

Tools and Technology

Mitigating Plague Risk in Utah Prairie Dogs: Evaluation of a Systemic Flea-Control Product

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ABSTRACT Plague, the disease caused by the bacterium *Yersinia pestis*, is a major threat to the Utah prairie dog (*Cynomys parvidens*), a species listed as threatened under the U.S. Endangered Species Act. Fleas are the primary vectors of plague, and flea control can stop the spread of plague epizootics and increase Utah prairie dog survival. We evaluated a newly developed grain-bait insecticide treated with the active ingredient imidacloprid. In 2009, we conducted a single application of the product in treatment plots within each of 4 study sites and sampled fleas from captured Utah prairie dogs on treatment and control plots at monthly intervals. We observed mixed results; the product generally was effective at reducing flea prevalence, abundance, and intensity on prairie dogs at some sites and not at others, and the effectiveness within a site varied over time. In 2010, we doubled the amount of bait on treatment plots, yet we still failed to observe a consistent decline in flea prevalence, abundance, and intensity on prairie dogs. At the application rates we evaluated, the imidacloprid product is likely not as effective at controlling fleas on Utah prairie dogs as the more commonly used topical insecticide containing deltamethrin. However, managers should also consider the risk of flea species developing resistance following the repeated application of a single flea-control product. Furthermore, because we observed a higher than expected diversity of flea species (8) on Utah prairie dogs, future work should be undertaken to investigate how other mammalian host species might mediate flea population dynamics, plague ecology, and the outcome of flea management approaches. © 2012 The Wildlife Society.

KEY WORDS *Cynomys parvidens*, flea control, imidacloprid, *Oropsylla*, plague, Utah.

Plague, caused by the bacterium *Yersinia pestis*, has historically been associated with major population declines in humans and other mammal species. Plague is believed to have originated in Asia, and is well-known because of 3 major human pandemics that have taken over 200 million lives (Perry and Fetherston 1997). Since the last pandemic in the 1800s, plague has spread to previously uninfected areas, including portions of Africa and the Americas (Gage and Kosoy 2005). In North America, plague is causing large reductions in the populations of several native rodent species, and, in particular, prairie dog (*Cynomys* sp.) populations (Gage et al. 1995, Biggins and Kosoy 2001, Antolin et al.

2002). Plague is a major threat to the Utah prairie dog (*C. parvidens*), which is listed as a threatened species under the U.S. Endangered Species Act. The Utah prairie dog is perhaps the rarest of all 5 species of prairie dog, occurring in relatively small, highly fragmented populations that exhibit relatively low gene flow and high genetic divergence (Brown et al. 2009). Recent findings suggest that plague is likely present in both epizootic and enzootic forms throughout the range of the Utah prairie dog (Biggins et al. 2010), and is a persistent threat to the recovery of the species (U.S. Fish and Wildlife Service 2009).

Fleas are the primary vectors of plague, and flea control can inhibit spread of epizootic plague (Seery et al. 2003, Hoogland et al. 2004). Further, flea control has been shown to increase individual survival in 3 prairie dog species (*C. ludovicianus*, *C. leucurus*, and *C. parvidens*) under non-epizootic conditions (Biggins et al. 2010). Currently, the most widely used pesticide for flea control is deltamethrin, which requires 4–6 g of powder to be injected into burrows within a prairie dog colony (Seery et al. 2003). Given the

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large extent and number of burrows on a prairie dog colony, application of deltamethrin can be costly and labor-intensive (Griebel 2009). Therefore, there is interest in finding means of controlling fleas in prairie dog populations that are more cost-effective.

Recently, a grain bait treated with the active ingredient imidacloprid (0.025% by vol) and marketed as Kaput[®] (Genesis Laboratories, Wellington, CO) has shown promising results in reducing fleas on black-tailed prairie dogs in the field (Jachowski et al. 2011). Imidacloprid is a chloronicotinyl insecticide that impairs insect nerve function by acting as a competitive inhibitor at nicotinic acetylcholine receptors of the nervous system. Within 24–48 hr after a mammal consumes bait treated with imidacloprid, the compound resides in the animal's blood-stream and typically kills 96–97.3% of fleas that ingest blood from the treated animal for up to 30 days in a laboratory setting (Poché et al. 2008). The product is a systemic flea-control agent that can be spread in a bait form above ground, and can be applied more rapidly than deltamethrin (Jachowski et al. 2011). Given that imidacloprid reduced fleas on black-tailed prairie dogs, it might also be effective in treating Utah prairie dogs. However, reduction in flea prevalence, abundance, and intensity post-treatment with imidacloprid was less pronounced and more variable than is reported to occur with treatment using topical insecticides (Jachowski et al. 2011).

The primary objective of this study was to evaluate the effectiveness of imidacloprid on Utah prairie dogs by evaluating whether treatment influenced flea prevalence, abundance, and intensity. Insight into which species of fleas are present on Utah prairie dogs and which are most affected by imidacloprid could be used to optimize the timing and location of future flea-control efforts. Plague epizootics are associated with rapid increases in flea abundance (Pauli et al. 2006, Tripp et al. 2009) and it has been hypothesized that elevated flea abundance on a host is correlated with the risk of plague being present in an enzootic or epizootic form (Lorange et al. 2005, Eisen et al. 2006). However, multiple flea species typically are present on prairie dogs and those flea species differ from each other in their phenology and patterns of seasonal abundance (Pizzimenti 1975, Tripp et al. 2009). In addition, flea species differ in their competence as vectors of plague (Eisen et al. 2009). Therefore, it is important to understand which flea species are most abundant on Utah prairie dogs, and how abundance is impacted by the application of imidacloprid. Thus, a second objective of this study was to assess whether some flea species are more affected than others by treatment with imidacloprid.

STUDY AREA

Utah prairie dog populations are managed as 3 recovery units (U.S. Fish and Wildlife Service 2009), consisting of prairie dog populations that are relatively isolated (Brown et al. 2009) and occur at different elevations (Fig. 1). Given that elevation can influence flea communities (Krasnov et al. 2001, Biggins et al. 2010), we attempted to include 2 sites from high- (>2,438 m) and low- (approx. 1,676 m)

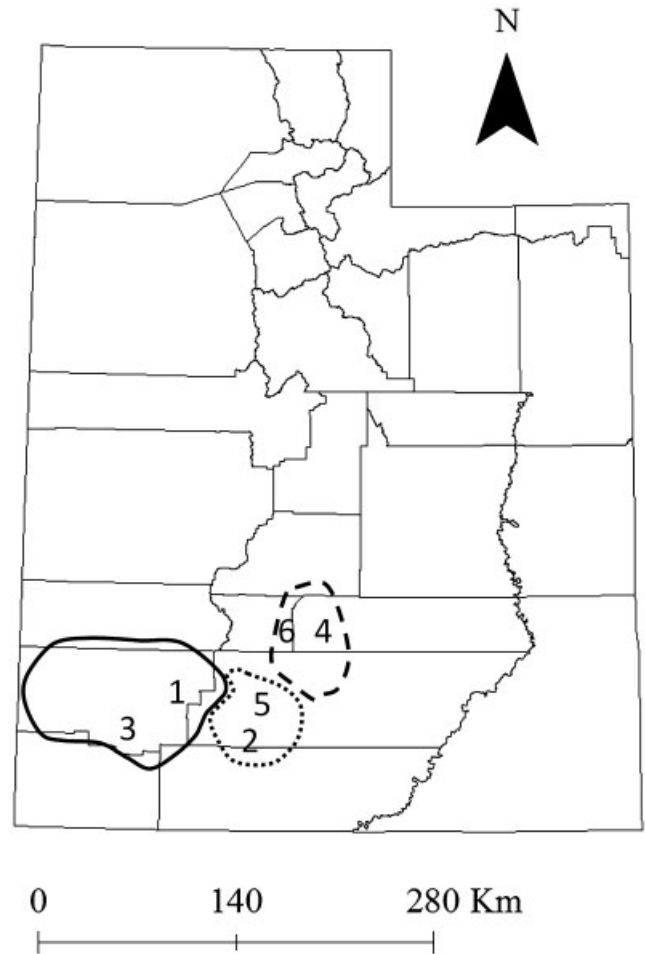


Figure 1. Location of 6 study sites in Utah, USA (county boundaries demarcated by solid lines). Polygons correspond to the extent of genotypically distinct populations of Utah prairie dogs, which also demarcate the extent of the West Desert (dark solid line), Paunsaugunt (dotted line), and Awapa Plateau (dashed line) recovery units. Sites are numbered in sequential order based on date of treatment with systemic flea-control product as (1) Dalley Farm, (2) East Creek, (3) Southern Utah University Farm, (4) Giles Hollow, (5) John's Valley, and (6) Forshea Draw.

elevation prairie dog colonies during each year. The low-elevation sites in 2009, Southern Utah University (SUU) Farm (1,707 m) and Dalley Farm (1,737 m), were in the West Desert recovery unit in Iron County, Utah, USA. Both sites contained clay loam soils with 0–5% slope and consisted of dry pasture dominated by crested wheatgrass (*Agropyron cristatum*). The high-elevation sites in 2009, East Creek (2,438 m) and Giles Hollow (2,438 m), were in the Paunsaugunt Recovery Unit, Garfield County, Utah, and in the Awapa Plateau Recovery Unit in Wayne County, Utah, respectively (Fig. 1). Soil conditions at the East Creek and Giles Hollow sites ranged from silty loam with 0–2% slope in the creek bottom to gravelly loam and 5–20% slope on the valley sides. These sites consisted of upland sage-steppe ecotype dominated by both cool- and warm-season grasses and with intermittent to low shrub cover of rabbitbrush (*Chrysothamnus viscidiflorus*) and sagebrush (*Artemisia tridentata*).

In September 2009, the East Creek (site 2) prairie dog population experienced a plague epizootic and was removed

from the study. In 2010, we also abandoned the Giles Hollow site (site 3) due to low trapping success on both treatment and control plots. These high-elevation sites were replaced by new sites, John's Valley (site 5; 2,268 m) and Forshea Draw (site 6; 2,774 m), which were located in the same counties and recovery units as the sites they replaced and had similar habitat attributes (Fig. 1). However, the John's Valley site (site 5) was only studied briefly after low trapping success and an unexplained decline in the prairie dog population. In total over both years combined, we sampled fleas from Utah prairie dog hosts at 6 different sites (Fig. 1).

MATERIALS AND METHODS

Field Methodology

We used a paired treatment and control design to evaluate the systemic flea-control product on each of our study sites. In May 2009, we established 2 study plots (treatment and control) at each of the 4 study sites. We selected study sites, and plots within sites, based on the expectation of capturing >50 prairie dogs within 5 consecutive trapping days based on visual assessments of prairie dog populations and our knowledge of likely trapping success (Jachowski et al. 2011). For each plot, we attempted to select an entire colony of prairie dogs (i.e., an aggregation of adjoining family groups) or 2 colonies in proximity to each other, such that treatment and control plots within a site had similar soil, landscape, and vegetation conditions. When 2 plots were selected within an individual colony, a buffer of ≥ 200 m was used between plot boundaries because of concerns that individual prairie dogs might move between treatment and control plots during our period of study. This resulted in treatment and control plots within sites that ranged in size from 2.2 ha to 190.2 ha, depending on density of prairie dogs.

We randomly assigned treatment or control status to plots at each site. Control plots were left untreated throughout the study. On each treatment plot, we scattered 56 g of imidacloprid-treated oat grain bait (following manufacturer recommendations) within 2.4 m of each prairie dog burrow opening. We applied bait once to each treatment plot during May and June 2009, when prairie dogs were observed to become consistently active above ground after winter hibernation. We treated 1 site per week so that the time of application was staggered, thereby allowing us to conduct intensive 5-day sampling of prairie dogs and fleas at individual sites at 30-day intervals post-treatment. In April and May 2010, we reapplied the bait to 3 of the 4 treatment plots that were treated in 2009, excluding the East Creek site (site 2). We doubled the bait application rate in 2010 by applying 56 g of treated grain bait to every burrow within each treatment plot on day 1 and repeating the application on day 5. In July 2010, we similarly applied bait to the treatment plot at the newly established Forshea Draw site (site 6).

From June to October we simultaneously sampled prairie dogs and their fleas in both treatment and control plots at each site. During each 5-day trapping session we placed 100 Tomahawk live traps (15.2 cm \times 15.2 cm \times 0.6 m) on the

treatment plot and 100 traps on the control plot close to actively used burrow openings. We set the traps at dawn each day and checked them at hourly intervals thereafter. Captured prairie dogs were anesthetized using a specially designed isoflurane vaporizer and fitted mask (Seven and Seven Anesthesia, Fort Collins, CO). While a prairie dog was sedated, we sampled fleas by brushing the entire body of the individual for 30–60 s using a flea comb (Biggins et al. 2010, Jachowski et al. 2011). We weighed each prairie dog, measured its hind foot length, determined its age and sex class, and attached a uniquely numbered metal tag to each ear. We determined age class as either juvenile (i.e., young that emerged from the natal burrow within the past 8 months) or adult based on size, pelage, reproductive status, and body condition (Hoogland 1995). Any prairie dog that was recaptured during the 5-day trapping session was released without further handling.

Laboratory Methodology and Data Analysis

Following counting fleas on prairie dogs in the field, fleas were collected and placed in individually marked bags and frozen for subsequent laboratory identification. Utilizing compound microscopes and following the taxonomic keys of Hubbard (1947), Stark (1958), and Lewis (2002), all fleas were identified to species and gender. Because some fleas were lost in transport from the field to the laboratory, the total number of fleas identified to species (Table 1) does not always correspond with the values used in conducting comparative statistical tests detailed below, which are based on field counts.

We evaluated effectiveness of the systemic flea-control product at a community (all flea species combined) and population (by individual flea species) scale. At the community scale, we compared prevalence (percent of sampled prairie dogs with fleas), abundance (no. of fleas divided by the total no. of prairie dogs sampled), and intensity (no. of fleas divided by the total no. of prairie dogs sampled with fleas) of fleas on prairie dogs between paired treatment and control plots during each trapping session. We used a Fisher's exact test to evaluate our null hypothesis that prevalence did not differ between treatment and control sites. We compared abundance and intensity between treatment and control pairings within each site during each month of sampling using a bootstrap 2-sample *t*-test (Rózsa et al. 2000). We calculated an index of discrepancy (Poulin 1993) using Program Quantitative Parasitology 3.0 (Reiczigel and Rózsa 2005) to determine the extent to which the distribution of fleas on prairie dogs differed from a uniform distribution. To determine whether fleas tended to be more aggregated on prairie dogs in treatment plots than in control plots, we used a nested analysis of variance (ANOVA), where index of discrepancy was the dependent variable, month and site were the independent variables, and plot type was the nested effect within each site.

We used the number of each species of flea collected from prairie dogs during each sampling occasion on each plot to calculate Shannon indices of diversity (Magurran 1988). The Shannon index (H_s) is a nonparametric measure that

Table 1. Total number of fleas (by species) and diversity of fleas (H_s , where higher values indicate more unique flea species present and higher evenness in count distribution among species) collected from Utah prairie dogs (*Cynomys parvidens*) on treatment (T) and control (C) plots at the Dalley farm (1), East Creek (2), Southern Utah University farm (3), Giles Hollow (4) and Forshear Draw (6) study sites (Fig. 1) sampled during 2009 (A) and 2010 (B). The Johns Valley site (5) is not included due to an unexplained decline in prairie dogs and failure to collect fleas.

Site	Month	Flea species												Total		H_s			
		<i>Oropsylla hirsuta</i>		<i>Oropsylla tuberculata cynomuris</i>		<i>Oropsylla labis</i>		<i>Oropsylla idahoensis</i>		<i>Oropsylla diaminus montana</i>		<i>Thrasis francisi</i>		T	C	T	C		
		T	C	T	C	T	C	T	C	T	C	T	C						
(A) 2009																			
1	Jun ^a	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0.257	0.082	
	Jul	52	123	0	0	0	0	0	0	4	2	0	0	56	125	0.082	0.088		
	Aug	407	257	0	0	0	0	0	0	5	3	1	1	413	261	0.122	0.059		
	Sep	441	320	0	0	0	0	0	0	12	2	0	1	453	323	0.013	0.014		
	Oct	1,662	534	0	0	0	0	0	0	3	1	0	0	1,665	535	0.601	0.601		
2	Jun	0	0	0	20	0	1	0	0	0	0	0	4	0	25	0.637	1.228		
	Jul	1	1	0	3	0	1	0	6	2	0	0	12	3	23	1.121	1.171		
	Aug	3	1	1	0	0	2	1	7	0	0	6	4	11	14	1.169	0.613		
	Sep	6	9	2	1	0	3	2	98	1	2	0	2	11	115	0.654	1.030		
3	Jul	269	327	0	0	0	0	0	0	12	143	75	369	356	839	0.783	0.791		
	Aug	104	96	0	0	0	0	0	0	8	5	45	102	157	203	1.049	0.844		
	Sep	93	124	0	0	0	0	0	0	53	11	46	101	192	236	0.395	0.587		
4	Jul	0	30	1	5	32	20	0	0	0	3	230	326	263	379	0.576	0.503		
	Aug	1	20	1	0	32	100	0	0	0	0	125	650	159	770	0.589	0.592		
	Sep	2	0	4	3	96	177	0	0	1	2	374	562	477	744				
(B) 2010																			
Site	Month	Flea species												Total		H_s			
		<i>Oropsylla hirsuta</i>		<i>Oropsylla tuberculata cynomuris</i>		<i>Oropsylla labis</i>		<i>Oropsylla diaminus montana</i>		<i>Thrasis francisi</i>		<i>Hoplopsyllus anomalus</i>		<i>Rhadinopsylla sectillis</i>		T	C	T	C
T	C	T	C	T	C	T	C	T	C	T	C	T	C	T	C				
1	May	5	11	0	52	0	0	0	0	0	0	0	0	0	0	57	11	0	0.463
	Jun	2	12	0	4	0	0	0	0	0	0	0	0	0	0	2	16	0	0.562
	Jul	4	150	0	0	0	0	0	0	0	0	0	0	0	4	150	0	0	
	Aug	83	249	0	0	0	0	0	0	0	0	0	0	0	83	249	0	0	
3	Jun	118	131	0	4	0	1	26	45	0	20	2	1	0	0	146	202	0.974	0.514
	Jul	236	327	0	1	0	0	36	20	0	12	21	15	0	0	293	375	0.510	0.589
	Aug	333	86	0	0	0	0	23	0	4	31	6	0	0	0	366	117	0.578	0.366
	Sep	491	0	0	0	1	0	45	0	0	0	3	0	0	0	540	0	0	0.330
6	Aug	0	4	1	0	1	0	0	0	3	85	0	0	0	5	89	0.950	0.183	
	Sep	1	0	2	2	2	0	0	0	9	173	0	0	0	2	14	177	1.029	0.114

^a After collection in the field, all fleas collected during this sampling period were lost in transit from the field to the laboratory for identification.

accounts for flea species richness and evenness of distribution among plots within sites. Values of H_s typically range between 1.5 and 3.5, with higher values indicating a greater number of unique species and a more even distribution of individuals among species. We evaluated whether flea diversity on prairie dogs differed between treatment and control plots using a nested ANOVA, where the H_s value was the dependent variable, month and site were the independent variables, and plot type was the nested effect within each site.

We evaluated the effect of the treatment on each species of flea individually by calculating the difference in abundance of each flea species between paired treatment and control plots during each sampling interval at each site. We used the log-transformed difference value as the dependent variable in an ANOVA procedure where month, treatment (single or double), site, and flea species were independent variables. We also included interactions of flea species and site, flea species

and treatment, and flea species and month in our model. When interactions were significant in our model, we fit factorial ANOVAs to each flea species individually to separately assess the supported interactive effects from our cross-species model. For all analyses, we considered effects significant at $P \leq 0.05$.

RESULTS

We collected and identified 8 flea species on Utah prairie dogs during this study (Table 1). Flea species richness varied among sites from 3 to 6 species in 2009 and 2 to 6 species in 2010. *Thrasis francisi*, *Oropsylla hirsuta*, *O. tuberculata cynomuris*, and *O. labis* were present on all 6 sites we included in the study. *Oropsylla hirsuta* generally was the most abundant species at low-elevation sites (sites 1 and 3) and *T. francisi* generally was the most abundant species at high-elevation sites (sites 2, 4, and 6). *Hoplopsyllus anomalus* was

Table 2. Summary of the number of Utah prairie dogs (*Cynomys parvidens*) sampled along with the prevalence, mean intensity (with 95% CIs), and aggregation index of discrepancy (*D*; Poulin 1993) of flea populations on prairie dogs at 30-day intervals post-treatment with imidacloprid in 2009 on Dalley Farm (1), East Creek (2), Southern Utah University Farm (3) and Giles Hollow (4) study sites (Fig. 1). Treatment plots received a single application of 56 g of imidacloprid-treated bait near every burrow on day 1. Control plots were sampled simultaneously to treatment plots, but they received no imidacloprid-treated bait.

Site	Month	Treatment					Control				
		No. sampled	Prevalence	Intensity		<i>D</i>	No. sampled	Prevalence	Intensity		<i>D</i>
				\bar{x}	95% CI				\bar{x}	95% CI	
1	Jun	102	0.049	1.6	1.00–1.80	0.949	50	0.120	1.17	1.00–1.33	0.877
	Jul	69	0.362 ^a	2.32	1.80–2.96	0.749	55	0.618	3.79	2.44–5.82	0.705
	Aug	63	0.903	7.43	6.07–9.11	0.458	48	0.813	6.46	4.31–9.72	0.649
	Sep	55	0.818	10.58	7.73–15.31	0.605	41	0.878	9.19	6.28–13.78	0.575
	Oct	37	1.000	45.73 ^a	36.78–61.08	0.438	30	1.000	17.37	13.67–21.80	0.357
2	Jun	43	0.000 ^a	0.00		0	46	0.239	2.18	1.45–3.27	0.862
	Jul	53	0.075 ^a	1.00	0.00–0.00	0.907	59	0.288	1.53	1.24–1.94	0.765
	Aug	50	0.160	1.50	1.00–2.00	0.859	49	0.265	1.46	1.15–1.69	0.764
	Sep	43	0.163 ^a	1.71 ^a	1.14–2.00	0.852	41	0.634	4.46	3.15–6.23	0.649
3	Jul	69	0.913 ^a	5.83 ^a	5.00–6.84	0.391	56	1.000	15.16	12.71–18.82	0.366
	Aug	50	0.900	3.58	2.91–4.51	0.441	49	0.837	5.05	3.78–7.05	0.561
	Sep	48	0.938	4.64 ^a	3.78–5.71	0.414	41	0.902	6.49	5.14–7.84	0.418
4	Jul	42	0.881	7.41	5.19–12.05	0.567	47	0.936	9.18	6.91–12.25	0.516
	Aug	43	0.698 ^a	5.17 ^a	3.07–8.93	0.706	50	0.940	16.83	12.79–22.66	0.519
	Sep	48	0.896	11.19 ^a	8.63–14.33	0.489	44	0.932	18.49	13.85–24.63	0.519

^a There was a significant difference (P -value ≤ 0.05) between treatment and control sites during the survey month. Differences in prevalence were tested with a Fisher's exact test and differences in intensity were tested with bootstrap 2-sample t -tests.

found only at low-elevation sites in 2010. *Rhadinopsylla sectillilis sectillilis* was found only at Forshea Draw (site 6 in 2010) and *O. idahoensis* only at East Creek (site 2 in 2009), both of which were high-elevation sites. *Oropsylla diamanus montana* was found at both high- and low-elevation sites (sites 1–3), but was most common on SUU Farm (site 3) during both 2009 and 2010.

The single application of imidacloprid-treated grain bait in 2009 had mixed effects on flea communities and populations (Table 2; Fig. 2). For all flea species combined, at Dalley Farm (site 1) we observed no effect of treatment at 30 days post-treatment ($P > 0.179$), followed by a 25% lower prevalence ($P = 0.006$) and 65% lower abundance ($P = 0.035$) of fleas on treatment plots compared to control plots at 60 days post-treatment. We observed no subsequent differences in flea populations between plots until 150 days post-treatment, when flea abundance was 3 times higher on the treatment plot compared to the control plot ($P = 0.002$; Table 2; Fig. 2). At East Creek (site 2) we detected the lowest flea abundance and intensity of all sites included in this study (Tables 2 and 3), but also observed the most consistent effects of treatment on fleas. Flea prevalence was 20% lower on treatment compared to control plot at 30 days ($P = 0.001$) and 60 days ($P = 0.007$) post-treatment and up to 47% lower ($P < 0.001$) at 120 days post-treatment (Table 2). In addition, we observed a small (1–3 flea) difference in flea abundance at 30 days ($P = 0.041$), 60 days ($P = 0.005$), and 120 days ($P = 0.006$) post-treatment (Table 2; Fig. 2). We observed a mass mortality of prairie dogs at East Creek on both treatment (18.4 ha) and control (13.4 ha) plots at the completion of September prairie dog trapping. The Centers for Disease Control (CDC) confirmed the presence of plague based on laboratory testing of a prairie dog carcass. At SUU

Farm (site 3), flea prevalence was 9% lower ($P = 0.032$) and flea abundance and intensity were 65% and 61% lower respectively ($P < 0.001$) on treatment compared to control sites at 30 days post-treatment. We observed no difference between treatment and control plots in all 3 flea metrics at 60 days ($P > 0.146$) or 90 days post-treatment ($P > 0.080$), with the exception of 29% lower flea intensity on treatment than control plots at 90 days post-treatment ($P = 0.039$). At Giles Hollow (site 4), we did not observe a difference in all 3 flea metrics between treatment and control plots at 30 days post-treatment ($P > 0.277$). However, at 60 days post-treatment, we observed a 24% lower flea prevalence ($P = 0.002$), as well as 69% lower flea intensity ($P = 0.001$) and 77% lower flea abundance ($P < 0.001$) on treatment plots compared to control plots. In addition, we observed 39% lower flea intensity ($P = 0.026$) and 42% lower flea abundance ($P = 0.026$) on treatment compared to control plots at 90 days post-treatment (Table 2; Fig. 2). In comparisons across all 4 sites, we did not observe an effect of treatment on flea aggregation ($F = 1.18$, $df = 4$, $P = 0.356$), but did observe that fleas tended to be more aggregated on prairie dogs at Dalley Farm (site 1) and East Creek (site 2; $F = 25.04$, $df = 3$, $P < 0.001$) and during the months of June and July ($F = 10.97$, $df = 4$, $P < 0.001$; Table 2).

Despite doubling the application rate of imidacloprid in 2010, we again failed to observe a consistent decline in flea populations across all treatment plots when all species were combined (Table 3; Fig. 3). At Dalley Farm (site 1), we observed 42% lower flea prevalence ($P < 0.001$), 93% lower flea abundance ($P = 0.006$), and 67% lower flea intensity ($P = 0.006$) on treatment compared to control plots at 30 days post-treatment, but did not detect a difference between plots at 60 days post-treatment ($P > 0.138$;

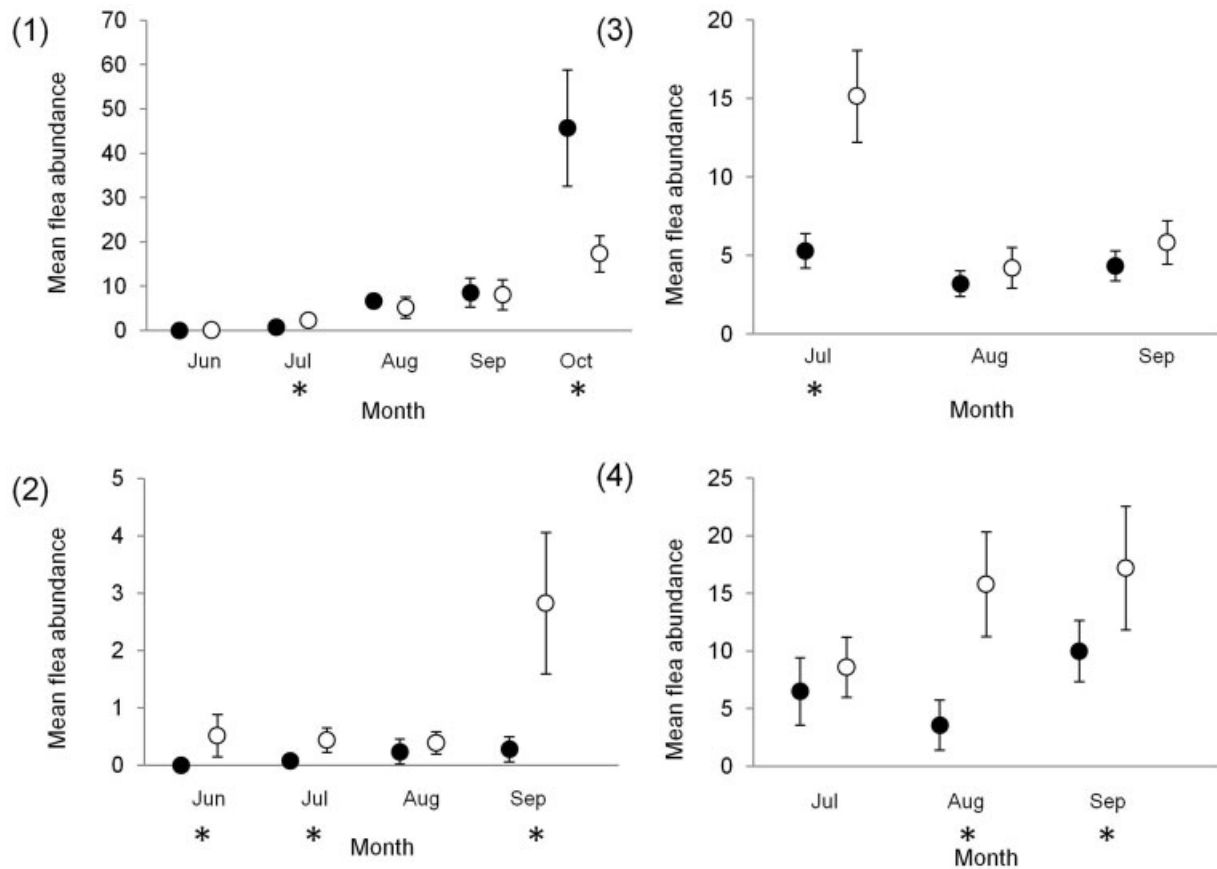


Figure 2. Mean abundance (with 95% CIs) of fleas on Utah prairie dogs on treatment (solid circles) and control (hollow circles) plots at Dalley Farm (1), East Creek (2), Southern Utah University Farm (3) and Giles Hollow (4) study sites (Fig. 1) during each monthly sampling session in 2009. All treatment plots were treated with a single application of imidacloprid. Asterisks underneath the month of sampling indicate a significant difference between treatment and control plots based on a bootstrap 2-sample *t*-test ($P \leq 0.05$).

Table 3; Fig. 3). We observed 44% lower flea prevalence on treatment compared to control plots at 90 days ($P < 0.001$) and 19% lower at 120 days ($P = 0.021$) post-treatment, but abundance ($P > 0.081$) and intensity ($P > 0.120$) did not differ (Table 3; Fig. 3). At SUU Farm (site 3), we did not observe a difference in our 3 flea metrics between plots at 30 days post-treatment ($P > 0.293$). At 60 days post-treatment, flea intensity and abundance was 39% lower on treat-

ment compared to control plots ($P < 0.013$). In contrast, at 90 days, flea intensity was 75% higher and flea abundance was 123% times higher on treatment than control plots ($P < 0.033$; Table 3; Fig. 3). Despite visual observations of adequate prairie dog populations prior to and during treatment, both Giles Hollow (site 4) and the newly added John's Valley (site 5) were abandoned at 30 days post-treatment following extremely low trapping success and an

Table 3. Summary of the number of Utah prairie dogs (*Cynomys parvidens*) sampled along with the prevalence, mean intensity (with 95% CIs), and aggregation index of discrepancy (*D*; Poulin 1993) of flea populations on prairie dogs at 30-day intervals post-treatment with imidacloprid in 2010 on Dalley Farm (1), Southern Utah University Farm (3), and Forshea Draw (6) study sites (Fig. 1). Treatment plots received a double application of imidacloprid-treated bait (56 g of bait applied near every burrow on day 1 and day 5). Control plots were sampled simultaneously to treatment plots, but they received no imidacloprid treated bait.

Site	Month	Treatment					Control				
		No. sampled	Prevalence	Intensity		<i>D</i>	No. sampled	Prevalence	Intensity		<i>D</i>
				\bar{x}	95% CI				\bar{x}	95% CI	
1	May	41	0.098 ^a	1.50 ^a	1.00–1.75	0.897	29	0.517	4.53	3.27–6.27	0.634
	Jun	46	0.043	1.00	0–0	0.978	40	0.150	2.83	1.50–5.67	0.888
	Jul	40	0.100 ^a	1.00	0–0	0.923	43	0.535	6.74	2.04–24.09	0.851
	Aug	58	0.690 ^a	3.13	2.33–4.32	0.623	50	0.880	7.18	4.80–12.45	0.623
3	Jun	54	0.667	4.17	3.08–7.17	0.627	41	0.683	7.25	4.11–14.96	0.753
	Jul	51	0.922	6.19 ^a	4.83–9.45	0.487	41	0.951	10.08	8.10–12.51	0.387
	Aug	42	0.905	9.79 ^a	7.08–13.68	0.538	31	0.710	5.59	4.00–7.41	0.553
	Sep	42	0.881	14.68	10.41–23.68	0.559	22	0.955	8.95	6.52–12.76	0.410
6	Aug	38	0.105 ^a	1.25	1.00–1.50	0.887	16	0.688	8.45	3.64–21.82	0.694
	Sep	41	0.317 ^a	1.38 ^a	1.00–1.85	0.735	29	0.828	7.38	4.79–11.29	0.585

^a There was a significant difference (P -value ≤ 0.05) between treatment and control sites during the survey month. Differences in prevalence were tested with a Fisher's exact test and differences in intensity were tested with bootstrap 2-sample *t*-tests.

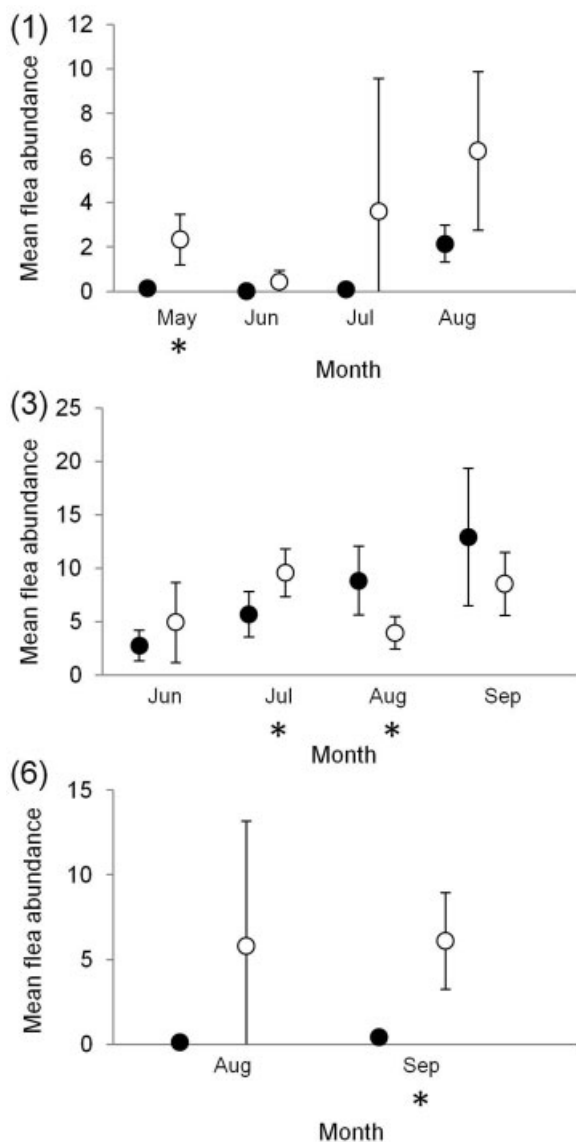


Figure 3. Mean abundance (with 95% CIs) of fleas on Utah prairie dogs on treatment (solid circles) and control (hollow circles) plots at Dalley Farm (1), Southern Utah University Farm (3), and Forshea Draw (6) study sites (Fig. 1) during each monthly sampling session in 2010. All treatment plots were treated with a double application of imidacloprid. Asterisks underneath the month of sampling indicate a significant difference between treatment and control plots based on a bootstrap 2-sample *t*-test ($P \leq 0.05$).

unexplained decline in above-ground activity of prairie dogs (CDC tests of a prairie dog carcass was negative for plague). At Forshea Draw (site 6), flea prevalence was 58% lower ($P < 0.001$) on treatment compared to the control plot at 30 days post-treatment, and 51% lower at 60 days post-treatment ($P < 0.001$; Table 3). In addition, we observed 93% lower flea abundance ($P = 0.021$) and 81% lower flea intensity ($P = 0.020$) on treatment compared to control plots at 60 days post-treatment (Table 3; Fig. 3). Similar to observations in the previous year, we did not detect a significant effect of treatment on flea aggregation within plots in 2010 ($F = 1.42$, $df = 3$, $P = 0.295$), but did find that fleas tended to be more aggregated on prairie dogs at Dalley Farm (site 1) and Forshea Draw (site 6; $F = 11.41$, $df = 2$, $P = 0.003$; Table 3).

Flea diversity on prairie dogs was not consistently lower or higher on treatment plots compared to control plots. We detected no difference in H_s between treatment and control plots within each site ($F = 0.35$, $df = 4$, $P = 0.842$) or month of sampling ($F = 2.40$, $df = 4$, $P = 0.088$) in 2009. However, we did find that H_s values differed among sites ($F = 21.92$, $df = 3$, $P < 0.001$), where H_s values were higher for East Creek (site 2) and SUU Farm (site 3) than for Dalley Farm (site 1; Table 1). In 2010, following the double treatment with imidacloprid, we observed differences in H_s values between treatment and control plots ($F = 7.47$, $df = 3$, $P = 0.007$). Only one species of flea occurred on prairie dogs in the treatment plot at Dalley Farm (site 1) and, thus, H_s was higher in control plots (Table 1). In contrast, H_s values were consistently higher in the treatment plot at Forshea Draw (site 6) during both months of sampling (Table 1). Similar to 2009, we detected no effect of month H_s values in 2010 ($F = 2.78$, $df = 4$, $P = 0.086$).

The effect of imidacloprid treatment on the abundance of each flea species was highly variable, and differed among species based on treatment type ($F = 4.04$, $df = 4$, $P = 0.027$), month ($F = 5.72$, $df = 23$, $P = 0.003$), and site ($F = 12.40$, $df = 6$, $P < 0.001$; Table 1). For *T. francisi*, we found that differences in abundance between control and treatment plots did not vary by treatment type ($F = 3.34$, $df = 1$, $P = 0.110$) or month of sampling ($F = 0.00$, $df = 3$, $P = 0.999$), but were lower on treatment plots compared to control plots at Forshea Draw (site 6), Giles Hollow (site 4), and SUU Farm (site 3) compared to East Creek (site 2) and Dalley Farm (site 1; $F = 18.47$, $df = 4$, $P = 0.001$). For *O. hirsuta*, we observed no effect of treatment type on abundance ($F = 0.24$, $df = 1$, $P = 0.647$), but observed that differences in abundance between treatment and control plots were greatest during July ($F = 6.71$, $df = 5$, $P = 0.045$) and lowest on East Creek (site 2; $F = 9.56$, $df = 4$, $P = 0.025$). The remaining 6 species of flea were encountered so infrequently that we were unable to individually test for differences in abundance between treatment and control plots across sites, months, and treatment types.

DISCUSSION

Effectiveness of imidacloprid in controlling fleas on Utah prairie dogs was highly variable among sites. The mixed results we observed among sites and flea species suggest that imidacloprid treatment at reported rates of application was only irregularly or marginally effective. Overall, oral treatment with an imidacloprid grain bait was less effective in this study than in a similar study performed on black-tailed prairie dogs (*C. ludovicianus*; Jachowski et al. 2011) and in previous evaluations on California ground squirrels (*Spermophilus beecheyi*; Borchert et al. 2009).

In comparison to topical flea-control products, imidacloprid-treated grain bait is likely less effective at controlling flea populations on Utah prairie dogs. Following treatment with the topical insecticide Pyreperm, Hoogland et al. (2004) reported that the number of Utah prairie dogs with no fleas present increased from approximately 45% pretreatment to

>90% post-treatment. Similarly, 10 months following treatment with the topical insecticide deltamethrin, Biggins et al. (2010) found that treated sites contained 85% fewer fleas than control sites. In contrast, we were only able to consistently reduce flea prevalence at 1 of our 4 sites in 2009 following a single application of imidacloprid, and 2 of our 4 sites in 2010 following a double application. Our systemic evaluations are similar to poor results observed in previous examinations of topical applications of imidacloprid, which have documented a 54% recovery in adult flea populations after 2 weeks and a complete recovery in 4 weeks (Metzger and Rust 2002). More frequent application of oral grain baits treated with imidacloprid could potentially improve results (Borchert et al. 2009). However, additional applications might increase costs beyond those of a single application of deltamethrin (Jachowski et al. 2011).

In contrasting potential flea-management options, managers should consider the potential for a flea species to develop resistance following repeated application of a single flea-control product. Many managers currently apply deltamethrin each year to prairie dog colonies to reduce flea abundance and mitigate risk of plague epizootics. For example, managers in South Dakota, USA, annually apply approximately 2,608 kg of deltamethrin on land occupied by a single large (4,513 ha) population of black-tailed prairie dogs (Griebel 2009). Managers of prairie dog populations in ≥ 7 other states in the western United States are similarly applying deltamethrin to prairie dog burrows for research and conservation purposes (Scott Larson, United States Fish and Wildlife Service, personal communication). While deltamethrin has been effective at limiting expansion of epizootic plague to treated areas, the repeated application of a single insecticide over multiple years can lead to development of chemical resistance (Roberts and Andre 1994). Therefore, it might become necessary in the future to use other flea control products due to the potential for flea species to develop resistance to insecticides (Bossard et al. 2002). Although resistance to imidacloprid (or future more effective products) also could develop, use of multiple insecticides might delay such resistance (Zhao et al. 2010).

Low abundance of fleas prior to and during an epizootic plague outbreak at East Creek (site 2) in September 2009 poses a challenge to current hypotheses regarding flea abundance thresholds and plague risk. Multiple studies have documented how flea control has halted the spread of epizootic outbreaks (Hoogland et al. 2004, Griebel 2009). However, flea control at East Creek during our study did not stop a plague epizootic despite a significant reduction in flea populations on the treatment plot, such that flea abundance was below currently hypothesized thresholds for epizootic plague outbreaks. Flea abundances >4 per individual are hypothesized to be required to maintain plague in a host population at an enzootic level and >9 per individual are required to maintain an epizootic (Lorange et al. 2005, Eisen et al. 2006). Prior to and during the epizootic outbreak during the September 2009 trapping session at East Creek, flea abundance averaged <3 per individual prairie dog on the control plot and <0.5 per

individual on the treatment plot (Fig. 2). This suggests either that we sampled fleas from prairie dogs immediately prior to a rapid increase in flea abundance typically associated with epizootic outbreaks of plague in prairie dogs (Pauli et al. 2006, Tripp et al. 2009), or that flea abundance was not a major factor in maintaining and spreading plague among prairie dogs on this site. In addition, the epizootic outbreak of plague at East Creek along with unexplained declines in prairie dog populations at Giles Hollow (site 4) and John's Valley (site 5) during our study suggests that plague effects, in either epizootic or enzootic form (Biggins et al. 2010), might be greater in prairie dog populations at high elevations ($>2,134$ m). Further research into how elevation influences the risk of plague could improve plague mitigation strategies.

We observed a high diversity of flea species on Utah prairie dogs. During a similar flea-control study on Utah prairie dogs, Biggins et al. (2010) reported that 3 flea species were commonly encountered: *O. tuberculata cynomuris*, *O. hirsuta*, and *T. francisi*. In addition to these species, which we commonly encountered, we found an additional 5 species of flea. Two of the 5 species were limited to <50 individual fleas (*H. anomalus* and *R. sectillis sectillis*), but the other 3 species (*O. labis*, *O. idahoensis*, and *O. diamanus montana*) were prominent components of the flea community (Table 1). Each of the 8 species was within its historically documented range; however, a majority of them are more commonly associated with rodent species other than Utah prairie dogs (Hubbard 1947, Stark 1958, Pizzimenti 1975). Seven of these 8 flea species have been documented to be competent plague vectors, although the degree of vector competency varies among flea species (Eisen et al. 2009). The diversity of flea species we encountered, together with the relatively high reported diversity of small mammals in this area (Rickart 2001), indicate the need to investigate how neighboring rodent communities, along with site-specific habitat factors (such as soil conditions and elevation), collectively contribute to patterns in flea diversity on Utah prairie dogs. This is of particular interest given that small mammals occurring on and near prairie dog colonies have been hypothesized to play a key role in maintaining and transmitting plague in prairie dog colonies (Salkeld et al. 2010).

MANAGEMENT IMPLICATIONS

Flea control remains the key tool to mitigate the effects of plague on prairie dog populations, and future efforts should be made to develop additional flea control products, as well as an improved understanding of flea ecology. Although imidacloprid is unlikely to be an effective flea-control product on Utah prairie dog populations, future evaluations of baits containing other more pharmacologically appropriate active ingredients should be undertaken. In particular, new, more effective products should be utilized on high-priority prairie dog populations and black-footed ferret (*Mustela nigripes*) reintroduction sites, where a single flea-control product is typically applied over multiple years and risk of fleas developing resistance is greatest. Timing and application of flea control products (and, potentially future plague vaccine treatments) to mitigate the effects of plague on Utah prairie

dogs could likely be improved by further research investigating the ecology of fleas in this system.

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