

## A PLAGUE EPIZOOTIC IN THE WHITE-TAILED PRAIRIE DOGS (*CYNOMYS LEUCURUS*) OF MEETEETSE, WYOMING

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**ABSTRACT:** Surveillance for sylvatic plague (*Yersinia pestis*) was conducted near Meeteetse, Wyoming (USA) from 24 May to 14 June 1985. Ten species of fleas were collected from white-tailed prairie dogs (*Cynomys leucurus*), and from their burrows and associated rodents. Five of these flea species and two adult prairie dogs were positive for plague. The progression of this plague epizootic appeared to be slower and the intensity was less than in previous epizootics in other prairie dog colonies. The plague epizootic occurred within the only known colony of black-footed ferrets (*Mustela nigripes*) and was a potential threat to the food source of this endangered species.

**Key words:** Plague, prairie dogs, fleas, Wyoming, rodents, *Yersinia pestis*, *Cynomys leucurus*, survey.

### INTRODUCTION

Plague is a flea-transmitted infection of rodents caused by the bacterium *Yersinia pestis*. The bacterium is maintained in rodent reservoir species (Poland and Barnes, 1979), but it also can survive in the soil of deserted burrows (Christie, 1982). Disease and disease-related declines among prairie dogs (*Cynomys* spp.) are reported. Although tularemia is important (Davis, 1935), the most devastating disease for prairie dogs is sylvatic plague (Barnes, 1982). Plague epizootics among the four species of prairie dogs (*C. gunnisoni*, *C. ludovicianus*, *C. leucurus* and *C. parvidens*) have been reported from different localities throughout the western United States (Lechleitner et al., 1968; Barnes et al., 1972; Clark, 1977; Barnes, 1982; Rayor, 1985) and thus all species appear to be highly susceptible to plague.

Mortality in a prairie dog colony may exceed 99% during a plague epizootic (Barnes, 1982; Rayor, 1985). These epizootics may be sporadic and localized in small prairie dog colonies, but in large, continuous colonies they may sweep across hundreds of km<sup>2</sup>. Colonies almost eradicated by the disease require 4 to 5 yr to

regain their former population sizes and then they will again become receptive to plague epizootics (Barnes, 1982). During a plague epizootic in a prairie dog population, the number of plague-infected, free-living fleas in the environment increases. Also sick and dead prairie dogs attract carnivores and raptors which act as flea carriers, spreading plague to other areas.

Plague is not new to the state of Wyoming; it was first reported in 1936 from a flea pool collected from an Uinta ground squirrel (*Spermophilus armatus*) trapped in Yellowstone National Park (Quan, 1982). Plague has since been detected in 11 counties among a variety of species including rodents, rabbits, and a mule deer (*Odocoileus hemionus*), as well as fleas, lice, and ticks collected from them (Table 1). Serologic evidence of plague infection in carnivores has been found also in three Wyoming counties (Centers for Disease Control, 1976-1982). A plague epizootic was observed in a population of white-tailed prairie dogs (*C. leucurus*) in 1968 in southeastern Wyoming (Clark, 1977).

Black-footed ferrets (*Mustela nigripes*)

TABLE 1. Distribution of animals and fleas positive for plague in the state of Wyoming from 1936 to 1985.\*

Location	Host/vector	Year
Yellowstone National Park	<i>Spermophilus armatus</i>	1936
Lincoln	"fleas" <sup>b</sup>	1938
Uinta	"fleas"	1938
Sweetwater	"fleas"	1939
Park	"fleas"	1940
Carbon	"fleas"	1943
Johnson	"fleas"	1943
Albany	"fleas"	1945
Laramie	"fleas"	1948
Albany	<i>Cynomys leucurus</i>	1968 <sup>c</sup>
Laramie and Niobrara	<i>Sciurus niger</i> / <i>Orchopeas howardi</i> <i>Spermophilus richardsoni</i> / <i>Opisocrostitis labis</i>	1976
Laramie and Niobrara	Dogs and <i>Canis latrans</i>	1977
Washakie	<i>Sylvilagus</i> spp. and human	1978
Park, Bighorn, Freemont, Laramie and Washakie	Dogs and <i>Canis latrans</i>	1979
Niobrara	<i>Odocoileus hemionus</i>	1980
Laramie	<i>Neotoma cinerea</i>	1980
Laramie	<i>Spermophilus tridecemlineatus</i> , domestic cat and human	1982
Meeteetse (present study)	<i>Cynomys leucurus</i> / <i>Opisocrostitis tuberculatus</i> <i>cynomuris</i> , <i>O. labis</i> , <i>O. idahoensis</i> , <i>Neopsylla inoptina</i> , <i>Rectofrontia</i> <i>fraterna</i>	1985

\* Data prior to 1985 from Centers for Disease Control, 1976–1982.

<sup>b</sup> Species not identified.

<sup>c</sup> Clark, 1977.

were discovered in a complex of white-tailed prairie dog towns in the Big Horn Basin of northwestern Wyoming in 1981 (Clark et al., 1985). White-tailed prairie dogs provide the main prey for this ferret population which was estimated to contain about 128 animals in 1984 (S. C. Forrest, unpubl. data). Because of the potential for the occurrence of plague in this area of Wyoming, surveillance for the identification of possible flea vector species and for the presence of plague in the white-tailed prairie dog population was conducted from 24 May to 14 June 1985. This report describes the results of that surveillance program.

#### MATERIALS AND METHODS

The study sites are located in the Big Horn Basin, Park County, Wyoming (43°50' to 44°15'N, 109°00' to 109°40'W). The black-footed ferret management area is approximately 29 km west of the town of Meeteetse (44°07'N, 109°07'W). The area, about 3,000 ha, contains 33 *C. leucurus* colonies (Fig. 1) with an esti-

mated mean density of 3.8 prairie dogs per ha (Clark et al., 1985) and is dominated by shrubs (*Artemisia tridentata*) and junegrass (*Koeleria cristata*) (Forrest et al., 1985). The average annual precipitation is 30 cm and mean monthly temperatures range from -6 to 7 C. Elevation ranges from 1,980 to 2,290 m (Forrest et al., 1985).

Fleas were collected from burrows with flannel squares (30 cm<sup>2</sup>) attached by a spring-loaded clip to the end of a cable 2 m long and 1.3 cm in diameter. When possible, cables were inserted their full length into the burrows and then withdrawn (Barnes et al., 1972). Each flannel square was immediately placed into a separate plastic bag. An attempt was made to obtain 10 flannel squares with fleas from each of six randomly selected prairie dog colonies. The plastic bags containing the flea samples were placed at -20 C for 20 min to inactivate the fleas so they could be collected from the flannel squares and placed in vials with field solution containing 2% sodium chloride and 0.001% Tween 80 (Barnes et al., 1972; Quan et al., 1981).

Fleas also were collected from rodents, trapped with Sherman and National live traps, by placing the rodents in an anesthetizing jar containing ether or by anesthetizing them with 18 mg/kg of ketamine hydrochloride (Ketaset, Bristol

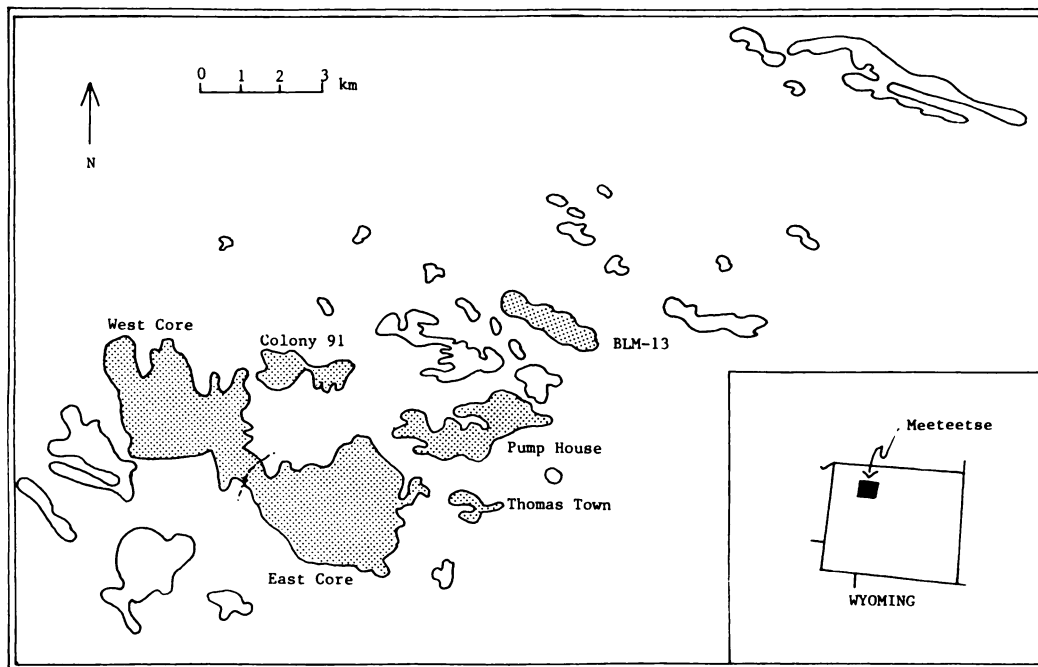


FIGURE 1. Distribution of white-tailed prairie dog (*Cynomys leucurus*) colonies in the Meeteetse complex, Park County, Wyoming (shaded areas indicate colonies surveyed during May and June 1985; map modified from Forrest et al., 1985).

Laboratories, Syracuse, New York 13201, USA). Animals were combed in a white enamel pan and fleas were placed in vials with the field solution. The vials with fleas were kept at 4 C until the fleas were identified and pooled according to host and locality. Pools containing from one to 25 fleas were triturated in sterile saline solution. Small aliquots (0.1–0.5 ml) of flea suspensions were inoculated subcutaneously into 6-wk-old male white laboratory mice (outbred Swiss mice from a pathogen-free colony established in 1967 at the Division of Vector-borne Viral Diseases, Centers for Disease Control, P.O. Box 2087, Fort Collins, Colorado 80522, USA) which were held for 21 days and observed daily for morbidity and mortality. Specimens of spleen and liver from moribund mice were inoculated directly onto blood agar plates and observed daily for the presence of bacterial colonies. Bacteria were identified by conventional procedures (Quan et al., 1979). Impression smears were prepared from spleen and liver tissues from prairie dog carcasses found at the study sites for microscopic examination by fluorescent antibody staining techniques (Quan et al., 1981). The remaining tissues were triturated with sterile saline and sand and were processed for bacterial isolation in the same way as the flea suspensions.

Blood samples were taken from all of the rodents captured during the surveillance program and from some rodents captured during subsequent studies on the evaluation of the plague status in the prairie dog complex (6 July to 19 August 1987). A blood sample (0.2 ml) was taken from the suborbital sinus of small rodents with a capillary pipet and from the heart of prairie dogs with a 1-ml syringe and 22 gauge needle and was absorbed on filter paper strips. After air drying, the strips were placed in individually marked envelopes. At the laboratory, the samples were eluted from the filter strips by extracting overnight in borate buffer and tested for antibody against *Y. pestis* by the passive hemagglutination (PHA) test (Wolff and Hudson, 1974). The minimal test titers were 1:32 for whole blood samples.

## RESULTS

From 165 sampled burrows, 86 (52%) had fleas with a mean of  $7.5 \pm 2.5$  (mean  $\pm$  standard error) fleas per infested burrow. Thirteen (15%) of these burrows had fleas that were positive for plague (plague positive) and were located in BLM 13, Thomas Town and East Core prairie dog colonies

TABLE 2. Distribution of fleas by species collected from burrows of white-tailed prairie dogs in Meeteetse, Wyoming, May and June 1985.

Location of burrows	Number of burrows sampled	% of burrows with fleas	Number of fleas per location	Mean fleas/infested burrow	% of total fleas	Flea species							
						OTC	OL	NI	RS	OI	HD	TP	RF
<b>Pitchfork Ranch</b>													
West Core	25	40	33	3.3	5.1	13	16	2	0	2	0	0	0
East Core	75	48	140	3.9	21.6	35	78	19*	1	5	2	0	0
<b>91 Ranch</b>													
Colony 91	10	100	113	11.3	17.4	20	80	2	1	6	1	3	0
Pump House	20	60	122	10.2	18.8	12	100	4	3	0	2	1	0
<b>Art Thomas Ranch</b>													
Thomas Town	20	40	47	5.9	7.2	32	4*	4	0	6*	0	1	0
<b>BLM</b>													
BLM 13	15	67	194	19.4	29.9	136*	36*	12	0	5	1	1	3*
Total	165	52	649	7.5	100	248*	314*	43*	5	24*	6	6	3*
						(16) <sup>b</sup>	(10)	(17)	(42)	(286)			

\* Plague positive fleas.

<sup>b</sup> Minimum infection rate (number positive fleas per 1,000 tested). OTC, *Opisocrostitis tuberculatus cynomurris*; OL, *O. labis*; NI, *Neopsylla incipina*; RS, *Rhadnopsylla sectilis*; OI, *Oropsylla idahoensis*; HD, *Hystriropsylla dippieii*; TP, *Thrassis pandorae*; RF, *R. fraternus*.

(Table 2, Fig. 1). Eight hundred forty-five fleas were collected from burrows and animals; 649 (77%), representing eight species, were collected from burrows (Table 2). Prairie dog colony BLM 13 had the highest mean number of fleas collected ( $19.4 \pm 4.3$ /burrow) and the West Core colony had the lowest mean number ( $3.3 \pm 0.8$ /burrow).

Of the 10 species of fleas identified (Tables 2, 3), six were collected from both prairie dogs and their burrows. The two most abundant species collected from burrows were *Opisocrostitis labis* (48%) and *O. tuberculatus cynomuris* (38%) (Table 2). These same two species were the most abundant on prairie dogs (Table 3) with 32% and 50%, respectively. In addition, these two species of prairie dog fleas were collected from Uinta ground squirrels and deer mice (*Peromyscus maniculatus*). Also *Thrassis pandorae*, a common Uinta ground squirrel flea, was collected from prairie dog burrows in one of the colonies (Tables 2, 3). *Monopsyllus exilis*, collected from deer mice and grasshopper mice (*Onychomys leucogaster*), as well as *M. wagneri* collected from deer mice, were not found on any of the other rodent species or in prairie dog burrows.

Of the 10 flea species collected, five were plague positive. *Opisocrostitis tuberculatus cynomuris*, *O. labis*, and *Oropsylla idahoensis* collected from both prairie dogs and burrows were positive; their minimum infection rates (MIR, minimum number positive per 1,000 tested) were 24, 19 and 122 from prairie dogs (Table 3) and 16, 10 and 42 from burrows (Table 2), respectively. Plague positive *Neopsylla inopina* and *Rhadinopsylla fraterna* were collected from burrows and their MIR were 17 and 286, respectively (Table 2).

Of 89 rodents examined, 59 (66%) had fleas (Table 3). From these 59 rodents, 250 fleas were collected with a mean of  $4.6 \pm 0.6$  fleas per infested rodent. The 32 prairie dogs examined were from the East Core and Pump House colonies (Fig. 1). All of them were infested (mean of  $5.8 \pm 0.7$

TABLE 3. Numbers of fleas by species from rodents captured in Meeteetse, Wyoming, May and June 1985.

Host species	Hosts		Flea species									
	Number with fleas	% with plague + fleas	OTC	OL	NI	RS	OI	HD	MW	ME	TP	RF
<i>Cynomys leucurus</i>	32	28.1	85* (24) <sup>b</sup>	54* (19)	15	2	8*	1	1	0	0	4
<i>Peromyscus maniculatus</i>	53	0	0	2	0	0	0	1	40	16	0	0
<i>Spermophilus armatus</i>	2	0	1	4	0	0	0	0	0	0	8	0
<i>Onychomys leucogaster</i>	2	0	0	0	0	0	0	1	0	7	0	0
Total	89	15.3	86	60	15	2	8	3	41	23	8	4

\* Plague positive fleas.  
<sup>b</sup> Minimum infection rate (number positive fleas per 1,000 tested). OTC, *Opisocrostitis tuberculatus cynomuris*; OL, *O. labis*; NI, *Neopsylla inopina*; RS, *Rhadinopsylla sectilis*; OI, *Oropsylla idahoensis*; HD, *Hystriochopsylla ditpicti*; MW, *Monopsyllus wagneri*; ME, *M. exilis*; TP, *Thrassis pandorae*; RF, *R. fraterna*.

fleas per prairie dog) and 28% had plague-infected fleas. Only 45% of the 53 deer mice were infested ( $2.5 \pm 0.3$  fleas per mouse) and none of the 59 fleas of four species collected were plague positive. None of the 13 fleas of three species collected from one of the two ground squirrels nor the eight fleas from the two grasshopper mice were plague positive.

One adult and one juvenile prairie dog on the East Core site and one adult on Colony 91 were found dead during early June. Only the two adults were positive for plague. While sampling burrows on Colony 91, decomposed carcasses of prairie dogs were observed outside the openings of the burrows; none were seen in any of the other colonies. However, very low prairie dog activity was observed while sampling burrows in the West Core, Thomas Town and BLM 13 colonies (Fig. 1).

A total of 109 deer mice, 37 prairie dogs, four Uinta ground squirrels, two grasshopper mice and one olive-backed pocket mouse (*Perognathus fasciatus*) were tested for plague antibody. All 153 rodent sera were negative for PHA antibody.

#### DISCUSSION

Of the 10 species of fleas collected between 24 May and 14 June 1985, five were found *Y. pestis* positive and all of the infected fleas were collected from prairie dogs or their burrows. The two most abundant flea species during that period were *O. tuberculatus cynomuris* and *O. labis*. Their abundance together with their distribution and infection rate indicate that these two species were probably responsible for spreading plague. Because *Y. pestis* was isolated only from prairie dogs and their fleas and since no antibody positive rodents were found, particularly deer mice which were frequently captured within the prairie dog colonies, it appears that the plague epizootic was in the prairie dog population. During an epizootic in a Gunnison's prairie dog (*C. gunnisoni*) complex in Colorado (Lechleitner et al., 1968) and

in white-tailed prairie dogs in southeastern Wyoming (Clark, 1977), populations of other small mammals living in the area were unaffected by plague.

White-tailed prairie dogs are thought to be as susceptible to plague as the other species of prairie dogs (Barnes, 1982). The absence of detectable antibody in the 37 prairie dogs captured in the study area indicates a lack of resistance in this species to *Y. pestis* and further supports previous information about the high susceptibility of prairie dogs to the plague organism (Lechleitner et al., 1968; Poland and Barnes, 1979). Plague epizootics have eliminated or greatly reduced populations of prairie dogs within one or two seasons (Clark, 1977; Barnes, 1982; Rayor, 1985). This epizootic could have started in 1984 when the absence of prairie dogs in some colonies at Meeteetse was noticed and the epizootic continued into 1985.

The absence from this area even during the warmest part of the summer (G. O. Maupin, unpubl. data) of *Opisocrostis hirsutus*, which is a common prairie dog flea in the western United States (Hubbard, 1947; Maupin, 1970; Barnes et al., 1972; Barnes, 1982), and the presence of *O. tuberculatus cynomuris* and *O. labis* could have influenced the transmission rate. *Opisocrostis hirsutus* occurs in warmer regions of the country at moderate elevations up to 2,740 m, is most abundant during the summer season and is replaced by *O. tuberculatus cynomuris* during the winter (Maupin, 1970). *Opisocrostis hirsutus* is an efficient vector of plague in the warmer regions (Maupin, 1970; Barnes et al., 1972). The Meeteetse area has a short, cool summer season which would favor *O. tuberculatus cynomuris* and other related species over the summer vector *O. hirsutus*. The interaction of these factors might have reduced the intensity of the plague epizootic in the white-tailed prairie dog population near Meeteetse. In reality, a plague epizootic under these circumstances probably progresses more slowly over several years, although the end result

of almost complete depopulation could be the same.

Lechleitner et al. (1968) studied the progression of an epizootic of plague in a population of Gunnison's prairie dogs (*Cynomys gunnisoni*) over a 3-yr period in southern Colorado. At the end of the study only a few individuals remained in two of the seven original colonies of prairie dogs. The elevation of this study area ranged from about 2,800 to 3,300 m, and the vectors were *O. tuberculatus cynomuris*, *O. labis*, and *O. idahoensis*. These same three species were the principal vectors in the Meeteetse area and the common summer flea vector, *O. hirsutus*, was absent just as was found in Meeteetse. In another location of southern Colorado, Rayor (1985) observed an explosive plague epizootic which exterminated an entire colony of Gunnison's prairie dogs (1,000 to 1,500 animals) within 2 mo after spring emergence from hibernation. The flea species incriminated as vectors during this epizootic were *O. hirsutus* and *O. tuberculatus cynomuris*. The results from the two previously studied plague epizootics and the 1985 epizootic in Meeteetse suggest that the flea vector species and/or environmental characteristics can influence the intensity and duration of an epizootic of flea-transmitted plague in prairie dog populations. The plague epizootic in white-tailed prairie dogs in southeastern Wyoming extended over two seasons (1967 to 1968) and reduced the population by more than 85% (Clark, 1977); however, the flea vector species were not determined during this epizootic.

There is no information on the susceptibility of black-footed ferrets to plague although other carnivores including the striped skunk (*Mephitis mephitis*) were found to suffer little or no morbidity to oral challenge (Rust et al., 1971; Barnes, 1982). The ferrets in their normal movement patterns were most likely involved in disseminating the disease by transporting infected fleas within and between prairie dog colonies. The ferrets could be af-

ected indirectly if a plague epizootic such as this continued in the prairie dog complex and reduced the prey base to below that required to maintain a healthy black-footed ferret population.

Because of the enzootic nature of the disease in Wyoming, continued surveillance for plague in the white-tailed prairie dog complex of Meeteetse will be necessary to monitor the presence and spread of plague. Also, it will be necessary to evaluate the disease management efforts which were conducted from July to October 1985, the results of which will be reported separately, in order to protect the food source of the remaining ferret population. In addition, if other colonies of black-footed ferrets are discovered, early and intensive surveillance for plague and other pathogens that might directly or indirectly affect the ferrets should be conducted.

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