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The Effects of Forest Fragment Size, Isolation, and Microhabitat Variables on Nest Box Use by Southern Flying Squirrels (*Glaucomys volans*) in Southern Illinois

Abstract

*We studied the effects of forest fragmentation on southern flying squirrels (*Glaucomys volans*) in southern Illinois, and examined factors influencing nest box use. Ten nest boxes were placed in each of 30 oak-hickory forest fragments. Nest boxes were checked monthly after installation. We measured habitat variables that described the nest box tree, the microhabitat surrounding the nest box tree, and landscape level characteristics of the fragments (isolation and area). Overall, 78 % of the 300 nest boxes were used. *Glaucomys volans* was captured in 24 of the 30 forest fragments, and evidence of squirrel use was found in 4 additional fragments. Only 2 fragments showed no evidence of squirrel use, suggesting *G. volans* may not be particularly sensitive to effects of fragmentation in a primarily forested landscape like southern Illinois. However, the 2 fragments apparently lacking squirrels were small and isolated. Stepwise logistic regression indicated that *G. volans* was more likely to use boxes that were on trees with a smaller diameter. Used boxes also occurred in areas with less ground cover, more hard mast trees, and fewer fallen logs than unused boxes.*

Keywords

Glaucomys volans, habitat fragmentation, nest boxes

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1. Introduction

Glaucomys volans is a nocturnal sciurid [15] that received relatively little study (but see [17, 24]) until artificial nest boxes were implemented as a research tool [12, 19, 26]. Artificial nest boxes simplified study of flying squirrels and increased our knowledge of their reproductive ecology [21], behavior [7], and home-range [27]. *Glaucomys* often uses boxes as dens during the day, making them easily accessible to researchers year-round, although summer use is often reduced due to increased temperatures [12].

Microhabitat variables that influence the use of cavities and nest boxes by *G. volans* have been studied in Arkansas [28, 29], Louisiana [9], Maryland [1, 8], Missouri [2], and Virginia [25]. Muul [19] reviewed habitats in which *G. volans* nests have been found, and suggested this species is not restricted to a certain forest or tree type. He noted that nest tree selection and cavity height followed that of woodpeckers (*Dendrocopos villosus* and *D. pubescens*), since *G. volans* does not excavate cavities.

Despite these studies, few if any variables have been found to be consistently important in predicting nest box use. Differences may reflect geographic variation as well as methods of analysis, but clearly more study over the geographic range of this species is needed. No recent studies have been completed on *G. volans* in Illinois, nor have studies addressed broader landscape-level variables that also may influence use of nest boxes. Forest fragmentation may cause significant barriers to dispersing animals, and there has been increased interest in determining how landscape level characteristics affect population dynamics and inter-patch movements of mammals [4, 6, 23].

Overall size and isolation of habitat patches are important factors that must be considered when determining what effects habitat fragmentation may have on a species [10]. Van Dorp and Opdam [32] used logistic regression and found woodlot size was the most important predictor of bird species occurrence. Studies of red squirrels (*Sciurus vulgaris*) in the Netherlands and Italy found woodlot size and isolation (distance to nearest 'source area') were factors that influenced presence or absence in habitat fragments [3, 33].

Glaucomys volans is usually associated with hardwood trees [35]. They are secondary-cavity nesters, and usually occupy woodpecker holes and other cavities [19]. Their diet consists primarily of hard mast, especially acorns [11]. Locomotion over long distances involves gliding from tree to tree in a descending fashion [7]. These traits make *G. volans* susceptible to forest fragmentation. Agricultural practices and increased urbanization have decreased forested area in Illinois from 38.2 % in 1820 to approximately 12 % in 1985 [16], and much of the remaining forest is distributed in isolated woodlots.

The objective of this study was to determine how area and isolation of forest fragments affect *G. volans* in southern Illinois. We also examined microhabitat variables that influence nest box use.

2. Methods

2.1 Study Areas

Thirty study sites were selected in and around the Shawnee National Forest in Jackson, Johnson, Union, and Williamson counties in southern Illinois, USA. Sites ranged in size from 6 to 5264 ha, and were primarily oak-hickory (*Quercus* spp. - *Carya* spp.) forests [37]. Seven sites were considered “very small” (6–10 ha), 7 were considered “small” (26–81 ha), 7 were considered “medium” (100–223 ha), and 9 were considered “large” (645–5264 ha). United States Geological Survey (USGS) maps (photo revised 1990) were used to identify and locate forest fragments. SigmaScan[®] (SPSS Inc., 444 N. Michigan Ave., Chicago, IL 60611) was used to measure areas from the USGS maps. Isolation was defined as the distance from a forest fragment to the next fragment of at least 5 ha. All sites were similar with respect to forest type, age, and topography, and all contained apparently suitable habitat for flying squirrels.

2.2 Nest Boxes and Capture Techniques

Roughly square grids with 10 nest boxes spaced 50 m apart were established on each of the 30 sites. Nest boxes were constructed with inside dimensions of 9 × 13 × 18 cm and an entrance hole 3.3 cm in diameter [13]. Boxes were mounted approximately 2.2 m high on the south sides of trees between March and June 1996. All nest boxes were placed on trees with a diameter at breast height of at least 32 cm.

Boxes were checked approximately every four weeks. A total of 3870 box checks were made between April 1996 and June 1997. Box use was classified following Heidt [12] and Muul [19] as: (1) nest site, (2) feeding station, (3) defecatoria, (4) combination use, or (5) no use. A box was considered a nest site if it contained nesting material characteristically used by *G. volans* (shredded bark and/or leaves). A feeding station was a box that contained acorns, hickory nuts, and other food items. Hard mast was examined for evidence of gnawings typical of *G. volans*. A box was considered a defecatoria if it contained *G. volans* feces, and classified as combination use if it showed evidence of more than one type of use. Usually, a combination box was initially a nest site, and then converted to a feeding station or defecatoria.

Captured squirrels were weighed, sexed, and marked with individually numbered ear tags. Reproductive condition (males = scrotal/non-scrotal, females = perforate vaginas, lactation, pregnant, or non-reproductive) was noted for each captured animal. Age classes were determined by body weight following Raymond and Layne [21]. Squirrels weighing over 50 g were considered adults, subadults were 25–50 g, and juveniles were less than 25 g. Animals were released at the point of capture.

2.3 Habitat Sampling

Vegetation associated with each nest box tree was sampled in June and July 1996 in 700-m² circular plots centered on the nest box tree. Diameters at breast height (DBH) of all trees ≥ 8 cm were measured. Trees were identified to genus, and placed into size classes of 8–10 cm DBH, 10–20 cm DBH, 20–30 cm DBH, 30–40 cm DBH, and > 40 cm DBH. Trees and snags were classified (as in [30]) into 9 stages representing a continuum from living tree (1) to stump (9). Additionally, logs (> 8 cm DBH) were assigned a decomposition classification [18] ranging from newly fallen (1) to near complete decomposition (5).

Species, height, bark texture (rated from 1 = smooth to 4 = very rough) [2], and DBH of each nest box tree was recorded. Canopy cover was estimated with a densiometer, and ground cover was estimated visually using 1 m² circular quadrats. Four estimates of canopy cover and ground cover were taken 5 m from each nest box tree in each cardinal direction. Habitat variables selected for statistical analysis were modified from Gilmore and Gates [8], Boardman [2], and Stone et al. [28].

2.4 Statistical Analysis

Stepwise logistic regression with forward selection (PROC LOGISTIC, Stepwise option, SAS [22]) was used to model nest box use by *G. volans*. Boxes used as nests, defecatoria, and feeding stations were included in the analysis. In addition, we looked at unused boxes versus those with any type of flying squirrel use. Initially, 47 habitat variables were considered for inclusion in the model. However, some variables exhibited strong colinearities. For example, relative density of hard mast trees was highly correlated with basal area of hard mast trees ($r = 0.96$). Thus, only relative density of hard mast trees was used as an independent variable. When variables were correlated, the variable used most commonly in other studies was selected. In 2 cases, variables were combined to create new variables (trees in the 2 largest DBH classes and snags in stages 2 and 3). Snags in decomposition classes 2 and 3 were grouped together because they were presumed best suited for use by *G. volans*, and there were few snags in other classes. No significant correlations existed among the 12 variables used as independent variables (Table 1). Independent variables generally did not deviate greatly from normality. Small deviations were tolerated given the robustness of the logistic regression technique, and the descriptive nature of the project. In an effort to maximize parsimony, the significance level for entry into the model was 0.05.

Spearman rank correlations (r) were used to determine if any significant associations existed between flying squirrel abundance and forest fragment size or isolation. Pearson product-moment correlations were used to determine if vegetative characteristics were related to fragment size [22].

Table 1

Abbreviations and descriptions of habitat variables used as independent variables in the logistic regression model of nest box use by *Glaucomys volans*.

Variable	Description and method of collection
Bark Texture Nest Box Tree	Bark texture of nest box tree: smooth (1), medium (2), rough (3), very rough (4) (as in [19]).
% Canopy Cover	Canopy cover estimated 4 times in each plot to the north, south, east, and west of the nest box tree with a densiometer.
DBH Nest Box Tree	Diameter (cm) at breast height (1.4 m) of nest box tree.
Distance Closest Tree	Distance to the closest tree (m) from the nest box tree that was ≥ 8 cm DBH.
% Ground Cover	Ground cover (%) visually estimated 4 times to the north, south, east, and west of each nest box tree with a 1-m ² circular hoop.
Height Nest Box Tree	Height (m) of the nest box tree measured with a clinometer.
No. Large Trees	The number of trees (≥ 35 cm DBH) in plot.
No. Logs	Number of logs in plot.
No. Snags	The number of snags in plot.
Relative Density	Number of trees in plot ≥ 8 cm DBH
Relative Density Hard Mast Trees	Number of hard mast producing trees (oaks, hickories, and walnuts) ≥ 8 cm DBH in plot.
Snags Grouped	The number of snags classified as snag stage 2 or 3 in plot.

3. Results

3.1 Captures and Use of Boxes

A total of 203 individual *G. volans* were captured in 98 boxes. There were 74 “nest only” boxes, 44 “feeding station only” boxes, 16 “defecatoria only” boxes, and 100 “combination usage” boxes. Squirrels were recaptured on 64 occasions. Number of captures varied monthly with a peak in October. Mean number of squirrels captured per used box also varied monthly, but was highest in November (3.4), December (2.8), and January (3.3). The largest aggregation of

9 squirrels (in one box) was found in November. In fragments in which *G. volans* was detected, between 40 and 100 % of the boxes were used. Individual squirrels were never captured in more than one fragment.

3.2 Patterns of Occupancy

Glaucomys volans was present in 28 of 30 forest fragments. We captured flying squirrels in 24 of the 30 fragments and noted definitive evidence of squirrel presence (i.e., nests and feeding stations in nest boxes) in 4 additional fragments. Squirrels were absent from the 2 most isolated sites.

Overall, 78 % of the 300 nest boxes were used by flying squirrels at some point during the study. No relationship existed between percentage of nest boxes used in a fragment and area or isolation (Table 2). In addition, total number of squirrels captured was not correlated with area or isolation (Table 2). Similarly, there was no relationship between number of recaptures per fragment and area or isolation (Table 2). Finally, number of individual squirrels captured per woodlot was not correlated with area or isolation (Table 2). Sample sizes were insufficient to calculate densities for more than 2 of the woodlots, thus comparisons of flying squirrel density among fragments were not possible.

Table 2
Spearman rank correlations between various measures of *Glaucomys volans* abundance and both area and isolation for 30 forest fragments in southern Illinois.

Variable	Area	Isolation
Number of Captures	0.110 (P > 0.50)	-0.252 (P > 0.10)
Number of Recaptures	0.141 (P > 0.20)	-0.321 (P > 0.10)
Number of Individuals	0.093 (P > 0.50)	-0.221 (P > 0.20)
Number of Male Captures	0.023 (P > 0.50)	-0.165 (P > 0.20)
Number of Female Captures	0.018 (P > 0.50)	-0.201 (P > 0.20)
Number of Subadult Captures	-0.233 (P > 0.20)	0.169 (P > 0.20)
Number of Litters	-0.077 (P > 0.50)	0.341 (P > 0.05)
Mean Litter Size	-0.103 (P > 0.50)	0.368 (P > 0.05)
% Nest Boxes Used	0.126 (P > 0.50)	-0.307 (P > 0.10)

3.3 Vegetation

Pearson correlation coefficients revealed no significant relationships among habitat variables and area (all $r < 0.347$, all $P > 0.05$). Therefore, vegetative characteristics were relatively uniform among fragments of different sizes (Table 3). Habitat variables included in this analysis were % canopy cover, % ground cover, relative density of trees, relative density of hard mast trees,

number of snags, and number of logs. Variables associated with each nest box tree including height, DBH, distance to closest tree, and bark texture, also were tested to determine whether they correlated with fragment size. These variables were not affected by fragment size (all $r < 0.348$, all $P > 0.05$). Since no significant correlations were found between fragment size and habitat characteristics, we assumed that the habitat was fairly similar among forest fragments.

Table 3
Correlations between mean habitat variables associated with 10 *Glaucomys volans* nest boxes placed in 30 habitat fragments in southern Illinois and the area of the fragment.

Variable	Pearson r	P-value
Distance to Closest Tree	-0.146	0.442
Diameter at Breast Height of Nest Box Tree	0.347	0.061
Height of Nest Box Tree	0.244	0.193
Bark Texture of Nest Box Tree	-0.288	0.123
% Canopy Cover	0.120	0.527
% Ground Cover	-0.343	0.063
Relative Density Trees	-0.178	0.346
Relative Density Hard Mast Trees	-0.098	0.608
# of Snags	-0.177	0.350
# of Logs	0.204	0.279

3.4 Factors Influencing Nest Box Use

Stepwise logistic regression identified 4 variables associated with overall nest box use (Table 4). These were DBH (negative relationship), percent ground cover (negative relationship), density of hard mast trees (positive relationship), and number of fallen logs (negative relationship). The model was concordant 72.1 % of the time, meaning it correctly predicted whether or not a box would be used 72 % of the time (see [22]).

Variables included in a model of boxes used for nests were DBH (negative relationship), height of the nest box tree (positive relationship), and number of snags (negative relationship) (Table 4). This model was 68.9 % concordant. Density of hard mast trees (positive relationship) was the only variable included in a model of boxes used as defecatoria (58.3 % concordant) (Table 4). No variables were significant in our model of boxes used as feeding stations.

Table 4
Parameter estimates, chi-square values (1 d.f.) and p-values for logistic regression model of probability of nest box use by *Glaucomys volans* in southern Illinois.

Variable	b	Chi-Square	p
Overall Box Use			
DBH of Nest Box Tree	-0.0198	5.1468	< 0.0233
% Ground Cover	-1.6496	6.1185	< 0.0134
Relative Density Hard Mast Tree	+0.0646	9.3564	< 0.0022
No. Logs	-0.0796	6.2788	< 0.0122
Box Used as Nest			
DBH of Nest Box Tree	-0.0433	19.5442	< 0.0001
Height of Nest Box Tree	+0.0522	5.1730	< 0.0229
No. Snags	-0.0782	6.4872	< 0.0109
Box Used as Defecatoria			
DBH of Nest Box Tree	-0.0385	5.0218	< 0.0250
Box Used as Feeding Station			
No Variable Entered			

4. Discussion

4.1 Factors influencing Nest Box Use

We found that DBH of nest box trees was an important predictor of overall nest box use, and of which boxes were selected for nests. This is in agreement with Stojeba [29], who found that DBH of nest box trees was an important variable associated with box use. Thorington and Thorington [31] examined *Microsciurus* and *Sciurillus*, and determined that increased length of the forelimbs may be an adaptation for vertical climbing and may allow for easier climbing on smaller diameter trees. Most likely, the same is true for *G. volans* which seemed to prefer trees with smaller DBHs.

Boxes in areas with a greater density of hard mast trees received greater overall use and greater use as defecatoria. This is reasonable since hard mast represents a large part of the squirrels' diet [11, 35]. This finding agrees with Stojeba [29] whose analysis included total food trees, "other" species of food trees, and total black oaks (*Quercus velutina*) as important factors explaining nest box use in Arkansas.

We found that nest boxes in areas with fewer snags were more likely to be used for nests by *G. volans*. Gilmore and Gates [8] also found that flying squirrel nests and feeding stations were more common in areas with fewer unbroken

snags with DBH > 15.2–22.9 cm. Because *G. volans* usually makes nests in woodpecker holes and other cavities [19] typically found in snags, it would be more likely to use a nest box in an area with few snags.

Glaucomys volans is known to use refugia (e.g., under root systems and trees) as secondary nests and retreats [19]. Gilmore and Gates [8] noted that *G. volans* was more likely to use a nest box in areas with fewer hard logs. We also found greater box use in areas with fewer logs. Nest boxes may represent supplemental refugia in areas with fewer subterranean retreats.

Previous studies have reported greater use by *G. volans* of areas with more ground cover [1, 25]. However, Boardman [2] found *G. volans* selected natural nest sites and nest boxes with less summer ground cover surrounding them. We also found reduced nest box use in areas with greater ground cover. Areas with greater ground cover generally have a less complex canopy structure [2] and thus may be less suitable as nesting sites for *G. volans*. It is not clear why our results and those of Boardman [2] differ from previous studies. Our study was designed to test landscape level differences, not microhabitat differences. All of our boxes were placed in habitat that was apparently suitable for squirrels. Had we sampled a greater variety of habitats, our results may have been different.

Variables identified by previous investigators as important to nest box use were also important in our study. These variables (DBH of the nest box tree, percentage of ground cover, number of logs, relative density of hard-mast trees) may have general biological significance for *G. volans*. However, a 3-year study of nest box use by Stone et al. [28] found most variables were not significant, consistently from year to year. Different results found by various studies on *G. volans* nest box use and habitat selection may be due to both temporal and geographic effects, as well as methodological differences. Alternatively, within relatively suitable habitat *G. volans* may be a habitat generalist.

4.2 Effects of Forest Fragment Size and Isolation

We found *Glaucomys volans* to be a nearly ubiquitous inhabitant of southern Illinois woodlots, occurring in 93 % of the forest fragments studied. Patch size did not appear to be a factor excluding squirrels from small sites. *Glaucomys volans* was present in 6 of 7 woodlots that were between 6 and 10 ha in area, leading us to conclude that area of habitat fragment (at least for fragments over 6 ha) may not be the most important factor in predicting squirrel occupancy in our woodlots.

However, *G. volans* was not present in the 2 most isolated woodlots. Populations in these woodlots may have become locally extinct, with recolonization unlikely due to distance from a potential source population. Fahrig and Merriam [5] designed a model of patch dynamics for white-footed mice (*Peromyscus leucopus*) in order to determine how population survival is affected by isolation. Their model predicted that mouse populations in isolated areas were more likely to have reduced growth rates and a greater probability of extinc-

tion. Field data for *P. leucopus* supported the model. The model may hold true for *G. volans* as well: we found squirrels were not present in woodlots that were isolated by more than 0.5 km.

Southern Illinois is primarily a forested landscape compared to northern and central Illinois. Maximum isolation between our woodlots was 643 m. In areas where distances between patches are relatively small (i.e. < 500 m), the probability of inter-patch dispersal by *G. volans* may be high. Unfortunately, this study was not designed to address dispersal. Landscape connectivity is often associated with persistence of species in fragmented landscapes [5]. Many species rely upon at least some level of connectivity (e.g. habitat corridors) in order for dispersal to take place. For species such as chipmunks (*Tamias striatus*) and white-footed mice, vegetated fencerows play an important role in connecting populations between woodlots [14, 34]. However, we were not able to document movement between patches, or use of habitat corridors.

Recently, Nupp and Swihart [20] reported *G. volans* in west-central Indiana were present only in continuous tracts of forest and woodlots > 6 ha which are near other woodlots. Our sites were larger than 6 ha in a primarily forested area (compared to west-central Indiana). While *G. volans* appears to be abundant in southern Illinois forest fragments, more work is needed on the effects of patch size and isolation; including determination of dispersal patterns, extent of habitat corridor use, and gene flow between woodlots. Additionally, long term studies on survival of flying squirrels in fragmented landscapes are needed.

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