

Ectoparasite and *Rickettsia tsutsugamushi*
Studies in Thailand

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OBJECTIVE : To establish and describe the chiggers and ticks that are vectors or potential vectors of human pathogens in Thailand, and to determine the geographical distribution of *Rickettsia tsutsugamushi* in natural populations of chiggers in Thailand.

BACKGROUND : This is a continuing project that began in the early 1960's. Emphasis during the early years was placed on establishing rodent-ectoparasite associations and collecting, classifying and determining the distribution of chiggers and ticks in Thailand. This early work served as the basis for a number of publications, of which Lakshana (1) and Lekagul and McNeely (2) have established a very firm taxonomic base on which epidemiological studies on *Rickettsia tsutsugamushi* in Thailand can proceed. More recent emphasis has now shifted to the distribution of strains of *R. tsutsugamushi* that occur in vector chiggers in Thailand. However problems still exist in identifying chigger specimens collected in Thailand, hence taxonomic studies on chiggers are continuing.

METHODS : Ectoparasites are collected from live trapped rodents and other small mammals by removal with forceps, by scraping or by holding the animals alive over a pan of water and allowing engorged ectoparasites to drop into the water. Engorged chiggers are normally preserved in alcohol and mounted on slides for study. Chiggers used for *R. tsutsugamushi* isolation attempts are preferably unengorged. Unengorged chiggers are usually found in leaf litter, on rotten logs and other favorable habitats frequented by rodents and other small mammals, and are easily collected by using 5" x 5" formica black plates. Collected unengorged chiggers are placed and kept alive in vials of water, which are then shipped to USAMRU-Kuala Lumpur for rickettsia isolation. A technique using direct immuno-fluorescence has recently been developed to detect rickettsia in naturally infected mites (3). Using this technique, the internal contents of each unengorged chigger can be screened for nine different strains of *R. tsutsugamushi*, i.e., Karp, Gilliam, Kato TC 586, TA 678, TA 686, TA 716, TA 763 and TH 1817. After the internal contents of the chigger has been tested for rickettsia, its exoskeleton is mounted in Hoyer's mounting media on a slide for identification.

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RESULTS : Between June 1977 - August 1978, 2,251 unengorged chiggers collected in Thailand by the black plate method, were identified and sent to USAMRU - Kuala Lumpur for rickettsia isolation (Table 1). These specimens were collected in the following provinces : Chiang Mai, Kanchanaburi, Nakhon Ratchasima, Prachin Buri, Surin and Ubon Ratchathani. A total of 13 species of chiggers in 5 genera were involved, with *Leptotrombidium* (L.) *deliense*, the primary *R. tsutsugamushi* vector in Thailand being the most abundant species. The results of the rickettsia isolation attempts are shown in Table 2. Slightly over 10% of the chiggers screened for *R. tsutsugamushi* were positive. The chigger species found positive for *R. tsutsugamushi* are listed in Table 3. *Leptotrombidium* (L.) *deliense*, the most commonly collected species, also yielded the most rickettsia isolations. The other species positive for rickettsia were : *Leptotrombidium* (L.) *miculum*, *arvinum*, *Microtrombicula chamlongi* and two new undescribed species collected in Chiang Mai, *Leptotrombidium* (L.) species A and species B. Data on *R. tsutsugamushi* strains found in Thailand will be described in the Annual Report from USAMRU - Kuala Lumpur.

In December 1977 a total of 343 *Leptotrombidium* (L.) *deliense*, 251 *L. (L.) scutellare* and 85 *L. (L.) striatum* were collected engorged from field rodents and shipped alive to the Walter Reed Army Institute of Research. These specimens were for ongoing research requiring progeny broods.

Reports of twelve new species of *Leptotrombidium* collected in Thailand are currently being prepared for publication. One of the new species is very similar to *L. (L.) deliense*, the primary vector of *R. tsutsugamushi* in Thailand. An additional 4 new species of *Leptotrombidium* and one new species of *Gahrliopia* were collected in the above black plate collections. A checklist of the ticks of Thailand is currently in manuscript form.

REFERENCES :

1. Lakshana, P. 1973. A checklist of the trombiculid mites of Thailand (Prostigmata : Trombiculidae). U.S. Army Med. Comp. - SEATO, Bangkok, Thailand. 44 p.
2. Lekagul, B. and J.A. McNeely. 1977. Mammals of Thailand. Assoc. Conser, Wildlife, Bangkok. 758 p.
3. Dohany, A.L., A. Shirai, D.M. Robinson, S. Ram and D.L. Huxsoll. Identification of *Rickettsia tsutsugamushi* in naturally infected chiggers (Acarina : Trombiculidae) by direct immunofluorescence. (in preparation).

Table 1. Unengorged chiggers collected in Thailand between June 1977 - September 1978, and sent to USAMRU - Kuala Lumpur for Rickettsia isolations.

Chigger Species	Locations						Total
	Chiang Mai	Kanchanaburi	Nakhon Ratchasima	Prachin Buri	Surin	Ubon Ratchathani	
No. of Collections	8	9	12	3	4	9	45
<i>Gahrlipeia (G.)</i> species A*	-	-	-	-	2	-	2
<i>Lept. (L.) deliense</i>	13	90	543	1	145	949	1,741
" " <i>fulleri</i>	-	-	21	-	-	-	21
" " <i>miculum arvinum</i>	46	4	8	-	-	-	58
" " species A	119	-	-	-	-	-	119
" " species B	4	-	-	-	-	-	4
" " species C	-	-	-	-	15	-	15
" " species D	1	1	-	-	-	-	2
<i>L. (Trom.) paniculatum</i>	-	1	-	9	-	-	10
<i>Microtrombicula chamlongi</i>	-	-	5	-	-	1	6
<i>Siseca rara</i>	-	-	16	-	-	-	16
<i>Walchiella oudemansi</i>	-	-	68	-	-	-	68
" <i>traubi</i>	-	-	1	-	-	-	1
Total	183	96	662	10	162	950	2,063

* Species with alphabetical designations are new and undescribed.

Table 2. *Rickettsia tsutsugamushi* isolations from Thai chiggers.*

Location	Total Chiggers Examined	Number Infected	Percent Infected
Chiang Mai	133	26	19.55
Nakhon Ratchasima	49	31	63.27
Prachin Buri	1	1	100.00
Ubon Ratchathani	579	26	4.50
Total	762	84	11.02

* The collections from Kanchanaburi, Surin and several collections from Khao Yai National Park have been deleted due to technical problems.

Table 3. Source and species of Thai chiggers infected with *Rickettsia tsutsugamushi*.

Location	Chiggers Species	Number Screened	Chiggers Infected	
			Number	Percent
Chiang Mai	<i>L. (L.) deliense</i>	52	12	23.08
	" " <i>miculum arvinum</i>	37	5	13.51
	" " species A	39	7	17.95
	" " species B	4	2	50.00
	" " species D	1	0	0
Nakhon Ratchasima	<i>L. (L.) deliense</i>	47	30	63.83
	<i>Microtrombicula chamlongi</i>	2	1	50.00
Prachin Buri	<i>L. (L.) deliense</i>	1	1	100.00
Ubon Ratchathani	<i>L. (L.) deliense</i>	579	26	4.49
Total		762	84	11.02