Notes

Evidence of Songbird Intoxication From Rozol® Application at a Black-Tailed Prairie Dog Colony

Nimish B. Vyas,* Craig S. Hulse, Carol U. Meteyer, Clifford P. Rice

N.B. Vyas, C.S. Hulse
U.S. Geological Survey, Patuxent Wildlife Research Center, Beltsville Lab, BARC-East Building 308, 10300 Baltimore Avenue, Beltsville, Maryland 20705

C.U. Meteyer
U.S. Geological Survey, 12201 Sunrise Valley Drive, Reston, Virginia 20192

C.P. Rice
U.S. Department of Agriculture, Agricultural Research Service, Henry A. Wallace Beltsville Agricultural Research Center-West, Building 001, 10300 Baltimore Avenue, Beltsville, Maryland 20705

Abstract

Concerns about avian poisonings from anticoagulant rodenticides have traditionally focused on secondary poisoning of raptors exposed by feeding on contaminated mammalian prey. However, ground foraging songbirds can be directly poisoned from operational applications of the anticoagulant rodenticide Rozol® (0.005% chlorophacinone, active ingredient) applied as a grain bait, at black-tailed prairie dog *Cynomys ludovicianus* colonies. A dead western meadowlark *Sturnella neglecta* recovered from the study prairie dog colony displayed hemorrhaging in brain and pectoral muscle tissue, and it contained chlorophacinone residue concentrations of 0.59 and 0.49 μg/g (wet weight) in the liver and intestinal contents, respectively. Chlorophacinone residues from two Rozol-colored songbird droppings found at the study colony were 0.09 and 0.46 μg/g (wet weight). The timing of the meadowlark mortality and the occurrence of discolored droppings show that songbird exposure and poisoning can occur weeks after a Rozol application.

Keywords: anticoagulant; chlorophacinone; rodenticide; songbird

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* Corresponding author: nvyas@usgs.gov

Introduction

More than 70 avian species use black-tailed prairie dog *Cynomys ludovicianus* colonies for nesting and foraging throughout the year (Kotliar et al. 1999). Black-tailed prairie dog colonies occupy more than 970,000 ha of prairie grasslands in the United States and extend into southern Saskatchewan, Canada, and into northern Mexico (Golden and Gober 2010; USFWS 2012). Many in the agricultural community consider black-tailed prairie dogs to be major pests (Kotliar et al. 1999), and in 2009, the U.S. Environmental Protection Agency (USEPA) granted full registration to the rodenticide Rozol Prairie Dog Bait® (0.005% active ingredient chlorophacinone, 2-[(*p*-chlorophenyl) phenylacetyl)] 1,3-indandione) for black-tailed prairie dog control (Section 3, USEPA registration number 7173-286) in Colorado, Kansas, Montana, Nebraska, New Mexico, North Dakota, Oklahoma, South Dakota, Texas, and Wyoming (Golden and Gober 2010; USEPA 2010b). Chlorophacinone is a first-generation anticoagulant rodenticide, and its mode of action involves inhibition of vitamin K epoxide reductase, resulting in the

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Figure 1. Rozol application in mid-March 2010 (photo taken 5 days post application) in a black-tailed prairie dog *Cynomys ludovicianus* burrow according to label. On the left of the burrow entrance is a combination of Rozol bait (green) and loose dirt on the ground surface from assumed prairie dog activity, making Rozol available to birds. Orange spray paint marks that a burrow has been treated with Rozol.

disruption of blood clotting, subsequent hemorrhaging, and death (Pelfrene 2001). The Rozol formulation is green-colored chlorophacinone-treated winter wheat *Triticum aestivum*, and it is approved for use from October 1 to March 15 of the following year.

Evidence of the adverse effects of Rozol to free-ranging avian species at black-tailed prairie dog colonies has been limited primarily to raptors; therefore, the concern for birds has traditionally focused on secondary poisonings of raptors from feeding on chlorophacinone-exposed prey (Golden and Gober 2010; USFWS 2012). Little is known about the risks posed to songbirds by this rodenticide. Herein, we provide documentation of exposure and effects to ground-foraging songbirds from operational applications of Rozol at a black-tailed prairie dog colony.

Methods and Results

Rozol exposure and its effects on songbirds were documented at a black-tailed prairie dog colony (approximately 17.3 ha) located on a private pasture approximately 1.3 km south of Vernon, Colorado. Within the colony, the areas used by black-tailed prairie dogs contained low, heavily grazed vegetation with patches of bare ground and taller vegetation; the colony was encircled by taller grasses and herbaceous vegetation. The colony was poisoned with Rozol according to the label by certified pesticide applicators of the Yuma County Pest Control District as part of their ongoing black-tailed prairie dog control program. Rozol was applied to burrows via a metal cup attached to a rigid pole. Approximately 216 kg of Rozol was applied, treating 4,080 black-tailed prairie dog burrows. Each burrow was marked with spray paint, and its GPS coordinates were recorded at time of treatment (see Figure 1). The colony was monitored for 12 d during a 29 d postapplication period in March–April 2010. East–west transects (approximately 3 m apart) were walked through the entire colony on days 1, 2, 4, 5, 12, 13, 24, 25, 26, and 27 postapplication. On days 23 and 29 postapplication, the transects were not followed because of prairie dog trapping in the colony, but the area was still perused on foot for evidence of effects. Each search lasted 2–4 h and was conducted in late afternoon as recommended on the Rozol label. One intact male western meadowlark *Sturnella neglecta* carcass was found in the grazed section of the black-tailed prairie

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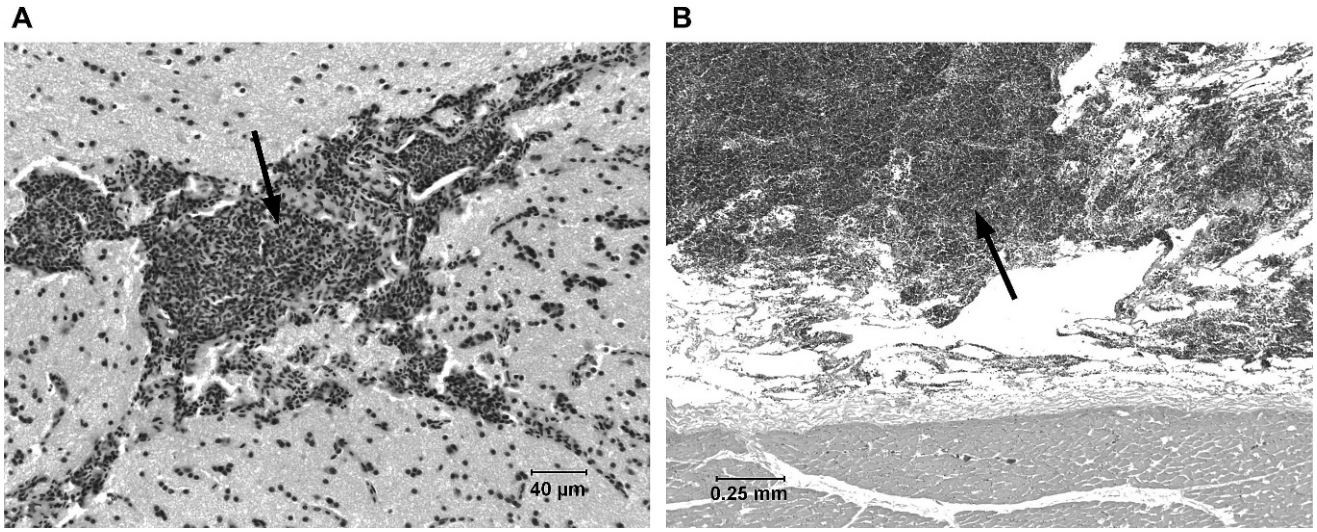


Figure 2. Microscopic hemorrhaging in tissues (arrows) from a western meadowlark *Sturnella neglecta* found dead (25 days post mid-March 2010 application) in a black-tailed prairie dog *Cynomys ludovicianus* colony that was treated with Rozol according to label. (**A**) Brain. (**B**) Pectoral skeletal muscle.

dog colony on day 25 postapplication. The western meadowlark showed no overt signs of trauma or external bleeding. Upon dissection, the bird seemed to have been healthy, with adequate subcutaneous fat, but it displayed signs of anticoagulant rodenticide poisoning, including frank internal bleeding and gross hemorrhaging in pectoral muscles and the brain. The western meadowlark's gizzard was replete with insect parts and did not show Rozol discoloration, but the intestinal contents were colored gray-green. Ten tissue samples including the skeletal muscle, heart, lung, kidney, spleen, brain, liver, and intestine were collected from the western meadowlark and fixed in 10% neutral buffered formalin. Hematoxylin and eosin–stained sections of tissues (American Histolabs Inc., Gaithersburg, MD) were sent to the U.S. Geological Survey National Wildlife Health Center for histopathological examination. The histopathological examination of the tissues revealed abundant microscopic hemorrhaging associated with the pectoral skeletal muscle and one focus of hemorrhage in the brain (Figure 2).

Evidence of songbird exposure to Rozol was derived by chlorophacinone residue analysis of the liver and intestinal contents from the western meadowlark and two Rozol-colored songbird droppings found at the black-tailed prairie dog colony on days 24 and 26 postapplication. The origin of the droppings was deduced based on their size, consistency, location, and the presence of avian species in the colony. Residue analysis was conducted at the U.S. Department of Agriculture, Beltsville Agricultural Research Center, Beltsville, Maryland, and methods followed Albert et al. (2010) as modified by Vyas et al. (2012). The limit of detection was 0.03 µg/g. Recoveries from spiked tissue samples averaged 106%. Chlorophacinone residue concentrations from the western meadowlark liver and intestinal contents were 0.59 and 0.49 µg/g, respectively, and chlorophacinone residue concentrations in the two droppings were 0.09 and 0.46 µg/g, respectively. All chlorophacinone values are presented based on the wet weight of the samples. This study was approved by the Institutional Animal Care and Use Committee of the Patuxent Wildlife Research Center.

Discussion

Mortalities have been reported in a wide variety of avian species that were attracted to seed-treated and pelleted bait formulations of various classes of rodenticides. These classes have included acute (nonanticoagulant) rodenticides such as zinc phosphide and strychnine (Wobeser and Blakley 1987; Apa et al. 1991; Warnock and Schwarzbach 1995; Shivaprasad and Galey 2001; Poppenga et al 2005; Proulx 2011); first-generation anticoagulant rodenticides such as chlorophacinone and diphacinone (Shivaprasad and Galey 2001; Eisemann and Swift 2006; Sánchez-Barbudo et al. 2012); and second-generation anticoagulant rodenticides such as brodifacoum and bromadiolone (Stone et al. 1999; USEPA 2004). Avian risk assessments are based on the rodenticide's toxicity and its availability to birds when used according to the label (USEPA 2010a). Therefore, the rodenticide application methodology is fundamental when calculating the expected exposure that birds will receive. For example, avian risks from rodenticide baits applied on the surface (either broadcasted or scattered around burrow entrance) would be expected for granivorous birds in addition to the risks to raptors from feeding on poisoned rodents (Sánchez-Barbudo et al. 2012). Conversely, risks to granivorous birds would be expected to be reduced if the rodenticide bait were placed down a burrow.

Because the Rozol Prairie Dog Bait label requires that the bait be placed at least 15.24 cm down the burrow (USEPA 2010b), concerns about avian poisonings from Rozol have focused on secondary poisoning of raptors exposed by feeding on contaminated mammalian prey.

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Figure 3. Rozol application in mid-January 2011 (photo taken about 7 days post application) in a black-tailed prairie dog *Cynomys ludovicianus* burrow according to label; however, the shape of the burrow entrance allows Rozol (green) to remain near the ground surface and be available to birds.

However, even when Rozol is applied down a burrow, it can still be available to ground-foraging birds in certain cases: we observed that Rozol can remain close to the ground surface in the burrow entrance because the applicator cannot navigate past the bend in a burrow entrance and that Rozol can be pushed out of the burrow entrance onto the surrounding ground surface by black-tailed prairie dogs (Figures 1 and 3). Our results show that ground foraging songbirds are exposed to and adversely affected by Rozol use at black-tailed prairie dog colonies.

In addition to the dead western meadowlark that we collected at the study site, we observed normal (unexposed to Rozol) and Rozol-colored horned lark droppings (source of droppings identified by the presence of large flocks of horned larks) at burrow entrances at other Rozol-treated colonies but not in untreated colonies (Figure 4). Although we did not witness it directly, we believe that ground-foraging songbirds may ingest the Rozol bait (winter wheat) either from the burrow entrances or from the ground surrounding the entrances. Hepatic residue concentrations are typically used to diagnose the cause of death from anticoagulant rodenticides. The western meadowlark's liver chlorophacinone concentration (0.59 μg/g) was within range of hepatic chlorophacinone levels (0.25–0.69 μg/g) reported from several free-ranging avian species suspected of succumbing from Rozol at black-tailed prairie dog colonies (Ruder et al. 2011; USFWS 2012). Although the chlorophacinone residues in the western meadowlark liver may seem small, it is of biological significance because first-generation anticoagulant rodenticides are designed for low-level multiple feedings over time (Ashton et al. 1986; Rattner et al. 2012; Vyas and Rattner 2012; Vyas et al. 2012).

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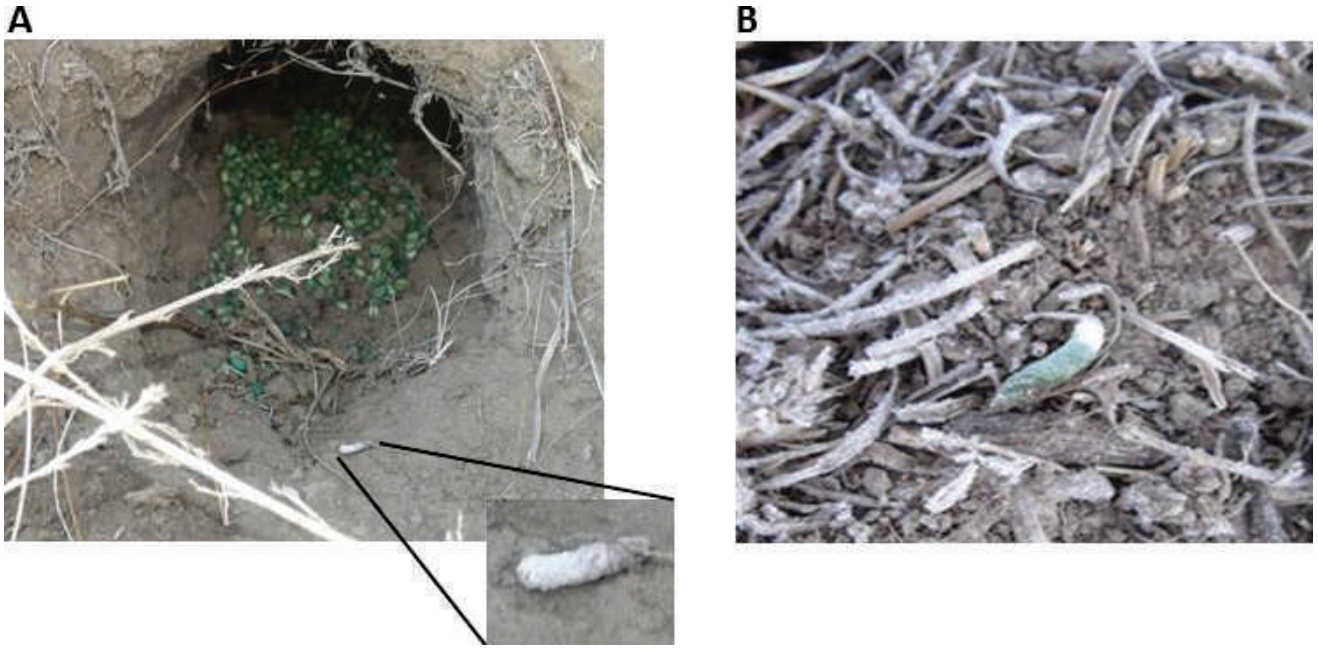


Figure 4. Horned lark *Eremophila alpestris* droppings at a black-tailed prairie dog *Cynomys ludovicianus* burrow that was treated with Rozol (green) according to label in mid-January 2011 (photo taken about 7 days post application). (**A**) Normal (unexposed to Rozol) dropping and (**B**) Rozol-colored dropping. Songbird droppings (normal and Rozol-colored) were seen in and near burrow entrances only at Rozol-treated colonies.

Avian mortality reports from the field confirm the hazards to birds from the operational use of Rozol. The few serendipitous avian carcass recoveries reported in literature (Ruder et al. 2011; USEPA 2010a; USFWS 2012) and the results reported for the dead western meadowlark found in this study raise the possibility of the adverse effects of Rozol on a larger scale. The paucity of incident data could be because Rozol is typically used on privately owned farms, pastures, and ranches where monitoring is not likely to occur. Furthermore, because the dead western meadowlark and the Rozol-colored songbird droppings were discovered on study days 24, 25, and 26 postapplication, it is reasonable to assume that under typical operational conditions, similar incidents would not be detected because carcass searches would not be expected to continue for 3 wk or longer postapplication (USEPA 2012a, 2012b). We believe the paucity of Rozol-related mortalities reported for songbirds is most likely due to insufficient monitoring (Vyas 1999).

Our results have raised a concern for ground foraging songbirds at Rozol-treated black-tailed prairie dog colonies, even when the rodenticide is used according to its label. Additional research and monitoring that integrates avian ecology and chlorophacinone's mode of action and chronic time course of response are needed to better understand the conditions that result in poisonings to mitigate the hazards of Rozol application to free-ranging songbirds.

Supplemental Material

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Reference S1. [USFWS] United States Fish and Wildlife Service. 2012. Final biological opinion for Rozol use on black-tailed prairie dogs registered under Section 3 of the Federal Insecticide, Fungicide and Rodenticide Act.

Found at DOI: http://dx.doi.org/10.3996/052012-JFWM-042.S1; also available: http://www.regulations.gov/#!documentDetail;D = EPA-HQ-OPP-2011-0909-0140 (2.21MB PDF).

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Found at DOI: http://dx.doi.org/10.3996/052012-JFWM-042.S3; also available: http://www.regulations.gov/#!documentDetail;D = EPA-HQ-OPP-2006-0955-0005 (963 KB PDF).

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Reference S5. [USEPA] U.S. Environmental Protection Agency. 2012b. Proposed Rozol Prairie Dog Bait Label.

Found at DOI: http://dx.doi.org/10.3996/052012-JFWM-042.S5; also available: http://www.regulations.gov/#!documentDetail;D = EPA-HQ-OPP-2012-0365-0003. (128 KB PDF).

Reference S6. [USEPA] U.S. Environmental Protection Agency. 2012a. Approval of revised label for Rozol Prairie Dog Bait.

Found at DOI: http://dx.doi.org/10.3996/052012-JFWM-042.S6; also available: http://www.regulations.gov/#!documentDetail;D = EPA-HQ-OPP-2012-0365-0009 (513 KB PDF).

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