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CLIMATIC SUITABILITY OF SAN JOAQUIN KIT FOX (*VULPES MACROTIS MUTICA*) DENS FOR SARCOPTIC MANGE (*SARCOPTES SCABIEI*) TRANSMISSION

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ABSTRACT: More than 460 cases of sarcoptic mange (*Sarcoptes scabiei*) in endangered San Joaquin kit foxes (SJKF, *Vulpes macrotis mutica*) have been reported in Bakersfield, California. Because SJKF are a den-obligate species, their dens have been proposed as a route of transmission. We determined whether SJKF den temperatures and humidities could support mite off-host survival based on previously published estimates of off-host mite survival times. We monitored SJKF dens for 6 d in summer and winter of 2017 and 2018 using temperature- and humidity-sensing data loggers placed within the dens. Motion-triggered cameras monitored animal use of and entrances into the dens. Linear regression models were fitted to the published mite survival data to predict estimated mite survival time (EMST) in SJKF dens based on observed mean temperature and humidity of the den. Den covariates including irrigation, type of den, and season were then fitted to a mixed effects linear model to predict EMST. The average EMST across various habitats in Bakersfield was 4.8 d; the longest EMST was 7.1 d for dens in habitats with irrigated grass in the winter. Den climatic conditions in Bakersfield may support off-host mite survival through a timeframe adequate for revisitation by another fox. The finding that irrigation may enhance EMST suggested that risk to foxes varied with den type and that mitigation strategies may need to vary with den types.

Key words: Climate suitability, dens, San Joaquin kit fox, sarcoptic mange, *Sarcoptes scabiei, Vulpes macrotis mutica*.

INTRODUCTION

Because of profound habitat loss and fragmentation, the San Joaquin kit fox (SJKF, *Vulpes macrotis mutica*) has a limited range in central California and is listed as a federally Endangered and California Threatened species (Williams et al. 1998; Cypher 2010; Cypher et al. 2013). San Joaquin kit foxes can adapt to urban living and have established populations within the cities of Taft, Coalinga, and Bakersfield (Cypher 2010). An epidemic of fatal sarcoptic mange (*Sarcoptes scabiei*) in SJKFs in Bakersfield was first detected in the spring of 2013 with 15 cases and 13 mortalities described (Cypher et al. 2017). To date, more than 460 cases have been documented. California Department of Fish and Wildlife in conjunction with the Endangered Species Recovery Program use slow release flumethrin-imidacloprid, oral and injectable ivermectin, and hospitalization of severe cases to try to mitigate the epidemic. These treatments are effective at clearing the infestation of individual SJKFs, but the foxes do not appear to become immune and reinfestation of an individual is frequent. The pathogenesis of sarcoptic mange is similar in most canid species (Scott et al. 2001; Curtis 2012), but unlike in dogs, there is no evidence of spontaneous recovery of SJKF, and mange in this species is expected to be fatal without intervention (Curtis 2012; Cypher et al. 2017).

Sarcoptic mange is caused by a mite that burrows in the epidermis of numerous mammal species and is most frequently transmitted through direct contact with infected individuals (Scott et al. 2001; Curtis 2012; Arlian and Morgan 2017). Environmental transmission is a rare but documented method of infestation (Scott et al. 2001; Curtis 2012). Mites will desiccate off the host and die when exposed to arid (<50% relative humidity) or hot (>47.5 C) environments (Mellanby et al. 1942; Arlian and Morgan 2017). In a 1989 study (Arlian 1989), the microclimate gradient in which mites can survive off-host was established. With lower temperatures, survival time can reach up to a month (Arlian et al. 1984; Arlian 1989; Arlian and Morgan 2017). Depending on environmental conditions, the mites can remain infectious anywhere from 24 to 36 h off-host (Arlian et al. 1984; Arlian 1989).

Outside of mating and interactions within their family unit, SJKFs have relatively low intraspecies physical contact, raising the question of how sarcoptic mange was so rapidly and extensively transmitted. The foxes obligately use dens year-round for raising young, eluding predators, avoiding temperature extremes, and conserving moisture (Cypher 2010). Each SJKF uses an average of 11 different dens per year throughout their home range (Koopman et al. 1998), with some individuals using as many as 49 (Reese et al. 1992; Cypher 2010). In addition to natural earthen dens, urban SJKFs will create dens under man-made structures, such as buildings and shipping containers, and within pipes (Cypher 2010). Because of high fox densities and substantial spatial overlap among urban family groups, dens may be used by members of multiple family groups, sometimes as rapidly as within consecutive days. Because of the ability of mange mites to survive off-host, SJKF dens potentially could constitute a route of environmental transmission if den climatic conditions are suitable.

To facilitate development of treatment plans for this on-going epidemic, our aim was to identify the microclimate conditions of Bakersfield SJKF dens during the coldest and hottest seasons. We hypothesized that this microclimate would be suitable for mite survival for at least 2–3 d on the basis of ranges previously established (Arlian et al. 1984; Arlian 1989). We also sought to identify any den-associated risk factors that might increase mite survival time. If dens are a potential route of transmission, then the dens might be treated in some manner to help limit the spread of this epidemic.

MATERIALS AND METHODS

Data collection

We chose the campus of California State University, Bakersfield, in Bakersfield, California, as our study site because of its high density of foxes and the presence of numerous mange cases. Data were collected in mid-July to early September 2017 (summer season) and late November to mid-January 2018 (winter season). Many of the same dens were measured in both seasons. If dens could not be remeasured in the second season, another den in close proximity was measured. We identified SJKF dens during a concurrent telemetry study that involved tracking foxes to dens. A portable sewer camera (Trojan Worldwide Inc. self-leveling mini color camera system, Houston, Texas, USA) was used to explore the SJKF dens and to place an iButton Hygrochron data logger (DS1923, Maxim Integrated, San Jose, California, USA), which measured temperature and relative humidity for 6 d of each season. The manufacturer states an accuracy of 0.6% for relative humidity and 0.5 C for temperature. Each iButton was placed within a loose mesh stainless steel tea infuser ball that was attached to a fishing line for retrieval from the den. A second iButton was staked at the den entrance, also within a mesh ball. Each iButton was programmed to take hourly readings.

In pilot runs, SJKFs often urinated on the surface iButton stake, causing a spike in relative humidity for several hours. To avoid this, a cardboard shade was placed over the stake. Use of a similar shade caused minimal changes to the measurements in wombat (*Vombatus ursinus*) burrows at all times except midday (Shimmin et al. 2002). A motion-triggered camera (Bushnell HD NatureView model 119740, Kansas City, Michigan, USA) was placed 1–2 m in front of each den to monitor the entrance. Dens were monitored for approximately 145 h, after which equipment was disinfected and placed at another den.

Data collected included duration of iButton placement, location of iButton placement (within a den tunnel or within a chamber), whether the den was a complex (cluster of dens within a 5-m radius of one another) or single den (single entrance with the closest den being greater than 5 m away), whether water from irrigation sprinklers sprayed den entrances, number of dens within a complex, number of tunnel bifurcations within the den, den type (grass, earth, or manmade), and evidence of current SJKF activity (e.g., trash, paw prints). Data on irrigation sprinkler presence, presence of other animals, presence of human activity (humans or cars), SJKF presence, and evidence of mange on SJKFs were obtained from the cameras. Data for weekly rainfall for the city of Bakersfield was obtained from the National Oceanic and Atmospheric Administration (2018). We used the data for mite survival times to 100% mortality from Arlian et al. (1984, tables 1–4).

Statistical analysis

The program R (R Development Core Team 2018) was used for statistical analyses, and $P \leq 0.05$ was used to infer statistical significance. We fitted a linear regression model to the mite survival times from Arlian et al. (1984). The model of best fit was chosen on the basis of a minimized Akaike information criterion (Burnham and Anderson 2004), predicted residual error sum of squares, P value, and coefficient of determination (r^2). This model was used to predict expected mite survival time (EMST) for each den from observed temperature and humidity. As dens tended to be relatively consistent overall in their microclimates, seasonal averages for these variables were used to describe the individual dens.

A mixed effects linear model was used to assess variables that might predict EMST, controlling for the repeated variable of dens, with package nlme in R (Pinheiro et al. 2013). Covariates from camera data were not included in the analysis because of multiple camera failures. Remaining covariates were assessed for multicollinearity by chi-square or analysis of variance as appropriate, and collinear variables were omitted, leaving season, complex presence, and den type in the model. With the use of forward selection and the same selection criteria as previously, the model of best fit was chosen. The variables from this model and the remaining variables measured via remote camera (presence of humans or cars, SJKFs, and mangy SJKFs) were then analyzed individually by Student's t-test to determine the relative risk of these covariates to EMST.

RESULTS

Descriptive statistics

Ninety-two SJKF dens were assessed for temperature and humidity conditions, after the time frame for one was truncated and four dens were removed from the study because data indicated that den and surface conditions did not differ and iButtons were found at the entrance of the den, likely from removal by ground squirrels (*Otospermophilus beecheyi*). In the truncated den measurement, only 112 h of data was available because a child (seen on camera) removed the logger. The final dataset consisted of 44 dens in the summer and 48 dens in the winter, with 17 dens measured in both seasons. Of these measured dens, 64% (58/92) had documented proximal human activity (humans or cars). Fifty-two percent (48/92) of dens were dug in bare dirt, 27% (25/92) were dug under grass, and 21% (19/92) utilized manmade structures (e.g., shipping containers, human-made dens, drains). Irrigation sprinklers affected 52% (48/92) of the dens.

A SJKF was seen on camera at 74% (63/85) of the sites where cameras did not fail, and 9% (7/76) of cameras detected a SJKF visibly affected with mange. The mangy SJKF sightings, thought to represent the same individual based on the severity of mange and the presence of a collar, showed the animal routinely visiting as many as eight den complexes in the same area nightly. Two confirmed different SJKFs were observed on three occasions visiting the same den at intervals of 3 d, 2 d, and 3 h. Once during the winter, two SJKFs were seen visiting a den complex together. Foxes typically ignored the monitoring equipment, although there were some reactions, including urinating on the stake nightly and pulling on the shade intermittently for up to 3–5 h during the first night of placement, although foxes never successfully removed it. Striped skunks (*Mephitis mephitis*, n=68), California ground squirrels (n=42), cats (*Felis catus*, n=28), and opossums (*Didelphis virginianus*, n=18) were the most common other species observed using the dens. Less commonly, raccoons (*Procyon lotor*, n=3) or burrowing owls (*Athene cunicularia*, n=1) were seen. On five occasions, a SJKF was observed on camera cohabitating with skunks.

Linear model of mite survival data

Mean den temperature was 29 C (SD 3.5) in summer and 12 C (SD 2.4) in winter (Table 1). Relative humidity of the dens in winter was 83% (SD 17.8%) and in summer 69% (SD 23.9%). Dens had significantly different microclimates than the surface controls, tending

TABLE 1. Ambient temperature and humidity levels (SD) recorded by iButton data loggers (Maxim Integrated, San Jose, California, USA) inserted into San Joaquin kit fox (*Vulpes macrotis mutica*) dens in Bakersfield, California, in summer (July–September 2017) and winter (November 2017–January 2018).

	Summer	Winter
No. of dens observed	44	48
Surface temperature mean (C)*	30.9 (3.6)	9.7 (2.7)
Surface temperature maximum (max, C)	62	42
Surface temperature minimum (min, C)	10	−10
Deep temperature mean (C)*	28.9 (3.5)	11.9 (2.4)
Deep temperature max (C)	39	28
Deep temperature min (C)	19	1
Surface relative humidity mean (%)	49.4 (18.4)	78.4 (8.7)
Surface relative humidity max (%)	100	100
Surface relative humidity min (%)	3	33
Deep relative humidity mean (%)	69.4 (23.9)	82.5 (17.8)
Deep relative humidity max (%)	100	100
Deep relative humidity min (%)	0.3	24.5
Estimated depth placed (m)	1.9 (0.6)	1.5 (0.5)
Mean weekly rain (cm)	0 (0)	0.09 (0.2)

* $P \leq 0.001$.

to have more consistent temperature ($P<0.001$) and high relative humidity ($P<0.001$; Fig. 1).

An additive model of log(days surviving+1) as the response variable was the best fitting model to account for the mite survival data collected by Arlian et al. (1984; Table 2). The model had adjusted r^2=0.88 and $P<0.001$. The linear equation was:

$$\log(\text{days of survival time} + 1) = 2.33 - 0.058X_{\text{temperature}} + 0.006X_{\text{RH}}.$$

This model indicated that a positive relationship between increasing relative humidity and increasing EMST by a factor of 0.006 on a log scale, as well as an inverse relationship with increasing temperature and decreasing EMST by a factor of 0.058. The model confirmed that EMST was maximized with decreasing temperature and increased relative humidity, as suggested (Arlian et al. 1984; Arlian and Morgan 2017). Both variables together had a significant effect on EMST and explained 87.7% of the variation ($P<0.001$; r^2 =0.88; Table 2).

Predictors of mite survival times

For all dens, the mean EMST was 4.79 d (SD 2.97) with a range of 0.52–9.92 d. The mean EMST was 1.97 d (range 0.54–3.84, SD 0.92) in the summer and 7.38 d (range 4.36–

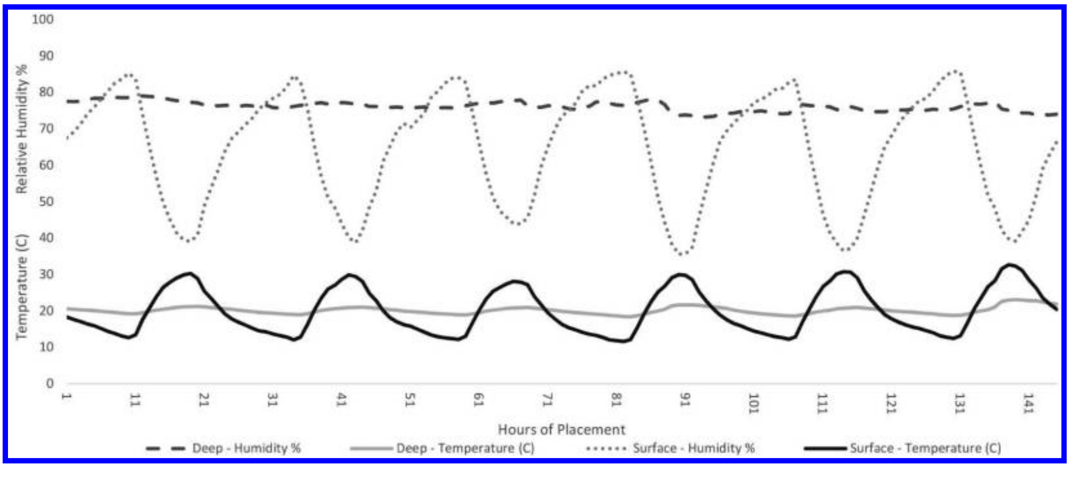

FIGURE 1. Hourly mean temperature and humidity in San Joaquin kit fox (*Vulpes macrotis mutica*) dens over 6 d for summer and winter seasons combined, Bakersfield, California.

TABLE 2. The best fitting linear model that accounts for female *Sarcoptes scabiei* mite survival times based on survival data and temperature and relative humidity (published in Arlian et al. 1989).

	Coefficient	SE	t	P
Intercept	2.333943	0.162200	14.39	<0.001
Temperature	−0.058332	0.003975	−14.68	<0.001
Relative humidity	0.005667	0.001967	2.88	0.007

Relative SE=0.307 (df=29), adjusted R^2=0.8773, $F_{2,29}$=111.8. P=2.336×10^{-14}, predicted residual error sum of squares=3.334668, Akaike information criterion=–72.735.

9.92, SD 1.41) in winter (Fig. 2A). Plots of depth of iButton placement within the den and EMST revealed no discernible relationship (Fig. 2B). There was also no association between SJKF presence (t=0.70, df=36.2, P=0.4862), human activity (t=–1.16, df=58.5, P=0.251), or mangy SJKF presence and the EMST (t=–0.62, df=6.9, P=0.557). However, grass dens had a higher mean EMST compared with other den types (Fig. 2C). Dens affected by sprinklers had a slightly higher average EMST than dens without sprinklers (Fig. 2D). An additive model using dummy

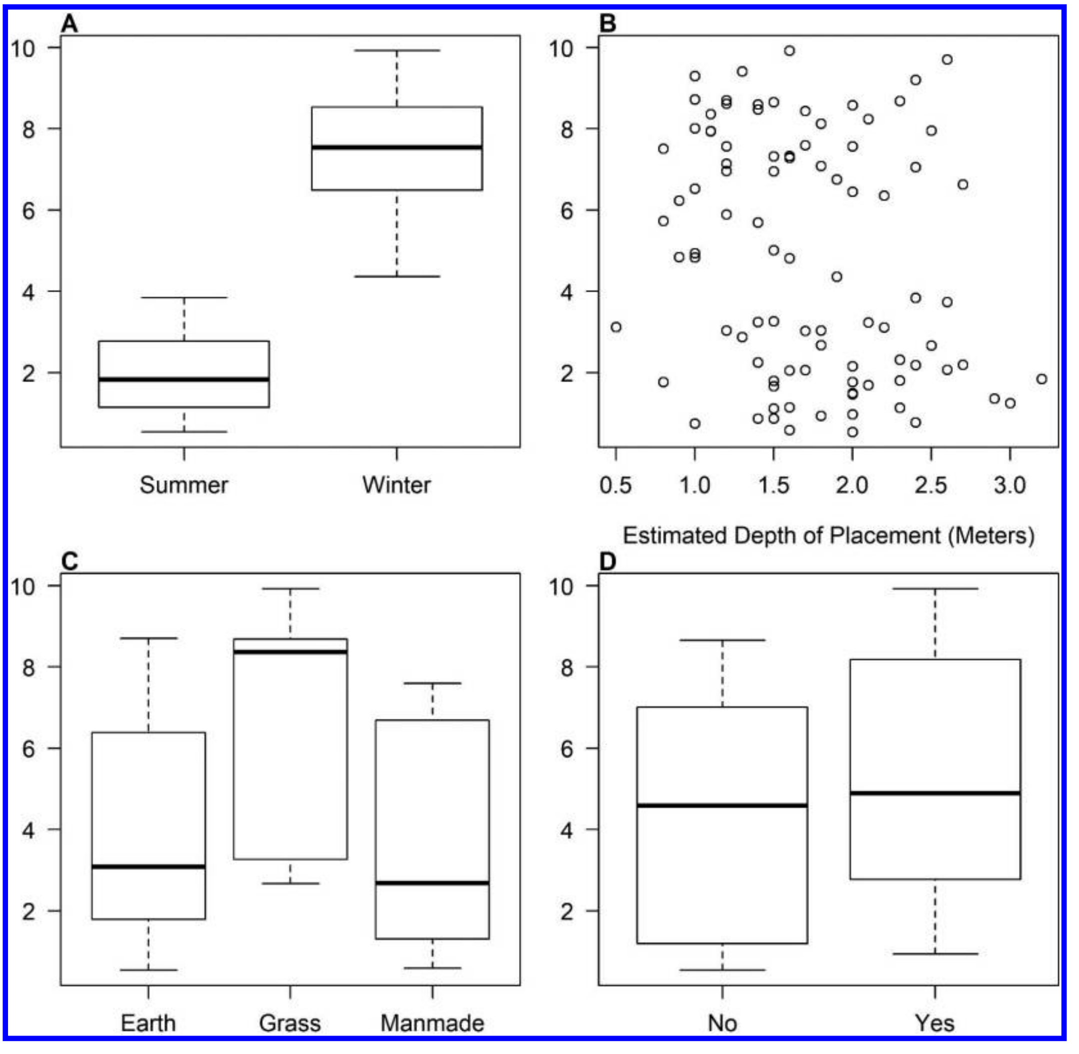

FIGURE 2. Plots of estimated survival time of *Sarcoptes scabiei* in San Joaquin kit fox (*Vulpes macrotis mutica*) dens of different conditions in Bakersfield, California. (A) Winter vs. summer; (B) whether dens were affected by water sprinklers; (C) at different iButton insertion depths; and (D) in different types of den.

variables for season and the den type had the best fit for predicting EMST. The assumptions for normality and evenly distributed residuals were tested and passed for this model. The presence of a den complex did not add significant information to the models. The final model was:

$$\begin{aligned}\mathrm{EMST} = 1.66 + 5.01X_{\mathrm{winter}} + 1.83X_{\mathrm{grassden}} \\ + 0.023X_{\mathrm{manmade}} + \mathrm{error(den)}.\end{aligned}$$

The model indicated that in a baseline earthen den in the summer, a mite will survive for an estimated 1.66 d. The covariate of winter would significantly increase EMST by 5.01 d ($P \leq 0.001$), and a grass den would significantly increase EMST by 1.83 d ($P \leq 0.001$) compared with an earthen den. The EMST in manmade dens was not significantly different from the baseline den ($P=0.93$; Table 3). Across all data, the model predicted that mites would survive maximally in grass dens in winter, whereas earthen dens in summer would be least likely to support mite survival.

TABLE 3. The best fitting linear mixed effects model using season and San Joaquin kit fox (*Vulpes macrotis mutica*) den construction type in Bakersfield, California, to explain estimated mite survival time interpolated from linear model of data (Arlian et al. 1989).

	Coefficient	SE	*t*	*P*
Intercept	1.661548	0.1622602	10.24	0
Season: winter	5.010295	0.1790319	27.99	0
Construction: grass	1.827845	0.2378466	7.69	0
Construction: manmade	0.023247	0.2703926	0.09	0.93

Akaike information criterion=257.35559, Bayesian information criterion=272.2199, log likelihood=–122.678.
Akaike information criterion=–72.735.

DISCUSSION

Although the primary method of transmission of mange among Bakersfield SJKFs is unknown, the obligate use and serial switching of dens by this species makes transmission within dens via fomites a plausible route of transmission. We found evidence that SJKF dens can support off-host survival of mites for approximately 5 d, identified risk factors for a den's ability to support prolonged off-host mite survival, and generated information on den ecology for this endangered species.

Previous work evaluating *S. scabiei* mite infectivity established that these mites can remain infective for approximately 36 h, or roughly 50% of the time they can survive off-host, dependent on environmental conditions (Arlian et al. 1984). Given the observed mean EMST predicted for SJKF dens, mites in dens might remain infective for at least 36–57 h. This infectious period may be even longer, because our estimates of EMST are conservative. For example, the depths to which we were able to insert iButtons in dens was limited, and conditions may be even more suitable for mites at greater depths, although our data did not indicate a relationship between depth of placement and environmental conditions. There was a wide range of EMST in dens, with season and den type being the most significant factors affecting mite survival time in the dens. Season had a particularly strong effect, with longer EMST in winter when temperatures are cooler and humidity is higher. Given this result, we might expect an increase in mange cases after winter. However, such a spike in cases would be difficult to detect, because of a not well-described period of latency between the time the SJKF is exposed until the disease is clinically observable. Additionally, transmission is likely influenced by increased contact rates that occur during spring breeding. The influence of season on mange infestation has been strongly supported in other wildlife populations, including the ibex (*Capra pyrenaica*) of Spain, coyotes (*Canis latrans*) in southern Texas, and raccoon dogs (*Nyctereutes procyonoides*) in Japan (Pence and Windberg 1994; Perez et al. 1997; Shibata and Kawamichi 1999). For ibex and raccoon dogs, this phenomenon was attributed to having a higher number of susceptible juveniles or a decline of cases in summer because of mortalities in winter (Perez et al. 1997; Shibata and Kawamichi 1999). In contrast, we suspect that the cool wet winters are the most important causal factor associated with mite survival and possible increases in SJKF cases.

The second factor we observed influencing EMST was the type of den. In the urban environment, SJKFs build dens in a variety of locations and substrates, including bare dirt, grass (e.g., irrigated lawns), or manmade structures. While manmade and earthen dens had similar EMST, grass dens had significantly higher EMST, likely because of sprinklers and shade from trees and buildings in the proximity of grass. The presence of sprinklers was in fact associated with longer EMST and correlated with grass dens. Increased moisture in the soil and air around dens likely increases den humidity and thus the EMST. Increased rainfall is associated with increased number of mange cases in Spanish ibexes (Perez et al. 1997). The presence of shade structures was not assessed in this study, but a correlation could be expected because grass, trees, and buildings are a common grouping in human development. Their increased presence would help decrease the ambient temperature of the local environment and thus decrease the temperature within the den, allowing mites potentially to live longer off-host. This factor could be playing a large role in this epidemic because the disease does not presently extend past the city boundaries. Further surveillance of the microclimate within the exurban dens would help assess this hypothesis.

Our study indicated that dens, particularly those in locations with irrigated grass and in the winter, could maintain mite off-host survival. These findings supported the hypothesis that SJKF dens could support pathogen persistence and propagation within the Bakersfield population, which suggested that certain targeted intervention strategies might aid in the management of the SJKF epidemic in Bakersfield. In black-tailed prairie dog (*Cynomys ludovicianus*) populations, dusting with acaricides for the plague flea vector of burrows has appeared to protect these populations from nearby outbreaks (Barnes et al. 1972; Seery et al. 2003). In wombat populations, medicated flaps placed over burrows to treat for sarcoptic mange did not result in a significant reduction in disease prevalence among wombats (Old et al. 2018). Another method of treating the dens may be through forced heated air applied for a prolonged period, which will desiccate mites and may be an effective treatment (Arlian and Morgan 2017). Given that the microclimate of manmade dens is not significantly different from that of natural earthen dens, use of such artificial dens (e.g., to route fox migration away from highly trafficked urban environments or to replace destroyed dens) is not increasing risk of mange. Furthermore, increasing the prevalence of artificial dens may also decrease the use of grass dens, thereby decreasing the potential for environmentally driven transmission of this mite. The use of these targeted interventions could make environmental control of this epidemic within this population a reality.

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