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Ectoparasite of Tupaia glis (Scandentia: Tupaiidae) from Lingai agricultural area, Terengganu

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ABSTRACT

Objective: To investigate ectoparasite fauna on the common tree shrew [*Tupaia glis* (*T. glis*)] in Lingai agriculture area, Terengganu.

Methods: The sampling was conducted once a month with five consecutive days from November 2012 to February 2013. Five mammal cage traps ($60 \text{ cm} \times 30 \text{ cm} \times 30 \text{ cm}$) and 30 rat cage traps ($45 \text{ cm} \times 15 \text{ cm} \times 15 \text{ cm}$) baited with banana, salt fish or fleshy-grilled coconut were used. One line transect was built and each trap was set up along the line transect with 5 m intervals. Ectoparasite was collected by combing host's fur vigorously and kept in vials containing 70% ethanol.

Results: Out of 23 hosts examined, 20 individuals (87%) of *T. glis* in Lingai agricultural area were infested by three species of ticks and two species of mites. It was found that *Laelaps echidninus* showed higher mean intensity (5) as compared to the other ectoparasites. However, the prevalence was higher on *Ixodes* sp. (43.5%) though its mean intensity was among the lowest (1.9) from the rest.

Conclusions: This study provides useful information of ectoparasite fauna infesting *T. glis* in the agricultural area. It is important to have knowledge regarding what type of ectoparasite infests small animals in agricultural area, which in turn can assist responsible agencies to take precaution if epidemic outbreaks caused by tick-borne zoonotic diseases occur in the future.

1. Introduction

The first record of *Tupaia glis* (*T. glis*) in 1780 was reported by William Ellis^[1]. It was identified as *Sciurus dissimilis* (Lyon 1913). In Malaysia, common tree shrew (*T. glis* Diard, 1820) is called tupai tanah or ground squirrel. It is a small mammal which is native to the tropical forests of Southeast Asia and currently 20 species have been recorded. The *T. glis* are active on the ground than in trees^[1], while squirrel is arboreal fauna. All *T. glis* are small, agile and omnivorous. Their food preference includes fruits and invertebrates especially arthropods^[2]. Adult *T. glis* shows cannibalistic behaviour that newborn or fresh dead adult *T. glis* are eaten by others^[3]. The International Union for Conservation of Nature has classified *T. glis* as a least concern species^[4]. This is due to their high adaptability towards habitat degradation. Tree shrews have a wide range of habitat utilization and usually found in dipterocarp forests, secondary forests, plantations, fruit orchards and near human settlements.

In Malaysia, *T. glis* is among identified animals which threatened fruit orchards, although the level of destruction is lesser as compared to synanthropic primates such as monkey. Nonetheless, the problem is not only based on plummeted production yield, yet involves the effect of disease transmitted by them. Currently, the studies on the *T. glis*-ectoparasite relationship are limited in Malaysia. Understanding types of ectoparasite-parasitized *T. glis* is

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not only important for our knowledge, but also provides beneficial information for us to predict the host behaviour and assists the authorities to come out with an effective plan focusing in human health security^[5]. This is because there was a case of plague outbreak among human in Vietnam caused by the small mammal and its ectoparasite which inhabited near human settlements^[6].

Previous studies from Gombak Forest Reserve, Malaysia, reported that T. glis was found infested by Laelaps sculpturata, Laelaps aingworthae and Dermacentor sp. and also reported that three chiggers species, namely, Gahrliepia (Walchia) sp., Leptotrombidium deliense and Ascoschoengastia sp. were found infested on T. gliss from Dusun Wildlife Reserve[7,8]. Two types of ectoparasites, chiggers and ticks, were reported to parasitize the T. glis in Ulu Muda Forest Reserve[9], while four species of ticks, one mesostigmatids species and eight species of chiggers were found to infest the T. glis in Panti Forest Reserve[5]. Although there were several studies done on the ectoparasites infested the T. glis in Malaysia, yet the information is still not enough especially in agricultural areas. Therefore, this study has been conducted with aims to identify the ectoparasites on T. glis in Terengganu with the focus area of Lingai agriculture area (LAA). Furthermore, this study also focuses the prevalence and mean intensity of T. glis ectoparasites.

2. Materials and methods

2.1. Study sites

This study was conducted in LAA (5°25'16.32" N, 102°59'58.46" E). The area is situated in 22.9 km from northwest of Kuala Terengganu and 16.1 km west from Universiti Malaysia Terengganu. The agricultural area is planted by rubber plantation and orchards. The orchards are planted by several types of fruit trees such as durian (*Durio* sp.), langsat (*Lansium parasiticum*), cempedak (*Artocarpus integer*), and bitter bean (*Parkia speciosa*).

2.2. T. glis sampling

The sampling was conducted once a month with five consecutive days from November 2012 to February 2013. Five mammals cage traps (60 cm \times 30 cm \times 30 cm) and 30 rat cage traps (45 cm \times 15 cm \times 15 cm) were used to capture the *T. glis*. One line transect was built and each trap was set up along the line transect with 5 m intervals. Traps were baited with either banana, salt fish or fleshy-grilled coconut. Traps were attended twice daily during day time at 08:00 and late evening at 17:00. Caught animals were kept individually in separate clean cloth bags to avoid the mixing of ectoparasites from one host to the other[5,8].

2.3. Ectoparasites collection

Caught hosts were then sedated with chloroform before examining

their ectoparasites. The sedated hosts were placed on a white plain paper and combed vigorously from the tail forward with a toothbrush. Not only parasites under hosts' furs were examined, but the ear and nasal canal were also thoroughly examined for chiggers. Ectoparasite especially ticks was dislodged by spraying 70% ethanol on it[10]. Dislodged ectoparasites which fell on the white plain paper were collected using fine pointed forceps or a moistened end of toothpick and kept in labeled vials containing 70% ethanol for preservation. A separate tube was used for the different host. All collected ectoparasites were deposited in Parasitology Laboratory of Institute of Tropical Aquaculture, Universiti Malaysia Terengganu for identification and curation.

2.4. Ectoparasites identification

All ectoparasites were sorted directly under a dissecting microscope. Identification was done by using Leica DM 750 and tabletop scanning electron microscope (TM-1000). All ectoparasites were identified for their genus and species by using available taxonomic keys[11,12].

2.5. Data analysis

The data such as the prevalence and mean intensity were analyzed by using the Quantitative Parasitology 3.0.

3. Results

A total of 23 individuals of *T. glis* were trapped, out of which 20 individuals (87%) were found to be infected by ectoparasites (Table 1). Five species of ectoparasites were identified comprising two orders, Metastigmata (ticks) and Mesostigmata (mites), and collected from *T. gliss* in LAA. *T. glis* predominantly were found to be infested by ticks with 19 individuals (82.6%) while only five (21.7%) *T. gliss* were infested by mites. *Ixodes* sp. showed the highest prevalence rate (43.5%) followed by *Haemaphysalis* sp. (34.8%) while *Amblyomma* sp. have the least prevalence (4.3%). Regarding the mean intensity, *L. echidninus* showed the highest intensity with five individuals per host while *Haemaphysalis* sp. and *Ixodes* sp. recorded the lowest intensity with 1.9 individuals per host, respectively.

Table 1

Composition of ectoparasites parasitized the tree shrews in LAA.

| Ectoparasite species | No. of | Ectoparasite | Ectoparasite | Prevalence | Mean |
|----------------------|--------|--------------|--------------|------------|-----------|
| | host | frequency | percentage | (%) | intensity |
| Amblyomma sp. | 1 | 2 | 3.51 | 4.3 | 2.0 |
| Haemaphysalis sp. | 8 | 15 | 26.32 | 34.8 | 1.9 |
| Ixodes sp. | 10 | 19 | 33.33 | 43.5 | 1.9 |
| L. echidninus | 2 | 10 | 17.54 | 8.7 | 5.0 |
| Laelaps sp. | 3 | 11 | 19.30 | 13.0 | 3.7 |
| Total | | 57 | 100.00 | | |

L. echidninus: Laelaps echidninus.

4. Discussion

In Malaysia, there have been substantial studies conducted especially in forest reserves to record, map and catalogue the geographical distributions of ectoparasites and their host preferences^[8,9,13-19]. Unfortunately, only a few are focusing on agricultural, plantation, urban or near human settlement areas^[7,20,21]. The short of studies of shrews-ectoparasite or rodent-ectoparasite relationship within or near human settlements should not be ignored as there is high possibility of shrews-borne or rodent-borne zoonotic diseases transmitted directly or indirectly to human through bites, urine and faeces.

The presence of ectoparasites on *T. glis* might be caused by a lack of grooming activities. Out of 110 observations on the behaviour of *T. glis*, just one grooming activity was observed (0.9%) while foraging was the most frequent activity (42.7% or 47 observations^[22]. Grooming was proven as the most effective way in reducing the ectoparasite load on animals whereas the number of ectoparasites would be on the wane as the frequency of grooming activities increases^[23].

In a different perspective, the presence of ectoparasites was also influenced by the availability of different forest biotopes as different species of ticks have their own habitat preferences. Some species of the genus *Haemaphysalis* prefer different ecological conditions (*e.g. Haemaphysalis turturis* and *Haemaphysalis kinneari* prefer semievergreen and semi-deciduous forests; *Haemaphysalis spinigera* prefer deciduous forests, teak and eucalyptus plantations)[2]. Ecotone zone, however, is the most preferred habitat of various ectoparasite species[24]. Though *T. glis* is considered as a kind of forest dwelling animal[25], availability of various food sources consisting of variety of fruits causes *T. glis* to move out of the forest. Thus, the probability of them to get infested by ectoparasites is high since they have to get through the ecotone zone between the forest and LAA. Furthermore, the ecotone zone is also considered as a preferred site for ectoparasite host(s) or reservoir to find food sources and habitats[26].

Some of the ectoparasites species identified in this study were found to be hosts for single or several pathogens, which have been identified to have medical and veterinary interests. *Ixodes* sp. which was found infested *T. gliss* in this study is expected to be *Ixodes granulatus* since it is the only species which have been reported to be parasitized rats, squirrels and shrews in all active stages (larva, nymph and adult)[7]. Infestation of *Ixodes granulatus* is significant as Langat virus has often been segregated from it[27,28]. Furthermore, the genus *Ixodes* was also found related as a causative agent of tickborne zoonotic diseases[29-31].

Knowledge of diseases carried by mites is still scanty and fragmentary. For example, although spiny rat mite (*L. echidninus*) is known as an agent to spread and transmit murine typhus among wild rodents, its role as a vector of human pathogens is perpetually undetermined. In South America, Junin virus which is known as a causative agent for Argentinean hemorrhagic fever had been

isolated from rodents infested by *L. echidninus*^[32]. Currently, there is no information whether *L. echidninus* in Asia harbor the Junin virus. Thus, dissemination of Junin virus in Southeast Asia remains uncertain. Henceforth, some literatures stated that *L. echidninus* predominantly infects murid rodents, especially species of *Rattus*, and if there is a report of *L. echidninus* associated with other animals, it only represents accidental and temporary association^[33]. However, if the results of LAA study should serve as a model, it clearly contradicts the statement^[33]. This is because although there is only two *T. gliss* individuals got infested by *L. echidninus*, the mean intensity was the highest as compared to other ectoparasites.

At the moment, most acarines are probably to have lived in close relationship with their host(s) in pristine and stable habitat such as in the forest reserve where human anthropogenic activities are unlikely to happen. Unfortunately, due to the development pressure, acarine which is a vector of zoonotic diseases will pose a danger to humans due to the changes of behaviours and habitats of their host(s). Habitat degradation will lead wild animals to live near human settlements with the ability of the host(s) to adapt with the new habitat within human vicinity, and contact between human-acarine host(s) cannot be circumvented. Therefore, probability of the occurence of tickborne disease transmission is very high. Currently, the information regarding ectoparasite and their principal host in Malaysia's urban areas is still lacking. However, due to the current rapid development, to identify risk, and emergence of new rickettsial organisms, mapping of animal hosts and arthropod vectors in a particular area is essential[34]. As such, this study will add valuable information to stimulate discussion among authorities and researchers to come out with efficient and long term solutions to prevent epidemic outbreaks caused by tick-borne zoonotic diseases in Malaysia.

Conflict of interest statement

We declare that we have no conflict of interest.

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References

- Fuchs E. Tree Shrews at the German Primate Center. *Primate Biol* 2015;
 2: 111-8.
- [2] Geevarghese G, Mishra AC. *Haemaphysalis ticks of India*. London: Elsevier Inc; 2011.
- [3] Sorenson MW. Behavior of tree shrew. In: Rosenblum LA, editor. *Primate behavior*. New York: Academic Press; 1970, p. 141-93.

- [4] Han KH. *Tupaia gliss*. The IUCN Red List of Threatened Species 2008.
 [Online] Available from: http://www.iucnredlist.org/details/41494/0
 [Accessed on 10th October, 2015]
- [5] Mariana A, Mohd KB, Halimaton I, Suhaili ZA, Shahrul-Anuar MS, Nor ZM, et al. Acarine ectoparasites of Panti Forest Reserve in Johore, Malaysia. *Asian Pac J Trop Biomed* 2011; 1(1): 1-5.
- [6] Moore SM, Monaghan A, Borchert JN, Mpanga JT, Atiku LA, Boegler KA, et al. Seasonal fluctuations of small mammal and flea communities in a Ugandan plague focus: evidence to implicate *Arvicanthis niloticus* and *Crocidura* spp. as key hosts in *Yersinia pestis* transmission. *Parasit Vectors* 2015; 8: 11.
- [7] Paramasvaran S, Sani RA, Hassan L, Krishnasamy M, Jeffery J, Oothuman P, et al. Ectoparasite fauna of rodents and shrews from four habitats in Kuala Lumpur and the states of Selangor and Negeri Sembilan, Malaysia and its public health significance. *Trop Biomed* 2009; 26(3): 303-11.
- [8] Madinah A, Fatimah A, Mariana A, Abdullah MT. Ectoparasites of small mammals in four localities of wildlife reserves in Peninsular Malaysia. *Southeast Asian J Trop Med Public Health* 2011; 42(4): 803-13.
- [9] Mariana A, Zuraidawati Z, Ho TM, Kulaimi BM, Saleh I, Shukor MN, et al. Ticks (Ixodidae) and other ectoparasites in Ulu Muda Forest Reserve, Kedah, Malaysia. *Southeast Asian J Trop Med Public Health* 2008; **39**(3): 496-506.
- [10] Hafiz SM, Marina H, Afzan AW, Chong JL. Ectoparasite from confiscated Malayan Pangolin (*Manis javanica* Desmarest) in Peninsular Malaysia. In: UMT 11th International Annual Symposium on Sustainability Science and Management. Malaysia: Penerbit UMT; 2012.
- [11] Mathison BA, Pritt BS. Laboratory identification of arthropod ectoparasites. *Clin Microbiol Rev* 2014; 27(1): 48-67.
- [12] Wall R, Shearer D. Veterinary ectoparasites: biology, pathology & control. 2nd ed. New Jersey: Wiley-Blackwell Sciences Ltd; 2001, p. 304.
- [13] Upham RW Jr, Nadchatram M. Three new species of trombiculid mites from Malaysia with a redescription and notes *Leptotrombidium muridia* (Womersley) (Acarina, Trombiculidae). *J Medi Entomol* 1968; 5(2): 195-203.
- [14] Mariana A, Zuraidawati Z, Ho TM, Mohd Kulaimi B, Saleh I, Shukor MN, et al. A survey of ectoparasites in Gunong Stong Forest Reserve, Kelantan, Malaysia. *Southeast Asian J Trop Med Public Health* 2005; 36(5): 1125-31.
- [15] Mariana A, Zuraidawati Z, Ho TM. [Ectoparasite fauna in Ulu Muda Forest Reserve]. Forest Biol Divers Ser 2005; 3: 89-98. Bahasa Malaysia.
- [16] Mariana A, Zuraidawati Z, Ho TM. Ecotoparasite fauna at several altitudes of Mount Jerai Forest Reserve. *Forest Biol Divers Ser* 2006; 6: 317-27.
- [17] Mariana A, Zuraidawati Z, Ho TM. Ectoparasite fauna in Pasir Raja Forest Reserve. *Forest Biol Divers Ser* 2006; **7**: 327-35.
- [18] Madinah A, Abang F, Mariana A, Abdullah MT, Mohd-Azian J. Interaction of ectoparasites-small mammals in tropical rainforest of Malaysia. *Community Ecol* 2014; **15**(1): 113-20.

- [19] Adrus M, Ahamad M, Abdullah MT. Detection of rickettsiae in engorged ticks from small mammals in Malaysia. *Borneo J Resour Sci Technol* 2014; 4(1): 34-41.
- [20] Ernieenor Faraliana CL, Ahamad M, Haron MS, Ming HT. Blood meal identification of field collected on-host ticks surrounding two human settlements in Malaysia. *The Experiment* 2013; **17**(2): 1177-83.
- [21] Nursyazana MT, Mohdzain SN, Jeffery J. Biodiversity and macroparasitic distribution of the wild rat population of Carey Island, Klang. *Trop Biomed* 2013; **30**(2): 199-210.
- [22] Kawamichi T, Kawamichi M. Spatial organization and territory of tree shrews (*Tupaia glis*). *Anim Behav* 1979; 27: 381-93.
- [23] Akinyi MY, Tung J, Jeneby M, Patel NB, Altmann J, Alberts SC. Role of grooming in reducing tick load in wild baboons (*Papio cynocephalus*). *Anim Behav* 2013; 85: 559-68.
- [24] Sponchiado J, Melo GL, Landulfo GA, Jacinavicius FC, Barros-Battesti DM, Cáceres NC. Interaction of ectoparasites (Mesostigmata, Phthiraptera and Siphonaptera) with small mammals in Cerrado fragments, Western Brazil. *Exp Appl Acarol* 2015; 66(3): 369-81.
- [25] Net Industries. Tree shrews. 2015. [Online] Available from: http:// science.jrank.org/pages/6952/Tree-Shrews.html [Accessed on 10th October, 2015]
- [26] Woodward J, Strong N, Coe FC, Cloughesy M. Wildlife in managed forests - Oregon forests as habitat. Oregon: Oregon Forest Resources Institute; 2011. [Online] Available from: http://www.wafarmforestry. com/sites/default/files/pdfs/Education/WildlifeMgdHabitat.pdf [Accessed on 10th October, 2015]
- [27] Mansfield KL, Johnson N, Phipps LP, Stephenson JR, Fooks AR, Solomon T. Tick-borne encephalitis virus – a review of an emerging zoonosis. J Gen Virol 2009; 90(Pt 8): 1781-94.
- [28] Kaslow RA, Stanberry LR, LeDuc JW. Viral infection of humans: epidemiology and controls. 5th ed. New York: Springer Science Business Media; 2014, p. 383-414.
- [29] Jaenson TG, Lindgren E. The range of *Ixodes ricinus* and the risk of contracting *Lyme borreliosis* will increase northwards when the vegetation period becomes longer. *Ticks Tick Borne Dis* 2011; 2(1): 44-9.
- [30] Chao LL, Liu LL, Shih CM. Prevalence and molecular identification of Borrelia spirochetes in Ixodes granulatus ticks collected from Rattus losea on Kinmen Island of Taiwan. Parasit Vectors 2012; 5: 167.
- [31] Herrmann C, Voordouw MJ, Gern L. *Ixodes* ricinus ticks infected with the causative agent of Lyme disease, *Borrelia burgdorferi sensu lato*, have higher energy reserves. *Int J Parasitol* 2013; **43**(6): 477-83.
- [32] Parodi AS, Rugiero HR, Greenway DJ, Mettler N, Martinez A, Boxaca M, et al. [Isolation of the Junin virus (epidemic hemorrhagic fever) from the mites of the epidemic area (*Echinolaelaps echidninus*, Barlese)]. *Prensa Med Argent* 1959; **46**: 2242-4. Spanish.
- [33] Furman DP. Laelapid mites (Laelapidae: Laelapinae) of Venezuela. Utah: Brigham Young University; 1972, p. 58.
- [34] Mohd Zain SN, Amdan SA, Braima KA, Abdul-Aziz NM, Wilson JJ, Sithambaran P, et al. Ectoparasites of murids in peninsular Malaysia and their associated diseases. *Parasit Vectors* 2015; 8: 254.