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Title: Investigating anti-predator defenses in chickadees along the Rocky Mountain Front Range

DFO mission statement project goals: Nest predation significantly influences bird ecology and evolution. However, documenting real-time predation events and understanding behavioral responses to nest predators remain challenging. My proposed work aims to fill these knowledge gaps by studying anti-predator defenses, specifically hissing behavior, in black-capped chickadees and mountain chickadees across an elevational gradient in the Rocky Mountain Front Range. This project involves recording predation/intrusion events using video cameras and analyzing the behavioral responses of chickadees. The study's outcomes will contribute to our understanding of predator-prey interactions, with implications for avian conservation and for understanding how predator communities influence bird behavior. The video cameras used in this project will also be used for educational purposes via online streaming of active chickadee nests, which we piloted successfully in Summer 2023.

Introduction:
Predation events are challenging to document in real time, leaving gaps in our understanding of anti-predator defenses in birds. Hissing behavior, observed in cavity-nesting birds during nest intrusions, is a notable defensive response. While well-studied in European tit species, limited research exists on this behavior in North American chickadees. Hissing occurs when an individual bird is trapped within a cavity by an intruder in the entry. When a bird hisses, they exhale while slapping their wings against the cavity and protruding their head. When studying defensive hissing behavior, researchers typically create models of potential predators or use their hand to elicit a reaction. Because predation events on cavity nesting birds are difficult to document we lack an understanding of the sequence of events that occur when a predator intrudes in a chickadee nest cavity, or whether chickadees respond differentially to different types of predators (e.g., a mammal versus a snake). **Using video cameras, I will document and study interactions between predators (both experimental and natural) and chickadees to determine the sequence of events and outcomes of nest intrusions (e.g., for what types of predators does a chickadee hiss, remain silent, or attack).**

Location of study:

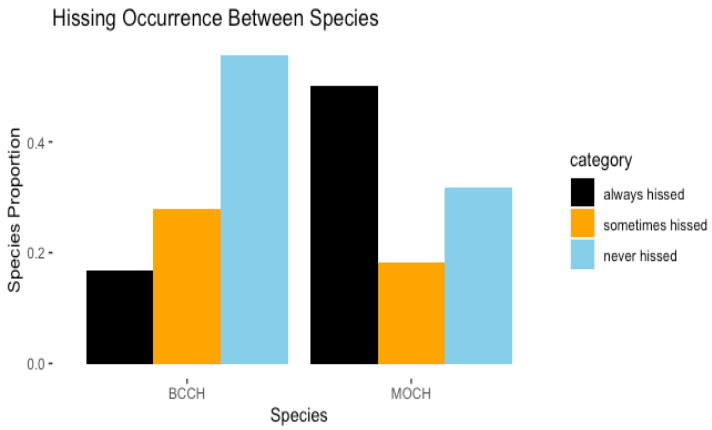

Figure 1 A histogram showing the proportion of Colorado's chickadee species that always hiss, sometimes hiss, and never hiss. (black-capped chickadee = 18, mountain chickadee = 22, n= 40)

The Boulder Chickadee Study (BCS) is a 416 nest box system that transects a 6500 foot elevational gradient of the Rocky Mountain Front Range in Boulder County, Colorado. Nest boxes are located in the city of Boulder (lowest elevation, private residents, CU Boulder campus), Sugarloaf Mountain (midelevation, private residents), Nederland and Ward, Colorado (highest elevation, CU Boulder Mountain Research Station, private residents). Studying hissing along an elevational gradient provides us with varying environmental contexts to analyze this anti-predator defense. Differential predator communities and variable predation risk along the gradient, as well as variation in temperature and metabolic stressors, may lead to the heterogeneous nature of hissing behavior that we documented in our population of chickadees in Boulder County in 2023 (Fig 1). In the Front Range of the Rocky Mountains mammalian species richness peaks at mid-elevations, however, as climate change progresses, birds at various elevations may begin to experience new predator species and be vulnerable to predation (e.g., fox squirrels are occurring at higher elevations than previously documented along our study transect, Karl Hendrickson, *pers comm*.). The elevation gradient across our study site will allow us to explore how patterns of hissing relate to elevation and predator species richness and understand how that may change with future predator range shifts.

Table 1. Project timeline	
Month in 2024	**Objective**
April	Install 13 live cameras (Boulder and Sugarloaf)
May-July	Record nest box interactions/predation events, install 7 cameras in Nederland and Ward BCS sites
August-October	Analyze video recordings
November-December	Visualize data for *The Lark Bunting* and present at December meeting

Methods:

Figure 2 A screenshot from the nest box camera recorded by a BCS participant. Using the app associated with the WIWA MW5, videos from live cameras can be downloaded and analyzed using general software video players.

Live camera installation –

Using the WIWA MW5 camera and application to view and record chickadee nests, I will study how intrusions of the cavity influence the behavior of birds inside the nest. I will install these video cameras inside chickadee nest boxes throughout the Boulder Chickadee Study (BCS). The protocol for installing video cameras has been developed and tested by a participant who livestreamed their chickadee nest box in summer 2023 (Figure 2). The WIWA MW5 camera records in 2K color video during the daylight and infrared when dark, as well as audio. Video recording can be controlled via the application system and will be set to triggered by motion or continuously (for outreach purposes.) Recordings will be stored on a MicroSD cards. I will set up and install cameras in nest boxes that we know are frequently used for nesting by chickadees based on our 5-year breeding database. Because the cameras record using Wi-Fi, nest boxes will be chosen based upon location within 50 feet of a Wi-Fi router (about 65% of our nest boxes have this capacity). When analyzing videos, I will identify intrusion events (experimental or natural) and record how the adult chickadee behaves during these intrusions using an established ethogram (Table 2). I will identify what predators are attacking nests, the behaviors of adult chickadees during and after an intrusion (i.e. whether or not a chickadee will fight back), the time-of-day natural intrusions most frequently occur, and the latency between an intrusion and hissing behavior.

Table 2. Ethogram for video analysis	
Bird head thrust	The head thrusts forward, the beginning of the hissing display
Wing slapping	Bird's wings quickly extend and retract –tail simultaneously expands and retracts
Hissing vocalization	Forced exhalation of bird in cavity
Intrusion	An animal besides the mate enters the cavity
Predator attack	An intruding animal attempts to harm the bird in cavity
Bird attack	Bird in the cavity attempts to harm the intruding animal
Hissing bout	Hissing vocalizations/behavior repeated until a pause of 5 seconds

I will deploy the 13 WIWA MW5 cameras strategically within nest boxes across the BCS to conduct comprehensive monitoring of avian nesting activities, moving the cameras to higher elevation nests as the breeding season progresses. By targeting nest boxes with a consistent history of chickadee nesting from 5 years of BCS data, I aim to maximize the

likelihood of documenting nest intrusions and predation events. Camera installation will reflect the dynamic nature of avian nesting behaviors, especially at different elevations. Starting in April, I will install 7 cameras in Boulder, CO. and 6 cameras in Sugarloaf Mountain residencies. By June, I will relocate these cameras to Nederland/Ward, CO. to continue monitoring and recording throughout the breeding season. To optimize data collection, I will use a flexible camera transfer strategy, facilitating the recording of a diverse range of nesting events across varied environmental conditions. Each camera will be left in place for three weeks, allowing for an extended monitoring period to collect a comprehensive dataset on both potential intrusions and successful nesting events.

Predator models –
Because capturing natural predation events will likely be infrequent, I will pair my observational study with an experimental study using 3D printed, realistic predator / nest invader models (e.g., a house wren, chipmunk, and a chickadee) . I will generate these predator models with a lab colleague who has experience creating 3D models of animals. I will use these predator / nest invader models to illicit defensive responses from nesting chickadees by inserting them into nest boxes and will analyze from the video recordings how anti-predator defenses in chickadees vary between natural predators/intruders and model predator intrusions, as well as between different types of experimental intrusions. To test for hissing, I will insert a predator model (house wren model, chipmunk model, and chickadee model) into the nest box entrance for 10 seconds to simulate an intrusion. I will then record the latency between model presentation and hissing vocalization as well as how many hisses a chickadee produces.

Outreach –
The deployment of video cameras in nest boxes throughout the BCS will not only allow us to understand hissing behavior and predator-prey interactions but will serve as an outreach resource for those with online access. During our last field season, participants who installed the WIWA MW5 also live streamed their nest (Figure 2.) I plan to livestream chickadee nests with high success rates based upon past BCS data as well as provide educational programs via Zoom and in local libraries focused on the natural history, nesting biology, and breeding biology of chickadees. Chickadees serve as great subjects for outreach because they are a common backyard bird with an easily identifiable plumage and vocalizations, making them relevant to people that reside in their range, especially Coloradoans.

Budget:

Budget Table	
Purchase	**Cost**
WIWA MW5 camera ($59 each) x13	$767
128 GBMicro SD card ($12 each) x13	$156
10ft USB cable (Type A to Micro) ($5 each) x13	$65
USB wall charger (3 for $12) x5	$60
100ft Outdoor extension cord ($25 each) x13	$325
3M residue-free Gaffers Outdoor Tape	$20

Quart size Ziploc Bag	$6
Wood	$20
5" x 7" poster board	$12
Push pins	$3
Local transportation along BCS	$566
Total Cost from DFO	$2,000

a. Letters of support / list of previous publications (optional)