

Field evaluation of imidacloprid as a systemic approach to flea control in black-tailed prairie dogs, *Cynomys ludovicianus*

David S. Jachowski¹✉ Sherry Skipper² and Matthew E. Gompper³

¹U.S. Fish and Wildlife Service, South Dakota Ecological Services Field Office, Pierre, SD 57501, U.S.A.

²U.S. Fish and Wildlife Service, Rocky Mountain Arsenal National Wildlife Refuge, Commerce City, CO 80022, U.S.A.

³Department of Fisheries and Wildlife Sciences, University of Missouri, Columbia, MO 65211, U.S.A.

Received 31 August 2010; Accepted 30 December 2010

ABSTRACT: Epizootic outbreaks of sylvatic plague have dramatically influenced prairie dog (*Cynomys sp.*) populations across North America. While a great deal of debate surrounds the cause and persistence of plague, flea control can stop the spread of plague epizootic outbreaks and even increase prairie dog survival under non-epizootic conditions. We investigated a newly-developed imidacloprid-treated grain bait that could potentially reduce flea infestations and mitigate the effects of plague on black-tailed prairie dogs (*C. ludovicianus*). We used a study design involving randomly assigned experimental and control study plots to assess the effectiveness of the systemic flea control product. We observed a significant difference in flea prevalence and abundance between experimental and control sites on three of the four sites treated with a single application of imidacloprid-treated grain bait for up to 90 days post-treatment. We observed an even greater reduction in flea infestations following the double application of treatment bait on two of three additional experimental sites. While we were unable to reduce flea infestations to the extent reported for more commonly used topical insecticides containing deltamethrin, imidacloprid might still be effective at reducing the risk of plague and halting epizootics. In addition, this systemic product can be more rapidly applied than topical insecticides, providing managers with a tool to quickly reduce flea infestations. Future research is needed to evaluate the effectiveness of different application timing and rates, the utility of the product in limiting plague, and the potential effects on non-target species that might also consume the treated bait. *Journal of Vector Ecology* 36 (1): 100-107. 2011.

Keyword Index: Sylvatic plague, imidacloprid, deltamethrin.

INTRODUCTION

Epizootic outbreaks of sylvatic plague have dramatically influenced prairie dog (*Cynomys sp.*) populations across North America (Biggins and Kosoy 2001, Antolin et al. 2002). Prairie dogs have disappeared from 90 to 95% of their former range (Forrest 2005) and all five species in the genus are listed or have been petitioned for listing under the Endangered Species Act. Sylvatic plague (hereafter referred to as plague), caused by the bacterium *Yersinia pestis*, is typically a flea-borne zoonotic disease characterized by epizootic outbreaks with periodic rapid transmission of *Y. pestis* between hosts resulting in nearly complete host population collapses, interspersed with enzootic periods during which *Y. pestis* persists at low or non-detectable levels in prairie dog communities (Biggins et al. 2010). Plague was introduced to the west coast of North America in 1900 and has continued to spread east (Adjemian et al. 2007), where epizootics have resulted in 85 to 100% declines in impacted prairie dog populations (Rayor 1985, Ubico et al. 1988, Pauli et al. 2006). In addition, once plague has been introduced to an area, epizootics are likely to reoccur at five to ten year intervals (Barnes 1982). Thus, plague is dramatically reducing prairie dog populations and hindering prairie dog conservation efforts. Declines in prairie dog populations also pose a threat to species dependent on prairie dogs, especially the critically endangered black-footed ferret

(*Mustela nigripes*) (Miller et al. 1994, Matchett et al. 2010).

Fleas are the primary vectors of plague, and large increases in flea abundance have been observed during epizootics (Anderson and Williams 1997, Pauli et al. 2006, Tripp et al. 2009). Flea control can stop the spread of plague epizootics (Seery et al. 2001, Hoogland et al. 2004) and even increase prairie dog survival under non-epizootic conditions (Biggins et al. 2010). Currently, the most widely used technique to mitigate the risk of epizootics is the application of the topical flea control pesticide DeltaDust® (Bayer Environmental Science, Research Triangle Park, NC), which contains deltamethrin powder (0.05% by volume) as the active ingredient. Application of DeltaDust (hereafter referred to as deltamethrin) is expensive and labor-intensive at large scales, costing up to \$68.91 per ha (Greibel 2009). In addition, deltamethrin is not a flea-specific insecticide and has lethal and sub-lethal effects on other arthropods (Croft 1990, Wardhaugh, et al. 1998, Desneux et al. 2007) and vertebrates (Alexander et al. 2002), which could have cascading effects on insect communities and insectivorous species.

We investigated a newly-developed, imidacloprid-treated grain bait that could potentially reduce flea loads and mitigate plague risk to prairie dogs. A systemic flea control insecticide has been developed that is composed of an oat grain bait treated with the active ingredient imidacloprid (0.025% by volume) and marketed as Kaput®

(Genesis Laboratories, Wellington, CO). Imidacloprid is a neonicotinoid compound that interferes with the nerve conduction system of insects. When an animal consumes the treated bait (hereafter referred to as imidacloprid), the active ingredient resides in the blood stream and within 24 to 48 h kills ectoparasites that ingest blood from a treated animal (Borchert et al. 2009, Poché et al. 2010). In a controlled captive setting, this systemic product was shown to kill 96–97.3% of fleas on treated prairie dogs for up to 30 days post-treatment (R. Poché, Genesis Laboratories, unpublished data). The objective of our study was to evaluate the efficacy of this imidacloprid product for controlling fleas on black-tailed prairie dogs for up to 90 days in a field setting.

MATERIALS AND METHODS

Study site

Our study site was the U.S. Fish and Wildlife Service managed Rocky Mountain Arsenal National Wildlife Refuge (RMA), located in Denver County northwest of the city of Denver, CO, U.S.A. The RMA contained a large prairie dog population consisting of over 30 distinct colonies that occupied a total area of 431 ha. A variety of topical chemical treatments for flea control in prairie dog colonies have been previously investigated at this site, including products containing permethrin (Beard et al. 1992) as well as pyriproxyfen and pyreperm. More recently, a test of topical application of a flea control powder containing deltamethrin showed that it significantly decreased flea infestations to the point that treated prairie dog populations were protected from plague epizootics for up to seven months post-treatment (Seery et al. 2003). Deltamethrin has a short persistence time in soils (Hill 1983) and no treatments had been applied in our study site for at least three years prior to our study; thus, we assumed there were no residual effects of previous insecticide treatments at our study site.

Experimental design

We used a completely randomized experimental design involving randomly assigned experimental and control study plots (Skalski and Robson 1992) to assess the effectiveness of imidacloprid. In May 2009, we established four experimental plots (E1, 2, 3, and 4) and four control study plots (C1, 2, 3, and 4) that were at least 3 ha in size (Figure 1). We attempted to select entire colonies (i.e., distinct aggregations of family groups with adjoining burrow systems; Hoogland 1995) or groups of colonies as study plots to avoid situations where prairie dogs move onto, away from, or between plots. We paired study plots based on colony size, vegetation type and cover, soils, and proximity, and then randomly assigned one of each pair as the control and the other as the experimental plot. In July 2009, we added three more experimental plots (E5, E6, and E7) that were paired with existing control plots (C1, C3, and C4, respectively) to evaluate a second round of imidacloprid application (Figure 1). Experimental and control plots were at least 250 m apart, with two exceptions: E2 and C2 were ~100 m apart and separated by a 30-cm-tall vinyl barrier that likely limited movement between plots (Witmer et al. 2008), and E7 and C4 were ~50 m apart and separated by a forested buffer strip with tall vegetation that we assumed impeded movement between plots during our period of study (Garrett and Franklin 1988, Northcott et al. 2008).

The null hypothesis that we tested was that there was no difference in mean flea prevalence, abundance, or aggregation on prairie dogs inhabiting experimental and control plots. We also investigated the potential influence of finer-scale differences in the effectiveness of imidacloprid at the individual prairie dog level. The distribution of fleas on black-tailed prairie dog populations tends to be aggregated, with some hosts more likely than others to be infested with fleas (Brinkerhoff et al. 2006). Therefore, we tested the null hypotheses that there was no difference in flea infestation between experimental and control plots based on host sex, age, or body condition.

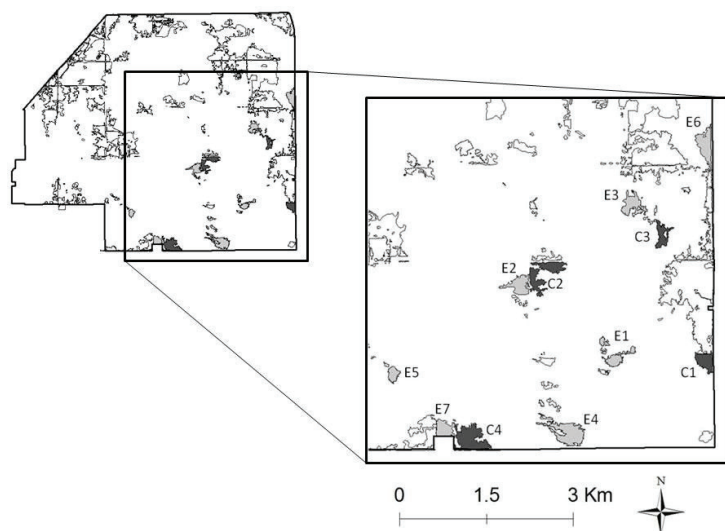


Figure 1. Study plots on the Rocky Mountain Arsenal National Wildlife Refuge, CO, with the spatial extent of black-tailed prairie dog (*Cynomys ludovicianus*) colonies in 2009 depicted with light solid lines and the boundary of the Arsenal depicted by a dark solid line. Seven experimental (abbreviated as “E” above and in grey) and four control (abbreviated as “C” above and in black) study plots were used in testing the effectiveness of imidacloprid flea control bait.

Bait application

Beginning in May, 2009, we applied imidacloprid-treated grain bait (prepared by Genesis Laboratories) to experimental plots. Previous work showed that uptake of 5 g of treated bait by an individual prairie dog resulted in approximately 96% flea control (T. Linder, Genesis Laboratories, personal communication). We followed label guidelines by scattering 56 g of grain bait within 2.4 m of each prairie dog burrow opening in each experimental plot. The size of the area and density of burrows differed between plots, but on average we applied 20.6 kg of treated grain bait per ha (sd=8.0, range 11.3 to 31.3). In July-August, we treated the three additional experimental plots (E5, E6, and E7) to evaluate the effect of double applications, where we applied 56 g on day 1 and reapplied 56 g on day 5. For all treatments, we dyed grain bait red to allow us to identify untreated burrows easily while applying the bait and to monitor bait uptake visually during daily visits to experimental plots (Figure 2).

Forage availability during the time of treatment appeared to be limited, and bait was readily consumed by prairie dogs. Herbaceous cover dominated both experimental and control plots, primarily consisting of field bindweed (*Convolvulus arvensis*) and Kochia (*Kochia scoparia*). Grass cover, primarily western wheatgrass (*Agropyron smithii*), was low across all plots. During daily revisits to experimental plots to check for bait uptake, we observed that all bait typically was eaten within one to three days post-treatment.

Sampling of fleas from prairie dogs

From June through September, 2009, we trapped prairie dogs on all plots at 30-day intervals post-treatment, with the goal of capturing 50 unique prairie dogs per plot during each five-day trapping session. At 06:30 each day, we set Tomahawk live traps (15.2 cm x 15.2 cm x 60 cm) at the openings of 80 active burrows on each plot. All traps were checked and closed at 09:30. We sampled three of the four single-application plots at 30, 60, and 90 days post-treatment. A single pairing (plots E1 and C1) was

only sampled at 30 days post-release due to field logistic issues. All double-application experimental plots were only sampled at 30 days post-treatment.

Upon capture we transferred each prairie dog to a specially designed induction chamber linked to an isoflurane vaporizer. While the individual was sedated, we used a flea comb (nine to ten teeth per cm) to dislodge fleas, combing the entire body of the prairie dog for 30 s (Biggins et al. 2010). We counted the number of fleas dislodged but did not identify individual fleas to species. Previous work in and near Rocky Mountain Arsenal identified *Oropsylla hirsuta* as the dominant flea species on *C. ludovicianus*, with *Thrassis fatus*, *Pulex simulans*, and *O. tuberculata* occurring in small numbers (Seery et al. 2003, Tripp et al. 2009). We recorded the sex, age class, hind foot length, and weight of each prairie dog. We classified prairie dogs as either juvenile (young that emerged from the natal burrow within the past eight months; Hoogland 1995) or adult based on size, pelage, reproductive status, and body condition. Prior to releasing each prairie dog at its capture location, we attached a uniquely numbered ear tag in both of its ears to avoid resampling.

Data analysis

We assessed the effectiveness of the treatment in two ways. First, we computed an effectiveness index developed by Abbott (1925): $\text{Efficacy} = 100 \times (a-b)/a$, where a is the mean abundance of fleas on prairie dogs on the control plot and b is the mean abundance of fleas on prairie dogs on the experimental plot. Second, we quantified flea prevalence (percent of examined animals infested by fleas), abundance (number of fleas observed on an individual host), and intensity (number of fleas observed on an infested host) on prairie dogs in each experimental and control plot during each monthly trapping session (Bush et al. 1997). We compared prevalence, mean abundance, and mean intensity of flea infestations on prairie dogs between paired experimental and control plots during each trapping interval using a Bootstrap 2-sample t-test. We computed an index of aggregation, k , to examine the extent to which flea infestation rates had an aggregated negative binomial distribution using the program, Quantitative Parasitology 3.0 (Reiczigel and Rózsa 2005). A k value >20 approximates a Poisson distribution, and as k approaches 0 the negative binomial distribution converges to the logarithmic distribution (Bliss and Fisher 1953, Shaw and Dobson 1995). We used linear regression to assess the potential relationship between prairie dog body condition and flea abundance on experimental and control plots. We estimated body condition for each captured prairie dog based on the relationship between weight (g) of an individual and length of its hind foot (cm) (Wirsing et al. 2002, Pauli et al. 2006). We regressed hind-foot length and weight of each prairie dog, fit a linear regression line, and calculated the residual distance of each prairie dog to the fitted line. We used the residual distance as a metric for body condition. We also assessed if differences in flea intensity on prairie dogs from experimental and control plots occurred due to sex or age



Figure 2. Black-tailed prairie dogs (*Cynomys ludovicianus*) consuming red dyed oat bait treated with imidacloprid insecticide in May, 2009.

class using one-way ANOVA.

RESULTS

We captured and sampled fleas from 1,151 prairie dogs on experimental and control plots (Tables 1 and 2). The number of prairie dogs sampled per plot ranged from 41 to 58, with a mean of 52.7. We captured an approximately equal ratio of male:female (male mean=26.7, female mean=25.9; paired t-test $p=0.59$) and adult:juvenile (adult mean=29.1, juvenile mean=23.2; paired t-test $p=0.15$) prairie dogs on each of our study plots.

We observed a significant difference in flea prevalence and abundance on prairie dogs between experimental and control plots on three of the four paired plots where treatment consisted of a single application of imidacloprid-treated grain bait. In the first paired plots (E1 and C1), our efficacy index was low (-103.9) and we observed similar prevalence (71.9% and 71.2%), but greater flea abundance (experimental mean=6.28, 95% CI=3.62; control mean=3.08, 95% CI=1.20) and intensity (experimental mean=8.73, 95% CI=5.05; control mean=4.41, 95% CI=1.56) on E1 compared to C1 30 days post-treatment. On all other paired plots we found that the product was more effective and observed decreased flea prevalence on experimental plots (Table 1) as well as significant differences in abundance at 30, 60, and 90 days post-treatment (boot-strap 2 sample t-test, $p < 0.02$) (Figure 3). We observed a significant difference ($p < 0.05$) in mean flea intensity between experimental and control on the second paired plots (E2 and C2) for up to 60 days and on the fourth paired plots (E4 and C4) for up to 90 days; however, we did not detect a significant difference ($p > 0.08$) in intensity between experimental and control on the third paired plots (E3 and C3) (Figure 3). Collectively, on three of the four experimental plots where we observed an effect of treatment, our findings indicate that flea abundance and intensity declined for at least 90 days following treatment (Figure 3).

We observed an even greater reduction in flea infestations following the double application of treatment bait on two of the three experimental plots. We observed significant differences in flea abundance and intensity ($p < 0.01$) on prairie dogs on plots E5 and E7 when compared with the corresponding control plots, C1 and C4 (Table 2). In these two double-treatments, we observed lower abundance, and intensity of fleas on prairie dogs at 30 days post-treatment than in single-treatment plots (Tables 1 and 2).

Flea infestation primarily occurred on a few individuals and was even more highly aggregated on experimental plots for up to 90 days post-treatment (Tables 1 and 2). However, there were no differences in which prairie dogs were infested by fleas between either control or experimental plots when the populations were subdivided by host sex, or age class, nor were there relationships between host body condition and flea abundance in experimental or control plots. For prairie dogs infested by fleas, body condition did not explain variation in the number of fleas on prairie dogs on experimental ($r^2=0.0032$, $df=677$, $F=2.18$, $P=0.1407$) or control ($r^2=0.0005$, $df=565$, $F=0.2761$, $P=0.5995$) plots. Adults typically had more fleas than juveniles on both experimental (adult mean=3.68, 95% CI=0.71, $n=367$; juvenile mean=1.97, 95% CI=0.52, $n=314$; $F=7.20$, $P=0.0001$) and control plots (adult mean=8.16, 95% CI=1.19, $n=331$; juvenile mean=4.82, 95% CI=0.74, $n=243$; $F=14.77$, $P=0.0001$). Male and female prairie dogs were similarly infested on both experimental (female =2.53, 95% CI=0.57, $n=333$; male mean=3.27, 95% CI=0.72, $n=359$; $F=1.50$, $P=0.2235$) and control (female mean=6.80, 95% CI=1.16, $n=288$; male mean=6.70, 95% CI=1.00, $n=283$; $F=0.10$, $P=0.7456$) plots.

DISCUSSION

The imidacloprid systemic flea control product generally depressed flea infestations for at least 90 days in

Table 1. Summary of efficacy index, flea prevalence, and aggregation (k) on prairie dogs (*Cynomys ludovicianus*) sampled 30 (June), 60 (July), and 90 (August) days post-treatment on paired, simultaneously sampled, control (C) plots, and on experimental (E) plots receiving a single treatment of imidacloprid grain bait.

Plot	June				July				August			
	Prairie dogs sampled	Prevalence	k	Efficacy index	Prairie dogs sampled	Prevalence	k	Efficacy Index	Prairie dogs sampled	Prevalence	k	Efficacy index
E2	52	78.8%	0.874	45.2	53	58.5%	0.880	74.6	50	72.0%	0.558	48.8
C2	41	87.8%	1.127		54	90.7%	1.431		52	88.5%	0.921	
E3	57	61.4%	0.566	55.4	51	52.9%	0.421	58.6	53	50.9%	0.447	47.0
C3	54	90.7%	1.093		54	83.3%	1.105		58	82.8%	1.368	
E4	50	88.0%	0.739	62.7	56	75.0%	0.634	56.7	55	52.7%	0.337	67.7
C4	53	94.3%	0.926		51	94.1%	1.635		53	84.9%	0.806	

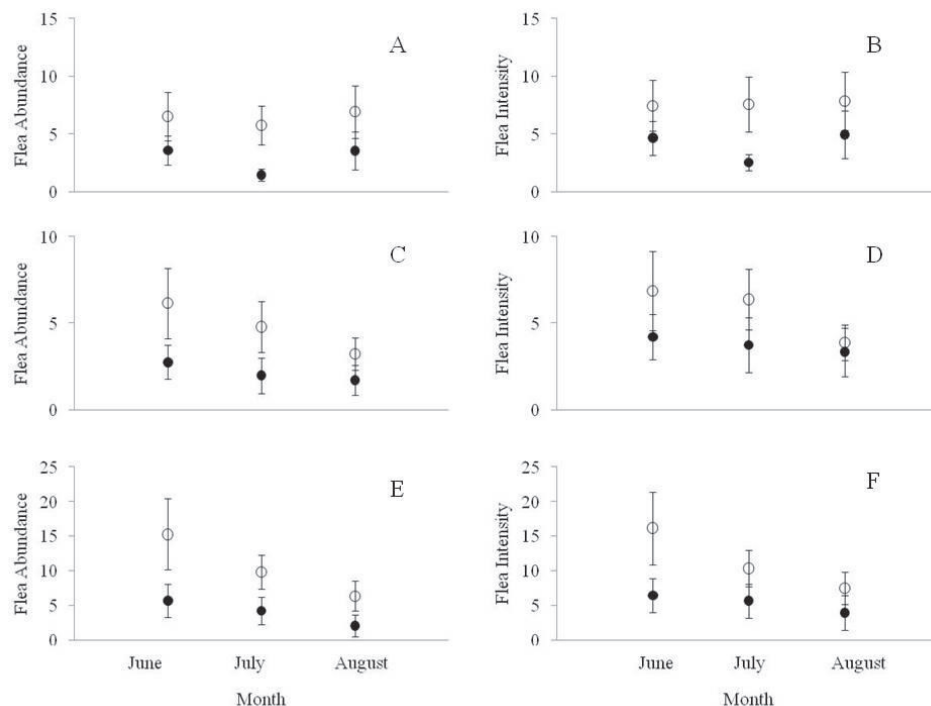


Figure 3. Mean (\pm 95% confidence intervals) flea abundance and intensity on prairie dogs (*Cynomys ludovicianus*) sampled 30, 60, and 90 days following a single treatment of imidacloprid bait on experimental (filled circles) and control (open circles) pairing 2 (A and B), pairing 3 (C and D), and pairing 4 (E and F).

Table 2. Summary of efficacy index, flea prevalence, abundance, intensity, and aggregation (k) on prairie dogs (*Cynomys ludovicianus*) 30-days post-treatment (August) on paired, simultaneously sampled, control (C) plots, and experimental (E) plots receiving double treatment of imidacloprid grain bait. Abundance and intensity values are reported as means with 95% confidence intervals.

Plot	Prairie dogs sampled	Prevalence	Abundance	Intensity	k	Efficacy index
E5	49	34.7%	0.78 (0.45, 1.35)	2.23 (1.29, 3.18)	0.368	83.5
C1	46	71.7%	4.72 (3.35, 6.87)	6.57 (4.45, 8.70)	0.673	
E6	53	71.7%	3.55 (2.23, 5.70)	4.94 (2.71, 7.18)	0.501	-10.6
C3	58	82.8%	3.21 (2.47, 4.26)	3.87 (2.87, 4.88)	1.368	
E7	56	35.7%	0.64 (0.39, 1.11)	1.80 (1.09, 2.50)	0.573	89.9
C4	53	84.9%	6.34 (4.75, 9.04)	7.47 (5.18, 9.75)	0.806	

a field setting. Laboratory tests show that imidacloprid remains in the blood-stream of prairie dogs fed imidacloprid-treated grain baits and kills fleas that bite prairie dogs for at least 30 days (R. Poché, Genesis Laboratories, unpublished data). Given that we observed a significant difference in flea infestation on prairie dogs at 90 days post-treatment, the product likely remains effective in the animal for a longer period of time or the reduction of fleas during the initial 30-day period delays the redevelopment of flea infestations. The more aggregated distribution of flea infestation on prairie dogs post-treatment also suggests that the product generally

was effective. On control plots, flea infestations were highly aggregated on a few individuals as found in previous investigations of black-tailed prairie dogs (Brinkerhoff et al. 2006). We would expect a similar aggregation index on experimental plots if the product generally was ineffective. However, following treatment, flea infestations were even more highly aggregated on a small number of individuals.

It is unclear why we did not detect a decline in flea infestations post-application for one single-application and one double-application experimental plot. Bait from the same batch/shipment was applied on all single-treatment

plots, and a second batch of treated bait was used for all double-treatment plots using an identical bait formulation. We assume that consumption of treated bait by prairie dogs on these two plots was similar to that on all other treated plots because: (1) the dominant vegetative cover was similar to other experimental plots and primarily consisted of *C. arvensis* and *K. scoparia*, both of which are less desired by prairie dogs in comparison to grass species such as *A. smithii* (Koford 1958), which was uncommon on all plots, and (2) all treated grain was eaten within one to three days of application (Figure 2). Thus, further evaluations are required to determine if and why the product might not consistently reduce flea infestations.

It was possible to apply imidacloprid more quickly than the more commonly used topical flea pesticides, which reduced personnel costs. Application of the topical insecticide deltamethrin, the product used most commonly to control flea infestations on prairie dogs, requires injection using a pressurized applicator of four to six g of powder into every burrow in a prairie dog colony (Beard et al. 1992, Seery et al. 2003, Biggins et al. 2010). Application of deltamethrin at a large scale using trained employees or contractors costs \$50.06/ha with an application rate of 0.9 h/ha using all-terrain vehicles (Greibel 2009). In comparison, based our study, imidacloprid application costs approximately \$77.09/ha with an application rate of 0.8 h/ha by walking. If all-terrain vehicles were used to apply imidacloprid, application costs could likely be further reduced. The rapidity with which imidacloprid can be applied could be an asset to managers who want to reduce flea infestations quickly, such as during an epizootic. In addition, since labor involved in applying flea control products typically is the most costly factor (71% of the total treatment cost; Greibel 2009), imidacloprid could be a more cost effective flea management tool than deltamethrin if the rate of application were increased and the cost of the product were reduced.

Neither a single nor double application of imidacloprid reduced flea infestations to the levels of the topical insecticide deltamethrin. In an evaluation of the application of the topical flea powder deltamethrin to black-tailed prairie dog burrow openings, Seery et al. (2003) reported a decline in flea prevalence on *C. ludovicianus* to ~16% by 30 days post-treatment and to “nondetectable levels” by day 84. Similarly, in a longer-term evaluation of the effects of deltamethrin on multiple prairie dog species, Biggins et al. (2010) observed a 96-98% reduction in fleas on prairie dogs at sites treated with deltamethrin at one month post-treatment and a 45-86% reduction at ten months post-treatment. We were able to reduce flea prevalence to a low of only 50.9% for a single application. Results from evaluations of the imidacloprid product on California ground squirrels (*Spermophilus beecheyi*) suggest that the application of larger quantities at more regular intervals could achieve higher reductions (>95%) in flea infestations (Borchert et al. 2009). The reduction in flea infestations we observed following a double-treatment (prevalence decreased to a low of 34.7%) suggests that additional applications could also

further reduce flea infestations. In addition, imidacloprid is likely to be more effective at decreasing flea infestations and plague risk when its application is timed to match spring and fall peaks in abundance of flea species (Wilder et al. 2008, Eisen et al. 2009, Tripp et al. 2009).

While we were unable to reduce flea infestations with imidacloprid to the level observed by others following treatment with deltamethrin, the product still might be effective at reducing the risk of plague and halting epizootics. Although a one-time annual application of imidacloprid is unlikely to result in short and long-term reductions of flea infestations as great as those produced by annual application of deltamethrin, short-term reductions might be sufficient to minimize immediate plague risks. Deltamethrin has been shown to be effective at stopping the spread of epizootic plague in prairie dog populations (Seery et al. 2003, Greibel 2009). Lorange et al. (2005) hypothesize that *Xenopsylla cheopis* infrapopulation sizes of >4.7 individuals per host are required to maintain the plague-causing *Y. pestis* infection in a population at an enzootic level, and that >9.4 individuals per host are required to support an epizootic. Eisen et al. (2006) suggested that densities of *O. montana* of 5.4 and 10.8 per host were required to maintain *Y. pestis* at enzootic and epizootic levels, respectively. We found that on the sites where treatments were effective, flea abundance stayed below enzootic thresholds for at least 90 days post-treatment. This suggests that imidacloprid can effectively limit the occurrence of plague at epizootic and enzootic levels. Because these hypothesized thresholds are based on laboratory experiments, further research is required to validate them in a field setting with prairie dogs.

Future evaluations of systemic flea control products in this system should also sample fleas that occur off-host or on other potential host species, and identify the species of flea collected. If sampling is limited to fleas on prairie dogs, it might not fully reflect flea dynamics, given that fleas spend a large portion of lives off-host (Krasnov 2008). The sampling of other potential host species and identification of fleas collected to species could also help in identifying the effect of this product on the larger flea community in this system. For example, the flea *Pulex simulans* is common to eastern Colorado and is a generalist that feeds on multiple hosts (Tripp et al. 2009). If this flea species were feeding on a non-prairie dog host, it could remain abundant in the study area and be a source of plague infection to prairie dogs.

We still have a poor understanding of the toxicity of imidacloprid on non-target species. We expect that prairie dogs consumed a majority of the bait we applied, because we applied a relatively small amount near burrow openings, observed prairie dogs consuming the bait, and visually confirmed that all bait was eaten within one to three days after application. However, this does not rule out the potential for detrimental effects on non-target birds and mammals although the product has a low oral toxicity (Tomlin 2000). Similar to previous field trials of this product (see Borchert et al. 2009), we did not observe injured or dead non-target species on or in the vicinity of our study site during the course of this study, but future studies should

more thoroughly evaluate treatment effects on non-target species. It should be noted, however, that deltamethrin also has potentially detrimental effects on non-target species (Alexander et al. 2002). A systemic pesticide, such as used in this study, is likely to be less harmful to non-target insects than topical insecticides. Effects of current wide-scale use of topical insecticides on invertebrate communities in prairie dogs colonies are poorly understood. There might also be indirect effects on species of conservation concern such as mountain plovers (*Charadrius montanus*) and burrowing owls (*Athene cunicularia*), which prefer to nest on prairie dog colonies and have offspring that forage on invertebrates near nest sites (Knopf 1996, Restani et al. 2001). Thus, managers need to weigh the costs and benefits of both flea control approaches when considering the effects of large-scale applications.

Acknowledgments

We thank Al Pfister, Henry Maddux, Pete Gober, and Scott Larson of the U.S. Fish and Wildlife Service for their support of this research project. We also thank the staff of the Rocky Mountain Arsenal National Wildlife Refuge for their support of field efforts. Dean Biggins and Joshua Millspaugh provided helpful guidance for designing and implementing the project. Richard Jachowski and Bruce Hastings provided helpful comments on this manuscript. Funding was provided by the U.S. Fish and Wildlife Service. Any use of trade, product, or firm names is for descriptive purposes only and does not imply endorsement by the U.S. Government. The findings and conclusions in this article are those of the authors and do not necessarily represent the views of the U.S. Fish and Wildlife Service.

REFERENCES CITED

- Abbott, W.S. 1925. A method of computing the effectiveness of an insecticide. *J. Econ. Entomol.* 18: 265-267.
- Adjemian, J.Z., P. Foley, K.L. Gage, and J.E. Foley. 2007. Initiation and spread of traveling waves of plague, *Yersinia pestis*, in the western United States. *Am. J. Trop. Med. Hyg.* 76: 365-375.
- Alexander, G.J., D. Horne, and S.A. Hanrahan. 2002. An evaluation of the effects of deltamethrin on two non-target species in the Karoo, South Africa. *J. Arid Environ.* 50: 121-133.
- Anderson, S.H. and E.S. Williams. 1997. Plague in a complex of white-tailed prairie dogs and associated small mammals in Wyoming. *J. Wild. Dis.* 33: 720-732.
- Antolin, M.F., P. Gober, B. Luce, D.E. Biggins, W.E. Van Pelt, D.B. Seery, M. Lockhart, and M. Ball. 2002. The influence of sylvatic plague on North American wildlife at the landscape level, with special emphasis on black-footed ferret and prairie dog conservation. *Trans. N. Am. Wild. Natl. Res. Conf.* 67: 104-127.
- Barnes, A.M. 1982. Surveillance and control of plague in the United States. *Sym. Zool. Soc. Lond.* 50: 237-270.
- Beard, M.L., S.T. Rose, A. Barnes, and J. Montenieri. 1992. Control of *Oropsylla hirsute*, a plague vector, by treatment of prairie dog burrows with 0.5% permethrin dust. *J. Med. Entomol.* 29: 25-29.
- Biggins, D.E. and M.Y. Kosoy. 2001. Influences of introduced plague on North American mammals: implications from ecology of plague in Asia. *J. Mammal.* 82: 906-916.
- Biggins, D.E., J.L. Godbey, K.L. Gage, L.G. Carter, and J.A. Montenieri. 2010. Vector control improves survival of three species of prairie dogs (*Cynomys*) in areas considered enzootic for plague. *Vector-borne Zoon. Dis.* 10: 17-26.
- Bliss, C.I. and Fisher, R.A. 1953. Fitting negative binomial distribution to biological data. *Biometrics* 9: 176-200.
- Borchert, J.N., R.M. Davis, and R.M. Poché. 2009. Field efficacy of rodent bait containing the systemic insecticide imidacloprid against the fleas of California ground squirrels. *J. Vector Ecol.* 34: 92-98.
- Brinkerhoff, R.J., A.B. Markeson, J.H. Knouft, K.L. Gage, and J.A. Montenieri. 2006. Abundance patterns of two *Oropsylla* (*Ceratophyllidae: Siphonaptera*) species on black-tailed prairie dog (*Cynomys ludovicianus*) hosts. *J. Vector Ecol.* 31: 355-363.
- Bush, A.O., K.D. Lafferty, J.M. Lotz, and A.W. Shostak. 1997. Parasitology meets ecology on its own terms: Margolis et al. revisited. *J. Parasitol.* 83: 575-583.
- Croft, B.A. 1990. *Arthropod Biological Control Agents and Pesticides*. Wiley Press, New York. 723 pp.
- Desneux, N., A. Decourtye, and J. Delpuech. 2007. The sublethal effects of pesticides on beneficial arthropods. *Annu. Rev. Entomol.* 52: 81-106.
- Eisen, R.J., S.W. Bearden, A.P. Wilder, J.A. Montenieri, M.F. Antolin, and K.L. Gage. 2006. Early-phase transmission of *Yersinia pestis* by unblocked fleas as a mechanism explaining rapidly spreading plague epizootics. *Proc. Natl. Acad. Sci.* 103: 15380-15385.
- Eisen, R.J., L. Eisen, and K.L. Gage. 2009. Studies of vector competency and efficiency of North American flea for *Yersinia pestis*: State of the field and future research needs. *J. Med. Entomol.* 46: 737-744.
- Forrest, S. 2005. Getting the story right: A response to Vermeire and colleagues. *Bioscience* 55: 526-530.
- Garrett, M.G. and W.L. Franklin. 1988. Behavioral ecology of dispersal in the black-tailed prairie dog. *J. Mammal.* 69: 236-250.
- Greibel, R.L. 2009. *Wall Ranger District 2009 Plague Management Report*. Nebraska National Forest, Buffalo Gap National Grassland, Wall Ranger District, Wall, SD. 13 pp.
- Hill, B.D. 1983. Persistence of deltamethrin in Lethbridge sandy clay loam. *J. Envir. Sci. Hlth.* 18: 691-703.
- Hoogland, J.L. 1995. *The Black-Tailed Prairie Dog: Social Life of a Burrowing Mammal*. Univ. Chicago Press, Chicago, IL. 557 pp.
- Hoogland, J.L., S. Davis, S. Benson-Amram, D. Labruna, B. Goosens, and M.A. Hoogland. 2004. Pyreperm kills fleas and halts plague among Utah prairie dogs. *Southwestern Nat.* 49: 376-383.

- Knopf, F.L. 1996. Mountain plover (*Charadrius montanus*). In: A. Poole and F. Gills (eds.) *The Birds of North America*, No. 211. The Academy of Natural Sciences, Philadelphia, Pennsylvania, and the American Ornithologists' Union, Washington, D.C. 16 pp.
- Koford, C.B. 1958. *Prairie Dogs, Whitefaces, and Blue Gramma*. Wildlife Monographs No. 2., 78 pp.
- Krasnov, B.R. 2008. *Functional and Evolutionary Ecology of Fleas: a Model for Ecological Parasitology*. Cambridge Univ. Press, Cambridge, UK.
- Lorange, E.A., B.L. Race, F. Sebbane, and B.J. Hinnebusch. 2005. Poor vector competence of fleas and the evolution of hypervirulence in *Yersinia pestis*. *J. Infect. Dis.* 191: 1970-1912.
- Matchett, M.R., D.E. Biggins, V. Carlson, B. Powell, and T. Roche. 2010. Enzootic plague reduces black-footed ferret (*Mustela nigripes*) survival in Montana. *Vector-borne Zoon. Dis.* 10: 27-35.
- Miller, B., G. Ceballos, and R. Reading. 1994. The prairie dog and biotic diversity. *Conserv. Biol.* 8: 677-681.
- Northcott, J., M.C. Anderson, G.W. Roemer, E.L. Fredrickson, M. DeMers, J. Truett, and P.L. Ford. 2008. Spatial analysis of effects of mowing and burning on colony expansion in reintroduced black-tailed prairie dogs (*Cynomys ludovicianus*). *Restoration Ecol.* 16: 495-502.
- Pauli, J.N., S.W. Buskirk, E.S. Williams, and W.H. Edwards. 2006. A plague epizootic in the black-tailed prairie dog (*Cynomys ludovicianus*). *J. Wildl. Dis.* 42: 74-80.
- Poché, R.M., J.N. Borchert, and J. Bruenning. 2010. Efficacy of systemic insecticide against fleas on Wyoming ground squirrels (*Spermophilus elegans*) and prairie dogs (*Cynomys ludovicianus*). *Vector-borne Zoon. Dis.* 10: 97-98.
- Rayor, L.S. 1985. Dynamics of a plague outbreak in Gunnison's prairie dog. *J. Mammal.* 66: 194-196.
- Reiczigel J. and L. Rózsa. 2005. Quantitative Parasitology 3.0. <http://www.zoologia.hu/qp/qp.html>
- Restani, M., L.R. Rau, and D.L. Flath. 2001. Nesting ecology of burrowing owls occupying black-tailed prairie dog towns in southeastern Montana. *J. Raptor Res.* 35: 296-303.
- Seery, D.B., D.E. Biggins, J.A. Montenieri, R.E. Enscoe, D.T. Tanda, and K.L. Gage. 2003. Treatment of black-tailed prairie dog burrows with deltamethrin to control fleas (Insecta: *Siphonaptera*) and plague. *J. Med. Entomol.* 40: 718-722.
- Shaw, D.J. and A.P. Dobson. 1995. Patterns of macroparasite aggregation in wildlife host populations. *Parasitology* 117: 597-610.
- Skalski, J. R. and D. S. Robson. 1992. *Techniques for Wildlife Investigations: Design and Analysis of Capture Data*. Academic Press, San Diego, CA. 237 pp.
- Tomlin, C.D.S. 2000. *The Pesticide Manual*. British Crop Protection Council, Farnham, U.K., 1,200 pp.
- Tripp, D.W., K.L. Gage, J.A. Montenieri, and M.F. Antolin. 2009. Flea abundance on black-tailed prairie dogs (*Cynomys ludovicianus*) increases during plague epizootics. *Vector-borne Zoon. Dis.* 9: 313-321.
- Ubico, S.R., G.O. Maupin, K.A. Fagerstone, and R.G. McLean. 1988. A plague epizootic in the white-tailed prairie dogs (*Cynomys leucurus*) of Meeteetse, Wyoming. *J. Wildl. Dis.* 24: 399-406.
- Wardhaugh, K.G., B.C. Longstaff, and M.J. 1998. Effects of residues of deltamethrin in cattle faeces on the development and survival of three species of dung-breeding insect. *Aust. Vet. J.* 76: 273-280.
- Wilder, A.P., R.J. Eisen, S.W. Bearden, J.A. Montenieri, D.W. Tripp, R.J. Brinkerhoff, K.L. Gage, and M.F. Antolin. 2008. Transmission efficiency of two flea species (*Oropsylla tuberculata cynomuris* and *Oropsylla hirsuta*) involved in plague epizootics among prairie dogs. *EcoHealth* 5: 205-212.
- Wirsing, A.J., T.D. Steury, and D.L. Murray. 2002. Relationship between body condition and vulnerability to predation in red squirrels and snowshoe hares. *J. Mammal.* 83: 707-715.
- Witmer, G., J. Gionfriddo, and M. Pipas. 2008. Evaluation of physical barriers to prevent prairie dog colony expansion. *Human-Wildl. Conf.* 2: 206-211.