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INFLUENCE OF PREDATORY BIRD SOUNDS ON THE EASTERN GREY SQUIRREL (*SCIURUS CAROLINENSIS*)

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ABSTRACT

The eastern gray squirrel (*Sciurus carolinensis*) are very common in many parts of the globe, especially North America. A few prior studies have examined how squirrels recognize predators. In this study, predatory bird calls from the red-tailed hawk (*Buteo jamaicensis*) and the Swainson's hawk (*Buteo swainsoni*) were played to squirrels. The northern cardinal (*Cardinalis cardinalis*) call was played as a control. The goal of this research was to determine if *S. carolinensis* are able to recognize native predatory birds from non-native predatory birds. After running chi-square tests and t-tests, it was determined that while the squirrels were able to differentiate between *B. jamaicensis* and *B. swainsoni*, they were not able to distinguish between *B. jamaicensis* and *S. carolinensis* significantly. They also froze for a significantly longer time when either hawk sound was played compared to the call from *C. cardinalis*. These results suggest that *S. carolinensis* may group hawk sounds together.

*Keywords: Eastern gray squirrel (*Sciurus carolinensis*), red-tailed hawk (*Buteo jamaicensis*), Swainson's hawk (*Buteo swainsoni*), northern cardinal (*Cardinalis cardinalis*), freeze, alarm sound*

INTRODUCTION

Eastern gray squirrels (*Sciurus carolinensis*) are widespread and found throughout North America and around the globe. They have a many type of predators, including red-tailed hawks (*Buteo jamaicensis*). They are necessary for the ecosystem as they help disperse plant seeds (Sawaya et al., 2018). This happens from squirrels burying nuts that they don't retrieve them later on.

Previous studies have shown that gray squirrels are able to distinguish predatory birds from nonpredatory birds (Lilly et al., 2019). Lilly and colleagues also found that abiotic noise did not affect the squirrel's behavior. A different study on squirrels and predatory bird sounds emphasized the different color morphs of squirrels, and they found that darker squirrels were more afraid of predatory noise and loud noises (a car alarm or buzzer) than a songbird call compared to lighter colored squirrels (Bohls and Koehnle, 2017). Bohls and colleagues also found that squirrels reacted differently when exposed to the predatory noise and loud noises, although that difference was not significant. Yet another study of squirrels and predatory birds found that the way the squirrels reacted was based on if they were familiar with the predator (Getschow et al., 2013). To test this, they looked at how the squirrel reacted to other animals (who also shared a common predator) and their alarm call.

It is necessary for survival that squirrels are able to quickly recognize and react to threats that exist in their environment. If new predators were introduced to an area, then those squirrels should learn over time to fear the new predator's call. At first one would not expect a new predatory call to trigger the same reaction that had already been developed to protect the squirrels from native predators. In this study, *S. carolinensis* response to bird predators and non-threatening native birds were evaluated. The hypothesis tested was that squirrels would be able to distinguish the calls of predatory native birds from

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those of predatory non-native birds. To test the hypothesis, wild squirrels found in residential areas were observed after being exposed to one of three bird calls: the northern cardinal (*Cardinalis cardinalis*), the red-tailed hawk (*Buteo jamaicensis*), and the Swainson's hawk (*Buteo swainsoni*). Afterwards, the data were analyzed.

METHODS AND MATERIALS

Field Site

Data was collected Montgomery County, Maryland, particularly North Bethesda and Bethesda, which are suburbs of Washington, DC. Residential neighborhoods were chosen as test sites due to the squirrels being more acclimated to humans. This allowed study team members to get close enough to subjects for accurate observation, and adequate sound volume. Data were collected between April 3, 2020 and April 25, 2020. All data were collected the four hours after sunrise or before sunset. This is due to the fact that most *S. carolinensis* are active during these times (Amspacheret al., 2019). Observers groups consisted of two people.

Data Collection

New squirrels were found and observed at a different field site each day of data collection. Once a squirrel was sampled, members of the study team walked at least one quarter of a mile away, to minimize the likelihood that the next subject would have heard the previous bird call. Each squirrel was exposed to one of three bird calls, which were played in sequence as new subjects were identified. The first bird call was from the non-native Swainson's hawk (*Buteo swainsoni*). The second bird call was from the control species, the northern cardinal (*Cardinalis cardinalis*). The third bird call was from the native red-tailed hawk (*Buteo jamaicensis*). The bird sounds were friendly calls made by adult birds (i.e., no hunting or territory calls were used). They were retrieved from the Cornell Lab of Ornithology, Macaulay Library (Ithaca, NY). The sound for the cardinal was recorded in Frederick, MD, the red-tailed hawk was recorded in Plumas, CA, and the Swainson's hawk came from Sutter, CA. An iPhone 7 Plus was used to play the recordings of the different birds, at full volume. The calls were repeated 5 times for a total of 15 seconds. The person playing the recording was blinded to the predator bird being played. Squirrels were not counted if they ran out of sight before our observation could be completed, or if a dog barked while they were being observed.

Data were recorded on the following variables: temperature (in Fahrenheit), weather conditions, time data collection started and ended, if squirrel froze or not (if so, the time they froze for), if the squirrel made an alarm noise, where the squirrel was at the start of the bird call, how the squirrel moved away (if it did), if the squirrel was eating while observed, the color of the squirrels, and if the squirrels demonstrated a tail flag. A tail flag is when a squirrel waves its tail in a particular whipping pattern as described by MacRae and Green (2014). The stopwatch on the iPhone was used to record the freeze time of the squirrels. The timer started when the squirrel froze and stopped when the squirrel moved. If any part of the squirrel moved (e.g. moving its mouth to eat) then the squirrel was not counted as frozen. The person playing the bird calls was not the same person timing how long the squirrels froze for. The start location of the squirrel was either on the ground, in a tree, or on a human structure (e.g., fences and roofs). The way squirrels moved away was recorded as running, hopping and walking, or not applicable (N/A).

Data analysis

SPSS was used to create a database and perform statistical analyses (IBM Corp., Version 25, Armonk, NY). First the data was cleaned. Then two versions of the variable (Freeze Time) were created (with and without zeroes). Later other variables for created, by combining the different ways squirrels fled and different combinations of our bird calls (such as merging all observations that were made during any hawk call). A Pearson's chi-square test was used to evaluate the categorical data. To compare the freeze time across the three groups, the data was skewed so a nonparametric test (Kruskal-Wallis test) was carried out. An ANOVA was also carried out for the normall distributed data. A Student's two-tailed t-test was conducted to analyze whether or not the freeze times between the hawks and cardinals were significantly different.

RESULTS

In this study, there were a total of 99 squirrels. Thirty-three squirrels were in each test group (*C. cardinalis*, *B. jamaicensis*, and *B. swainsoni*). During the 23-day data collection period, 33% squirrels were sampled in the morning and 67% squirrels were sampled in the afternoon. Overall, the temperatures during our observations ranged from 46 to 72 ºF, with the average being 56.73 ºF (s.d. 7.8). The squirrels were found in various locations, with 73%

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starting out on the ground, 25% starting in trees, and 2% starting on some human structure. There was no significant difference in the squirrels freezing based on where they started out. While the calls were finished playing, 79% of the squirrels fled, while 21% didn't flee. The squirrels that fled did so in a variety of ways; 54% of squirrels ran away, and 25% hopped or walked away. Some squirrels were eating or holding food when observed, and they made up 19% of our sample. There was no significant difference in the squirrels freezing or not when they were eating. As for coloration, 75% were gray squirrels and 10% were black squirrels. For 15% of our observations, color was not recorded. Only one squirrel made an alarm noise and two squirrels flagged their tails.

Several comparisons were tested between whether the squirrel froze or not and the different birds. In every instance, observed squirrels were most likely to freeze when the sound of the native predator was played and least likely to freeze when the songbird was played. This difference was not always statistically significant. When squirrel freeze was compared across all three bird sounds, differences were not found to be statistically significant (x^2=4.36, df=2, p=0.11). The proportion of squirrels that froze for any hawk was 73% the proportion that froze for froze for *C. cardinalis* was 55% (x^2=3.27, df 1, P=.07). When the squirrel freeze was compared between *B. swainsoni* and *C. cardinalis songs*, (67% that froze upon hearing the non-native predator call, and 55% froze in reaction to the native songbird's call (x^2= 1.02, df=1, p=.31). When squirrel freeze was compared between groups exposed to the calls of *B. swainsoni* and *B. jamaicensi*, 67% froze for the non-native hawk and 79% froze in response to the native hawk (x^2=1.2, df=1, p=.27. When squirrel freeze was compared between the *B. jamaicensis* and the *C. cardinalis calls*, they did differ significantly (see Table 1). More squirrels froze when exposed to the native hawk rather than the native songbird.

Table 1. Freeze comparison between B. jamaicensis and C. cardinalis. Frequencies of whether the squirrel froze or didn't freeze for the particular bird are shown. Percentages are shown in parenthesis. The chi-square value and a two-sided p-value is shown to the right.

Freeze	*B. jamaicensis* n (%)	*C. cardinalis* n (%)	Chi-Square	P-value (2 sided)
Yes	26 (78.8)	18 (54.5)	4.364	0.037
No	7 (21.2)	15 (45.5)		

The results from comparing the times of freezing yielded interesting results. This study's data set includes squirrels that did not freeze. Therefore, these results in two ways: one including all squirrels (with some freeze times being represented by zero values) and for the other analysis, we only included squirrels that did freeze.

The three variables were compared using ANOVA and Kruskal-Wallis. The ANOVA compared the freeze times without zeros replacing the times for the squirrels that didn't freeze. The Kruskal-Wallis compared the freeze times with zeros replacing the times for the squirrels that didn't freeze. The results from ANOVA indicate that there was no significant difference between the three groups (p=0.180). The results from Kruskal-Wallis indicate that there was a significant difference between the three groups (p=0.030) when the significance level was 0.05. To see which groups had the difference, two groups were compared to each other as opposed to three.

When squirrel freeze time was compared between the songbird and any predator, it was only significant when zeros were included (Table 2). When zeros were included, the F-test result for Levene's Test of Equal Variances was 7.417, with a p value of 0.008, indicating that equal variances could not be assumed. When zeros were not included, the F-test result was 3.462, with a p-value of 0.97, indicating that the assumption of the equal variances was not violated. These results influenced which t test results are reported in the following table.

Table 2. Squirrel freeze times observed: C. cardinalis calls compared to hawk calls. The third column shows the average. Columns five to 11 show the results from the t-test.

	Bird	Mean		t-test for Equality of Means						
				t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Zeros substituted for those who didn't freeze	Both Hawks	10.73	Equal variances not assumed	2.747	96.77	0.007	6.01	2.19	1.67	10.35
	Control (*C. cardinalis*)	4.72								
Zeros not substituted	Both Hawks	14.75		1.639	64.00	0.106	6.10	3.72	-1.34	13.54

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for those who didn't freeze	Control (*C. cardinalis*)	8.65	Equal variances assumed							

When squirrel freeze time was compared between *C. cardinalis* to the *B. swainsoni*, it was only significant when zeros were included (Table 3). When zeros were included, the F-test result for Levene's Test of Equal Variances was 4.726, with a p value of 0.033, indicating that equal variances could not be assumed. When zeros were not included, the F-test result was 1.853, with a p-value of 0.181, indicating that the assumption of the equal variances was not violated. These results influenced which t test results are reported in the following table.

Table 3. Squirrel freeze times observed: C. cardinalis calls compared to B. swainsoni calls. The third column shows the average. Columns five to 11 show the results from the t-test.

	Bird	Mean		t-test for Equality of Means						
				t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Zeros substituted for those who didn't freeze	Non-native hawk (*B. swainsoni*).	8.54	Equal variances not assumed	1.582	54.43	0.119	3.82	2.41	-1.02	8.65
	Control (*C. cardinalis*)	4.72								
Zeros not substituted for those who didn't freeze	Non-native hawk (*B. swainsoni*).	12.80	Equal variances assumed	1.224	38.00	0.229	4.15	3.39	-2.71	11.02
	Control (*C. cardinalis*)	8.65								

When squirrel freeze time was compared between *B. jamaicensis* to the *B. swainsoni*, it was not significant in either of the groups (Table 4). When zeros were included, the F-test result for Levene's Test of Equal Variances was 1.641, with a p value of 0.205, indicating that the assumption of the equal variances was not violated. When zeros were not included, the F-test result was 1.101, with a p-value of 0.300 indicating that the assumption of the equal variances was not violated. These results influenced which t test results are reported in the following table.

Table 4. Squirrel freeze times observed: B. jamaicensis calls compared to B. swainsoni calls. The third column shows the average. Columns five to 11 show the results from the t-test.

	Bird	Mean		t-test for Equality of Means						
				t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Zeros substituted for those who didn't freeze	Native hawk (*B. jamaicensis*)	8.5361	Equal variances assumed	-1.252	64	0.215	-4.38939	3.50484	-11.39	2.61
	Non-native hawk (*B. swainsoni*).	12.9255								
Zeros not substituted for those who didn't freeze	Native hawk (*B. jamaicensis*)	12.8041	Equal variances assumed	-0.831	46	0.410	-3.60129	4.33162	-12.32	5.12
	Non-native hawk (*B. swainsoni*).	16.4054								

When squirrel freeze time was compared between *C. cardinalis* to the *B. jamaicensis*, it was only significant when zeros were included (Table 5). When zeros were included, the F-test result for Levene's Test of Equal Variances was 8.990, with a p value of 0.004, indicating that equal variances could not be assumed. When zeros were not included, the F-test result was 4.321, with a p-value of 0.044, indicating that the assumption of the equal variances was not violated. These results influenced which t test results are reported in the following table.

Table 5. Squirrel freeze times observed: C. cardinalis calls compared to B. jamaicensis calls. The third column shows the average. Columns five to 11 show the results from the t-test.

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	Bird	Mean		t-test for Equality of Means						
				t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference	95% Confidence Interval of the Difference	
									Lower	Upper
Zeros substituted for those who didn't freeze	Native hawk (*B. jamaicensis*)	12.9255	Equal variances not assumed	2.615	44.71	0.012	8.21	3.14	1.88	14.53
	Control (*C. cardinalis*)	4.7200								
Zeros not substituted for those who didn't freeze	Native hawk (*B. jamaicensis*)	16.4054	Equal variances not assumed	2.016	38.58	0.051	7.75	3.85	-0.03	15.53
	Control (*C. cardinalis*)	8.6533								

DISCUSSION

The results partially support the hypothesis that squirrels are able to distinguish predatory native birds from predatory non-native birds. The gray squirrels were able to differentiate between *B. jamaicensis* and *B. swainsoni*, but not statistically significantly. This suggests some difference of recognition, in the direction of our hypothesis. With more statistical power, the differences observed may have been significant. They were also unable to distinguish *C. cardinalis* from *B. swainsoni*. These comparisons of insignificant results were apparent through both the chi-square test and the t-test.

The squirrels were able to differentiate *B. jamaicensis* from *C. cardinalis*. This was evident through the chi-square test as well as the t-test. When comparing the freeze times between the *B. jamaicensis* to the *C. cardinalis*, they were only significant when zeros were substituted in for the squirrels that didn't freeze. So, when only looking at the squirrels that froze, it wasn't statistically significant. This could be due to the data being more normal than when all of the zeros are added. They were also able to tell the difference between a call from *C. cardinalis* and a call from either of the two hawks, but the only way this was confirmed was through the t-test.

There were some limitations to this research, including the subjectivity involved in deciding if a squirrel froze, the fact that some squirrels could not be counted (due to environmental conditions) and confounding variables. To increase the reliability of our judgement about whether a squirrel froze, one person assigned that determination (who used consistent methods). Some squirrels froze after the recording stopped. Those squirrels didn't count, as only the squirrels that started freezing while hearing the sound was recorded. Some squirrels froze as they were approached, which might have influenced their reaction. In some cases, other people not part of this study would walk by them before the squirrels heard the recording. This could have potentially made the squirrels more attentive to their surroundings than normal. Another factor was that during many of our observations, songbirds could be heard in the background. In a different study by Getschow and colleagues, squirrels paid attention to songbird calls as one indicator of the safety of an area (2013). Future studies could try to take this into account and look at whether the time of year influences patterns of squirrel freezing. The data in the present study was collected during the spring, when there are more songbirds and more young squirrels that need protection.

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