Light Reflectance Patterns of Decayed Wood with Implications for the Visual Ecology of Woodpeckers

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by
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The undersigned, appointed by the dean of the Graduate School, have examined the thesis entitled
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and hereby certify that, in their opinion, it is worthy of acceptance.

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ABSTRACT

Birds rely on eyesight for many aspects of their behavior and ecology, and a majority of diurnal species have thus evolved complex visual systems that include sensitivity to near ultraviolet (UV; 300-399 nm) wavelengths. The benefits of UV sensitivity to birds have been linked primarily to foraging and signaling. Behavioral studies of UV sensitivity have been conducted largely with passerine test subjects. Woodpeckers are a globally distributed avian subfamily (Picinae) that is ecologically and economically important. They are considered keystone taxa because the cavities that they excavate are utilized by dozens of other species of birds, mammals, and reptiles. Additionally, woodpeckers are responsible for millions of dollars in damage to anthropogenic structures annually. Despite their importance both as primary cavity excavators and nuisance animals, little work on their visual systems has been published. We developed a novel foraging-based behavioral assay designed to test UV sensitivity in woodpeckers, using the Pileated Woodpecker (*Dryocopus pileatus*) as a model organism. We acclimated 21 wild-caught *D. pileatus* to foraging for frozen mealworms within 1.2 m sections of peeled cedar (*Thuja spp.*) poles. We then tested the functional significance of multiple UV-reflective cues by placing frozen mealworms behind increased UV covers (0.07% MgCO$_3$, wt/wt), decreased UV covers (0.07% MgCO$_3$ + UV Killer$^\text{®}$ or UV Killer), or decayed red pine substrates within the same 1.2
m poles in independent experiments. We recorded four response variables for each experimental substrate presented to study subjects, and analyzed these using generalized linear mixed models. Behavioral responses were greater towards both increased UV and decreased UV substrates in three experiments. Study subjects therefore reliably attended to two distinct UV conditions of a substrate. When we analyzed results from cue-naïve subjects separately, those birds showed a preference towards decreased UV substrates, suggesting that woodpeckers may be pre-disposed to foraging from decreased UV substrates. In experiments with decayed wood, study subjects exhibited greater behavioral responses towards decayed wood even after UV reflectance was reduced. This indicates that increased UV reflectance of our decayed substrates was not essential for our subjects to identify decayed from not decayed substrates. Woodpeckers are known to transport spores of decay fungi on their bills, feathers, and feet. Some species exhibit preferences for placing cavities in trees infected by particular species of fungi, however, a mechanism that might allow such a specific level of detection is unknown. In another analysis, we produced aspen, oak, and pine substrates with multiple species of decay fungi and measured light reflectance spectra from the resulting decayed substrates. We then employed an established model of color discrimination to assess the spectral differences between these substrates as perceived by a hypothetical woodpecker. Two decay fungi known to be associated with woodpecker cavities produced substrates above the threshold of discrimination when compared with control and other decayed substrates. Most decayed substrate comparisons (12 of 14) were also above threshold, which indicates decayed wood appears visually different to woodpeckers based on the fungi responsible for the decay. Together, these studies provide evidence that the UV condition of substrates may be a useful foraging cue for woodpeckers, and that woodpecker-fungus mutualisms may be considerably more complex than previously thought.
THESIS FORMAT

The data chapters of this thesis were written as independent manuscripts prepared for submission to peer-reviewed journals. As a result, these chapters contain some redundant material and are followed by separate literature cited sections. Additionally, I use the plural pronoun “we” rather than “I”.
CHAPTER 1
INTRODUCTION

Woodpeckers (Piciformes: Picidae: Picinae, Leach 1820) are a group of primary cavity excavators that occur globally in forest and woodland habitats. They often are considered to be keystone taxa because the cavities that they excavate are in turn used by dozens of other species of birds, mammals, and reptiles (Aubry and Raley 2002), and thus woodpeckers participate in hierarchically-structured cavity web communities (Martin and Eadie 1999, Martin et al. 2004). In addition to such cavity web associations with other vertebrates, some woodpecker species are known to place cavities within trees that have been decayed by particular species of wood decay fungi, typically Basidiomycete heart rots (Jackson and Jackson 2004). While such associations with decay fungi have been documented for decades (Steirly 1957, Shigo and Kilham 1968, Conner et al. 1976, Winternitz and Cahn 1983, Parks et al. 1996, Huss et al. 2002, Farris et al. 2004), the mechanisms behind these associations are not well understood (Jackson and Jackson 2004).

Woodpecker associations with fungi are not limited to cavity sites. Some woodpeckers preferentially forage within decayed substrates, presumably because decayed substrates contain a higher arthropod biomass (Conner et al. 1994). Additionally, foraging-type damage patterns are often found on buildings in areas of wood trim or siding that are experiencing fungal decay, even though overt evidence of insect activity is absent from these areas (O’Daniels, pers. obs.). Recent studies have indicated that woodpeckers can transport fungal spores on their bills, feathers, and feet (Farris et al. 2004), and that cavities excavated by woodpeckers contain different fungal communities than cavities not excavated by woodpeckers (Jusino et al. 2015).
These findings suggest that at least some woodpecker-fungi relationships may be more complex
than previously thought.

Animal-pollinated plants have evolved mechanisms of attracting pollinators based on the
sensory capabilities of those pollinators including visual and olfactory cues or signals, and
gustatory rewards (Schiestl 2005). Woodpeckers transport fungal spores and could fill a role
analogous to pollinators, thus, woodpeckers and fungi may have evolved mutualistic
relationships similar to those between plants and pollinators. However, very little information on
any sensory capabilities of woodpeckers has been published. In order to investigate potential
mutualisms between fungi and woodpeckers, an understanding of woodpecker sensory
capabilities is imperative. Additionally, woodpecker excavating behaviors are responsible for
millions of dollars in damage to anthropogenic structures annually (Harness and Walters 2004),
and available control measures are often limited in effectiveness. Therefore, a better
understanding of woodpecker sensory ecology may also lead to better damage control measures.

In this thesis, I examined several aspects of woodpecker vision using the Pileated
Woodpecker (*Dryocopus pileatus*) as a model organism. In Chapter 2, I tested *D. pileatus’
visual sensitivity to ultraviolet (UV) wavelengths using a behavioral assay that I developed
specifically for this study. I was interested in UV sensitivity because avian repellents paired with
UV cues have shown promise in conditioning avoidance behaviors in other nuisance species
(Werner et al. 2012, 2014), and similar repellent-UV cue combinations could be useful in
woodpecker damage situations, provided woodpeckers are similarly sensitive to UV
wavelengths. I also examined the role of UV wavelengths in discriminating decayed from not
decayed (sound) pine substrates, using the same behavioral assay.
In Chapter 3, I employed the receptor noise-limited (RNL) model of color discrimination (Vorobyev and Osorio 1998) to assess whether wood substrates decayed by different fungi might produce discriminable substrates when viewed by woodpeckers. Mechanisms that allow woodpeckers to select substrates decayed by specific fungi are unknown (Jackson and Jackson 2004), and the objective of this study was to determine if visual cues or signals produced by decay fungi could be perceived by woodpeckers and, thus, play a role in foraging-site or cavity-site selection.

**Literature Cited**


CHAPTER 2
FUNCTIONAL VISUAL SENSITIVITY TO ULTRAVIOLET WAVELENGTHS IN PILEATED WOODPECKERS (DRYOCOPUS PILEATUS), AND ITS INFLUENCE ON FORAGING SUBSTRATE SELECTION.

Abstract

Most diurnal bird species are sensitive to near ultraviolet (UV) wavelengths, and benefits of UV sensitivity are linked primarily to foraging and signaling. Studies of avian visual ecology based on modeling theoretical perception of feathers and objects as birds see them have become prevalent in recent decades. Controlled behavioral studies investigating UV signals remain few, but in general focus on species that are important ecologically, economically, or recreationally. Woodpeckers participate in keystone processes through their excavating behaviors, and also create millions of dollars in damage to anthropogenic structures through those same behaviors. Despite woodpeckers’ importance as primary cavity excavators and nuisance animals, little work on their visual systems has been published. We developed a novel foraging-based behavioral assay designed to test UV sensitivity in woodpeckers, using the Pileated Woodpecker (Dryocopus pileatus) as a model organism. We acclimated 21 wild-caught woodpeckers to foraging for frozen mealworms within 1.2 m sections of peeled cedar (Thuja spp.) poles. We then tested the functional significance of multiple UV-reflective cues by placing frozen mealworms behind increased UV covers, decreased UV covers, or decayed red pine substrates within the same 1.2 m poles in separate experiments. Behavioral responses were greater towards both increased and decreased UV substrates in three experiments. Study subjects therefore reliably attended to two distinct UV conditions of a substrate. Cue-naïve subjects showed a preference towards decreased UV substrates, suggesting that woodpeckers may be pre-disposed to foraging from decreased UV substrates. Behavioral responses were greater towards decayed
pine substrates (increased UV) than sound pine substrates suggesting that decayed pine can be a useful foraging cue. When both decayed and sound pine substrates were treated with a liquid that decreases UV reflectance, behavioral responses were still greater towards decayed substrates. This result may suggest that UV signals were not important in discriminating decayed from sound pine. We combined our behavioral results with perceptual model data to estimate two parameters of *D. pileatus*’ visual system, SWS1 $\lambda_{\text{max}}$ and the LWS Weber fraction. Those results suggest that *D. pileatus* may possess a violet sensitive system comparable to the Great Spotted Woodpecker (*Dendrocopus major*), and that the noise in its visual neural mechanisms may be similar to that of domestic Red Junglefowl (*Gallus gallus*). Woodpecker responses to increased UV substrates appear to have been conditioned, suggesting that UV wavelengths can be a useful conditioning cue for *D. pileatus*. The finding that cue-naïve subjects preferred decreased UV substrates has implications for ecological interactions with fungi. Woodpeckers transport fungal spores, and communication methods analogous to those of plant-pollinator mutualisms (i.e. UV-absorbing patterns) may have evolved to support mutualistic relationships between woodpeckers and fungi.

**Introduction**

Birds rely on sight for many aspects of their life history and thus have evolved complex visual systems. Diurnal avifauna possess perhaps the most sophisticated color vision system among vertebrates (Goldsmith 1990), and most, if not all birds are sensitive to near ultraviolet (UV, 300-400 nm) wavelengths (Ödeen et al. 2011; Burns and Shultz 2012). Avian visual systems are typically categorized as either UV-sensitive (UVS) or violet-sensitive (VS), depending on the wavelength of maximum absorbance ($\lambda_{\text{max}}$) for the first of two short wave-sensitive visual pigments (SWS1; Vorobyev and Osorio 1998). The UVS system is
characterized by an SWS1 \( \lambda_{\text{max}} \) from 355nm – 373nm (Hart and Hunt 2007), and is found in higher Passerines, Paleognaths, Psittaciformes, and Laridae (Ödeen and Håstad 2003). All other avian taxa are presently presumed to possess a VS system (Ödeen et al. 2011), characterized by an SWS1 \( \lambda_{\text{max}} \) from 402nm – 426nm (Hart and Hunt 2007).

Measurements of \( \lambda_{\text{max}} \) values for visual pigments can be obtained by microspectrophotometry (MSP; Jane and Bowmaker 1988), or estimated from total DNA (Wilkie et al. 2000, Ödeen and Håstad 2003), giving a sense of a species’ visual system. These data can then be incorporated into models to estimate the saliency of wavelengths or the discriminability of colors, in species of interest (e.g. Vorobyev and Osorio 1998, Endler and Mielke 2005). Kemp et al. (2015) recognized such modeling as a valuable first step, but stressed the importance of behavioral studies in visual research. Behavioral studies may not categorize a subject’s visual system as UVS or VS, but they can demonstrate the ability to detect and respond to UV cues and thus provide potentially useful insights into visual systems.

The benefits of UV sensitivity to birds have been linked to orientation, foraging, and signaling (Bennett and Cuthill 1994). There is ample behavioral evidence indicating that UV wavelengths inform mate choice and foraging decisions for both UVS and VS species. Female preference for UV-reflective males over UV-blocked or UV-reduced males has been demonstrated in the European Starling (*Sturnus vulgaris*; Bennett et al. 1997), Zebra Finch (*Taeniopygia guttata*; Hunt et al. 1997), Bluethroat (*Luscinia svecica*; Johnsen et al. 1998), Blue Tit (*Parus caeruleus*; Hunt et al. 1999), Pied Flycatcher (*Ficedula hypoleuca*; Siitari et al. 2002), Eastern Bluebird (*Sialia sialis*; Siefferman and Hill 2003), Budgerigar (*Melopsittacus undulatus*; Zampiga et al. 2004), Wild Turkey (*Meleagris gallopavo*; Hill et al. 2005), and King Penguin (*Aptenodytes patagonicus*; Jouventin 2008). As a plumage or integument characteristic, UV
reflectance may signal male fitness in terms of resource acquisition or overall health (Zampiga et al. 2004; Siefferman and Hill 2005; Hill et al. 2005; Jouventin 2008).

Behavioral studies also have demonstrated the potential importance of UV wavelengths to foraging in frugivores, insectivores, and raptors. For example, removal of the waxy cuticle layer of the bilberry (*Vaccinium mytillus*), which reduces UV reflectiveness, reduces preference for bilberries by Redwings (*Turdus iliacus*; Siitari et al. 1999). In foraging experiments with two cryptically-colored caterpillars, captive wild-caught Blue Tits located their first prey items faster when UV light was included compared to when UV light was excluded (Church et al. 1998).

Behavioral studies of UV sensitivity have been conducted largely with Passeriform study subjects, with one or two species from the Anseriformes (Parrish et al. 1981), Columbiformes (Wright 1972), Coraciiformes (Parrish et al. 1984), Falconiformes (Viitala et al. 1995), Galliformes (Maddocks et al. 2001), and Psittaciformes (Zampiga et al. 2004) also represented. Species evaluated thus far primarily represent those of ecologic, economic, or recreational interest. Woodpeckers (Piciformes: Picidae: Picinae, Leach 1820) are a globally distributed, ecologically and economically important avian group, yet little work on their visual systems has been published. To our knowledge the only woodpecker species to have had its visual system categorized is the Great Spotted Woodpecker (*Dendrocopos major*; GSWO), which has an estimated SWS1 $\lambda_{\text{max}}$ of 405 nm (Ödeen and Håstad 2003), suggesting a VS system.

As primary cavity excavators, woodpeckers are a foundational link to hierarchical nest web communities (Martin and Eadie 1999) because the cavities they create are used by dozens of other vertebrate species, and represent resources from which those secondary users would otherwise be excluded (Aubry and Raley 2002, Steeger and Dulisse 2002, Arnett et al. 2010).
Woodpeckers are known to carry fungal spores on their bills and feathers, and may facilitate fungal colonization of wood substrates as well (Farris et al. 2004). Through their excavating behaviors, woodpeckers often fill the role of keystone species (Bednarz et al. 2004) by creating cavities and by contributing to the decomposition of woody debris.

When directed towards anthropogenic structures, woodpecker excavating behaviors can cause structural damage and impose significant financial costs. Worldwide costs attributed to repair and replacement of structures damaged by woodpeckers total at least in the millions of dollars (US) annually (Harness and Walters 2004). The Pileated Woodpecker (*Dryocopus pileatus*; PIWO) is the largest extant woodpecker in North America, with a range that spans much of the north and east of the continent, and often is identified as the cause of the most severe damage to wooden utility infrastructure (Dennis 1964, Tupper et al. 2011). Avian repellents paired with UV-reflecting and UV-absorbing compounds have shown promise in reducing avian crop damage (Werner et al. 2012, Werner et al. 2014). One such repellent, 9,10 anthraquinone, is UV-absorbing and has shown effectiveness at deterring woodpecker damage in a limited trial (Cummings et al. 2004). However, whether *D. pileatus*, or any other woodpecker species, is sensitive to UV wavelengths is unknown. A better understanding of the cues woodpeckers initially use to locate potential excavation sites may lead more effective control measures.

Conner et al. (1994) described the preference of several species of woodpeckers, including *D. pileatus*, to forage by excavation within wood substrates that were softer (decayed) than adjacent unselected substrates, and these selected substrates contained a higher arthropod biomass than the unselected substrates. It is not known how woodpeckers identify decayed substrates or select specific sites on a given substrate for foraging (Jackson and Jackson 2004), but they may use cues outside of human perception (Zahner et al. 2012). One potential cue is
UV reflectance, and wood that has been decayed by fungi can exhibit a UV reflectance pattern that differs from uninfected wood (Klapstein et al. 1989). We hypothesized that woodpeckers possess UV sensitivity, and that they use UV reflectance characteristics of wood to identify decayed substrates for foraging and possibly cavity excavation.

Herein, we present a study that attempts to address these hypotheses. We developed a novel foraging-based behavioral assay designed to determine whether altering the UV reflectance of a wood substrate influences selection of foraging substrates by woodpeckers. We conducted five two-choice experiments with captive, wild-caught *D. pileatus*. We first artificially increased (Experiment 1) and decreased (Experiments 2 and 3) the UV reflectance of experimental substrates relative to control substrates. We predicted that study subjects would respond differently to control and UV-altered treatment substrates. We also tested whether treating increased UV substrates with a UV-decreasing substance affected study subjects’ ability to discriminate these treatments from unaltered control substrates (Experiment 2). Finally, we tested whether decayed wood is a useful foraging cue for woodpeckers (however, not specifically a visual cue), and if decreasing UV reflectance of both decayed and control substrates diminishes woodpeckers’ ability to discriminate between these substrates (Experiments 4 and 5). We predicted that study subjects would respond differently to decayed and control substrates, and that decreasing the UV reflectance of decayed and control substrates would have little to no effect on discrimination. This last prediction was based on Experiments 1 and 2 (occurred prior to Experiment 5), on work with other avian species (Lytinnen et al. 2001, Werner et al. 2012), and in part on our inability to completely control potential confounding influences (see 5.2).

1. General Methods

1.1 Experimental Design
1.1.1 Test Apparatus

One test pole was prepared for each test subject, and the same test pole was presented to each subject throughout the duration of experiments in which that subject participated. Human sebaceous oils are UV-absorbing, therefore poles were handled with gloves for the duration of all experiments. The test pole was a 20 cm diameter x 120 cm section of cedar (*Thuja* sp.). Each test pole contained a total of 12 pre-drilled holes (2.2 cm diameter x ~ 7.5 cm depth) at 45° from vertical. Four holes were evenly spaced around the test pole at each of three heights (30, 60, 90 cm), and each set of holes was offset from the others so they did not align vertically. At each height, holes were randomly assigned to either treatment or control group (two each per height), and assignments were re-randomized each trial. Treatment holes received ~ 3 g frozen mealworms and were sealed with a treatment substrate. Control holes received ~ 3 g of mealworm bedding material (sawdust, frass, etc.) to control for olfactory and resonance cues, and were sealed with a control substrate. For each of Experiments 1-5, experimental substrates were randomly assigned a position in a test pole after the appropriate preparation (see below).

1.1.2 Corks

Since woodpeckers may preferentially forage from softer wood substrates, we used natural corks (Size #10, Carolina Biological, Burlington, NC, USA) as a surrogate for soft wood. All corks for Experiments 1 & 2 were soaked in demineralized water for 24 hours, after which each was randomly assigned to either the treatment or control group. We had previously determined that soaking was necessary to ensure retention of the increased UV treatment by the corks. Treatment corks were further prepared as either increased UV (Experiment 1) or decreased UV (Experiments 2 & 3), while control corks received no additional preparation. As
with the test poles, all corks were handled only with nitrile gloves for the duration of experiments.

Increased UV treatment corks were created by submersion in a warmed 0.07% magnesium carbonate (MgCO$_3$; Sigma-Aldrich, St. Louis, MO, USA) suspension (wt/wt, Werner et al. 2012) for 20 sec, with constant agitation by magnetic stir bar. MgCO$_3$ is a naturally occurring compound that is odorless and tasteless to humans and exhibits its peak reflectance below 300 nm. The suspension was warmed on the lowest setting of a laboratory hot plate for at least 5 min prior to application with a stirring speed of 6. Treatment and control corks were then dried for a minimum of 2 hr at 200$^\circ$C in a laboratory drying oven, or a minimum of 24 hr under a fume hood at room temperature. At the 0.07% MgCO$_3$ concentration, ~70% of the treated corks exhibited no human visible (white) residue. Any corks exhibiting a human visible residue were excluded from further use, and for consistency the same person (SO) prepared and selected all of the corks. Decreased UV treatment corks (Experiments 2 and 3) were prepared by submerging corks in a bath of UV Killer® (Atsko, Inc., Orangeburg, SC, USA; henceforth, UVK) for 20 sec with constant agitation. Corks for Experiment 2 were pre-treated with MgCO3, but not for Experiment 3. Each cork was prepared at least 24 hr in advance of the trial in which it was used.

Prior to each trial, the surface reflectance of each treatment and control cork was measured using an Ocean Optics USB2000+ microspectrophotometer calibrated for 200-850 nm with a QR400-7-UV-BX reflectance probe and a PX-2 pulsed xenon light source (Ocean Optics; Dunedin, FL, USA). The probe was calibrated against white (WS-1 Spectralon) and black (the dark) standards, and was re-calibrated after each group of six corks. Measurements were taken haphazardly at 3 points on the widest end of the cork. We used a modified black rubber stopper
to hold the probe at a fixed distance (5 mm) and angle (90°), and to eliminate ambient light. Due to the rugose nature of the cork surface, we only collected measurements from the flattest, smoothest portions of the cork surface to ensure a constant, fixed distance from the probe.

1.1.3 Wood Wafers atop Corks

For Experiments 4 & 5, we inoculated red pine (*Pinus resinosa*) wafers with *Porodaedalea* (syn. *Phellinus*) *pini*, a wood decay fungus that is associated with Red-cockaded Woodpecker (*Picoides borealis*) cavity sites (Steirly 1957, Jackson 1977, Conner and Locke 1982). Wafers were 20 x 20 x 3 mm, and had been cut perpendicular to the transverse plane (across the grain) to facilitate colonization of the entire wafer. All wafers were autoclaved in a 2% malt extract (2M) broth and placed in petri dishes (plates) on 2M agar, 7 wafers per plate. Plates sat for 7 days in the dark at indoor, ambient conditions to monitor for contamination. We then assigned plates to either control or treatment conditions. All treatment plates were inoculated on the same day by agar block transfer from pure cultures of *P. pini*, while control plates remained unmanipulated. All plates were then returned to dark storage, and monitored periodically for contamination.

After ~5 months, treatment wafers were extracted from plates and fungal mycelia were manually scraped from wafer surfaces. Control wafers also were manually scraped such that any tool marks left on wafers would be similar between treatments and controls. Scraped wafers were placed on paper towels and allowed to desiccate for at least 72 hr before further use. Upon the dryness of wafers, each was attached to the 25 mm side of a Size #10 laboratory-grade cork (Carolina Biological) with WeldBond adhesive (F.T. Ross & Sons, Ltd., Markham, Ontario, CAN). We maintained the same orientation of wafers throughout this process, such that the
surface initially in contact with agar (bottom) was the bottom surface while drying and was the surface adhered to corks.

Surface reflectance of each control and treatment wafer was measured in the manner previously described, except that reflectance was collected at six points on the top surface. Additionally, wafer reflectance was collected prior to the first trial in which it was used, but not before each trial. Undamaged wafers were re-used because we did not have enough control or treatment wafers to conduct 10 trials with each test subject. After each trial, any wafers with visible damage were excluded from further use.

1.2 Training

For each of Experiments 1-5, all study subjects were trained to forage for mealworms (*Tenebrio* spp. larvae) within a test pole prior to participating in experiments. Each subject participated in two training periods each day from ~ 0800 – 0900 MDT and from ~1600 – 1700 MDT for three consecutive days (six sessions), immediately prior to the experiments in which they participated. All subjects were food-deprived prior to the start of each training session to ensure adequate food motivation (Koenig 1991). Maintenance diets were removed at ~ 1900 MDT the evening before each morning session and ~ 1300 MDT prior to each afternoon session. Additionally, the daily ration of mealworms was removed from the maintenance diet, and mealworms were offered only during subsequent training or testing periods. Water was provided *ad libitum*. See Appendix 1 for details regarding the capture, care, and use of study subjects and description of the testing facilities.

Each training session consisted of preparing the test poles by placing ~ 1.5 g frozen mealworms in each of the 12 holes with no obstructions. We accurately weighed out 50% of the daily ration (18.0 +/- 0.1 g (2014); 20.0 +/- 0.1 g (2015)) for each bird prior to the start of each
session, but no effort was made to ensure that each hole received equal amounts. The enrichment poles were then removed from the aviaries and replaced with the test poles. Each subject was allowed to explore and forage from the test pole for ~ 60 min during each training session, and each session was recorded with digital video. At the end of each training session, we removed the test poles and replaced the enrichment poles. To verify that establishing conditions (i.e. mealworm consumption) were met, we collected and weighed any remaining mealworms from each test pole at the conclusion of each training session.

1.3 Statistical Analyses

1.3.1 Behavioral Analyses

For each of Experiments 1-5, we examined the fate of each substrate (i.e. cork or wafer) presented to study subjects. The dependent measures for detection of UV cues paired with food rewards included contact with corks (yes/no), removal of corks from test poles (yes/no), cork handling time (CHT), and the order in which corks were removed. Data for all response variables were collected after reviewing video recordings of trials. Values of CHT were calculated by summing the amount of time (whole seconds) that a subject physically interacted with a substrate prior to removing it from the test pole. Values of CHT that were two or more orders of magnitude greater than the median value were considered outliers and excluded from analyses. We created generalized linear mixed models (GLMMs) using the lme4 package (Bates et al. 2015) within the R environment (version 3.2.3, R Core Team 2015) to analyze each dependent variable, and we considered differences significant at $\alpha < 0.05$. The GLMMs included cork type (treatment/control) as a fixed variable and random variables for both subject ID and trial number to account for non-independence due to repeated measures. We assumed a binomial distribution for contact and removal variables, and a Poisson distribution for handling time and
ranked variables. We used logistic regression on the binomial response variables to generate probability estimates for each substrate condition. Denominator degrees of freedom were calculated with the Satterthwaite method in the lme4 package.

For most variables analyzed, the null hypothesis was no difference between control and treatment values and the alternative hypothesis was treatment values greater than control values. For the order removed variable, we expected treatment corks would be removed earlier than control corks resulting in an alternative hypothesis of treatment value less than control value. Any woodpecker-substrate interactions that occurred after 60 minutes were excluded from analysis.

1.3.2 Spectral Analyses

Mean reflectance spectra of each substrate were analyzed for their departure from the overall mean reflectance spectrum of the opposite substrate condition, over 300-700 nm (i.e. each treatment substrate vs. mean control substrate). We compared departures from means separately for 300-400 nm (UV) and 400-700 nm (human visible) using general linear models and linear mixed effects models, and we considered differences significant at \( \alpha < 0.05 \). Analyses were conducted in R using the packages lme4, lsmeans (Lenth and Hervae 2015), and multcompView (Graves et al. 2015).

When unpredicted differences were detected by statistical models, we employed the receptor noise-limited (RNL) color discrimination model of Vorobyev and Osorio (1998) to determine whether those differences might translate into perceptual differences for the study subjects. The RNL model has been widely used to estimate avian color discrimination capabilities (e.g. Eaton and Lanyon 2003, Eaton 2005, Igic et al. 2012, Olsson et al. 2015). The model calculates the difference between points (\( \Delta S \)) within a theoretical color space as defined
by the quantum catch of each receptor type within the retina of the species of interest. Values of 
\( \Delta S \geq 1.0 \) are considered to be distinguishable. We ran the model with differing values of SWS1 
\( \lambda_{\text{max}} \) and long-wave sensitive (LWS) Weber fraction to cover the range of relevant published 
values. See Appendix 2 for details on the RNL model and parameters used in this study.

2. Experiment one: increased UV cork treatments, MgCO\(_3\)

2.1 Methods

To test whether increased UV substrates are a useful foraging cue for PIWO, we 
employed a two-choice behavioral assay using six adult (5 M, 1 F), trained, cue-naïve PIWO. If 
so, then behavioral responses for increased UV corks (N=360) would be greater than those for 
control corks (N=360). Each subject participated in 10 trials (2 per day) from 31 May – 4 June 
2014, following the previously described methods, schedule of food-deprivation, and trial times 
(General Methods 1.2). Test poles contained increased UV and control corks.

To reduce the likelihood that MgCO\(_3\) transferred from corks to poles would confuse birds 
during subsequent trials, we rinsed each hole and the immediate surrounding surface area with 
demineralized water. Poles were left inverted until the next trial to facilitate drying. Subject 108 
sustained extensive abrasions on the tongue between Trials 9 and 10, and was placed under 
veterinary care, thus data from Trial 10 for this study subject were excluded from analyses.

2.2 Results and discussion

Study subjects were more likely to contact treatment corks than control corks (T = 86.0%, 
C = 76.2%; \( F_{1,691.85} = 11.1, P = 0.0008; \) Fig. 2.1A), and more likely to remove treatment corks (T 
= 83.0%, C = 69.8%; \( F_{1,691.83} = 16.6, P < 0.0001; \) Fig 2.1F). With one outlier removed, CHT of 
treatment corks was greater than control corks (T = 13.1, C = 9.5; \( F_{1,542.79} = 27.1, P < 0.0001; \) 
Fig. 2.1K). There was no difference in the order that corks were removed (\( P = 0.111 \)). These
behavioral responses were aligned with our predictions, with the exception of the order removed response.

To test whether cue-naïve subjects exhibited a predisposition to forage at increased UV substrates, we analyzed data from Trial 1 separately. Probability estimates for cork contact ($P = 0.25$), corks removed ($P = 0.75$), and the order removed ($P = 0.92$) were not different. However, after removing one outlier, the estimate for CHT was greater for control corks than treatment corks ($T = 37.8, C = 44.6; F_{1, 53.121}, P < 0.0001; \text{Fig. 2.2G}$).

Spectrally, control and treatment corks from Experiment 1 were different in the UV range ($Z\text{ ratio} = 17.58, P < 0.0001$) and were not different in the human visible range ($Z\text{ ratio} = 0.16, P = 0.9999; \text{Fig. 2.3A}$). Based on preliminary work with these substrates, these were the expected spectral differences.

We measured the reflectance of each cork presented to study subjects, and thereby ensured that differences between control and treatment corks occurred in the UV range (300-400 nm) rather than in the human visible range (400-700 nm). Since MgCO$_3$ is considered odorless and tasteless (to humans), and because we controlled for mealworm odor and differences in resonance between control and treatment holes, we conclude that these subjects responded to the spectral differences between control and treatment corks as they perceived them. After one trial, study subjects did not demonstrate a preference toward increased UV treatments, and in the case of handling time, the preference was toward control substrates. However, after a total of 10 trials, subjects’ responses aligned with our $a\ priori$ predictions. This suggests that the observed responses toward increased UV corks were conditioned, and that a predisposition toward decreased UV substrates may in fact be present in PIWO.

3. Experiment two: decreased UV cork treatments, MgCO$_3$ + UV Killer
3.1 Methods

To test whether decreasing the UV reflectance of increased UV substrates negatively impacts the ability to locate food in previously conditioned PIWOs, we employed another two-choice behavioral assay. If so, then behavioral responses should be similar (i.e. not different) between control and treatment substrates. We used the same subjects from Experiment 1 (4 M, 1 F), except for study subject 108 which had been removed from trial participation. Test poles contained decreased UV (N=300) and control (N=300) corks. Treatment corks were prepared by submerging MgCO$_3$-treated corks in a bath of UVK for 20 sec. All other parameters were as previously described. Trials took place from 8 June – 12 June 2014.

3.2 Results and discussion

Study subjects were more likely to contact treatment corks than control corks (T = 94.1%, C = 88.7%; $F_{1, 585} = 7.982, P = 0.004$; Fig. 2.1B) and were more likely to remove treatment corks (T = 93.9%, C = 88.0%; $F_{1, 585} = 9.005, P = 0.003$; Fig. 2.1G). Handling time was greater for treatment corks than control corks (T = 8.8, C = 7.3; $F_{1, 493.64} = 32.108, P < 0.0001$; Fig. 2.1L). There was no difference in the order that corks were removed ($P = 0.45$). These behavioral responses were aligned with our predictions (see Introduction), with the exception of the order removed response.

Spectrally, control and treatment corks from Experiment 2 were different over 300-400nm (Z ratio = -23.55, $P < 0.0001$) and over 400-700 nm (Z ratio = -3.72, $P = 0.0011$; Fig. 2.3B). We anticipated there would be no difference over 400-700 nm. Since the difference from 400-700 nm was unpredicted, we used the RNL model to assess whether this difference might have been perceptible to the study subjects.
Comparing the mean treatment and control reflectance spectra produced a ΔS value of 1.3 (above threshold; Table 2.1) under the GSWO model (LWS Weber fraction = 0.05; see Appendix 2 for details). When we set the UV portion of these mean reflectance spectra equal to each other in the model, so that the only differences were in the 400-700nm range, ΔS dropped to 0.3 (below threshold; Table 2.1) suggesting that any differences over that range were not perceptible.

The RNL model results enabled us to estimate two aspects of Pileated Woodpecker visual systems, SWS1 λ_{\text{max}} and the LWS Weber fraction. Threshold values of ΔS were produced only when the GSWO model LWS Weber fraction was \leq 0.06 (ΔS = 1.0), and no threshold values were produced under the average VS model (Table 2.1). Since our subjects did behaviorally discriminate between control and treatment substrates, these perceptual model results suggest that the SWS1 λ_{\text{max}} of the Pileated Woodpecker is very similar, or identical, to that of the Great Spotted Woodpecker. This finding is consistent with the conservative nature of avian visual systems (Kemp et al. 2015).

In this experiment, we aimed to test whether decreasing the UV reflectance of MgCO₃-treated substrates would impair our subjects’ ability to locate food items. We selected the UVK product for the reduction of UV wavelengths because we believed it would produce corks with reflectance spectra similar to controls, and because there was no human visible residue on the corks when dried. The UVK treatment did not result in spectrally similar control and treatment corks (Fig. 2.3B). Rather, the treatment corks exhibited a depression in UV reflectance compared to the controls. The performance of the study subjects suggests they were able to distinguish this spectral relationship equally as well as the opposite relationship to which they were previously conditioned.
Aspects of Pileated Woodpecker sensory ecology such as olfaction and taste perception are presently undescribed. Though we thought it unlikely that either of these senses aided study subjects in distinguishing control and treatment corks in our experiments, because we included MgCO$_3$ substrates in both experiments, we could not definitively rule out the possibility of confounding influences. We therefore found it prudent to initiate an experiment that attempted to rule out properties of MgCO$_3$ other than spectral characteristics (i.e. Experiment 3).

4. **Experiment 3: decreased UV cork treatment, UV Killer**

4.1 Methods

To test whether PIWO may have identified treatment corks in Experiments 1 or 2 based upon a property of MgCO$_3$ other than UV reflectance (taste, odor, etc.), we conducted another two-choice behavioral assay. If PIWO were responding to the presence of MgCO$_3$ and not to the spectral difference between treatment and control substrates, then behavioral responses should be similar (i.e. not different) between treatment and control substrates when MgCO$_3$ is not present on treatment substrates. This experiment was conducted with five (M) adult, trained, cue-naïve PIWO randomly selected from a population of 15 birds. Experimental substrates were decreased UV corks (N=300; General Methods 1.1.2) and control corks (N=300). All other conditions were as previously described. Trials took place from 20 May – 25 May 2015.

4.2 Results and discussion

Study subjects were more likely to contact treatment corks than control corks ($T = 88.0\%$, $C = 75.3\%$, $F_{1,585.09} = 15.9$, $P < 0.0001$; Fig. 2.1C) and more likely to remove treatment corks ($T = 85.8\%$, $C = 73.4\%$, $F_{1,585.08} = 14.2$, $P = 0.0002$; Fig. 2.1H). With two outliers removed, CHT of treatment corks was greater than control corks ($T = 9.2$, $C = 5.7$, $F_{1,514.99} = 201.8$, $P < 0.0001$; Fig. 2.1M). There was no difference in the order that corks were removed ($P = 0.2$). These
behavioral responses were aligned with our predictions (see Introduction), with the exception of the order removed response.

To test whether cue-naive subjects exhibited a predisposition to forage at decreased UV substrates, we analyzed the data from Trial 1 of this experiment separately. Probability estimates for cork contact were not different ($P = 0.32$), but the cork removed probability was greater for treatments than controls ($T = 87.4\%, \ C = 63.9\%; \ F_{1, \ 54} = 4.2, \ P = 0.042$; Fig. 2.2E). After removing one outlier, CHT was greater for treatment corks than control corks ($T = 9.2, \ C = 5.7; \ F_{1, \ 44.782} = 18.7, \ P < 0.0001$; Fig. 2.2H). There was no difference in the order removed ($P = 0.33$). These results support the greater control substrate (decreased UV) handling time observed with cue-naive subjects in Experiment 1.

Spectrally, treatment and control corks from Experiment 3 were different over 300-400nm ($Z$ ratio = -21.54, $P < 0.0001$), and did not differ over 400-700nm ($Z$ ratio = 1.35, $P = 0.53$; Fig. 2.3C). These were the expected spectral differences.

Subjects in Experiment 3 responded similarly to those in Experiment 2, both sets of birds demonstrated greater behavioral responses towards decreased UV treatment corks than towards control corks. Since MgCO$_3$ was not included in this experiment, there could be no influence of its odor, taste, or other sensory-related properties. We therefore conclude that subjects in both experiments with decreased UV substrates were responding to the reduction of UV reflectance relative to the control substrates, and not merely to the presence of MgCO$_3$.

5. Experiment 4: increased UV wood wafers, wood decay fungi

5.1 Methods

To test whether decayed wood is a useful foraging cue for PIWO, we conducted a two-choice behavioral assay. If so, then behavioral responses towards decayed (treatment) substrates
would be greater than towards control substrates. We used 10 (8 M, 2 F) adult, trained, cue-naïve individuals selected randomly from a population of 15 available study subjects, but stratified by aviary position (5 West, 5 East). Experimental substrates were decayed (N = 236) and control (N = 239) red pine wafers adhered to corks (Size #10, Carolina Biological), with all other conditions as previously described. Trials took place from 4 May – 8 May 2015.

5.2 Results and Discussion

Study subjects were more likely to contact treatment corks than control corks (T = 97.4%, C = 94.7%, F_{1, 1174} = 13.9, P = 0.0002; Fig. 2.1D) and more likely to remove treatment corks (T = 86.6%, C = 75.5%, F_{1, 1172.1} = 23.8, P < 0.0001; Fig. 2.1I). Handling time of treatment corks was greater than control corks (T = 6.5, C = 6.1; F_{1, 1071.5} = 4.3, P = 0.037; Fig. 2.1N). There was no difference in the order that corks were removed (P = 0.18). These behavioral responses were aligned with our predictions, with the exception of the order removed response.

To test whether cue-naïve subjects exhibited a predisposition to forage at decayed substrates, we analyzed the data from Trial 1 separately. There was no difference in any of the response variables (Fig. 2.2), suggesting that the observed responses over 10 trials were conditioned.

Spectrally, treatment and control wafers from Experiment 4 were different over 300-400nm (Z ratio = 5.12, P < 0.0001) and over 400-700nm (Z ratio = -7.83, P < 0.0001; Fig. 2.3D). Based on preliminary work with *P. pini*-decayed and control red pine substrates, these were the expected spectral differences.

In this experiment, we were interested in whether our subjects could distinguish between *P. pini*-decayed and sound pine wafers. The wafers used in these trials were desiccated for at least eight days prior to use, in an effort to remove any influence of the decay fungi other than
the spectral condition of the wafers. However, analysis of volatile organic compounds, often produced by wood decay fungi, was beyond the scope of this study. Therefore, we cannot conclude that subjects relied on only a visual cue (UV or otherwise) to distinguish between treatment and control wafers, just that they were able to make the distinction.

6. Experiment five: decreased UV decayed and sound pine wafers, UV Killer

6.1 Methods

To test whether PIWO could discriminate between decayed and sound wood wafers after both were treated with UVK, we conducted another two-choice behavioral assay. If so, then behavioral responses toward decayed substrates should be greater than toward sound substrates. Subjects were those used in Experiment 4. Experimental substrates were decayed (N=197) and sound (N=190) red pine wafers adhered to corks, both of which were treated with UVK (20 sec bath). Trials took place from 12 May – 16 May 2015.

6.2 Results and Discussion

Study subjects were more likely to contact decayed wafers than sound wafers (D = 96.3%, S = 93.2%, F_{1, 1180} = 12.2, P = 0.0004; Fig. 2.1E) and more likely to remove decayed wafers (D = 83.1%, S = 73.0%, F_{1, 1180} = 23.8, P < 0.0001; Fig. 2J). There was no difference in handling time between decayed and sound wafers (P = 0.12; Fig. 2O), nor was there a difference in the order that wafers were removed (P = 0.18). These behavioral responses aligned with our initial predictions for probability of contact and probability of removal, but not for the handling time and order removed responses.

After treatment with UV Killer, decayed and sound wafers were different over both the 300-400nm (Z ratio = 5.52, P < 0.0001) and 400-700nm (Z ratio = -11.4, P < 0.0001; Fig. 2.3E) portions of the spectrum. For much of the UV range, the relative reflectance of both substrates...
was below 5% (threshold of vision, Eaton and Lanyon 2003), and where it was above that, there was little difference (Fig. 2.5). Regardless, the difference over the UV range was not predicted, so we again employed the RNL model to determine if this translated into a perceptible difference for our subjects. The ΔS values comparing the mean decayed and sound substrate reflectance spectra were 2.4 and 1.4 (both above threshold) under the Average VS and GSWO models, respectively. When we set the UV portion of the mean reflectance spectra to equal in the model, so that the only differences were in the 400-700nm range, there was no change in ΔS (Table 2.1). We therefore conclude that the perceived differences between decayed and sound substrates were entirely in the 400-700 nm range, and that differences in the UV range were not perceptible to our subjects.

In this experiment, we were interested in determining whether reducing differences in UV reflectance between substrates would influence the study subjects’ ability to discriminate between decayed and sound wood wafers. While spectral differences were detected in the UV range, these did not translate into perceptible differences. The differences in contact and removal probability between Experiments 4 and 5 were less than 1%. Substrate handling time was not different in this experiment, but it was different in Experiment 4. These experiments were conducted sequentially with the same subjects, and since there was no difference in contact or removal between Experiments 4 and 5, we attribute the change in handling time between experiments to our subjects becoming more adept at removing the corks rather than being influenced by the UV condition of the substrates. We therefore conclude that our subjects were not influenced by the reduction of UV reflectance (via UVK) of the wood wafers.

7. General Discussion
Our results show that PIWO study subjects were able to distinguish between treatment and control substrates in each of the five experiments. In the first three experiments (i.e. chemically-treated corks), we altered the UV reflectance of the substrates, and any variation to the human visible reflectance was not perceptible according to the RNL model. We had controls in place to account for both olfactory and resonance cues, and also accounted for any non-visual confounding influences of MgCO₃ by removing it from the substrates (Experiment 3). We therefore conclude that Pileated Woodpeckers are sensitive to UV wavelengths, and that this sensitivity translated into a useful foraging cue in our experiments. To our knowledge, this is the first documentation of UV sensitivity in the Piciformes, a widespread avian family of both ecological and economic importance.

Further, study subjects responded in the same manner whether the substrates’ UV reflectance was increased or decreased. Thus, a change in UV reflectance relative to the untreated control substrates, regardless of the direction of the change, is demonstrated to be perceived by PIWOs. This result mirrors those found in behavioral trials with Red-winged Blackbirds (*Agelaius phoeniceus*; Werner et al. 2012), and has implications for ecological interactions with wood decay fungi as well as for developing damage control strategies.

We tested the importance of UV reflectance in discriminating decayed from sound red pine substrates in Experiments 4 and 5. Our subjects did not change their behavior when UV reflectance was reduced in both decayed and sound substrates. Thus, for red pine decayed by *P. pini*, the difference in reflected light from 400-700 nm was sufficient for PIWOs to make the discrimination. There are at least 10,000 species of wood decay fungi (Hibbett and Donoghue 2001), and some woodpecker species exhibit a preference for placing nest and roost cavities within trees decayed by specific fungi (Steirly 1957, Shigo and Kilham 1968, Conner et al. 1976,
Winternitz and Cahn 1983). It is possible that each decay species could create a specific substrate reflectance pattern, and that pattern could vary between tree species as well (O’Daniels, Chpt. 3). A larger sampling of decayed substrates may reveal combinations of decay fungi and tree species that produce substrates for which UV reflectance or absorbance is greatly different from surrounding uninfected wood.

Cue-naïve subjects in Experiment 3 (decreased UV corks) discriminated between decreased UV and control substrates, and the handling time of control corks by cue-naïve subjects in Experiment 1 was greater than that of increased UV treatment corks. These results suggest that decreased UV substrates (e.g. UV-absorbing) may contain information for Pileated Woodpeckers, possibly as a foraging cue. Many flowering plant species exhibit UV-absorbing “nectar guides” on their petals (Fig. 2.4) which have co-evolved with UV-sensitive insect pollinators. Woodpeckers are known to transport spores of fungi present at cavity locations (Farris et al. 2004, Jusino et al. 2015). It is possible that woodpeckers and fungi have evolved mutualisms analogous to those between plants and pollinators, and that such mutualisms could enlist similar communication methods.

Reducing UV reflectance of decayed and sound wood did not alter our subjects’ response, therefore it is unlikely that products which only alter substrate UV reflectance would deter or repel woodpeckers from anthropogenic structures. However, products which pair a negative consequence with a UV cue could be useful in conditioning woodpecker avoidance of treated substrates. In behavioral tests with Red-winged Blackbirds, a UV-absorptive cue paired with 9, 10-anthraquinone (negative post-ingestive consequences) showed a synergistic effect in repellency (Werner et al. 2014). Given that our subjects’ response to the UV condition of
experimental substrates mirrored that of Red-winged Blackbirds, we speculate Pileated Woodpeckers would display a similar response towards a repellent/UV cue combination.

There was no difference in the order that control and treatment substrates were removed in any of the five experiments we conducted. One possible explanation for this is that we simply did not conduct enough trials to detect a difference in substrate removal order. Another explanation is that incorrect choices were merely unrewarded and not negatively reinforced, so the cost for wrong choices was not sufficient to alter foraging-site selection in terms of the order available substrates were accessed. Future studies could explore these hypotheses by modifying our trial protocols.

By using the RNL model to analyze our spectral data, we unexpectedly were able to provide behaviorally supported estimates of Pileated Woodpecker visual system components, SWS1 $\lambda_{\text{max}}$ and the LWS Weber fraction. A value of the SWS1 $\lambda_{\text{max}}$ for the species can be estimated genetically to verify the similarity with GSWO that our modeled data suggests. Estimates of Weber fractions can only be generated with behavioral experiments.

The LWS Weber fraction of the Red-billed Leiothrix (Leiothrix lutea) has been estimated at 0.1, and this value has been widely used in publications incorporating the RNL model despite the suggestion that Weber fractions likely vary by species (Vorobyev 2003, Olsson et al. 2015). Recently, behavioral experiments with domestic Red Junglefowl (Gallus gallus) suggested that an LWS Weber fraction of 0.06 is appropriate for that species (Olsson et al. 2015). Therefore, an LWS Weber fraction of 0.06 for the Pileated Woodpecker would be within the range of published values for avian species. Since we did not design our experiments with the goal of estimating Weber fractions, further behavioral experiments may be required to confirm these
results. Still, our findings represent the first Weber fraction estimates for a woodpecker, and add to the avian visual physiology literature.

8. Conclusions

We have demonstrated that Pileated Woodpeckers can be conditioned with UV cues, but may be pre-disposed to target decreased UV substrates when foraging. Knowledge of woodpecker sensory systems is vital for understanding ecological interactions with fungi which form the basis for cavity nest webs, and our study provides an initial framework for understanding woodpecker visual ecology. Other sensory systems such as olfaction may be important to these interactions as well.

A behavior can be defined as an organism’s response to some environmental stimulus (Levitis et al. 2009). Therefore, if a particular animal behavior is to be somehow manipulated, or “controlled”, an understanding of that organism’s sensory thresholds to external stimuli is imperative. Results from this study should be beneficial for future research seeking to modify woodpecker excavation behaviors through visual cues.

Finally, our behavioral assay represents a novel approach to woodpecker research. With minimal training, our subjects readily performed the task of removing corks in search of food items. This methodology offers the ability to research similar visual ecology questions with other species of woodpecker and primary cavity excavators (e.g. nuthatches). Additionally, our design can be easily modified to investigate other sensory modalities (e.g. olfactory, vibrational) which may be equally important to woodpeckers. We view this study as a starting point, and hope that our work will lead to supplemental research regarding the sensory ecology of woodpeckers.
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Literature Cited


http://CRAN.Rproject.org/package=multcompView


Appendix 1

Methods for the live-capture and care of woodpeckers used in this study, and a description of the testing facilities.

We captured individual PIWO from wild populations in Arkansas (n=20) and Missouri (n=1), USA, from 17 March – 8 May, 2014, and from 31 March – 10 April, 2015, from suitable habitats using mist nets combined with audio recordings of PIWO calls following methods described by York et al. (1998). We conducted behavioral experiments at the Outdoor Animal Research Facility of the National Wildlife Research Center (NWRC), Ft. Collins, Colorado, USA. Live-captures were carried out under authority of the following state and federal permits: US Fish & Wildlife Service Scientific Collection Permit # MB019065-2; USDA Forest Service Scientific Research Permit #’s SO-FW-FY14-15 and SO-FW-FY15-04; Arkansas Game & Fish Commission Scientific Collection Permit # 011520141; and Missouri Department of Conservation Wildlife Collector Permit # 15961. Once captured, woodpeckers were placed in individual temporary outdoor holding pens for 1-14 days until transport to NWRC was possible. Woodpeckers were transported to NWRC in individual holding cages (50 cm x 61 cm x 50 cm) on 21 March, 2014, 18 April, 2014, 8 May, 2014, and 12 April, 2015. Each transport cage was fitted with a cage liner to prevent slipping, and each bird was supplied with maintenance diet *ad libitum* during transport. At NWRC, woodpeckers were housed separately in outdoor aviaries (2.6 m x 2.6 m x 5.3 m). All temporary and permanent aviaries were lined with nylon mesh nets to reduce injury potential. After arriving at NWRC, all subjects underwent a 14 day acclimation/quarantine period during which the daily maintenance diet and fresh water were provided *ad libitum.*
The daily maintenance diet consisted of 20 g canned dog food (beef), 35 g live mealworms, and 50 g mixed fruit (apples, bananas, grapes, oranges) (Tupper et al. 2010; 40 g live mealworms in 2015). Each PIWO had free access to food, water, and 1 untreated 20 cm diameter x 90 cm section of southern yellow pine (*Pinus* sp.) enrichment pole. During testing periods, food restrictions were implemented (see General Methods 1.2). The capture, care, and use of birds were approved by NWRC’s Institutional Animal Care and Use Committee (NWRC study protocols QA2242, QA2398). Permit language precluded test subject release, therefore at the conclusion of testing, subjects were either transferred to an unrelated NWRC study protocol or euthanized per stipulations of US Fish & Wildlife Service Scientific Collecting Permit # MB019065-2.
Appendix 2

The receptor noise-limited (RNL) model of color discrimination (Vorobyev and Osorio 1998) was used to assess unpredicted differences between control and treatment substrate reflectance spectra. The RNL model was implemented through the pavo package (version 0.5-2, Maia et al. 2013) in R as it is described in Vorobyev et al. (1998). We created control and treatment reflectance spectra with equalized UV values by averaging the relative reflectance between control and treatments from 300-400 nm, and compared the ΔS values of the original and UV-equalized spectra as calculated by the model.

There is no published MSP data for any woodpecker species, but we have an estimate of SWS1 $\lambda_{\text{max}}$ for GSWO at 405 nm (Ödeen and Håstad 2003). Therefore, we compared ΔS values generated by two different models of woodpecker visual perception. The first model was based on the receptor quantum catches for the average VS bird (vis= “avgV” in pavo). The second model (GSWO) was created with the sensmodel() function in pavo, using $\lambda_{\text{max}}$ values of 405, 452, 505, and 565 for each receptor. The latter three values are the average values for SWS2, MWS, and LWS cones reported for VS species in Endler and Mielke (2005). We included average absorbance values for T, C, Y, and R oil droplets (oiltype = c(“T”, “C”, “Y”, “R”)) and for avian ocular media (om = “bird”) in the GSWO model. Additional parameters for both models included forest shade irradiance (illum = “forestshade”), the mean reflectance of four test poles as the background spectra, and default values for the density of receptor types to calculate ΔS. The Weber fraction is an estimate of the noise inherent in the neural mechanisms of vision, and is a limiting model parameter. We ran the models with LWS Weber fraction values ranging from 0.05 to 0.1, which covers the range of published values for avian species.
Figure 2.1. Results of generalized linear mixed model (GLMM) analyses (point = mean, error bars = standard error) for contact, removed, and cork handling time (CHT) response variables (Gray = Control, Black = Treatment) for trials with Pileated Woodpeckers (*Dryocopus pileatus*) in 2014 and 2015. All relationships were different (*P* < 0.05; see text for specific values), except for CHT of Experiment 5 (panel O, *P* = 0.18).
Fig. 2.2. Results of generalized linear mixed model (GLMM) analyses (point = mean, error bars = standard error) for contact, removed, cork handling time (CHT), and order removed response variables (Gray = Control, Black = Treatment) for Trial 1 only in experiments with cue-naïve Pileated Woodpeckers (*Dryocopus pileatus*) in 2014 and 2015. Experiments 2 & 5 were not conducted with cue-naïve individuals. ** denotes differences ($P < 0.05$; see text for specific values).
Fig. 2.3. Departure from the mean relative reflectance of the opposite substrate condition (Gray = Control, Black = Treatment) at 1 nm intervals over 300 – 700 nm for each substrate used in trials with Pileated Woodpecker (*Dryocopus pileatus*) in 2014 and 2015. Panels: A = Experiment 1 (increased UV; MgCO$_3$), B = Experiment 2 (decreased UV; UV Killer + MgCO$_3$), C = Experiment 3 (decreased UV; UV Killer only), D = Experiment 4 (increased UV; decayed red pine), E = Experiment 5 (decreased UV; decayed red pine + UV Killer).
Figure 2.4. Human visible (left) and UV-only (right) photographs of the same flower illustrating a UV-absorbing nectar guide. The darker interior portion of petals in the right picture is yellow (UV-absorbing) whereas the lighter outer portion is yellow + UV. Photo credit: Sean O’Daniels
Fig. 2.5. Mean relative reflectance of decayed (N = 197) and sound (N = 190) pine wafers used in behavioral trials with Pileated Woodpecker (*Dryocopus pileatus*; Experiment 5, 2015) over 300-700 nm, 10 nm average. Relative reflectance values below 5% are considered to be not visible.
Table 2.1. Results of RNL model analyses of substrates (LWS Weber fraction = 0.05) used in trials with Pileated Woodpeckers in 2014 and 2015. In Experiment 2, substrates were perceptibly different according to the Great Spotted Woodpecker (GSWO) model (ΔS = 1.3), but were not perceptibly different when UV was set to equal (ΔS = 0.3), demonstrating that UV reflectance was the driver of behavioral discrimination. In Experiment 5, there was no change in ΔS when UV was set to equal, demonstrating that there were no perceptual differences from 300-400 nm. See Appendix 2 for description of visual model parameters.
CHAPTER 3
LIGHT REFLECTANCE OF DECAYED WOOD VARIES BY DECAY SPECIES
AND MAY BE A USEFUL CUE FOR WOODPECKERS

ABSTRACT

Studies of visual ecology in recent decades have provided increased understanding that many organisms visualize their surroundings differently than humans. To glean ecologically relevant information from studies of signals (color or otherwise), a signal should be viewed and analyzed from the perspective of the intended signal receiver. The receptor noise-limited (RNL) color discrimination model has been widely used in studies of feather coloration, egg shells, flowers, and fruit to model how those objects appear to birds. The appearance of wood substrates is likely relevant to at least some birds with life histories that require regular interactions with wood for food and shelter. Woodpeckers often select decayed wood for cavity placement or foraging. Some species appear able to detect trees decayed by specific fungi when placing cavities, but the mechanism that would allow for such specificity is unknown. We hypothesized that decay fungi known to be associated with woodpecker cavity sites will alter substrate reflectance in a species-specific manner that is visually discriminable by woodpeckers. Since woodpecker excavations routinely damage anthropogenic structures, we also hypothesized that some common in-service decay fungi (those found in anthropogenic structures) would not be discriminable from some cavity-associated fungi. To evaluate spectral changes created by the decay process, we grew 10 species of wood decay fungi from pure cultures on substrates from 3 tree species. We autoclaved 20mm x 20mm x 3mm wood wafers in broths of 2% malt dextrose (2M) or 0.5% potato dextrose (PD), placed each wafer in a petri dish on 2M or PD agar, and inoculated each dish with one organism. Control wafers were treated identically, but were not inoculated. Replicates were grown for 5 months under indoor ambient, dark conditions. Each
wafer was then manually cleared of fungal mycelia and desiccated in air. We then collected reflectance spectra from each wafer. We used the RNL model to compare spectra of decayed substrates with spectra of control substrates, and also compared spectra of substrates decayed by different fungi. These analyses showed 6 of 10 decayed substrate/control comparisons were above the threshold of discrimination, and 12 of 13 decayed substrate comparisons were also above threshold. Further, all 8 comparisons of in-service decay substrates with cavity-associated decay substrates were above the threshold of discrimination. We conclude that woodpeckers should be able to visually detect decayed wood, and should also be able to visually detect species-specific differences in wood substrates decayed by these fungi, including in-service decay. Visual confusion between the in-service and cavity-associated fungi selected for this study is therefore an unlikely explanation for woodpecker damage to anthropogenic structures. This study provides evidence for a visual mechanism that allows woodpeckers to identify and select substrates decayed by specific fungi, which has implications for understanding ecologically important woodpecker-fungi mutualisms.

INTRODUCTION

That color signals are important to birds has been evident at least since the days of Aristotle (350 BCE), although our understanding of those signals has historically been interpreted through the eyes of humans. Studies of avian visual ecology have become prevalent in recent decades, driven both by the advent of new technologies and an increased understanding that most organisms visualize their surroundings differently than humans. To glean ecologically relevant information from studies of signals (color or otherwise), a signal should be viewed and analyzed from the perspective of the intended signal receiver. For this, one must have knowledge of the sensory capabilities of that intended receiver, or potential eavesdroppers.
Avian Visual Systems

Diurnal birds possess tetrachromatic vision due to the presence of four types of single cone cells (short wave-sensitive 1, short wave-sensitive 2, medium wave-sensitive, and long wave-sensitive; SWS1, SWS2, MWS, LWS) within the retina, and a wavelength sensitivity range from 300-700 nanometers (nm; Eaton 2005). For comparison, human trichromatic visual sensitivity ranges from 400-700 nm (Osorio and Vorobyev 2008). Avian visual sensitivity includes the near ultraviolet (UV; 300-400 nm), owing to the presence of the fourth cone type (SWS1), which is absent from the primate retina (Osorio and Vorobyev 2008). Within birds, the wavelength sensitivities of the LWS, MWS, and SWS2 cones are relatively constant (Endler and Mielke 2005). However, the SWS1 cone varies between two conditions, based on the wavelength of maximum absorbance ($\lambda_{\text{max}}$), creating dichotomous visual systems between broad taxonomic groups (Ödeen and Håstad 2003). The ‘ultraviolet sensitive’ (UVS) species exhibit an SWS1 $\lambda_{\text{max}}$ from 355-373 nm, whereas the ‘violet sensitive’ (VS) species exhibit a $\lambda_{\text{max}}$ from 402-426 nm (Hart and Hunt 2007). The UVS system is found in Paleognaths, higher Passerines, Psittaciformes, and Laridae, with all other species presently presumed to possess a VS system (Ödeen et al. 2011). Behavioral studies with VS species have shown that regardless of the SWS1 $\lambda_{\text{max}}$, UV sensitivity may extend to at least 360 nm (Parish et al. 1981, 1984, Blackwell et al. 2012).

These $\lambda_{\text{max}}$ values can be measured directly by microspectrophotometry (MSP; Jane and Bowmaker 1988) or estimated from total DNA (Wilkie et al. 2000, Ödeen and Håstad 2003). Such data can then be incorporated into visual models to estimate the saliency of particular wavelengths or the discriminability of colors (e.g. Vorobyev and Osorio 1998, Endler and Mielke 2005). Retinal physiology data (e.g. MSP) are available for very few species, and as a
consequence, MSP data from one species are often used to represent visual sensitivity in others that are assumed to have similar visual systems (e.g. Eaton and Lanyon 2003). Based on our contemporary understandings of avian visual physiology, this approach is not necessarily unwarranted (but see Renoult et al. 2010), and has been acknowledged as a valuable first step for visual ecology studies (Kemp et al. 2015).

*Visual Model*

The receptor noise-limited (RNL) color discrimination model of Vorobyev and Osorio (1998) has been widely used in studies of feather coloration (e.g. Vorobyev et al. 1998, Eaton and Lanyon 2003, Eaton 2005, Benites et al. 2007, Seddon et al. 2010, Burns and Shultz 2012). The RNL model calculates the difference between points (reflectance spectra; ΔS) within a theoretical color space, and can be applied to di-, tri-, and tetrachromatic visual systems. The model assumes that a) discrimination of colors within the color space is limited by noise originating in the receptors, and b) no visual signal occurs when the stimulus and background differ only in intensity (Eaton 2005). The RNL model requires an estimate of the noise inherent within the system of interest, as well as information on the spectral sensitivities (λ max) for, and relative numbers of, photoreceptor types to predict discrimination between pairs of reflectance spectra.

The RNL model also has been used in discrimination studies of objects encountered by birds, such as egg shells (Igic et al. 2012), flowers (Herrera et al. 2008), and fruits (Schaefer et al. 2007). Reflectance spectra of wood substrates have been incorporated into some previous studies as background spectra (e.g. Eaton 2005), but no studies have specifically examined whether birds can discriminate between spectra of different wood substrates. The reflectance
spectra of wood substrates are likely to be relevant to at least some primary cavity excavators with life histories that require regular interactions with wood substrates for food and shelter.

*Woodpecker Ecology*

Woodpeckers (Piciformes: Picidae: Picinae, Leach 1820) comprise approximately 183 species within 24 genera (Winkler and Christie 2002), and are an ecologically important group that is globally distributed across forest and woodland systems. As primary cavity excavators, woodpeckers are a foundational link to nest web communities (Martin and Eadie 1999) because they excavate cavity resources which dozens of other vertebrate species also use (Aubry and Raley 2002, Steeger and Dulisse 2002, Arnett et al. 2010). Nest webs are hierarchically-structured models that map the interdependence of cavity-nesting communities (Bednarz et al. 2004). The 22 extant woodpecker species native to the United States and Canada appear to be even more important to their nest webs, when compared with woodpecker assemblages from other regions (Cockle et al. 2011).

Woodpeckers also aid in the breakdown of dead trees, fallen logs, and coarse woody debris by creating openings for moisture, fungi, and other decomposition agents (Aubry and Raley 2002). They are known to transport fungal spores on their bills and feathers, and may facilitate fungal colonization of wood substrates (Farris et al. 2004). Additionally, associations between woodpeckers and decay fungi are known from cavity locations (Jusino et al. 2015). Woodpeckers typically create nest and roost cavities in standing dead timber or dead limbs, and rarely in live portions (Jackson and Jackson 2004). Cavity placements of widely distributed woodpecker species are necessarily plastic in terms of cavity height and orientation, tree or limb diameter, tree species, canopy cover, surrounding vegetation, and distance from edge (Winternitz and Cahn 1983, Aubry and Raley 2002, Jackson and Jackson 2004).
In contrast, cavity placement appears to be relatively uniform with regard to the condition of the wood substrate selected (Bednarz et al. 2004). Several species of woodpeckers select wood substrates experiencing fungal deterioration for both nest and roost cavities, usually by a Polyporaceae (Basidiomycota) heart rot (reviewed in Jackson and Jackson 2004, Zahner et al. 2012). Presumably, this softer wood is easier to excavate, and therefore less energy is expended for cavity construction, conferring a higher degree of fitness on those individuals that are able to detect softer heartwood (Conner et al. 1994, Zahner et al. 2012).

At least two species of woodpeckers exhibit a well-documented preference for placing cavities in substrates infected by particular species of heart rot fungi, including the endangered Red-cockaded Woodpecker (*Picoides borealis*) with red heart fungus (*Porodaedalea pini*; Steirly 1957, Jackson 1977, Conner and Locke 1982), and the Yellow-bellied Sapsucker (*Sphyrapicus varius*) with the false tinder fungus (*Phellinus igniarius* var. *populinus*; Shigo and Kilham 1968, Kilham 1971). Evidence of deterioration by heart rots often is not obvious to human observers (Jackson and Jackson 2004), and woodpeckers’ use of “conks”, or the fruiting bodies of Polyporaceae, as visual landmarks has been discounted for some species by previous work (Conner et al. 1976, Rudolph et al. 1995, Huss et al. 2002). Differences in resonance between decayed and sound wood have been suggested as one means by which woodpeckers may detect the presence of heart rots (Conner et al. 1976, Rudolph et al. 1995, Zahner et al. 2012). However, this hypothesis remains untested, and selection of trees infected with specific decay fungi seems unlikely by resonance alone.

Woodpeckers may use visual signals to select excavation sites. Reflectance spectra of decayed wood differ from sound wood, and different decay organisms impart differing substrate reflectances (Klapstein et al. 1989). We hypothesized that decay fungi known to be associated
with woodpecker cavity sites alter substrate reflectance in a species-specific manner that is visually discriminable by woodpeckers. Woodpeckers can also create significant damage when excavating within anthropogenic structures (Harness and Walters 2004), and woodpecker damage to wood siding and trim in particular, is often associated with areas of decay (O’Daniels, pers. obs.). Therefore, we were interested in whether reflectance spectra of substrates decayed by fungi associated with anthropogenic structures (in-service decay) were visually similar to substrates decayed by woodpecker cavity-associated fungi such that those substrates could be mistaken for substrates decayed by a cavity-associated fungus.

We used the RNL model to assess whether reflectance spectra of decayed substrates could be discriminated by a hypothetical woodpecker based on the fungi responsible for the decay. We predicted that all of the decayed substrates would produce reflectance spectra that were discriminable from control substrates, and that substrates produced by cavity-associated fungi would differ from all other cavity-associated substrates of the same tree species. We also predicted that some in-service decay fungi would produce substrates that were not discriminable from decayed substrates produced by some of the cavity-associated fungi.

**Methods**

**Wood Substrates**

To examine the change in reflected light between decayed and sound wood, we selected ten decay organisms and three wood substrates (Table 3.1). Wood substrates used in this study were 20 mm x 20 mm x 3mm wafers of quaking aspen (*Populus tremuloides*), northern red oak (*Quercus rubra*), and red pine (*Pinus resinosa*). The wafers were cut perpendicular to the transverse plane (across the grain) to facilitate colonization by fungi. All wafers used in this study were cut from the same individual trees. The decay organisms were selected based on the
substrates available to us, and are either associated with woodpecker cavities in the literature or are common in-service decay fungi.

All wafers were autoclaved in broths of either 2% malt extract (2M; red oak, red pine) or 0.5% potato dextrose (PD; aspen). Sterile wafers were transferred to petri dishes (plates) containing either 2M agar or PD agar (1 per plate). Plates were sealed with Parafilm to prevent desiccation and allow gas exchange, and placed in ambient indoor, dark conditions to monitor for contamination. After seven days, plates were randomly assigned to either control or treatment groups.

Treatment plates were inoculated with one decay organism by agar block transfer from pure cultures (Center for Forest Mycology Research Culture Collection, US Forest Service, Madison, WI). Control plates were unmanipulated. We created 6 replicate plates (wafers) of each decay organism and 5 replicate control wafers of each tree species. After inoculation, plates were re-sealed, returned to dark storage for 5 months, and were periodically monitored for contamination. After ~5 months, treatment wafers were extracted from plates and all fungal mycelia were manually scraped from the wafer surfaces. Control and treatment wafers were placed on paper towels and allowed to desiccate for at least 72 hr before further use.

The surface reflectance of each treatment and control wafer, relative to a white standard, was measured using an Ocean Optics USB2000+ microspectrophotometer calibrated for 200-850 nm with a QR400-7-UV-BX reflectance probe and a PX-2 pulsed xenon light source (Ocean Optics; Dunedin, FL, USA). The probe was calibrated against white (WS-1 Spectralon) and black (the dark) standards, and was re-calibrated between each set (i.e. 5 or 6) of wafers. Reflectance measurements were taken haphazardly from 6 points on each wafer, which were averaged to create a mean reflectance spectrum for each wafer. We used a modified black
rubber stopper to hold the probe at a fixed distance (5 mm) and angle (90°), and to eliminate ambient light.

**Visual Model**

The RNL model was implemented through the *pavo* package (version 0.5 – 4, Maia et al. 2013) within the R environment (version 3.2.3, R Core Team 2015). Specific details of the model can be found elsewhere (Vorobyev et al. 1998), but the Weber fraction, an estimation of the noise within each receptor type (i.e. signal to noise ratio), is a limiting function of color discrimination (Lind et al. 2013). Larger Weber fractions generally indicate greater noise. The RNL model requires an input for the Weber fraction of the LWS cone only, with values for the other cone types calculated based on their relative abundances. Estimates of Weber fractions are available for very few bird species, and are likely to vary by species (Vorobyev 2003). Published values for the Weber fraction of avian LWS mechanisms range from 0.06 for domestic Red Junglefowl (*Gallus gallus*; Olsson et al. 2015) to 0.1 for the Red-billed Leiothrix (*Leothrix lutea*; Lind et al. 2013). Behavioral data suggests that an LWS Weber fraction close to that of the Red Junglefowl may be appropriate for Pileated Woodpeckers (*Dryocopus pileatus*; O’Daniels, Chpt. 2). The majority of published studies based on the RNL model have used an LWS Weber fraction of 0.1, we therefore present modeled data for both 0.06 and 0.1 LWS Weber fractions.

To our knowledge, there are no published MSP data for any woodpecker species, but Ödeen and Hästad (2003) estimated SWS1 λ_{max} for the Great Spotted Woodpecker (*Dendrocopos major*) to be 405 nm. Therefore, we modeled color discrimination based on the receptor quantum catches for the average VS bird under woodland shade irradiance (Endler 1993). We selected the woodland shade irradiance because it also describes the light
environment of coniferous forests, which along with woodlands, are typical woodpecker habitats. Additionally, even the densest of temperate forests experience prolonged periods with no leaf canopy precisely when many woodpeckers excavate cavities.

We compared the mean reflectance spectrum of each wafer (smoothing parameter = 0.2) to all other wafers of the same substrate (QA, RO, or RP) using the RNL model to generate mean ΔS with 95% confidence intervals from all possible pairwise comparisons, both within wafer (e.g. all control aspen wafers) and among wafer types (e.g. P. igniarius vs. P. tremulae). The units of ΔS are ‘just noticeable differences’ (JNDs), with ΔS ≥ 1.0 JND considered to be discriminable. We used 1-sample, 2-tailed t-tests to test for ΔS values that were different from 1.0 (Igic et al. 2012) with α < 0.05, and applied Bonferroni corrections to account for multiple pairwise comparisons.

We modeled substrate reflectance against three different background spectra, an idealized spectrum (default in pavo), a peeled cedar (Thuja sp.) log, and the mean reflectance spectrum of each control substrate (e.g. all aspen substrate comparisons conducted with mean control aspen spectrum as background). All of these backgrounds produced identical results. Values reported are with the mean reflectance of each control substrate as the background.

Results

For illustration, we calculated the percent difference in mean reflectance spectra of decayed and control substrates (Figs. 3.1-3.3) by the formula:

\[(Treatment – Control) / Control\]

With an LWS Weber fraction of 0.06, aspen substrates decayed by P. tremulae were above threshold when compared with control substrates (ΔS = 1.5, \(P < 0.001\)) and substrates decayed by P. igniarius (ΔS = 1.6, \(P < 0.001\)). P. igniarius substrates were not above threshold
compared with controls (Table 3.2). No aspen substrate comparisons exceeded the threshold of discrimination with an LWS Weber fraction of 0.1 (Table 3.3).

With an LWS Weber fraction of 0.06, two of four decayed red oak substrates were above threshold compared with control substrates (\(F. fomentarius\) (\(\Delta S = 2.0, P < 0.001\)), \(S. pachyodon\) (\(\Delta S = 2.5, P < 0.001\)), and five of six comparisons of decayed red oak substrates were also above threshold (Table 3.4). When an LWS Weber fraction of 0.1 was considered, only \(S. pachyodon\) was above threshold compared with controls (\(\Delta S = 1.5, P < 0.001\)), and three of six comparisons of decayed substrates were above threshold. (Table 3.5).

With an LWS Weber fraction of 0.06, three of four decayed pine substrates were above threshold compared with control substrates: \(C. puteana\) (\(\Delta S = 1.3, P < 0.001\)), \(G. trabeum\) (\(\Delta S = 2.8, P < 0.001\)), \(P. pini\) (\(\Delta S = 2.8, P < 0.001\)). All six comparisons of decayed red pine substrates were also above threshold (Table 3.6). With an LWS Weber fraction of 0.1, two of four substrates were above threshold compared with controls, and four of six comparisons of decayed substrates were also above threshold (Table 3.7).

**Discussion**

Our data indicate that decayed and sound wood substrates appear visually different to woodpeckers based on the species of fungi responsible for the decay. Our results show that red pine substrates decayed by \(P. pini\) and northern red oak substrates decayed by \(S. pachyodon\) are above the threshold of discrimination compared with most other substrates in our study, even when more conservative visual model parameters are considered. These results are substantial because both fungi are known to be associated with woodpecker cavities (Jackson and Jackson 2004), and woodpecker detection by the use of external basidiocarps (conks) is unlikely for these two fungi (Connor et al. 1976, Rudolph et al. 1995). Additionally, Pileated Woodpeckers
successfully discriminated between *P. pini* wafers and control red pine wafers in captive behavioral trials (O’Daniels, Chpt. 2).

The results for other woodpecker-associated decay fungi were more equivocal. Aspen substrates decayed by *P. igniarius* were not different from control substrates, regardless of model parameters, and this result was contrary to our predictions. We expected *P. igniarius* substrates to be discriminable from controls based on published associations with woodpecker cavity locations. However, *P. tremulae* substrates were above threshold when compared with both control and *P. igniarius* substrates with an LWS Weber fraction of 0.06. *P. tremulae* was somewhat recently separated from *P. igniarius* (Jackson and Jackson 2004), so it is possible that earlier studies of sapsucker cavity sites were in fact decayed by *P. tremulae* rather than *P. igniarius*. The fact that these fungi produce discriminable substrates under certain conditions may also be a significant finding if future research were to show that *P. tremulae* is found at more woodpecker cavity sites than *P. igniarius*.

*F. pinicola* has been associated with woodpecker cavities located in fir, hemlock, and spruce species in Northwestern North America (Huss et al. 2002), and we expected it to produce multiple above-threshold substrate comparisons, similar to the results with *P. pini* and *S. pachyodon*. After Bonferroni corrections with an LWS Weber fraction of 0.06, *F. pinicola* substrates were not different from control red pine, but were above-threshold compared with the other three fungi. However, with an LWS Weber fraction of 0.1, *F. pinicola* was discriminable only from *P. pini* (Table 3.8). Our experimental substrates were from red pine which does not occur in Northwestern North America. When *F. pinicola* decays other tree species (i.e. fir, hemlock, spruce), the reflectance of the resulting decayed substrates may have a different
appearance and produce more above-threshold (i.e. discriminable) substrate comparisons. This logic applies to any combination of tree species and decay fungi.

Comparisons of substrates decayed by in-service fungi (*B. adusta*, *C. puteana*, *G. trabeum*, and *T. versicolor*) and those decayed by woodpecker-associated fungi produced above threshold values in every case with an LWS Weber fraction of 0.06. Only *F. pinicola* – *G. trabeum* and *F. fomentarius* – *T. versicolor* were not above the threshold with an LWS Weber fraction of 0.1, and while *F. fomentarius* is a heart rot, it is not presently known to be associated with woodpecker cavities. Our results do not offer support for the idea that these in-service decay fungi could be visually mistaken for a “preferred” decay fungi when they occur on anthropogenic structures, but by only examining 10 decay fungi out of at least 10,000 (Hibbett and Donoghue 2001), we cannot rule the possibility out.

The majority of fungi examined in this study are heart rots. Heart rot fungi are important to woodpeckers because they decay the interior heartwood of a tree, without affecting the integrity of the more exterior sapwood (Kilham 1971, Conner et al. 1994). This fact seemingly calls into question the feasibility of a visual cue produced by a heart rot that is detectable and useful for woodpeckers. However, the decay fungi have to enter the tree somehow in order to access the heartwood, and this often occurs at the site of an injury such as a broken branch or tree top (Jackson and Jackson 2004). Red-cockaded Woodpecker cavities are typically located directly below a branch stub where a decay fungi, often *P. pini*, entered the tree (Fig. 3.4; Conner et al. 2004). It may be that in such instances, the reflectance of the exposed heartwood where the decay fungi enters becomes altered in the manner that we have presented here. This well studied system seems to lend validation to the idea of such an external cue (“tree selection hypothesis”).
Jusino et al. 2015), and portable spectrometers can be used in the field to investigate this hypothesis.

Many decay fungi produce species-specific combinations of volatile organic compounds (volatiles), and insects use these volatiles to locate decaying trees (Raffa and Smalley 1995). No published data on olfactory abilities exist for any woodpecker species, but recent research supports the idea that olfaction is an important sense for a wide range of avian taxa (Mihailova et al. 2014). Significant olfactory ability in woodpeckers is not a mutually exclusive theory in the detection of decayed wood substrates. Indeed, visually detectable cues could compliment olfactory cues, as is often the case in plant-pollinator relationships (Schiestl 2005). Since woodpeckers are known to transport fungal spores, they could and perhaps should, be considered to fill analogous roles to pollinators.

Several woodpecker species worldwide are considered to be threatened or endangered, and the population status is declining or unknown for several others (IUCN Red List 2015). Given that woodpeckers are important components of the ecosystems that they inhabit, a better understanding of woodpecker sensory ecology, and any mutualistic relationships in which they participate, will likely improve conservation efforts and management strategies.

**Conclusion**

We have presented evidence that decay fungi create varying substrate reflectances by species, and that such variations are likely visually detectable by woodpeckers. This is the first description of decayed wood substrate reflectance in an ecologically relevant context. The idea that some woodpeckers select trees decayed by particular fungi is supported by decades of field observations involving multiple species of woodpecker and decay fungi (e.g. Stierly 1957, Shigo and Kilham 1969, Conner et al. 1976, Jackson 1977). While we do not present direct evidence
that woodpeckers are visually able to discriminate between wood decayed by different fungi, our results suggest that it is theoretically possible. Visual cues may not be used at all, or may be used in conjunction with olfactory or resonance cues. We hope these findings will spur further research into woodpecker sensory ecology, including olfaction, and woodpecker-fungi mutualisms.

**Acknowledgments**

We would like to thank J. Bruhn and E. Fernandez-Juricic for productive discussions at the outset of this research, and R. Maia for analytical advice and help navigating *pavo*. Additionally, we thank J. Endler for sharing irradiance data. Funding for this study was provided by Arkion Life Sciences, the Avian Power Line Interaction Committee, and Critter Control, Inc. SO was partially supported by a Trans World Airlines Graduate Scholarship.

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Renoult, J.P., A. Courtiol, and F. Kjellberg. 2010. When assumptions on visual system evolution


Figure 3.1. Difference in mean relative reflectance of decayed and control quaking aspen (Populus tremuloides) substrates over 300 – 700 nm (10 nm avg.) created in 2015. Pi = Phellinus igniarius, Pt = Phellinus tremulae. Control and P. igniarius N = 5; P. tremulae N = 6.
Figure 3.2. Difference in mean relative reflectance of decayed and control northern red oak
(Quercus rubra) substrates over 300 – 700 nm (10 nm avg.) created in 2015. Ba = Bjerkandera
adusta, Ff = Fomes fomentarius, Sp = Spongipellis pachyodon, Tv = Trametes versicolor.
Control, B. adusta, F. fomentarius, and T. versicolor N = 5; S. pachyodon N = 6.
Figure 3.3. Difference in mean relative reflectance of decayed and control red pine (*Pinus resinosa*) substrates over 300 – 700 nm (10 nm avg.). Cp = *Coniophora puteana*, Fp = *Fomitopsis pinicola*, Gt = *Gleophyllum trabeum*, Pp = *Porodaedalea pini*. Control N = 5; C. *puteana*, F. *pinicola*, G. *trabeum*, and P. *pini* N = 6. *P. pini* – Control comparison is from wafers created in 2014, all other wafers were created in 2015 with separate controls.
Exposed heartwood with reflectance possibly altered by *P. pini*

**Fig. 3.4.** Diagram of typical RCW cavity location. Modified from Conner et al. (2004), Figure 1.
**TABLES**

**Table 3.1.** List of decay fungi used in this study. RP = red pine (*Pinus resinosa*), RO = northern red oak (*Quercus rubra*), QA = quaking aspen (*Populus tremuloides*).

<table>
<thead>
<tr>
<th>Decay Fungi</th>
<th>Wood Substrate</th>
<th>Relevance</th>
<th>Woodpecker Species</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Phellinus</em> igniarius</td>
<td>QA</td>
<td>Woodpecker, Heart Rot</td>
<td><em>Sphyrapicus</em> spp.</td>
<td>Shigo and Kilham (1968)</td>
</tr>
<tr>
<td><em>Bjerkandera</em> adusta</td>
<td>RO</td>
<td>In-Service</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Fomes</em> fomentarius</td>
<td>RO</td>
<td>Heart Rot</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Spongipellis</em> pachyodon</td>
<td>RO</td>
<td>Woodpecker, Heart Rot</td>
<td><em>Dryocopus pileatus</em> (Pileated)</td>
<td>Conner et al. (1976)</td>
</tr>
<tr>
<td><em>Trametes</em> versicolor</td>
<td>RO</td>
<td>In-Service</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Coniophora</em> puteana</td>
<td>RP</td>
<td>Heart Rot, In-Service</td>
<td>not identified</td>
<td>Parks et al. (1996)</td>
</tr>
<tr>
<td><em>Fomitopsis</em> pinicola</td>
<td>RP</td>
<td>Woodpecker, Heart Rot</td>
<td><em>Picoides villosus</em> (Hairy)</td>
<td>Huss et al. (2002)</td>
</tr>
<tr>
<td><em>Gleophyllum</em> trabeum</td>
<td>RP</td>
<td>In-Service</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Porodaedalea</em> pini</td>
<td>RP</td>
<td>Woodpecker, <em>Picoides borealis</em></td>
<td>Heart Rot (Red-cockaded)</td>
<td>Steirly (1957)</td>
</tr>
</tbody>
</table>
Table 3.2. Receptor noise-limited model results of QA substrates; LWS Weber fraction = 0.06. Mean $\Delta S$ (95% CI); $N =$ number of pairwise comparisons used to generate mean $\Delta S$. Bold values indicate $\Delta S > 1$ JND (Bonferroni-adjusted $P < 0.05$).

<table>
<thead>
<tr>
<th>Referent Substrate</th>
<th>Comparison Substrate</th>
<th>$\Delta S$ (95% CI)</th>
<th>$N$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aspen</td>
<td>Phellinus igniarus</td>
<td>1.7 (0.8 - 2.5)</td>
<td>10</td>
</tr>
<tr>
<td>Aspen</td>
<td>Phellinus tremulae</td>
<td>1.6 (0.6 - 2.5)</td>
<td>10</td>
</tr>
<tr>
<td>Phellinus igniarus</td>
<td>N = 25</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Phellinus tremulae</td>
<td>1.4 (0.9 - 1.9)</td>
<td>1.6 (1.3 - 1.8)</td>
<td></td>
</tr>
<tr>
<td>Phellinus</td>
<td>1.5 (1.3 - 1.8)</td>
<td>1.6 (1.4 - 1.9)</td>
<td>1.0 (0.7 - 1.3)</td>
</tr>
<tr>
<td>N = 30</td>
<td>N = 30</td>
<td>N = 15</td>
<td></td>
</tr>
</tbody>
</table>
Table 3.3. Receptor noise-limited model results of QA substrates; LWS Weber fraction = 0.1. Mean ΔS (95% CI), N = number of pairwise comparisons used to generate mean ΔS. * denotes ΔS < 1 JND (Bonferroni-adjusted $P < 0.05$). No comparisons were above the threshold of 1.0.

<table>
<thead>
<tr>
<th>Referent Substrate</th>
<th>Comparison Substrate</th>
<th>Phellinus igniarius</th>
<th>Phellinus tremulae</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aspen</td>
<td>Phellinus igniarius</td>
<td>0.8 (0.5 - 1.1)</td>
<td>0.9 (0.4 - 1.5)</td>
</tr>
<tr>
<td>Aspen</td>
<td>Phellinus tremulae</td>
<td>0.9 (0.8 - 1.1)</td>
<td>1.0 (0.9 - 1.1)</td>
</tr>
<tr>
<td>Aspen N = 10</td>
<td>Phellinus igniarius N = 25</td>
<td>N = 10</td>
<td></td>
</tr>
<tr>
<td>Aspen N = 10</td>
<td>Phellinus tremulae  N = 30</td>
<td>N = 30</td>
<td>N = 15</td>
</tr>
</tbody>
</table>
Table 3.4. Receptor noise-limited model results of RO substrates; LWS Weber fraction = 0.06.

Mean ΔS (95% CI); N = number of pairwise comparisons used to generate mean ΔS. Bold values indicate ΔS > 1 JND (Bonferroni-adjusted $P < 0.05$), ‡ denotes values not significant after Bonferroni correction.

<table>
<thead>
<tr>
<th>Referent Substrate</th>
<th>Comparison Substrate</th>
<th>Red</th>
<th>Bjerkandera</th>
<th>Fomes</th>
<th>Spongipellis</th>
<th>Trametes</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Oak</td>
<td></td>
<td>adusta</td>
<td>fomentarius</td>
<td>pachyodon</td>
<td>versicolor</td>
</tr>
<tr>
<td>Red</td>
<td>0.7 (0.4 – 1.0)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Oak</td>
<td>N = 10</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bjerkandera</td>
<td>1.5 (1.1 - 1.9)‡</td>
<td>1.4 (1.0 - 1.9)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>adusta</td>
<td>N = 25</td>
<td>N = 10</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fomes</td>
<td>2.0 (1.7 – 2.3)</td>
<td>3.3 (2.8 – 3.8)</td>
<td>1.5 (0.8 - 2.3)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>fomentarius</td>
<td>N = 25</td>
<td>N = 25</td>
<td>N = 10</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Spongipellis</td>
<td>2.5 (2.3 – 2.8)</td>
<td>4.0 (3.6 - 4.3)</td>
<td>1.2 (0.8 - 1.6)</td>
<td>0.7 (0.5 - 0.9)‡</td>
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<td></td>
</tr>
<tr>
<td>pachyodon</td>
<td>N = 30</td>
<td>N = 30</td>
<td>N = 30</td>
<td>N = 15</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Trametes</td>
<td>0.9 (0.7 – 1.0)</td>
<td>1.9 (1.5 – 2.3)</td>
<td>1.8 (1.5 – 2.1)</td>
<td>2.3 (2.0 – 2.6)</td>
<td>1.0 (0.8 - 1.3)</td>
<td></td>
</tr>
<tr>
<td>versicolor</td>
<td>N = 25</td>
<td>N = 25</td>
<td>N = 25</td>
<td>N = 30</td>
<td>N = 10</td>
<td></td>
</tr>
</tbody>
</table>
Table 3.5. Receptor noise-limited model results of RO substrates; LWS Weber fraction = 0.1. Mean ΔS (95% CI); N = number of pairwise comparisons used to generate mean ΔS. Bold values indicate ΔS > 1 JND (Bonferroni-adjusted P < .05); * denotes ΔS < 1 JND (Bonferroni-adjusted P < 0.05); ‡ denotes values not significant after Bonferroni correction.

<table>
<thead>
<tr>
<th>Referent Substrate</th>
<th>Comparison Substrate</th>
<th>Red</th>
<th>Bjerkandera</th>
<th>Fomes</th>
<th>Spongipellis</th>
<th>Trametes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Red</td>
<td>Oak</td>
<td>0.4</td>
<td>(0.2 - 0.6)*</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Oak</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bjerkandera</td>
<td>adusta</td>
<td>0.9</td>
<td>(0.7 - 1.2)</td>
<td>0.9</td>
<td>(0.6 - 1.1)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fomes</td>
<td></td>
<td>1.2</td>
<td>(1.0 - 1.4) ‡</td>
<td>2.0</td>
<td>(1.7 - 2.3)</td>
<td>0.9</td>
</tr>
<tr>
<td>fomentarius</td>
<td></td>
<td>N = 25</td>
<td>N = 25</td>
<td>N = 10</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Spongipellis</td>
<td>pachyodon</td>
<td>1.5</td>
<td>(1.4 - 1.7)</td>
<td>2.4</td>
<td>(2.1 - 2.6)</td>
<td>0.7</td>
</tr>
<tr>
<td></td>
<td></td>
<td>N = 30</td>
<td>N = 30</td>
<td>N = 30</td>
<td>N = 15</td>
<td></td>
</tr>
<tr>
<td>Trametes</td>
<td>versicolor</td>
<td>0.5</td>
<td>(0.4 - 0.6)*</td>
<td>1.1</td>
<td>(0.9 - 1.4)</td>
<td>1.1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>N = 25</td>
<td>N = 25</td>
<td>N = 25</td>
<td>N = 30</td>
<td>N = 10</td>
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</table>
Table 3.6. Receptor noise-limited model results of RP substrates; LWS Weber fraction = 0.06.

Mean ΔS (95% CI); N = number of pairwise comparisons used to generate mean ΔS. Bold values indicate ΔS > 1 JND (Bonferroni-adjusted \( P < 0.05 \)); * denotes ΔS < 1 JND (Bonferroni-adjusted \( P < 0.05 \)); ‡ denotes values not significant after Bonferroni correction.

<table>
<thead>
<tr>
<th>Referent Substrate</th>
<th>Comparison Substrate</th>
<th>Coniophora</th>
<th>Fomitopsis</th>
<th>Gleophyllum</th>
<th>Porodaedalea</th>
</tr>
</thead>
<tbody>
<tr>
<td>Red Pine</td>
<td>puteana</td>
<td>1.3 (1.2 - 1.3)</td>
<td>0.4 (0.3 - 0.5)*</td>
<td>N = 30</td>
<td>N = 15</td>
</tr>
<tr>
<td>Coniophora puteana</td>
<td>N = 30</td>
<td>N = 15</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fomitopsis pinicola</td>
<td>1.4 (1.0 - 1.9) ‡</td>
<td>1.6 (1.4 - 1.7)</td>
<td>1.8 (1.2 - 2.4) ‡</td>
<td>N = 30</td>
<td>N = 36</td>
</tr>
<tr>
<td>Gleophyllum trabeum</td>
<td>2.8 (2.6 - 3.0)</td>
<td>2.0 (1.9 - 2.2)</td>
<td>1.9 (1.5 - 2.3)</td>
<td>0.9 (0.7 - 1.1)</td>
<td>N = 30</td>
</tr>
<tr>
<td>Porodaedalea pini</td>
<td>2.8 (2.6 - 3.1)</td>
<td>4.2 (3.9 - 4.5)</td>
<td>3.9 (3.4 - 4.4)</td>
<td>2.4 (2.1 - 2.7)</td>
<td>1.4 (1.1 - 1.7) ‡</td>
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<td>N = 30</td>
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</tbody>
</table>
Table 3.7. Receptor noise-limited model results of RP substrates; LWS Weber fraction = 0.1.

Mean $\Delta S$ (95% CI); $N$ = number of pairwise comparisons used to generate mean $\Delta S$. Bold values indicate $\Delta S > 1$ JND (Bonferroni-adjusted $P < 0.05$); * denotes $\Delta S < 1$ JND (Bonferroni-adjusted $P < 0.05$).

<table>
<thead>
<tr>
<th>Referent Substrate</th>
<th>Comparison Substrate</th>
<th>Red</th>
<th>Coniophora</th>
<th>Fomitopsis</th>
<th>Gleophyllum</th>
<th>Porodaedalea</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pine</td>
<td>Coniophora puteana</td>
<td>0.2</td>
<td>(0.2 - 0.3)*</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td></td>
<td>Coniophora puteana</td>
<td>N = 10</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pine</td>
<td>Fomitopsis pinicola</td>
<td>0.8</td>
<td>(0.7 - 0.8)*</td>
<td>0.3 (0.2 - 0.3)*</td>
<td></td>
<td></td>
</tr>
<tr>
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<td></td>
</tr>
<tr>
<td>Pine</td>
<td>Gleophyllum trabeum</td>
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<td>(0.6 - 1.1)</td>
<td>0.9 (0.8 - 1.0)</td>
<td>1.1 (0.7 - 1.4)</td>
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<tr>
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<td>N = 30</td>
<td>N = 36</td>
<td>N = 36</td>
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</tr>
<tr>
<td>Pine</td>
<td>Porodaedalea pini</td>
<td>1.7</td>
<td>(1.6 - 1.8)</td>
<td>1.2 (1.1 - 1.3)</td>
<td>1.1 (0.9 - 1.4)</td>
<td>0.5 (0.4 - 0.7)*</td>
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<td></td>
<td>Porodaedalea pini</td>
<td>N = 30</td>
<td>N = 36</td>
<td>N = 36</td>
<td>N = 36</td>
<td>N = 15</td>
</tr>
<tr>
<td>Pine</td>
<td>Porodaedalea pini</td>
<td>1.7</td>
<td>(1.6 - 1.8)</td>
<td>2.5 (2.3 - 2.7)</td>
<td>2.3 (2.0 - 2.6)</td>
<td>1.4 (1.3 - 1.6)</td>
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<td>Porodaedalea pini</td>
<td>N = 30</td>
<td>N = 36</td>
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<td>N = 15</td>
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</tbody>
</table>
Woodpeckers (Piciformes: Picidae: Picinae, Leach 1820) are an ecologically and economically important avian group. Woodpecker excavating behaviors (cavity formation, foraging, and drumming) are the basis for numerous ecological relationships with vertebrates (Martin et al. 2004), invertebrates (Otvos et al. 1965), and fungi (Farris et al. 2004), but those same excavating behaviors are responsible for millions of dollars in damage to anthropogenic structures annually (Harness and Walters 2004). In this study, I investigated aspects of Pileated Woodpecker (PIWO; *Dryocopus pileatus*) vision in an attempt to better understand their wavelength sensitivity range, interactions with decayed wood, and the potential to manipulate or control woodpecker excavating behaviors by altering ultraviolet (UV) reflectance of substrates.

I found that PIWO are sensitive to the UV condition of a substrate, to the extent that PIWO can be conditioned with both an increased UV cue (i.e. MgCO$_3$) and a decreased UV cue (i.e. UV Killer). The behavioral responses to UV reflectance demonstrated by PIWO study subjects were similar to those reported for other avian taxa (Werner et al. 2012). I also found that UV cue-naïve study subjects exhibited a preference for substrates with lower UV reflectance (i.e. UV-absorbing) than alternative substrates, and this finding suggests that decreased UV reflectance (relative to nearby substrates) may inform foraging site decisions for PIWO. The UV condition of red pine decayed by *Porodaedalea pini* did not appear to influence study subjects’ ability to detect decay. Therefore, attempts to control woodpecker excavations merely by altering the UV condition are likely to be ineffective. However, UV cues paired with negative consequences could be successful in deterring woodpecker excavations in unwanted locations (i.e. anthropogenic structures).
The results of my behavioral experiments can be combined with the receptor noise-limited model (RNL; Vorobyev and Osorio 1998) to provide estimates of two components of the PIWO visual system, SWS1 $\lambda_{\text{max}}$ and the LWS Weber fraction. The wavelength of maximum absorbance ($\lambda_{\text{max}}$) for the SWS1 opsin is used to categorize avian visual systems as either ultraviolet-sensitive (UVS) or violet-sensitive (VS), and our results suggest that PIWO should have an SWS1 $\lambda_{\text{max}}$ identical or very similar to that of the Great Spotted Woodpecker ($Dendrocopus major$) at 405 nm. A value for PIWO SWS1 $\lambda_{\text{max}}$ can be estimated genetically, but this finding aligns with our contemporary understanding of the conservative nature of avian visual systems.

The noise inherent within neural processing mechanisms is estimated by the Weber fraction, and is an essential component of the RNL model (Vorobyev and Osorio 1998). Estimates of Weber fractions can only be determined from behavioral studies, and published values for avian species range from 0.06 to 0.1 (Olssen et al. 2015) for long wave sensitive (LWS) cones. My behavioral trials suggest that a Weber fraction of $\leq 0.06$ is appropriate for PIWO LWS cones, which fits within that published range. However, because we did not design our experiments specifically for the purpose of determining the LWS Weber fraction for PIWO, further work may be required to confirm this finding.

Some species of woodpeckers preferentially place cavities within trees that have been decayed by particular species of fungi. Although these associations have been documented through decades of field observations with multiple species of woodpecker and fungi, no mechanism has been identified that might allow such a species-specific detection by woodpeckers. I used the RNL model to assess how a theoretical woodpecker might perceive the spectral differences between wood substrates exposed to different decay fungi. The model
predicted that substrates decayed by *P. pini* or *Spongipellis pachyodon* should be discriminable from substrates decayed by several other fungi and from undecayed substrates. Both of these fungi are known to be associated with woodpecker cavities, and other means of visual detection have been ruled out (Conner et al. 1976, Rudolph et al. 1995).

Other species of decay fungi for which woodpeckers seem to select did produce discriminable substrates under less conservative model parameters. Further, since I only analyzed three different substrates, it is possible that other combinations of tree species and decay fungi may also produce discriminable substrates. Nonetheless, the finding that woodpeckers will perceive some decayed substrates as different from other decayed substrates provides a possible method by which they can identify and select these substrates for cavity placement.

Given that woodpeckers contribute to keystone ecosystem processes (Bednarz et al. 2004), any mutualisms or other such interactions in which they participate are also likely to be ecologically important. By assessing some visual capabilities of PIWO with regards to UV and decayed wood substrates, I have provided an initial framework to develop a better understanding of woodpecker sensory ecology and woodpecker-fungi interactions. Woodpeckers and fungi may participate in relationships that are analogous to those of plants and pollinators. If true, olfactory cues may be as, or more important than visual cues. My behavioral assay represents a novel approach for woodpecker research, and can be easily modified to investigate other sensory modalities with Pileated Woodpeckers or other species of cavity excavators.

**Literature Cited**


