Recent Trends in Plague Ecology

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Abstract

Plague (*Yersinia pestis* infection) presents serious risks not only to humans but also to wildlife species such as prairie dogs (*Cynomys* spp.) and the critically endangered black-footed ferret (*Mustela nigripes*). The effects of plague are sufficiently serious to hamper recovery of ferrets and prairie dogs in areas that experience repeated epizootic activity. In order to more effectively manage and reduce plague risks for both wildlife and humans, we must improve our understanding of what factors influence the distribution of plague, the transmission and spread of epizootics, and the ability of the plague bacterium to maintain itself indefinitely in some populations of rodent hosts and their flea (Insecta: Siphonaptera) vectors. This article provides a review of our current knowledge of plague ecology. We also describe how recent research advances are providing significant new knowledge and methodologies that can help us better manage plague risks and reduce the impact of the disease on mammalian populations, including those of conservation interest.

Keywords: disease ecology, flea, plague, rodent, *Yersinia pestis*, zoonosis

Introduction

Plague is a flea-borne zoonotic disease caused by the bacterium *Yersinia pestis* (Gage, 1998). The disease is best known as the cause of devastating pandemics, including the Black Death of the Middle Ages. These same pandemics, as well as other more regional outbreaks, also provide striking demonstrations of plague's ability to spread rapidly across vast geographic areas, a process that occasionally results in the establishment of long-term foci of infection among suitable populations of susceptible mammalian hosts and competent flea vectors. At present, active plague foci are found in many countries in Asia, Africa, and the Americas (Gage, 1998; Tikhomirov, 1999; World Health Organization, 2004). In the United States, evidence of plague infection has been identified during recent decades in mammals or fleas in 17 western States (fig. 1).

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Although most evidence suggests that virtually any mammal exposed to *Y. pestis* is likely to become infected, the true vertebrate hosts are certain species of rodents (Pollitzer and Meyer, 1961; Gage and Kosoy, 2005). Plague-related mortality can vary greatly between rodent species and even among populations within the same species. In some rodent species mortality approaches 100 percent (Poland and Barnes, 1979). Although certain other rodents appear to be more resistant to plague, even supposedly resistant populations can experience mortality rates in excess of 40 percent (Rivkus and others, 1973). Mortality can also be high among various nonrodent species found naturally infected with *Y. pestis*. Wild and domestic felines, as well as some lagomorphs (hares, rabbits, and pikas), are extremely susceptible (Gage and others, 1994; Gage and Kosoy, 2005). Identification of high seropositivity rates among other nonrodent species, such as coyotes (*Canis latrans*), badgers (*Taxidea taxus*), and feral hogs (*Sus scrofa*), suggests that these species are at least moderately resistant to plague-related mortality (Gage and others, 1994). While most nonrodent species, with the exception of a few lagomorphs and the house shrew (*Suncus murinus*) of southeastern Asia and Madagascar, are not significant hosts of plague, certain mammalian predators and birds of prey probably play important ecological roles by transporting infected fleas from one region to another (Gage and others, 1994).

Elton's (1958) classic book on the ecology of invasions mentions plague as an example of an agent that can spread explosively across vast areas, infecting not only commensal rats (*Rattus* spp.) and "wild" rodents but also other mammals, including humans (Gage and others, 1995; Gage and Kosoy, 2005). Within the past two decades, an increasing number of biologists have become aware of the devastating effects plague has on certain mammal species of conservation interest (Biggins and Kosoy, 2001a,b). Mortality among infected black-tailed prairie dogs (*Cynomys ludovicianus*) reportedly approaches 100 percent during epizootics, and other prairie dog species (*Cynomys* spp.) also are quite vulnerable to the disease (Kartman and others, 1962; Lechleitner and others, 1962, 1968; Rayor, 1985; Ubico and others, 1988; Anderson and Williams, 1997; Cully, 1997; Cully and others, 1997, 2000; Girard and others, 2004; Stapp and others, 2004). Recent evidence also indicates that plague epizootics can cause significant reductions in genetic diversity among prairie dog populations (Trudeau and others, 2004). In some situations plague has both direct and indirect impacts on wildlife populations. Prairie dogs and their endangered predator, the

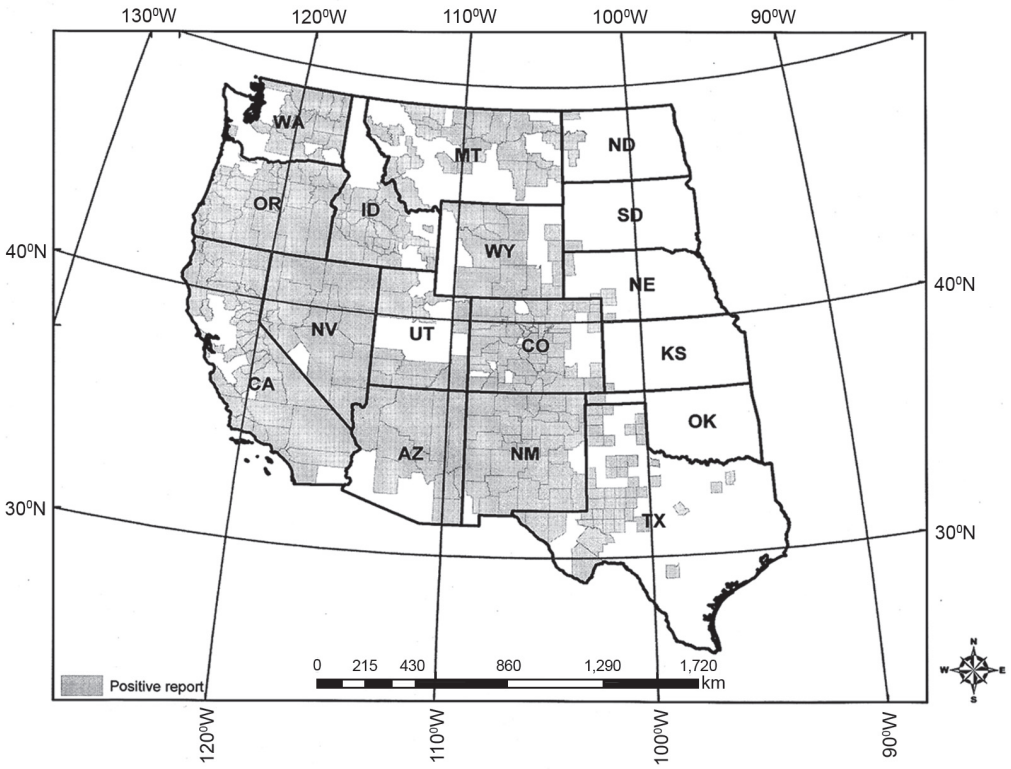

Figure 1. Counties with plague-positive mammals or fleas (1970–present). Figure courtesy of Centers for Disease Control and Prevention.

black-footed ferret (*Mustela nigripes*), are both severely affected by plague, and recovery efforts for black-footed ferrets are hampered not only by the fact that plague outbreaks eliminate the ferrets' prey but also because the ferrets themselves are extremely susceptible to the disease (Williams and others, 1994; Biggins and Kosoy, 2001b; Biggins and Godbey, 2003). The devastating impact of plague on these and other mammalian species of conservation interest has resulted in a renewed emphasis on identifying means for managing plague, including techniques as diverse as insecticidal control of vector fleas and immunization of animals with recombinant vaccines (Creekmore and others, 2002; Seery and others, 2003; Mencher and others, 2004; Rocke and others, 2004). Biggins and Godbey (2003) also discuss partial solutions to the problem of black-footed ferret recovery, including means for increasing breeding in captive populations, increasing survival of released animals, and taking advantage of South Dakota sites that are located slightly east of the known distribution of plague.

In order to more effectively manage and reduce human and wildlife risks associated with plague, we must improve our understanding of the factors that influence transmission, the occurrence and spread of epizootics, and the ability of plague to maintain itself in natural foci. This article provides a brief update on our current knowledge of plague ecology and describes how recent research has contributed to a better understanding of the topic and improved methodologies for studying plague. Also discussed are the many gaps in our knowledge of how plague is maintained in natural foci, what roles certain rodent and vector species play in transmission dynamics, and how environmental factors influence the occurrence, spread, and persistence of epizootics.

The Plague Bacterium and Its Origins

Yersinia pestis is a gram-negative bacterium belonging to the family Enterobacteriaceae. Unlike other members of this group, which are transmitted through fecal-contaminated food and water and live in the guts of their hosts, *Y. pestis* is typically spread from host to host through the bites of infectious fleas and inhabits the blood, as well as lymphatic and reticuloendothelial systems, of its hosts. This dramatic shift in mode of transmission and vertebrate host habitat appears to have been associated, at least in part, with the acquisition of genes that encode virulence and transmission factors. Homologous genes for some *Y. pestis* virulence factors can be found in other species of *Yersinia*, including *Y. pseudotuberculosis*. The origin of genes encoding other virulence or transmission

factors is not always clear, but most evidence suggests they were acquired through horizontal transfer of genetic material from other enteric bacteria (Prentice and others, 2001; Gage and Kosoy, 2005). The virulence factors of *Y. pestis* play important roles in enabling host invasion, dispersal within the host, or development of high level bacteremias that greatly increase the likelihood that blood-feeding fleas will imbibe sufficient *Y. pestis* to become infected and later transmit the plague bacterium to other hosts. *Yersinia pestis* transmission factors promote survival of the plague bacterium in the guts of vector fleas and its transmission by these insects. For a more thorough review of virulence and transmission factors and their role in maintaining the natural transmission cycle of plague, see reviews by Perry and Fetherston (1997), Hinnebusch (2003), and Gage and Kosoy (2005).

Until relatively recently, it was believed that the plague bacterium first appeared many millions of years ago, perhaps as early as the upper Oligocene or lower Miocene (Kucheruk, 1965). According to Kucheruk (1965), plague initially arose in cricetid rodents living in semidesert and desert environments. He based these conclusions on an analysis that indicated that the predominant plague hosts in Asia, Africa, and the Americas belonged to the Cricetidae, a family that at the time of Kucheruk's publication included gerbillines, cricetines, arvicolines, and sigmodontines. While this suggestion is still generally accepted, other former Soviet researchers have recently proposed that *Y. pestis* first evolved in marmots (*Marmota* spp.) and their fleas (Suntsov and Suntsova, 2000).

Recent molecular studies clearly indicate that *Y. pestis* is very closely related to the gut microbe *Y. pseudotuberculosis* (Bercovier and others, 1980; Trebesius and others, 1998). The high degree of relatedness between these two bacteria strongly suggests that they have diverged only recently, as suggested by Achtman and others (1999), who proposed that *Y. pestis* might have arisen as a clone of *Y. pseudotuberculosis* only 1,500–20,000 years ago (Achtman and others, 1999; Wren, 2003). This last finding is particularly interesting because of its implications for the degree of coadaptation or coevolution that might have occurred between *Y. pestis* and its hosts and vectors. The recently reported genomic sequences of three *Y. pestis* strains also reveal many interesting features of this bacterium and support the contention that the *Y. pestis* genome is still in a state of rapid flux and might be undergoing reductive evolution as it loses the ability to express certain genes that remain active in *Y. pseudotuberculosis* but are not required for *Y. pestis* to be maintained in a vector-borne transmission cycle (Wren, 2003). Indeed, it has been suggested that the disruption of genes still expressed in *Y. pseudotuberculosis* might be essential for *Y. pestis* to survive in a vector-vertebrate host life cycle (Wren, 2003).

The actual geographic origin of the plague bacterium was a subject of considerable speculation during much of the 20th century. Based on the analysis of plague hosts cited in the previous paragraph, Kucheruk (1965) felt that *Y. pestis* probably appeared in either North American or Asian cricetids. More recent lines of reasoning, however, suggest that a North American origin is highly unlikely. First, epidemiologic evidence strongly indicates that plague did not exist in the United States prior to the last pandemic when rat-infested ships introduced *Y. pestis* to the San Francisco area around 1900 (Link, 1955; Barnes, 1982). Second, microbiological evidence indicates that North American isolates almost invariably reduce nitrates to nitrites but fail to acidify glycerol, which identifies them as belonging to the orientalis biovar that was involved in the late 19th and early 20th century pandemic mentioned above (Devignat, 1951; Guiyoule and others, 1994). Even more convincing results have been provided by recent molecular investigations, including ribotyping and single nucleotide polymorphism analyses, which indicate that United States strains are genetically similar to other orientalis biovar strains collected from areas in other continents that also experienced rat-associated outbreaks during the last pandemic (Guiyoule and others, 1994; Achtman and others, 2004). In general, most lines of evidence, including levels of strain diversity within particular geographic regions, suggest an Asian origin for *Y. pestis*, although the plague bacterium clearly has existed in Africa for more than a millennium and probably considerably longer.

The availability of appropriate methodologies for detecting and analyzing variations among plague strains will have a significant impact on our ability to understand the evolution of plague and how strain differences influence various aspects of *Y. pestis* biology, including its ecology, virulence, and modes of transmission. Early attempts to analyze variation among plague strains relied primarily on phenotypic characteristics, such as reactivities in various biochemical tests, virulence for different types of laboratory animals, production of selected virulence factors, or apparent host associations (Devignat, 1951; Tumanskii, 1957, 1958; Levi, 1962; Stepanov, 1975; Kozlov, 1979). More recently, investigators have analyzed variation among *Y. pestis* strains by using DNA probes, ribotyping, multiple locus variable number tandem repeat assays (MLVA), and analyses of IS100 elements and single nucleotide polymorphisms (Guiyoule and others, 1994; Gorshkov and others, 2000; Klevytska and others, 2001; Motin and others, 2002; Achtman and others, 2004; Girard and others, 2004). Many of the above studies were intended primarily to demonstrate the feasibility of using a particular system for analyzing variation and, thus, examined mostly strains from established reference collections. By contrast, Girard and others (2004) used MLVA to track the spread of plague during an actual epizootic in prairie dogs in northern Arizona. These authors also used their MLVA results, in conjunction with other field and laboratory data, to construct a mutation-rate model that suggested that plague dynamics in their systems consisted of a rapid expansion phase, which was associated with population growth and dispersal, followed by a persistent phase characterized by lower reproduction and dispersal rates. The identification of additional markers should be favored by the recent publication of the complete genomic sequences of three *Y. pestis* strains (Parkhill and others, 2001; Deng and others, 2002; Song and others, 2004).

The phenotypic and genetic studies cited in the previous paragraph identified differences among strains from different foci and host sources, but they fail to answer the question of whether the observed differences among *Y. pestis* strains simply reflect geographic variation or actually provide evidence that regional variants of *Y. pestis* are indeed adapted to a particular host species. Fortunately, the new molecular methodologies described earlier should provide researchers with valuable tools for answering this question as well as other important ecological and evolutionary questions. Analyses of North American strains should be particularly interesting because, as indicated previously, *Y. pestis* apparently has existed in this continent for only a little over 100 years, and the few orientalis biovar strains that were introduced at that time probably were highly similar, having originated in the same region of southwest China. Because the diversity among these invading strains of *Y. pestis* was very low, researchers have an interesting opportunity to examine how *Y. pestis* changes over time and whether this bacterium is likely to exhibit different characteristics, such as increased or decreased virulence, when it is associated with a particular host or vector species.

Plague Transmission Cycles and Maintenance of Plague in Natural Foci

Figure 2 presents a generalized illustration of the natural transmission cycle of plague. In order for flea-borne transmission of plague bacteria to occur, a flea must take a blood meal from a rodent with a heavy *Y. pestis* bacteremia, become infected with plague bacteria, and later transmit this bacterial infection to another susceptible rodent host. Some researchers assume that rodent-to-flea-to-rodent transmission can occur indefinitely in so-called enzootic cycles that cause few apparent deaths among the purportedly resistant rodent hosts (enzootic or maintenance hosts) of these cycles. According to this same concept of plague maintenance and transmission, the disease occasionally spills over to other much more susceptible hosts (epizootic or amplification hosts) that often die in rapidly spreading epizootics, thereby posing increased plague risks for other mammals, including humans (Poland and Barnes, 1979; Poland and others, 1994). As indicated in fig. 2, *Y. pestis* occasionally is transmitted through consumption of infected prey or, perhaps, as a result of inhaling infectious respiratory droplets from animals with pneumonic plague and cough. The roles of these last two modes of transmission in maintaining natural foci have not been determined but are generally assumed to be less important than flea-borne transmission. A few researchers also have proposed that hosts can acquire plague as a result of digging in soil containing viable *Y. pestis* (Mollaret, 1963). Such infections presumably would be acquired through breaks in the skin or inhalation of *Y. pestis* stirred up by an animal's digging activities.

Rodent Hosts of Plague

Based on early observations in India and elsewhere (Pollitzer, 1954), plague initially was believed to exist in

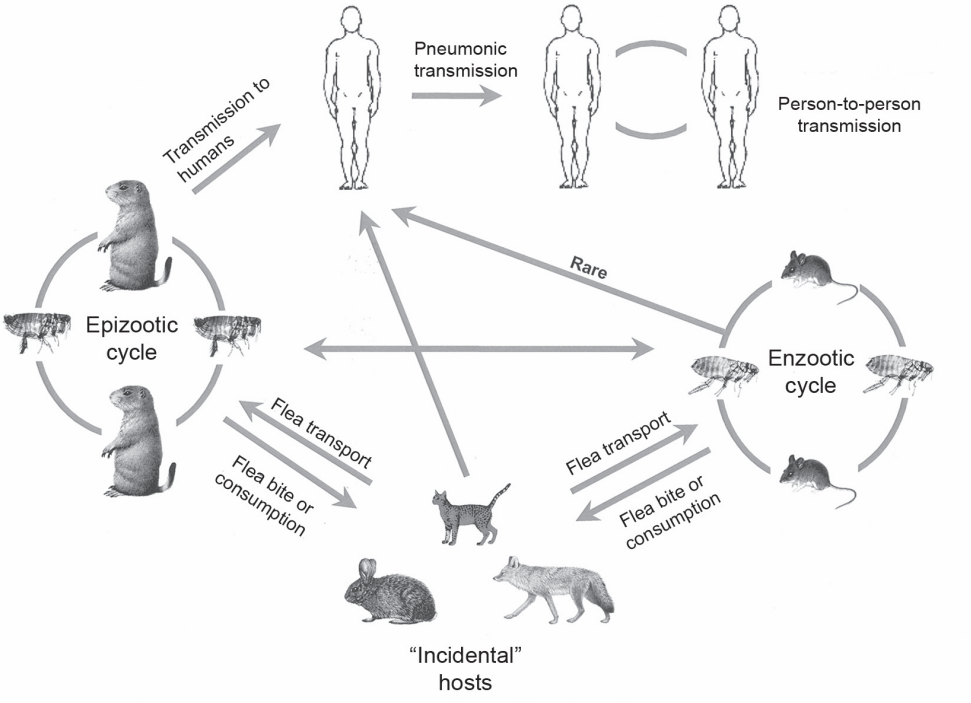

Figure 2. Generalized plague transmission cycle for the United States. Figure courtesy of Centers for Disease Control and Prevention.

nature almost exclusively in commensal rats (primarily *Rattus rattus* and *R. norvegicus*) and rat fleas (primarily *Xenopsylla cheopis*), but it soon became clear that *Y. pestis* also could be found in a variety of wild (noncommensal) rodents and their fleas. In the first decade of the 20th century, McCoy (1908) reported plague among California ground squirrels (*Spermophilus beecheyi*), and others noted soon thereafter that although epizootic activity among rats had largely disappeared, the disease continued to persist in other small mammals around the San Francisco Bay area (Link, 1955). In Asia, Zabolotny (1915) suggested the possibility of wild rodent foci, noting that pneumonic plague outbreaks in Manchuria probably originated from hunters handling tarbagans (*Marmota sibirica*) rather than as a result of human exposure to infectious rat fleas. Later studies confirmed that *Y. pestis* could persist among a variety of rodent species and their fleas without the involvement of commensal rats (Pollitzer, 1954; Pollitzer and Meyer, 1961).

Following the recognition that certain wild rodents are the major hosts of plague, researchers began to ask what characteristics allow particular rodent species to play important roles in the ecology of plague while others play little or no role. At first glance the number of potential rodent hosts is surprisingly high. Pollitzer (1960) identified 203 rodent species or subspecies reported to be naturally infected with *Y. pestis*, a list that could now be slightly extended. However, only a few of these species can be considered truly important hosts of plague, primarily those belonging to the families Sciuridae and Muridae. Among the sciurids, the predominant plague hosts include members of certain genera of burrow-dwelling squirrels (*Spermophilus* [formerly *Citellus*], *Cynomys*, *Ammospermophilus*) and chipmunks (*Tamias*, including *Eutamias* and *Neotamias*). Within the Muridae a number of species in the subfamilies Murinae, Gerbillinae, Arvicolinae, and Sigmodontinae are considered to be important hosts in various regions (Kucheruk, 1965; Gage, 1998; Gratz, 1999).

Among the topics discussed in this paper, probably the most neglected by recent researchers has been the response of native rodent species to *Y. pestis* infection and the roles these animals play in the long-term maintenance of plague foci in different regions. Although many rodents are mentioned in the literature as major plague hosts, the actual evidence to support these claims is often weak, particularly for those putative host species found in certain regions where relatively little research has been done (Gage, 1998; Gratz, 1999). Factors believed to influence the suitability of a particular rodent host for plague include the degree of its population-level resistance to *Y. pestis*-related mortality, its ability to serve as a source of infection for suitable flea vectors, the presence of large numbers of fleas on many members of the host population throughout much of the year, and occupation of burrows or nests that support development and maintenance of high flea populations (Pollitzer and Meyer, 1961; Gage and Kosoy, 2005).

Among these factors, one of the most contentious has been the degree to which population-level resistance to *Y. pestis*-related mortality is essential for the maintenance of plague by one or more rodent species in a particular focus (Pollitzer and Meyer, 1961; Gage and Kosoy, 2005). Host resistance to plague is undoubtedly influenced by many factors, including species, genetic factors within and among populations of a particular species, age, breeding status, prior immunity, physiologic condition, and probably other considerations. When assessing the importance of resistance, it is clear that its presence could favor the survival of host populations in plague-affected areas, although other factors also could operate to reduce mortality and prevent total die-offs among these animals, including the presence of patchy environments that could provide refuges for subpopulations within a larger metapopulation. Seasonal changes in the activities of susceptible hosts or competent flea vectors also might temporarily interrupt or slow down transmission to the point where host populations could be sustained from year to year by recruitment of new individuals (Pollitzer and Meyer, 1961).

One problem encountered in discussing resistance among plague hosts is the somewhat confusing use of the term itself. Host populations that are considered resistant rarely, if ever, are uniformly resistant to *Y. pestis*-related mortality but typically consist of a mixture of somewhat resistant individuals that become infected but recover and other animals that are more susceptible and succumb to plague. For example, mortality rates among great gerbils (*Rhombomys opimus*), which are considered resistant hosts, typically are 40–60 percent (Petrunina, 1951; Rivkus and others, 1973). Although this figure appears high, it is significantly lower than the mortality rates experienced by many other rodents, including other sympatric species of gerbils in the genus *Meriones*. Others have demonstrated that resistance can be associated with past exposure to plague (Birukova, 1960; Thomas and others, 1988; Levi, 1994). Several experiments demonstrated differences in plague resistance between populations of midday gerbils (*Meriones meridianus*) from different sides of the Volga River (Birukova, 1960; Levi, 1994). Levi (1994) compared median lethal doses (LD50) of *Y. pestis* for live-caught gerbils from a population on the west side of the river and another from the east side and found that in three trials, the LD50 values for populations on the west side were 2, 4, and 216 colony forming units (CFUs) while those on the east side of the Volga exhibited LD50 values of 3.397×10^6, 1.000×10^6, and 39.400×10^6 CFUs. Captive-born hybrids of representative individuals (F1 generation) from both populations exhibited intermediate levels of resistance, as did the offspring of these individuals (F2 generation), suggesting that the observed resistance had a genetic basis. According to Levi (1994), these experiments helped explain how midday gerbils are able to serve as primary hosts for plague on the east side of the Volga but have a lesser role west of the river. Interestingly, these same populations of midday gerbils did not differ in their sensitivities to infection with the agents of tularemia and brucellosis. These authors also noted that two populations of another gerbil species, the tamarisk gerbil (*Meriones tamariscinus)*, from the western and

eastern sides of the Volga were found to be highly sensitive to plague infection (LD50 values of 6.800×10^2 and 5.000×10^2 CFUs, respectively).

In North America, Thomas and others (1988) demonstrated that captive-born northern grasshopper mice (*Onychomys leucogaster*) from a plague-free region of Oklahoma were much less resistant to plague than were mice of the same species from a north-central Colorado population that had been exposed to plague. In another North American paper, Quan and Kartman (1956) demonstrated that different populations of deer mice (*Peromyscus maniculatus*) and California voles (*Microtus californicus*) varied in their susceptibility to *Y. pestis*. Differences in susceptibility have been demonstrated to have a genetic basis in California voles (Hubbert and Goldenberg, 1970). Although the above data indicate that populations of some rodent species are highly resistant to *Y. pestis*, others, such as those of the black-tailed prairie dog, nearly always succumb to infection whenever they are struck by plague epizootics (Poland and Barnes, 1979; Biggins and Kosoy, 2001a,b).

Regardless of whether resistant hosts must be present in order for plague foci to persist, flea-borne transmission of plague bacteria among rodents depends on the presence of animals that are capable of serving as sources of infection for feeding fleas. Experimental results indicate that fleas are likely to become infected with *Y. pestis* only after feeding on animals that have very high bacteremias ($>10^6$ *Y. pestis*/mL blood) (Burroughs, 1947; Engelthaler and others, 2000). In general, animals that have such high bacteremias often appear moribund, and few, if any, survive their infections. Thus, resistant animals that develop little or no bacteremia following infection probably are unlikely to serve as significant hosts for infecting fleas. Resistant individuals that survive infection could, however, still play important ecological roles by serving as hosts for maintaining flea populations and contributing offspring to the next generation of hosts. While many of the offspring of these animals also might be resistant, it is possible that at least some of their littermates will be susceptible.

Some animals might not be completely resistant, at least in the sense of being able to rapidly clear themselves of infection, but rather survive their initial bout of illness and go on to develop a chronic infection with *Y. pestis*. While evidence for chronic infections among North American species is almost nonexistent, the phenomenon has been observed in laboratory rats infected with nonencapsulated plague (F1-minus) strains (Williams and others, 1975; Williams and Cavanaugh, 1983). If wild rodents were chronically infected with fully virulent *Y. pestis* and later experienced a recrudescence of infection, perhaps as a result of breeding stress or decreased immune function in older individuals, they could develop a fatal bacteremia of sufficient magnitude to infect feeding fleas.

As noted above, differences of opinion exist about the importance of resistance among host populations. Some researchers have suggested that the role a particular host population plays in plague maintenance can be inferred largely from its level of resistance (Pollitzer, 1954; Pollitzer and Meyer, 1961; Rall, 1965). For example, great gerbils are believed to be the major hosts of plague in certain central Asian desert foci. The percentage of resistant animals among great gerbil populations in these foci has been reported to be 40–60 percent, a level that is higher than that found in gerbils of the genus *Meriones*, which occur in the same foci (Rivkus and others, 1973). In other situations resistance does not appear to differ greatly among various potential host species, making it difficult to assert that one host is more important than another based strictly on the observed levels of host resistance. For example, resistance was similar among great gerbils (50–80 percent), little susliks (*Spermophilus pygmaeus*) (50–70 percent), and midday gerbils (44–60 percent) in a Kazakh steppe focus (Atshabar, 1999).

Others have argued that the importance of resistance can be overemphasized and that other mechanisms can lead to the persistence of plague among highly susceptible host species (Pollitzer, 1954). While plague might kill most animals in a highly susceptible population, survival can be influenced by age, season, or physical condition, thus allowing some hosts to survive and reproduce. Rodents also might be able to become infected shortly before entering hibernation, develop a latent infection as their body temperatures drop, and then not experience severe illness or die of plague until they reawaken in the spring (Gayskii, 1944; Pollitzer, 1954; Pollitzer and Meyer, 1961). Maevskii and others (1999) also reported that *Y. pestis* could be isolated from the "mummified" carcasses of long-tailed susliks (*S. undulatus*) for 7.5 months after these animals first entered hibernation. Spatial isolation among colonies or subpopulations of highly susceptible hosts also could allow plague to be maintained in metapopulations of these animals (Pollitzer and Meyer, 1961; Gage and Kosoy, 2005). In those foci where highly susceptible hosts live in a mosaic of distinct habitat patches, plague is unlikely to kill all of the susceptible animals in each patch or go from patch to patch without at least some delay, thereby allowing the disease to persist by spreading from patch to patch at a rate that is low enough to allow host populations in previously affected patches to recover before once again being exposed to *Y. pestis* infection.

Types of Plague Hosts

Another unresolved question about the role of different rodent hosts in the natural cycle of plague is whether a single host or multiple hosts are required for long-term maintenance of natural transmission cycles. Fenyuk (1940, 1948) believed that certain rodent species and their fleas could maintain plague in the absence of other rodent species and referred to such animals as primary hosts. Secondary hosts were those species that routinely become infected but are incapable of supporting long-term maintenance of *Y. pestis* in a particular focus. Although secondary hosts are by definition incapable of maintaining plague foci in the absence of primary hosts, some proponents of this concept believe they are important in spreading the disease during epizootics.

Expanding on the primary host hypothesis, Rall (1965) proposed the concept of monohostality wherein maintenance of plague in a particular focus depends on the presence of a single rodent species and its fleas. Probably the most commonly cited examples of monohostal foci are those involving great gerbils in central Asia (Petrov, 1959). Acceptance of this proposal has not been universal, and maintenance of plague in other Asian foci has been suggested by other investigators to involve multiple host species (polyhostal foci) (Kalabukhov, 1965). The "Daurian enzootic area" of central Asia represents a proposed polyhostal focus, with Siberian marmots (*M. sibirica*), Daurian susliks (*S. dauricus*), pikas (*Ochotona* spp.), and voles (*Microtus* spp.) all presumedly playing important roles in maintaining this plague focus (Kalabukhov, 1965). The question of whether various United States foci are monohostal or polyhostal has received little attention. Although existing evidence does not allow firm conclusions to be made, *Y. pestis* infections are frequently identified in multiple rodent species in the western United States, particularly in certain southwestern States (New Mexico, Colorado, Arizona) and some mountainous regions of California and nearby areas (Barnes, 1982; Gage and others, 1995), suggesting that at least some of these foci are polyhostal.

American workers have rarely used the terms primary and secondary hosts or monohostality and polyhostality. Instead, the most commonly cited concept categorizes rodent hosts as either enzootic or epizootic (Poland and Barnes, 1979; Poland and others, 1994). Supporters of this concept suggest that enzootic hosts and their fleas maintain plague during interepizootic periods and share certain features, including heterogeneous population responses to *Y. pestis* infection, low mortality following infection, long multiestrous breeding seasons with high reproductive potential, short life expectancies, flea infestations during all seasons, and a relatively high likelihood that antibody will be detected within the population. The most commonly proposed enzootic hosts are various species of *Peromyscus* and *Microtus*. By contrast, epizootic hosts are considered to have low to moderate resistance to *Y. pestis* infection, often experience high morbidity and mortality when infected, exhibit relatively little population-level heterogeneity to infection, and often experience heavy infestations with one or more species of vector flea that are likely to peak in abundance during the warmer months of the year, which is the time when transmission rates also appear to be highest. Proposed epizootic hosts include various species of *Cynomys*, *Spermophilus*, *Ammospermophilus*, *Tamias*, and *Neotoma* (Barnes, 1982; Gage and others, 1995).

In reality, evidence to support the enzootic-epizootic host concept is often lacking or questionable. Obviously, epizootics with dramatic die-offs do occur among proposed epizootic hosts, but corresponding data to indicate that supposed enzootic hosts, such as deer mice or voles, are essential for the maintenance of plague during interepizootic periods is largely lacking. Another plausible alternative is that plague does not rely on any one host for its maintenance in a particular focus during the intervals between epizootics, but rather circulates at much reduced rates among most, if not all, of the same hosts that commonly become infected during epizootics. Under such circumstances, a fair amount of mortality could occur among these hosts during interepizootic periods but go virtually undetected because of the lack of routine rodent surveillance in most plague-enzootic areas.

The Role of Fleas in Transmitting *Yersinia pestis*

Because of its obvious role in rat-associated bubonic plague outbreaks during the last pandemic, many early studies concentrated on the role of the Oriental rat flea (*Xenopsylla cheopis*) as a vector of plague. Within two decades after Yersin's 1894 discovery of the plague bacterium, Bacot and Martin (1914) demonstrated that *Y. pestis* proliferates in the midgut and proventriculus of an infected flea, forming recognizable colonies within a few days after the fleas ingest an infectious blood meal. They also showed that *Y. pestis* colonies can proliferate in an infected flea to such an extent that its proventriculus, a globular spine-filled structure at the end of the foregut, becomes blocked by a mass of bacteria and blood cell remnants. Once blockage of the proventriculus occurs, blood is no longer able to pass through the foregut to the midgut or "stomach" of the flea, resulting in its eventual starvation. Because the blocked rat flea is starving, it will repeatedly attempt to feed on almost any available mammalian host, including humans. As the flea repeatedly fails in its efforts to ingest blood, it attempts to clear the proventricular blockage by regurgitating, a process that does not clear the block but can dislodge plague bacteria from it. These dislodged bacteria and a small amount of ingested blood are then flushed back into the bite wound, resulting in infection of the host. Fleas that fail to become blocked were found to transmit at much lower rates or not at all, which led to the currently accepted dogma that the only efficient plague vectors are those that become blocked.

Within the past decade the molecular basis by which *Y. pestis* promotes blocking in infected *X. cheopis* has become clear (Hinnebusch, 1997, 2005). Hinnebusch and others (1996) demonstrated that *Y. pestis* strains containing mutations in certain genes (hmsR and hmsH) found in the hemin storage (hms) locus were incapable of forming blockages in infected *X. cheopis* fleas. The hemin storage locus derives its name from the ability of strains that possess a functional hms locus to bind hemin to their surfaces. In general, hemin-binding strains appear to be more "sticky" than strains that cannot bind hemin and are, thus, more likely to form clumps of *Y. pestis* in the flea's gut or adhere to the cuticular spines in its proventriculus (Bibikova, 1977; Hinnebusch and others, 1996). Other investigators have demonstrated that blocking depends on temperature, with fleas rarely becoming blocked,

or actually clearing themselves of blockages, when maintained at temperatures above 27.5°C (Cavanaugh, 1971; Hinnebusch and others, 1998).

Additional studies have demonstrated that survival of plague bacteria in flea midguts depends on the expression of a gene (ymt) found on the largest of the three *Y. pestis* plasmids (approximately 110 kb) (Hinnebusch and others, 2002). The product of this gene (Ymt), which is a phospholipase D, has been referred to as murine toxin because of its high toxicity for murines (rats and mice) but not other types of rodents or mammals belonging to other orders. The study by Hinnebusch and others (2002), however, suggests that the true function of Ymt is to promote the survival of *Y. pestis* in the flea vector and that its toxicity for murines is merely coincidental. Even more recent studies have suggested that colonization of flea guts by *Y. pestis* might depend on biofilm formation by the plague bacterium (Darby and others, 2002; Jarrett and others, 2004).

This research has greatly improved our understanding of how *Y. pestis* promotes its transmission by flea vectors, but we still have little knowledge of why some flea species, including those found on wild rodents and presumed to be important vectors, vary so greatly in their ability to transmit plague (Gage and Kosoy, 2005). Taxonomic affinities appear to provide little guidance, as demonstrated by the pulicid fleas of the genus *Xenopsylla*. The Oriental rat flea (*X. cheopis*) and a less widely distributed African rat flea (*X. brasiliensis*) are both highly efficient vectors, but their congener *X. astia*, which is common on rats in the Indian subcontinent and southeastern Asia, is a very poor vector (Pollitzer, 1954). Many decades ago, it was hypothesized that the structure or arrangement of the proventricular spines might be important determinants of a flea's ability to transmit *Y. pestis* (Eskey and Haas, 1940). In support of this contention, Korzun and Nikitin (1997) reported that blocking in a ground squirrel flea, *Citellophilus tesquorum*, was positively associated with high levels of fluctuating asymmetry among the proventricular spines of these fleas.

Although the structure of the proventricular spines might very well influence the blocking process, it does not explain why *Y. pestis* appears to be unable to survive and develop in the guts of certain fleas. Among the poorest plague vectors are a number of flea species commonly associated with man and his domestic animals, including the so-called human flea (*Pulex irritans*), the cat flea (*Ctenocephalides felis*), the dog flea (*C. canis*), and sticktight fleas (*Echidnophaga gallinacea*) (Pollitzer, 1954). For example, *P. irritans* often clear themselves of infection within days after ingesting an infectious blood meal and rarely become blocked. Although these insects can transmit plague, they appear to do so only when large numbers of fleas are placed on susceptible hosts within a few hours after being allowed to feed on a *Y. pestis*-infected animal, suggesting that hosts are infected through the introduction of plague bacteria on contaminated flea mouthparts (mechanical transmission) rather than by the feeding of blocked fleas (Pollitzer, 1954; Blanc, 1956). It should be noted that despite its poor vector competency, some authorities believe that *P. irritans* is a significant vector of plague to humans in those situations where people live in unsanitary, heavily flea-infested homes that are often shared with domestic animals (Pollitzer, 1954; Blanc, 1956). These findings raise the possibility that infected but unblocked fleas on wild rodents also might transmit plague bacteria under certain circumstances.

Fleas found on wild rodent hosts also vary considerably in their ability to support *Y. pestis* infections and transmit plague bacteria (Eskey and Haas, 1940; Douglas and Wheeler, 1943; Burroughs, 1944, 1947; Holdenried, 1952; Pollitzer, 1954; Kartman and Prince, 1956; Kartman, 1957; Kartman and others, 1958a,b; Pollitzer, 1960; Pollitzer and Meyer, 1961; Engelthaler and others, 2000). While some wild rodent fleas appear to block at high rates and become infectious soon after ingesting a *Y. pestis*-containing blood meal, other species require considerably longer periods of time to become blocked. The time required for blocking to occur in some species is sufficiently long that most of the infected fleas are likely to die before block formation actually occurs. A recent comparison of the development of *Y. pestis* infections in *X. cheopis* and *Oropsylla montana*, a ground squirrel flea, demonstrated that *Y. pestis* colonies became established very early in the course of infection in both the proventriculus and the midgut of infected *X. cheopis* (Engelthaler and others, 2000). In *O. montana*, however, *Y. pestis* colonies initially appeared only in the midguts of infected fleas, which meant that the midgut infection had to proliferate and spread considerably before colonization of the proventriculus could occur. Because colonization of the proventriculus is delayed, the average time required for blocking to occur in *O. montana* is much longer than in *X. cheopis*. The failure of many *O. montana* to become blocked and the fact that these fleas transmit at much lower rates than *X. cheopis* are particularly interesting because *O. montana* is considered to be the primary vector of plague to humans in the United States. Published results of experimental infection and transmission studies (see citations at the beginning of this paragraph) done with other species of wild rodent fleas suggest that the situation observed for *O. montana* is more typical than that seen with *X. cheopis*. Of particular interest are the limited studies done with ground squirrel, prairie dog, and woodrat (*Neotoma* spp.) fleas, which typically indicate that most of these fleas are relatively poor plague vectors compared to *X. cheopis* (see earlier citations in this paragraph). While many wild rodent fleas reportedly block and transmit at low rates, a few, such as *Hystrichopsylla dippei*, appear to be quite efficient vectors (Kartman and others, 1958b). Although the studies cited earlier in this paragraph make it obvious that *X. cheopis* is an exceptional plague vector, this does not mean that *Y. pestis* is always successful in its attempts to colonize and establish a stable infection in this flea. Engelthaler and others (2000) found that by 6 weeks after ingesting a *Y. pestis*-infected blood meal, 60 percent of all *X. cheopis* had cleared themselves of infection. Despite this fact, however, the infection rates observed in *X. cheopis*

6 weeks after taking an infectious blood meal were still much higher than those observed in *O. montana* (60 percent versus 15 percent, respectively).

Many of these studies raise questions about whether transmission by blocked fleas is actually essential for the rapid spread of *Y. pestis* during epizootics or for the interepizootic maintenance of plague. One possibility is that in some situations partially blocked fleas could transmit at sufficiently high rates to be important vectors. Burroughs (1947) and Engelthaler and others (2000) demonstrated that *O. montana* fleas were capable of transmitting within 4 days after feeding on an infectious host, a much shorter time than that required for blockage in these species, but perhaps too long for strictly mechanical transmission of viable *Y. pestis* on contaminated mouthparts to occur. Burroughs (1947) and others (Voronova, 1989; Degtyareva and others, 1990; Gan and others, 1990; Bazanova and others, 1991) list additional examples of the transmission of *Y. pestis* by partially blocked or apparently block-free fleas.

The role that mechanical transmission might play in natural foci also should be reexamined. As noted previously, early studies of potential plague vectors indicated that some fleas, such as the human flea, rarely became blocked but occasionally transmitted plague when fleas that had fed on an infected host were quickly transferred in large numbers to susceptible hosts, a finding that is typically interpreted as evidence for mechanical transmission (Pollitzer, 1954; Blanc, 1956). Later studies, particularly those of Burroughs (1944, 1947) and Kartman and others (1958a,b) also provided evidence that common North American rodent fleas are capable of transmitting *Y. pestis* by mechanical means. Quan and others (1953) provided interesting evidence that even *X. cheopis* is capable of mechanically transmitting plague bacteria. Based on the results of the studies noted earlier and others, Burroughs (1947) and Kartman and others (1958a,b) suggested that mechanical transmission might be important, particularly during epizootics when host densities are high and the likelihood that fleas will rapidly transfer from dead hosts to susceptible ones is also high. Kartman and others (1958a,b) further suggested that the bulk of transmission during epizootics occurs through mechanical means while transmission of plague during interepizootic periods is accomplished by those rodent fleas that are capable of becoming blocked and transmitting at high efficiencies. In particular, he cited *Malaraeus telchinum*, a flea that is extremely abundant on mice and voles in some regions of the West, as a likely mechanical vector during epizootics and *Hystrichopsylla dippei*, a far less abundant but much more efficient plague vector, as an important vector during interepizootic periods. Unfortunately, others have not pursued this hypothesis, and it would be very interesting to know whether other "pairs" or groups of fleas play similarly complimentary roles during epizootic and interepizootic periods. It also would be worthwhile to determine whether the rapid rates of transmission observed during plague epizootics in prairie dogs or other highly susceptible hosts are due to mechanical transmission or transmission by blocked fleas. The former can take place virtually immediately after a flea has fed on a heavily bacteremic host, but the latter typically requires an extrinsic incubation period of 2 or more weeks before fleas can become blocked and, therefore, capable of efficiently transmitting. Alternatively, hosts might become infected by consuming other animals that have died of plague or through respiratory contact with hosts that have pneumonic plague.

Although laboratory experiments can help determine whether a particular flea species is likely to be an important vector, other factors also need to be considered in determining the actual role a potential vector will play in nature (Gage, 1998; Gage and Kosoy, 2005). Obviously, fleas that feed on hosts that are seldom infected with plague, or live only in plague-free areas, are unlikely to be important. Fleas that are highly host-specific might be very important for transmitting plague among members of a particular host species but would rarely spread the disease to other hosts. The seasonality and abundance of the flea's hematophagous adult stage also are likely to be important. Many important vectors occur most abundantly on their hosts during those warm months when plague transmission also peaks. Another potentially important factor is the ability of fleas to survive in off-host environments while waiting for an alternative host to appear.

Maintenance of Plague Between Transmission Seasons and Between Epizootics

Figure 2 provides a basic overview of the plague transmission cycle but unfortunately conveys almost no information on the relative roles different components play in maintaining plague between transmission seasons or during interepizootic periods when little or no *Y. pestis*-related illness is apparent among the normal hosts of the disease. At least four different hypotheses can be advanced for long-term maintenance of plague (Gage and Kosoy, 2005): continuous enzootic transmission among rodent hosts and their fleas at more or less steady rates except during irregularly occurring epizootics; chronic infection of rodents with eventual relapses of the disease in these animals and subsequent infection of vector fleas following these relapses; prolonged survival of infected fleas in host nests or burrows; and indefinite survival of *Y. pestis* in soil, soil protozoa, or perhaps even plant tissues. The following sections discuss the above hypotheses of plague maintenance.

Are Rodents Merely Amplifying Hosts or True Reservoirs of Infection?

In order for plague to be maintained through continuous enzootic transmission, the rodent hosts and flea vectors must

both be present and active throughout the year. In temperate regions some plague hosts enter hibernation or become much less active during winter months, which could interrupt the *Y. pestis* transmission cycle. For example, marmots (*M. sibirica* and certain other *Marmota* spp.), which are thought to be critically important plague hosts in some Asian foci, hibernate for many months and, thus, are unlikely to become infected after entering hibernation or support ongoing transmission during this period. If their fleas also become inactive during winter months or lack the opportunity to acquire new infections from hibernating hosts, transmission could be interrupted. One possible solution to this dilemma could be the survival of *Y. pestis* in hibernating animals (Gayskii, 1944; Pollitzer, 1954). According to this hypothesis, a *Y. pestis*-infected animal might enter hibernation prior to becoming ill, thus slowing or temporarily halting the progression of *Y. pestis* infection as a result of the effects of low host body temperature on the growth of the pathogen or its virulence. Upon reawakening in the spring, the infection could reactivate, causing the animal to become ill and develop a *Y. pestis* bacteremia of sufficient magnitude to infect feeding fleas, thereby continuing the cycle of rodent-to-flea-to-rodent transmission for another year. While this explanation seems plausible and does have some experimental support, little is known about its importance in natural foci. Also, such an explanation is unlikely to be important in tropical or subtropical foci. If hibernating animals die of plague before reawakening in the spring, it is also possible that plague bacteria could survive in their dried tissues for many months after the animals have died (Maevskii and others, 1999).

According to some investigators, rodents that do not hibernate might develop chronic infections and act as reservoirs for maintaining plague from one transmission season to the next (Pollitzer, 1954; Pollitzer and Meyer, 1961). Experimental evidence suggests that individual great gerbils in central Asia survive infection and then develop granuloma-like lesions in their livers and perhaps other tissues that contain viable *Y. pestis* (Suleimenov, 2004). These plague bacteria-containing lesions can reportedly persist for many months, thereby allowing latent infections to become reactivated during the spring as adult hosts experience increased stress due to breeding or decreased immune system function due to old age. Great gerbils that experience reactivation of their infections are believed to circulate sufficient *Y. pestis* in their bloodstream to infect feeding fleas. One of the practical problems encountered in evaluating the importance of presumed chronic infections in rodents under field conditions is whether lesions observed in the tissues of suspected carrier hosts are really indicative of chronic infection or simply a sign of resolving infections.

Some researchers have argued that plague could be maintained through the winter months by continuous transmission between certain hosts and their fleas. Such a pattern of transmission has been proposed for deer mice (*P. maniculatus*) and their allies (other *Peromyscus* spp.) or various species of voles (*Microtus* spp. and others) (Poland and Barnes, 1979; Poland and others, 1994). Deer mice and other mice of the genus *Peromyscus* remain active in all seasons, are often infested with fleas during the winter months, and reproduce throughout much of the year, which results in the ongoing introduction of susceptible animals into local mouse populations. Whether populations of *Peromyscus* or voles can indeed maintain plague through continuous rodent to flea to rodent transmission is at present uncertain. In a 13-month study (March 1954–April 1955) of 1,458 *Microtus californicus* found dead in a San Mateo County plague focus, *Y. pestis* was identified in the tissues of these animals during 10 of the 13 months. The only months when positive animals were not identified were December 1954 (n = 52), March 1955 (n = 33) and April 1955 (n = 27) (Kartman and others, 1962). Considering the relatively low number of dead animals examined during those 3 months, *Y. pestis* might have indeed been present all year in at least some voles within this focus.

Fleas as Reservoirs of Plague

While no one disputes that fleas are the only significant vectors of plague, they also could act as long-term reservoirs by maintaining *Y. pestis* in off-host environments during the intervals between transmission seasons or during periods of host hibernation (Gage and Kosoy, 2005). Many studies indicate that infected but unblocked, and even blocked, fleas can survive for many months in off-host environments. In one study, infected *Ctenopthalmus breviatus* survived for up to 396 days when held on wet sand at temperatures of 0–15°C (Golov and Ioff, 1926, 1928). Other studies indicated that *Oropsylla silantiewei* could survive for as long as 558 days without feeding while *Citellophilus tesquorum* and *Neopsylla setosa* did so for 275 and 180 days, respectively (cited by Kozlov, 1979). Sharets and others (1958) reported that *Rhadinopsylla ventricosa* fleas remained infected with *Y. pestis* for at least 420 days. Bazanova and Maevskii (1996) succeeded in maintaining more than half of all *C. tesquorum altaicus* fed on infected susliks (*Spermophilus undulatus*) over a period from mid-September to mid-June, which provided sufficient time for these fleas to survive through the hibernation period of their hosts. One female in their experiments survived through two winters, living for a total of 411 days after being fed on an infected suslik. Even more importantly, when infected *C. tesquorum altaicus* that had been starved through the hibernation period of their hosts were later allowed to feed, they succeeded in transferring plague to these animals, thus demonstrating that these fleas could act as both vectors and reservoirs of infection. In North America, Kartman and others (1962) reported the recovery of infected *Oropsylla labis* (syn. *Opisocrostis labis*) and *O. tuberculata cynomuris* (syn. *Opisocrostis tuberculatus cynomuris*) from abandoned prairie dog burrows for more than a year after their hosts had died of plague.

Survival of Plague in Soils, Soil Protozoa, Plant Tissues, or Other Unusual Sites

Some investigators have proposed that plague might survive during interepizootic periods in the soil of burrows (Mollaret, 1963). In one experiment, four species of gerbils (*Meriones libycus*, *M. persicus, M. tristrami*, and *M. vinogradovi*) developed plague after being allowed to dig burrows in laboratory enclosures containing soils contaminated on the previous day with *Y. pestis* in a broth culture (Mollaret, 1963). In other experiments, it was claimed that plague survived many months in both sterilized and nonsterilized soils (Mollaret, 1963; Baltazard, 1964). According to supporters of this hypothesis, rodents can become infected by burrowing in soils that are contaminated with the remains or excreta of infected mammals or fleas. Other researchers have expressed skepticism about this hypothesis, noting methodological concerns about the few studies that have been advanced in its support or that the observed patterns of disease spread and host population recovery often fail to agree with the suggestion that new epizootics are initiated through contact of animals with contaminated soils (Gage and Kosoy, 2005). Also, unlike anthrax or certain other soil-dwelling organisms, *Y. pestis* does not form a sporelike structure, and most evidence suggests that plague bacteria die relatively quickly outside their hosts or vectors (Brubaker, 1991; Perry and Fetherston, 1997). Recently, some have presented evidence that plague might be able to survive in soil protozoa rather than in a free state in soils (Nikul'shin and others, 1992; Nersesov and Tsikhistavi, 1997; Domaradsky, 1999; Pushkareva, 2003). Recent studies also have shown that plague bacteria can form biolfilms on a nematode species (*Caenorhabditis elegans*) commonly used in laboratory studies (Darby and others, 2002), but we know of no evidence indicating that soil nematodes become infected under natural conditions. Others have suggested that *Y. pestis* might survive in plant tissues (Rivkus and others, 1993; Litvin, 1997) or in a latent nonculturable state in soils (Suchkov and others, 1997). Although none of these hypotheses has received strong support, they cannot be completely rejected on the basis of currently available data and are worthy of additional research.

Factors Affecting Rates of Plague Transmission and Incidence of Epizootics

One of the most striking aspects of plague is its ability to spread explosively among susceptible animal populations and across landscapes during epizootics. Almost equally striking is the fact that these relatively brief periods of intense transmission are followed by much longer intervals when the lack of obvious mortality among highly susceptible hosts makes it seem as if the disease has completely disappeared from a particular focus. In most instances, however, at least some *Y. pestis* transmission can still be identified in the suspect area through use of a sensitive monitoring technique, such as serosurveys of coyotes or other rodent-consuming carnivores (Gage and others, 1994). While much remains to be learned about the conditions that lead to plague epizootics or halt their progress, transmission rates can be affected by such factors as host resistance, densities of host and vector populations, the length of time that *Y. pestis* can persist in off-host flea populations, the vector competency of local flea species, the length of the extrinsic incubation period before fleas become infective for hosts, the likelihood that rodents will become chronically infected, periods of host inactivity (hibernation or aestivation), and seasonal changes and other climatic factors that influence the timing of host and vector life cycles as well as their survival and reproduction (Pollitzer, 1954; Pollitzer and Meyer, 1961; Poland and Barnes, 1979; Poland and others, 1994; Gage, 1998; Gage and Kosoy, 2005).

One of the most important questions in plague ecology is what conditions lead to the onset of epizootics. Modeling studies of human rat-associated plague suggest that if plague can persist in small rat subpopulations, it will spill over at irregular intervals to other susceptible rat subpopulations, causing epizootics and increased risks of flea-transmitted bubonic plague in humans (Keeling and Gilligan, 2000a,b). In these studies, persistence was favored by a high proportion of resistant individuals, and short-lived epizootics occurred when plague was introduced into subpopulations composed primarily (>80 percent) of susceptible individuals. In a more recent modeling study using rodent plague surveillance data from Kazakhstan, Davis and others (2004) reported that the invasion and persistence of plague in great gerbil populations was related to rodent density. They also found that as populations fell below certain thresholds, plague was likely to disappear from an area that had been invaded earlier in the course of an epizootic.

The suggestion that rodent population densities affect the invasion and persistence of plague in host populations is not surprising but still leaves open the question of what factors initially cause rodent populations in plague foci to increase and epizootic activity to become likely. Human plague risks typically increase greatly during epizootics, and the occurrence of increased numbers of human cases is generally believed to reflect increased epizootic activity. Parmenter and others (1999) analyzed human plague in New Mexico and found that human risks were correlated with increases in cool season precipitation from the previous year. They attributed this increase in human risk to a trophic cascade effect where increased cool season precipitation led to increased food availability for rodents. It was hypothesized that as food availability increases, so do survival and reproduction of

rodent hosts and perhaps flea vectors of plague. In agreement with the results of Davis and others (2004), they postulated that increased rodent numbers increase the risk of epizootics, as well as human cases. In a later study, Enscore and others (2002) demonstrated that both late winter precipitation and threshold temperatures were associated with human plague risks in the Four Corners region of the American Southwest. These last authors suggested that the trophic cascade model of Parmenter and others (1999) be modified to include threshold temperature effects that might affect not only rodent populations but also flea survival and reproduction. In particular, they suggested that years with exceptionally high numbers of days above certain threshold temperatures were likely to be those with low flea populations because of the negative effects of hot summer temperatures on flea survival and reproduction, or perhaps the ability of these insects to transmit plague (Cavanaugh, 1971; Cavanaugh and Marshall, 1972; Enscore and others, 2002). Collinge and others (2005b) attempted to test the generality of the trophic cascade model (Parmenter and others, 1999) as modified by Enscore and others (2002) and found that the occurrences of reported plague events in prairie dogs were not associated with certain climatic variables in Boulder County, Colo., but were associated with precipitation and temperature effects in a Phillips County, Mont., site. The authors concluded that the timing and magnitude of precipitation and temperature might influence the occurrence of plague in some but not all areas. They also reported that the best climatic predictors in the Montana site corresponded well with those noted in the above studies of human plague cases in the southwestern United States. In another Colorado study, Stapp and others (2004) demonstrated that epizootics in prairie dogs living on grasslands in north-central Colorado were associated with El Niño events.

Landscape Ecology of Plague

The influence of landscape structure on plague distribution and dynamics has been investigated in only a few of the world's plague foci. Bibikov and others (1963) stated that localities where plague infection can be maintained for a long period of time occupy relatively small portions of the territories that are endemic for plague, and speculated that, for unknown reasons, these sites present more auspicious conditions for the circulation of *Y. pestis* than other sites that are only affected sporadically. In other studies, Alexeev (1991) and Karimova (2002) used landscape characteristics for typing plague foci in desert zones of Kazakhstan and central Asia. Medzykhovsky and others (2001) demonstrated an association between the distribution of plague epizootics in the trans-Uralian steppe regions of eastern Kazakhstan and certain soil and grass characteristics. Serzhanov and others (1982a) found that places where plague persists over long periods of time in central Asian deserts are closely associated with landscapes characterized by abundant underground water lying near the surface (hydrologic lenses). These authors also demonstrated a correlation between the dynamics of plague epizootics and groundwater characteristics in nine different landscapes in Turkmenia. Based on these observations, Serzhanov and others (1982b) proposed the use of hydrothermal indices for the ecological typing of plague foci. In another interesting study, Rotshild (2001) hypothesized that levels of trace metals in natural environments influence the distribution and occurrence of plague. His hypothesis was based on multiple observations in the Altai Mountains, Tuva (eastern Siberia), the Kyzyl Kum desert in Uzbekistan, and a sandy semidesert area of the Caspian lowlands where he found correlations between epizootic plague activity and decreased or increased concentrations of Fe, Co, and Ti and low concentrations of Cu, Ni, and V.

In the United States, plague foci are known to occur in a variety of landscapes in numerous western mountain ranges, the High Plains, and intermountain grasslands (Barnes, 1982). Although plague might make brief epizootic intrusions into some areas, it remains conspicuously absent from certain extremely hot desert regions of the southwestern States, including the Sonoran Desert in southern Arizona. Although the reasons for plague's absence in these areas are unknown, it is tempting to speculate that the extremely hot, dry conditions in these desert areas are likely to limit transmission by fleas because these insects probably face severe desiccation when they are not closely associated with a host or protected burrow system or when they attempt to quest at burrow entrances in such exceptionally hot and dry environments.

A so-called "plague line" appears to exist at about the 100th meridian of longitude, a line that along much of its length marks the zone of transition from the tall grass prairies to the short grass habitats of the High Plains (Barnes, 1982). Among the factors that might influence the location of this "plague line" are rodent and flea diversity and changes in burrow microclimates or other features of burrow ecology. Although some recognized plague hosts occur on the plains, including black-tailed prairie dogs, thirteen-lined ground squirrels (*Spermophilus tridecemlineatus*), spotted ground squirrels (*S. spilosoma*), and southern plains woodrats (*Neotoma micropus*), the diversity of important plague hosts clearly decreases as one moves away from the Rocky Mountains onto the High Plains. By contrast, numerous rodent hosts of plague occur in relatively close proximity to each other in the lower elevation coniferous woodlands, foothills, and nearby plains. Many of these species, including woodrats, prairie dogs, ground squirrels, and chipmunks, live in burrows or complex nests that are often heavily infested with fleas. Another factor that might be important is the habitat complexity found near the Rockies and on High Plains sites nearest to these mountains. The more varied and patchy habitats around the Rockies could provide partial barriers and slow the movement of plague from one habitat to another, thus providing a limited refuge for some rodent populations and increasing

the likelihood that sufficient hosts will survive epizootics and keep transmission going from one season the next. As one moves on to the High Plains, however, the habitats appear to be more homogeneous with fewer barriers to the spread of plague, which could result in rapidly spreading epizootics that kill nearly all susceptible rodents and leave few individuals to support ongoing transmission. Plague probably is unlikely to persist in areas with such relatively homogeneous habitats but could, perhaps, repeatedly invade them when widespread epizootics sweep across the landscape.

Regional or local landscape ecology studies are almost nonexistent in the plague foci of the western United States. A single recent study by Collinge and others (2005a) used logistic regression to analyze two long-term data sets on plague occurrence in prairie dogs. The first of their two study sites was located in Boulder County, Colo., a region subject to rapid human development, and the second was in Phillips County, Mont. Associations were found at both sites between plague occurrence, landscape parameters, and colony characteristics. The best models from both sites predicted positive effects on plague occurrence of proximity to colonies that experienced plague and negative effects of road, stream, and lake cover.

Conclusions

Although some important findings, such as those describing how *Y. pestis* promotes its transmission by flea vectors, have occurred in recent years, many aspects of our understanding of plague ecology have progressed little since the mid-20th century. This is surprising when one considers the exciting new advances in many relevant fields or technologies, including molecular biology, immunology, population genetics, microbiology, geographic information systems, remote sensing, and mathematical modeling. Among the many interesting issues that have yet to be addressed satisfactorily are the degree to which *Y. pestis* exhibits adaptations to major hosts and vectors or vice versa, the relative roles of various factors in determining levels of host resistance, the roles many rodent species play in plague maintenance, the structure of plague foci in North America and elsewhere, the true significance of mechanical transmission or the transmission of plague bacteria by partially blocked fleas, the reasons why different flea species vary so greatly in vector competency, and the roles that climatic variables, landscape features, host and vector densities, or other factors play in influencing the spread of plague or the occurrence of epizootics. Fortunately, many of these questions can now be addressed, as interest in plague and funding for its study have increased as a result of recent concerns about the use of plague as a weapon of bioterrorism and the recognition that *Y. pestis* can adversely impact many wildlife species.

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