Seroprevalence of Rickettsial Infection in Commensal Rodents and Shrews Trapped in the Bangkok Metropolitan Area

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Abstract

Murine typhus and scrub typhus are important human rickettsial diseases in Thailand. Small mammals, including many species of rodents and shrews, serve as the reservoir host of rickettsial diseases. *Rickettsia typhi* can be transmitted to humans by fleas causing murine typhus, while infection with *Orientia tsutsugamushi* causing scrub typhus in humans is transmitted by chiggers. The prevalence of rickettsial infection depends on the geographic area. The seroprevalence of antibody to *R. typhi* and *O. tsutsugamushi* was studied in commensal rodents and shrews trapped in markets in the Bangkok Metropolitan Area (BMA). *R. typhi* and *O. tsutsugamushi* antigen prepared in the yolk sac of embryonated eggs were used to determine the specific antibody in trapped animals' sera by using fluorescein isothiocyanate (FITC)-anti rat immunoglobulins as a second antibody. Antibody to *R. typhi* was found in 25 (5%) of 500 sera tested and no antibody to *O. tsutsugamushi* was detected. *R. typhi* antibody titer ranged from 40-1280 and was found in *Rattus norvegicus* (4.2%), *Rattus rattus* (0.4%), *Rattus exulans* (0.2%), and *Mus musculus* (0.2%) trapped in 8 of 47 markets in the BMA. *R. typhi* antibody was commonly found in *R. norvegicus*. The authors concluded that murine typhus is an important rickettsial disease and *R. norvegicus* is an important reservoir species of rodents found in markets of the BMA.

Key word : Murine typhus, Scrub typhus, *Orientia tsutsugamushi, Rickettsia typhi, Rattus norvegicus,* Bangkok Metropolitan Area

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Human rickettsial diseases are caused by the obligatory intracellular bacteria of the family Rickettsiaceae. Small mammals, including many species of rodents and shrews, serve as the reservoir hosts of rickettsiae, and the organisms are transmitted to humans by arthropod vectors, i.e., mites, ticks, fleas and lice. Murine typhus, scrub typhus, tick borne typhus, Q-fever and human ehrlichioses are human rickettsial diseases found in Thailand[1-4]. The most common rickettsial diseases reported in Thailand are murine typhus and scrub typhus. Murine typhus in humans, caused by *Rickettsia typhi*, is transmitted from reservoir hosts to humans by fleas. Commensal rodents and shrews are animals which live close to man and they are the reservoir hosts of *R. typhi*. The seroprevalence of *R. typhi* infection in human residents in the area where the animals were trapped was found to be 0-20 per cent[2]. Scrub typhus, caused by *Orientia tsutsugamushi*, is transmitted to humans by chigger (mite larvae) bites. Chiggers are reservoirs of *O. tsutsugamushi* and also act as vectors while rodents and shrews are accidental hosts which can transmit the disease to humans[5]. High prevalences of antibody to *O. tsutsugamushi* were found in 59 per cent of healthy Thais in the northern area of Thailand and 77 per cent of healthy Thais in the northeastern area[6,7]. Since rickettsial diseases can be transmitted to humans from infected commensal rodents and shrews and the prevalence of rickettsial infection in humans and animals depends on the geographic area. The authors aimed to investigate the seroprevalence of rickettsial infections caused by *R. typhi* and *O. tsutsugamushi* in commensal rodents and shrews trapped in markets in the BMA. The seroprevalence of rickettsial infection in these rodents and shrews may indicate the risk of rickettsial infection for people in the BMA who live close to reservoir hosts.

MATERIAL AND METHOD

Five hundred animals in 47 fresh food markets in districts of the BMA were trapped from June, 1999 to May,2000. Based on the density of the population and location, the districts in the BMA were grouped into three regions, i.e., the inner, middle and outer region. The inner region was the most crowded area. Animal species were identified. Serum was taken from the animal' s heart and kept at -20°C until the *O. tsutsugamushi* and *R. typhi* antibody tests were performed. *R. typhi* and three strains of *O. tsutsugamushi*, i.e., Karp, Gillium and Kato were grown in chick embryonated egg yolk sacs. Antigen was prepared by harvesting the organisms from the yolk sacs, suspending homogeneously in phosphate buffered saline (PBS) and dropping onto 12-well glass slides. The animal sera were diluted to 1 : 20 in PBS to test for antibody to *R. typhi* and *O. tsutsugamushi*. *R. typhi* and *O. tsutsugamushi* antibody was detected by an indirect immunofluorescence test using FITC-rabbit anti-rat immunoglobulins (Dako, Copenhagen, Denmark). Twofold dilution of positive antibody sera was performed in order to determine the antibody titer.

RESULTS

Samples of 458 tested specimens were obtained from *Rattus norvegicus*, 28 from *Rattus rattus*, 5 from *Rattus exulans*, 5 from *Suncus murinu*, and 4 from *Mus musculus*. Antibody to *R. typhi* was found in 25 (5%) of 500 specimens tested and no antibody to *O. tsutsugamushi* was detected. The frequencies of *R. typhi* antibody positive sera detected from each species of rodent and shrew are shown in Table 1. Most of the positive *R. typhi* antibody samples were found in the sera of *R. norvegicus* (4.2%) followed by *R. rattus* (0.4%) and *R. exulans* (0.2%) or *M. musculus* (0.2%). The distribution of *R. typhi* antibody titers in the 25 positive specimens is demonstrated in Fig. 1. The median titer was 320. Three high *R. typhi* antibody titers (1280) were obtained from sera from *R. norvegicus, R. rattus and R. exulans* respectively, which were trapped in a market in Phra Nakorn district. The distribution of infected animals from each market is shown in Table 2.

DISCUSSION

Many species of rodents and shrews serve as reservoirs or accidental hosts for rickettsiae. Murine

Table 1. *R. typhi* antibody in the 500 sera of rodent and shrew species trapped in markets in the BMA.

Animal species	No. of positive *R. typhi* antibody samples	%
Rattus norvegicus	21	4.2
Rattus rattus	2	0.4
Rattus exulans	1	0.2
Mus musculus	1	0.2
Suncus murinus	0	0
Total	25	5

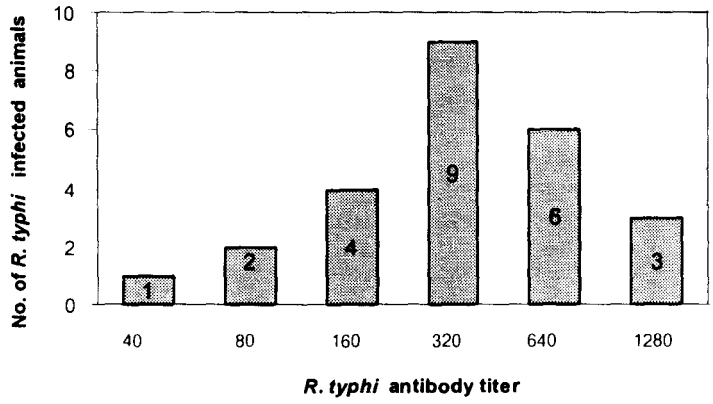

Fig. 1. Distribution of antibody titers to *R. typhi* in animal sera. The antibody titers ranged from 40-1,280.

Table 2. *R. typhi* antibody (Ab) in sera of animals trapped in markets in the districts of the Bangkok Metropolitan Area.

Districts	BMA region	No. of sera tested	No. of positive *R. typhi* Ab sera	%	Animal species (No. of positive *R. typhi* Ab sera)
Phra Nakhon	Inner	25	15	60	*R. norvegicus* (11), *R. rattus* (2), *R. exulans* (1), *M. musculus* (1)
Bang Rak	Inner	8	2	25	*R. norvegicus* (2)
Yan Nawa	Inner	9	2	23	*R. norvegicus* (2)
Min Buri	Outer	17	2	12	*R. norvegicus* (2)
Din Daeng	Inner	10	1	10	*R. norvegicus* (1)
Lak Si	Middle	10	1	10	*R. norvegicus* (1)
Nong Chok	Outer	11	1	9	*R. norvegicus* (1)
Bangkok Noi	Inner	22	1	5	*R. norvegicus* (1)
Other	Inner, middle, outer	388	0	0	
Total	Inner, middle, outer	500	25	5	*R. norvegicus* (21), *R. rattus* (2), *R. exulans* (1), *M. musculus* (1)

typhus is a more important rickettsial disease than scrub typhus in the BMA because the antibody to *R. typhi* was more commonly found in rodents and shrews than the antibody to *O. tsutsugamushi*. The prevalence of the antibody to *R. typhi* in commensal rats and shrews trapped in markets in the BMA showed that *R. norvegicus* is an important rodent species which serves as a reservoir host to transmit *R. typhi* to humans. *R. typhi* antibody was detected in animals trapped in 8 markets. A high prevalence of *R. typhi* antibody was found in the markets in Phra Nakorn district of the BMA which had a large number of animals trapped and a large human population. Murine typhus in humans usually occurs in association with commensal rats, the *Rattus* spp rats are the primary reservoir and the oriental rat flea *(Xenopsylla cheopis)* is the vector. Rat fleas become infected when feeding on a rickettsaemic rat. *R. typhi* proliferates in the midgut cells of the flea and is excreted in the feces. Humans become infected when a flea bites and the *R. typhi* in its feces contaminates a bitten wound. The disease is an occupational harzard for those working in rat-infected areas such as markets or ports[8]. In Thailand, *R typhi* was first isolated from rats in 1964

(9). *R. typhi was isolated* from 3 per cent (35/1,215) of *R. exulans* which was the most common species of domestic rat trapped in the north and northeast part of Thailand, but no *R. typhi* has been isolated from animals trapped in the central provinces of Thailand. In Korat, a province in the northeastern part of Thailand, *R. typhi* has been isolated in 9.1 per cent of house rats trapped in areas where patients with rickettsial disease were living(10). The most important reservoir species of murine typhus is *R. norvegicus* which was the most frequently trapped rodent species in markets in the BMA. In a survey of rodents in Egypt, antibody to *R. typhi* was detected in 25 per cent of *R. norvegicus* and 11 per cent of *R. rattus*(11). A high seroprevalence has also been reported in rodents trapped in the harbor area of the provincial capital, Jayapura, in Indonesia(12). Recent infection with *R. typhi* in rodents could not be identified since the authors did not perform a test for IgM antibody. However, it was observed that the most frequent antibody titer in infected rodents was 320. A high antibody titer (1,280) might represent recent infection which was detected in 0.6 per cent (3/500) of trapped animals. Three high antibody titers were detected from the serum of *R. norvegicus*, *R. rattus*, and *R. exulans* which were trapped from a market in Phra Nakorn district in June. The warm and humid climate may enhance proliferation of fleas, therefore, increasing the infection rate in rodents. The authors did not investigate whether there is a seasonal variation in *R. typhi* infection in rodents in the present study. Seasonal variation of murine typhus in humans is not recognized in Thailand, but a disease peak occurs in summer and early fall in temperate countries(13,14).

No antibody to *O. tsutsugamushi* was detected in animals trapped in markets in the BMA. Most endemic *O. tsutsugamushi* infections in Asia are acquired through agricultural exposure. Small ground-dwelling mammals, especially wild rats, are hosts for the transmission of *O. tsutsugamushi* to humans. Scrub typhus can be found in the suburbs of Bangkok which provide typical habitats for these rodents. Humans become infected by intruding the habitats of infected chigger vectors. The prevalence of antibody to *O. tsutsugamushi* found in rodents trapped in north, northeast, south and central parts of Thailand, excluding Bangkok is 5.6 per cent of animals caught(15). The most common *O. tsutsugamushi* infected animal species is *Bandicota. indica* followed by *R. rattus*, *R. exulans*, *R. norvegicus*, *B. savilei*, field rats which are rarely found in the BMA. In Ratchaburi and Nakhon-Pathom provinces, located about 60-90 kilometres west of Bangkok, *O. tsutsugamushi* has been isolated from 29.5 per cent of rats caught including the following species: *B. indica*, *B. bengalensis*, and *R. rattus* (16). In Chiang Mai province in northern Thailand *O. tsutsugamushi* was isolated from 12.9 per cent of trapped rodent species including *B.* indica and *R. rattus thai*(16). Although *R. rattus* was trapped in markets, the predominant species found in markets in the BMA was *R. norvegicus*.

SUMMARY

Murine typhus is a more important human rickettsial disease than scrub typhus in the BMA. *R. typhi* antibody was found in the sera of *R.* norvegicus, *R. rattus*, *R. exulans*, and *M. musculus* trapped in 8 of 47 markets of BMA. *R. norvegicus* is an important reservoir rodent species transmitting *R. typhi* to humans who live close to them. It would be interesting to study the prevalence of murine typhus in individuals living close to markets with a high prevalence of *R. typhi* infected rodents. In addition, a study of flea infestation in these individuals may help the prevention and control of murine typhus in the BMA.

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ความชุกของการติดเชื้อริคเก็ตเซียในสัตว์แทะที่อาศัยอยู่เขตกรุงเทพมหานคร

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มิวรีนไทฟัสและสครับไทฟัสเป็นโรคติดเชื้อริคเก็ตเซียที่สำคัญในประเทศไทย มิวรีนไทฟัสมีสาเหตุจาก *Rickettsia typhi* มีสัตว์แทะเป็นรังโรคและติดต่อสู่คนโดยมีหมัดหนูเป็นพาหะ ส่วนสครับไทฟัสมีสาเหตุจาก *Orientia tsutsugamushi* มีสัตว์แทะเป็นรังโรคเช่นกันและแต่มีไรอ่อน (chigger) เป็นพาหะนำโรค การตรวจพบแอนติบอดีย์ในสัตว์แทะแสดงถึงมีการติดเชื้อในสัตว์นั้น อัตราความชุกของการติดเชื้อริคเก็ตเซียในสัตว์แทะขึ้นกับแหล่งพื้นที่สำรวจ การศึกษานี้มีวัตถุประสงค์เพื่อหาความชุกของการติดเชื้อ *R. typhi* และ *O. tsutsugamushi* ในสัตว์แทะที่จับจากตลาดในเขตกรุงเทพมหานครโดยวิธีการตรวจหาแอนติบอดีย์จำเพาะด้วยวิธีอิมมูนเรืองแสง แอนติเจนที่ใช้ทดสอบเตรียมโดยเพาะเลี้ยงเชื้อ *R. tyhi* และ *O. tsutsugamushi* ในถุงไข่แดงของไข่ไก่ และใช้ FITC–rabbit anti–rat immunoglobulins เป็นแอนติบอดีย์ตัวที่สอง ผลของการตรวจซีรั่มจากหนู 500 ตัวพบแอนติบอดีย์ต่อ *R. typhi* ได้ร้อยละ 5 (25 ตัว) และตรวจไม่พบแอนติบอดีย์ต่อ *O. tsutsugamushi* แอนติบอดีย์ต่อ *R. typhi* ที่ตรวจพบมีแอนติบอดีย์ไตเตอร์อยู่ระหว่าง 40–1280 จากหนูท่อ (*Rattus norvegicus*) หนูท้องขาว (*Rattus rattus*) หนูจี๊ด (*Rattus exulans*), หนูหริ่งบ้าน (*Mus musculus*) โดยพบได้ร้อยละ 4.2, 0.4, 0.2 และ 0.2 ตามลำดับ หนูที่ติดเชื้อ *R. typhi* นั้นจับได้จาก 8 ตลาดจากจำนวน 47 ตลาดที่ทำการสำรวจ การศึกษานี้สรุปผลได้ว่า ในเขตพื้นที่กรุงเทพ–มหานคร *R. typhi* เป็นเชื้อก่อริคเก็ตเซียที่สำคัญ โดยมีหนูท่อเป็นรังโรคสำคัญ

คำสำคัญ : มิวรีนไทฟัส, สครับไทฟัส, *Rickettsia typhi*, *Orientia tsutsugamushi*, หนูท่อ (*Rattus norvegicus*), เขตกรุงเทพมหานคร

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