Interaction between rodent species in agro-forestry habitats in the western Usambara Mountains, north-eastern Tanzania, and its potential for plague transmission to humans

R.H. Makundi, B.S. Kilonzo and A. W. Massawe*

Pest Management Center, Sokoine University of Agriculture, PO Box 3110, Morogoro, TANZANIA
*Corresponding author, email: rmakundi@suanet.ac.tz

Abstract. This study was carried out to determine rodent species composition and abundance, the interaction between them, and the possible implication in plague dissemination to humans. Over 2000 rodents were captured, identified and the relative species abundance determined. These animals belonged to six species, namely *Mastomys natalensis*, *Arvicanthis nairobe*, *Lophuromys flavopunctatus*, *Grammomys dolichurus*, *Mus* sp. and *Praomys* sp. They were distributed in two principal habitats, namely fallow land and forest. The distribution of the species overlapped, indicating interaction between them, but their abundance varied considerably between the habitats. Three species of fleas were collected from rodents. Of these, *Dinopsylus lypusus* was most abundant, followed by *Leptopsylla aethiopica* and *Nosopsyllus fasciatus*.

Rodent population densities declined rapidly in August and September and were followed by outbreaks of human plague in October. The observations made in the current study suggest that declining rodent population abundance leads to more 'free' fleas which probably seek alternative hosts, including humans. This consequently facilitates an increase in the transfer of plague from rodents to humans. The study further indicated that *M. natalensis* and *A. nairobe* form a continuum between forest-inhabiting rodent species and peri-domestic premises which therefore creates an avenue for transferring the disease from a potential forest reservoir to the human population.

The presence of specific anti-plague immunoglobulin (IgG and IgM) antibodies in blood sera of rodents was tested by enzyme-linked immunosorbent assay (ELISA). The presence of *Yersinia pestis* DNA was tested by polymerase chain reaction (PCR). Both tests revealed that *M. natalensis, A. nairobe*, *Rattus rattus* (captured in houses) and *L. flavopunctatus* were the potential rodent reservoirs of plague in the western Usambara Mountains. *Grammomys dolichurus* and *Praomys* sp. tested negative for plague, but more specimens will be tested to confirm this finding.

Introduction

The western Usambara Mountains, in north-eastern Tanzania, have experienced dramatic ecological changes brought about by government decisions made 40 years ago to open the mountains for agriculture (J. Bell, unpublished report). As a result of these decisions, human pressure on the remaining fragments of the natural montane rainforest has become a serious problem. The immediate impact of opening up the forest for agricultural activities is the destruction of natural habitats and fragmentation of others, which have, in general, some effects on species composition, diversity and distribution. Equally important is that some species are likely to disappear, while opportunistic species, particularly *Mastomys natalensis* and *Arvicanthis nairobe*, find suitable conditions in the newly cultivated land for immediate colonisation. Factors related to land management practices also affect how rodent species interact with each other and with the human population. Rodent–human interaction in the western Usambara Mountains is more evident in the form of crop damage and the spread of diseases, mainly sylvatic plague. Plague has been known in East Africa for many years (Roberts 1935), but remains persistently epidemic in only a few localities. In Tanzania, persistent plague outbreaks during the last two decades have occurred only in the western Usambara Mountains (Kilonzo et al. 1997). In view of the persistence and epidemic nature of the disease, several studies have been carried out in the past to elucidate the factors involved in maintaining the disease in the area (Kilonzo and Mhina 1982; Kilonzo and Msangi 1991; Kilonzo et al. 1992, 1997). It has been suggested that some species of rodents found in the area are potential reservoirs and carriers of the disease (Kilonzo and Mhina 1983; Njunwa et al. 1989; Kilonzo et al. 1992). Rodent fleas, of which several species have been identified, are suspected to be the principal vectors of the disease (Njunwa et al. 1989; Kilonzo et al. 1992; Makundi and Kilonzo 1994). House-infesting fleas, particularly *Pulex irritans*, occur abundantly, but their role in the transmission of the disease in Lushoto District is not well understood. This species has been reported to be important in inter-human transmission in other countries (Karimi and Farhang-Azal 1974; Twigg 1978). Ecological factors responsible for the outbreaks and persistence of plague in Lushoto District have not been adequately studied. Previous studies investigated the breeding patterns of some of the rodent species (Makundi 1995). Ecological studies on a wider area were initiated in the western Usambara Mountains in May 1998. The long-term objective of these studies is to develop ideas for ecological approaches to management of both rodents and fleas in the plague outbreak foci.

We aimed to show that species interaction does occur in suitable habitats due to habitat fragmentation and overlap, and that both savanna and forest species are most likely to be reservoirs of plague in the areas, by examining the following hypotheses:

(i) the existence of suitable reservoirs of plague with a stable population in the natural forest reserve is responsible for maintenance of the disease;

(ii) the interaction of savanna and forest-inhabiting rodent species enables the transfer of plague from forest animals to peri-domestic rodent species; and

(iii) both forest and savanna species in the area are equally exposed to plague bacteria, *Yersinia pestis*, and are responsible for its dissemination to people.

Materials and methods

The study area

The study was carried out in Shume Ward, Lushoto District, in the western Usambara Mountains. Shume Ward is located north of Lushoto town (04°42'16"S, 38°12'16"E) and has experienced persistent outbreaks of human plague. The climate of the area has pronounced temperature and humidity differences during the year. The coldest months are June to September with the lowest temperature usually in July (mean temperature = 19°C). It is usually warmer between December and March when the mean temperature ranges from 25–26°C. The rainfall pattern is characterised by two discernible rainy seasons, with the wet season extending from late February/early March to end of May. Short rains fall mainly in November and December, but usually start towards the end of October and extend to January. July to mid-October is usually a dry period.

Trapping procedures

Removal trapping of rodents was carried out for 12 months between May 1998 and May 1999. Trapping was carried out for 6 days each month using Sherman live-traps baited with peanut butter mixed in maize bran. The traps were placed in four lines, each consisting of 25 traps, and approximately 10 m apart in the fallow land. In the forest, there were 2 lines of 25 traps each, 10 m apart and 10 m between trap stations. The traps were inspected every morning, with captured animals removed and taken to the laboratory for processing. The animals were brushed to remove fleas, which were later identified. Additionally, 10 traps baited with peanut butter were placed in houses (1 trap/house) for three consecutive nights to capture *Rattus rattus*. Sera from captured animals were used to test for anti-plague antibodies.

A correlation analysis was carried out between the flea load on hosts and the incidence of plague between October and March.

Tests for *Yersinia pestis* DNA and anti-plague antibodies

Venous blood was collected from the orbital sinus of rodents and centrifuged to separate serum. *Mus* sp. was excluded because sufficient sera could not be obtained. All specimens of *R. rattus* (n = 12) captured in houses were included in these tests. The sera were preserved at –20°C and selected samples were tested for the presence of anti-plague immunoglobulin (IgG and IgM) antibodies by enzyme-linked immunosorbent assay (ELISA) and for *Yersinia pestis* deoxyribonucleic acid (DNA) by polymerase chain reaction (PCR), as described by Chu (2000). Human plague cases were obtained from the records of the local district hospital.

Results and discussion

The two principal rodent habitats (forest and fallow) were characterised by fragmentation brought about by deforestation and agricultural activities. Cultivated land interspersed the two principal habitats forming a continuum with peri-domestic areas and human settlements. Separation between these three main rodent habitats was minimal, and therefore allowed rodent movements between them (Figures 1 and 2).

Within the fallow land, *M. natalensis* and *A. nairobe* were the dominant species. Other species found in fallow habitats in relatively smaller numbers included *Grammomys dolichurus*, *Mus* sp. and *Lophuromys flavopunctatus* (Figure 2). Temporal and spatial fluctuations of rodent numbers occurred. Species inhabiting the forest, despite the lower trapping effort relative to the effort in fallow habitats, were at lower densities throughout the year. The distribution of *M. natalensis* and *A. nairobe* extended into the forest while *L. flavopunctatus* was also found in the fallow land close to the forest.

Three species of fleas were collected from rodents. These were, in order of abundance, *Dinopsylus lypusus*, *Leptopsylla aethiopica* and *Nosopsyllus fasciatus.* The monthly mean numbers of fleas (flea index) for all rodent hosts collected are shown in Figure 3. There was a marked increase in the abundance of fleas on rodents between August and September, and more fleas on rodents in October than in any other month. Fleas were found mainly on *M. natalensis*, *A. nairobe* and *L. flavopunctatus*. No fleas were found on *Mus* sp., and relatively few were collected on *Praomys* sp. and *G. dolichurus*.

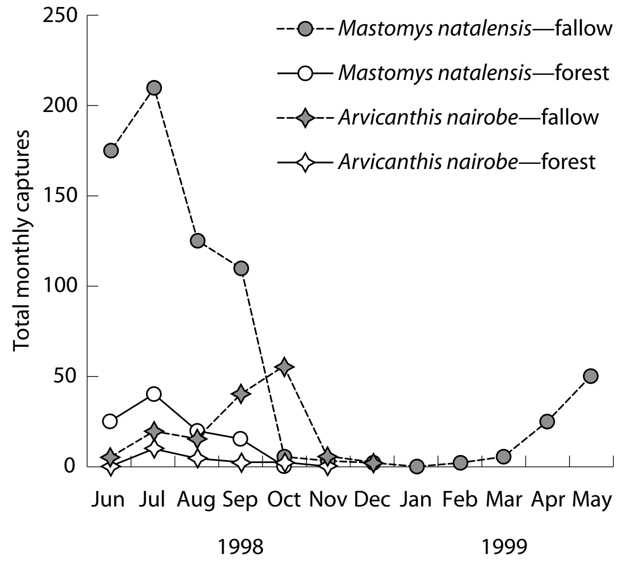

Figure 1. Abundance of *Mastomys natalensis* and *Arvicanthis nairobe* in fallow and forest habitats in Shume Ward, Lushoto District, Tanzania in 1998–1999.

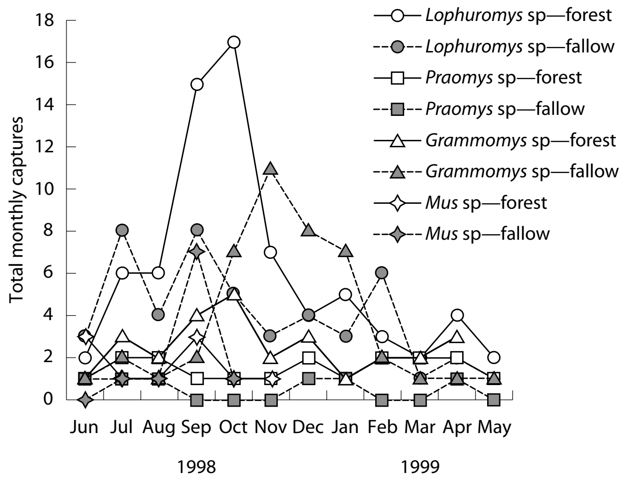

Figure 2. Abundance of non-savanna species of rodents in fallow and forest habitats in Shume Ward, Lushoto District, Tanzania, in 1998–1999.

The data show considerable variation in abundance of rodents from one season to another, and within and between habitats. Seasonal variations in the abundance of the population of *M. natalensis* were well pronounced, with the highest densities occurring after the rainy season and during the onset of the dry season in July. It is noteworthy that fluctuations of rodent populations in the forest, particularly *L. flavopuncatus*, were just as dramatic relative to the lower populations present. Although these species occupy a more stable habitat, it is uncertain how fragmentation and agricultural encroachment have affected their population dynamics. More is known about the ecology of *M. natalensis* and the factors causing population fluctuations in other areas of Tanzania (Leirs 1992; Christensen 1996; Leirs et al. 1996, 1997; Mwanjabe and Leirs 1997) than for the rodent species found in the western Usambara Mountains.

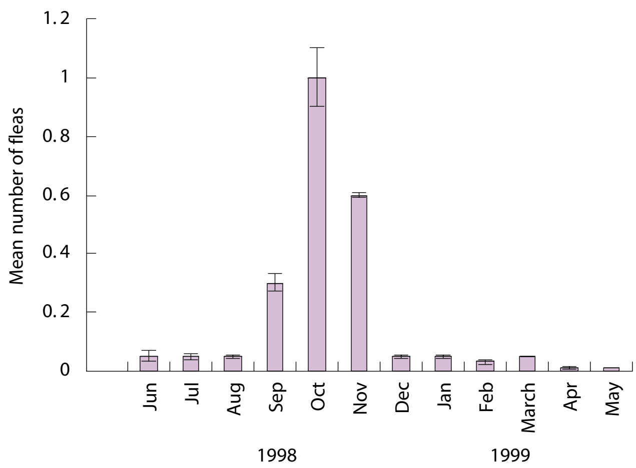

Figure 3. Changes in abundance of fleas on rodent hosts in Shume Ward, Lushoto District, Tanzania.

There has been a great human impact on the ecology of the western Usambara Mountains caused by agricultural encroachment into the natural forest. This has led to modifications of habitats, making them less suitable for forest-inhabiting species, but more favourable for colonisation by savanna species, specifically *M. natalensis* and *A. nairobe*. The distribution of these species overlaps with that of species found in the remaining forested area. This overlap of habitats leads to unrestricted interaction and potentially makes it possible for exchange of fleas and the disease pathogen between them. The presence of specific anti-plague IgG and IgM antibodies and *Y. pestis* DNA in rodent sera is shown in Table 1. Rodent species that tested positive for *Y. pestis* DNA and anti-plague antibodies probably are the natural reservoirs of the disease. It could also be inferred that the species inhabiting the forest are most likely to maintain the disease throughout the year since a residual population is present when the savanna species populations are extremely low. Before the disease outbreak, the most important link between forest and peri-domestic species appears to be *M. natalensis* and *A. nairobe* because they occurred in relatively large numbers and due to their close proximity to both peri-domestic and forest-inhabiting rodent species. Both species were positive for *Y. pestis* DNA, and anti-plague IgM and IgG antibodies, indicating previous and current infection/exposure.

It is likely that the decline in rodent populations, particularly *M. natalensis* and *A. nairobe* from October–March, led to more fleas seeking alternative hosts. The increasing number of fleas on rodent hosts between September and October was associated with increasing human plague cases in October and November. However, the number of fleas on rodents declined throughout December to May. Plague prevalence was also low in December, but increased again in January and declined in subsequent months. The declining numbers of rodent hosts, particularly *M. natalensis* and *A. nairobe*, did not lead to a marked increase in the flea load on remaining hosts, but there was an increase in human plague cases, particularly in November and January (Figure 4).

Table 1. Presence of anti-plague immunoglobulin (IgG and IgM) antibodies and *Yersinia pestis* DNA in rodent sera in the western Usambara Mountains, north-eastern Tanzania (NT = not tested).

Species	No. of sera (samples) tested	Percentage with *Y. pestis* DNA	Percentage positive	
			IgG	IgM
Mastomys natalensis	86	10.5	9.4	15.3
Arvicanthis nairobe	25	28.0	23.1	26.9
Lophuromys flavopunctatus	14	7.1	7.7	15.4
Rattus rattus	12	16.7	8.3	16.7
Grammomys dolichurus	14	0	NT	NT
Praomys sp.	6	0	NT	NT

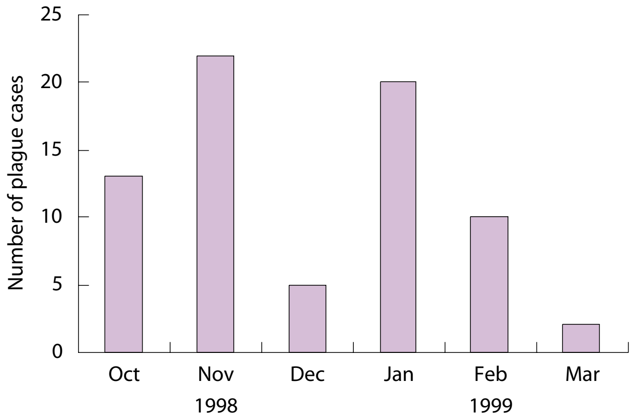

Figure 4. Human plague cases during the 1998–99 outbreak in Shume Ward, Lushoto District, Tanzania.

The correlation between flea loads on rodents and plague cases in humans (r = 0.397, p = 0.43) was weak and not significant, indicating that once the disease outbreak occurs, transmission to humans may not be mediated by fleas but possibly by inter-human transmission. This suggests that inter-human transmission of the disease, probably mediated by the human flea (*Pulex irritans*) in houses, was responsible for the plague cases recorded in January after the initial rodent to man transmission between October and November. Therefore, it appears that during plague outbreaks in the Usambara Mountains, there are two distinct phases in transmission of the disease. In the first phase, fleas from rodents play a central role in disseminating the disease to man, while in the second phase, the disease is disseminated between people by *P. irritans*. This follows a trend similar to that reported by Karim and Farhang-Azad (1974) and Twigg (1978), and probably occurs when there are few natural hosts of the disease in the fallow and cultivated areas, particularly in November to March. However, inter-human transmission by droplet infection cannot be ruled out as contributing to the disease outbreaks, especially when the disease advances to pneumonic status among some patients (Kilonzo et al. 1997).

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