Acta Chiropterologica, 11(1): 157–162, 2009
PL ISSN 1508-1109 © Museum and Institute of Zoology PAS
doi: 10.3161/150811009X465776

Host-parasite interactions of tropical bats in Puerto Rico

KRISTLE KRICHBAUM[1], SARAH PERKINS[1, 3], and MICHAEL R. GANNON[2]

[1]*Center for Infectious Disease Dynamics, 208 Mueller Laboratory, The Pennsylvania State University, University Park, PA 16802, USA*
[2]*Department of Biology, The Pennsylvania State University, Altoona College, Altoona, PA 16601, USA*
[3]*Corresponding author: E-mail: sep18@psu.edu*

Bats are receiving increasing attention in the parasitological world due to their potential role as reservoir hosts for zoonotic pathogens. However, details of the macroparasitic fauna (helminths and ectoparasites) are currently recorded and studied. Here, we start to address this paucity of data by detailing a study where we have documented the macroparasite fauna of a sample of tropical bats (*Mormoops blainvillei*, *Pteronotus quadridens*, and *Monophyllus redmani*) from Puerto Rico. Additionally, we investigated the possible host characteristics influencing the prevalence and intensity of macroparasite infection. Macroparasites were collected and identified from three species of bat, which were thoroughly washed and dissected. The overall parasite community of all three bat species consisted of a range of ectoparasites as well as the cestode *Vampirolepis christensoni* and the nematode *Capillaria pusilla*, although there was considerable variation in the parasite community of each individual species. We discovered bat flies of a previously undescribed species of the *Nycterophilia* genus as well as new parasite records for all three species of bats. All parasites had an aggregated distribution within the host population. Differences were observed in the intensity of the helminths between bat species, but not for ectoparasite prevalence. As the helminth intensity increased so the ectoparasite intensity decreased. Overall, the helminth intensity was female-biased and increased, for both sexes with increasing body mass; no sex-bias or body mass effects were associated with ectoparasite prevalence.

Key words: *Mormoops blainvillei*, *Pteronotus quadridens*, *Monophyllus redmani*, macroparasites, parasite infection, tropical bats, Puerto Rico, Caribbean

INTRODUCTION

Despite having nearly one thousand known extant species in the order of Chiroptera (Wilson and Reeder, 2005), bats remain little studied in terms of the parasites and pathogens they harbor, particularly those from tropical regions. This lack of documentation of the parasite fauna of bats is of some concern since tropical bats have been implicated in the emergence of several important zoonotic infections, such as Nipah and Hendra virus as well as a possible sylvatic reservoir for Ebola (Calisher *et al*., 2006). As a consequence, recent studies have focused on documenting microparasitic infections of bats, while at times overlooking the macroparasitic infections (but see Gannon and Willig, 1995). Macroparasites are a group of parasites which typically live in or on the host body and include ectoparasites and intestinal helminths. This group of macroparasites has been shown to be important in regulating host populations and may interact with other parasites within the host to alter pathogen dynamics (Christensen *et al*., 1987; Bentwich *et al*., 1999, 2000; Cox, 2001). As such, an investigation into the macroparasitic fauna of bats is undeniably important.

We provide here a documentation of the macroparasitic infections of three different species of tropical bat from Puerto Rico. The bats species studied include *Mormoops blainvillei* (Antillean ghost faced bat), *Pteronotus quadridens* (sooty mustached bat), and *Monophyllus redmani* (Greater Antillean long tongued bat). Puerto Rico is the smallest and most eastern of the Greater Antillean islands. As such it stands as an island fulcrum between the Greater and Lesser Antilles and contains flora and fauna of both. The bat fauna and distribution on the island have been previously well documented (Gannon *et al*., 2005).

We investigated which host characteristics may be associated with macroparasitic infection. For example, increasing body mass has been associated with increasing parasite intensity (Arneberg *et al*.,

1998; Wilson *et al*., 2002; Perkins *et al*., 2003). Additionally, where sexual size dimorphism is male-biased so parasitic intensity can be expected to be male-biased and so we also determined if a sex-bias in infection occurred (Moore and Wilson, 2002).

In this manuscript we first provide a description of the helminth and ectoparasite species associated with three different species of tropical bat from Puerto Rico. Secondly, we describe the differences in macroparasite infections and determined if we could relate the differences in parasite intensity and prevalence between the species to characteristics to host characteristics including body mass, species and host sex.

Materials and Methods

Collection of the bats occurred on the island of Puerto Rico during August 2004. A harp trap was placed outside the entrance of the Culebrones Cave, located at Mata de Platano Field Station (Municipality of Arecibo, Barrio Dominguito, field station operated by InterAmerican University, Bayamon, Puerto Rico) between approximately 19:00 and 01:00 hours. Culebrones cave is uniquely tropical, being a 'hot cave'; as defined by Silva-Taboada (1977, 1979), having one reduced entry with little air circulation, a high bat population density, relative humidity above 90% and a constant temperature ranging between 28°C and 40°C year round. This cave is known to contain seven of the 13 species of bats known from Puerto Rico (Gannon *et al*., 2005).

Of the 24 individual bats captured, four belonged to *Monophyllus redmani*, 12 to *Mormoops blainvillei*, and the remaining eight to *Pteronotus quadridens*. All three of these species are West Indian endemics and ubiquitous throughout the Greater Antillean Islands (Gannon *et al*., 2005)

Upon capture, bats were sacrificed, fixed in formaldehyde, wrapped in paper towels and sealed individually in plastic bags. Penn State University Animal Care Committee approved protocols were followed for all animal handling. All collected animals were deposited as voucher specimens in the Field Museum of Natural History in Chicago, IL at the conclusion of the research. Each bat carcass was washed vigorously with water over three stainless steel mesh sieves: the top sieve had an aperture of 9.5 mm and the bottom two had apertures of 212 micrometers. The bottom two sieves were rinsed and the debris collected in a Petri dish, which was then examined under a dissecting microscope. The ectoparasite specimens collected were stored and preserved as voucher specimens in 70% ethanol. The gastrointestinal tract was removed and cut from the colon to the stomach. Each section of the gastrointestinal tract was examined carefully, using a binocular microscope and all intestinal helminths removed and preserved in 10% mixture of glycerol in 70% ethanol for identification.

We examined the frequency distribution of the intensity of helminths to determine if the distribution was consistent with a negative binomial distribution. To test for differences in parasite prevalence among species and sex, we used parasite prevalence or intensity as the response variable. For all analyses we used generalized linear models (GLM's), with the appropriate error distribution, which were carried out in Splus (Insightful Corporation, USA).

Results

In this sample of bats several ectoparasites and two intestinal helminths were recorded (Table 1). However, not all parasites were distributed evenly among the bats collected, for example one bat species, *M. redmani*, was helminth-free. The ectoparasite community collected from the bats consisted of mites, flies, chiggers and a springtail. All three bat species shared one common parasite; the *Periglischrus* mite. Flies observed on *P. quadridens* and *M. redmani* were identified as members of the *Nycterophilia* genus, but were a previously unidentified species and likely constitute a new species to science (Carl Dick, personal communication). The remaining ectoparasites were collected on *P. quadridens* and *M. blainvillei*. *Pteronotus quadridens* hosted a Chirodiscidae mite and a Collembolan springtail. *Mormoops blainvillei* hosted two types of fly (*Trichobius* and nematoceran) and one trombiculid chigger. Ectoparasite prevalence was greatest among *M. redmani* ($\bar{x}$ ± SE, 4.00 ± 1.15) and smallest among *M. blainvillei* ($\bar{x}$ ± SE, 0.25 ± 0.13). The two helminth species observed in the sample were the cestode *Vampirolepis christensoni*, which was found exclusively in *M. blainvillei* and the nematode *Capillaria pusilla*, which was more prevalent than the cestode *V. christensoni*. The frequency distribution of the macroparasite intensity followed a negative binomial distribution (Fig. 1).

Table 1. Descriptive statistics of parasite species from three species of tropical bat. Mean, standard error of the mean (SE), and prevalence (prev) of all parasites are given. The sample size of ectoparasites observed from different species was low; for this reason the data were grouped together as 'ectoparasites'

Bat species	Cestode (*Vampirolepis christensoni*)			Nematode (*Capillaria pusilla*)			All helminths			All ectoparasites		
	$\bar{x}$	SE	prev	$\bar{x}$	SE	prev	$\bar{x}$	SE	prev	$\bar{x}$	SE	prev
P. quadridens	0.00	0.00	0.00	0.25	0.16	0.25	0.25	0.16	0.25	2.50	1.80	0.25
M. blainvillei	0.83	0.75	0.17	2.50	1.35	0.42	3.33	1.64	0.50	0.25	0.13	0.63
M. redmani	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	4.00	1.15	1.00

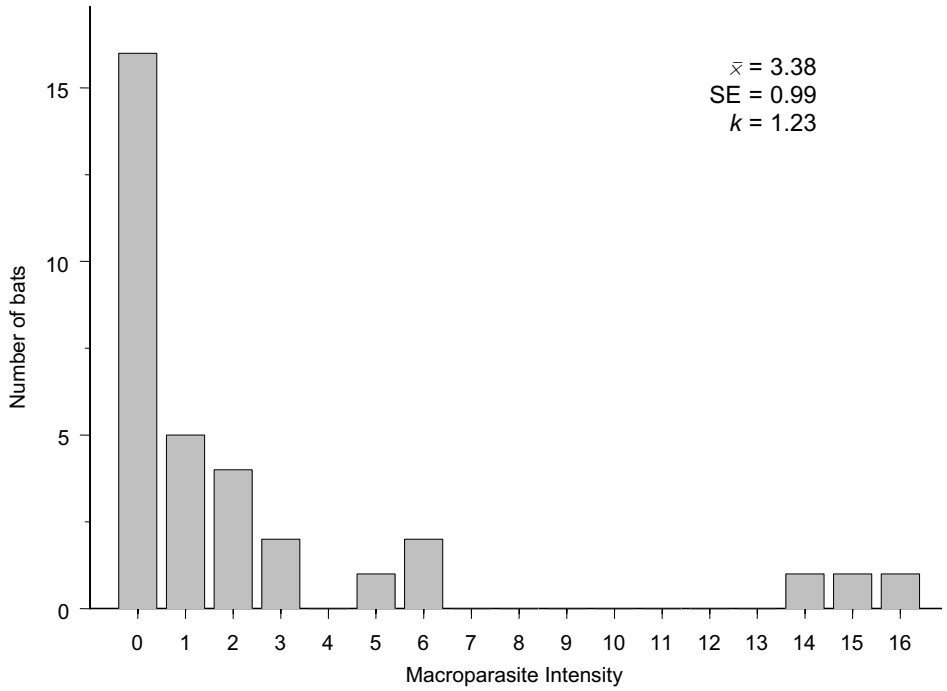

FIG. 1. Frequency distribution of macroparasites (helminths and ectoparasites) in tropical bats of Puerto Rico, following a negative binomial distribution. The mean (x), standard error (SE) and k, of the negative binomial are given in the top right of the figure. Note that the majority of bats have low parasite intensity, whilst relatively few have heavy parasite intensity

We used a GLM, with helminth intensity as the response variable and ectoparasite intensity as the explanatory variable to test for an association between the two groups of parasites. We found a significant negative relationship between the intensity of helminths and ectoparasites (Negative binomial GLM: $\chi_{22} = 6.97$, $P = 0.01$) such that when helminth intensity was high ectoparasite intensity was low and vice versa (Fig. 2).

A significant difference in the mean macroparasite intensity among the bat species occurred ($\chi_{21} = 5.84$, $P < 0.05$), with *M. redmani* hosting highest macroparasite intensity and *P. quadridens* the lowest (Table 1). Using multiple comparisons we observed a significant difference in macroparasite intensity occurred between these two species only. Focusing this analysis on the differences in helminth intensity only (i.e. excluding the ectoparasites) we observed a significant difference between the bat species (Negative binomial GLM: $\chi_{21} = 10.35$, $P < 0.05$, but notably *M. redmani* was helminth-free and therefore there was no variation to test for. Therefore the significant difference in helminth intensity existed between *M. blainvillei* and *P. quadridens* only (Table 1). A greater proportion of the overall macroparasite intensity was a function of the intensity of the ectoparasites, rather than the helminths (Table 1). However, there was no significant difference in ectoparasite intensity among the bat species sampled (Gaussian GLM: $F_{2, 21} = 2.27$, $P \geq 0.05$).

Host body mass had a strong positive relationship with overall macroparasite intensity (Negative binomial GLM: $\chi_{22} = 7.14$, $P < 0.01$ — Fig. 3). The same pattern was seen for ectoparasite intensity (Negative binomial GLM: $\chi_{22} = 7.96$, $P < 0.01$), but not for helminth intensity, where no significance was detected (Negative binomial GLM: $\chi_{22} = 1.67$, $P > 0.05$). Finally, we determined whether there was an association between host sex or body mass and macroparasite intensity. Female bats had a significantly higher macroparasite intensity than males (Negative binomial GLM: $\chi_{22} = 9.10$, $P < 0.01$ — Fig. 4). However, no significant difference was detected between the sexes, for helminths alone (Negative binomial GLM: $\chi_{22} = 3.19$, $P > 0.05$) or ectoparasites alone (Negative binomial GLM: $\chi_{22} = 2.22$, $P \geq 0.05$).

DISCUSSION

The purpose of this study was to identify the macroparasite species associated with a sample of tropical bats from a 'hot cave' in Puerto Rico and to carry out a preliminary investigation of how

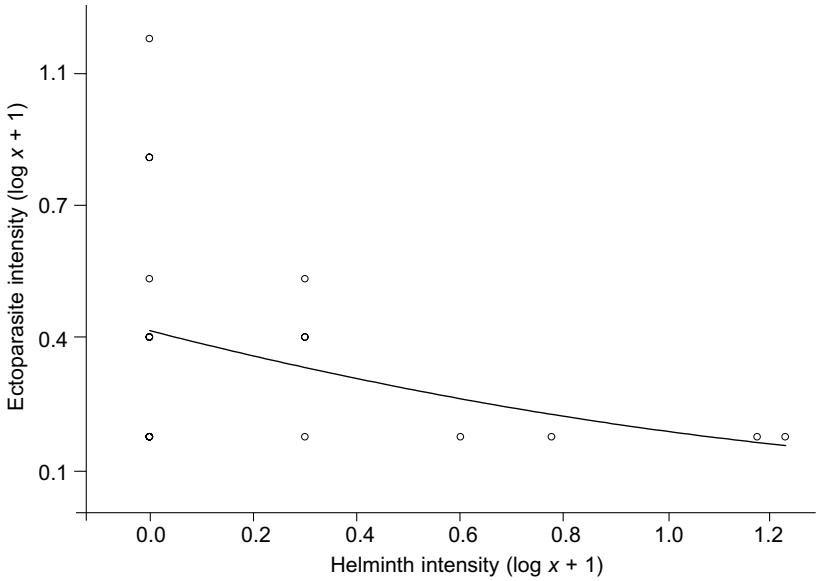

FIG. 2. The association between ectoparasite and helminth intensity (log x + 1) from three species of tropical bats of Puerto Rico. The best fit line is a polynomial regression, described by the equation $y = 0.322 - 0.400x + 0.100^x$. Note a negative relationship between ectoparasite and helminth intensity such that high helminth intensity corresponds with low ectoparasite intensity

macroparasite infection may be related to host factors. We identified two helminths that are commonly found in these bat species, several ectoparasite species that have not previously been described for these bat species (C. Dick, personal communication) and an undescribed species of bat fly, belonging to the genus *Nycterophilia*.

Previously, *M. redmani* has been recorded to host several species of bat fly of the *Trichobius* genus as well as the bat fly *Nycterophilia parnelli* in addition to two Spelaeorhynchidaes and the mites *Jamesonia rosikyi*, *Periglischrus iheringi*, and *Periglischrus vargasi* (Homan and Jones, 1975; Gannon *et al.*, 2005). The only example of these previously recorded macroparasite that we observed on *M. redmani* was a *Periglischrus* mite; however, we also found *Nycterophilia* cf. *coxata* which is a new species record for *M. redmani.* In contrast, six species of

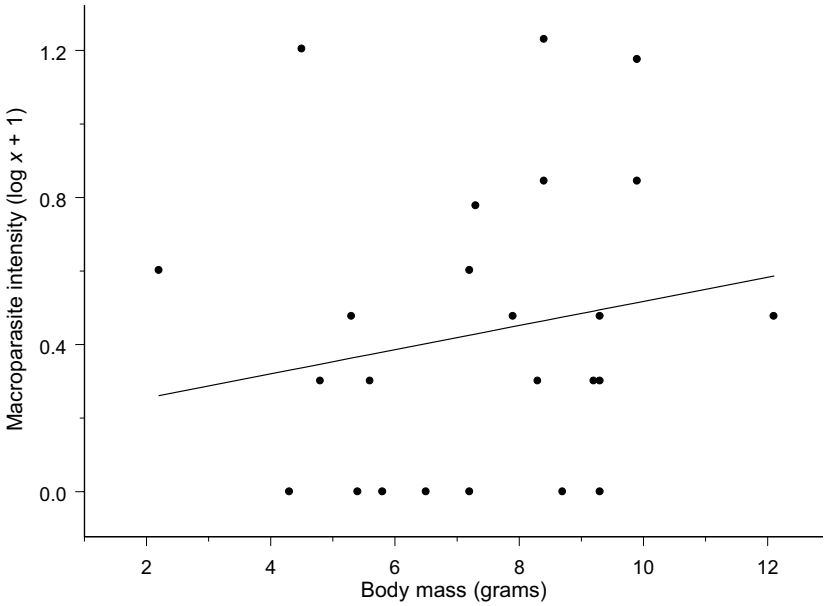

FIG. 3. Regression of (log) mean macroparasite intensity and body mass of three different tropical bats: *M. blainvillei*, *M. redmani*, and *P. quadridens*. The regression line is fit by the equation $y = 0.256 + 0.026x$

mite (*Chiroptomyssus cubnsisk*, *Steatonyssus ceratognatus*, *Cameronieta torrei*, *Eudusbabekia saquei*, *Lawrenceocarpus micropilus* and *Antricola silvai*), and two bat fly parasites (*Trichobius dusbabeki* and *T. frequens*) that have previously been recorded for *P. quadridens* (Peterson and Hůrka, 1974), were not observed on our specimens. *Monophyllus blainvillei* is known to host several arachnid parasites including *Cameronieta standtmanni*, *Antricola silvai*, *Parantricola marginatus*, and *Lawrenceocarpus mormoops* (Lancaster and Kalko, 1996). None of our *M. blainvillei* specimens carried these parasites. The two helminth species in the sample presented here are commonly reported in these bat species (Homan and Jones, 1975; Rodríguez-Durán and Kunz, 1992; Lancaster and Kalko, 1996).

The ectoparasites listed in previously published literature for these bat species were not congruent with those from our specimens. This could be because our sample sizes were small. Additionally parasites are typically aggregated in their host population and a small sample of bats may preclude observations of the most parasitized individuals, so missing certain parasite species (Shaw *et al*., 1998). Conversely, we discovered a new species record — an observation that may have arisen from our sampling method. We used a 'washing method' in that we washed bats over fine meshed sieves to collect what were quasi-microscopic mites, chiggers, springtails and flies, thereby carrying out a more thorough collection procedure as opposed to the typical technique of brushing bats. It is clear that

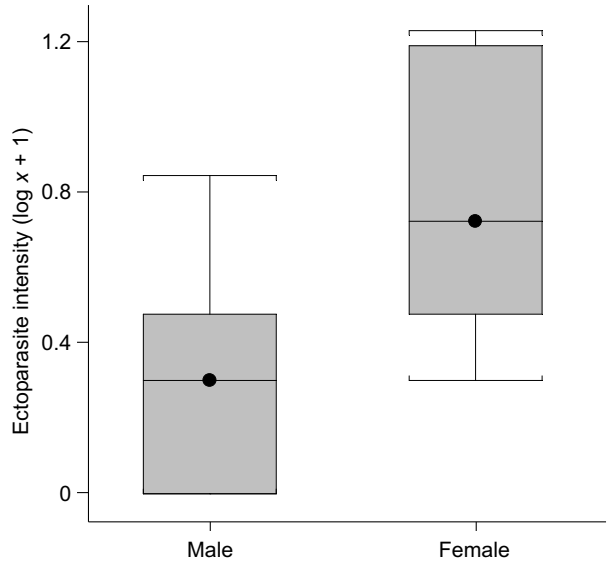

FIG. 4. Box plot of the sex differences in (log) mean macroparasite burden of three different tropical bats species of Puerto Rico: *M. blainvillei*, *M. redmani*, and *P. quadridens*

the parasites recorded here do not consistently match previously published records and indeed this was impetus this research; simply that there have been few investigations into parasites of these hosts.

Monophyllus redmani often roosts with either *M. blainvillei* or *P. quadridens* over 50% of the time in a study of bats in seventeen different hot caves in Puerto Rico (Rodríguez-Durán, 1998) but despite this finding we observed that few ectoparasites were shared between the bat species with the exception of the Periglischrus mites which were shared by all three species of tropical bat. These species differences in the macroparasite community of the bats may occur due to differences not investigated here including bat roosting location in the cave. The cave sampled in this study was a 'hot cave' with three large chambers, each increasing in temperature with progression deeper into the cave. The hotter areas of the cave may support greater numbers of ectoparasites, which are more active than those found in cooler parts. If each bat species inhabits different parts of the cave then we may expect that the ectoparasite intensity and/or macroparasite community will reflect their location in the cave. *Monophyllus redmani* hosted the highest ectoparasite intensity of the three species sampled in this study. This species is an obligate hot cave species (Gannon *et al*., 2005). Indeed this leads us to an interesting hypothesis that observed differences in ectoparasite intensity and the macroparasite species diversity of bats could be related to whether certain bat species are roosting in hot or 'cold' caves and this would be an interesting direction for future research.

We used the data gathered to carry out a preliminary investigation into the ecological characteristics of hosts with respect to macroparasite infection. As is common with host-parasite distributions (Shaw *et al*., 1998) the helminth intensity was highly aggregated in the bat population (k value = 1.23). We found significant differences in the helminth intensity between bat species, which itself was significantly female-biased. A female-bias in infection is contradictory to previous meta-analytic studies that, when also looking for trends across taxa, have found an overall male-bias in infection; a female-bias tends to exist only where the female is the larger of the species (Moore and Wilson, 2002). However, there is no sexual size dimorphism in any of these bat species (Gannon *et al.*, 2005) but a female-bias in parasitism may arise due to trade-offs in immunity versus reproduction efforts and maternal care. In support of this hypothesis in other mammals we find evidence of high energetic costs associated with

pregnancy and maternal care (Festa-Bianchet, 1989) and a mechanistic explanation for increased macroparasite intensity may be explained by these energy trade-offs plus an increased female susceptibility due to the immunosuppressive effects of hormones produced during the parturition and lactation (Dobson and Meagher, 1996).

We observed a negative relationship between ectoparasite and helminth intensity, suggesting a potential trade-off in infection. Much work is starting to emerge in disease ecology on the interaction between the parasite communities of hosts, and putatively, negative correlations may be due to indirect competition between the parasite species or interaction via the hosts' immune response (Bentwich *et al*., 1999, 2000). Again, this is a subject of future research.

Because this study was preliminary in nature, coupled with the difficulty of obtaining specimens from Puerto Rico and the decline of species richness of bats in the tropics, the sample size we had to work with was limited. It should be noted that some of the insignificant results we received may be due to a lack of statistical power associated with small sample sizes. These data represent a preliminary study and point toward future research in this little explored field. Our new species observations highlight how studies of this kind are of particular importance. They allow us to obtain voucher specimens of both bats and their parasites and contribute to the growing pool of knowledge of the parasitic fauna of tropical bats.

Acknowledgements

We are very grateful to Mike Kinsella for identification of helminth samples and to Dr. Carl Dick from the Department of Zoology at the Museum of Natural History in Chicago, IL for identification of the ectoparasites samples from the bats. Finally we extend our gratitude to InterAmerican University for permission to use Mata de Plátano Field Station and allowing access to the cave.

Literature cited

Arneberg, P., A. Skorping, and A. F. Read. 1998. Parasite abundance, body size, life history and the energetic equivalence rule. The American Naturalist, 151: 497–513.

Bentwich, Z., A. Kalinkovich, Z. Weisman, G. Borkow, N. Beyers, and A. D. Beyers. 1999. Can eradication of helminthic infections change the face of AIDS and Tuberculosis? Immunology Today, 20: 485–487.

Benwtich, Z., G. Maartens, D. Tortens, A. A. Lal, and R. B. Lal. 2000. Concurrent infections and HIV pathogenesis. AIDS, 14: 2071–2081.

Calisher, C. H., J. E. Childs, H. E. Field, K. V. Holmes, and T. Schountz. 2006. Bats: important reservoir hosts of emerging viruses. Clinical Microbiology Reviews, 19: 531–545.

Christensen, N. O., P. Nansen, B. O. Fagbemi, and J. Monrad. 1987. Heterologous antagonistic and synergistic interactions between helminths and between helminths and protozoans in concurrent experimental infection of mammalian hosts. Parasitological Research, 73: 387–410.

Cox, F. E. G. 2001. Concomitant infections, parasites and immune responses. Parasitology, 122: 23–S38.

Dobson, A., and M. Meagher. 1996. The population dynamics of brucellosis in the Yellowstone National Park. Ecology, 77: 1026–1036.

Festa-Bianchet, M. 1989. Individual differences, parasites and the costs of reproduction for bighorn ewes (*Ovis canaensis*). Journal of Animal Ecology, 58: 755–795.

Gannon, M. R., and M. R. Willig. 1995. Ecology of ectoparasites from tropical bats. Environmental Entomology, 24: 1495–1503.

Gannon, M. R., A. Kurta, M. Rodríguez-Durán, and M. R. Willig. 2005. Bats of Puerto Rico: an island focus and a Caribbean perspective. Texas Tech University Press, Lubbock, 239 pp.

Homan, J. A., and J. K. Jones. 1975. Monophyllus redmani. Mammalian Species, 57: 1–3.

Lancaster, W. C., and E. K. C. Kalko. 1996. Mormoops blainvillii. Mammalian Species, 544: 1–5.

Moore, S. L., and K. Wilson. 2002. Parasites as a viability cost of sexual selection in natural populations of mammals. Science, 297: 2015–2018.

Perkins, S. E., I. M. Cattadori, V. Tagliapietra, A. P. Rizzoli, and P. J. Hudson. 2003. Empirical evidence for key hosts in persistence of a tick-borne disease. International Journal for Parasitology, 33: 909–917.

Peterson, R. V., and K. Hůrka. 1974. Ten new species of bat flies of the genus *Trichobius* (Diptera: Streblidae). Canadian Entomologist, 106: 1049–1066.

Rodríguez-Durán, A. 1998. Nonrandom aggregations and distribution of cave-dwelling bats in Puerto Rico. Journal of Mammology, 79: 141–146.

Rodríguez-Durán, A., and T. H. Kunz. 1992. Pteronotus quadridens. Mammalian Species, 395: 1–4.

Shaw, D. J., B. T. Grenfell, and A. P. Dobson. 1998. Patterns of macroparasite aggregation in wildlife host populations. Parasitology, 117: 597–610.

Silva-Taboada, G. 1977. Algunos aspectos de la seleccion de habitat en el murcielago *Phyllonycteris poeyi* Gundlach en Peters 1861 (Mammalia: Chiroptera). Poeyana, 168: 1–10. [in Spanish].

Silva-Taboada, G. 1979. Los murciélagos de Cuba. Académia de Ciencias de Cuba, Havana, 423 pp. [in Spanish].

Wilson, D. E., and D. M. Reeder (eds.). 2005. Mammal species of the World: a taxonomic and geographic reference, 3rd edition. The Johns Hopkins University Press, Baltimore, 2142 pp.

Wilson, K., O. N. Bjornstad, A. P. Dobson, S. Merler, G. Poglayen, S. E. Randolph, A. F. Read, and A. Skorping. 2002. Heterogeneities in macroparasite infections: patterns and processes. Pp. 6–44, *in* The ecology of wildlife diseases (P. J. Hudson, A. Rizzoli, B. T. Grenfell, H. Heesterbeek, and A. P. Dobson, eds.). Oxford University Press, New York, 197 pp.

Received 23 June 2008, accepted 25 November 2008