

Epidemiological and Ecological Studies of Scrub Typhus in Royal Thai Army Field Training Facilities

Principal Investigators : David E. Johnson, MAJ, MC
Bruce A. Harrison, MAJ, MSC
Panita Lakshana Tanskul, M.Sc.
John W. Crum, MAJ, MSC
Paibul Buspathumrong, COL, MC, RTA
Vichai Sangkasuwan, COL, MC, RTA
Alexander L. Dohany, MAJ, MSC

Associate Investigators : Richard G. Andre, MAJ, MSC
Michael C. Callahan, SSG
Suvath Hanchalay, B.A.
Inkam Inlao
Robert S. Kennedy, SFC
Illa Muul, MAJ, MSC
Vichit Phunkitcha
Chantana Ratanawaraha, B.Sc.
Chirapa Saengruchi, 1st LT, RTA
Withoon Thiemanun

OBJECTIVES :

1. To prospectively determine the susceptibility and the exposure risk of Royal Thai Army personnel to *Rickettsia tsutsugamushi* during field training exercises;
2. To determine the prevalence of *Rickettsia tsutsugamushi* in selected species of small mammals and chiggers from areas and habitats utilized by troops during training;
3. To determine if there is a seasonal effect influencing the susceptibility risk of Royal Thai Army personnel;
4. To evaluate the use of regional, habitat and seasonal data for predicting human exposure and risk potential to scrub typhus in Thailand.

BACKGROUND : Outbreaks of scrub typhus in the Royal Thai Army have been isolated epidemic events. On the basis of hospitalized cases, personnel at greatest risk appear to be those undergoing primary field training in the central or Korat plateau areas or those in security forces deployed in the field (Sangkasuwan-personal communication). Sangkasuwan, *et al.* (1) found *R. tsutsugamushi* in every area sampled in Thailand and also concluded that there was seasonality in the frequency of rickettsial isolations from chiggers and mammals. A similar seasonal pattern has been observed in human cases in Thailand (2).

* USAMRU-Kuala Lumpur, Institute for Medical Research, Malaysia.

The ecological complexity of Thailand offers unique opportunities for comparative studies to assess the influence of various ecological factors on the presence, prevalence and risk of *R. tsutsugamushi* to man. Recent development of a serological test for *R. tsutsugamushi* (3), more reliable than the standard Weil-Felix test, now provides a good basis for field studies of scrub typhus in Thailand.

METHODS : As a pilot project, class 1-4, 138 soldiers undergoing Special Forces training at Pak Chong, Nakhon Ratchasima province, contributed blood samples before and after their training. Training consisted of four weeks of mixed classroom and field problems and four days of jungle bivouac approximately 15-20 km. from the base camp. The troop training includes daily contact with forested or grassy areas and a bivouac in similar areas. Total field exposure was calculated for each individual and background information on age, rank, occupation (both in and outside of the military), home, travel and previous medical history was taken. The latter of the two blood samples was collected two weeks after the end of training. The blood samples were tested by the Weil-Felix (4) and by the indirect immunofluorescence tests (3).

Rodents and other small mammals were live-trapped in each habitat associated with troop training. Fifty to 100 traps were placed in 2-3 habitats each night. All small mammals collected were identified to species, sexed, aged, bled via cardiac or retro-orbital puncture and examined for ectoparasites. Rodent rickettsial identification will be done at AFRIMS by the IFA technique (3). Attached chiggers were gently scraped off and placed in vials containing 70% ETOH. Chiggers were counted and collated according to host species and habitat. Unengorged chiggers were collected in the troop training areas by black plates. Chiggers collected from black plates were kept alive in vials of water and sent to USAMRU-Kuala Lumpur, for rickettsia isolation by the micro direct fluorescent antibody (MDFA) technique (5).

RESULTS : The initial part of the study involved a thorough reconnaissance of the two study areas. The terrain surrounding the base camp is government owned and is hilly and primarily covered with secondary evergreen forests, although disturbed areas with early regenerating vegetation (woody plants, grass) are also found in several of the training areas. The government owned property is completely surrounded by privately owned rolling hills which have been cleared of forest and are planted in agricultural crops, primarily maize. Several training areas utilized by the troops are on the edge of the government property and adjacent to the maize fields. The soil in the base camp training areas is primarily red-orange clays. The forests in the area are fairly well preserved with considerable humus and leaf litter, indicating that fires are uncommon.

The troop bivouac areas are located on non-government property in a valley at the foot of forested mountains. These sites consist primarily of grass (lalang) and isolated patches of regenerating and secondary evergreen forest. The bivouac areas are usually located in between or next to private agricultural property. The civilians in the area annually burn the maize stalks and adjacent grass and forest areas. Consequently, there is little humus and leaf litter present. The soil type in the bivouac areas is a sandy loam which dries very quickly.

The first entomological-rodent trapping studies occurred during the period 18 June - 26 July 1978, and were conducted during two separate 10 day periods. The first period, 18-27 June, corresponded with the first 10 days of troop training around the base camp. The second period, 19-26 July, corresponded with the troop bivouac period in a small village area, Ban Wang See Sod, about 15-20 km from the base camp.

During the first entomological study period three basic habitats in nine training areas were trapped for small mammals, i.e., early regenerating grass, early regenerating evergreen and secondary evergreen. A total of 132 small mammals of eight species were trapped in these habitats over the 10 day period (Table 1). *Rattus rattus* was the most commonly trapped mammal. A total of 21 collections (black plate) were made for unengorged chiggers. These collections resulted in 168 chiggers which were sent to USAMRU-Kuala Lumpur for rickettsia isolations. Of the 168, 7.2% of the specimens were found positive for *Rickettsia tsutsugamushi* (Table 3). The infected specimens included nine *Leptotrombidium (L) deliense* and three *Leptotrombidium (L) miculum arvinum*. During the second entomological study period three basic habitats in and adjacent to two bivouac areas were trapped, i.e. early regenerating grass (lalang), secondary evergreen and banana orchard. A total of 100 small mammals of five species were captured in these habitats over the 10 day period (Table 2). Again, *R. rattus* was by far the most commonly trapped mammal. A total of 14 black plate collections were made in the areas for unengorged chiggers, resulting in only 20 specimens. These specimens were sent to USAMRU-Kuala Lumpur and all were found negative for *R. tsutsugamushi* (Table 3). The low chigger densities on the black plates in this study area were probably due to the low leaf litter-humus content on/in the sandy-porous soil. Although rain fell nearly every day and chiggers were very abundant on the trapped mammals, the disturbed nature of the soil and vegetation was apparently not conducive to high chigger densities in the absence of the mammals or their burrows.

Of 138 soldiers, none showed Weil-Felix titers suggestive of recent infection (≥ 160) in the pre-training serum sample. No specimens had an IFA titer of 1:50 or greater. Post-training specimens demonstrated a four fold or greater titer rise in six soldiers by the Weil-Felix examination (all were titer rises from $< 1:20$ to 1:40), but all soldiers were negative by the IFA technique.

The rodent sera from both study areas remain to be examined for *R. tsutsugamushi* antibody levels. Engorged chiggers taken from the mammals captured during both study periods are being mounted on slides for later identification.

Table 1. Small mammals collected, by habitat, in the Pak Chong Special Forces Camp Training Areas, 18-27 June 1978

| Habitat | Trap Night | Mammals | | | | | | | | | | Total | |
|------------------------------|---------------|------------------------------|------------------------------|-------------------------------|-------------------------|--------------------------|----------------------------|---------------------------|------------------------|-----|-----|-------|-----|
| | | <i>Bandicota saviler</i> | <i>Meretes berdnorei</i> | <i>Rattus bukit bukit</i> | <i>Rattus losea</i> | <i>Rattus rattus</i> | <i>Rattus sabarnus</i> | <i>Rattus sumifer</i> | <i>Inpaya glis</i> | | | | |
| Early Regenerating Grass | 760 | 1 | - | 2 | 4 | 63 | 1 | 4 | 4 | 4 | 4 | 4 | 79 |
| Early Regenerating Evergreen | 216 | - | - | - | - | 5 | - | 7 | - | - | - | - | 12 |
| Secondary Evergreen | 858 | - | 2 | 3 | - | 11 | 2 | 7 | 16 | 7 | 16 | 41 | 41 |
| Total | 1,834 | 1 | 2 | 5 | 4 | 79 | 3 | 18 | 20 | 132 | 132 | 132 | 132 |

Table 2. Small mammals collected, by habitat, in the Royal Thai Army Bivouac Area in Ban Wang See Sod, 17-26 July 1978

| Habitat | Trap Nights | Mammals | | | | | Total |
|-----------------------------------|----------------|-----------------------------|---------------------------|-------------------------------|--------------------------|-------------------------|-------|
| | | <i>Bandicota indica</i> | <i>Mus cervicolor</i> | <i>Rattus bukit bukit</i> | <i>Rattus rattus</i> | <i>Taparia glis</i> | |
| Early Regenerating Grass (Lalang) | 792 | 1 | 6 | 1 | 73 | 1 | 82 |
| Secondary Evergreen | 329 | - | - | - | 10 | 6 | 16 |
| Banana Orchard | 44 | - | - | - | 1 | 1 | 2 |
| Total | 1,165 | 1 | 6 | 1 | 84 | 8 | 100 |

Table 3. Chiggers and *Rickettsia tsutsugamushi* isolations from Pak Chong, Royal Thai Army Training Center

| Chigger species | Pak Chong | | | | | | Total | |
|---------------------------------|-----------------|-------------------|------------------|-------------------|-----------------|-------------------|-------|-------|
| | S.F. Camp | | Ban Wang See Sod | | Total | | | |
| | Number screened | Chiggers Infected | Number screened | Chiggers Infected | Number screened | Chiggers Infected | | |
| | No. | % | No. | % | No. | % | | |
| <i>L. (L.) deliense</i> | 144 | 9 | 6.25 | 18 | - | 162 | 9 | 5.56 |
| <i>L. (L.) miculium curvium</i> | 23 | 3 | 13.04 | - | - | 23 | 3 | 13.04 |
| <i>L. (L.) species C</i> | - | - | - | 1 | - | 1 | - | - |
| <i>L. (Trom.) paniculatum</i> | 1 | - | - | - | - | 1 | - | - |
| <i>Ascho. (Lau.) indica</i> | - | - | - | 1 | - | 1 | - | - |
| Total | 168 | 12 | 7.14 | 20 | - | 188 | 12 | 6.38 |

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