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Prevalence of Antibody to *Orientia tsutsugamushi* in Dogs Along Thai - Myanmar Border

Mongkol Chenchittikul, Decha Pangjai and Paijit Warachit

ABSTRACT

Three hundred and ninety six dog sera collected from ten provinces along Thai-Myanmar border in 1997 were tested for antibody to *Orientia tsutsugamushi* by Indirect Fluorescent Antibody test (IFA). The average percentage of positive antibody (Ab) to scrub typhus in all provinces was 39.65%. The Ab prevalence to scrub typhus in each province ranged from 21.95 to 62.16%, northern provinces had the highest prevalence (54.00%) whereas southern provinces had the lowest prevalence (24.39%). Geometric mean titer (GMT) of the Ab in dog sera were 1:191 (range from 1:100 to 1:594). GMT of the Ab in Chiengrai province was the highest titer (1:594) and Chumphon province was the lowest titer (1:100).

Key words : dog, IFA, *Orientia tsutsugamushi*,prevalence, scrub typhus

INTRODUCTION

Scrub typhus, caused by *Orientia tsutsugamushi* (*Rickettsia tsutsugamushi*), is a febrile disease (Frances *et al.*, 1997), transmitted to man by the bite of an infected, larval mite (chigger) of the family Trombiculidae, order Acarina. Numerous cases were reported from Kwe Noi Valley and Phuket in 1943 (Oaks *et al*., 1983). However, scrub typhus in Thai patient was first confirmed by serology in 1952 (Thainua, 1952). Since then, it has been recognized as endemic in Thailand. Since 1973, every cases had to reported as the surveillance disease to the Division of Epidemiology, Ministry of Public Health. Recently, the reported cases in 1995-1997 were 1,203, 1,210, and 2,064 respectively (Division of Epidemiology, 1995; 1996; 1997).The highest prevalence of scrub typhus was in the north and the lowest prevalence was in the south.

Rodents have been recorded as being natural host of scrub typhus rickettsia (Azad and Beard, 1998). Dogs infected with *O. tsutsugamushi* were found in the endemic area of Vietnam and Peninsular Malaysia (Alexander *et al*., 1972; Huxsoll *et al*. 1977). In Thailand, isolation of *O. tsutsugamushi* from rodents had been reported (Traub *et al*., 1954). Seroprevalence of scrub typhus in human also had been carried out, demonstrated that human was exposed to *O. tsutsugamushi* (Johnson *et al*., 1982), but serosurveys on dogs had not been conducted. This report presents the serosurvey of scrub typhus in dogs along Thai-Myanmar border.

MATERIALS AND METHODS

Sera

Serum samples of 396 Dogs were collected from ten provinces in November 1997 along Thai-Myanmar border, Thailand for an epidemiological

National Institute of Health, Department of Medical Sciences, Ministry of Public Health, Nontaburi 11100, Thailand.

survey of plague (Figure 1). Two villages from each province were selected randomly for collecting the samples. Dogs were privately owned and belong to inhabitants of the respective study areas. Blood samples were collected from cephalic vein and allowed to clot. The serum samples were maintained at -20°C prior to testing.

Serological test

The sera were tested for antibodies specific to the three prototype (Gilliam, Karp and Kato) strains of *O. tsutsugamushi*, *Rickettsia typhi* Wilmington strian and Thai tick typhus TT118 strain. An indirect fluorescent antibody test (IFA) using fluorescein conjugated goat anti-dog IgG (Kirkegaard Perry Laboratories Inc., America) were employed in the IFA. The IFA was performed as described previously (Tamura *et al*., 1984). Serum dilution, beginning at 1:25 were test for screening. Positive sera were further diluted two fold serially, titer ≥1:50 was considered as positive serodiagnosis (Huxsoll *et al*., 1977).

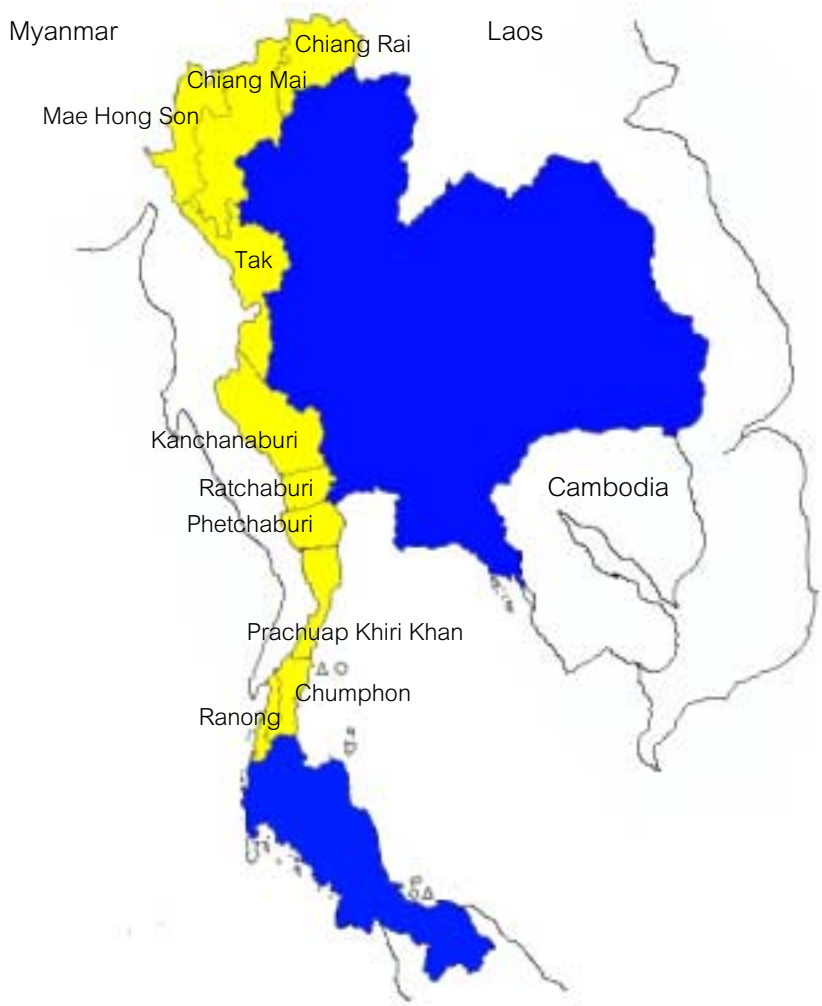


Figure 1 Map of Thailand showing ten provinces along Thai-Myanmar border from which specimens were collected.

RESULTS

Of the 396 dog serum samples, the number of antibody positive against *O. tsutsugamushi* was 157 (39.65%). All sera were negative for *R. typhi* and TT118. The samples collected from 10 provinces ranged from 37-42 cases/province (average 39.6). The prevalence of antibodies to scrub typhus were varied (Table 1) by provinces (range from 21.95 to 62.16%). Chiang Mai had the highest prevalence with 62.16% where as Prachuap Khiri Khan and Chumphon had the lowest with 21.95%. Geometric mean titer (GMT) of the Ab in dog sera were 1:191 (range from 1:100 to 1:594). GMT of the Ab in Chiengrai province was the highest titer (1:594) and Chumphon province was the lowest titer (1:100).

Table 2 showed the prevalence of antibodies to scrub typhus in each village (range from 4.76 to 84.21). The highest prevalence of antibodies was 84.21% at Muang Na village, Chiang Mai where as Klong Loi village in Prachuap Khiri Khan showed the lowest with a prevalence of 4.76%. For GMT, the highest titer was 1:635 at Rong village, Chiang Rai. The lowest titer was 1:71 at Wang Ta Kian village, Tak.

Table 3 showed the prevalence of antibodies against scrub typhus rickettsia in dog in three different geographical parts of Thailand. Northern provinces were the highest, with a prevalence of 54.00% and southern provinces were the lowest, with a prevalence of 24.39%. GMT of the Ab in dog sera in three parts were highly significant difference ($p<0.01$). However, it was not different between central and northern parts ($p>0.05$). The GMT in northern part (1:220) was highly significant difference ($p<0.01$) from southern part (1:111). But GMT in central part (1:190) was only significant difference from southern part ($p<0.05$).

From the Annual Epidemiological report, in 1997 the reported cases in the studied provinces (Northern : Chiang Mai, Chiang Rai, Tak, Mae Hong Son, Central : Phetchaburi, Ratchaburi, Kanchanaburi, Prachuap Khiri Khan, Southern : Chumphon, Ranong) are 56,404,166,23,1,11,2,5,1 and 0 respectively (Division of Epidemiology, 1997). The sum of reported cases in three parts of Thailand were presented in Figure 2. This figure also showed the GMT of dog antibodies in each part.

DISCUSSION

The prevalence of antibodies to scrub typhus in dogs along Thai-Myanmar border, Thailand were 39.65 %. The data presenting here have demonstrated that natural infections of scrub typhus occur in dogs in every province along Thai-Myanmar border, Thailand. All villages in this study were rural areas. This correlated to the former study by Johnson *et al.* (1982) which showed 77% of seroprevalence in rural Thai village. And also, *O. tsutsugamushi* could be isolated from patients and rodents in rural areas (Trishnananda *et al*., 1964).

The prevalence of scrub typhus antibodies in three parts of Thailand was correlated well with the number of reported patient cases. Northern part was the high risk areas than central and southern parts, respectively.

The prevalence of antibodies to scrub typhus were not only varied by province but also by village. The highest prevalence of antibodies was 84.21% at Muang Na village, Chiang Mai where as Klong Loi village in Prachuap Khiri Khan showed the lowest with a prevalence of 4.76% . For GMT, the highest was 1:635 at Rong village, Chiang Rai, but the lowest was 1:71 at Wang Ta Kian village, Tak.

The present study correlated well with the previous studies. Alexander *et al*. (1972) showed 45% of antibodies against *O. tsutsugamushi* in

Table 1 The prevalence of antibodies to scrub typhus in dogs in 10 provinces.

Province	Number examined	Antibodies titer									GMT[a)]	% Positive
		<1:25	1:25	1:50	1:100	1:200	1:400	1:800	1:1600	1:3200		
Chiang Rai	37	15	1	0	1	2	7	7	3	1	594	21/37 (56.76)
Chiang Mai	37	14	0	1	5	7	6	2	0	2	279	23/37 (62.16)
Mae Hong Son	37	8	7	7	9	3	3	0	0	0	107	22/37 (59.46)
Tak	39	24	0	6	2	6	1	0	0	0	110	15/39 (38.46)
Ratchaburi	41	25	1	1	4	1	3	4	1	1	348	15/41 (36.59)
Phetchaburi	42	28	2	2	1	6	2	1	0	0	189	12/42 (28.57)
Prachuap Khiri Khan	41	32	0	1	4	2	1	1	0	0	159	9/41 (21.95)
Kanchanaburi	40	17	3	7	4	6	1	1	1	0	132	20/40 (50.00)
Chumphon	41	25	7	2	3	3	1	0	0	0	126	9/41 (21.95)
Ranong	41	22	8	4	3	4	0	0	0	0	100	11/41 (26.83)
Total	396	210	29	31	36	40	25	16	5	4	191	157/396 (39.65)

[a)] GMT, Geometric Mean titer

Table 2 The prevalence of antibodies to scrub typhus in dogs in 20 villages.

Provinces	Villages	No. examined	No. positive (%)	GMT[a)]
Chiang Rai	Viang Hom	20	12 (60.00)	566
	Rong	17	9 (52.94)	635
Chiang Mai	Aruno Tai	18	7 (38.89)	135
	Muang Na	19	16 (84.21)	383
Mae Hong Son	Mae Sa Riang	20	12 (60.00)	100
	Mae Ja Ton	17	10 (58.82)	115
Tak	Rim Mei	19	7 (36.84)	181
	Wang Ta Kian	20	8 (40.00)	71
Ratchaburi	Bo Moo	20	7 (35.00)	400
	Tago Lang	21	8 (38.09)	308
Phetchaburi	Teng Nuea	21	4 (19.05)	200
	Suan Yai Pattana	21	8 (38.10)	183
Prachuap Khiri Khan	Dan Sing Khon	20	8 (40.00)	168
	Klong Loi	21	1 (4.76)	100
Kanchanaburi	Tai Mhuang	20	9 (45.00)	147
	Pra Chum Mai	20	11 (55.00)	121
Chumphon	Ran Tad Phom	19	3 (15.79)	79
	Santi Nimitr	22	6 (27.27)	159
Ranong	Nai Krung	20	5 (25.00)	115
	Had Tun	21	6 (28.57)	89
Total		396	157 (39.65)	191

[a)] GMT, Geometric Mean titer

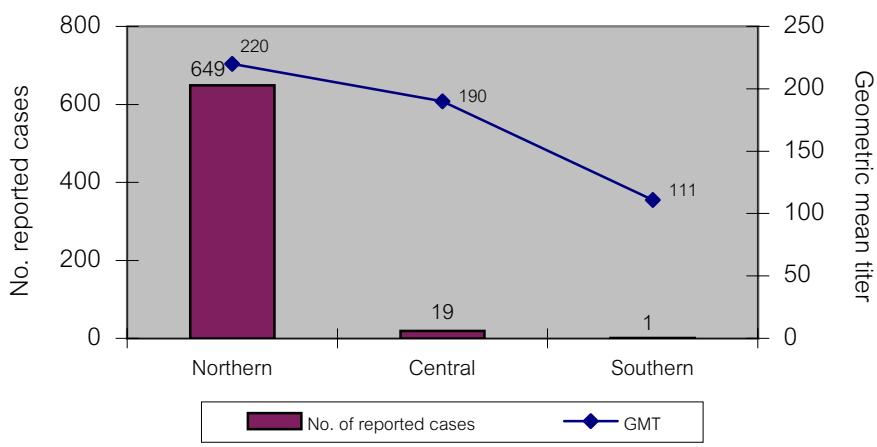


Figure 2 Number of reported cases and geometric mean titer of dogs in three studied parts.

Table 3 The prevalence of antibodies to scrub typhus in dogs in 3 parts.

Part	Number examined	Antibody titer %								Possitive	GMT[a)]	
		<1:25	1:25	1:50	1:100	1:200	1:400	1:800	1:1600	1:3200			
North	150	61	8	14	17	18	17	9	3	3	81/150 (54.00)	220	
Central	164	102	6	11	13	15	7	7	2	1	56/164 (34.15)	190	$P<0.01$
South	82	47	15	6	6	7	1	0	0	0	20/82 (24.39)	111	$P<0.05$
Total	396	210	29	31	36	40	25	16	5	4	157/396 (39.65)		

[a)] GMT, Geometric Mean titer

military scout and tracker dogs worked in rural areas in Vietnam. Huxsoll *et al*. (1977) also showed the seroprevalence antibodies in dog in Malaysian rural areas.

Dogs usually live in the same habitat as human and are frequently exposed to the same disease organisms. But their movements in the scrub areas are more frequently than man. So, they are more risk of infection by scrub typhus rickettsia than man. It is recommended to use dogs as sentinels for scrub typhus and also in determining areas of potential risk to man.

Antibodies against *R. typhi* were not detected in this study. This supported that murine typhus was a disease of urban areas where man and rats shared the same habitat in Southeast Asia but not in rural areas (Brown *et al*., 1988; Sankasuwan *et al*., 1969). Okabayashi *et al*. (1996) also showed that wild rats in rural areas of Thailand were not found antibodies against *R. typhi*. However, it was contrast to 38% of sero-prevalence to *R. typhi* in dogs in rural areas in Malaysia and 0.4% of prevalence in the Suez Canal area in Egypt (Huxsoll *et al*., 1977; Soliman *et al*., 1989).

Antibodies against TT118 were also not found in this study. This disease may be more prevalence in the areas near the forest. Two of three reported cases were working and living near the forest areas (Sirisanthana *et al*., 1994). However, Okabayashi *et al*., (1996) showed a 64.71% prevalence of antibody to Thai tick typhus (TT118) in wild rats in rural areas of Thailand.

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