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ORIGINAL PAPER

Ectoparasitic insects and mites on Yunnan red-backed voles (*Eothenomys miletus*) from a localized area in southwest China

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Abstract Ectoparasitic insects and mites on Yunnan red-backed voles (*Eothenomys miletus*) in Dali prefecture, Yunnan Province, southwest China, were studied between 2003 and 2004. In total, 34,389 individuals of 86 species of ectoparasitic insects (seven species of fleas and five species of sucking lice) and mites (23 species of gamasid mites and 51 species of chigger mites) were collected from 916 individual hosts. The diversity of ectoparasites on this single rodent species in such a small area was much higher than in previous reports, which concerned more host species and greater geographical areas. The majority of the ectoparasites were chigger mites, which accounted for 59.3 % of the parasite species and 87.4 % of the individual parasites. Most voles harbored parasites with an overall prevalence (*P*) of 82.5 % and mean abundance (MA) of 37.5 parasites per host. The dispersion coefficient (*C*) and patchiness index (*m**/*m*) were used to study the spatial patterns of the seven dominant parasite species, and all seven had aggregated distributions. The species abundance distribution of the ectoparasites on the vole was fitted by Preston's lognormal distribution (R^2=0.82), and the total expected parasite species was estimated from this plot as 167 species. Yunnan red-backed voles harbor many ectoparasites as revealed by examination of a large host population. Future field investigations should sample large numbers of host individuals to assess ectoparasite populations.

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Introduction

The Yunnan red-backed vole (also called Yunnan Chinese vole or large oriental vole), *Eothenomys miletus* (Thomas 1914), is mainly distributed in the Himalayan Ranges of southwest China. As a dominant rodent species in farmlands and scrublands, *E. miletus* is abundant in the Hengduan Mountainous Regions of Yunnan Province, including Dali, where the current study was based (Luo et al. 2004; Zhu et al. 2010). Besides destroying crops and plants, the Yunnan red-backed vole is also an important reservoir host of plague, murine typhus, scrub typhus, hemorrhagic fever with renal syndrome (HFRS), and some other zoonoses (Yang and Yang 1985; Yu and Xu 1988; Guo and Dong 2008; Zhang et al. 2008).

Rodents and other small mammals often harbor two taxa of insects (fleas and sucking lice) and two taxa of mites (chigger mites and gamasid mites). These ectoparasitic arthropods are important vectors of some zoonoses (zoonotic diseases) and vector-borne diseases. Fleas are known as the vectors of plague, murine typhus (endemic typhus), and flea-borne spotted fever. Sucking lice (for example, the human louse *Pediculus humanus*) are associated with the transmission of epidemic typhus, epidemic relapsing fever, and trench fever. Gamasid mites may play a potential role in transmitting hemorrhagic fever with renal syndrome (HFRS; also called epidemic hemorrhagic fever, EHF), and chigger mites are the transmission vectors of scrub typhus, or tsutsugamushi disease (Deng et al. 1993; Wu et al. 1996; Li et al. 1997; Jin 1999; Song 1999; Brouqui and Raoult 2006; Wang and Ye 2006; Bitama et al. 2010).

The ectoparasites of rodents and other small mammals were previously reported in some local investigations, which usually covered a wide geographical region and included many host species (Ritzi and Whitaker 2003; Storm and Ritzi 2008; Oguge et al. 2009; Paramasvaran et al. 2009; Changbunjong et al. 2010; Zuo et al. 2011; Zhan et al. 2013). The previous investigations, however, often neglected to make detailed analyses of the ectoparasites on a single host species in a localized area. We studied ectoparasites on the Yunnan red-backed vole in a localized area surrounding Erhai Lake in Dali. Besides analyzing the infestation and spatial pattern of the ectoparasites on the vole, this paper also explored the maximum likelihood number of parasitic species harbored by this single species living in a localized area.

Materials and methods

Collection and identification of ectoparasites

The study site included the area surrounding Erhai Lake: which measures 10×45 km at maximum width and length [100°05′E–100°17′E, 25°36′N–25°58′N]. The study area covered about 450 km^2 at an altitude of around 1,976 m. Yunnan red-backed voles were captured with mouse traps from three sampling sites (Xiaguan, Wase, and Xizhou) surrounding Erhai Lake in 2003 and 2004. The traps were set in different habitats (farmland, scrubland, woodland, etc.) in the evening and then checked the following morning. The trapped voles were placed in white cloth bags and brought to the laboratory, where the ectoparasites were collected from them and separately preserved in labeled vials containing 70 % ethanol (Zuo and Guo 2011; Zuo et al. 2011; Huang et al. 2013; Zhan et al. 2013). Chigger mites were especially collected by a curette and lancet from the auricles and external auditory canals of the host ears, where chigger larvae are usually attached (Li et al. 1997; Guo et al. 2006). All the voles were identified according to their morphological features (Huang et al. 1995; Huang et al. 2013). All the ectoparasites were dehydrated, transparentized, mounted on to glass slides, and finally identified to species under microscopes (Deng et al. 1993; Li et al. 1997; Jin 1999; Wu 2007; Huang et al. 2013). Voucher ectoparasitic insects and mites, together with representative voles, are deposited in the specimen repository of the Institute of Pathogens and Vectors, Dali University.

The capturing of the voles (rodents) was officially permitted by the local authority of wildlife service in Dali Prefecture, Yunnan Province, China. The use of animals for research was also officially approved at the Institute of Pathogens and Vectors, Dali University.

Basic statistical analysis

Constituent ratios (Cr), prevalence (P), and mean abundance (MA) were calculated for each parasite species (Ritzi and Whitaker 2003; Storm and Ritzi 2008; Huang et al. 2013; Zhan et al. 2013). The dispersion coefficient (C) and patchiness index (m^*/m) were used to determine the spatial patterns of the dominant parasite species among different individual voles (Kuno 1991; Lopez 2005; Guo et al. 2006; Seal et al. 2006).

Description of species abundance distribution and total species estimation

In a semi-logarithmic rectangular coordinate system, the x-axis (indicating the parasite individuals) was marked with log intervals based on $\log_3 N$ (Table 4 in "Results"). The y-axis was an arithmetic scale, indicating the parasite species. Preston's lognormal distribution model was used to fit the theoretical curve of species abundance distribution (Preston 1948; Greig-Smith 1983; Baltanas 1992; Guo 1999; Guo et al. 2006) as follows: $\widehat{S}(R) = S_0 e^{-[a(R-R_0)]^2} (e = 2.71828\ldots\ldots)$, where $\widehat{S}(R)$ is the theoretical species number in R-th log interval, R_0 the mode log interval, and S_0 the species number at R_0. The value of α (spread constant) was determined according to the determination coefficient (R^2) and the fitting effect (Ni 1990; Guo et al. 2006).

Based on the species abundance distribution, the total expected parasite species (S_T) and the probably missed parasite species in the field sampling (S_M) were approximated by $S_T = (S_0\sqrt{\pi})/a$ and $S_M = S_T - S_A$ ($\pi = 3.14159\ldots\ldots$; S_A = the actually collected parasite species) (Preston 1948; Greig-Smith 1983; Baltanas 1992; Guo 1999).

Results

Ectoparasitic insects and mites present on Yunnan red-backed voles

The 916 Yunnan red-backed voles included 488 males and 428 females, and they were captured from the woodland

(483 individuals, 52.7 %), scrubland (245 individuals, 26.8 %), and farmland (188 individuals, 20.5 %). In total, 34,389 individual ectoparasitic insects and mites were collected from the voles. These ectoparasites were identified as comprising 86 different species from 27 genera in eight families (Table 1). In total, 82.5 % of the voles was infested by parasites (total P=82.5 %) with a high mean abundance (total MA=37.5 parasites per host). Of the four ectoparasite taxa (fleas, sucking lice, chigger mites, and gamasid mites), the majority of species (Cr=59.3 %) and individuals (Cr=87.4 %) were chigger mites with much higher P and MA than other three taxa (Table 2). Of 86 parasite species, some transmitting vectors (or potential transmitting vectors) of zoonoses were found, including six species of fleas that are vectors of plague and murine typhus, five species of gamasid mites that are potential transmitting vectors of HFRS, and six species of chigger mites that are transmitting vectors of scrub typhus (Table 1).

Dominant ectoparasite species and their spatial patterns

One dominant species appeared in each of the taxa of fleas, sucking lice, and gamasid mites, which accounted for 93.2, 99.3, and 81.7 % of the total corresponding taxa (Table 3). There were four dominant species of chigger mites (Table 3), and they accounted for 69.5 % of the total 51 chigger mite species. These seven dominant ectoparasite species all

Table 1 Collection details of ectoparasitic insects and mites from the Yunnan red-backed vole (*E. miletus*) in the localized area of Dali, Yunnan Province, southwest China

Taxonomic taxa of ectoparasites	Collected species and individuals
Fleas	Total fleas: 1,317 individuals; 7 species, 6 genera, 2 families
Ctenophthalmidae	*Ctenophthalmus quadratus*[a] (1,227); *Ctenophthalmus yunnannus* (2); *Neopsylla specialis*[a] (58); *Stenischia huminis*[a] (20)
Leptopsyllidae	*Leptopsylla segnis*[a] (3); *Frontopsylla spadix*[a] (5); *Paradoxopsyllus custodies*[a] (2)
Sucking lice	Total lice: 1,485 individuals; 5 species, 2 genera, 2 families
Hoplopleuridae	*Hoplopleura edentula* (1,475)
Polyplacidae	*Polyplax reclinata* (2); *Polyplax serrata* (5); *Polyplax spinulosa* (1); *Polyplax* sp. (2)
Gamasid mites	Total gamasid mites: 1,538 individuals; 23 species, 11 genera, 3 families
Laelapidae	*Laelaps chini* (1,256); *Laelaps echidninus*[a] (46); *Laelaps guizhouensis* (34); *Laelaps nuttalli* (8); *Laelaps paucisetosa* (10); *Laelaps traubi* (9); *Haemolaelaps cordatus* (1); *Androlaelaps casalis*[a] (3); *Androlaelaps fahrenholzi*[a] (33); *Androlaelaps singularis* (6); *Eulaelaps substabularis* (3); *Gymnolaelaps weishanensis* (3); *Haemogamasus oliviformis* (23); *Haemogamasus sexsetosus* (39); *Hirstionyssus sunci*[a] (1); *Hypoaspis chianensis* (4); *Hypoaspis lubrica* (1); *Hypoaspis miles* (3); *Hypoaspis pavlovskii* (17)
Macronyssidae	*Ornithonyssus bacoti*[a] (1)
Aceocejidae	*Proctolaelaps pygmaeus* (21); *Lasioseius medius* (2); *Lasioseius* sp. (14)
Chigger mites	Total chigger mites: 30,049 individuals; 51 species, 8 genera, 1 family
Trombiculidae	*Leptotrombidium scutellare*[a] (7,962); *Leptotrombidium sinicum* (5,690); *Leptotrombidium eothenomydis* (3,163); *Leptotrombidium apodemi* (3); *Leptotrombidium apodevrieri* (8); *Leptotrombidium bambicola* (30); *Leptotrombidium baoshui* (1); *Leptotrombidium bishanense* (121); *Leptotrombidium deliense*[a] (71); *Leptotrombidium densipunctatum* (726); *Leptotrombidium deplanoscutum* (1); *Leptotrombidium dianchi* (22); *Leptotrombidium gongshanense* (368); *Leptotrombidium hiemalis* (524); *Leptotrombidium imphalum*[a] (10); *Leptotrombidium jianshanense* (1); *Leptotrombidium jinmai* (587); *Leptotrombidium kaohuense*[a] (9); *Leptotrombidium longchuanense* (33); *Leptotrombidium longimedium* (15); *Leptotrombidium neotebraci* (1); *Leptotrombidium robustisetum* (51); *Leptotrombidium rupestre*[a] (28); *Leptotrombidium rusticum* (782); *Leptotrombidium saltuosum* (1); *Leptotrombidium shuqui* (41); *Leptotrombidium suense* (44); *Leptotrombidium xiaguanense* (25); *Leptotrombidium xiaowei* (48); *Leptotrombidium xishani* (1); *Leptotrombidium yongshengense* (11); *Leptotrombidium yui*[a] (311); *Leptotrombidium yunlingense* (29); *Herpetacarus hastoclavus* (2,037); *Herpetacarus tenuiclavus* (1); *Trombiculindus bambusoides* (144); *Trombiculindus chilie* (2); *Trombiculindus spinifolatus* (1); *Trombiculindus yunnanus* (1,168); *Helenicula simena* (4,057); *Helelenicula globularis* (4); *Ascoschoengastia leechi* (31); *Ascoschoengastia* sp.(6); *Gahrliepia linguipelta* (927); *Gahrliepia radiopunctata* (1); *Gahrliepia silvatica* (9); *Gahrliepia yunnanensis* (316); *Walchia acutascuta* (1); *Walchia ewingi* (114); *Walchia koi* (511); *Chatia acrichela* (1)
Total ectoparasites	34,389 individuals; 86 species, 27 genera, 8 families

The number in the bracket is the collected individuals of each parasite species

[a] The transmitting vectors or potential transmitting vectors of the zoonoses, plague, murine typhus (endemic typhus), hemorrhagic fever with renal syndrome (HFRS; also called "epidemic hemorrhagic fever, EHF" in China), and scrub typhus (tsutsugamushi disease)

Table 2 The constituent ratios (Cr), prevalence (*P*), and mean abundance (MA) of the four taxa of ectoparasites on the Yunnan red-backed vole (*E. miletus*) in the localized area of Dali, Yunnan Province, southwest China

Ectoparasite categories	Species and constituent ratios (Cr)		Individuals and constituent ratios (Cr)		Prevalence (*P*)		Mean abundance (MA)	
	Species	Cr (%)	Individuals	Cr (%)	Infested voles	*P* (%)	Parasite individuals	MA
Fleas	7	8.1	1,317	3.8	418	45.6	1,317	1.4
Sucking lice	5	5.8	1,485	4.3	158	17.3	1,485	1.6
Gamasid mites	23	26.7	1,538	4.5	334	36.5	1,538	1.7
Chigger mites	51	59.3	30,049	87.4	560	61.1	30,049	32.8
All insects	12	13.9	2,802	8.2	486	53.1	2,802	3.1
All mites	74	86.0	31,587	91.9	657	71.7	31,587	34.5
Total parasites	86	100.0	34,389	100.0	756	82.5	34,389	37.5

exhibited an aggregated distribution among the host with both C and m^*/m much higher than 1.0 (Table 3).

Species abundance distribution and total species estimation of ectoparasites

The species abundance distribution of the ectoparasite community (all the insects and mites on *E. miletus*) was successfully fitted by Preston's lognormal distribution with R^2=0.82. The theoretical curve of the species abundance distribution showed a gradually descending tendency from the rare parasite species (the highest point) to the dominant parasite species (the lowest point) (Table 4; Fig. 1). The theoretical curve equation was $\widehat{S}(R) = 16e^{-[0.17(R)]^2}(S_0 = 16;\ \ R_0 = 0; \alpha = 0.17; e = 2.71828\ldots\ldots)$. Based on $S_T = (S_0\sqrt{\pi})/a$ and $S_M = S_T - S_A$, the total expected species of ectoparasitic insects and mites on *E. miletus* was estimated to be about 167 species (S_T =167). The missed species was about 81 species (S_M=81).

Discussion

Infestation of ectoparasitic insects and mites on Yunnan red-backed voles

In the present study, the sample size of 916 host animals of a single species was far larger than previous studies which have included multiple species sampled over much wider geographical areas (Ritzi and Whitaker 2003; Storm and Ritzi 2008; Oguge et al. 2009; Paramasvaran et al. 2009; Changbunjong et al. 2010). From this large sample, we collected 86 species of ectoparasitic insects and mites. This number also exceeded the numbers observed in previous studies using smaller sample sizes (Ritzi and Whitaker 2003; Storm and Ritzi 2008; Oguge et al. 2009; Paramasvaran et al. 2009; Changbunjong et al. 2010). For example, from 14 species and 204 individuals of rodents and shrews in Malaysia, 20 species of ectoparasites were collected from five

Table 3 The dominant species of ectoparasitic insects and mites on the Yunnan red-backed vole (*E. miletus*) in the localized area of Dali, Yunnan Province, southwest China

Dominant parasite species	Individuals	Cr (%)	*P* (%)	MA	*C*	*m*/m*
Fleas						
Ctenophthalmus quadratus	1,227	93.2	43.7	1.3	8.0	6.3
Sucking lice						
Hoplopleura edentula	1,475	99.3	16.8	1.6	40.3	25.4
Gamasid mites						
Laelaps chini	1,256	81.7	30.6	1.4	11.9	8.9
Chigger mites						
Leptotrombidium scutellare	7,962	26.5	28.1	8.7	139.1	16.9
L. sinicum	5,690	18.9	26.2	6.2	86.2	14.7
Helenicula simena	4,057	13.5	31.2	4.4	43.8	10.7
L. eothenomydis	3,163	10.5	11.5	3.5	76.7	22.9

Constituent ratios (Cr %) here means the constituent ratios of the dominant parasite species in the corresponding taxa of ectoparasites on the vole hosts. The *P* (%), *MA*, *C*, and *m*/m* stand for the infestation prevalence, mean abundance, dispersion coefficient, and patchiness index of the dominant parasite species, respectively

Table 4 The statistical parameters for fitting the theoretical curve of species abundance distribution of ectoparasitic insects and mites on the Yunnan red-backed vole (*E. miletus*)

Log intervals (*A*)	Individual ranges in each log interval (*B*)	Midpoint values of each individual range (*C*)	Actual parasite species	Calculated theoretical parasite species
0	1	1	16	16.00
1	2–4	3	14	15.54
2	5–13	9	12	14.25
3	14–40	27	16	12.34
4	41–121	81	9	10.08
5	122–364	243	3	7.77
6	365–1,093	729	7	5.65
7	1,094–3,280	2,187	6	3.88
8	3,281–9,841	6,561	3	2.52
9	9,842–29,524	19,683	0	1.54

"*C*" is the midpoint of *B*; $3^A=C$; $\log_3 C=A$

taxa (Paramasvaran et al. 2009). Another survey documented 54 species of ectoparasites present on 166 individuals of 12 species of small mammals (Storm and Ritzi 2008).

The species diversity of ectoparasites on *E. miletus* in the present study was much higher than that on *Rattus norvegicus* (brown rat) in the same locality (the areas surrounding Erhai Lake), in which 47 species and 8,040 individuals of ectoparasites were collected from 431 individuals of *R. norvegicus*, including 23 species of chigger mites, 16 species of gamasid mites, 6 species of fleas, and 2 species of sucking lice (Dong et al. 2009). The overall prevalence (P=82.5 %) and mean abundance (MA=37.5 parasites per host) of the ectoparasites on *E. miletus* (Table 2) were also higher than those on *R. norvegicus* (P=71.0 %, MA=18.65 parasites per host) (Dong et al. 2009). Moreover, the species number (51 species), P (61.1 %), and MA (32.8) of chigger mites (Table 2) were even much higher than those on *Rattus flavipectus* (*Rattus tanezumi*, Asian house rat) from six counties of Yunnan Province, in which 31 chigger mite species were collected from 451 rats with P=18.18 % and MA=4.69 (Guo et al. 2006).

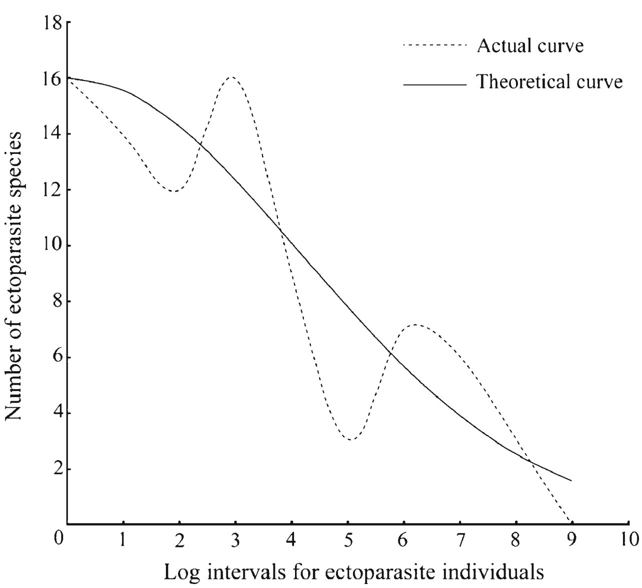

Fig. 1 The species abundance distribution of ectoparasitic insects and mites on the Yunnan red-backed vole (*E. miletus*) fitted by Preston's lognormal distribution with the theoretical curve equation of $\widehat{S}(R) = 16\, e^{-[0.17(R)]^2} \left(S_0 = 16; R_0 = 0; \alpha = 0.17; e = 2.71828\ldots\ldots; R^2 = 0.82\right)$

These results therefore show that Yunnan red-backed voles have very high and diverse infestations of ectoparasitic insects and mites. This may be related to several factors: (1) In contrast to *R. norvegicus* and *R. flavipectus* which lives almost exclusively in houses and residential areas (Huang et al. 1995), *E. miletus* is a typical wild rodent and is found in diverse habitats including the woodland, scrubland, and farmland (Yang and Yang 1985; Huang et al. 1995). As a wild rodent species, it may have a lot of opportunity to contact other voles, rodents, and small mammals. Some ectoparasites with low host specificity may frequently invade them by cross infestation, and this allows the vole to harbor more parasites. In addition, gamasid mites actually include some free-living species in the soil, litter, and humus (Pan and Deng 1980; Huang et al. 2013), and some free-living gamasid mites (i.e., *Proctolaelaps pygmaeus*, *Lasioseius medius*, and *Lasioseius* sp. in Table 1) were collected from the vole. These free-living gamasid mites are certainly not true ectoparasites, but they can crawl onto the body surface of some small mammals by accident (Huang et al. 2013). This also increases the number of "parasite" species on the vole. (2) The large host sample (916 voles) increased the probability of detecting additional uncommon ectoparasite species. Therefore, a large host sample is strongly recommended in future field investigations. (3) The larvae of chigger mites often reside on the auricles and external auditory canals of the host ears (Li et al. 1997; Guo et al. 2006), and they are too tiny to be seen by the naked eye. In our collection, both ears of each vole were carefully inspected, and this allowed us to find abundant chigger mites, i.e., 51 species with 30,049 individuals. These may have been overlooked in previous investigations.

Dominant ectoparasites on Yunnan red-backed voles

Of the four taxa of ectoparasites on *E. miletus*, the majority were chigger mites with a high species diversity, more than one dominant species and abundant individuals. This was similar to the situation on *R. norvegicus* (Dong et al. 2009). Many species of chigger mites infesting a single host species imply the lower host specificity of chigger mites than the other three taxa of ectoparasites.

Species abundance distribution and total species estimation

Preston's lognormal distribution model is a classical way to fit the species abundance distribution, which aims to illustrate the relationship between species and individuals in a certain community (Preston 1948; Greig-Smith 1983; Baltanas 1992; Guo 1999). The theoretical curve of species abundance distribution for the ectoparasites on *E. miletus* showed a gradually descending tendency, and this revealed that most parasite species were rare with very few individuals, but a few dominant species had abundant individuals. Based on the theoretical curve, the total ectoparasitic insect and mite species on the vole was estimated to be 167 species. Hence, about 81 parasite species were missed in the field investigation. Consequently, although we captured nearly 1,000 voles, this number was sufficient to collect only about half species of ectoparasites present. This further emphasizes the necessity of collecting large samples of host individuals to adequately assess the populations of their ectoparasites.

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