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Empirical assessment of a threshold model for sylvatic plague

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Plague surveillance programmes established in Kazakhstan, Central Asia, during the previous century, have generated large plague archives that have been used to parameterize an abundance threshold model for sylvatic plague in great gerbil (*Rhombomys opimus*) populations. Here, we assess the model using additional data from the same archives. Throughout the focus, population levels above the threshold were a necessary condition for an epizootic to occur. However, there were large numbers of occasions when an epizootic was not observed even though great gerbils were, and had been, abundant. We examine six hypotheses that could explain the resulting false positive predictions, namely (i) including end-of-outbreak data erroneously lowers the estimated threshold, (ii) too few gerbils were tested, (iii) plague becomes locally extinct, (iv) the abundance of fleas was too low, (v) the climate was unfavourable, and (vi) a high proportion of gerbils were resistant. Of these, separate thresholds, fleas and climate received some support but accounted for few false positives and can be disregarded as serious omissions from the model. Small sample size and local extinction received strong support and can account for most of the false positives. Host resistance received no support here but should be subject to more direct experimental testing.

Keywords: ***Yersinia pestis*; mathematical model; abundance threshold; fade out; vector-borne disease; invasion**

1. INTRODUCTION

Plague (flea-borne *Yersinia pestis* infection) is endemic in wild rodent populations throughout much of Central Asia. Monitoring systems were established in Soviet Central Asia *ca* 1950 to detect epizootics of plague in the rodent communities and to protect humans. These systems continue today. Surveillance consists of sampling the wild rodent and flea populations, and then attempting to isolate *Y. pestis* bacteria from rodent blood and organs and their fleas. When plague is found in areas close to human habitation, the risk of transmission to humans is reduced by treating rodent burrows with insecticide. The natural dynamics of plague have also been recorded in sparsely inhabited areas. In addition, serological data on the presence of plague antibody in rodents are available from the early 1970s, and ecological (mainly abundance) data on flea and rodent (especially great gerbil, *Rhombomys opimus*) populations have also been recorded since monitoring began. The resulting archives are unique in terms of the number of years sampled (1949–1996), the spatial extent of the sampling and the level of details available. A number of studies have developed various statistical approaches to analyse different aspects of these data (Davis *et al.* 2004; Frigessi *et al.* 2005; Park *et al.* 2006; Stenseth *et al.* 2006; Samia *et al.* 2007). The archives have also motivated more intensive capture–mark–recapture fieldwork where great gerbil populations were visited monthly and individuals were tested serologically for plague on each capture (Begon *et al.* 2006; Davis *et al.* in press).

A predictive model for plague was developed using a subset of the surveillance data from the PreBalkhash

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plague focus in southeastern Kazakhstan (Davis *et al.* 2004). This focus is one of many desert foci in Central Asia where great gerbils are regarded as the primary host (Gage & Kosoy 2005). The model is a rare example of applying the concept of a critical abundance threshold to a wildlife disease (Lloyd-Smith *et al.* 2005). It is based on a single threshold for invasion and persistence of plague, where the predictor is a weighted sum of abundance 1 year ago and abundance 2 years ago. Hence, information on abundance for the current and the previous year can be used to predict whether plague will be present in the following year. There is an explicit threshold, below which the chance of an epizootic (and hence of finding plague) is zero. Above the threshold, the likelihood that plague would be found increases as the predictor increases.

The predictive power of the model, however, was only tested on the same data as were used to derive it. Here, therefore, we test the model with further data collected at other sites in the same focus. Unsurprisingly, its performance is not perfect. We identify a series of hypotheses to explain the imperfect performance, test these and thus seek to both improve the predictive model and further our understanding of the plague–great gerbil system. Moreover, Lloyd-Smith *et al.* (2005) described the study of Davis *et al.* (2004) as one of the only two 'success stories' in the work on thresholds for wildlife diseases, but identified issues that should be addressed by all such studies in the future. Six of these aspects are considered here: separating invasion and persistence thresholds; the spatial structure of populations; environmental reservoirs; environmental variation (climate); the role of vectors; and host–pathogen coevolution.

2. PERFORMANCE OF MODEL

2.1. The plague archives and the model

For the purposes of monitoring plague, the whole of Kazakhstan is divided into 40 km×40 km squares ('large squares', LSQ). Each LSQ is further divided into four primary squares and each primary square into four 10 km×10 km sectors. Each record (plague prevalence in a sample of rodents or a measure of flea or gerbil abundance) is associated with a sector. Samples of fleas and gerbils were collected from early spring to late autumn, but not in the hottest months of summer, and each record is associated with a season (spring or autumn) and a year. In any one season, only a fraction of sectors were visited, and the selection of sectors was not random. Sectors were sometimes visited for which there had been local reports of dead or dying rodents, and once plague was isolated from a particular sector, there were attempts to establish the geographical extent of the epizootic. Some sectors were visited regularly and others rarely. Attempts to construct time-series at the sector scale result in severely fragmented series, even for the most frequently visited sectors. Therefore, constructing time-series from these data requires aggregation at either the primary or LSQ scale.

Great gerbil abundance was recorded as a density, obtained by estimating the fraction of burrow systems occupied, the average number of great gerbils per burrow system and the density of burrow systems per hectare. The last of these is considered a constant for each primary square, measured once by recording the diameters of 30 burrow systems lying on a single transect and dividing by 30 to obtain the average area per burrow system. Whether a burrow system is occupied or not can be determined by the signs found on the surface of the burrow system. The fraction occupied ('occupancy') was determined by visiting 30 burrow systems and recording how many were occupied. Occupancy is, in its own right, a simple measure of great gerbil abundance. The number of gerbils per burrow was estimated by counts of the maximal number simultaneously visible at 10 occupied burrow systems, observed from 20 to 30 m away for 15–30 min during good weather (so as to minimize the impact of variation in the activity level). Flea abundance was recorded in several ways. Whenever samples were collected to detect plague, counts of fleas were taken from captured gerbils to estimate the number of fleas per gerbil. Occasionally, flea density was estimated by collecting all fleas from 10 burrow systems, including fleas on great gerbils, fleas at the burrow entrances and fleas in the burrow system. This requires digging up and hence destroying whole burrow systems. Flea densities, like gerbil densities, were calculated by multiplying the average number of fleas in a burrow system by the fraction of burrow systems occupied and burrow system density.

A nonlinear (threshold) autoregression model was fitted to the time-series data on the presence/absence of plague (Davis *et al.* 2004). The presence or absence of plague was inferred solely from whether isolation attempts from great gerbils were successful. The serological data were not used since they were available for only part of the series and only past exposure to plague can be inferred from the presence of antibody. The model represents the simplest case in which there is a single threshold for both invasion and persistence of the pathogen (figure 1). The nonlinear regression function was the cumulative Weibull distribution function, which is given by

$$f(x) = \begin{cases} 0 & x \le \gamma, \\ 1 - \exp\left(-\left(\dfrac{x-\gamma}{\eta}\right)^{\beta}\right) & x > \gamma, \end{cases} \tag{2.1}$$

where γ represents a threshold value and η and β are the shape parameters. The predictor, x, is $0.59y_{t-1} + 0.87y_{t-2}$, where y_t is the proportion of great gerbil burrows occupied in year t obtained by taking the mean of spring and autumn estimates of occupancy. Occupancy performed at least as well as gerbil density in this model (Davis *et al.* 2004), but there are also practical reasons for focusing on the model using occupancy as the measure of abundance; field data on occupancy are simple and cheap to collect and arguably more reliable, since density calculations multiply occupancy by (unreliable) counts of animals observed active at a burrow.

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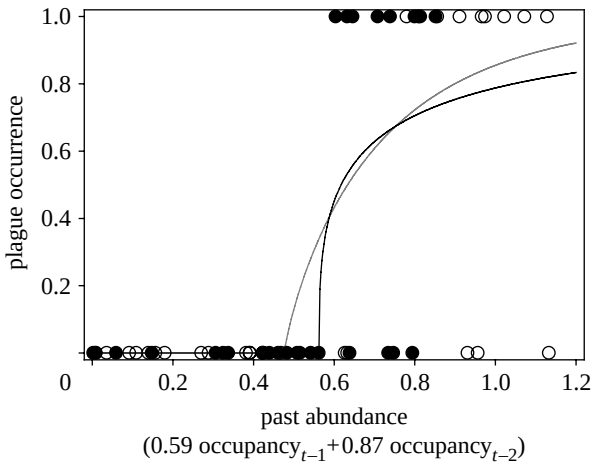

Figure 1. The original relation (solid grey line) between plague occurrence and past abundance measured in terms of burrow-system occupancy rates and the re-estimated relation (solid black line) having removed the final year of each observed outbreak, together with data on the presence/absence of plague at the two sites (with the removal of final years of outbreaks); Bakanas plain (open circles; LSQ 91) and Akdala plain (filled circles; LSQ 105).

The data used to fit the various models presented by Davis *et al.* (2004), and on which model selection was performed, come from two LSQs (91 and 105) within the PreBalkhash plague focus. This is a large (approx. 50 000 km^2) desert and semi-desert focus, situated southeast of crescent-shaped Lake Balkhash in eastern Kazakhstan. At these sites, monitoring was almost continuous from 1955 to 1996. The data collected outside these two areas provide an opportunity to test whether the threshold quantified for LSQs 91 and 105 is relevant for the whole of the focus. It is also possible to examine how the threshold relationship seen at the LSQ scale (40×40 km) performs at a finer primary square scale (20×20 km).

2.2. Testing the model

Occasions when spring and autumn estimates of occupancy for the previous 2 years were recorded *and* gerbils were tested for plague are required to evaluate the model. Where a seasonal estimate of occupancy was missing, but was available for the years before and after, a simple average of these was used. At the LSQ scale, this accounted for 40 out of 1564 estimates, and at the small square scale for 188 out of 3239 estimates. Davis *et al.* (2004) considered a sample of less than 100 gerbils too small to infer the absence of plague, as prevalence was often less than 1%. A similar approach is taken here. On a small number of occasions (which were retained), plague was detected, even though less than 100 gerbils were tested.

Four sets of occasions were collated: two using the data from LSQs 91 and 105 (aggregated at large and primary square scales) and two using the data from the rest of the focus (again aggregated at large and primary square scales). For the data from LSQs 91 and 105, the threshold model is already known to describe the data well. These results are presented again for comparison. For all four sets, we calculated numbers of occasions when the predictor was above the threshold and plague was isolated (confirmed positives), when the predictor was below the threshold and plague was not isolated (confirmed negatives), when plague was isolated but the predictor was below the threshold (false negatives) and when the predictor was above the threshold but plague was not isolated (false positives). We also calculated, from 1967 when data were available, the proportion of occasions falling into the last category, but for which there was serological evidence of plague. Finally, for each set of occasions, we calculated the expected number of years when plague should have been isolated, $e(P)$, by summing, across occasions, the probabilities given by the threshold model. That is, we acknowledge that even above the threshold, the model does not predict that plague should be isolated on all occasions (the probability was between zero and one, not one), and that some occasions when the predictor was above the threshold but plague was not isolated simply reflect this. In this sense, testing the threshold (above which plague *can* occur) is different from testing the threshold model. In the latter case, the estimated number of 'false positives' is the difference between the number of confirmed positives and $e(P)$.

2.3. Results

In LSQs 91 and 105, the model performed almost as well at the primary square scale as at the LSQ scale at which it was derived (table 1). The proportions 'confirmed' were 0.71 and 0.76, respectively, and there were only one and zero false negatives in the two cases. Notably, outside LSQs 91 and 105, too, at both spatial scales, the proportion of false negatives was low. In this case, however, the proportions when the predictor was above the threshold but plague was not isolated were much higher, i.e. 0.53 and 0.57 at the large and primary square scales, respectively, compared with 0.22 and 0.29 for LSQs 91 and 105. When comparing $e(P)$ in each case with the number of confirmed positives, we find that in LSQs 91 and 105 at the LSQ scale, the performance of the model was effectively perfect, as expected. At the primary square scale, $e(P)$ predicts 42 positive observations compared with 35 confirmed, i.e. an estimated 7 false positives, 6% of the total. Outside LSQs 91 and 105, at the large and small primary scales, respectively, the differences between $e(P)$ and the number of confirmed positives were 115.7 (27%) and 167.6 (31%). Hence, the threshold model performs well in predicting when plague will *not* be present, both at a different scale to that at which it was derived and when applied to different datasets, but it predicts plague will be found more often than it actually is.

3. EXPLANATIONS FOR THE HIGH NUMBERS OF FALSE POSITIVES

Evaluating possible explanations for the high number of false positive results has a number of purposes: it may suggest improvements to the model and its predictive powers; it may further our understanding of the

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Table 1. Performance of the model at two spatial scales. Data from LSQs 91 and 105 were used for model construction and selection. 'Elsewhere' refers to other large or primary squares in the PreBalkhash focus. Numbers following proportions refer to numbers of occasions. $e(P)$ is explained in the text.

	large square scale (40×40 km)		primary square scale (20×20 km)	
	91 and 105	elsewhere	91 and 105	elsewhere
occasions	65	421	110	537
plague predicted and isolated (confirmed positives)	0.38 (25)	0.17 (71)	0.32 (35)	0.15 (83)
plague neither isolated nor predicted (confirmed negatives)	0.38 (25)	0.29 (120)	0.39 (43)	0.27 (143)
plague isolated but not predicted (false negatives)	0.02 (1)	0.02 (9)	0.0 (0)	0.01 (5)
plague predicted but not isolated (false positives)	0.22 (14)	0.52 (221)	0.29 (32)	0.57 (306)
$e(P)$	25.1	186.7	42.0	250.6

plague–great gerbil system; and it addresses several of the issues raised in the review of Lloyd-Smith *et al.* (2005). Note again from table 1, however, that the number of false positives is lower than first appears: it is *not* the number of occasions when plague is predicted but not isolated. The model above the threshold, like the system whose behaviour it seeks to capture, is probabilistic. Even if the model were 'perfect', there would still be occasions when plague is predicted (with a probability less than one) but not isolated.

3.1. Separating invasion and persistence thresholds

In a host population that fluctuates in abundance, like the gerbils, a pathogen is predicted to invade when abundance rises above its invasion threshold (provided the pathogen has the opportunity to arrive in the population), persist for as long as it stays above its persistence threshold, but then 'fade out' when it drops below that persistence threshold (Lloyd-Smith *et al.* 2005). Briefly, an invasion threshold refers to the abundance of a wholly susceptible population within which a single infected individual could initiate an epizootic, whereas a persistence threshold refers to the abundance of a population in which an infection is already established, above which the flow of new susceptibles is sufficient to counteract the loss of susceptibles to infection, preventing the infection from fading out. That fade-out, however, is unlikely to be instantaneous, so the disease may still be observed after the threshold has been passed in a declining population. Thus, by relating the presence/absence of plague simply to host abundance, the invasion and persistence thresholds are necessarily confounded, and, at the end of epidemics, abundances are likely to be classified as above the threshold when they are actually below, leading to an underestimate of the threshold, which in itself will generate false positives. A direct test for separate invasion and persistence thresholds failed because more complex models with two thresholds failed to converge (see Davis *et al.* 2004). Hence, we assessed the importance of these factors by excluding the final year of each observed epizootic and refitting the Weibull threshold model (2.1). The threshold was re-estimated but the weights given to occupancy 1 and 2 years previously were fixed, so that any effect of removing end-of-outbreak data would be observed as a change in the threshold and the two models would be directly comparable (the weights and the threshold of the model are not independent). Relationships between annual occupancy estimates and the probability that plague was detected in gerbils (1, plague detected that year; 0, plague not detected) were assessed with generalized nonlinear regression models (Lindsey 2001) with binomial error. To account for serial dependence among years, year was added as a first-order autoregressive variable in a generalized nonlinear autoregression model (Lindsey 1999). Years where less than 100 gerbils were captured were omitted from the analyses because prevalence among gerbils was regularly less than 0.01.

Omitting the final year of each outbreak in LSQs 105 and 91 (7 years were omitted) and refitting the model raised the estimated threshold from 0.476 to 0.562 (figure 1). The 95% confidence interval for the re-estimated threshold is broad compared with that for the original model ((0.276, 0.811) compared with (0.355, 0.572)) and the estimates for the shape parameters also differ substantially. Fitting this new threshold on the data from the less sampled sites outside LSQs 105 and 91 reduces the number of false positives from 115.7 (27% of occasions) to 107.6 (26% of occasions) but doubles the number of false negatives (from 2 to 4%). Because these sites were sampled far less often, constructing time-series and distinguishing between the beginning and end of outbreaks so as to test the new threshold as only a predictor of invasion was not feasible.

3.2. Sample sizes

One straightforward explanation for at least some of the false positives is that plague was present but not observed simply because sample sizes were too low. Before examining this hypothesis, we first clarify that though there *is* a correlation between sample size and occupancy ($r=0.614$, $n=66$, $p<0.001$), the threshold model is not a consequence of this correlation, whereby plague was absent below the threshold simply because that is when sample sizes were low. In fact, sample size has no relationship with either the presence of plague (logistic regression, $\chi^2_1=0.457$, $p=0.499$) or the predictor based on past occupancy ($r=0.131$, $n=64$,

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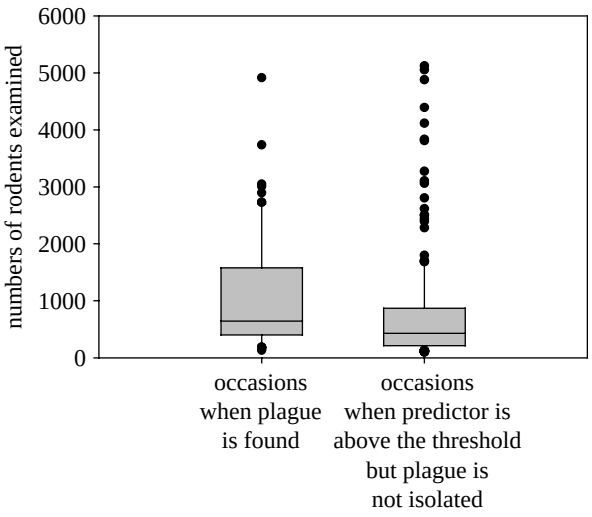

Figure 2. A box plot of the number of rodents examined when plague is isolated and the numbers examined when the predictor is above the threshold but it is not isolated (calculated for LSQs). Occasions when the number examined was less than 100 were not included since these were not used either when fitting the original model or when constructing table 1. The difference between the two groups is highly significant (Mann–Whitney *U*-test, $U=5893.5$, $p=0.001$, $N=71\,222$).

$p=0.302$), where we have once again been careful to avoid inferring absence of plague when sample sizes are very small (less than 100).

We test the hypothesis that false positives are associated with low sample sizes by restricting the data to those occasions where the threshold model predicts plague will be found. Figure 2 shows box plots of the numbers of rodents examined when plague was isolated, and the numbers when the predictor was above the threshold but plague was not isolated (sample sizes ≥ 100). Sample sizes were, on average, larger when plague was isolated (Mann–Whitney $U=5893.5$, $p=0.001$), but there were also numerous occasions when very large samples (several thousands of rodents) yielded no plague isolates.

Serological testing came into use in the early 1970s. The serological test is more sensitive than the bacteriological test in the sense that isolating plague bacteria requires an animal to be going through a period of bacteraemia, whereas antibodies are present consistently following a short period after initial infection, though antibodies may of course indicate a past rather than a current infection. Nonetheless, serological testing makes it more unlikely that an epizootic would go undetected. At the LSQ scale, there were 86 occasions in which (i) more than 100 gerbils were examined, (ii) plague was predicted by the model, and (iii) some gerbils tested seropositive. Of those 86, there were 45 (52.3%) in which all the gerbils examined were bacteriologically negative, even though some tested seropositive for plague. Serological testing detects not only an ongoing epizootic, but also the memory of a recent epizootic and 12 of the occasions were identified as being a final year of seropositive gerbils at the end of an epizootic. Even if these 12 are excluded, then still 33 out of 74 (44.6%) square-years that were identified as 'plague predicted but not isolated', i.e. false positives, were highly likely to be missed epizootic years due to low sample sizes and the difficulties in isolating bacteria from infected animals.

3.3. Local extinction of plague

At least one infectious individual (or other infectious material) is required for an epizootic to begin. If plague becomes locally extinct, then depending on the mechanism for the initiation of an epizootic, there may be long periods when host abundance is high but plague is absent. Many mechanisms that enable plague bacteria to persist over inter-epizootic periods were proposed when Fedorov (1944) first called the attention to the problem. One suggestion is that the bacterium can survive in the soil, or in invertebrates living in the soil, for long periods (Drancourt *et al.* 2006) and plague re-emerges among burrowing rodents when digging in infected soil. Others have favoured 'reinvasion' of plague from outside the area concerned, carried by either the reservoir host (Fedorov 1944; Naumov *et al.* 1959) or other species (Kalabukhov 1969; Shevchenko *et al.* 1969). The plague archives include several time periods when gerbil abundance was high over the whole focus and epizootics were detected in some squares but not others. If plague must reinvade, then the spatial pattern of observed epizootics should show a 'zone' of plague, within which epizootics occur and outside which they do not because these squares have not yet been invaded. That is, a 'zone' would suggest that false positives may occur simply because plague is not yet present in populations where it is capable of invading.

Data were pooled for the period 1977–1980. Populations in all squares were above the abundance threshold, but plague was isolated in only a subset of LSQs (5). Their spatial arrangement (figure 3) shows the type of spatial pattern associated with local extinction and reinvasion; the squares form a tight zone within which plague was found and outside of which it was not. A sharp decline in gerbil abundance over the whole focus in 1980 may explain why plague failed to spread any further and the epizootic as a whole died out. A similar pattern occurred between 1989 and 1995 (when the threshold condition was met in more than 90% of the squares), but plague was only found in the eastern parts of the focus. Figure 4 shows the cumulative distribution curves for nearest-neighbour distances between the infected squares in 1979, 1989 and 1995, along with a 99% confidence envelope of simulated complete spatial randomness (Diggle 2003). In each case, the observed nearest-neighbour distribution curve is outside the upper bound of the 99% simulation envelope. That is, the infected squares are highly significantly clustered together in a way that is consistent with plague spreading across the landscape from one or more points of invasion, rather than simply re-emerging locally across the focus once conditions allow. This indicates, in turn, that some false positives can be attributed to the local absence of plague bacteria. For example, in the period 1977–1980,

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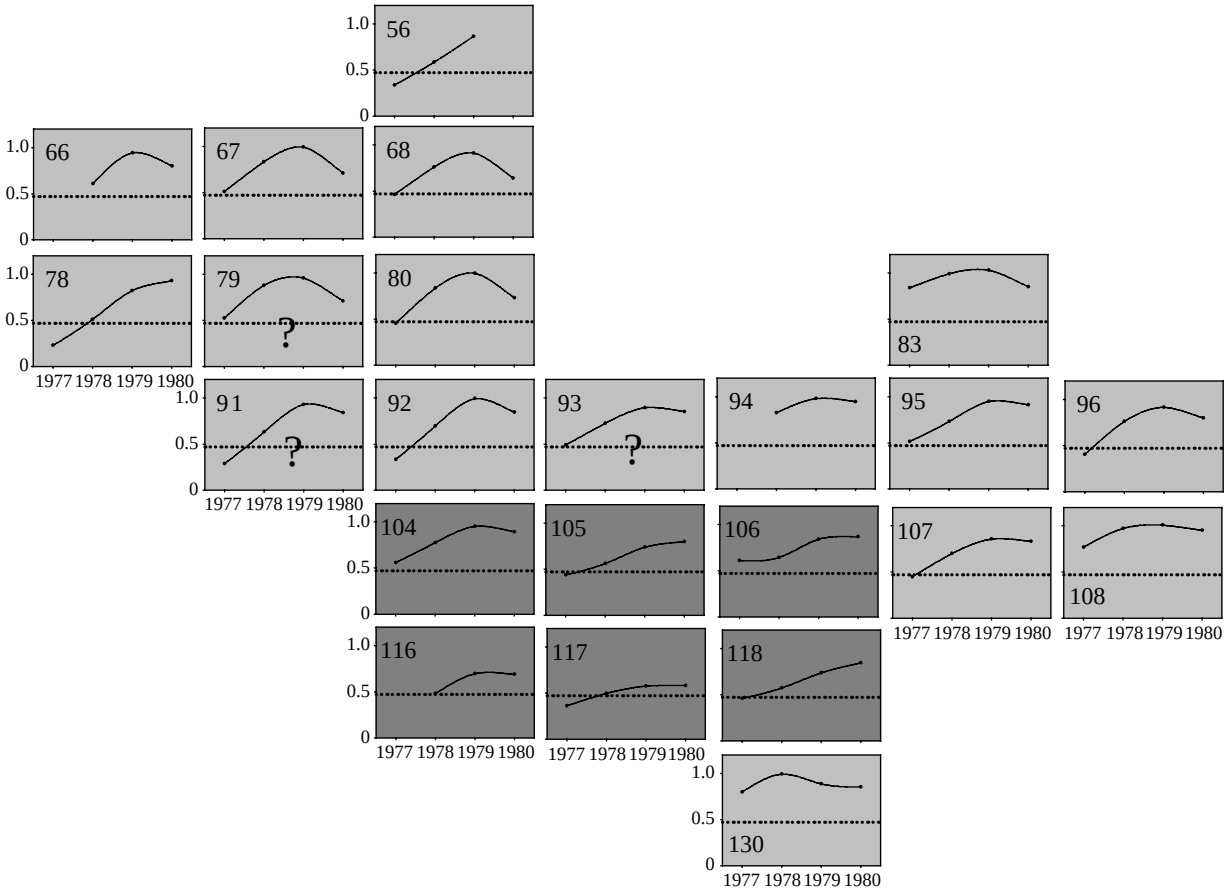

Figure 3. Each set of axes shows the predictor (see equation (2.1)) for a single LSQ for the period 1977–1980. The spatial arrangement of the axes is the same as the geographical arrangement of the LSQs in the PreBalkhash focus. The threshold value for the predictor is shown on each axis as a horizontal dotted line. Clearly, gerbil abundance was high over the whole of the focus. The backpane of each set of axes indicates whether plague was detected bacteriologically (dark grey indicating it was and light grey that it was not). A question mark on the axes indicates that a small fraction (less than 0.1%) of gerbils tested serologically positive.

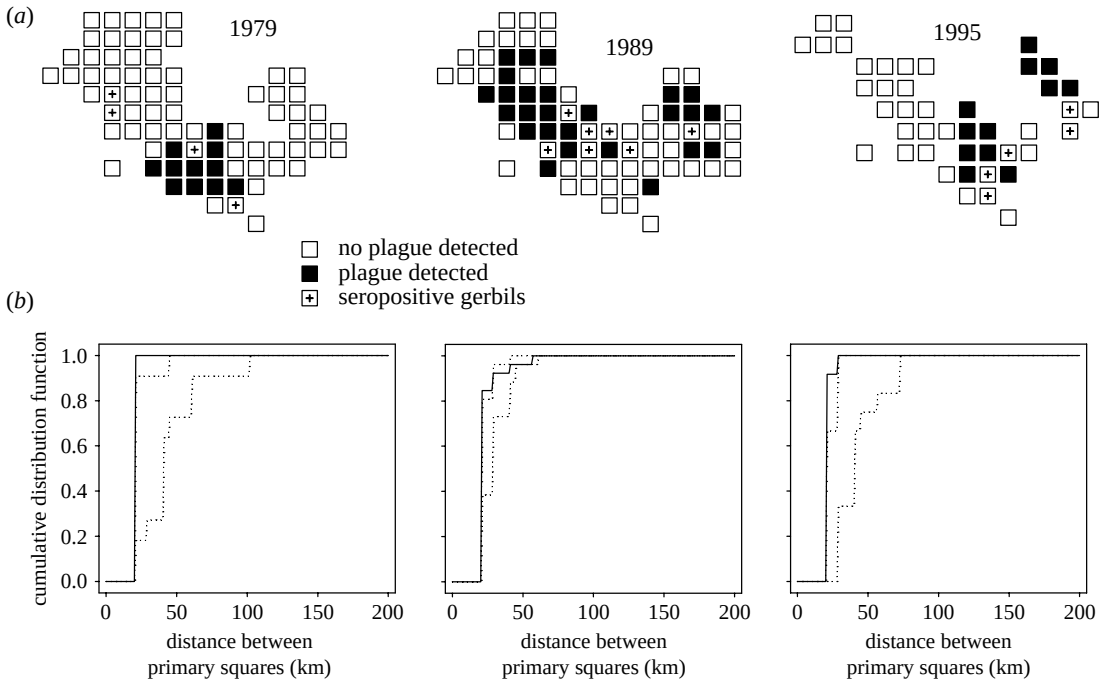

Figure 4. (*a*) Spatial arrangement of infected squares and (*b*) nearest-neighbour cumulative distribution curves for infected squares in the years 1979, 1989 and 1995. The nearest-neighbour distribution curves are plotted together with 99% simulation envelopes for complete spatial randomness (the number of squares equal to the number infected is chosen at random from all visited squares and a nearest-neighbour cumulative distribution function calculated; this is repeated 99 times to form upper and lower envelopes from maximum and minimum values). The years represent those in which plague was predicted for almost all squares and plague was detected in more than 10 of those squares. (*b*) The proportion of squares with populations above the abundance threshold.

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Table 2. AIC values for the set of logistic models with and without measures of flea abundance. Because the different measures of flea abundance are not available for the same years, the number of years used in the three subsets differs and the AIC values cannot be compared. All data are taken from LSQs 91 and 105.

flea abundance measure	intercept[a]	+gerbil predictor[b]	+flea measure[c]	+gerbil +flea[d]
previous autumn fleas	17.14	14.45	15.77	14.45
spring fleas	17.53	12.75	17.41	12.74
fleas/rodents	18.91	14.99	18.41	15.43

[a] Logit(outbreak) = a.
[b] Logit(outbreak) = $a + b \times$ gerbil occupancy.
[c] Logit(outbreak) = $a + c \times$ flea measure.
[d] Logit(outbreak) = $a + b \times$ gerbil occupancy $+ c \times$ flea measure.

there were 55 LSQ-years in which plague was predicted but not found, and 48 (87%) of these occurred outside the cluster of infected squares identified in figure 3.

3.4. Fleas

The many fleas that inhabit the burrow systems of the great gerbil are responsible for the transmission of plague between gerbils. Low abundance of fleas is therefore a very natural explanation for a delay in, or even the absence of, an epizootic. To assess whether flea abundance affects the presence/absence of plague for years in which burrow occupancy is above the threshold, we fitted linear logistic regression models with the probability that plague was detected in gerbils as the dependent variable. As explanatory variables, we used burrow occupancy and three measures of flea abundance: flea density in the previous autumn; flea density in spring; and numbers of fleas per gerbil (calculated by dividing estimates of flea density by estimates of gerbil density). Model comparison was based on the Akaike's information criterion (Burnham & Anderson 1998). The criterion is based on a data analysis philosophy that no model is true, rather the truth is far more complex than any model used. AIC can be used to compare different types of candidate models, given the response data (y) are equal. Models that differed in AIC by 2 or less were considered to be equally well supported by the data. However, because the different measures of flea abundance are not available for the same years, the number of years used in the three subsets differs and hence the AIC values cannot be used to compare models based on different measures of flea abundance. To account for serial dependence among years, year was added as a first-order autoregressive variable. In all three subsets, burrow occupancy significantly improved the models (table 2). An increase in occupancy increases the likelihood of detecting plague. Adding the flea measures does not further improve the models. When only flea abundance measures are introduced in the models, only flea density in the previous autumn significantly explains plague. However, this was strongly correlated with occupancy ($F_{1,119} = 20.06$, $p < 0.001$), which explains why flea density does not add extra information to the model.

3.5. Climate

Climate could affect plague dynamics by acting on great gerbil abundance, flea abundance or the plague bacterium directly (Stenseth *et al.* 2006). Climate does indeed appear to affect gerbil abundance (unpublished results), but the omission of climate from the threshold model cannot explain the false positives on these grounds, since abundance itself, whether or not it is affected by climate, is already integral to that model.

While flea abundance appears unable to explain why plague is absent from high-density gerbil populations, it may play a large role in determining the intensity of epizootics once they begin, and since flea abundance is sensitive to climatic variables, this is one way climate may affect plague dynamics. Stenseth *et al.* (2006) analysed the same archives but focused on prevalence levels during epizootics and their relationship to climate. Using a threshold very similar to that used here (but with separate conditions for spring and autumn), they found that *Y. pestis* prevalence in great gerbils increased with warmer springs and wetter summers. In particular, a 1°C increase in average spring temperature was predicted to lead to a more than 50% increase in bacteriological prevalence. They argued, moreover, that fleas played a major role in driving this effect, on the grounds that, for a subset of their data for which flea abundance was monitored, a model including spring and autumn flea abundance performed better (in terms of AIC) with the climate covariates suppressed than with them included.

In summary then, despite the evidence for *indirect* effects of climate via the host and vector populations, analyses of the available climate data for the Pre-Balkhash focus have so far suggested no direct effects of climate on the presence/absence of plague that could account for the observed false positives.

3.6. Host resistance

The great gerbil is known to have a heterogeneous response to infection with plague (Gage & Kosoy 2004), with some individuals dying soon after infection, others recovering and still others able to block the spread of infection completely. If populations with a recent history of high plague prevalence (and hence strong selection pressure) contain a high proportion of genotypically resistant gerbils, then this could explain the failure of plague to invade populations that appear to be above the abundance threshold. As a first attempt to assess this explanation, five epizootics were identified (separated by years in which plague was not isolated from any of the great gerbils collected): 1949–1951; 1956–1966; 1970–1974; 1979–1980; and 1988–1995. For

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Table 3. For each pair of epizootics, squares that were monitored during both epizootics and were positive for the first epizootic were classified as either '++' (involved in both epizootics) or '+−' (involved in the first but not in the second). Expected values are based on overall proportions of positive squares in the second epizootic.

first epizootic	second epizootic	++	+−	$E[++]$	$E[+-]$
1949–1951	1956–1966	7	0	5.8	1.2
1956–1966	1970–1974	10	9	8.7	10.3
1970–1974	1979–1980	5	5	2.3	7.7
1979–1980	1988–1995	3	1	2.7	1.3

great gerbils to live for more than 2 years is relatively unusual though by no means unknown and hence the gaps between the five epizootics do not represent large numbers of generations. If resistance plays a role in determining whether plague invades an area, then squares involved in one epizootic would be less likely to be involved in the next. For each consecutive pair of epizootics, squares that were monitored during both epizootics and were positive for the first epizootic were classified as either '++' (involved in both epizootics) or '+−' (involved in the first but not the second). The ratio of '++' to '+−' squares was then compared with the overall fraction of positive squares in the second epizootic (table 3). This revealed no evidence that resistance played a role in determining the spatial extent of these epizootics. In fact, for each of the four pairs, the number of '++' squares is higher than expected (though not statistically significant), implying there is a slight tendency for plague to be found in the same LSQs for consecutive epizootics. There are two LSQs where plague was found for all five epizootics (LSQ 105, analysed in Davis *et al.* (2004) and LSQ 117, which is adjacent to LSQ 105).

4. DISCUSSION AND CONCLUSION

It is now clear that the threshold model for plague epizootics in great gerbil populations in Central Asia, based solely on great gerbil abundance, works well in terms of predicting when plague will be absent. This is true both within the datasets used to generate the model and beyond them. However, outside the original dataset, an estimated approximately 30% of the predictions that plague will be present are 'false positives'. Simply listing the possible reasons for these is itself a useful exercise in suggesting how the threshold model might be improved, and in identifying the shortcomings of the simple theoretical concept of an abundance threshold when applied to a natural wildlife population (Lloyd-Smith *et al.* 2005). Of the reasons examined here, a number are plausible, and may even account for a small proportion of the false positives, but have received no support as serious omissions from the model, and, we suggest, can safely be disregarded. Conflating invasion and persistence thresholds and omitting explicit consideration of either climate or flea abundance come into this category.

Of the remaining reasons, small sample size appears to be a potential explanation for almost half of the false positive predictions, and while this is practically rather than scientifically important, this importance should not be underestimated. In the public health management of the plague–great gerbil system, 'accurate prediction' is itself more of practical than of scientific importance, since the ultimate aim of the threshold model is to direct pest controllers to sites and years where plague is a potential medical threat. It is important that public health managers understand that even when plague has not been detected, it may nonetheless be present, especially when the model predicts its presence and sample sizes are relatively small. Hence, controlling gerbil fleas in areas close to human habitation because plague is predicted would be sound practice, even if the presence of plague could not be confirmed. That is, these results reinforce rather than undermine the public health utility of the predictive model.

Of the other two reasons considered here, the local absence of bacteria necessary to initiate an epizootic and the local resistance of hosts following a previous epizootic, only the first received support. We emphasize though that we were unable to directly test the second hypothesis. A priority for future research therefore is to monitor levels of resistance in sites of recent epizootics by direct experimental means rather than through correlations. The performance of the model may furthermore be greatly improved by letting thresholds be site specific. This would effectively allow spatial heterogeneity in the landscape, soil type and climate, which, for instance, creates variation in burrow-system, gerbil and flea densities, to be realized in the model as variation in the threshold. The statistical modelling required for this approach is beyond the scope of the present article and is the subject of the future work. But whatever the underlying reason(s), it appears that 'location' should be able to explain a high proportion of the false positive predictions, and that as those reasons are elaborated, they should be incorporated into the predictive model.

Lloyd-Smith *et al.*'s (2005) review of critical thresholds in wildlife populations pointed to three specific shortcomings in the study by Davis *et al.* (2004). Of these, two, conflating invasion and persistence thresholds and omitting explicit consideration of vectors (fleas), have been dealt with directly here. The third is also arguably a feature of the current study, i.e. failure to account for the 1- or 2-year lag between breaching an abundance threshold and the outbreak of overt plague infection. Note, however, first, that the lag, while lacking an indisputable cause, is a practical asset (rather than a shortcoming) of the model in so far as it gives the model the predictive quality of an 'early

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warning system'. Second, that a lag in a *statistical* model may be inevitable, given that breaching a threshold *initiates* an epidemic and, as we have argued, plague may often be present but not detected as a result of low prevalence and/or insufficient sample sizes. Finally, the lag between when the population falls to lower levels of abundance and when plague is no longer detected is an entirely different lag and requires a different explanation. However, this lag may be easier to understand, since in the context of abundance thresholds, a sharp fall in abundance is associated with the beginning of fade-out rather than a sudden end to plague transmission. Hence, allowing the fact that some transmission will continue even at low levels of abundance (given an epizootic has occurred) and taking into account the lifespan of great gerbils, a 2-year lag is, if anything, surprisingly short.

The recent suggestion that plague reservoirs in the soil are important in initiating new epizootics at the end of inter-epizootic periods (Drancourt *et al.* 2006) receives no direct support from the present analysis. The reappearance of plague from soil reservoirs would require the reservoirs to remain viable for 5–8 years, but infect such low numbers of great gerbils in that same period that the presence of plague goes undetected. Given the large sample sizes for most LSQs from which absence is inferred, it seems unlikely that plague reservoirs in the soil are a common feature of the focus. Indeed, the very existence of a sharp abundance threshold in the plague–great gerbil system argues against an environmental reservoir, since models including a reservoir lack such thresholds throughout most of their parameter space (Bowers *et al.* 1993).

Overall, then, previous omissions in a rare 'success story' in the study of disease thresholds in wildlife (Lloyd-Smith *et al.* 2005) have been addressed. Possible reasons for false positives being generated by the Davis *et al.* (2004) model have been examined and divided into the unimportant and the potentially important ones. Further study of the latter is a priority in the search for a fuller understanding of plague dynamics in Central Asia.

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