CORRELATION OF HOST SPECIFICITY, ENVIRONMENTAL FACTORS AND ORIENTAL RAT FLEA ABUNDANCE

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Abstract. Fleas are the vectors of many communicable diseases that are normally found in oriental rats. Climate and environmental changes influence the habitat and migration patterns of vectors. In this study, the oriental rat flea abundance, represented as total flea index, was determined in correlation to host specificity and various environmental factors. The number of hosts and fleas calculated from 3 specific habitats (shipping area, decayed area, and market area) from July 2010 to June 2011. The results showed that the common hosts in the shipping area and decayed area were *Rattus rattus* and *R. exulans*, with the total flea indexes of 3.36 and 1.58. *R. norvegicus* was the most common host identified in the market area. Fleas were virtually absent in rat hosts collected from the market area. Both the density of reservoir hosts and the total flea index were positively correlated with the mean annual rainfall and temperature. These data could be useful for control of rat populations in each specific habitat.

Keywords: *Xenopsylla cheopis*, rat flea, rodent, flea index, climate, Thailand

INTRODUCTION

Fleas are blood-feeding ectoparasites of warm-blooded animals. Nearly three-fourths of all known species of fleas infest rodent and insectivores. Some of them are important to humans as disease vectors (Marquardt, 2004). Flea behaviors are of profound epidemiological importance in the transmission of disease agents. The main diseases that fleas can transmit to humans are bacterial or rickettsial. Fleas also serve as hosts to parasitic worms, and can cause allergic dermatitis or other conditions as a result of their feeding activities (Service, 2004). Among the diseases transmitted by fleas, plague is a highly virulent disease which has killed millions during three historic human pandemics and still re-emerges in the present (Stenseth *et al*, 2008). In 2009, China reported a cluster outbreak of pulmonary plague cases in the remote town of Ziketan, Qinghai Province which led to the deaths of three people (WHO, 2009). International and domestic travel and trade lead to the spread of plague. The surveillance of commensal rats and flea species is quantified by calculating the "Flea index" (an average number of fleas of all species per host)

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as the risk indicator of plague.

Xenopsylla cheopis, the ectoparasitic flea of rats and shrews, is the most important vector for transmitting plague and murine typhus to man. The species is thought to have originated in Egypt, but during the 19th century spread to all parts of the world as parasites of rats infesting ships' cargos. A high incidence of plague infected *X. cheopis* in a given focus greatly increases the risk of transmission to humans. *X. cheopis* most commonly parasitizes *Rattus* species but is frequently found on other rodent species in and around houses. In urban areas, the reservoirs of plague are the domestic rats *R. norvegicus* and *R. rattus* and the insectivore *Suncus murinus* (Gratz, 1999).

Although plague has not occurred in Thailand since 1952, the appearance of flea vectors and incidence of plague is still reported intermittently in neighbor countries (Bin Saeed *et al*, 2005; Joshi *et al*, 2009; Pham *et al*, 2009) including an outbreak in China. This highlights the need to monitor and prevent animal carriers of the plague entering Thailand (The Malaysian Insider, 2009).

This study evaluated the flea species infesting commensal rodents in Bangkok, Thailand and assessed factors influencing their distribution. Associations between flea abundance, its host-dependency, host habitat, and environmental factors were examined. The distribution of *X. cheopis* and calculated flea index will be used to determine the incident of the diseases. This study aims to identify any links between climate and rat populations.

MATERIALS AND METHODS

Study design

The investigation was conducted on the second week of each month for a period of one year, from July 2010 to June 2011. The study sampled 3 specific habitats in Bangkok, namely a shipping port, houses in a decayed area and fresh food markets.

Bangkok port was the shipping port collection site. This port is located on the west side of the Chao Phraya River between km +26.5 and km +28.5 Khlong Toei District. It is well connected with road and rail systems, which transport cargo between the port and the hinterland quickly and economically. Rong Moo community was the houses in decayed area collection site in this category. This community is located on Thang Rotfai Sai Kao Road, Khlong Toei District. It consists of 500 families in 12 acres (30 rai). It is part of The Khlong Toei slum, the oldest and biggest low socioeconomic community in Bangkok (Jongpiputvanich *et al*, 1998). Rung Charoen market was the last collection site in this category. This market is located in Yan Nawa District.

Rodent trapping and flea collection

Small rodents, rats and shrews, were trapped using cage mouse traps. This type of trap could be set in several locations by a single collector, and were baited with dried squids or fish balls and randomly set in different habitats. Traps were checked approximately 2 to 3 hours after sunset and again at 24 hours after setting. The traps containing hosts were placed in a plastic bag or other closed container with cotton balls soaked with ethyl ether until the host became unconscious. The host was removed from the bag while the bag was examined for parasites that left the host body. The host body was then brushed, combed or tapped above a white tray containing water until ectoparasites ceased to appear. The ectoparasites were collected using fine-tipped forceps. The ears, nostrils, mouth and other body openings of the host were examined under stereomicroscope for fleas that may have entered in an attempt to escape the fumigant. Exposed parts of the host such as the ears, nose, feet and tail were also examined for fleas that may be attached by their mouthparts or embedded in the host tissues (Lewis and Siphonaptera, 1993; Shayan and Rafinejad, 2006).

Morphological identification of rodents and their ectoparasitic fleas

Rodent species identification was completed by morphological characterization following the methods of Marshall (1988) and Aplin *et al* (2003). Flea specimens were placed on glass slides and identified by physical characteristics under stereomicroscope, identified to the species level using the description and keys of Lewis and Siphonaptera (1993). After identification, flea specimens were photographed to document significant characteristics specific to genus and species.

Data analysis

Shannon-Wiener diversity index (H) was used to analyze species diversity among 3 different collection sites:

$$H = -\sum_{i=1} (Pi \ln Pi)$$

This index accounts both abundance and richness of the species present. The proportion of species i relative to the total number of species (Pi) is calculated and then multiplied by the natural logarithm of this proportion (InPi). The result is summed across species and multiplied by -1 (Smith, 2002). H- index will increase when the numbers are rising and the distribution of each species is consistent. H is equal to 0 when there is only one type in the dataset. However, in practice, the H- index is smaller than 5.

Percentage of trap success was calculated from number of trapped rodents, divided by number of cages, multiplied by 100. This index indicates the density of reservoir hosts.

$$\text{Percent trap} = \frac{\text{Total amount of trapped rodent}}{\text{Number of cages/ days of trapping}} \times 100$$

The most basic information obtained from flea and rodent surveys is the number of fleas of different species found on various species of hosts. This raw data can be used to calculate various indices, including:

Specific flea index = number of fleas of species *A* collected from host species *Y*, divided by the number of individuals of host species *Y* examined (multiplication of this index by 100 gives the percentage index).

Total flea index = Total number of fleas collected (regardless of species), divided by the total number of hosts examined.

The specific flea index is the most widely used of the above indices. It can be used in conjunction with other rodent and vector surveillance data to estimate human and epizootic risks. For example, it has been reported that a specific flea index of greater than 1 for *X. cheopis* on rats represents a potentially dangerous situation with respect to increased plague risk for humans (Gage, 1999), and that an outbreak is likely to occur if the specific flea index is higher than 5 (Singchai *et al*, 2003).

The number of rats and fleas from each collection site were used to calculate the total flea index and specific flea index.

$$\text{Total flea index} = \frac{\text{Total number of fleas collected from the rats examined}}{\text{Total no. of rats examined}}$$

$$\text{Specific flea index} = \frac{\text{Number of fleas of species } A \text{ collected from host species } Y}{\text{Number of individuals of host species } Y \text{ examined}}$$

RESULTS

A total of 283 rodents were collected from 3 different specific habitats: shipping port (Bangkok port), houses in a decayed area (Rong Moo community), and a fresh food market (Rung Charoen market) between July 2010 and June 2011. Three rat species (*Rattus norvegicus, R. exulans* and *R. rattus*) and one shrew species (*Suncus murinus*) were found at these sites. The most prevalent host species differed at each sample site. These were *R. rattus* (49.6%) at the Bangkok port site, *R. exulans* (71.5%) at the Rong Moo community and *R. norvegicus* (54.3%) at Rung Charoen market (Table 1).

The highest diversity of rats and shrews was found at the port site (H = 0.96) when compared with Rung Charoen market (H = 0.83) and Rong Moo community (H = 0.68).

The mean percentages of trap success rates of the Bangkok port, Rong Moo community and Rung Charoen market sites in the study were: 36.45, 48.13 and 18.85, respectively. The highest percentage of trap success was 70, which appeared in the Rong Moo community in March (Table 2).

A total of 591 flea samples were collected from rats and shrews in the 3 different habitats. Flea samples were identified to species level by description and the keys of Lewis and Siphonaptera (1993). All of them were *X. cheopis*. Fifty-one point four percent of these fleas were found on *R. rattus*, 31.8% on *R. exulans*, 16.4% on *R. norvegicus* and 0.3% on *S. murinus* hosts. The flea abundance on each host species was different in each habitat and related to the predominant host species. Overall flea abundance was higher in the shipping port (68.9%) than the decayed area (31.1%)

Table 1
The abundance of rodent species and fleas, specific flea index of 3 different specific habitats in Bangkok.

Habitat	Host (*n*, %)	No. of flea	Specific flea index
Shipping area	RN (14, 11.6)	97	6.93
	RR (60, 49.6)	220	3.67
	RE (47, 38.8)	90	1.91
	SM (0, 0)	0	0
Decayed area	RN (0, 0)	0	0
	RR (30, 25.9)	84	2.8
	RE (83, 71.5)	98	1.18
	SM (3, 2.9)	2	0.67
Market	RN (25, 54.3)	0	0
	RR (2, 4.3)	0	0
	RE (0, 0)	0	0
	SM (19, 41.3)	0	0
Total	283	591	

RN, *Rattus norvegicus*; RR, *Rattus rattus*; RE, *Rattus exulans*; SM, *Suncus murinus.*

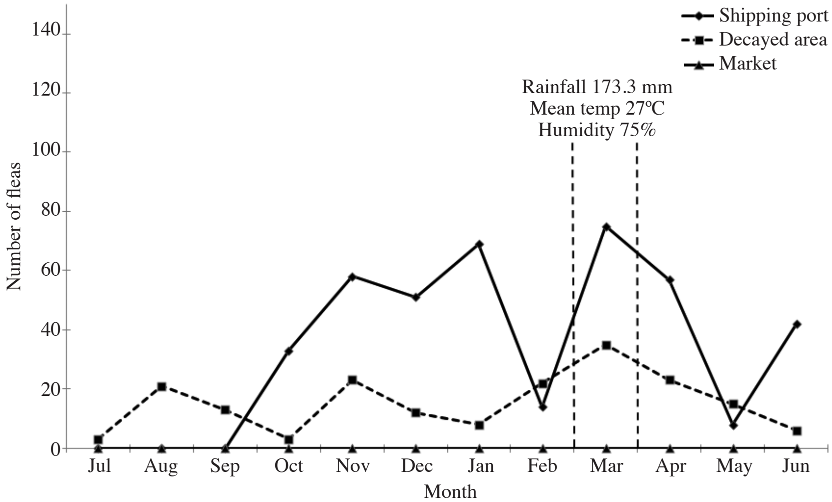

Fig 1–Comparison of flea abundance of each habitat within 12 observed months (July 2010 to June 2011).

and the market (0%) (Table 1). The highest number of fleas recorded in a month was 75 for the Bangkok port and 35 for the Rong Moo community, both occurring in March (Fig 1). No fleas were found in hosts caught in Rung Charoen market during the study (Table 1 and Fig 1).

The raw data was used to calculate various indices, including total flea index and specific flea index. The mean total flea index values were 3.36 for Bangkok port, 1.58 for Rong Moo community and 0 for Rung Charoen market within the 12 observed months. The highest total flea index was 11.4 which appeared in Bangkok port in April. The comparison of total flea index values and the climate of each specific habitat throughout the study is shown in Table 2.

For the average specific flea index value, the most common host at the Bangkok port was found in *Rattus norvegicus* followed by *R. rattus* and *R. exulans*. The most specific flea index value of Rong Moo community was found in *R. rattus* followed by *R. exulans* and *Suncus murinus*. The average specific flea index value of Rung Charoen market in every rodent species = 0 (Table 2). The highest specific flea index was 13.0 which appeared in *R. norvegicus* and *R. rattus* in the Bangkok port (data not shown).

DISCUSSION

The family Muridae includes rats (*Rattus*, *Bandicota*, etc) and mice (*Mus*), both of which are common rodents in Southeast Asia (Marshall and Pantuwatana, 1966). Rats and mice in Thailand can be classified as commensal or field animals according to their natural habitats. The majority of commensal rats and mice previously reported in Thailand were *R. norvegicus*, *R. exulans*, *R. rattus* and *Mus musculus* (Imvithaya *et al*, 1998). This study found that the most abundant species of rats and shrews in 3 different specific habitats in Bangkok differed slightly from the previous report *R. rattus* was the most common species identified in the shipping port, while *R. exulans* was the most prevalent in houses in a decayed area and *R. norvegicus* was the most common in the fresh food markets. This investigation showed the habitat preferences of each rodent. *R. rattus* was most commonly found in and around human dwellings, livestock yards and storage facilities. *R. exulans* is usually confined to villages, often seen climbing on the walls and roofs of houses. *R. norvegicus* is a terrestrial, burrowing species with poor climbing skills, often found close to water and feeding on refuse and stored food (Aplin *et al*, 2003).

Table 2
Percentage of trap success, flea index and climate condition of Bangkok port, Rong Moo community and Rung Charoen market (July 2010 to June 2011).

Month	Collection sites						Temperature (°C)	Rainfall (mm)	Relative humidity (%)
	Bangkok port		Rong Moo community		Rung Charoen market				
	% trap success	Total flea index	% trap success	Total flea index	% trap success	Total flea index			
In 2010									
July	20	0	25	0.6	45	0	29.4	348.7	78
August	0	0	35	3	15	0	28.6	343.1	80
September	0	0	50	1.3	15	0	28.8	409.5	80
October	43.33	2.54	50	0.3	15	0	27.9	256.3	79
November	50	3.87	60	1.9	15	0	28.2	30.6	65
December	62.96	3	45	1.3	15	0	27.5	22.7	68
In 2011									
January	66.67	3.45	40	1	10	0	27	0.3	59
February	30	1.56	55	2	25	0	28.5	21.8	72
March	55	6.82	70	2.5	5.26	0	27	173.3	75
April	16.67	11.4	52.38	2.1	30	0	29.1	134.7	76
May	20	1.33	30	2.5	20	0	29.6	296.5	77
June	63.33	2.21	65	0.46	15	0	29.1	411.9	78
Average	36.45	3.36	48.13	1.58	18.85	0			

Suncus murinus was not observed in the shipping port. *R. norvegicus* was absent from the decayed housing area and *R. exulans* was not recorded in the fresh food markets. These results further confirm that each species of rodent prefer to exist in a different habitat.

Rattus sp is able to breed throughout the year. The peak breeding season is in summer, but where the climate is more strongly seasonal breeding is restricted to the wetter months. The gestation period ranges from 21 to 29 days, and the young rats will leave the nest when they are 3-4 months old (Parker, 1990). In Bangkok, the hot season is in February - April and the highest rainfall occurs in May (TMD, 2011). The highest number of all rat species in our study appeared in June and July, supporting the theory that climate is related to reproductive success.

Bangkok port was the site with a higher diversity of rodents species compared to the other sites (H = 0.96). These results may be due to the various conditions in Bangkok port which allow the habitation of many rodent species. These habitat types include warehouses, export department, open storage areas, cafeteria and staff houses which are each suitable for different rodent species.

The Rong Moo community was the site with a higher density of reservoir hosts than the other sites. The highest density of reservoir hosts in this site appeared in March. It is noteworthy that density of reservoir hosts was related to flea number which was also highest in March. This places this site at higher risk of spreading diseases via rat fleas than other sites, especially during this period.

Experimentally, it has been shown that *X. cheopis* flourish in climatic conditions of 20-25ºC (68-77ºF) with about 70% humidity for egg laying (Shrewsbury, 2005). This flea dislikes temperatures much higher than 26.7ºC (80°F). Usually Bangkok in March has an average daily temperature between 24.9-33.7ºC, but in March 2011 the average temperature was lower than normal by 1-3ºC, and the rainfall amount of this month was also higher than normal (TMD, 2011). This data showed that the highest flea numbers in March were related to the climate, which was more suitable for the fleas.

Fleas are prevalent in the major cities, including Bangkok. In this study the most abundant, *X. cheopis*, was associated with its primary hosts, *R. norvegicus* and *R. rattus*. Both these hosts are typically found in urban areas and other anthropogenic habitats. However, no flea samples were found in rat and shrews collected from Rung Charoen market. This may be due to the rodents often being exposed to water when the market is cleaned, or the regulations of the market for controlling insect disease vectors.

The flea index values of all specific habitats were calculated in 2 indices, total flea index and specific flea index. In each separate month, the total flea index of Bangkok port were >1, with the highest value (11.4) occurring in April (Table 2). This value represents a potentially dangerous situation with respect to increased plague risk for humans (Gage, 1999). The highest specific flea index (13.0) at this site was found in *R. norvegicus* and *R. rattus*. Although the specific flea index in *R. norvegicus* was greater, *R. rattus* (the predominant rat species of Bangkok port) showed a higher number of fleas. This result implies that the fleas prefer certain rodent species over others. This investigation has thus shown an association between flea abundance, climate, host availability and host habitat.

The average total flea index of Rong Moo community also represented a situation of increased plague risk. This index value was >1 in almost every month (except in June and July) with the highest value (2.5) occurring in March and May. The highest specific flea index (6.5) at this site was found in *R. rattus*.

The results of all flea index analyses suggest that Bangkok port is the most risky site for plague occurrence compared with the decayed housing and the market. Another study also reported the overall total flea index of 6 Thailand international seaports was 0.1, with Leam Chabang international port having the highest flea index of 0.9 (Cheewakriengkrai and Parsartwit, 2004). In addition to the high flea index, Bangkok port is Thailand's major port for maritime cargo transport, which increases the likelihood of importing an etiologic agent of disease.

A previous study about the occurrence of plague in Vietnam and its association with ecological factors showed that the incidence of plague peaked during the dry season, with 63% of cases occurring from February through April (Pham *el al*, 2009). The risk of plague occurrence was associated with an increased monthly flea index (for months with the flea index >1) and increased rodent density. This study provided similar findings, showing that the highest density of reservoir hosts in Rong Moo community appeared in March and the highest total flea index of Bangkok port and Rong Moo community occurred in April and March, respectively. Ecological factors found in this study can be interpreted as the chance of plague occurring in Thailand as it has been in Vietnam (Bin Saeed *et al*, 2005; Joshi *et al*, 2009; Pham *et al*, 2009), which has similar climate.

Although our study revealed some concerning results, an outbreak of plague still did not occur due to the absence of the plague pathogen, *Yersinia pestis*, in Thailand. Plague still occurs in Asian countries such as India (Joshi *et al*, 2009), Vietnam (Pham *et al*, 2009) and recently in China (WHO, 2009); all of which have traded with each other and Thailand. Plague may re-emerge in Thailand via infected rats (transported, for example, by ships) arriving in a new city and transmitting the infection to local house rats and their fleas, which then serve as source of human infection (Stenseth *et al*, 2008).

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