

BARRED OWL PREDATION OF WESTERN SPOTTED SKUNKS

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ABSTRACT—The potential for trophic cascades triggered by recent range expansion of the Barred Owl (*Strix varia*) to the Pacific Northwest has caused concern among conservationists and managers. Barred Owl predation of small forest carnivores is a particular concern because these carnivores typically have low population growth rates relative to their body size owing to long interbirth intervals, which may result in sensitivity to increased mortality. The Western Spotted Skunk (*Spilogale gracilis*) is a common small carnivore in forests of the Pacific Northwest that may be a prey item for Barred Owls, and previous research suggests that avian predation can be a primary cause of mortality for congeneric spotted skunks (*Spilogale* spp.). We report a confirmed predation event of a Western Spotted Skunk by a Barred Owl and 3 additional predation events that we suspect were due to Barred Owls based on circumstantial evidence. During a Western Spotted Skunk research study, we recovered the radio collar of an adult male skunk from the top of a tall snag and located intestines and avian feces at the base of this snag. DNA metabarcoding revealed that the avian feces contained Western Spotted Skunk and Barred Owl DNA. Barred Owls are a novel predator of the Western Spotted Skunk in forests of the Pacific Northwest and may have both direct and indirect negative impacts on Western Spotted Skunk populations.

Key words: DNA metabarcoding, HJ Andrews Experimental Forest, mortality, Oregon, *Spilogale gracilis*, *Strix varia*, Willamette National Forest

The invasion of a novel predator can significantly destabilize native ecosystems by altering food-web dynamics through displacement of native predators and reduction of native prey populations. Invasive predators typically reach higher densities and have a broader diet than native predators and exert stronger top-down pressures on native prey species, which can result in local extinctions in extreme situations (Finke and Snyder 2010). In the Pacific Northwest, Barred Owls (*Strix varia*) have recently invaded most forested ecosystems and have displaced and contributed to the decline of the native Northern Spotted Owl (*Strix occidentalis caurina*) (Wiens and others 2014; Lesmeister and others 2018; Franklin and others 2021). Current

density estimates of Barred Owls in some areas are 3–8 times greater than historical populations of Northern Spotted Owls (Wiens and others 2021). Although the potential for a trophic cascade triggered by this Barred Owl expansion has been speculated (Holm and others 2016), empirical observations of these changes in the ecosystem and the consequences of this invasion have not been documented for many species besides the Northern Spotted Owl.

Given their broad diet (Livezey 2007; Wiens and others 2014), Barred Owls may have both direct mortality effects and non-lethal effects on potential prey populations (Lima 1998). This is particularly interesting with respect to small forest-adapted carnivores such as weasels (Mustelidae) and skunks (Mephitidae) that are vulnerable to avian predators such as owls (Lesmeister and others 2010; Linnell and others 2017). Even if small forest carnivores only comprise a limited portion of the avian predator's diet, small carnivore populations may be sensitive to increases in mortalities owing to low population density and the slow life history (such as low reproductive effort at 1 litter per year) of these small-bodied species (Mead 1968; King and others 2017). Moreover, non-lethal changes to their behavior can alter the caloric balance of small carnivores (Salo and others 2008), which have high metabolic rates and require calorie-rich diets to thermoregulate (King and Powell 2007; Zub and others 2009).

Nocturnal avian predators have been suspected of being one of the primary causes of mortality and drivers in space use of several small forest-adapted carnivores (Bull and Heater 2001; Lesmeister and others 2009, 2010, 2013; Linnell and others 2017), but determining the specific cause of a mortality can be difficult when only sparse forensic evidence is left behind at a mortality location. Even with radio tracking and prompt investigations once a mortality signal is emitted from a collar, causes of mortality are often attributed to a predator based on indirect evidence from clues left at radio-collar recovery locations. For example,

avian predators are implicated based on the presence of fresh avian excrement or suitable trees for perching and local information about predators in the area (Lesmeister and others 2010). Incorporating modern molecular approaches such as fecal DNA metabarcoding into assessments of cause-specific mortality can help identify the predator and verify that it consumed the focal species (Wengert and others 2013; Hopken and others 2016).

Here, we report confirmation of a predation event by a Barred Owl on a Western Spotted Skunk (*Spilogale gracilis*) that we were monitoring via radio-telemetry in western Oregon. We also report circumstantial evidence of 3 additional Western Spotted Skunk mortality events that were likely caused by Barred Owls. Despite often being reported as a common forest carnivore (Carey and Kershner 1996), the natural history and population trends of Western Spotted Skunks are poorly understood. In the eastern United States, the once common Eastern Spotted Skunk (*S. putorius*) has experienced dramatic range-wide declines, and a paucity of literature on the species hindered management and conservation assessments (US Fish and Wildlife Service 2012; Eastern Spotted Skunk Cooperative Study Group 2019). Although the main cause of Eastern Spotted Skunk population declines remains unknown, a number of possible mechanisms have been proposed including land-use change, wide-spread use of pesticides, historical overharvest, disease outbreak, and changes in predator communities (Gompper and Hackett 2005; Gompper 2017). Western Spotted Skunk populations also may be in decline (Cuarón and others 2016), but it remains unknown if any of these potential mechanisms are affecting this process. Owls are common predators of Eastern Spotted Skunks (Errington and others 1940; Lesmeister and others 2010; Hassler and others 2021), but only 1 study has confirmed Western Spotted Skunks in the diets of Barred Owls in their expanded range west of the Rocky Mountains (Wiens and others 2014). This raises questions about how the range expansion of Barred Owls into the Pacific Northwest may affect the population dynamics of the Western Spotted Skunk and trigger other trophic cascades (Livezey 2009; Holm and others 2016).

We radio-collared 25 Western Spotted Skunks in the Willamette National Forest around the HJ

Andrews Experimental Forest near Blue River, Oregon, between 2017 and 2019 (Fig. 1). All animal capture and handling was conducted in accordance with the guidelines set by the American Society of Mammalogists (Sikes and others 2016), and were approved by the USDA Forest Service Institutional Animal Care and Use Committee (IACUC #2016-015) and the Oregon Department of Fish and Wildlife. We captured Western Spotted Skunks as part of a larger study on the spatial ecology and foraging ecology of skunks in the temperate rainforest ecosystem in western Oregon. Once captured, we chemically immobilized skunks using an intramuscular injection of 15 mg/kg ketamine HCl (Zoetis Services LLC, Parsippany, NJ) and 40 mcg/kg dexmedetomidine HCl (Zoetis Services LLC, Parsippany, NJ). We fitted each skunk with 2 metal ear tags (Monel size 1; National Band and Tag Co., Newport, KY) and a very high frequency (VHF) radio-collar (M1545, 16 g; Advanced Telemetry Systems, Isanti, MN) with a 12-h motion-sensitive mortality sensor. Following capture, we monitored skunks on a nightly basis and investigated mortalities as soon as possible when a mortality signal was heard. Consequently, mortality signals were typically investigated within 24 h, allowing for accurate identification of cause of death based on evidence recovered at the site and from the carcass. Collar recovery, however, sometimes occurred much later owing to difficult retrieval locations and weather conditions.

On 7 November 2018 at 21:00, we detected a mortality signal from a radio-collar on an adult male Western Spotted Skunk that we captured on 18 September 2018 (mass = 445 g). Although we could not locate the collar initially, we conducted a thorough search of the area on 8 November 2018 and suspected the radio-collar was located at the top of a tall snag. On 10 November 2018, we conducted a search for the skunk's carcass with detection dogs, who located 3 pieces of evidence (1 set of intestines and 2 avian fecal samples: F38-164 and F38-165) 19.1 m away from the snag with the radio-collar (Fig. 2). We identified the animal intestines as those of a Western Spotted Skunk from morphology and similarity to intestines of necropsied Western Spotted Skunks. Samples were frozen until we processed them in the laboratory. We recovered the radio-collar from the top of the tall snag

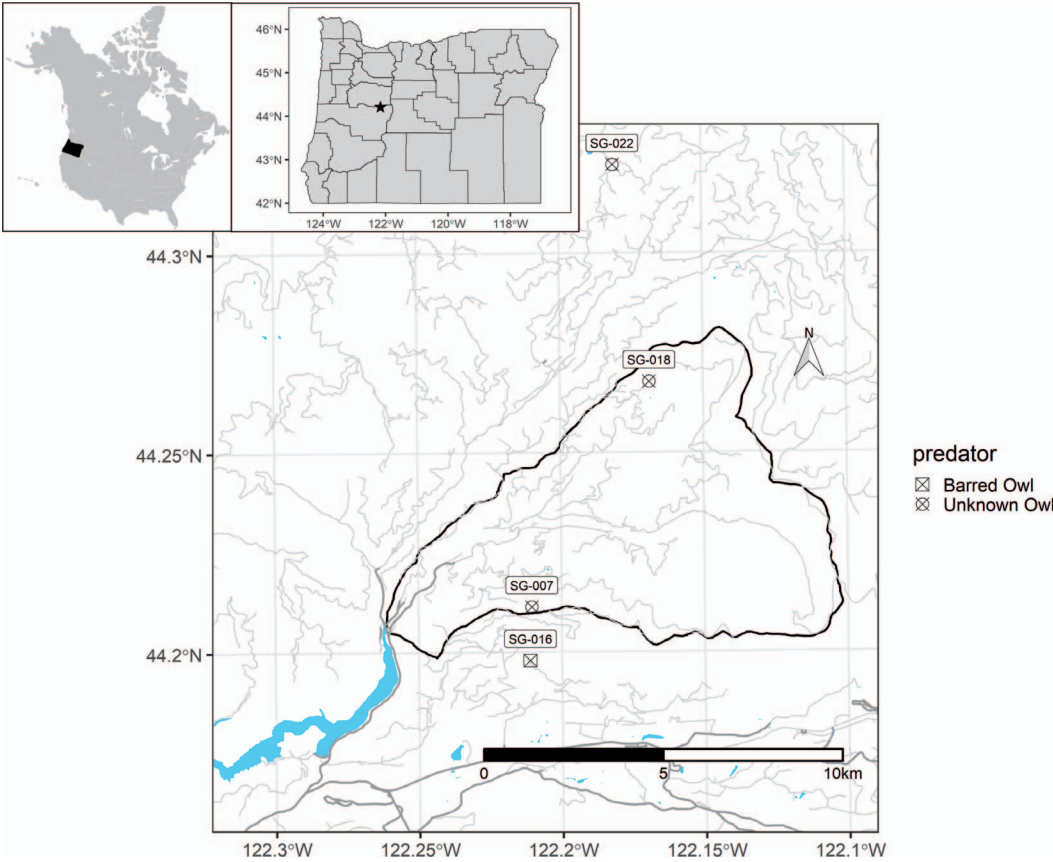


FIGURE 1. Study area located in the Willamette National Forest in western Oregon. Boundary of the HJ Andrews Experimental Forest shown in black lines. Roads are shown in grey lines, paved roads are shown in thicker grey lines. Radio-collar recovery locations of Western Spotted Skunk mortalities that were likely killed by owls shown with crosses. Confirmed Barred Owl predation represented by cross inside square.

using tree-climbing equipment on 5 December 2018 (Fig. 2).

We conducted DNA metabarcoding (Ji and others 2013) on the 2 fecal samples recovered by the detection dog teams (methods described in Eriksson and others 2019; Massey and others 2021). Briefly, we extracted DNA, amplified the 12S gene of the mitochondrial DNA with universal vertebrate primers using polymerase chain reaction (PCR) in 3 replicates, and sequenced amplicons on the Illumina HiSeq 3000 at the Center for Quantitative Life Sciences at Oregon State University. We taxonomically assigned unique sequences to species using a basic local alignment search tool (BLAST; www.ncbi.nlm.nih.gov/blast) against the 12S vertebrate sequences in GenBank and a custom 12S library of local vertebrates from the Pacific

Northwest (Eriksson and others 2019). Final species reported for each fecal sample represent consensus between at least 2 of the 3 PCR replicates, and sequence read numbers for each species exceeded 1% of the total number of reads for the sample. DNA metabarcoding revealed that the 2 fecal samples both contained DNA from Western Spotted Skunk and Barred Owl (Table 1). Feces also contained DNA of Humboldt's Flying Squirrel (*Glaucomys oregonensis*) (sample F38-164) and Townsend's Chipmunk (*Neotamias townsendii*) (sample F38-165).

In 2018 and 2019, we recovered 3 additional radio-collars from Western Spotted Skunks that we suspect were depredated by owls: a 365-g female detected on 2 January 2018 (SG-007), a 685-g male detected on 19 December 2018 (SG-022), and a 445-g female detected on 30 January

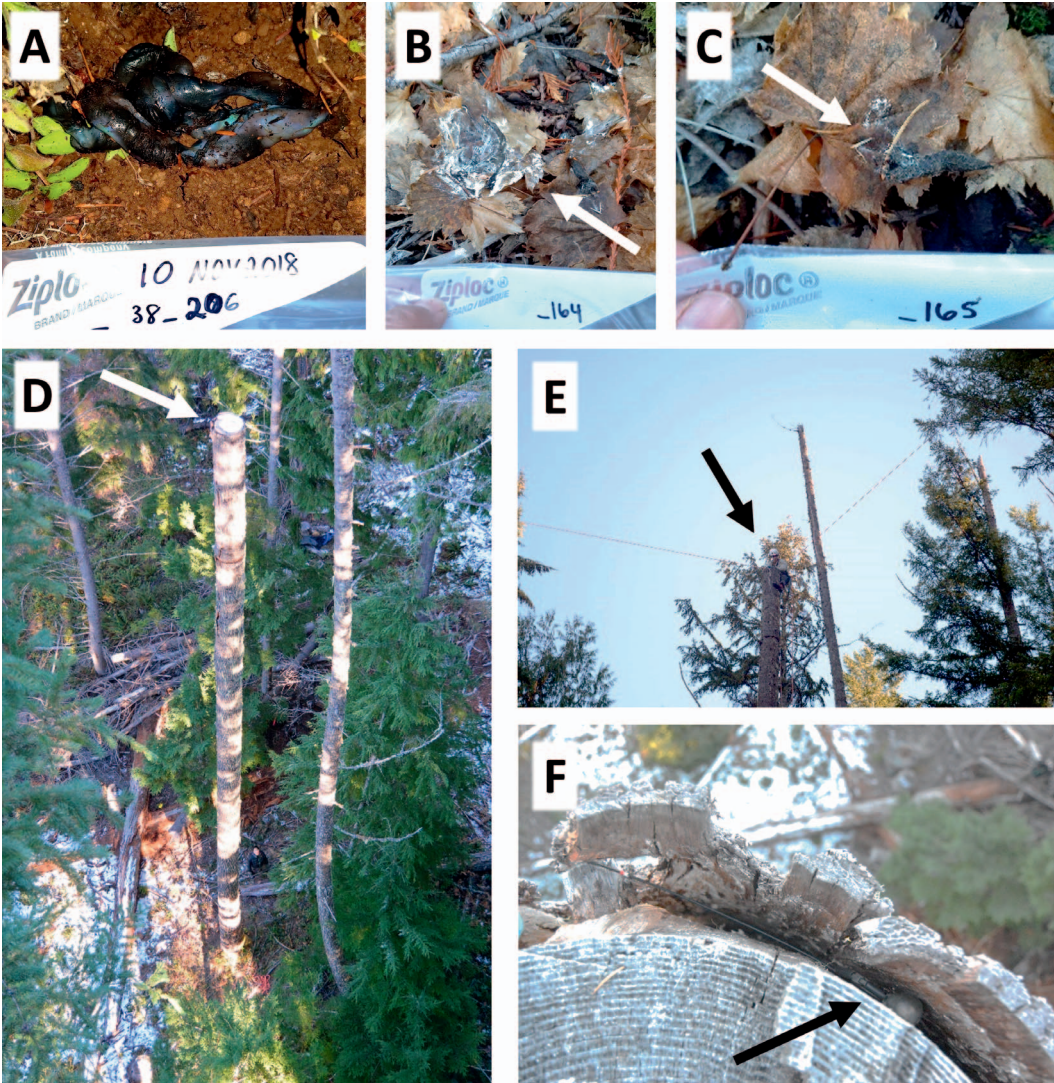


FIGURE 2. Evidence of Western Spotted Skunk predation. Evidence located by detection dogs near radio-collar recovery location on 10 November 2018: A) apparent Western Spotted Skunk intestines; B and C) avian predator feces. Recovery of Western Spotted Skunk radio-collar from snag on 05 December 2018: D) view of snag from above; E) view of collar retrieval from ground; and F) location of Western Spotted Skunk radio-collar, lodged inside bark on top of snag.

2019 (SG-018) (Fig. 1). We recovered these collars from the ground with little evidence. Collars from 2 of the skunks were found resting on the forest floor with no chew marks or carcass remnants in areas with large trees and snags. The collar for the 445-g female was recovered next to a skinned whole skull in an area with many large snags, also without tooth marks that are a typical sign of mammalian predation

events (Lesmeister and others 2010). In contrast to these 3 collar recovery sites, we documented 2 mammalian predation events during the study, which consisted of bloody kill sites and heavily disturbed forest floor. Parts of skunk carcasses were often found, and the braincases of the skulls were crushed, leaving only the mandible and a partial skull. All probable predations by avian predators occurred in the late autumn or

TABLE 1. Vertebrates identified through DNA metabarcoding of the 12S mitochondrial region from avian feces collected by detection dogs on 10 November 2018. Average number of reads is the mean number of identical sequences from the 3 replicates of each sample.

Sample	Species	Sequence	Average number of reads
F38-164	<i>Spilogale gracilis</i>	TTAGCCGTAAACACAAACAATTAGTATAACAAAACCTG TTCGCCAGAGAACTACTAGCAACAGCCTGAAACTC AAAGGACTTGGCGGTGCTTTATATCCCT	65,636
	<i>Glaucomys oregonensis</i>	TTAGCCCTAAACACAAATATTTAACTAACAAAAATATT CGCCAGAGTACTACTAGCAATTGCTTAAAACTCAAA GGACTTGGCGGTGCTTTATATCCCT	11,912
	<i>Strix varia</i>	CCGGCCCTAAATCCTGATATTCATCACCCTAGAACAT CCGCCAGGGTACTACGAGCACAAACGCTTAAACC CTAAGGACTTGGCGGTGCCCCAAACCCAC	2950
F38-165	<i>Strix varia</i>	CCGGCCCTAAATCCTGATATTCATCACCCTAGAACAT CCGCCAGGGTACTACGAGCACAAACGCTTAAACC CTAAGGACTTGGCGGTGCCCCAAACCCAC	8023
	<i>Neotamias townsendii</i>	TTAGCCCTAAACACAAATACTCAATAAACAAAGAGTAT TCGCCAGAGTACTACTAGCAATAGCCTAAAACCTCA AAGGACTTGGCGGTGCTTTACATCCCT	1161
	<i>Spilogale gracilis</i>	TTAGCCGTAAACACAAACAATTAGTATAACAAAACCTG TTCGCCAGAGAACTACTAGCAACAGCCTGAAACTC AAAGGACTTGGCGGTGCTTTATATCCCT	748

winter, when the skunks would have had reduced deciduous leaf cover. In combination with fewer prey items for owls in the winter, reduced cover may have increased the skunks’ vulnerability to avian predation. This increase in mortality rates during the winter also has been observed in other small carnivores such as the Short-Tailed Weasel (*Mustela erminea*) and the Long-Tailed Weasel (*Neogale frenata*) (Linnell and others 2017).

The skunks we tracked in this study were between 325 g and 855 g and almost exclusively nocturnal (M. Tosa, unpubl. camera trapping and radio-telemetry data), unlike sympatric forest carnivores, so we posited that any avian predators were likely medium- or large-bodied and nocturnally active. Potential nocturnal avian predators known to inhabit the HJ Andrews Experimental Forest include Barred Owls, Northern Spotted Owls, and Great Horned Owls (*Bubo virginianus*). Great Horned Owls are sparse in the study area, whereas the Barred Owl population is now the most abundant and commonly observed nocturnal avian predator. The Northern Spotted Owl populations have been declining at 5.6% annually from 1993 to 2018 and are now infrequently detected (Franklin and others 2021). The study area is also rarely visited by the Great Grey Owl (*Strix nebulosa*).

Although Great Horned Owls are known predators of Western Spotted Skunks (Errington and others 1940), Hassler and others (2021) genetically confirmed a Barred Owl depredation of a collared Eastern Spotted Skunk in West Virginia, and Wiens and others (2014) identified 10 Barred Owl pellets (out of 4306) collected between 2007 and 2009 from the Coast Range of Oregon that consisted of Western Spotted Skunk remains. In our study area, we found Barred Owl feathers with a strong skunk odor in June 2018, and there have been many accounts of Barred Owls covered in skunk odor from the Barred Owl removal study (2015–2019) across Washington, Oregon, and California (R. Baumbusch, Oregon State University, Corvallis, Oregon, pers. comm.). Although stomach content analyses of removed Barred Owls have not revealed bones or fur of Western Spotted Skunks (R. Baumbusch, pers. comm.), the stench covering these owls suggests that, at the very least, skunks continue to encounter this novel predator. Given the presence of Spotted Skunk remains in Barred Owl pellets, Barred Owls pose a legitimate threat, highlighting the potential for non-lethal effects on this small carnivore. In contrast, the Northern Spotted Owl has rarely been documented as consuming Western Spotted Skunks (1 out of 22,421 pellets; Forsman and others 2004) (Errington 1932; Livezey 2007).

Avian predation events may have significant impacts on Western Spotted Skunk population dynamics. Together, these 4 avian mortalities accounted for 36.3% of confirmed or suspected mortalities and 16% of all Western Spotted Skunks tracked during the study. Thus, the continued increase in Barred Owl abundance and densities raise concerns about the possible negative impacts of Barred Owls on Western Spotted Skunk populations and changes in the food web, vertebrate community, and ecosystem caused by the invasion of the Barred Owl in the Pacific Northwest (Holm and others 2016). This study raises additional concerns about the potential negative impact of Barred Owls on other forest carnivores, including the threatened Humboldt Marten (*Martes caurina humboldtensis*).

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