

first observations suggest that villages with a high plague incidence are connected through typical fertile valley bottoms, i.e. Gleysols and Fluvisols, and that hamlets (part of a village) in this valley bottom have had more human plague cases. Soil and plant samples are being analyzed to test if factors that define the microclimate (in this study, bulk density, soil texture, pH, and organic carbon, and concentrations of chemical elements in soil and plant) are linked with plague occurrence in Lushoto. Our results give an indication that a landscape ecological study approach can provide insights into the persistence of plague and how its distribution can be affected by landscape features, and therefore in this case, might open the track towards a better understanding of the underlying ecology of plague's distribution in Lushoto.

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PRELIMINARY STUDY OF FLEAS ON RODENTS IN THREE COLORADO COUNTIES

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Some species of fleas are found on multiple species of rodents, thus increasing the likelihood of spreading disease from one host to another. As part of a preliminary study, fleas were collected off live-trapped and kill-trapped rodents from October 2003 through August 2007 in Elbert, El Paso, and Pueblo Counties, Colorado. Twelve species of fleas representing three families were collected off nine species of rodents. The two most common species of fleas, *Aetheca wagneri* and *Orchopeas leucopus* were found on four species of mice (*Peromyscus* spp.), Mexican woodrats (*Neotoma mexicana*) and rock squirrels (*Spermophilus variegatus*). *Oropsylla montana* was found in high numbers on rock squirrels and, because of its high incidence, is an important vector in plague transmission (Lewis 2002). In addition, we found small numbers of *Hoplopsyllus anomalus*, a known plague vector, on rock squirrels. We also found high numbers of *Foxella ignota*, a primary parasite of northern pocket gophers (*Thomomys talpoides*), which is a minor vector of plague (Pigage et al. 2005). Although pocket gophers are frequently found in the same areas as other rodents, we did not collect *F. ignota* from any other host. Our current goal is to investigate rodent populations that live in or near black-tailed prairie dog (*Cynomys ludovicianus*) colonies and their fleas as well as some predators of the prairie dogs in order to examine flea exchange.

References:

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DETERMINATION OF THE BLOOD TITER LEVELS OF IMIDACLOPRID AND EFFECTIVENESS AGAINST *XENOPSYLLA CHEOPIS* FLEAS ON LABORATORY RATS (*RATTUS NORVEGICUS*)

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Rats have been long considered a reservoir host of flea-borne diseases, especially plague. Systemic insecticides commonly used for flea control in veterinary medicine could also be applied to control flea populations on rats. Imidacloprid [1-(6-chloro-3-pyridylmethyl)-N-nitroimidazolidin-2-ylideneamine] is a versatile and effective insecticide with diverse and growing number of applications ranging from agricultural uses to the treatment of pets and control of household pests. We report here the evaluation of imidacloprid as an effective insecticide to systemically control *Xenopsylla cheopis* fleas on laboratory rats and thus to mitigate flea-borne diseases. A high performance liquid chromatography (HPLC) method with reverse phase separation for determining imidacloprid level in blood of rats was developed. Imidacloprid was

detected by UV at 270 nm with the Limit of Detection at 0.018 µg/mL, Limit of Quantification at 0.051 µg/mL, and mean recovery of 97.6%. The method was validated for imidacloprid concentration range from 0.02 to 0.82 µg/mL. For testing imidacloprid as an insecticide, single doses of 0.2 mg (group 1) and 0.4 mg (group 2) were orally delivered to two treatment groups (n = 5) of rats. The control group (n = 3) was given an aliquot of pure solvent. After 3 hours of delivery ~20 fleas were applied to each rodent using flea chambers placed on animals. Fleas were allowed to feed for 3 hours and then removed. Blood was collected into 3 mL EDTA tubes. Liquid samples for HPLC were prepared by liquid-liquid extraction of imidacloprid from blood by dichloromethane. The imidacloprid level was found to be 0.47 ± 0.049 µg/mL and 0.89 ± 0.188 µg/mL for group 1 and group 2, respectively. No traces of imidacloprid were found in the control group. Flea mortality in group 1 was 78.0% and 80.5% after 24 hours and 48 hours of imidacloprid exposure, respectively. Flea mortality in group 2 was 72.2% and 77.8% after 24 hours and 48 hours of imidacloprid exposure, respectively. Mortalities of fleas in the control group were 10.5% and 12.6% respectively. The reported HPLC/flea application method allows determination of the effective imidacloprid doses that need to be delivered to host rats to reach effective imidacloprid concentrations in host blood and lethally control *Xenopsylla cheopis* fleas.

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SOCIO-ECONOMIC RISK FACTORS ASSOCIATED WITH HUMAN PLAGUE CASES IN NEW MEXICO

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Plague, caused by the bacterium *Yersinia pestis*, is a zoonotic disease that is rare in the United States, but can be highly fatal in humans, when left untreated. The majority of human cases in recent decades have been acquired in New Mexico. A recently developed habitat suitability model for the disease identified areas which support a diverse assemblage of rodent hosts for plague as those most at risk for human infections in this state (Eisen et al. 2007). Here, we combine known environmental risk factors with socio-economic features of U.S. census block groups in a Geographic Information Systems (GIS) model to further refine the area within New Mexico which poses the highest plague risk for humans. The socio-economic risk factors identified included proportion of housing units with incomplete plumbing, proportion of housing units built 40 or more years prior to a census, and poverty rate. The overall accuracy of our model was about 82%, and reduced the area considered at highest risk from about 17% to between 2 and 7% of New Mexico. This reduction in the area identified as high-risk highlights the potential importance that human behavioral or lifestyle factors play in *Y. pestis* infection risk, particularly when these factors are ones that might influence the likelihood of native rodent species invading peridomestic habitats. Moreover, such integration of environmental and socio-economic risk factors may aid in the targeting of limited public health resources to areas where prevention and control efforts may be most effective.

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SEASONAL AND SPATIAL CHANGES IN FLEA COMMUNITIES OF BLACK-TAILED PRAIRIE DOGS OF NORTHWESTERN MEXICO

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Host population structure and density, and changes in flea community structure and composition, are considered to be important in the dynamics of vector borne diseases in prairie dogs, including plague and tularemia. In Mexico, neither tularemia nor plague is recognized despite the presence of environmental conditions similar to those

found in the southern USA where these diseases are frequently reported. We carried out a survey of fleas parasitizing black-tailed prairie dogs at 8 160×160 m quadrants in 4 prairie dog colonies in northwestern Chihuahua, Mexico during the dry and rainy seasons of 2006. The colonies of the survey are included at the core of one of the largest black-tailed prairie dog colony complexes in North America, and differ in size, isolation degree of isolation and host density. Fleas were collected directly from the pelage of the trapped prairie dogs, along with some blood samples to search for evidence of plague or tularemia infection. All captured animals were measured and released at the site of capture. We trapped 51 prairie dogs and collected 119 fleas belonging to 5 flea species, including *Echidnophaga gallinacean*, *Pulex simulans*, *Pulex sp.*, *Thrassis fatus*, and *Oropsylla hirsuta*. *Pulex* was the dominant genus among the prairie dog colonies surveyed, comprising 57% of the total fleas, followed by *Oropsylla hirsuta* with 38%. Seasonal changes in flea community structure were recorded. During the dry season, *Oropsylla hirsuta* was the dominant flea, comprising 42% of all fleas collected, followed by *Pulex* spp (31 %) and *Pulex simulans* (24%). During the wet season, *Pulex simulans* dominated the flea community with 70% occurrence, followed by *Pulex* spp. with 20%. The smallest and most isolated colony of prairie dogs exhibited the lowest diversity of flea species. Changes in flea community structure due to changes in host densities, degree of isolation and changes in flea communities due to seasonal changes may produce changes in vector competence for infectious agents. These changes also may create a different pattern of disease occurrence between prairie dog colonies from northern Mexico and southern USA. Recognizing the role of flea communities, vector competence and host population structure is the key to understanding and predicting disease outbreaks. Further analyses are needed, including molecular and serologic tests for plague and tularemia in fleas and prairie dogs of northern Mexico. In future studies, we will assess the relationship of flea community dynamics and disease prevalence in both fleas and hosts with different spatial and temporal scales. The results of this research will contribute to the development of predictive models for prairie dog colonies in northern Mexico.

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ECOLOGY OF RODENTS AND FLEAS ASSOCIATED WITH BLACK-TAILED PRAIRIE DOGS IN AREAS WITH PLAGUE

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The aim of this study was to identify rodent-flea complexes that might be important in the transmission and maintenance of plague in the black-tailed prairie dog (*Cynomys ludovicianus*) ecosystem. We characterized the relationship between fleas and their rodent hosts in the presence of prairie dog colonies (referred to as on-colony grids) and compared them to adjacent assemblages in the surrounding grasslands (referred to as off-colony grids). We evaluated the rodent-flea relationship by quantifying prevalence (proportion of rodents infested with fleas) and estimated the probability of infestation based on observed prevalence, flea load (number of fleas per infested rodent), and mean intensity of fleas on rodents (number of fleas of a given species per infested rodent). Because prairie dog burrows provide refugia for fleas, we hypothesized that prevalence, flea load, and intensity would be higher for rodents that are associated with black-tailed prairie dog colonies. Rodents were trapped at off- and on-colony grids, resulting in the collection of 4,509 fleas from 1,430 rodents in six study areas. The rodent community composition varied between these study areas. Flea species richness was not different between prairie dog colonies and the surrounding grasslands ($p=0.883$) but was positively correlated with rodent species richness ($p=0.055$). Prairie dog colonies did not increase the prevalence of fleas ($p>0.10$). Flea loads on rodents did not vary between off- and on-colony grids at three of the study areas ($p>0.10$). Based on the prevalence, infestation rates, and flea loads, we identified *Peromyscus maniculatus*, *Onychomys leucogaster*, and two *Neotoma* species as important rodent hosts for fleas and *Aetheca wagneri*, *Orchopeus leucopus*, *Peromyscopsylla hesperomys*, *Pleochaetis exilis*, and *Thrassis fatus* as the most important fleas associated with these rodents. These rodents and fleas have been implicated in plague (Biggins and Kosoy 2001, Gage and Kosoy 2005) and their presence in areas without a known history of plague suggests that the current distribution of plague is not limited by the distribution of these rodents and fleas. Prairie dog colonies did not seem to facilitate transmission of fleas between rodent hosts, and the few rodent-flea associations exhibited significant differences between off- and on-colony grids. How-

ever, the presence of prairie dog fleas on rodents at both off- and on-colony grids suggests the potential for intra and interspecific transmission of fleas between rodent hosts, and between rodents and prairie dogs.

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FLEA LOADS ON BLACK-TAILED PRAIRIE DOGS (*CYNOMYS LUDOVICIANUS*) DURING PLAGUE EPIZOOTICS IN COLORADO

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Plague, caused by the bacterium *Yersinia pestis*, is primarily a disease of wild rodents and their fleas. Black-tailed prairie dogs (*Cynomys ludovicianus*) are highly susceptible and mortality on individual towns often reaches 100%. Flea load, or the number of fleas per host, fluctuates seasonally and transmission of the pathogen during epizootics is likely to become more efficient as flea load increases. Fleas were collected from live-trapped black-tailed prairie dogs on towns before and during plague epizootics and tested by PCR for the presence of *Y. pestis* DNA. The predominant fleas infesting Black-tailed prairie dogs were *Oropsylla hirsuta*, *Oropsylla tuberculata cynomuris*, and *Pulex simulans*, with greatest flea abundance occurring in March and October. Flea load and infestation intensity increased during epizootics and was highest on prairie dogs with *Y. pestis*-infected fleas. The seasonal occurrence of epizootics among black-tailed prairie dogs was found to coincide with seasonal peaks in mean flea load.

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PLAGUE ACTIVITY IN CALIFORNIA: A SUMMARY OF STATEWIDE PUBLIC HEALTH SURVEILLANCE, 1984-2007

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Yersinia pestis, the causative bacterial agent of plague, entered California by way of infected rats and humans who disembarked at the major port cities during the early 1900s. Over the succeeding decades, plague rapidly adapted to indigenous wildlife and spread throughout the western United States. Since 1930 the California Department of Public Health (CDPH) has maintained an integrated statewide plague surveillance program that encompasses investigations of clinical plague in humans and domestic felids, evaluation of epizootic activity in rodents, and serologic monitoring of wild carnivores. This poster provides a 24-year summary of surveillance data collected in California from 1984 through 2007: Twenty-four human cases of plague occurred from 14 counties, two of which were fatal. *Y. pestis* was isolated from 82 domestic pets in 12 counties. We recorded at least 68 epizootic events among wild rodents in 21 counties. Rodent species most frequently involved in plague maintenance and transmission were California ground squirrels (*Spermophilus beecheyi*), Douglas' squirrels (*Tamiasciurus douglasii*), shadow chipmunks (*Tamias senex*), and yellow-pine chipmunks (*Tamias amoenus*). Carnivores frequently detected with serum antibodies to *Y. pestis* were black bears (*Ursus americanus*), bobcats (*Lynx rufus*), and coyotes (*Canis latrans*). Animals evaluated for plague activity totaled 34,537 and provided 2,355 positive results. Through the use of this cooperative statewide plague surveillance program, CDPH has been able to respond effectively to human cases and epizootic activity when they occur. The program has also enabled CDPH to gain a greater understanding of the epidemiology of the disease.