U.S. FISH AND WILDLIFE SERVICE
DIVISION OF ENVIRONMENTAL QUALITY

REGION 6

RETENTION TIME OF CHLOROPHACINONE IN THE TISSUES OF BLACK-TAILED PRAIRIE DOGS

EXPOSED TO CHLOROPHACINONE BAIT

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Prepared August, 2011, by:
Matthew S. Schwarz[1]
Joy Gober[2]

[1]U.S. Fish and Wildlife Service
Division of Environmental Quality
South Dakota Field Office
Pierre, South Dakota 57501

[2]US Fish & Wildlife Service
Natural Resource Program Center
1201 Oakridge Drive, Suite 320
Fort Collins, CO 80525

Reviewer's Comments

The current study (Witmer, 2011) was funded by the U.S. Fish and Wildlife Service through the Environmental Contaminants Program as an Environmental Contaminants Investigation. The objectives of this investigation were:

1) To determine chlorophacinone tissue residues in prairie dogs at incremental time periods post-exposure from a limited feeding.

2) To determine potential exposure of predators from consuming prairie dogs killed by chlorophacinone at incremental time periods post-exposure.

The current study provides data on Objective 1: however, the limited exposure of bait to prairie dogs (i.e., short feeding duration and limited amount of bait) is not reflective of actual prairie dog consumption rates when chlorophacinone bait is used in the field as a rodenticide. Therefore, while inferences can be made towards Objective 2, they are limited because the current study was not designed to replicate how prairie dogs forage on poisoned bait in the field. We recognize that to examine the issue of secondary poisoning of predators and scavengers from consumption of poisoned prairie dogs, it was necessary to start with a controlled exposure experiment that could provide levels of chlorophacinone in various prairie dog tissues at specific time intervals post exposure. However in doing so, study Objective 2 was not fully satisfied because the limited feeding duration in the current study underestimates exposure that may occur from repeated small daily doses consumed during an actual field application of chlorophacinone to control prairie dogs. Prairie dogs that feed on chlorophacinone bait over several days following a field application would likely have higher tissue residues than those in the current study. Secondary exposure risk to prairie dog predators would also be expected to occur for a longer duration and at higher uptake levels than the current study results suggest.

Chlorophacinone bait availability is expected to be greater after a field application to control prairie dogs than in the current study. In the current study, prairie dogs in group T1 were provided bait for two consecutive days (53 grams total) and excess bait was removed. Prairie dogs were then either euthanized at predetermined time intervals or maintained on a clean diet for up to 25 days. For field applications, the Rozol label recommends a dose of 53 grams per active burrow and there are on average 3.9 active burrow-entrances per black-tailed prairie dog (Biggins et al., 1993). Inactive burrows may also be mistakenly baited resulting in further bait availability. Furthermore, there is

no effort to remove excess bait or provide clean food during field applications and thus prairie dogs may continue to consume bait after they have accumulated a lethal dose. As noted on page 35 of the report, it has been observed that “animals continue to feed on the baits for several days, then become lethargic and eventually stop feeding." Lee and Hygnstrom (2007) reported finding prairie dogs carcasses 10 - 25 days after field applications.

Observations during the current study also indicate that prairie dogs were exposed to less bait than what would typically occur during a field application. For example, only 3 of 36 poisoned prairie dogs died and only 9 showed signs of being poisoned. The current study also reported that only 56 percent of T1 animals showed evidence of hemorrhaging. Although first generation indandiones (i.e., chlorophacinone and diphacinone) can cause mortality without showing hemorrhaging in mammals, the low number of dead and morbid prairie dogs in the current study indicates a lower dose was received than reported elsewhere for field applications. For example, Lee and Hygnstrom (2007) reported prairie dog population reductions between 85 – 96 percent following field applications of chlorophacinone.

Previous reports (Lee and Hygnstrom, 2007; Primus, 2007) that include chlorophacinone residues in prairie dog tissues after a field application also suggest that prairie dogs in the current study consumed less bait than what would likely occur under a field application. The current study reported a mean concentration of chlorophacinone in liver of 0.82 ± 0.70 micrograms per gram (µg/g) in prairie dogs euthanized 11 days after bait (0.005% chlorophacinone) was first presented. In comparison, eight prairie dog carcasses collected 10 to 25 days after a field application of similar bait had a mean concentration of chlorophacinone in liver of 2.19 ± 1.80 µg/g (Lee and Hygnstrom, 2007; Primus, 2007). Eight additional prairie dog carcasses collected from other non-experimental field sites (dates of application unknown) reported a mean concentration of chlorophacinone in liver of 5.86 ± 1.88 (Primus, 2007). Residues in live rodents are expected to be greater than in their carcasses, especially for relatively short-lived, chronic rodenticides such as diphacinone and chlorophacinone as the rodent may continue to consume bait above a lethal dose and close to the onset of morbidity. The longer the lag time (between exposure and death), the more time is available for the target rodent to continue consuming bait. Therefore, chlorophacinone residues in live prairie dogs could be substantially higher than those in carcasses, but further study is needed to predict residue levels in live prairie dogs that have undergone multiple feedings of poisoned bait.

Repeated small daily doses of anticoagulants are also known to result in higher toxicity than a single acute dose (Godfrey et al., 1981; Jackson and Ashton, 1992). For example, the median lethal dose (LD50) from a single exposure of chlorophacinone to Norway rats

(*Rattus norvegicus*) is 20.5 milligrams per kilograms (mg/kg) whereas a 5-day daily dose LD50 is twenty times lower at 0.95 mg/kg (Jackson and Ashton, 1992). We expect that the difference in toxicity between a single acute dose and repeated daily doses would also apply to non-target species that feed on prairie dogs. Additionally, the extended period of 1 to 4 weeks between application of poisoned bait and observed deaths of prairie dogs (Vyas, 2010, Lee and Hygnstrom, 2007) extends the time during which prairie dogs are available to non-target predatory animals.

Results from the current study indicate that tissue concentrations of chlorophacinone were highly variable among individuals necropsied on the same day (not evident in Figures 3 and 4 but note standard deviations in Table 1). Silberhorn and others (2003) also reported large variations in residues in individual ground squirrel carcasses even for those squirrels that died on the same day. All the prairie dogs in the current study were exposed at the same time so the variability may be due to individual differences in the amount of bait consumed, size, and metabolism and excretion of chlorophacinone. Due to variability in individual residue loads, it is recommended that future assessments include at least 10 individuals collected on the same day post application.

Toxicity to chlorophacinone among individuals also appeared to be highly variable and not necessarily related to the amount of bait consumed. Observational data on prairie dog response after consuming the maximum amount of bait (53 grams) ranged from appearing normal to severe incapacitation and death. Animal health logs indicate that the three prairie dogs found dead had appeared normal during prior observation checks and only one showed signs of external bleeding. Variability in the susceptibility of target species to chlorophacinone toxicity may also apply to non-target species and should be further evaluated.

Although chlorophacinone residues in the current study likely underestimate exposure from field applications, dead and euthanized prairie dogs still had residue levels that were high enough to warrant concern for secondary exposure to non-target birds or mammals. Non-target species that feed on prairie dogs would be exposed to chlorophacinone in whole-body and liver tissues. In the current study, prairie dogs had concentrations of chlorophacinone in whole-body and liver that ranged from 0.053 - 1.78 µg/g and 0.061 - 8.407 µg/g, respectively. Liver chlorophacinone concentrations exceeded those previously reported for common voles (0.082 - 3.800 µg/g) but secondary exposure risk to vole predators was not evaluated (Vidal et al., 2009). Whole-body concentrations in prairie dogs from the current study were also greater than those estimated in laboratory rats (0.18 – 0.81 µg/g) that resulted in the death of 11 of 20 domestic ferrets when fed upon for five consecutive days (Ahmed et al., 1996 as cited by Erickson and Urban, 2004).

Observations from the current study indicate that chlorophacinone is metabolized over time so that death can occur after the parent compound is nearly gone. Prairie dogs that lived the longest but eventually were euthanized based on condition had liver chlorophacinone levels similar to "trace amounts" reported for wildlife mortality incident investigations. For example, two prairie dogs (KQ-18 and KQ-26) ate 36 and 53 grams of bait during the first two days, respectively. They were then euthanized due to their condition on days 22 and 26 and had liver concentrations of chlorophacinone of 0.265 and 0.187 µg/g, respectively. These concentrations are similar to those previously reported as "trace" amounts of chlorophacinone (e.g., 0.25 µg/g) in wild raptors opportunistically found. For example, a bald eagle found near a chlorophacinone poisoned prairie dog town in Nebraska had a liver concentration of 0.3 µg/g chlorophacinone and forensic necropsy results indicated that the eagle died from chlorophacinone ingestion (USFWS, 2007). Chlorophacinone was also detected at 0.18 µg/g in a red-tailed hawk from New York State (Stone et al., 2003). Other raptors, for which chlorophacinone exposure may have contributed to death, include a ferruginous hawk and great-horned owl, both collected from Kansas with “trace amounts” of 0.25 µg/g chlorophacinone (USFWS, 2009).

The current study indicates that lethargy can persist in poisoned prairie dogs for several days before they die or need to be euthanized based on morbidity. Despite only a single exposure to rozol bait, many of the prairie dogs suffered from delayed incapacitation. For example, prairie dog KQ26 was lethargic for 16 days starting 10 days after exposure and was euthanized on Day 26 due to poor condition. Incapacitation in these animals occurred despite receiving a clean maintenance diet post exposure to rozol. Prolonged lethargy would likely result in increased susceptibility to predation and these same prairie dogs that are more easily captured by predators may present the highest risk of secondary exposure if they continue to eat chlorophacinone after receiving a lethal dose and thus accumulate higher tissue residues.

The current study included a T2 group of three prairie dogs that were provided chlorophacinone bait *ad libitum* for two days. Valid conclusions cannot be made from this T2 group. The small sample size of this group (n = 3) and high variability in both the amount of bait consumed per individual (i.e., range of 7.0 - 54.6) and tissue concentrations (see Table 2B) preclude statistical analysis. The prairie dog that consumed only 7.0 g of bait had the lowest tissue concentrations of chlorophacinone in the T2 group and ingested much less bait than any other prairie dog in either treatment group (the next lowest was 27.5 g of bait consumed), leading to the question of whether some other factor was affecting this test animal. Furthermore, the time period of two days for *ad libitum* exposure is less than what would be expected in a field application.

We do not agree with the current study conclusions that "the highest risk of secondary exposure to chlorophacinone residues by non-target animals consuming prairie dogs exposed to the bait would occur within a few days after bait application and would drop quickly thereafter." As specified above, the current study is not representative of prairie dog exposure to chlorophacinone from a typical field application and prairie dogs that continue to consume bait after they have accumulated a lethal dose may have the highest chlorophacinone tissue residues. This would result in risk of secondary exposure to non-target animals over a more extended time period.

The current study also suggests that "because birds are less susceptible to chlorophacinone poisoning than mammals, secondary risks are probably higher for predatory or scavenging mammals (coyotes) than for predatory birds" and based this conclusion on a review by Primus and others (2001). The risk assessment by Primus and others (2001) did not evaluate sub-lethal effects leading to indirect mortality, which is our greatest concern regarding avian consumption of chlorophacinone poisoned prairie dogs. Sub-lethal effects have been documented in raptors exposed to anticoagulants and can occur despite low tissue residue concentrations. For example, American kestrels (*Falco sparverius*) administered diphacinone and with liver residues just above the diphacinone method detection limits of 0.263 and 0.280 µg/g diphacinone had histological evidence of hemorrhage in lung and liver (Rattner et al., 2011a). Golden eagles (*Aquila chrysaetos*) fed muscle from diphacinone-treated sheep exhibited extreme weakness, hemorrhages, and ataxia (Savarie et al., 1979). These studies indicate that raptors are susceptible to indandione's multiple modes of action which include both the blocking of prothrombin formation and the uncoupling of oxidative phosphorylation (Van Den Berg and Nauta, 1974). Ample evidence exists to indicate that avian predators and scavengers are susceptible to secondary toxicity risks and additional study is needed to further evaluate the issue.

Management Recommendations

More data that are representative of field conditions are needed to adequately evaluate Objectives 1 and 2. We recommend a more robust assessment of chlorophacinone residues in prairie dogs that mimics operational application exposures of chlorophacinone bait. The assessment is needed to determine residues in prairie dogs that receive repeated small doses of chlorophacinone and should include at least 10 individuals that are euthanized as soon as they exhibit signs of lethargy or morbidity.

Studies indicate that avian lethality tests required to register first generation indandione rodenticides can result in toxicity values that ultimately underestimate risk and that new test requirements are needed. Standardized tests for avian lethality that are required by the U.S. Environmental Protection Agency (USEPA) to support pesticide registration include the single-dose acute oral toxicity test and the five-day sub-acute dietary toxicity test that are used to derive an LD50 and median lethal concentration (LC50), respectively (USEPA, 2007). First generation indandione rodenticides have a mode of action that results in cumulative effects over several days of feeding, thus the required single-dose acute oral toxicity test tends to result in large LD50s values that ultimately underestimate risk (Ashton et al., 1986; Jackson and Ashton, 1992). The standardized five-day sub-acute dietary toxicity test includes multiple exposures over several days but has little value as a quantitative descriptor of lethal toxicity and is more of a measure of vulnerability to a contaminated diet, with results that can be highly dependent on a species willingness to eat the bait and ability to cope with reduced nutriment (Hill, 1993; Mineau et al., 1994; Hoffman, 2003). Studies that do not follow required methodologies for registration but provide supplemental information, such as the previously mentioned five-day sub-acute oral toxicity tests (Godfrey et al., 1981; Jackson and Ashton, 1992), indicate that a repeated low dose oral sub-acute toxicity test for anticoagulant rodenticides can result in a more toxic LD50 than a single-dose acute oral test. Likewise, a dietary toxicity test that measured the diphacinone-treated diet consumed daily by Eastern screech-owls (*Megascops asio*) found that repeated low-dosage exposure over seven days increased diphacinone toxicity by more than an order of magnitude compared to an acute oral toxicity test (Rattner et al., 2011b; N. Vyas pers comm.). These studies indicate a need to change current required avian oral and dietary lethality tests for first generation indandione rodenticides to include multiple day low-dose exposures that measures individual daily dosage. Factors associated with extrapolating laboratory derived risk quotients to the field can further underestimate risk (Matz et al, 1998; Vyas et al., 2006), and this may be especially true when considering the sub-lethal effects from first generation indandione rodenticides. Thus, methods for the lethality tests should also be expanded to include observational periods for sub-lethal effects and protocols that include gross pathology and histopathological examination of tissues to evaluate internal hemorrhaging. The USEPA has the responsibility and authority under the Federal Insecticide, Fungicide, and Rodenticide Act to determine the potential of a pesticide to cause adverse effects and require further testing when needed (USEPA, 2007). We recommend that USEPA develop new standardized testing requirements for first generation indandione rodenticides and require additional long term field studies to allow for a more adequate determination of whether continued registration approval is warranted for use of first generation indandione rodenticides to control prairie dogs.

Active surveillance is needed to further examine the extent of non-target mortalities from the use of anticoagulant rodenticides to control prairie dogs. Lee and Hygnstrom (2007) included searchers for non-target carcasses on and immediately around baited plots while performing field assessments designed to assess the efficacy of chlorophacinone and did not report any indications that avian non-targets were adversely affected from feeding on poisoned prairie dogs. However, recovery of poisoned raptors from baited prairie dog downs is expected to be highly unlikely given the chronic nature of chlorophacinone that allows wide ranging birds to move away from the site of application.

Additional assessments of secondary risks to avian species from exposure to chlorophacinone are needed and should consider interspecific differences in exposure and susceptibility. Although there is a paucity of sub-lethal threshold effects data following repeated exposure for birds of prey to chlorophacinone; a few studies of diphacinone toxicity to raptors (Savarie et al., 1979; Mendenhall and Pank, 1980; Rattner et al., 2011a) indicate that they may be especially sensitive to anticoagulants. Acute diphacinone toxicity tests indicate that American kestrels are over 20 times more sensitive than Northern bobwhite (*Colinus virginianus*), and over 30 times more sensitive than mallards (*Anas platyrhynchos*), two test species required by USEPA for pesticide registration (Rattner et al., 2010 and 2011a). Furthermore, golden eagles appear to be even more sensitive to diphacinone than kestrels (Savarie et al., 1979; Rattner et al., 2011a). Mendenhall and Pank (1980) observed differences in diphacinone toxicity between great-horned owls and barn owls and suggested that explanations for such a discrepancy may include interspecific differences in susceptibility or differences in prey species that result in dissimilar exposure. These studies indicate that future assessments on the effects of chlorophacinone on avian species that consume prairie dogs should include multiple species. Ferruginous hawks may be especially susceptible to anticoagulant use on prairie dogs as they are a primary predator of prairie dogs and have been frequently reported near prairie dog towns poisoned with anticoagulants. In 2010, Audubon of Kansas reported finding the remains of 17 dead hawks in 2009 following anticoagulant use to control prairie dogs in the area. Unfortunately, these carcasses were not recovered for necropsy or chemical analysis.

Based on sub-lethal effects to non-target species as reported from laboratory studies as well as reported mortalities and concerns based on opportunistic recoveries (Littrell, 1990; Ruder et al., 2008), there is clearly a need for field studies that evaluate anticoagulant exposure and effects to the many species that may consume poisoned prairie dogs. Littrell (1990) ranked exposure to diphacinone/chlorophacinone second only to strychnine as the most hazardous vertebrate pesticide to non-targets based on his 10 years of experience in reviewing vertebrate pesticides. Ruder and others (2008)

reported three mortality events involving several species, including wild turkeys (*Meleagris gallopavo*), a raccoon (*Procyon lotor*), and an American badger (*Taxidea taxus*) after a chlorophacinone application to control black-tailed prairie dogs in Kansas. The authors concluded that their opportunistic findings of non-target mortalities likely underestimate actual non-target losses and warrant further investigation. This conclusion seems justified as a four year survey of possible anticoagulant poisonings of wildlife in France that was based on a wildlife disease surveillance network yielded 59 confirmed diagnoses for bromadiolone and 41 for chlorophacinone (Berny et al., 1997 as cited by Stone et al., 1999). A similar surveillance network is needed to evaluate non-targets after anticoagulant use to control prairie dogs in the United States, especially given all of the avian predators that key in on and consume prairie dogs including golden eagles, northern goshawks, northern harriers, peregrine falcons, prairie falcons, Cooper's hawks, ferruginous hawks and red-tailed hawks. A few laboratory studies indicate that some of these species survive after being fed anticoagulant poisoned rodents, at least until time of necropsy (Savarie et al., 1979; Mendenhall and Pank, 1980; Radvanyi et al., 1988). However, the sub-lethal effects described in these studies (e.g., fatigue, wing-dropping, and lung, heart and liver hematomas) are likely to result in decreased survival or reproduction and need to be evaluated under field conditions.

Chlorophacinone is slow acting and non-target species are often highly mobile, thus tracking individual birds via radio or satellite telemetry is needed to evaluate secondary toxicity to avian predators that consume chlorophacinone poisoned prairie dogs. Avian carcasses are also quickly scavenged in the wild (Vyas, 1999) and tracking may aid in recovering intact carcasses for necropsy and residue analyses. Tracking studies also have the added benefit of allowing for evaluation in the field where animals also are exposed to other stressors and can also help assess potential sub-lethal effects that can include decreased survival and reproduction.

Conclusions

The current study provided some information on chlorophacinone residue concentrations over time and observed effects from toxicity; however, the limited exposure of chlorophacinone in the current study underestimates what would occur during a field application. Chlorophacinone loss from metabolism and excretion indicates that prairie dogs that continue to consume bait over several days will likely have higher chlorophacinone residues than prairie dogs that are found dead or euthanized after several days of being too sick to eat. Field observations and results from previous studies indicate that field applications would likely result in higher prairie dog residue burdens

than indicated in the current study and risk of secondary exposure to prairie dogs predators could last for weeks after application. Predators that consume prairie dogs would also have an increased risk to secondary toxicity from repeated small doses above what is inferred from a single reference dose.

Study results indicate that injury from exposure to chlorophacinone may not be related to concentrations of chlorophacinone in whole-body or liver tissues as measured at the time of death. Chlorophacinone is ingested, results in internal and sometimes external hemorrhaging and is then metabolized and excreted. Internal bleeding in non-targets from repeated exposure to chlorophacinone likely results in sub-lethal effects that contribute to increased mortality without resulting in high concentrations in tissue. Thus even low concentrations of chlorophacinone detected in poisoned carcasses may be indicative of cause of death, either directly or indirectly, and should be considered with other biological evidence in determining whether harmful exposure to anticoagulants occurred.

This study was an acute dietary toxicity test that provided some useful information, as previously noted. However, chlorophacinone lethality increases with multiple low-dose feedings and prairie dogs exposed to chlorophacinone bait under field conditions can live for several weeks before death occurs. Therefore, further study is needed to adequately evaluate Objectives 1 and 2. We recommend a more robust assessment of chlorophacinone residues in prairie dogs that are exposed to repeated low doses of chlorophacinone and are immediately euthanized after they exhibit signs of morbidity. Previous studies indicate that raptors are likely more sensitive to the first generation indandione rodenticides than traditional avian test species and further evaluation of threshold effects from repeated daily doses are needed. Lastly, we recommend that future field assessments incorporate tracking techniques to evaluate decreased survival and reproduction for multiple avian species.

References

Ahmed MS, Daroch J, Carlet L, Whaley D. 1996. Secondary hazard study using chlorophacinone-killed laboratory rats fed to domestic ferrets (*Mustela putorius furo*).
Unpublished report submitted to EPA by LiphaTech, Inc., Milwaukee, WI. 84 pp.

Ashton AD, Jackson WB, Peters H. 1986. Comparative evaluation of LD50 values for various anticoagulant rodenticides. *Tropical Pest Management* 32:187-197.

Berny PJ, Buronfosse T, Buronfosse F, Lamarque F, Lorgue G. 1997. Field evidence of secondary poisoning of foxes (*Vulpes vulpes*)and buzzards (*Buteo buteo*) by bromadiolone, a four-year survey. *Chemosphere* 35:1817-1829.

Biggins DE, Miller BJ, Hanebury L, Oakleaf R, Farmer A, Crete R, Dood A. 1993. A technique for evaluating black-footed ferret habitat. In: Oldemeyer J, Biggins DE, Miller BJ, and Crete R, (eds), Management of prairie dog complexes for reintroduction of the black-footed ferret, pp 73-88. USFWS, Biological Report 134 Washington, D.C.

Erickson W, Urban D. 2004. Potential risks of nine rodenticides to birds and nontarget mammals: A comparative approach. Office of Pesticides Programs Environmental Fate and Effects Division. U.S. Environmental Protection Agency, Washington, DC. 230 pp.

Godfrey MER, Reid TC, McAllum HJF. 1981. The oral toxicity of brodifacoum to rabbits. *New Zealand Journal of Experimental Agriculture* 9:23-25.

Hill EF. 1993. Acute and subacute toxicology in evaluation of pesticide hazard to avian wildlife In: Kendall RJ and Lacher TE, (eds), Wildlife Toxicology and Population Modeling, pp 45-67. Lewis Publishers, Boca Raton, FL.

Hoffman DJ. 2003. Wildlife toxicity testing. In: Hoffman DJ, Rattner BA, Burton GA Jr, Cairns J Jr, (eds), Handbook of Ecotoxicology, pp 75-110. Lewis Publishers, Boca Raton, FL, USA.

Jackson WB, Ashton AD. 1992. A review of available anticoagulants and their use in the United States. *Proceedings of the Fifteenth Vertebrate Pest Conference* 15:156-160.

Lee CD, Hygnstrom SE. 2007. Field efficacy and hazards of rozol bait for controlling black-tailed prairie dogs (*Cynomys ludovicianus*). Laboratory Project Identification 06076. Kansas State University Research and Extension, Manhattan KS and University of Nebraska, Lincoln, NE. 56 pp.

Littrell EE. 1990. Effects of field vertebrate pest control on nontarget wildlife (with emphasis on bird and rodent control). In: Davis LR and Marsh RE, (eds), Proceedings of the 14th Vertebrate Pest Conference, pp 58-61. University of California. http://digitalcommons.unl.edu/vpc14/55

Matz AG, Bennett RS, Landis WG. 1998. Effects of azinphos-methyl on Northern bobwhite: A comparison of laboratory and field results. *Environmental Toxicology and Chemistry* 17:1364-1370.

Mendenhall VM, Pank LF. 1980. Secondary poisoning of owls by anticoagulant rodenticides. *Wildlife Society Bulletin* 8:311-315.

Mineau P, Jobin B, Baril A. 1994. A critique of the avian 5-day dietary test (LC50) as the basis of avian risk assessment. Canadian Wildlife Service Technical Report Series 215. Canadian Wildlife Service, Environment Canada, Ottawa, ON, CA.

Primus T, Eisemann J, Matschke G, Ramey C, Johnston J. 2001. Chlorophacinone residues in rangeland rodents: as assessment of the potential risk of secondary toxicity to scavengers. Pp. 164-180 In Johnston J, (ed), Pesticides and Wildlife. American Chemical Society Symposium Series 177. American Chemical Society, Washington, D.C.

Primus TM. 2007. Determination of chlorophacinone residues in prairie dog whole body and liver tissues. Project Number QA-1405. U.S. Department of Agriculture National Wildlife Research Center. Fort Collins, CO. 58 pp.

Radvanyi A, Weaver P, Massari C, Bird D, Broughton E. 1988. Effects of chlorophacinone on captive kestrels. *Bulletin of Environmental Contamination and Toxicology* 41:441-448.

Rattner BA, Horak KE, Warner SE, Day DD, Johnston JJ. 2010. Comparative toxicity of diphacinone to northern bobwhite (*Colinus virginianus*) and American kestrels (*Falco sparverius*). Proceedings, 24th Vertebrate Pest Conference, Sacramento, CA, USA, February 22- 25, 2010, pp 145-152.

Rattner BA, Horak KE, Warner SE, Day DD, Meteyer CU, Volker SV, Eisemann JD, Johnston JJ. 2011a. Acute toxicity, histopathology, and coagulopathy in American kestrels (*Falco sparverius*) following administration of the rodenticide diphacinone. *Environmental Toxicology and Chemistry* 30(5):1213-1222.

Rattner BA, Horak KE, Eisenreich KM, Lazarus RS, Eisemann JD, Johnston JJ. 2011b. Comparative toxicity and risk evaluation of the anticoagulant rodenticide diphacinone in various species of birds. Society of Environmental Toxicology and Chemistry Europe 21st Annual Meeting. Abstract 311 ET10B-2. May 15-19, 2011, Milan, Italy.

Ruder MG, Poppenga RH, Bryan II JA, Bain M, Pitman J, Keel MK. 2008. Intoxication of nontarget wildlife with rodenticides in Northwestern Kansas. *Journal of Wildlife Diseases* 47(1):212-216.

Savarie PJ, Hayes DJ, McBride RT, Roberts JD. 1979. Efficacy and safety of diphacinone as a predacide. In Kenaga EE, (ed), Avian and Mammalian Wildlife Toxicology, pp 69-79. STP 693 American Society for Testing Materials, Philadelphia, PA.

Silberhorn EM, Schnabel DL, Salmon TP. 2003. Ecological risk assessment for grain-based field-use anticoagulant rodenticides registered by the California Department of Food and Agriculture for special local needs. California Department of Food and Agriculture, Sacramento, CA. 109 pp.

Stone WB, Okoniewski JC, Stedelin JR. 1999. Poisoning of wildlife with anticoagulant rodenticides in New York. *Journal of Wildlife Diseases* 35:187-93.

Stone WB, Okoniewski JC, Stedelin JR. 2003. Anticoagulant rodenticides and raptors: recent findings from New York, 1998-2001. *Bulletin of Environmental Contamination and Toxicology* 70:34–40.

U.S. Environmental Protection Agency (USEPA. 2007. Pesticides; data requirements for conventional chemicals. Federal Register 72(207): 60934-60988. http://edocket.access.gpo.gov/2007/pdf/E7-20826.pdf

U.S. Fish and Wildlife Service (USFWS). 2007. Veterinary medical examination report. Lab Case # 07-000015. National Fish and Wildlife Forensics Laboratory, Ashland, Oregon. 6 pp.

U.S. Fish and Wildlife Service (USFWS). 2009. Veterinary medical examination report. Lab Case # 09-000049. National Fish and Wildlife Forensics Laboratory, Ashland, Oregon. 11 pp.

Van Den Berg G, Nauta WT. 1975. Effects of anti-inflammatory 2-aryl-1,3-indandiones on oxidative phosphorylation in rat liver mitochondria. *Biochemical Pharmacology* 24(7):815-821.

Vidal D, Alzaga V, Luque-Larena J, Mateo R, Arroyo L, Vinuela J. 2009. Possible interaction between a rodenticide treatment and a pathogen in common vole (*Microtus arvalis*) during a population peak. *Science of the Total Environment* 408:267-271.

Vyas NB. 1999. Factors influencing estimation of pesticide-related wildlife mortality. *Toxicology and Industrial Health* 15:186-191.

Vyas NB, Spann JW, Hulse CS, Borges SL, Bennett RS, Torrez M, Williams BI, Leffel R. 2006. Field evaluation of an avian risk assessment model. *Environmental Toxicology and Chemistry* 25: 1762-71.

Vyas NB. 2010. Annual Report 2010: Characterization of avian hazards following chlorophacinone (Rozol®) use for prairie dog control. U.S. Geological Survey, Patuxent Wildlife Research Center. 39 pp.

Witmer G. 2011. Retention time of chlorophacinone in the tissues of black-tailed prairie dogs exposed to chlorophacinone bait. Final Report: QA-1682. USDA/APHIS/WS National Wildlife Research Center, Fort Collins, CO. 59 pp.