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## Observations on the current status of plague endemicity in the western Usambara mountains, north-east Tanzania

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### Summary

Epidemiological investigations were executed at 6 locations in the western Usambara mountains in north-east Tanzania from November 1980 to May 1982. Rodent, human and dog sera were checked for agglutinating plague antibodies, using the passive haemagglutination test. Fraction I plague antigen was similarly tested for in rodent organ macerates. *Bubo* aspirates and rodent organ-smears were microscopically examined for plague bacilli. A total of 257 rodents and 191 fleas were collected. *Rattus rattus*, *Praomys natalensis* and *Arvicanthis niloticus* were the commonest rodent species. Flea ectoparasites mostly comprised of *Xenopsylla brasiliensis* and *Dinopsyllus lypusus*. Specific plague antibodies were detected in 2.8%, 10% and 2.9% of the rodents examined in November 1980, November 1981 and May 1982 respectively. Similarly, plague antibodies were found in 0.6%, 1.2% and 0.5% of the human sera tested in September 1981, November 1981 and May 1982 respectively. All the dog sera were negative. The results suggested past contact with plague and existence of a persistent plague focus in the area. Health education for villagers, maintenance of a plague surveillance programme and regular seminars for medical and health personnel in the area were recommended in order to prompt early detection, reporting and control of an outbreak.

**Key words:** agglutinating plague antibodies; rodents; plague endemicity; fleas.

### Introduction

The first recorded outbreak of human plague in the western Usambaras in north-east Tanzania occurred in April 1980 (Kilonzo and Mhina, 1982). This

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outbreak which was mostly bubonic and which involved 49 cases with 11 deaths (Mkami, 1980 – pers. comm.), was attributed to possible introduction of the pathogen from the then active foci in southern Kenya (Kilonzo and Mhina, 1982). Most villagers in the affected area have close economic and social relationships with villagers at Rongai in Rombo district and South Pare in Same district, both in north-east Tanzania (Mtera, 1980 – pers. comm.). Since the two areas constitute endemic plague foci (Msangi, 1968), it is possible that the disease spread from there. Whatever the origin, however, the infection remained active in the western Usambara mountains for about six months (Mkami, 1980 – pers. comm.).

In order to establish the status of its endemicity, periodic studies were initiated in the affected villages and their surroundings three months after the last victim of the first outbreak recovered. The aim of the present paper is to report the bacteriological, serological and entomological observations made in these studies.

### Materials and Methods

All the investigations were performed at Mkunki (the village affected by the first outbreak) and the neighbouring villages of Shume-Nywelo, Manolo, Gemai, Viti and Lokome in the western Usambara mountains of Tanga Region, Tanzania (Fig. 1). The choice of these villages was based on the close social and economic relationship of villagers with those of Mkunki and on the periodic occurrences of human plague after the first outbreak. The investigations were done in November 1980, March, June, September and November, 1981 and May 1982.

Commensal and field rodents were live – trapped in the selected villages, using box traps baited with roasted chicken mash. The animals and their fleas were anaesthetised with ether, identified, counted and processed as described before (Kilonzo, 1976; Kilonzo and Mhina, 1982).

Cardiac blood was collected from each animal carcass and serum was separated and tested for agglutinating plague antibodies, using the Passive Haemagglutination (PHA) and Passive Haemagglutination Inhibition (PHAI) tests as described elsewhere (WHO, 1970; Cavanaugh and Bahmanyar, 1976; Kilonzo and Mhina, 1982). All the positive sera were adsorbed with 50% formalin-treated sheep erythrocytes to remove any heterogenous antibodies, and then re-tested to detect specific plague antibodies. The lowest serum dilution used was 1:20.

Pieces of heart, spleen, liver, axillary and inguinal glands of each rodent carcass were removed and used for making impression smears. The smears were fixed with a 1:1 solution of ether: methanol, stained with 2% methylene blue and examined for bipolar-staining bacilli. The remaining organ pieces were subjected to ether-extraction of proteins, using the technique of Larson et al. (1951). The protein extracts were serologically tested for presence of Fraction I plague antigen, using the passive haemagglutination test mentioned above.

The flea specimens were occasionally crushed and smeared on microscope slides, dried, fixed, stained and examined for bipolar staining bacilli as described for rodent-organ smears.

Male and female human subjects of varying ages (villagers and school children) who voluntarily accepted to have their blood examined were involved in the studies. Venous blood (2 ml) was aseptically collected from each subject and serum was separated and tested as described for rodents. Occasionally, venous blood and consequently serum was obtained from domestic dogs and similarly tested for agglutinating plague antibodies.

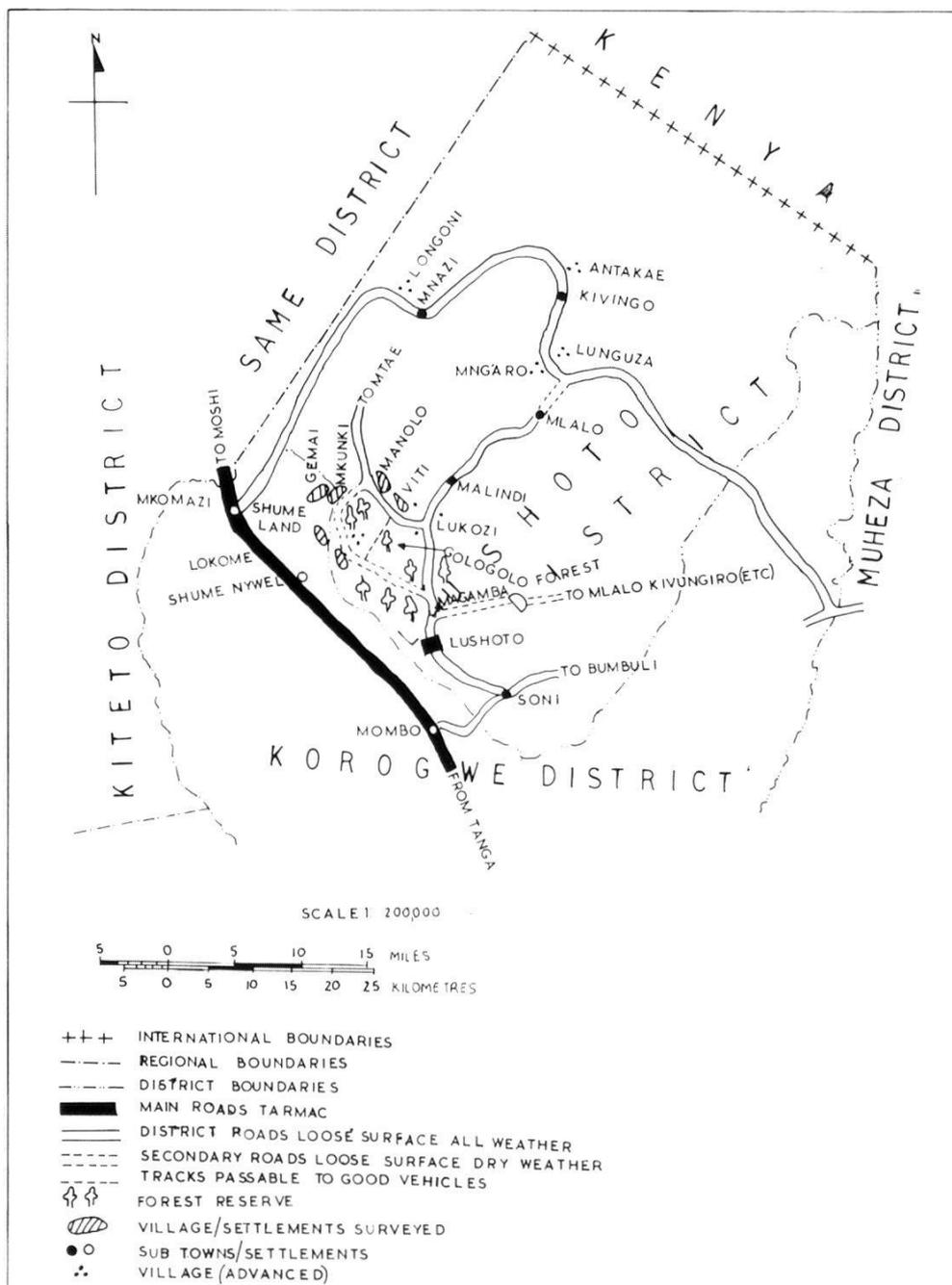


Fig. 1. Villages surveyed for plague in the western Usambaras.

## Results

A total of 257 rodents, 191 fleas and 810 human sera were examined (Tables 1 and 2). The rodents consisted of *Rattus rattus* (49.8%), *Praomys natalensis* (36.6%), *Arvicanthis niloticus* (11.3%), *Lophuromys sikapusi* (1.6%), *Tatera robusta* (0.4%) and *Gramomys dolichurus* (0.4%). The fleas comprised of *Xenopsylla brasiliensis* (42.4%), *Dinopsyllus lypusus* (41.4%), *Ctenophthalmus calceatus* (5.2%), *Echidnophaga gallinacea* (6.8%), and *Nosopsyllus fasciatus* (4.2%). Of all the rodents captured, 89 (34.6%) hosted fleas, making a total and

Table 1. Flea infestation and plague infection of rodents captured in the western Usambara mountains from November 1980 to May 1982

| Rodent species             | Species and Nos. fleas |                |                |              |                         |                        |                  |                     |                      |                     |       |             |                |
|----------------------------|------------------------|----------------|----------------|--------------|-------------------------|------------------------|------------------|---------------------|----------------------|---------------------|-------|-------------|----------------|
|                            | No. caught             | % of each spp. | No. with fleas | % with fleas | No. and % positive sera | <i>X. brasiliensis</i> | <i>D. typsus</i> | <i>C. calceatus</i> | <i>E. gallinacea</i> | <i>N. fasciatus</i> | Total | Total index | Infested index |
| <i>R. rattus</i> .....     | 128                    | 49.8           | 43             | 33.6         | —                       | 76                     | 7                | 2                   | 13                   | —                   | 98    | 0.8         | 2.3            |
| <i>P. natalensis</i> ..... | 94                     | 36.6           | 33             | 35.1         | 2 (2.1%)                | 3                      | 52               | 2                   | —                    | 3                   | 60    | 0.6         | 1.8            |
| <i>A. niloticus</i> .....  | 29                     | 11.3           | 10             | 34.5         | 2 (6.9%)                | 2                      | 17               | 5                   | —                    | 3                   | 27    | 0.9         | 2.7            |
| <i>L. sikapusi</i> .....   | 4                      | 1.6            | 2              | 50           | —                       | —                      | 3                | 1                   | —                    | —                   | 4     | 1.0         | 2.0            |
| <i>T. robusta</i> .....    | 1                      | 0.4            | 1              | 100          | —                       | —                      | —                | —                   | —                    | 2                   | 2     | 2.0         | 2.0            |
| <i>G. dolichurus</i> ..... | 1                      | 0.4            | —              | —            | —                       | —                      | —                | —                   | —                    | —                   | —     | —           | —              |
| Total                      | 257                    | 100            | 89             | 34.6         | 4 (1.6%)                | 81                     | 79               | 10                  | 13                   | 8                   | 191   | 0.7         | 2.1            |
| % of each species of flea  |                        |                |                |              |                         | 42.4                   | 41.4             | 5.2                 | 6.8                  | 4.2                 | 100   |             |                |

Table 2. Serological observations of human plague in the western Usambara mountains from November 1980 to May 1982

| Age groups<br>(Years) | Numbers of sera examined |         |       | Numbers of positive sera |          |          |
|-----------------------|--------------------------|---------|-------|--------------------------|----------|----------|
|                       | Males                    | Females | Total | Males                    | Females  | Total    |
| 0-5 .....             | 22                       | 20      | 42    | -                        | 1 (5%)   | 1 (2.4%) |
| 6-15 .....            | 197                      | 141     | 338   | -                        | 2 (1.4%) | 2 (0.6%) |
| 16-25 .....           | 85                       | 93      | 178   | -                        | -        | -        |
| 26-35 .....           | 54                       | 51      | 105   | -                        | -        | -        |
| 36-45 .....           | 24                       | 18      | 42    | -                        | -        | -        |
| Above 45 .....        | 51                       | 54      | 105   | -                        | 1 (1.9%) | 1 (1.0%) |
| Total                 | 433                      | 377     | 810   | -                        | 4 (1.1%) | 4 (0.5%) |

infested-flea indices of 0.7 and 2.1 fleas per rat, respectively (Table 1). Two (6.9%) *A. niloticus* captured in November 1980 contained agglutinating plague antibodies at a serum titre of 1:40. Similar antibodies were detected in 2 (2.1%) *P. natalensis* captured in November 1981 and May 1982, respectively.

The antibody titres of the two positive *P. natalensis* were respectively 1:40 and 1:160. Organ impression smears of 6 animals (4 *P. natalensis*, 1 *R. rattus* and 1 *L. sikapusi*) contained bipolar-staining bacilli. All the rodent-organ extracts lacked fraction I plague antigen. A total of 11 domestic dogs were serologically examined for plague antibodies in September 1981 but they were all negative.

Of all the human subjects examined, 4 (0.5%), all females, contained agglutinating plague antibodies (Table 2). These antibodies the titres of which ranged from 1:20 to 1:80 were detected in September 1981 (1 case), November 1981 (2 cases) and May 1982 (1 case). Furthermore, typical bipolar staining bacilli were observed in axillary bubo aspirates from a 25-year-old lady who, in November 1981, was clinically feverish and had a severe headache. Historical investigations carried out indicated that none of the positive cases had travelled outside her home district during the previous one year.

## Discussion

According to Suzuki and Hotta (1979) an agglutinating antibody titre (when tested with fraction I plague antigen) of 1:16 is specific for *Yersinia pestis*. In contrast, however, Kanatov (1975) incriminated 1:20 as the specific titre for *Y. pestis* agglutinating antibodies. The presence of agglutinating plague antibodies in 2 *A. niloticus*, 2 *P. natalensis* and 4 people at serum dilutions ranging from 1:20 to 1:160 therefore strongly suggests past contact with plague. Since none of the positive subjects had travelled outside the district during the past

Table 3. Personal details\* of human plague cases/suspects during the outbreak of October/December 1981

| Case No. | Age (Years) | Sex (M/F) | Date of onset           | Date of death or recovery | Reported main signs and/or symptoms         | Remarks/Village  |
|----------|-------------|-----------|-------------------------|---------------------------|---|--|
| 1        | 1           | F         | 28. 10. 81              | 30. 10. 81                | Fever, headache                             | Infant at Gemai  |
| 2        | 15          | F         | 4. 11. 81               | 6. 11. 81                 | Fever, headache                             | Student at Gemai, Pr. School   |
| 3        | 11          | F         | 6. 11. 81               | 8. 11. 81                 | Fever, headache                             | Student at Gemai, Pr. School   |
| 4        | 23          | M         | First week of Nov. 1981 | 13. 11. 81                | Fever, headache                             | Student teacher at Gemai Pr. School. Felt ill while at Bendera (Bottom of the hill). |
| 5        | 11          | F         | 14. 11. 81              | 16. 11. 81                | Fever, headache                             | Allegedly treated unsuccessfully at Same Hospital. Died upon his return to Gemai     |
| 6        | 40          | F         | 15. 11. 81              | 18. 11. 81                | Fever, headache                             | Student at Gemai Pr. School<br>Villager at Gemai                                     |
| 7        | 5           | M         | 16. 11. 81              | Recovered (5. 12. 81)     | Inguinal buboes (observed)                  | Son of case No. 6, cured   |
| 8        | 20          | F         | 20. 11. 81              | Recovered (5. 12. 81)     | Fever, headache, axillary buboes (observed) | Wife of case No. 4. <i>Y. pestis</i> bacilli observed in bubo aspirates, cured       |
| 9        | 5           | F         | 28. 11. 81              | Recovered (11. 12. 81)    | Fever, headache, inguinal/axillary buboes   | <i>Y. pestis</i> bacilli found in bubo aspirates                                     |

\* Extracted from the Tanga Regional Health Officer's notes of December 1981

| Case No. | Age (Years) | Sex (M/F) | Date of onset | Date of death or recovery | Reported main signs and/or symptoms | Remarks/Village   |
|----------|-------------|-----------|---------------|---------------------------|-------------------------------------|---|
| 1        | 13          | F         | 24. 4. 82     | 27. 4. 82                 | Fever, headache, inguinal buboes    | Mbughui, Viti, visited Manka near Gemai before she became sick        |
| 2        | 12          | F         | 26. 4. 82     | 2. 5. 82                  | Headache, cervical buboes           | Gundi, Gemai  |
| 3        | 13          | M         | 1. 5. 82      | 3. 5. 82                  | Cervical buboes                     | Gundi, Gemai, first case reported to health authorities               |
| 4        | 19          | F         | 2. 5. 82      | 4. 5. 82                  | Fever, headache, inguinal buboes    | Gundi, Gemai  |
| 5        | 32          | F         | 6. 5. 82      | Recovered (12. 5. 82)     | Fever, headache, inguinal buboes    | Langoni, Gemai  |
| 6        | 28          | F         | 7. 5. 82      | Recovered (12. 5. 82)     | Fever, headache, inguinal buboes    | Gundi, Gemai, <i>Y. pestis</i> bacilli seen in aspirates              |
| 7        | 30          | F         | 7. 5. 82      | Recovered (12. 5. 82)     | Fever, headache, pneumonia          | Langoni, Gemai  |
| 8        | 16          | M         | 10. 5. 82     | 12. 5. 82                 | Fever, headache, inguinal buboes    | Mbughui, Viti, same household as case No. 7                           |
| 9        | 10          | M         | 10. 5. 82     | Recovered                 | Buboes                              | Mbughui, Viti, bipolar bacilli in bubo aspirates                      |
| 10       | 6           | F         | 17. 5. 82     | Recovered                 | Buboes                              | Lokome, Shume-Nywelo, aspirates negative                              |
| 11       | 46          | F         | ?             | Recovered                 | Buboes                              | Lokome, Shume-Nywelo, aspirates positive for <i>Y. pestis</i> bacilli |
| 12       | 4           | M         | ?             | Recovered                 | Buboes                              | Lokome, Shume-Nywelo  |
| 13       | 9           | F         | ?             | Recovered                 | Buboes                              | Lokome, Shume-Nywelo  |
| 14       | 10          | F         | ?             | Recovered                 | Pneumonia                           | Gemui   |
| 15       | 64          | M         | 25. 5. 82     | Recovered                 | Buboes                              | Gundi, Gemai  |
| 16       | 20          | F         | 25. 5. 82     | Recovered                 | Buboes                              | Kwamshinga, Gemai   |
| 17       | ?           | F         | ?             | Recovered                 | Buboes                              | Gemui   |
| 18       | 30          | M         | 29. 5. 82     | Recovered                 | Buboes                              | Gundi, Gemai  |

one year, the results also indicate that the infections occurred locally. Furthermore, the occurrence of human plague outbreaks in the area in October 1981 and in April 1982 and the presence of bipolar-staining bacilli in bubo aspirates of one patient in November 1981 suggest that the disease persisted endemically since the 1980 outbreak and hence an active plague focus probably exists in the area. Maintenance of this focus can be attributed to the presence of *R. rattus*, *P. natalensis* and *A. niloticus*, the rodent species already incriminated as suitable reservoirs of plague in Tanzania (Msangi, 1968; Kilonzo and Mtoi, 1982; Kilonzo and Mhina, 1982). Similarly, *X. brasiliensis* and *D. lypusus*, the most efficient flea vectors of plague in the country (Msangi, 1968; Kilonzo and Mtoi, 1982) were partly responsible for the maintenance and transmission of the disease in the area.

The long persistence of plague outbreaks in this area (Tables 3 and 4) is probably associated with the general belief among villagers that the disease is caused by witchcraft and evil spirits and their consequent reluctance to report outbreaks promptly. Indeed most villagers were uncooperative during the surveys, especially in November 1980 and June 1981. Health education for villagers and their leaders is therefore necessary and a plague surveillance service is needed for the area so that outbreaks can be foreseen and counter-measures taken promptly. Furthermore, regular seminars for health and medical personnel in the district should be held in order to acquaint them with various diagnostic and control methods.

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