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Anthraquinone-based repellent for horned larks, great-tailed grackles, American crows and the protection of California's specialty crops

Scott J. Werner*, Shelagh T. DeLiberto, Anna M. Mangan, Susan E. Pettit, Jeremy W. Ellis, James C. Carlson

United States Department of Agriculture, Animal and Plant Health Inspection Service, Wildlife Services, National Wildlife Research Center, 4101 LaPorte Avenue, Fort Collins, CO 80521-2154, USA

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ABSTRACT

Specialty crops include fresh and dried fruits, vegetables, tree nuts, and horticultural and nursery crops. California accounts for 28% of the specialty crop acreage in the United States of America, including 72% of U.S. lettuce production (*Lactuca sativa* L.), 27% of U.S. melon production and 100% of U.S. almond production (*Prunus dulcis* L.). We conducted controlled feeding experiments to evaluate an anthraquinone-based repellent for horned larks (*Eremophila alpestris* L.), great-tailed grackles (*Quiscalus mexicanus* Gmelin) and American crows (*Corvus brachyrhynchos* Brehm) associated with the depredation of California's lettuce, melon and almond crops, respectively. We observed 38–100% feeding repellency among horned larks offered wheat seeds (*Triticum* spp. L.) treated with 168–3010 ppm anthraquinone during the concentration-response experiment. Great-tailed grackles exposed to rice seeds (*Oryza sativa* L.) treated with 2060–35,400 ppm anthraquinone exhibited 90–100% repellency. We observed 80–100% repellency among American crows offered almonds treated with 2980–31,500 ppm anthraquinone. We predicted a threshold concentration of 5200 ppm anthraquinone for American crows offered treated almonds. Our laboratory efficacy data provide a reliable basis for planning future field applications of anthraquinone-based bird repellents for the protection of specialty crops. Supplemental field efficacy studies are necessary for the registration of avian repellents and the management of agricultural depredation caused by wild birds.

1. Introduction

Within the United States of America, the designation of commodity and specialty crops is intended to highlight the differences between non-perishable crops such as grains (e.g. corn, *Zea mays* L.; soybeans, *Glycine max* L.) and the foods people eat directly (http://www.ucsusa.org/publications/ask/2011/fruits-and-veg.html#.VDcA03KKBMw). The value of farm-level specialty crop production in 2012 totaled nearly $60 billion, representing approximately one-fourth of the value of U.S. crop production, yet specialty crops encompass only 3% of harvested cropland in the U.S. (USDA NASS, 2009; Johnson, 2014). In addition to the relatively high value of specialty crops, the production costs per ha are higher for specialty crops than non-specialty crops (GAO/RCED, 1999).

Specialty crop production within the U.S. is mainly located in California, Florida, Washington, Oregon, North Dakota and Michigan (Johnson, 2014). California accounts for 28% of the specialty crop acreage in the U.S. (USDA NASS, 2009), including 72% of U.S. lettuce production (*Lactuca sativa* L.), 27% of U.S. melon production (cantaloupe and honeydew [*Cucumis* spp. Naudin], and watermelon [*Citrullus lanatus* Matsum. & Nakai]) and 100% of U.S. almond production (*Prunus dulcis* L.; USDA NASS, 2014). Several bird species cause monetary losses to agricultural production in California. Gebhardt et al. (2011) identified Alaudidae (larks), Corvidae (crows, jays), Fringillidae (finches) and Turdidae (thrushes) as some of the primary bird families that cause damage to economically important crops in California.

Several birds can cause damage to California's economically-important lettuce, melon and almond crops (Table 1). Horned larks (*Eremophila alpestris* L.) uniquely consume lettuce seeds, uproot seedlings, and graze seedling leaves (i.e. cotyledons). Damaged lettuce seedlings are typically stunted or disfigured, and thus disrupt harvest schedules. Great-tailed grackles (*Quiscalus*

* Corresponding author.
E-mail address: Scott.J.Werner@aphis.usda.gov (S.J. Werner).

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Table 1
Economically-important specialty crops for California agriculture associated with bird depredation (NASS 2014).

Specialty crop	Hectares planted (California)	Production Value	Yield loss per damaged hectare[a] (%)	Depredating bird species[b,c,d]
Lettuce	81,340	$1.7 billion	6.1	Horned Lark
Melons	25,700	$334.5 million	4.2	Great-tailed Grackle/ American Crow
Almonds	339,940	$5.8 billion	5.1	American Crow

[a] Data from Gebhardt et al., 2011.
[b] Hasey and Salmon, 1993; Hamby and Zalom, 2013.
[c] Koehler, 1962; Clark, 1976; York et al., 2000.
[d] Rappole et al., 1989; LeBoeuf, 2002.

mexicanus Gmelin) have been observed to damage citrus groves in Texas (Hobbs and Leon, 1987; Johnson et al., 1989; Glahn et al., 1997). Bird damage to young and ripening melons makes the fruit unfit for harvest (LeBoeuf, 2002). Crows can cause damage to agricultural production by consuming crop seeds and seedlings (Heckmanns and Meisenheimer, 1944; Kennedy and Connery, 2008). Crows consume almonds in orchards as they mature and they cause almonds to fall to the ground, thus making them unsuitable for harvest (Gardner, 1926; Emlen, 1937; Hasey and Salmon, 1993). Recent studies also suggest that increasing bird damage in almond orchards is correlated with an increasing infestation of navel orangeworms (*Amyelois transitella* Walker), an insect that feeds directly on the nut meat of almonds and thus makes them unmarketable (Hamby and Zalom, 2013).

Damage to specialty crops has motivated the use of several bird damage management techniques, including chemical repellents. Although methiocarb effectively reduced horned lark damage to lettuce seedlings in aviary tests (Cummings et al., 1998) and a field enclosure study (York et al., 2000), methiocarb is no longer registered as a bird repellent in the U.S. for use on agricultural crops. Methyl anthranilate, a naturally occurring compound, did not provide effective repellency when applied as a foliar spray (i.e. CO_2 backpack sprayer and/or tractor-mounted sprayer) to lettuce and cantaloupe (Umeda and Sullivan, 2001), and ripening rice and sunflower (Werner et al., 2005). Recent laboratory efficacy studies have estimated the threshold concentration of anthraquinone as a chemical repellent for Canada geese (*Branta canadensis* L.), red-winged blackbirds (*Agelaius phoeniceus* L.), ring-necked pheasants (*Phasianus colchicus* L.; Werner et al., 2009), common grackles (*Q. quiscula* L.; Werner et al., 2011), European starlings (*Sturnus vulgaris* L.; Tupper et al., 2014) and wild turkeys (*Meleagris gallopavo* L.; Werner et al., 2014a).

The present study was designed to evaluate the repellency of an anthraquinone-based repellent and develop an anthraquinone concentration-response relationship for horned larks, great-tailed grackles and American crows (*Corvus brachyrhychos* Brehm) in captivity. These bird species-specific, concentration-response relationships will provide the basis for future field studies to better determine the utility of anthraquinone-based repellents for protecting California's specialty crops from bird damage. The capture, care and use of all birds associated with these experiments were approved by the Animal Care and Use Committee of the United States Department of Agriculture's (USDA) National Wildlife Research Center (NWRC Study Protocols QA-1825, QA-1902 and QA-1939; S.J. Werner- Study Director).

2. Methods

The anthraquinone-based repellent used for each of three feeding experiments included 50% 9,10-anthraquinone (Avipel® Shield, Arkion Life Sciences, New Castle, DE, USA). Horned larks and American crows were maintained in individual cages, and great-tailed grackles were maintained in a group during quarantine and holding by the NWRC Animal Care Unit. All birds were quarantined for a minimum of five days prior to testing. Water was provided *ad libitum* to each test subject throughout the study (quarantine, holding, acclimation, testing). A nutrient-complete maintenance diet was provided *ad libitum* to each test subject throughout quarantine and holding. The maintenance diet for horned larks included 45% millet, 33% crushed poultry feed, 11% wheat and 11% cracked corn. The maintenance diet for great-tailed grackles included equal parts of cracked corn, milo, safflower, small-kibble dog food and rice. The maintenance diet for American crows included dry dog food (i.e. mixed kibble).

For each of three feeding experiments, test subjects acclimated within individual cages for five days subsequent to quarantine and holding. During the acclimation period, one bowl of unadulterated test diet was presented *ad libitum* within each cage at approximately 0800 h, daily. During the three days subsequent to the acclimation period (i.e. pre-test), one bowl of unadulterated test diet (30 g for horned larks, 75 g for great-tailed grackles and American crows) was presented within each cage at approximately 0800 h, daily. Daily consumption of the test diet was measured for each test subject ($\pm$0.1 g) throughout the pre-test (including spillage and desiccation; Werner et al., 2009).

We ranked birds based upon average pre-test consumption and assigned them to one of several test groups such that each group was similarly populated with birds that exhibited high–low daily consumption. We randomly assigned test treatments (i.e. Avipel® Shield-treated test diet) among groups. Test treatments were formulated by applying aqueous suspensions to test diets (60–100 ml/kg) using a rotating mixer and household spray equipment (Werner et al., 2009). A 100-g sample of each formulated test diet was collected within 24 h of each feeding experiment and then submitted the NWRC Analytical Chemistry Unit for their quantification of anthraquinone residues among test treatments (i.e. high performance liquid chromatography; Werner et al., 2011, 2014a,b).

The dependent measure of our feeding experiments was calculated as test consumption of repellent-treated test diet relative to average pre-test consumption of untreated test diet (i.e. percent repellency = [one − (group-average test consumption/group-average pre-test consumption)] * 100). Logarithmic regression procedures (Proc Reg, SAS v9.2) were used to analyze repellency as a function of anthraquinone concentration. Repellent dose (mg anthraquinone/kg body mass) and threshold repellent concentration (ppm anthraquinone) were estimated for bird species that exhibited significant concentration-response relationships including $\leq$80% and $\geq$80% repellency (Werner et al., 2009).

2.1. Horned larks and anthraquinone seed treatment

The purpose of this experiment was to develop an anthraquinone concentration-response relationship for horned larks in captivity. Rather than a foliar repellent application to emergent lettuce seedlings under field conditions (York et al., 2000; Cummings et al., 2006), wheat seeds were selected as the test diet based upon our previous observations of seasonal food selection and energetic requirements of horned larks under captive and field conditions. Thus, this experiment involved concentration-response testing among individually-caged horned larks ($N = 54$) offered whole wheat seeds treated with the Avipel® Shield repellent.

On the day subsequent to the pre-test (i.e. test), one bowl (30 g