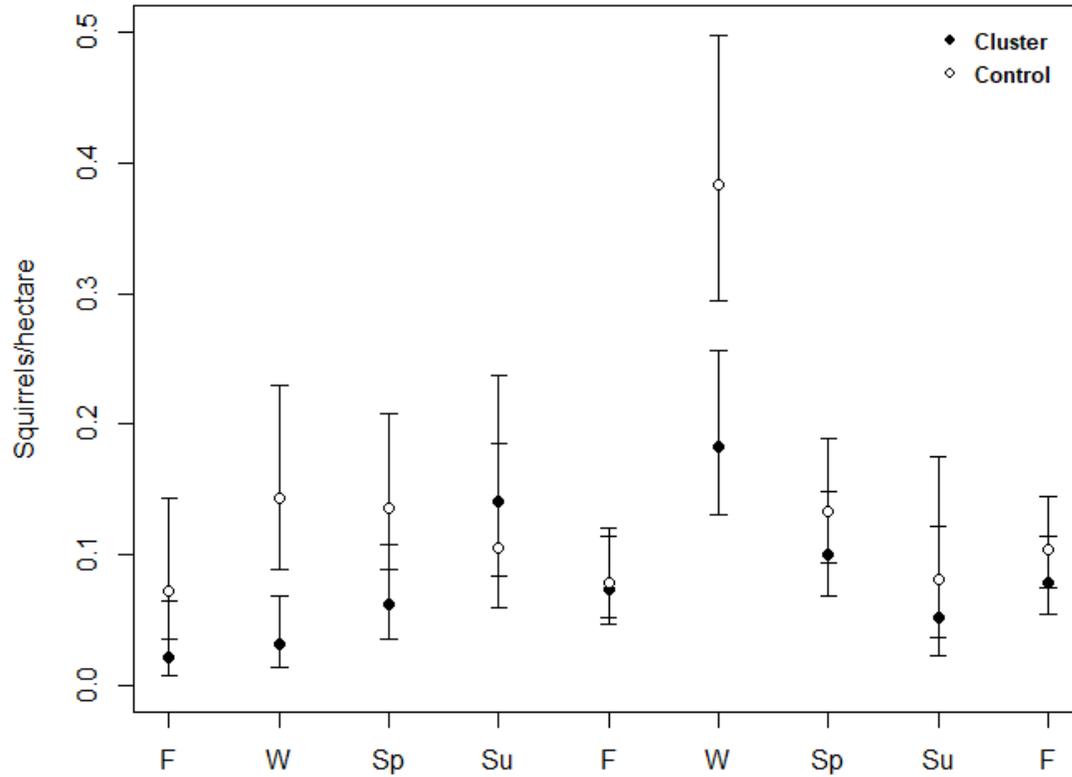


Figure 2.5 Seasonal density estimates

Density estimates from the best performing model for the density of southern flying squirrels at the U.S. Fish and Wildlife Service Sam D. Hamilton Noxubee National Wildlife Refuge in Mississippi. Estimates are given with 95% *CI* per trapping occasion for control and cluster partition grids. X-axis shows season with F = fall (October), W = winter (January), Sp = spring (April), and Su = summer (July) and starting in fall 2015 until summer 2017.

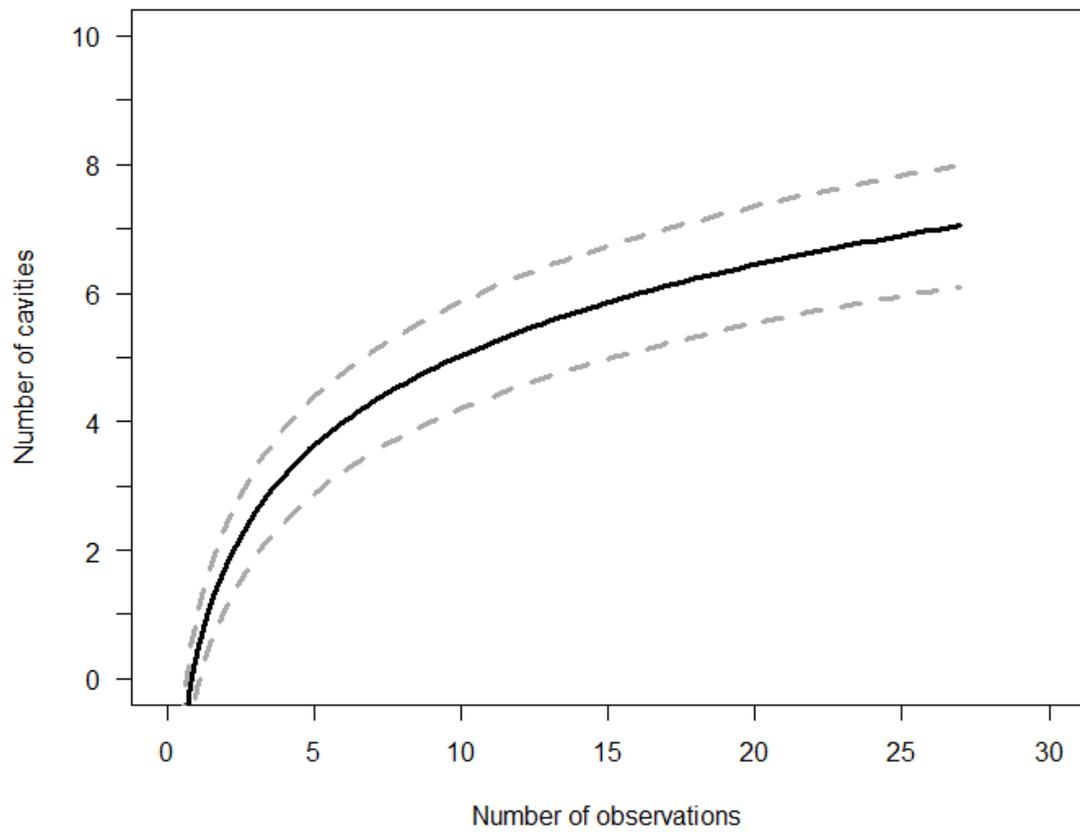


Figure 2.6 Cavities over time

Accumulation curve for the number of cavities, boxes, or drays used by southern flying squirrels (*Glaucomys volans*; solid line) with 95% CI (dashed line) at the U.S. Fish and Wildlife Service Sam D. Hamilton Noxubee National Wildlife Refuge in Mississippi.

References

- Adams, I. T., and G. D. Campbell. 1996. Improved radio-collaring for southern flying squirrels. *Wildlife Society Bulletin* 24:4–7.
- Akaike, H. 1973. Information theory as an extension of the maximum likelihood principle. Pages 267–281 in B.N. Petrov and F. Csaki, editors. *Second international symposium on information theory*, Akademiai Kiado, Budapest.
- Bendel, P.R., and J. E. Gates. 1987. Home range and microhabitat partitioning of southern flying squirrel (*Glaucomys volans*). *Journal of Mammalogy* 68:243–255.
- Borgo, J. S., M.R. Conover, and L. M. Conner. 2006. Nest boxes reduce flying squirrel use of red-cockaded woodpecker cavities. *Wildlife Society Bulletin* 34:171–176.
- M. R. Conover, and L. M. Conner. 2010. Flying squirrel removal does not reduce their use of simulated red-cockaded woodpecker nest clusters. *Southeastern Naturalist* 9:813–820.
- Conner, R. N., B. A. Locke. 1982. Fungi and red-cockaded woodpecker cavity trees. *The Wilson Bulletin* 94:64–70.
- K. A. O’Halloran. 1987. Cavity-tree selection by red-cockaded woodpeckers as related to growth dynamics of southern pine. *The Wilson Bulletin* 99:398–412.
- D. C. Rudolph. 1995. Excavation dynamics and use patterns of red-cockaded woodpecker cavities: relationships with cooperative breeding. Pp. 343–352 in *Red-cockaded woodpecker: recovery, ecology, and management* (D. L. Kulhavy, R. G. Hooper, and R. Costa, eds.). Center for Applied Studies, College of Forestry, Stephen F. Austin State University. Nacogdoches, Texas, USA.
- D. C. Rudolph, D. Saenz, and R. R. Schaefer. 1996. Red-cockaded woodpecker nesting success, forest structure, and southern flying squirrels in Texas. *Wilson Ornithological Society* 108:697–711.
- D.C. Rudolph, and J.R. Walters. 2001. *The red-cockaded woodpecker: surviving in a fire maintained ecosystem*. University of Texas Press, Austin, TX.
- Dennis, J. V. 1971. Species using red-cockaded woodpecker holes in northeastern South Carolina. *Bird-banding* 42:79–87.

- Efford, M. G. (2016). secr: Spatially explicit capture-recapture models. R package version 2.10.4
- Fridell, R. A., and J. A. Litvaitis. 1991. Influence of resource distribution and abundance on home range characteristics of southern flying squirrels. *Canadian Journal of Zoology* 69: 2589–2593.
- Garroway, C. J., J. Bowman, P. J. Wilson. 2013. Complex social structure of southern flying squirrels is related to spatial proximity but not kinship. *Behavioral Ecology and Sociobiology* 67:113–122.
- Gelman, A. and J. Hill. 2007. *Data analysis using regression and multilevel/hierarchical models*. Cambridge University Press, New York, NY.
- Harlow R. F., and A.T. Doyle. 1990. Food habits of southern flying squirrels (*Glaucomys volans*) collected from red-cockaded woodpecker (*Picoides borealis*) colonies in South Carolina. *American Midland Naturalist* 124:187–191.
- Jackson, J. A., M.R. Lennartz and R. G. Hooper. 1979. Tree age and cavity initiation by red-cockaded woodpeckers. *Journal of Forestry* 77:102–103.
- Laves, K.S. and S.C. Loeb. 1999. Effects of southern flying squirrels *Glaucomys volans* on red-cockaded woodpecker (*Picoides borealis*) reproductive success. *Animal Conservation* 2:295–303.
- S.C. Loeb. 2006. Differential estimates of southern flying squirrels (*Glaucomys volans*) population structure based on capture method. *American Midland Naturalist* 155:237–243.
- Lockhart, B. R., J. M. Guldin, and T. Foti. 2010. Tree species composition and structure in an old bottomland hardwood forest in south-central Arkansas. *Castanea* 75:315–329.
- Loeb, S. C. 1993. Use and selection of red-cockaded woodpecker cavities by southern flying squirrels. *Journal of Wildlife Management* 57:329–335.
- S. L. Reid, and D. J. Lipscomb. 2012. Habitat and landscape correlates of southern flying squirrel use of red-cockaded woodpecker clusters. *Journal of Wildlife Management* 76:1509–1518.
- MacArthur, R. H., and E. R. Pianka. 1966. On optimal use of a patchy environment. *The American Naturalist* 100:603–609.
- Mech, L. D. 1983. *A handbook of animal radiotracking*. University of Minnesota Press, Minneapolis, MN, USA.

- Mitchell, L. R., C. R. Chandler, and L. D. Carlile. 2005. Habitat as a predictor of southern flying squirrel abundance in red-cockaded woodpecker cavity clusters. *Journal of Wildlife Management* 69:418–423.
- Montague, W.G., J.C. Neal and J.E. Johnson. 1995. Techniques for excluding southern flying squirrels from cavities of Red-cockaded Woodpeckers. Pages 401–409 in D. L. Kulhavy, R. G. Hooper, and R. Costa, editors. *Red-cockaded Woodpecker: recovery, ecology, and management*. Center for Applied Studies, College of Forestry, Stephen F. Austin State University. Nacogdoches, Texas, USA.
- Nakagawa, S. and H. Schielzeth. 2013. A general and simple method for obtaining R^2 from generalized linear mixed-effects models. *Methods in Ecology and Evolution* 4:133–142.
- Nebeker, T. E., M. Pelligrine, R. A. Tisdale, and J. D. Hodges. 1995. Site/stand factors associated with red-cockaded woodpecker colonies on the Noxubee National Wildlife Refuge, Mississippi. Pages 431–435 in D. L. Kulhavy, R. G. Hooper, and R. Costa, editors. *Red-cockaded woodpecker: Recovery, ecology, and management*. Center for Applied Studies, College of Forestry, Stephen F. Austin State University. Nacogdoches, Texas, USA.
- Nudds, T. D. 1977. Quantifying the vegetative structure of wildlife cover. *Wildlife Society Bulletin* 5:113–117.
- Olson, D. F. Jr. 1974. *Quercus L. Oak*. USDA Forest Service Agriculture Handbook 450. USDA Forest Service, Washington, DC, 88 pp.
- Risch, T. S., and M. J. Brady. 1996. Trap height and capture success of arboreal small mammals: evidence from southern flying squirrels (*Glaucomys volans*). *American Midland Naturalist* 136:346–351.
- Smith, R. L. and T. M. Smith. 2001. *Ecology and field biology*. 6th edition. Addison Wesley Longman, San Francisco, CA, USA.
- Sollberger, D. E. 1943. Notes on the breeding habits of the eastern flying squirrel (*Glaucomys volans volans*). *Journal of Mammalogy*, 24:163–173.
- Sork, V. L., J. E. Bramble. 1993. Prediction of acorn crops in three species of North American oaks: *Quercus alba*, *Q. rubra* and *Q. velutina*. *Annales des sciences forestières* 50(Suppl1)128s–136s.
- Stone, K. D., G. A. Heidt, P. T. Caster, and M. L. Kennedy. 1997. Using geographic information systems to determine home range of the southern flying squirrel (*Glaucomys volans*). *American Midland Naturalist* 137:106–111.

- Thomas, R. B. and P. D. Weigl. 1998. Dynamic foraging behavior in the southern flying squirrel (*Glaucomys volans*): test of a model. *American Midland Naturalist* 140:264–270.
- Tryon, E. H. and K. L. Carvell. 1958. Regeneration under oak stands. West Virginia University Agricultural Experiment Station Bulletin. West Virginia University Agricultural Experiment Station, Morgantown, WV, USA.
- U.S. Fish and Wildlife Service. 2003. Recovery plan for the red-cockaded woodpecker (*Picoides borealis*): second revision. U.S. Fish and Wildlife Service, Atlanta, GA. 296 pp.
- U.S. Fish and Wildlife Service. 2014. Comprehensive conservation plan and environmental assessment for Sam D. Hamilton Noxubee National Wildlife Refuge. U.S. Fish and Wildlife Service, Atlanta, GA. Volume 1.
- Walters, J. R. 1990. The red-cockaded woodpecker: a “primitive” cooperative breeder. Pgs. 67-101 in *Cooperative breeding in birds: long term studies of ecology and behavior* (P.B. Stacey and W. D. Koenig, Editors). Cambridge University Press, Cambridge, United Kingdom.
- Ware, S., C. Frost, and P. D. Doerr. 1993. Southern mixed hardwood forest: the former longleaf pine forest. (W.H. Martin, S.G. Boyce, and A. C. Echternacht, Editors). *Biodiversity of the southeastern United States: lowland terrestrial communities*. John Wiley and Sons. Inc., New York, NY.
- Weigl, P. D. 1978. Resource overlap, interspecific interactions and the distribution of the flying squirrels *Glaucomys volans* and *G. sabrinus*. *American Midland Naturalist* 100:83-96.
- Winston, P. W. 1956. The acorn microsere, with special reference to arthropods. *Ecology* 37:120–132.
- Zar, J.H. 1999. *Biostatistical Analysis*, Fourth Edition. Prentice Hall, Upper Saddle River, NJ.

CHAPTER III
SOUTHERN FLYING SQUIRREL (*GLAUCOMYS VOLANS*) DIET COMPOSITION
IN RED-COCKADED WOODPECKER (*PICOIDES BOREALIS*)
CLUSTERS OF THE SOUTHEAST U.S.

Abstract

Southern flying squirrels (SFS; *Glaucomys volans*) have a negative impact on red-cockaded woodpeckers (RCW; *Picoides borealis*), yet little is known about SFS diet relative to their presence in RCW clusters. Information on SFS diet is important to form targeted management to limit negative interactions between squirrels and RCWs. I examined the diets of SFSs in the U.S. Fish and Wildlife Service, Sam D. Hamilton Noxubee National Wildlife Refuge in east-central Mississippi (n = 10 SFSs), U.S. Forest Service, Oakmulgee Ranger District of the Talladega National Forest in west-central Alabama (n = 10 SFSs), Alabama Department of Conservation and Natural Resources, Coosa Wildlife Management Area in central Alabama (n = 9 SFSs), and the Sehoy Plantation in east-central Alabama (n = 10 SFSs) using analysis of the stable isotopes of carbon and nitrogen. Analysis for isotopic ratios of $^{14/15}\text{N}$ (δN^{15}) and $^{12/13}\text{C}$ ($\delta^{13}\text{C}$) in livers of SFS, and their potential food items, yielded evidence that oaks (*Quercus* sp.) and hickories (*Carya* sp.) comprised 18–36%, and 22–63% of SFS diet, respectively, across all locations. SFSs also showed little variation in $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ relative to food items consumed suggesting that SFSs have a small dietary breadth. The narrow dietary breadth

shown across study sites indicates that targeted management against specific food items may be a viable option to deter SFSs from RCW cluster habitat.

Introduction

The southern flying squirrel (SFS; *Glaucomys volans*) negatively affects the endangered red-cockaded woodpecker (RCW; *Picooides borealis*) through nest predation and cavity usurpation (Conner et al. 1996; Dennis 1971; Laves and Loeb 1999; Loeb 1993; U.S. Fish and Wildlife Service 2003). RCWs use groups of living pine trees referred to as a ‘cluster’ (Walters 1990). Cluster trees are ≥ 60 years old (Conner and Locke 1982; Conner and O’Halloran 1987; Nebeker et al 1995) and may take months to years to excavate (Jackson et al 1979; Conner and Rudolph 1995). Currently, there is no published record of an RCW evicting a SFS from a cavity (Dennis 1971; Montague et al. 1995). Thus, SFSs present an issue for recovery efforts of RCWs with greater negative impacts on smaller populations of RCWs (Kappes 2008; Laves and Loeb 1999). These harmful interactions have prompted studies examining habitat correlates of SFSs to RCW cluster trees (Loeb 1993, Conner et al. 1996, Loeb et al. 2012) but with varying degrees of success at finding significant habitat characteristics. Perhaps a more direct assessment of important components of RCW habitats that contribute to SFS occupancy may come from examining the diets of SFSs within these areas.

Relatively few studies have evaluated SFSs’ feeding habits relative to RCW cluster habitat. Such investigations can increase understanding of the SFS’s life history, affording novel management options limiting negative interactions between this species and RCWs. For instance, SFSs are known to be generalist consumers (Dolan and Carter 1977), yet it is likely that SFSs consume different resources throughout the year. By

depleting a resource that SFSs use when RCWs are nesting, a land manager may decrease the suitability of that area for SFSs and thus promote RCW nest success.

Previous studies of SFSs' food preference conducted in captivity revealed differing preference for oaks and hickories, depending on ambient temperature during trials (Thomas and Weigl 1998; Helmick et al. 2014). These studies provide evidence that oaks are generally preferred relative to hickory nuts until temperatures are sufficiently low. However, as these studies were conducted with squirrels in captivity, results may not be consistent in natural systems. To this end, Harlow and Doyle (1990) examined SFS diets from RCW clusters via stomach contents but were unable to find animal matter other than insect exoskeletons due to what they suspected to be rapid digestion of soft animal tissue. Additionally, discerning plant tissue may be difficult once this matter has been partially digested. To clarify and supplement this previous work discerning SFS diet *in situ*, I used stable isotopes to identify possible food items SFSs could assimilate from their natural environment.

Stable isotopes occur naturally in living and non-living things (DeNiro and Epstein 1978, Hoefs 2009). Organisms integrate isotopes into their tissues after consuming them, creating discrepancies between lighter and heavier isotopes within their tissues (DeNiro and Epstein 1978, 1981). These discrepancies can then be used to identify the contribution of various food items to the diet using linear and non-linear models built with varying levels of complexity (Phillips et al. 2014).

In this study, I compared the ratios of the carbon isotope $^{12/13}\text{C}$ ($\delta^{13}\text{C}$) and nitrogen isotope $^{14/15}\text{N}$ ($\delta^{15}\text{N}$) in the liver of SFSs, to the isotopes of potential food items in the environment. I analyzed stable isotope composition of SFSs during the RCW

nesting season to reconstruct their diets and identify important food types. By identifying those food items that most prominently contribute to SFS diets it may be possible to manage these resources on the landscape to limit the suitability of RCW habitat for SFSs.

Methods

Study Site

Sampling for this study was conducted at four sites within Mississippi and Alabama: the Sam D. Hamilton Noxubee National Wildlife Refuge in northeastern Mississippi (hereafter; Noxubee), Oakmulgee Ranger District of the Talladega National Forest in west-central Alabama (hereafter; Oakmulgee), Coosa Wildlife Management Area in central Alabama (hereafter; Coosa), and the Sehoj Plantation in east-central Alabama (hereafter; Sehoj)(Fig. 2.1). Ecosystems of each site are comprised largely of a mix of pine and hardwoods and managed for good quality foraging habitat as determined by their ecoregion in the RCW recovery plan (U.S. Fish and Wildlife Service, 2003) and include prescribed fire (approximately every three years) and forest thinning.

Each site varies in dominant tree compositions. Noxubee is comprised of a mix of primarily loblolly pine (*Pinus taeda*) and some shortleaf pine (*P. echinata*) with dominant hardwoods including sweetgum (*Liquidambar styraciflua*), white oak (*Quercus alba*), southern red oak (*Q. falcata*), water oak (*Q. nigra*), willow oak (*Q. phellos*), post oak (*Quercus stellata*), cherrybark oak (*Q. pagoda*), mockernut hickory (*Carya tomentosa*), shagbark hickory (*C. ovata*), and black gum (*Nyssa sylvatica*). Oakmulgee and Coosa are both dominated by longleaf pine (*P. palustris*) with hardwoods comprising of southern red oak, white oak, post oak, chestnut oak (*Q. montana*), blackjack oak (*Q. marilandica*), swamp white oak (*Quercus bicolor*). sweetgum and mockernut hickory.

Sehoy is dominated by shortleaf pine with scattered loblolly and longleaf pines with hardwoods comprising of the same hardwoods as Noxubee but also include greater amounts of swamp white oak.

Squirrel Collection

Each location donated euthanized SFSs collected through their removal operations. These SFSs were evicted from active and inactive red-cockaded woodpecker cavities in active clusters to ensure cavity space for RCWs. Collection from Noxubee occurred in the fall from October to late November of 2015, and again in the spring from late March to late May 2016. This seasonality of sampling allowed me to compare SFS isotope values as an approximation of diet change between sites and among seasons for Noxubee. SFSs from all other sites were collected in and around March of 2017. Once SFS were obtained, I excised their livers using a scalpel. I then froze livers individually at -20°C, and later freeze-dried these samples for storage, typically for 6–12 months.

Food collection

I collected possible food items from each study site based on food items that SFSs are known to consume (Dolan and Carter 1977; Harlow and Doyle 1990). Collected food items were acorns (*Quercus sp.*), hickory nuts (*Carya tomentosa* and *C. ovata*), and insects including Hemiptera (*Boisea sp.*), Blatodea (*Parcoblatta pensylvanica*), Orthoptera (*Neoconocephalus sp.*), and Coleoptera (*Phyllophaga sp.*). The exact species of insects that SFSs consume is largely unexplored, thus I sampled insects from a wide array of orders to obtain as much diversity of the food as possible. I collected food items close to the area where SFSs were captured to best approximate potential food items

available to the individual SFSs sampled. I also collected the same species of food items in each study site, when possible, to facilitate equitable comparisons among study sites.

I binned food items into the three categories: Oak (*Quercus sp.*), Hickory (*Carya sp.*), and Insect. Food items were also freeze-dried for storage for the same duration of time as the SFS livers.

Vegetation Analysis

Hardwood habitat patterns surrounding clusters where squirrels were obtained may explain patterns in the consumption of hardwood diet items. For this reason, I created a 1km circular buffer around the center of each cluster a SFS was obtained from in each site. I chose a 1km buffer based on what is a reasonable amount of space that SFSs could use based on previous literature's estimates on home range size and movements (McCabe 1947, Bendel and Gates 1987; Fridell and Litvaitis 1991; Stone et al. 1997; Garroway et al. 2013). I used National Land Cover Data (NLCD; Homer et al. 2015) within these buffers to obtain percentage of hardwood (relative to all other land cover types) by adding the hardwood and woody wetland cover type (corresponding land cover index 41 and 90 respectively) with one half of the mixed hardwood/pine land cover type (land cover index 43).

Isotope Analysis

Each food item was analyzed to determine isotopic ratios of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$, allowing analysis of specific contributions of each food category to SFS diets. I used 80% standard ellipses and ellipse area to describe and represent the data using *SIBER* package (Jackson and Parnell 2011) for program R (R Core Development Team, 2016). Standard

ellipse area comparisons afford insight into diet breadth, i.e. food items that differ from each other or similar food items from other more distant places.

Stable isotope analyses were completed at the Analytical Chemistry Laboratory within the Institute of Ecology at the University of Georgia (Georgia, USA), or the Chemical Tracer Laboratory at the University of Windsor (Ontario, CA). Stable isotope analyses were conducted using a Delta Plus isotope-ratio mass spectrometer (ThermoFinnigan, San Jose, CA, U.S.A.) coupled with an elemental analyzer (Costech, Valencia, CA, U.S.A.). Values of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ were quantified in relation to four internal laboratory standards, an NIST standard (#1577 bovine liver) and spinach (*Spinacia oleracea*) which was run every 12 samples. Vienna Pee Dee Belemnite carbonate and atmospheric nitrogen were the standard reference materials for ^{13}C and ^{15}N , respectively. The standard deviation of the NIST standard 1577 ($n = 9$) was 0.05‰ for $\delta^{13}\text{C}$ and 0.12‰ for $\delta^{15}\text{N}$ and for spinach ($n = 9$) was 0.04‰ for $\delta^{13}\text{C}$ and 0.18‰ for $\delta^{15}\text{N}$.

I used Bayesian mixing models generated in program MixSIAR (Moore and Semmens 2008; Stock and Semmens 2016) using the GUI in program R to determine the most parsimonious combination of food categories fitting the $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ found in each SFS's liver. I used uninformed priors and 1×10^6 iterations to obtain model estimates. To acquire a generic estimate of percent contribution of each food category to SFS, across the suite of study sites, I treated sampling site as a random effect in MixSIAR.

Since SFSs are known to be opportunistic foragers, I used two sets of isotope discrimination factors, either omnivorous ($\delta^{13}\text{C} = 2.15\text{‰}$ (SD = 0.08), $\delta^{15}\text{N} = 2.95\text{‰}$ (SD

= 0.29)) or vegetarian ($\delta^{13}\text{C} = 1.20\text{‰}$ (SD = 0.55), $\delta^{15}\text{N} = 3.10\text{‰}$ (SD = 0.18)), based on estimates obtained for a closely allied species; *Rattus norvegicus* (Kurle et al. 2014). Because discrimination factors are not available for SFS, I employed those in Kurle et al. (2014), comparing fit among mixing models. This was done to determine which discrimination factors best fit the data using Deviance Information Criteria (DIC) to identify the best fit model, that with the lowest DIC.

Results

The omnivorous discrimination factor explained more of the variation than the competing vegetarian discrimination factor model (DIC: 182.53 and 184.33 respectively). Hickory was estimated to contribute the most to the diet of SFSs at $39.70 \pm 18.60\%$ ($\bar{x} \pm \text{SD}$) compared to Insect $33.20 \pm 17.00\%$ or Oak $27.00 \pm 19.20\%$ (Table 2.1 and 2.2).

Standard 80% ellipses showed little variation in SFS isotope ranges for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ (Fig. 2.2) despite the wide range in possible food items and amount of hardwood habitat (Fig. 2.3). The ellipses created around SFSs collected from fall and spring at Noxubee resulted in ellipse areas of 5.64 and 12.25 respectively, an approximately 280% increase between these seasons (Fig. 2.4).

The percentage of hardwood land cover surrounding RCW cluster centers where SFSs were captured varied between study sites with highest to lowest hardwood land cover corresponding to Noxubee ($51.59 \pm 17\%$ [$\bar{x} \pm \text{SD}$]), Oakmulgee ($45.69 \pm 11.00\%$), Coosa ($34.74 \pm 6.00\%$), and Sehoj ($9.60 \pm 6.00\%$) respectively (Fig. 2.3).

Discussion

The best performing MixSIAR model was one that used the omnivorous discrimination factor thus conforming to previous records of SFSs consuming both vegetation and animal matter (Dolan and Carter 1977; Harlow and Doyle 1990; Helmick et al. 2014). However, for each site the mean contribution of oak and hickory mast to the diet of SFSs was relatively constant, as opposed to insect contributions that varied among sites (Table 2.1). These differences may reflect the importance of hard mast as a food across all sites unlike insects that may be opportunistically consumed. However, the positioning of SFS isotope values relative to food items reflects a stronger relationship to oaks. Hickories were consistently lower in $\delta^{13}\text{C}$ than expected if the trophic enrichment of 2.15‰ for ^{13}C obtained from Kurle et al. (2014) is correct (except Noxubee; Fig. 2.2). MixSIAR results showed a great deal of variation in model estimates. Therefore, it is not clear if and to what degree each food item is consumed.

Oaks are known to be important food items for squirrels (Dolan and Carter 1977; Johnson et al. 2009) and the MixSIAR model provided results suggesting that mean estimates of oak contribution to SFS diet roughly followed the amount of hardwoods near the clusters where SFSs were captured (Fig. 2.3). Sehoj differed from the other sites in the lower amount of hardwood land cover relative to the mean contribution of oaks to SFS diets. This may indicate the importance of oaks to SFS diets due to this discrepancy in availability. Sehoj was also unique in its increased difference in $\delta^{15}\text{N}$ relative to oaks but due to its alignment with oaks in $\delta^{13}\text{C}$ these SFSs may be consuming more nitrogen rich parts of the oak such as the buds or the SFSs could be consuming insects that feed on oaks and thus increase $\delta^{15}\text{N}$.

MacArthur and Pianka (1966) theorized that as the distance between suitable foraging locations increased, and search effort for preferred food items increased, an optimal forager would increase the number of food items consumed (diet breadth). Due to the similarity and narrow range in each sites' isotope value for SFSs, it is likely that in all sites SFSs were able to obtain their preferred food items despite hardwoods ranging from 4–77% across all sites, and within Sehoj with locations ranging from 3–17% hardwood. It is worthwhile to note however, that hardwood land cover does not necessarily relate to the amount of suitable forage comprising the hardwoods but instead this should be viewed as a surrogate for suitable foraging habitat. For example, there is evidence that weather plays a major role in the production of acorns (Sork and Bramble 1993). Thus, in the fall of 2016, there could have been a modest or high crop yield from favorable weather conditions across the region that could explain the SFSs' narrow diet breadth. Oaks also vary in the amount of acorns produced annually, usually with a large acorn crop every 3 or more years (Olson 1974; Sork and Bramble 1993, Straub et al. 2016). White oaks generally have shorter cycles and germinate in the fall whereas red oaks have longer cycles and germinate the following spring. For this reason, squirrels have been found to consume white oaks first and cache red oaks (Steele et al. 2005). This may be the reason I observed Noxubee SFSs increasing the range of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ from fall 2015 to the spring of 2016 (Fig. 2.4). Mixing model results indicated that SFS sampled at Noxubee were relatively specific in fall (perhaps towards white oaks), and then incorporated a larger variety of food items in their diet in spring as they depleted their cache of acorns.

Complete hardwood removal to limit harmful interactions of SFSs on RCWs may be less than ideal in most cases, but targeted species removal may still be feasible. In all sites SFSs align with oaks' $\delta^{13}\text{C}$ (Fig. 2.2, Table 2.2) suggesting they are most likely feeding on acorns, oak buds, or insects that might also be feeding on oak. Harlow and Doyle (1990) found similar choice of a natural population of SFSs consuming acorns. Thus, perhaps lowering the amount of oaks could help deter SFSs from RCW clusters as it would be more energetically costly to use cavities there as mentioned previously. Future studies should incorporate DNA analysis to determine the specific foods (including oaks) consumed. Once identified, subsequent experiments can manipulate their availability in the environment to determine if it is a viable management option to selectively remove oaks around RCW cluster partitions.

Tables and Figures

Table 3.1 Landcover and diet proportions by site

| Site | Proportion of Hardwood | | | Oak | | | Hickory | | | Insect | | | | |
|-----------|---------------------------|------|-----------|------|-----------|-----------|---------|-----------|-----------|--------|-----------|-----------|----|--------|
| | \bar{X} | SD | \bar{X} | SD | 95% CI | \bar{X} | SD | 95% CI | \bar{X} | SD | 95% CI | \bar{X} | SD | 95% CI |
| Noxubee | 0.52 | 0.17 | 0.37 | 0.36 | 0-1.0 | 0.23 | 0.19 | 0-0.56 | 0.40 | 0.29 | 0-0.93 | | | |
| Oakmulgee | 0.46 | 0.12 | 0.28 | 0.25 | 0-0.78 | 0.63 | 0.26 | 0.16-1.0 | 0.09 | 0.08 | 0-0.24 | | | |
| Coosa | 0.35 | 0.06 | 0.18 | 0.15 | 0-0.46 | 0.39 | 0.14 | 0.15-0.61 | 0.43 | 0.10 | 0.28-0.61 | | | |
| Sehoy | 0.10 | 0.06 | 0.23 | 0.24 | 0-0.75 | 0.36 | 0.19 | 0-0.62 | 0.41 | 0.14 | 0.17-0.64 | | | |
| Global | - | - | 0.27 | 0.19 | 0.03-0.65 | 0.40 | 0.19 | 0.10-0.72 | 0.33 | 0.17 | 0.07-0.64 | | | |

Proportion of hardwood land cover and MixSIAR results for mean contribution of food items to the diet of SFs from each location (reported with standard deviation).

Table 3.2 Oak isotope values by site

| | Noxubee | | Oakmulgee | | Coosa | | Sehoy | | $\bar{\chi}$ Across Sites | |
|--------------------|-----------------------|-----------------------|-----------------------|-----------------------|-----------------------|-----------------------|-----------------------|-----------------------|---------------------------|-----------------------|
| | $\delta^{13}\text{C}$ | $\delta^{15}\text{N}$ | $\delta^{13}\text{C}$ | $\delta^{15}\text{N}$ | $\delta^{13}\text{C}$ | $\delta^{15}\text{N}$ | $\delta^{13}\text{C}$ | $\delta^{15}\text{N}$ | $\delta^{13}\text{C}$ | $\delta^{15}\text{N}$ |
| <i>Q. alba</i> | -26.95 (1) | -0.74 (1) | -26.52 (2) | -3.86 (2) | -23.79 (2) | -4.54 (2) | - | - | -25.51 | -3.50 |
| <i>Q. bicolor</i> | - | - | - | - | - | - | -24.33 (1) | -0.01 (1) | -24.33 | -0.01 |
| <i>Q. falcata</i> | - | - | -26.73 (2) | -2.745 (2) | -22.54 (1) | -5.06 (1) | - | - | -25.33 | -3.52 |
| <i>Q. lyrata</i> | -25.21 (1) | -0.98 (1) | - | - | - | - | - | - | -25.21 | -0.98 |
| <i>Q. montana</i> | - | - | -26.06 (1) | -2.05 (1) | -27.99 (2) | -3.025 (2) | - | - | -27.35 | -2.7 |
| <i>Q. nigra</i> | - | - | - | - | - | - | -26.67 (1) | -0.03 (1) | -26.67 | -0.03 |
| <i>Q. pagoda</i> | -26.07 (1) | -2.85 (1) | - | - | - | - | - | - | -26.07 | -2.85 |
| <i>Q. phellos</i> | -26.91 (1) | -2.47 (1) | - | - | - | - | -25.54 (2) | -1.38 (2) | -26.00 | -1.74 |
| <i>Q. stellata</i> | -29.19 (1) | -0.29 (1) | - | - | - | - | -31.48 (1) | -4.65 (1) | -30.34 | -2.47 |

Oak (*Quercus*) species used across sites, represented with their average $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ with sample size in parenthesis.

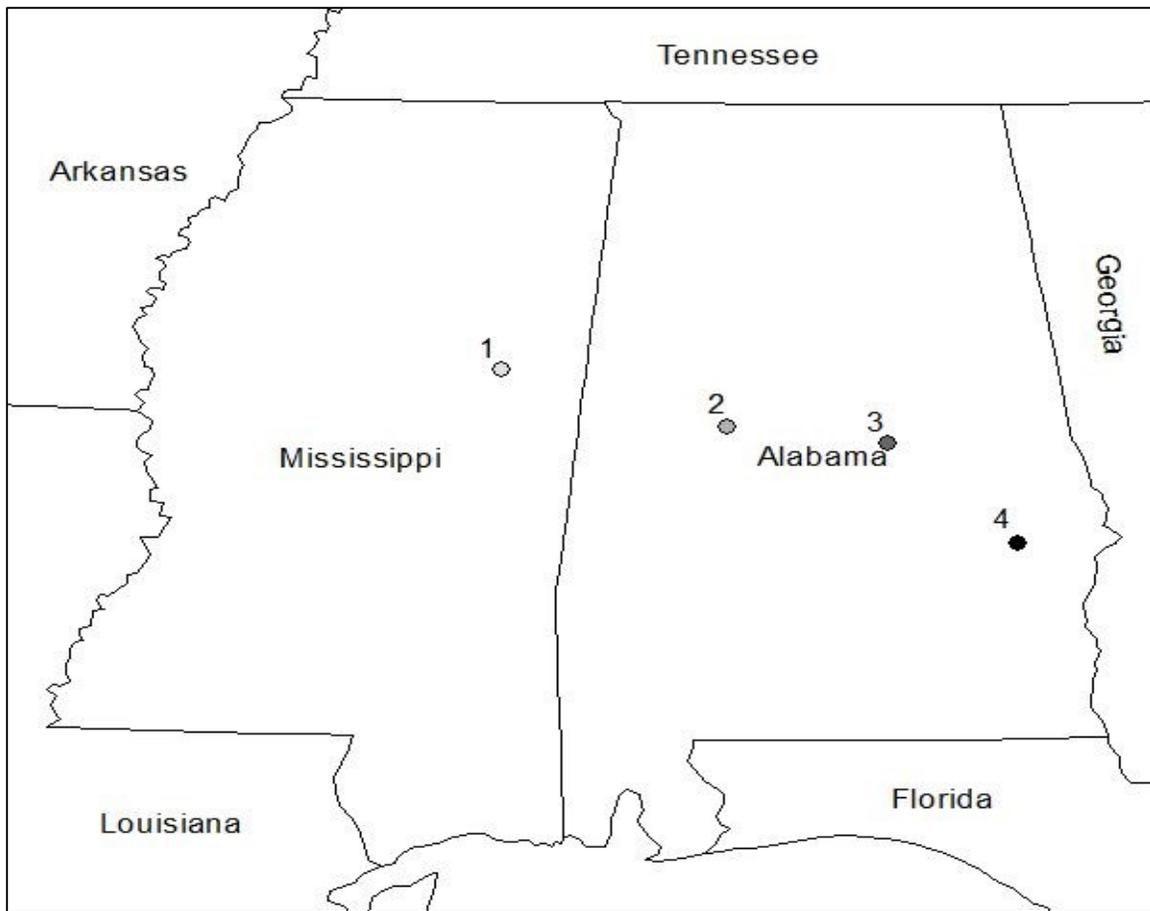


Figure 3.1 Site locations

Geographic location of the four study sites across Mississippi and Alabama where southern flying squirrels (*Glaucomys volans*) and food items were obtained; 1) U.S. Fish And Wildlife Service Sam D. Hamilton Noxubee National Wildlife Refuge, 2) U.S. Forest Service Talladega National Forest Oakmulgee District, 3) Coosa Wildlife Management Area, and 4) Sehoj Plantation.

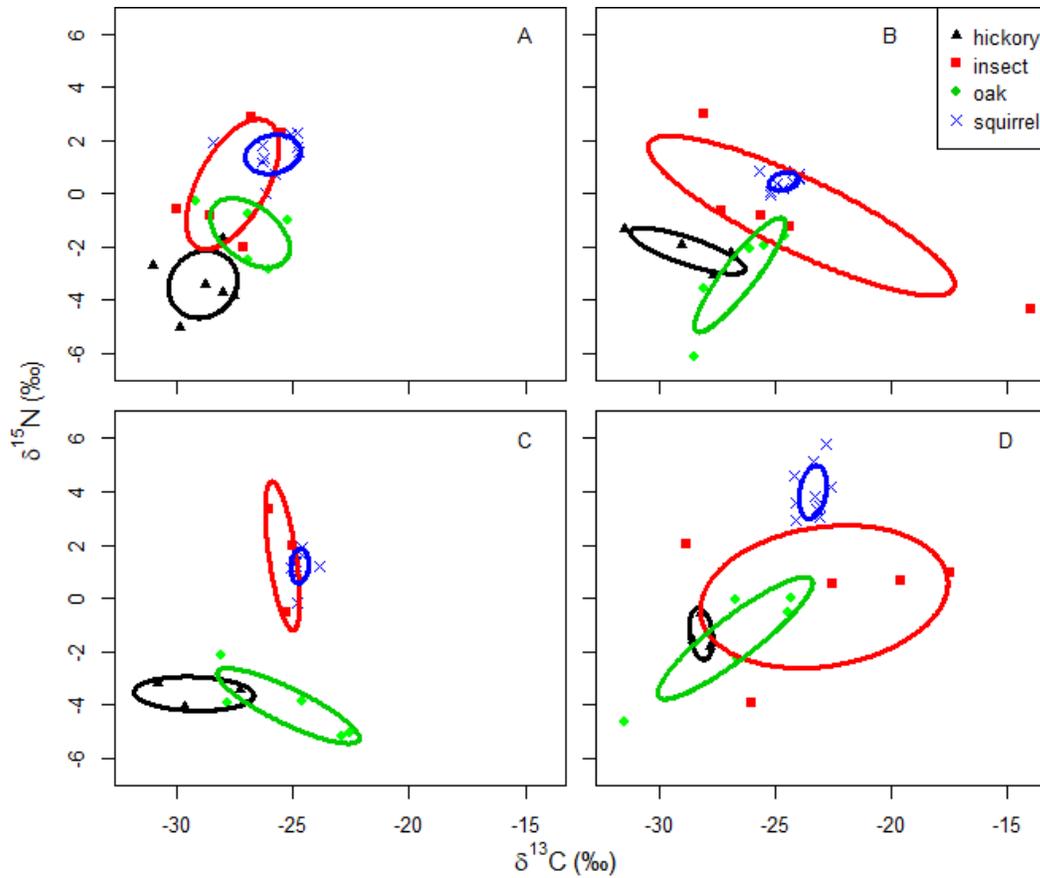


Figure 3.2 Squirrel and diet isotope ellipses by site

Standard 80% isotope ellipses constructed for southern flying squirrel (*Glaucomys volans*) livers and diet items sampled from a) U.S. Fish and Wildlife Service Sam D. Hamilton Noxubee National Wildlife Refuge (2016), b) U.S. Forest Service Talladega National Forest Oakmulgee District (2017), c) Alabama Department of Conservation and Natural Resources Coosa Wildlife Management Area (2017), and d) Sehoy Plantation (2017) during the spring season.

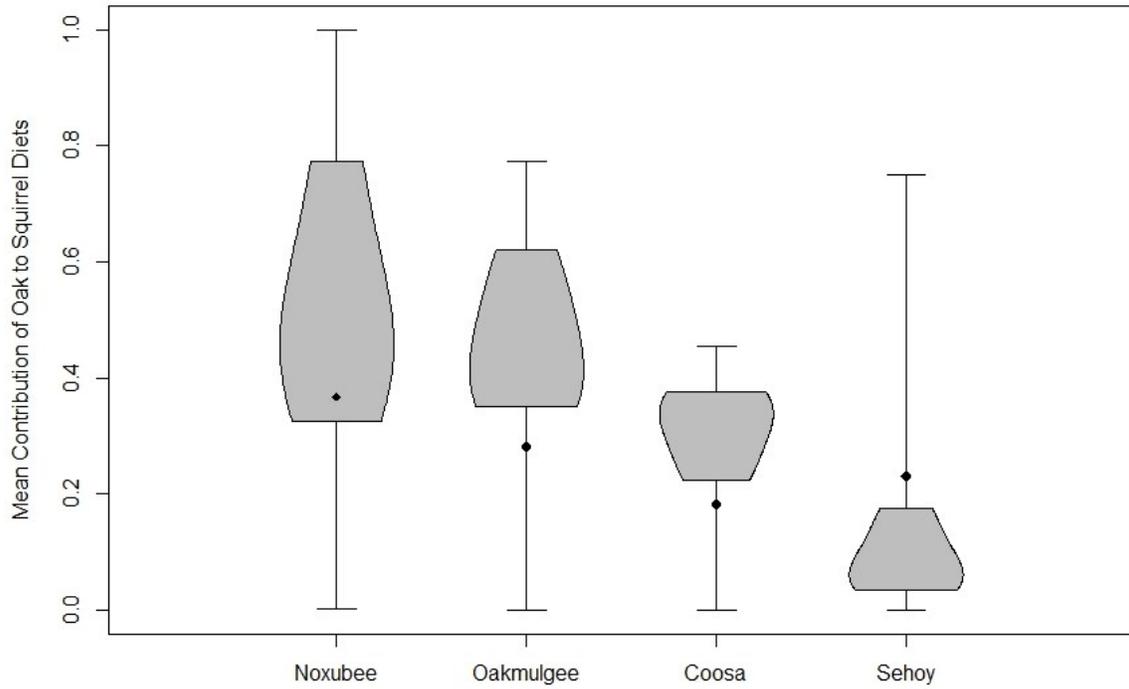


Figure 3.3 Oak/hardwood violin plot by site

Violin plot reflecting mean contribution of oak (*Quercus sp*) to the diet of southern flying squirrel (*Glaucomys volans*) in each sample site (x-axis). Shaded areas represent the 100% kernel densities of the proportion of hardwood land cover (NLCD cover types [Homer et al. 2015] 41, 90 and half of 43) in the areas sampled for southern flying squirrels.

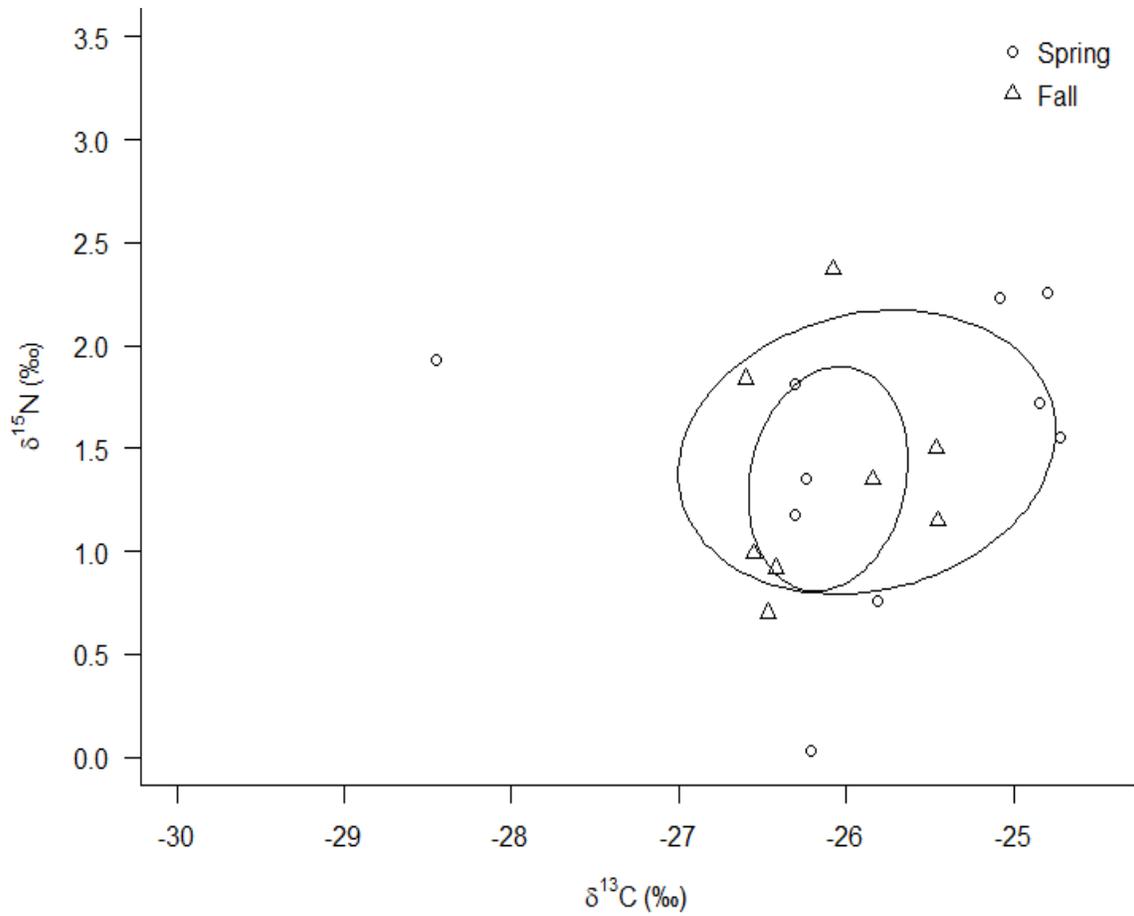


Figure 3.4 Seasonal isotope ellipses for Noxubee

Comparison of fall 2015 (inner ellipse) and spring 2016 (outer ellipse) isotope values for southern flying squirrel (*Glaucomys volans*) at Sam D. Hamilton Noxubee National Wildlife Refuge, Mississippi.

References

- Bendel, P. R., and J. E. Gates. 1987. Home range and microhabitat partitioning of southern flying squirrel (*Glaucomys volans*). *Journal of Mammalogy* 68:243–255.
- Borgo, J. S., M. R. Conover, and L. M. Conner. 2010. Flying squirrel removal does not reduce their use of simulated red-cockaded woodpecker nest clusters. *Southeastern Naturalist* 9:813–820.
- Caut, S., E. Angulo and F. Courchamp. 2007. Discrimination factors ($\Delta^{15}\text{N}$ and $\Delta^{13}\text{C}$) in an omnivorous consumer: effect of diet isotopic ratio. *Functional Ecology* 22:255–263.
- Conner, R. N. and B. A. Locke. 1982. Fungi and red-cockaded woodpecker cavity trees. *The Wilson Bulletin* 94:64–70.
- K. A. O'Halloran. 1987. Cavity-tree selection by red-cockaded woodpeckers as related to growth dynamics of southern pine. *The Wilson Bulletin* 99:398–412.
- D. C. Rudolph. 1995. Excavation dynamics and use patterns of red-cockaded woodpecker cavities: relationships with cooperative breeding. Pp. 343–352 in *Red-cockaded woodpecker: recovery, ecology, and management* (D. L. Kulhavy, R. G. Hooper, and R. Costa, eds.). Center for Applied Studies, College of Forestry, Stephen F. Austin State University. Nacogdoches, Texas, USA.
- D. C. Rudolph, D. Saenz, and R. R. Schaefer. 1996. Red-cockaded woodpecker nesting success, forest structure, and southern flying squirrels in Texas. *The Wilson Bulletin* 108:697–711.
- DeNiro, M. and J. S. Epstein. 1978. Influences of diet on the distribution of carbon isotopes in animals. *Geochimica et Cosmochimica Acta* 42:495–506.
- J. S. Epstein. 1981. Influences of diet on the distribution of nitrogen isotopes in animals. *Geochimica et Cosmochimica Acta* 42:495–506.
- Dennis, J. V. 1971. Species using red-cockaded woodpecker holes in northeastern South Carolina. *Bird-banding* 42:79–87.
- Dolan, P. G. and D. C. Carter. 1977. *Glaucomys volans*. *Mammalian Species*. American Society of Mammalogists 78:1–6.

- Fridell, R. A., and J. A. Litvaitis. 1991. Influence of resource distribution and abundance on home range characteristics of southern flying squirrels. *Canadian Journal of Zoology* 69: 2589–2593.
- Garroway C. J., J. Bowman, and P. J. Wilson. 2013. Complex social structure of Southern Flying Squirrels is related to spatial proximity but not kinship. *Behavioral Ecology and Sociobiology* 67:113–122.
- Gilmore, R. M., and J.E. Gates. 1985. Habitat use by the southern flying squirrel at a hemlock-northern hardwood ecotone. *Journal of Wildlife Management* 49:703–710.
- Harlow R. F., and A.T. Doyle. 1990. Food habits of southern flying squirrels (*Glaucomys volans*) collected from red-cockaded woodpecker (*Picoides borealis*) colonies in South Carolina. *American Midland Naturalist* 124:187–191.
- Helmick, K. R., T. L. Barrett, and G. W. Barrett. 2014. Dietary resource preference of the southern flying squirrel (*Glaucomys volans*). *American Midland Naturalist* 171:371–374.
- Herrera, L.G., E. Gutierrez, K.A. Hobson, B. Altube, W.G. Diaz and V. Sanchez-Cordero. 2002. Sources of assimilated protein in five species of New World frugivorous bats. *Oecologia* 133:280–287.
- Hoefs, J. 2009. *Stable isotope geochemistry*. 3rd ed. Springer-Verlag Berlin Heidelberg. Berlin, Germany.
- Homer, C.G., J.A. Dewitz, L. Yang, S. Jin, P. Danielson, G. Xian, J. Coulston, N.D. Herold, J.D. Wickham, and K. Megown. 2015. Completion of the 2011 national land cover database for the conterminous United States-representing a decade of land cover change information. *Photogrammetric Engineering and Remote Sensing* 81:345–354.
- Jackson, A. L. and A.C. Parnell. 2011. Comparing isotopic niche widths among and within communities: SIBER- stable isotope Bayesian ellipses in R. *Journal of Animal Ecology* 80:595–602.
- Jackson, J. A., M.R. Lennartz and R. G. Hooper. 1979. Tree age and cavity initiation by red-cockaded woodpeckers. *Journal of Forestry* 77:102–103.
- Johnson, P. S., S. R. Shifley, and R. Rogers. 2009. *The ecology and silviculture of oaks*. CABI International New York, New York, USA.
- Kappes, J.J. Jr. 2008. Cavity number and use by other species as correlates of group size in red-cockaded woodpeckers. *Wilson Ornithological Society*. 120: 181–189.

- Kurle, C. M., P. L. Koch, B. R. Tershy, and D. A. Croll. 2014. The effects of sex, tissue type, and dietary components on stable isotope discrimination factors ($\Delta^{13}\text{C}$ and $\Delta^{15}\text{N}$) in mammalian omnivores. *Isotopes in Environmental and Health Studies* 50:307–321.
- Laves, K.S. and S.C. Loeb. 1999. Effects of southern flying squirrels *Glaucomys volans* on red-cockaded woodpecker *Picoides borealis* reproductive success. *Animal Conservation* 2:295–303.
- Loeb, S. C. 1993. Use and selection of red-cockaded woodpecker cavities by southern flying squirrels. *Journal of Wildlife Management* 57:329–335.
- S. L. Reid, and D. J. Lipscomb. 2012. Habitat and landscape correlates of southern flying squirrel use of red-cockaded woodpecker clusters. *Journal of Wildlife Management* 76:1509–1518.
- MacArthur, R. H., and E. R. Pianka. 1966. On optimal use of a patchy environment. *The American Naturalist* 100:603–609.
- McCabe, R. A. 1947. Homing of flying squirrels. *Journal of Mammology* 28:404.
- Moore, J.W. and B.X. Semmens. 2008. Incorporating uncertainty and prior information into stable isotope mixing models. *Ecology Letters* 11:470–480.
- Nebeker, T. E., M. Pelligrine, R. A. Tisdale, and J. D. Hodges. 1995. Site/stand factors associated with red-cockaded woodpecker colonies on the Noxubee National Wildlife Refuge, Mississippi. Pages 431–435 in D. L. Kulhavy, R. G. Hooper, and R. Costa, editors. *Red-cockaded woodpecker: recovery, ecology, and management*. Center for Applied Studies, College of Forestry, Stephen F. Austin State University. Nacogdoches, Texas, USA.
- Olson, D. F. Jr. 1974. *Quercus L. Oak*. USDA Forest Service Agriculture Handbook 450. USDA Forest Service, Washington, DC, 88 pp.
- Phillips, D. L., R. Inger, S. Bearhop, A. L. Jackson, J. W. Moore, A. C. Parnell, B. X. Semmens, and E. J. Ward. 2014. Best practices for use of stable isotope mixing models in food-web studies. *Canadian Journal of Zoology* 92:823–835.
- Schwartz, W.W., and E.R. Schwartz. 1959. *The wild mammals of Missouri*. University of Missouri Press and Missouri Conservation Commission, Kansas City MO.
- Sork, V. L., J. E. Bramble. 1993. Prediction of acorn crops in three species of North American oaks: *Quercus alba*, *Q. rubra* and *Q. velutina*. *Annales des sciences forestières* 50(Suppl1)128s–136s.

- Steele, M. A., B. C. McCarthy, and C. H. Keiffer. 2005. Seed dispersal, seed predation, and the American chestnut. *Journal of the American Chestnut Foundation* 19:47–54.
- Stock, B. C. and B. X. Semmens. 2016. MixSIAR GUI user manual. Version 3.1. <https://github.com/brianstock/MixSIAR/>. doi:10.5281/zenodo.47719.
- Stone K. D., G. A. Heidt, P. T. Caster, and M. L. Kennedy. 1997. Using geographic information systems to determine home range of the southern flying squirrel (*Glaucomys volans*). *American Midland Naturalist* 137:106–111.
- Straub, J. N., R. M. Kaminski, A. G. Leach, A. W. Ezell, and T. Leininger. 2016. Acorn yield and masting traits of red oaks in the lower Mississippi river alluvial valley. *Forest Science* 62:18–27.
- U.S. Fish and Wildlife Service. 2003. Recovery plan for the red-cockaded woodpecker (*Picoides borealis*): second revision. U.S. Fish and Wildlife Service, Atlanta, GA. 296 Pp.
- U.S. Fish and Wildlife Service. 2014. Comprehensive conservation plan and environmental assessment for Sam D. Hamilton Noxubee National Wildlife Refuge. U.S. Fish and Wildlife Service, Atlanta, GA. Volume 1.
- Walters, J. R. 1990. The red-cockaded woodpecker: a “primitive” cooperative breeder. Pp. 67–101 in *Cooperative breeding in birds: long term studies of ecology and behavior* (P.B. Stacey and W. D. Koenig, eds.). Cambridge University Press, Cambridge, United Kingdom.
- Zar, J.H. 1999. *Biostatistical Analysis*, Fourth Edition. Prentice Hall, Upper Saddle River, NJ.

CHAPTER IV
AN ANALYSIS OF NON-LETHAL DETERRENTS TO SOUTHERN FLYING
SQUIRREL (*GLAUOMYS VOLANS*) USE OF CAVITIES

Abstract

The southern flying squirrel (SFS; *Glaucomys volans*) can negatively affect the endangered red-cockaded woodpecker (RCW; *Picoides borealis*) by displacing individuals from tree cavities and through destruction of eggs and young. To limit this interaction, I tested several non-lethal odor deterrents for excluding SFSs from nest boxes. Deterrent trials consisted of one SFS within an outdoor cage where they were given a choice of two nest boxes: a control (only water applied) and a box with a scent. Scents included grey rat snake (*Pantherophis spiloides*) feces, peppermint, and synthetic weasel musk. Binomial tests to determine if the probability of using the scented box for the final cavity to rest in was different than chance (50:50) and showed significant results for snake feces ($\hat{p} = 0.28$, $P = 0.05$), marginal results for synthetic weasel musk ($\hat{p} = 0.4$, $P = 0.25$), and insignificant results for peppermint ($\hat{p} = 0.47$, $P = 0.50$). Chi-square statistics for number of entrances and investigations showed no significant differences than expected by chance ($P > 0.05$). This study indicates SFSs react negatively to the smell of grey rat snakes. Further investigation should be conducted to determine the optimal quantity of scent and applicability to RCW cavities.

Introduction

The red-cockaded woodpecker (RCW; *Picoides borealis*) is an endangered species endemic to southeastern pine forests (U.S. Fish and Wildlife Service, 2003). RCWs create cavities in living, old-growth pines with hearts typically weakened by red heart-rot fungus (*Phellinus pini* [Thore ex. Fr.]) (Conner and Locke 1982). RCWs create cavities in groups of trees referred to as ‘clusters’ (Walters 1990). It can take a group of RCWs a few months to more than a year to make a single cavity (Jackson et al 1979, Conner and Rudolph 1995) making cavity space an important part in their management. The southern flying squirrel (SFS; *Glaucomys volans*) is a threat to RCWs through nest predation and cavity usurpation (Conner et al. 1996; Dennis 1971; Laves and Loeb 1999; Loeb 1993; U.S. Fish and Wildlife Service 2003).

SFSs present a unique conservation challenge for RCW managers because SFSs can glide to RCW cavities making mechanical barriers on cavity trees ineffective. To date, there is no known method for successfully excluding SFSs from RCW cavities (Montague et al. 1995). An alternative to mechanical barriers for barring SFS from RCW cavities is to use olfactory deterrents. Borgo et al. (2006) was able to show that SFSs use scent to determine safety of potential cavities to rest in. Their findings reveal that SFSs are less likely to use cavities inoculated with the scent of grey rat snake (*Pantherophis spiloides*) than cavities that are not treated with an olfactory deterrent. Although Borgo et al. (2006) found an effect of gray rat snake musk on cavity selection. However, until musk can be harvested or replicated synthetically, the use of musk as a deterrent remains inefficient and troublesome.

I expand upon Borgo et al.'s (2006) findings by testing the interaction between snake scent and cavity choice by SFSs. The current study differs from Borgo et al. (2006) in that I used snake feces instead of snake musk as feces are more easily obtained and have a more practical application for management. Additionally, I tested other deterrents that have very pungent odors, such as peppermint and weasel musk, to assess their utility as potential deterrents to SFS entering cavities used by RCW.

Methods

Deterrent Scent Trials

For the deterrent trials, I constructed eight outdoor enclosures each measuring 2.40m x 2.40m x 2.40m using treated pine wood and steel wire mesh. SFSs were donated by the U.S. Fish and Wildlife Service during normal duties from the Sam D. Hamilton Noxubee National Wildlife Refuge (hereafter; Noxubee) located in east-central Mississippi. Food consisting of acorns, hickory seed, rodent mix, and water were provided to each SFS *ad libitum* while they are in captivity. Polyester fibers were placed in each box (~0.50g) for thermal regulation. SFSs were given two days in the enclosure before trials are initiated to become familiar to the placement of test boxes.

I placed two nest boxes on opposing walls approximately 1.50m above the ground in each enclosure for each trial. Each nest box was made out of untreated pine with internal dimensions 10cm x 10cm x 25cm with a 3.80cm diameter entrance on the left side of each nest box. Nest boxes were left untreated with deterrent during the two-day acclimation period. I re-used boxes throughout the study (15 boxes total), and for each trial gave SFSs a different combination of boxes to avoid familiarity with boxes they encountered in a previous trial, or to those encountered the previous night in a cross-over

design. Due to prior evidence that SFSs may select boxes that smell like conspecifics (Borgo et al. 2006), boxes were scrapped before each trial with clean sandpaper to remove any feces or SFS musk. Between each trial SFSs were given one night in which they had two boxes with no scent to allow the SFSs to re-acclimate to the boxes on either side. Since SFSs are nocturnal, trials were conducted one half hour after sunset and continued until one half hour before sunrise.

Each treatment was applied via a 7ml vial (1.30cm diameter by 7.60cm long) with a plant-fiber sponge (weighed for consistency to 0.25g) inside the vial and placed within the wall of the box under the entrance hole (Fig. 3.1). The control for each trial was water placed in a vial in a similar manor as the treatment. Vials were shaken vigorously to coat the sponge before setting them in the corresponding box. In this way, I hoped to increase the surface area of the mixture and therefore increase the evaporative surface for which the smell can disperse. I analyzed avoidance behavior of SFS for each scent tested by recording 1) the final box choice, 2) number of times each box is entered, and 3) the number of investigations. Investigations were defined as events where SFSs spent ≥ 1 second at the entrance hole and either sniffing the entrance or not moving. In this way, I hope to capture a sense of hesitation that may be associated with each box. Data were recorded using video from motion-triggered infrared trail cameras (Bushnell, Kansas USA). Due to the limitations of the cameras and the speed of the SFSs, I was unable to accurately estimate entrance and exit times to calculate total time spent in each box.

I used grey rat snake (*Pantherophis spiloides*) feces from a wild-caught, captive snake for the snake scent trial (Trial 1). The snake was fed a diet of SFSs that had been euthanized, frozen and thawed. SFSs provided to the snake were euthanized as part of the

U.S. Fish and Wildlife Service RCW management. Feces collected from the snake were frozen, then freeze-dried, and then pulverized to homogenize. The resulting fecal sample was weighed to 0.50g, added to 5ml of water, and placed in the vial with the sponge just prior to the trial.

For Trial 1, the SFS was given a choice of two boxes. The vial containing the feces mixture was placed at the entrance hole of the treatment box. The control box had only water and the sponge with the vial at the entrance hole (same control for all trials). Trial 2 consisted of a control and a synthetic weasel musk-treated box with only 5ml of musk (J.R. and Sons, Monroeville OH, USA) added to the vial with the sponge and placed beneath the entrance hole in the same manner as Trial 1. Trial 3 consisted of a control and a peppermint treatment. Peppermint is a strong smell that has been used in commercial anti-squirrel products before (Olmos 2012) and serves as a pungent, non-predator scent. The treatment vial contained 5ml of peppermint oil and placed within the entrance of the treatment box. Each SFS was given a random order of trials such that each possible combination was executed at least once during the study (randomized complete block design) to control for the possible effect of the order in which the SFS receives the scents.

Statistical Analysis

I left SFSs in the cages over multiple days, and thus SFS may have selected a box based on the box used in the previous night. Additionally, a SFS can become acclimated to scented boxes over time if the SFS has gone through multiple trials. To account for this possible bias I conducted a generalized linear mixed effects model with previous box choice, the trial sequence (1st, 2nd, or 3rd), and deterrent scent as fixed effects and the SFS

as a random effect with the binomial response being final box choice. *Post hoc* Tukey honest significance difference (HSD) test was used to determine how covariates affected the final box choice. I tested the probability of a SFS choosing a treatment box to a probability of 0.5 using a binomial test. I used a Chi-square test to determine if the number of entrances and investigation events differed than expected by 50:50 chance (Zar 1999). All statistical tests were considered significant at $\alpha \leq 0.05$.

Results

Deterrents varied in how they affected SFS cavity selection. The location of the previous box choice was independent of the next nights' box choice (Tukey HSD, $P = 0.9$). The probability that a SFS selected a particular treatment as the final box choice differed relative to whether the deterrent used was snake feces, weasel musk, or peppermint oil: snake feces (\hat{p} , estimated probability of use = 0.28, 95% $CI = 0-0.5$, $P = 0.05$, $n = 18$), outperformed synthetic weasel musk ($\hat{p} = 0.40$, 95% $CI = 0-0.61$, $P = 0.25$, $n = 20$), and peppermint ($\hat{p} = 0.47$, 95% $CI = 0-0.69$, $P = 0.50$, $n = 17$; Fig. 3.2).

Generalized linear mixed effects model revealed no significant effect of previous box choice (the side of the enclosure that the SFS chose in the previous night), trial order or between specific deterrent on the final box site choice ($P > 0.05$). Additionally, there was no detectable difference in SFS use of boxes among treatments (Tukey HSD, $P > 0.50$). Chi-square tests revealed no significant differences between control or treatment boxes for any trial for number of entrances or investigations ($P > 0.05$).

Discussion

Results of this study support the findings of Borgo et al. (2006) for grey rat snake scent as a SFS deterrent. It is interesting to note that my study used snake feces whereas Borgo et al. (2006) used snake musk, yet both snake feces and snake musk seem to deter SFSs. Of the three deterrents tested, only snake feces was shown to decrease the probability of a SFS choosing the cavity (Fig. 3.2). Synthetic weasel musk did not appear to deter SFS significantly but it did perform better than peppermint extract, which showed little observable effect on box choice. These results re-confirm that SFS choose cavities based partly on smell (Borgo et al. 2006). It is interesting to note that the two deterrents that showed avoidance were those based on predator scents. It may be that SFS displayed a stronger reaction to snake feces due to the abundance of grey rat snakes from the capture site. Comparatively, there are very few Mustelids in this area. Thus SFSs may be unfamiliar with weasel musk and thus did not display a strong avoidance of boxes treated with weasel musk scent. The lack of significant difference for weasel musk could also be due to the chemical make-up of the synthetic compounds that may not accurately represent true weasel musk.

Although a SFS may avoid choosing a cavity based on its scent, it doesn't necessarily prevent entrances into the cavity. SFSs may not choose RCW cavities as a nest but even if entering for a short time, they may break or consume RCW eggs or injure nestling. Subsequently, I quantified the number of entrances SFS make into control and treatment boxes for each trial. I found no significant difference in entrance rate per treatment (Chi-square, $P > 0.05$). One explanation for this lack of significant difference may owe to the squirrels moving faster than the cameras could record. On several

occasions, SFSs in the enclosure entered and exited boxes faster than the camera could detect them, with the cameras often taking video after the SFS had entered the box. I would advise future studies to use continuous video to obtain estimates that are more accurate.

My study methods differed from Borgo et al. (2006) in that I applied the scent to the entrance of the box, whereas Borgo et al. (2006) applied the scent to the inside of the box. As my results were similar to Borgo et al.'s (2006), these findings indicate that it may not be necessary to apply the scent to the inside of the cavity to have an effect. Application of a scent inside an RCW cavity could coat the eggs in the nest thus increasing potential harm to the developing birds, but an entrance-hole application would not harm the eggs. The way in which I applied the scent had the benefit of limiting direct contact with any animal entering the cavity.

To my knowledge, only one field study has been conducted to test the effectiveness of grey rat snake musk as a deterrent in an RCW cluster. Stober and Conner (2007) applied grey rat snake musk in 6–7ml doses of 1% solution to the outside of an RCW cavity every seven days to determine its effects on SFS occupancy of the cavity. Stober and Conner (2007) did not find a significant effect but this may be due to several reasons. The spray application of grey rat snake musk to the outside of the cavity could cause the scent to disperse too quickly due to wind, rain, or through gravitational action. The concentration used in Stober and Conner (2007) may not have been sufficient to last a seven-day treatment rotation. The current study used only 0.50g of feces in 5ml of water and yet that amount and concentration was sufficient to notice a difference in treated and control boxes over the course of one night. This concentration over multiple

days may have neutral or diminishing effects on SFSs. Further studies must optimize the concentration of the scent and time to create the greatest deterrent effects. For these reasons, more tests are needed before it should be broadly applied to RCW cavities. I believe the evidence provided in this study should be sufficient to warrant further investigation into grey rat snake scents as odor deterrents to SFSs.

Tables and Figures

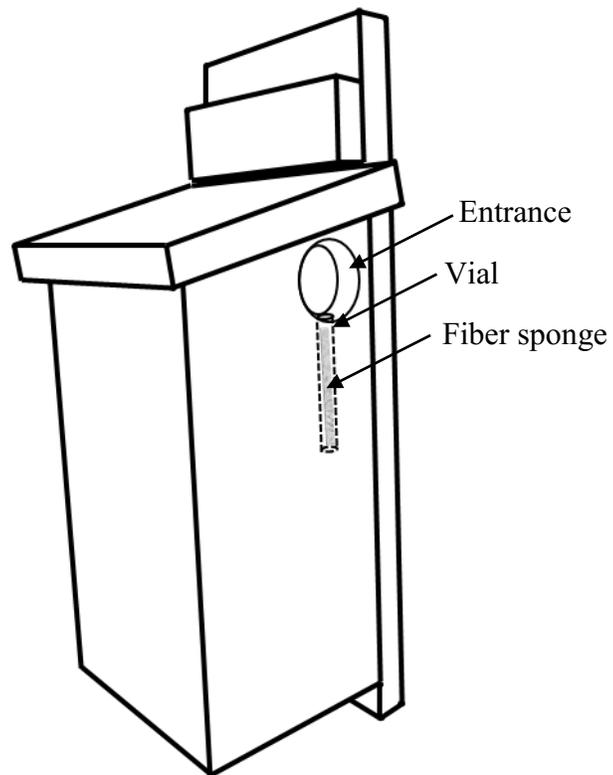


Figure 4.1 Trial box design.

Control and treatment box design for southern flying squirrel (*Glaucomys volans*) trials. Vials containing the scent or water (control) were placed within the wall of the box, below the entrance hole.

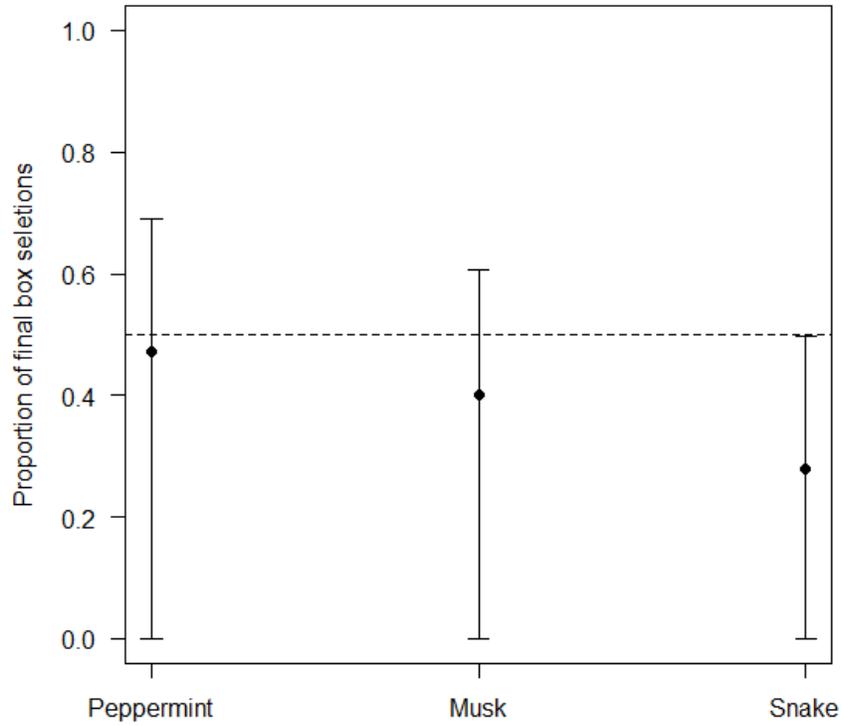


Figure 4.2 Final cavity selection.

Proportion of southern flying squirrel (*Glaucomys volans*) final box selections for the treated box for peppermint (n = 17), synthetic weasel musk (n = 20), and snake feces (n = 18). Dashed line represents the null hypothesis that there is no difference in the proportion of treatment choices.

References

- Borgo, J. S., L. M. Conner, and M. R. Conover. 2006. Role of predator odor in roost site selection of southern flying squirrels. *Wildlife Society Bulletin* 34: 144–149
- Conner, D. C. Rudolph, D. Saenz, and R. R. Schaefer. 1996. Red-cockaded woodpecker nesting success, forest structure, and southern flying squirrels in Texas. *Wilson Ornithological Society* 108:697–711.
- Dennis, J. V. 1971. Species using red-cockaded woodpecker holes in northeastern South Carolina. *Bird-banding* 42:79–87.
- Jackson, J. A., M.R. Lennartz, and R. G. Hooper. 1979. Tree age and cavity initiation by Red-cockaded Woodpeckers. *Journal of Forestry* 77:102–103.
- Laves, K.S. and S.C. Loeb. 1999. Effects of southern flying squirrels *Glaucomys volans* on red-cockaded woodpecker *Picoides borealis* reproductive success. *Animal Conservation* 2:295–303.
- Loeb, S. C. 1993. Use and selection of red-cockaded woodpecker cavities by southern flying squirrels. *Journal of Wildlife Management* 57:329–335.
- Montague, W.G., J.C. Neal and J.E. Johnson. 1995. Techniques for excluding southern flying squirrels from cavities of Red-cockaded Woodpeckers. Pages 401–409 in D. L. Kulhavy, R. G. Hooper, and R. Costa, editors. *Red-cockaded Woodpecker: recovery, ecology, and management*. Center for Applied Studies, College of Forestry, Stephen F. Austin State University. Nacogdoches, Texas, USA.
- Olmos, Mario. 2012. Scented birdseed as a squirrel deterrent. US patent 20120177778 A1.
- Stober, J. M., and M. L. Conner. 2007. Scent deterrence to reduce southern flying squirrel kleptoparasitism of red-cockaded woodpecker cavities. *Human-Wildlife Conflicts* 1: 45–48.
- Walters, J. R. 1990. The red-cockaded woodpecker: a “primitive” cooperative breeder. Pp. 67–101 in *Cooperative breeding in birds: long term studies of ecology and behavior* (P.B. Stacey and W. D. Koenig, eds.). Cambridge University Press, Cambridge, United Kingdom.
- Zar, J.H. 1999. *Biostatistical Analysis*, Fourth Edition. Prentice Hall, Upper Saddle River, NJ.