Chapter 6
Parasitic Bat Flies (Diptera: Streblidae and Nycteribiidae): Host Specificity and Potential as Vectors

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Abstract Host specificity gauges the degree to which a parasite occurs in association with host species and is among the most fundamental properties of parasite–host associations. The degree of specificity is indicative of myriad properties of the host and parasite and of their ecological and evolutionary relationships. Bat flies are highly specialized bloodfeeding ectoparasites of bats worldwide and were historically viewed as unspecific. In the bat fly—bat system, numerous properties actually or potentially interrupt the linkage of parasite to host and should thus decrease specificity. Such properties of bat flies include a life history strategy requiring females to leave the host, an off-host pupal stage, and high dispersal capability of many species. For hosts, properties include high diversity, mobility, sociality, and multispecies roosting environments. These and other biological and ecological characteristics of bats and flies should together facilitate interspecific host transfers and over time lead to nonspecific host–parasite associations. Despite these properties, large and carefully executed biodiversity surveys of mammals and parasites unequivocally demonstrate the high host specificity of many bat flies, and molecular sequence data promise to demonstrate that many cases of lowered specificity are misunderstood due to unresolved parasite species boundaries. On the other hand, experimental approaches have suggested that host specificity is context dependent and may be lessened in cases of ecological disturbance and in particular when novel host–parasite associations are created. Evolution and maintenance of specificity in bat flies depends in part on the encounter and compatibility properties of bats and on the reproductive potential of the flies on available host species. Moreover, the degree to which parasites have coevolved immunological compatibility with their hosts, thereby diminishing immunological surveillance and response, may also serve to maintain high host

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specificity. Although worldwide bat species on average harbor higher diversity of parasites and pathogens than any other mammalian group, the likelihood of bat flies vectoring disease agents across host species of bats, and particularly to distantly related mammals such as humans, may be relatively small.

Keywords Bat flies • Host specificity • Ectoparasites • Immunological compatibility • Vector • Streblidae • Nycteribiidae

6.1 Introduction

Host specificity is perhaps the most fundamental emergent property of any parasite–host association and is a measure that gauges the degree to which a particular parasite species is limited to, or occurs in strict association with, a particular host species (Poulin 1998a). Parasitic organisms vary in the degree to which they spend time in or on their host, and the degree to which they are found in association with one or more species of host (Marshall 1981). One on extreme, when a parasite species is associated strictly with a single host species, it is understood to be highly specific. Conversely, when a parasite species is associated with a variety of unrelated host taxa, it clearly exhibits low specificity. As a general rule, permanent parasites that spend their entire life cycle on or in a particular host tend to be more specific, whereas temporary parasites that spend little time coupled with their host are less specific (Price 1980). In reality, however, parasitic organisms span a broad spectrum of host specificity. To capture this phenomenon in a more quantitative sense in relation to host taxonomy, Wenzel et al. (1966) introduced the following terms: Monoxenous parasites are restricted to a single host species. Stenoxenous parasites are restricted to a narrow range of closely related hosts (e.g., congeneric hosts). Oligoxenous parasites are restricted to a broader taxonomic range of hosts (e.g., confamilial hosts). Finally, polyxenous ("heteroxenous" sensu Jobling 1949) parasites are not host taxon restricted and occur on a variety of unrelated hosts (Wenzel et al. 1966).

As captured by Wenzel et al. (1966), host specificity is more than a function of the number of host species associations per parasite species but explicitly refers to the host's evolutionary (and taxonomic) associations. It is therefore an attempt to meaningfully incorporate the degree of host relatedness into measures of specificity. Various other measures of specificity (i.e., S_{TD} sensu Poulin and Mouillot 2003) attempt to account for host relatedness. In particular, S_{TD} measures the average taxonomic distinctness of all host species associated with a particular parasite species. While such a measure attempts to capture taxonomic or phylogenetic distinctness, it does not capture ecological distinctness of hosts (Poulin and Mouillot 2003). The degree to which parasites are specific to hosts depends not only on the number of host species or their evolutionary distinctness but also on myriad ecological factors including the parasites' life history strategy, dispersal characteristics, host mobility and social biology, and on dynamics of the nesting or roosting environment of the host (Dick and Patterson 2006; Patterson et al. 2007; Dittmar et al. 2009; Dick and Miller 2010). Finally, there is a strong and intractable relationship between any parasite species' degree of host specificity and the likelihood of that parasite switching to a novel host. Given that many parasites vector pathogens, host-switching is an important means of new host recruitment for parasites and infectious agents alike. This chapter provides an overview of historical and current understanding of the degree of host specificity for a prominent group of bat parasites (Diptera: Nycteribiidae and Streblidae), of the dynamics between bats and these parasites, and finally, the likelihood with which bat flies may be able to move pathogens within and among host species.

6.2 Biology of Bat Flies

6.2.1 Overview

Bat flies (Diptera: Hippoboscoidea) are highly specialized ectoparasites and only associate with bats (Mammalia: Chiroptera) (Figs. 6.1–6.6). They live in the fur and on the wing membranes where they feed on host blood. During the course of their life cycle, however, they necessarily decouple from their host for periods of time, including during female deposition of a prepupae on the roost substrate, and during the nonmotile pupal stage itself. Bat flies are nominally divided into two cosmopolitan families, Streblidae and Nycteribiidae, but an emerging phylogenetic consensus suggests that while bat flies are monophyletic, the families themselves are not natural groups (Dittmar et al. 2006; Petersen et al. 2007). Both families reach the zenith of their diversity within tropical latitudes, are less diverse in the subtropics, and rather impoverished in temperate regions. However, this latitudinal richness gradient appears more pronounced in the Western Hemisphere. Nycteribiids (ca. 275 species) are more speciose in the Eastern Hemisphere, whereas the streblids (ca. 230 species) are richer in the Western Hemisphere (Dick and Patterson 2006).

6.2.2 Life History

Relatively little is known regarding life history and reproductive biology of bat flies, and what is known is based on a very limited number of studies involving only a few species. Nonetheless, some generalizations likely apply to all bat flies. Hippoboscoid Diptera (including bat flies) reproduce via viviparous puparity (Hagan 1951). Eggs are fertilized inside the female, and the three larval stages develop within the female, likely nourished by intrauterine "milk" glands. When internal development is complete, female flies leave the host bat, seek and find a

Fig. 6.1 *Archinycteribia octophthalma* Theodor (Nycteribiidae: Archinycteribiinae) from *Penthetor lucasi*, female, dorsal view (Sarawak)

Fig. 6.2 *Cyclopodia inclita* Falcoz (Nycteribiidae: Cyclopodiinae) from *Pteropus tonganus*, male, dorsal view (Fiji)

suitable deposition site (typically, within the confines of the bat's roosting environment), and deposit a single, terminal (3rd instar) larva on this substrate. Once deposited, the larva ("prepupa") immediately forms a puparium. Following a pupal stage ca. 3–4 weeks in duration (Ching and Marshall 1968), an unfed (teneral) adult fly emerges and must locate and colonize a host. Ching and Marshall (1968) reported prepupal deposition directly on the host's body. But in these atypical cases, the pupae were distorted in shape and most failed to develop into an adult fly or were removed by host grooming.

The most thorough treatment of the life history of a bat fly species is Overal's (1980) study of the life cycle of *Megistopoda aranea* (Streblidae) in Panama. In this species, prepupae were deposited in the roost, but usually in close proximity of bats (*Megistopoda* spp. are incapable of flight). Following an average of 23 days, the adult fly emerged from the puparium and located a host for a blood meal before

Fig. 6.3 *Nycterophilia* n. sp. (Streblidae: Nycterophiliinae) from *Brachyphylla cavernarum*, female, lateral view (St. John, US Virgin Islands)

Fig. 6.4 *Megistopoda aranea* (Coquillett) (Streblidae: Trichobiinae) from *Artibeus jamaicensis*, male, lateral view (Panama)

mating. Postpartum females were observed mating within minutes after depositing a prepupa. The time interval between successive depositions of prepupae was about 10 days. Another detailed study was conducted by Fritz (1983), on the Costa Rican species *Trichobius joblingi*, *Speiseria ambigua*, and *Strebla guajiro* (all Streblidae). These species also deposited prepupae on roost substrates away from the host, with the pupal stage duration of 17, 20, and 19 days for these species, respectively. Recently, Dittmar et al. (2009, 2011) confirmed prepupal deposition away from the roost for other *Trichobius* spp. (Mexico, Puerto Rico), as well as *Nycterophilia* spp. (Puerto Rico). A few studies also have been conducted on temperate (North American) streblid species. Generally, life histories of temperate species mirrored those in tropical regions. Unlike tropical bats, however, temperate bats hibernate during winter months, and bat flies remained physically and reproductively active on hibernating bats whose body temperatures ranged from 2 to 6 °C (Ross 1960; Reisen et al. 1976; Caire and Hornuff 1986).

Fig. 6.5 *Aspidoptera phyllostomatis* (Perty) (Streblidae: Trichobiinae) from *Artibeus planirostris*, female, dorsal view (Ecuador)

Fig. 6.6 *Megastrebla gigantea* (Speiser) (Streblidae: Brachytarsininae) from unknown host, female, dorsal view (Philippines)

The life cycle of *Basilia hispida* (Nycteribiidae) has been described in some detail by Marshall (1970). In this species, flies reached sexual maturity 5–6 days after emergence from the puparium. Like that observed in the streblid *Megistopoda aranea*, male *B. hispida* often not only copulated with postpartum females immediately following prepupal deposition but also were observed copulating with newly emerged and unfed (teneral) females. A single copulation was sufficient to produce several offspring, suggesting that female *B. hispida* may store sperm. At intervals of ca. 9 days, mature females deposited prepupae on roost substrate, which were depressed onto the substrate by use of their abdomen. Prepupal deposition occurred between 0900 and 1800 h, apparently stimulated in part by temperatures elevated by the presence of bats in the roost. Development of pupae depended upon whether bats were present or absent. When bats were present, teneral adults emerged as soon as 25 days after deposition. When bats were absent or their presence sporadic, pupal development was nearly twice as long (up to 46 days). Newly emerged adults began walking about (all nycteribiids are wingless) until encountering a host. Upon colonization of a host, teneral flies began feeding within 20 min. The total life span of *B. hispida* averaged 136 and 195 days for males and females, respectively, with 5 days prepartum, 9 days in the larval stages, 25 days in the pupal stage, and 97 days (males) or 156 days (females) in the adult stage (Marshall 1970). When experimentally removed from hosts, *B. hispida* died within 5–25 h; mature females were less resistant after removal from the host than were teneral flies.

The breeding biology of another nycteribiid, *Eucampsipoda sundaica*, was described by Ching and Marshall (1968). Most features of the life cycle resembled that of *B. hispida*, but during mating, males attached to females for up to ½ h, during which time the female remained fully ambulatory. The interval between successive prepupal depositions was three times faster in *E. sundaica* than *B. hispida* (3 versus 9 days, respectively) (Ching and Marshall 1968). Peterson and Wenzel (1987) suggested that the life cycle of bat flies was rather uniform across species. In general, the life history strategy of bat flies appears to reflect their obligate association with bats, where vulnerable immature stages generally remain coupled with the host inside the female fly, and completion of the life cycle, constrained by the relatively vulnerable pupal stage, requires a somewhat stable roosting environment away from the risks of host grooming (Patterson et al. 2007).

6.2.3 Host as Habitat

One prevailing or defining characteristic of parasitism is that the parasite is to some degree dependent upon their host. As such, the host itself is often considered to be the "habitat" for the parasite. The host's body itself provides a broad array of environmental stimuli and resources to the parasite and plays a major role in shaping the life history, ecology, and behavior of the parasite. Moreover, the habitat affects dynamics of parasite populations, assemblages, or community-level interactions among multiple parasite species (Patterson et al. 2008a, b; Tello et al. 2008; Dick and Miller 2010).

Certain parasites are highly dependent on habitat variables such as host body size and correlated characters. This is known as "Harrison's Rule" (Eichler 1963) and states that size of parasites and hosts are positively correlated. This rule is evident in highly habitat-specific parasites such as bird wing lice (Phthiraptera), where lice transferred to larger or smaller hosts experience significant reduction in fitness (Bush and Clayton 2006). Although bat flies are not as host microhabitat specific as bird wing lice, they do exhibit a degree of general habitat specificity, living on either the wing and tail membranes, or in the fur, of the bats that they infest (Dick and Patterson 2006). In these general habitats, they appear to feed in a similar manner but tend to conform to three general morphological forms (one adapted to patagia and two adapted to fur) that are related to different evasion strategies to counter-effect host grooming efforts. Although such specificity to particular host macrohabitats may help maintain some degree of specificity, across host taxa these broad categories (e.g., patagia and haired regions) of macrohabitat should not prevent cross-host switching of flies that specialize on particular macrohabitats. The nature and specificity of parasitic associations is strongly influenced by the ecology and behavior of both host and parasite (Brooks and McLennan 1993; Marshall 1981; Poulin 1998a).

6.2.4 Roost as Habitat

Variation in parasitic life history traits and host specificity may result from the capacity of the parasite to perceive and respond to a much larger set of environmental variables than those presented strictly by the host itself (Heeb et al. 2000). In addition to the behavior and ecology of both associates, environmental features shape the dynamics of parasite fitness. The greater environment of parasites has dimensions and linkages that extend from its immediate environment such as the host itself to the surrounding ecosystem in which the host lives (Pilosof et al. 2012). Many parasitological studies have focused on the "host as habitat" and have largely ignored the greater nesting or roosting environment of hosts. While a plethora of studies document and explore the former, the latter is still poorly understood and documented. For example, studies of fleas have highlighted instances where parasitism varied with soils and other habitat variables (Krasnov et al. 1997, 1998) and where such relationships are more strongly associated with host species than with habitat variables (Krasnov et al. 2006). Flea species, however, vary tremendously in the permanence of their coupling with hosts (Medvedev and Krasnov 2006; Whiting et al. 2008).

The dimensionality of the off-host environment varies greatly with the degree to which host and parasite remain in direct physical contact throughout their life cycles. For example, all phases of the life cycle of bird and mammal chewing lice take place strictly on a host individual. Lice have drastically reduced visual systems, are relatively immobile off the host, and typically exhibit high fidelity to individual host species (Clayton et al. 2004). These characteristics often correspond with observations of parallel clade formation, or cospeciation (Hafner and Page 1995; Hafner et al. 1998; Reed et al. 2007; Light and Reed 2009). Ixodid ticks, conversely, spend up to 90 % of their lives off-host and most species parasitize a broad range of vertebrate hosts (Klompen et al. 1996; Poulin 1998b). When such a parasite's life cycle interrupts the linkage with a host, its potential for ecological proliferation and coevolution is limited. Bat flies, although obligate ectoparasites, are holometabolous and must metamorphose in order to complete their development. Given that pupation takes place off the host, in the bat's roost, and lasts a few weeks, there is a necessary decoupling of parasite and host. This means that bat flies spend possibly up to a third of their adult life span off the host and in the immediate roosting environment, mainly as pupae (Dick and Patterson 2006). In order to be specific to a particular host species, newly emergent flies must locate not just any bat but a bat of a particular species.

Roosting habits of bat species vary greatly in relation to bat fly development. Although colony size, composition, occupancy, and interbat spacing may all be expected to influence parasitism, of particular importance is the structure of roosts themselves. The types of roosts that bats choose span remarkable ecological amplitude. Different species seek shelter in a variety of roosts, including leaf tents, tree foliage, bark, and cavities, termite nests, rock crevices, houses, mines, culverts, and caves (Kunz and Lumsden 2003). Considering this variety of roosting structures, they vary in durability from very short lived to permanent. For example, while foliage-type roosts offer abundant and ubiquitous roosting locations, they tend to be ephemeral and bats spend only a few days or weeks in a given roost (Timm 1987). At the other extreme roosts such as caves or mines, while often uncommon and patchily distributed, represent somewhat permanent roosting spaces that can be utilized by many successive generations of bats (Kunz 1982). Roosts also vary in the amount of exposure and protection they offer roosting bats, in a manner that should affect the roost fidelity of bats: foliage and leaf tents are relatively exposed and open, while caves mines are enclosed and protected.

The roosting biology of bats thus influences the quality and quantity of parasitism by bat flies (Lewis 1995; Marshall 1981; ter Hofstede and Fenton 2005; Reckardt and Kerth 2006; Patterson et al. 2007), which in turn influences the likelihood of vectoring pathogens. Because bat flies pupate in the roost itself, the duration of bat residency at roosts determines to a great degree the likelihood that newly emerged flies are reassociated with the original host population of their parents. Although many factors affect roost-site fidelity in bats (Lewis 1995), durability must play an important role. Both ecological and coevolutionary associations of bats and bat flies thus depend on the fly's ability to place pupae in a location conducive to emergent offspring quickly and reliably reassociating with individuals of the host species (Dittmar et al. 2011). Roost fidelity of bats also varies according to the durability of its roost and the shelter it affords from predation and adverse weather (Kunz 1982; Kunz and Lumsden 2003). In this context, it has been shown that the number of bat fly species parasitizing a bat, the proportion of host individuals parasitized, and the number of parasites per infested host all increase with the permanence and protection of bat roosts (Patterson et al. 2007). However, when investigating the mobility of bat flies in relation to the aforementioned variables, it was found that dispersal ability as measured by the presence or absence of functional wings did not affect evolutionary associates, or the prevalence and intensity of parasitization by bat flies (Patterson et al. 2007). Although such variables have not been assessed in the light of host specificity among bat flies, we expect this measure to be unrelated also to dispersal ability of the parasites.

6.2.5 Historical View of Host Specificity

The degree to which nycteribiid and streblid bat flies are host specific has long been debated (Jobling 1949; Theodor 1957; Wenzel et al. 1966; Marshall 1981; Poulin 1998a; ter Hofstede et al. 2004). Early studies (Jobling 1949; Theodor 1957) insisted that bat flies are not particularly host specific. At the time, only 9 of the 36 known species of New World streblid flies were thought of as strictly monoxenous and even then, only one species was documented sufficiently to confirm its level of specificity (Jobling 1949). Thus, monoxeny of the majority of those nine species was doubtful. In an apparent nod to the importance of roosting dynamics, lack of specificity was explained due to the fact that many species of bats often roost together, giving flies ample opportunity to parasitize multiple host species (Jobling 1949). Close and consistent proximity of host bats should increase the likelihood of exchange of parasites among different bat taxa, where bat flies would effectively utilize multiple taxa as one large host metapopulation. Jobling (1949) noted that host preference was distinct among some fly species, and he forwarded the concept of the “principal host” to describe the observed preference of certain flies for certain host species. However, it was claimed that some fly species had “so completely adapted” to different species and genera of bats that it was impossible to ascertain a principal host (Jobling 1949).

It has become increasingly clear that historical treatments of host specificity among bat flies were hampered by several factors, including (1) limited collection records, (2) poor identifications (and limited taxonomic understanding) of both parasites and hosts, (3) lack of voucher collections of both parasites and hosts, and (4) poorly controlled collection protocols to minimize interhost contamination. It has been argued that the single most important requirement for determining host specificity is the systematic assembly of large, geographically representative collections (Wenzel et al. 1966). Indeed, early assessments of bat fly specificity primarily were based on haphazardly accumulated museum collections, gathered over long periods of time and by numerous workers whose foci often were not on issues regarding host specificity (Marshall 1976). During the initial phase of one large survey (Panama; Wenzel et al. 1966), field workers routinely placed bat specimens of multiple species into the same receptacle, and only later would these hosts be sampled for ectoparasites. Moreover, surveys routinely employed workers not properly trained in collection of ectoparasites (R. L. Wenzel, personal communication).

Nonetheless, the early studies set the foundation for our understanding of host specificity. Based on a sample of 12,000 Panamanian streblid flies, Wenzel et al. (1966) found that about 55 % of the 66 streblid species were associated with a single host species (monoxenous sensu stricto), and that an additional 15 % were monoxenous (sensu *lato*), known rarely to "stray" onto ecologically associated hosts such as roost associates. Of the remaining 30 %, about half were oligoxenous and the remaining 15 % were polyxenous. A decade later, a very large and systematic collection effort was undertaken in Venezuela (Smithsonian Venezuelan Project), resulting in ca. 36,000 streblid specimens. Wenzel (1976) treated 116 Venezuelan bat fly species and assessed host specificity. Seventy-five percent of those species were strictly monoxenous, while the remaining 25 % were steno-, oligo-, or polyxenous. In Malaysia, Marshall (1980) reported that 72 % and 64 % of streblids and nycteribiids, respectively, were recorded from a single host, with the remaining fly species restricted to sets of congeneric species (stenoxenous). It was during this time and based on the results of very large collections that workers on bat flies began to refute claims of low specificity forwarded previously by Jobling (1949) and Theodor (1957).

6.3 Contemporary View of Host Specificity

6.3.1 Field Surveys and Natural History Collections

In general and as collection data have grown in both quality and quantity, the trend has been one of an increasing recognition of host specificity for bat flies (Marshall 1981; Hutson 1984; ter Hofstede et al. 2004; Dick 2007; Dick and Patterson 2007). In the past couple of decades, surveys of mammals and ectoparasites have been designed and executed specifically to assess host specificity (among other things) and with an eye to reducing potential for cross-host contamination of parasite samples (Presley 2004; Dick and Gettinger 2005). Such survey protocols mandated strict isolation of each host individual and that cleanliness steps are taken to enhance our ability to accurately discern patterns in host–parasite specificity. Nevertheless, our knowledge of specificity among ectoparasites largely depends on mammal–ectoparasite surveys that are broad in scope and varied in objective. Given the nature of such surveys, literature based on them often contains dubious host–parasite associations (e.g., Wenzel et al. 1966; Wenzel 1976; Autino et al. 1999; Komeno and Linhares 1999; Graciolli and de Carvalho 2001; Dick and Gettinger 2005).

Regarding sampling and host specificity, increased acknowledgment of the reality and likelihood of contamination invoked the use of terms such as "nonprimary" or "transitory" hosts (e.g., Wenzel et al. 1966; Wenzel 1976). Often, such nonprimary or transitory associations were assumed and the cause of such associations not pursued further. They were explained as: (1) rare but natural host transfers, (2) disturbance (accidental) transfers made while the host or parasite was being sampled, or (3) contamination from parasite sampling events (Wenzel et al. 1966). Usually no attempt was made to discriminate among the possible causes of such associations. Moreover, other workers sometimes ignored these issues and listed indiscriminately any parasite species reported from a host species (e.g., Guerrero 1997). Even when host–parasite associations are deemed nonprimary, such assessments may be based on raw percentages of flies on hosts (e.g., Wenzel 1976). Statistical assessments of host specificity of ectoparasites remain rare. While host-specificity indices have been proposed (Miles et al. 1957; Stark and Kinney 1969; Rohde 1980), they all can be problematic and unreliable, particularly in cases where parasite species are monoxenous and others stenoxenous and oligoxenous (Poulin 1998a). Understanding the nature of host–parasite associations depends upon the accuracy of host specificity appraisals. The degree to which parasite species are specific greatly informs studies of host–parasite ecology, evolution, and cospeciation, and as such, data regarding host specificity and interhost contamination of parasites must be addressed quantitatively. Quantitative assessments of host specificity would allow the use of statistical tests to determine the likelihood that observed associations between host and parasite are ecological phenomena or whether they could occur by chance. Moreover, insights into historical patterns and processes, most notably phylogenetically framed ones, are obscured when reliable and accurate estimates of host specificity under natural conditions are unavailable (Brooks and McLennan 1993).

Two general scenarios exist to explain the association of a given parasite species and a given host species. First, the parasite may have intrinsically come to be associated with that host (e.g., by colonization of that particular host after emergence from a puparium), or by vertical transfer in a cophyletic sense (e.g., the parasite is associated with a particular host because its mother is associated with that host). Alternatively, it may be in association with that host species due to human influence of some kind (i.e., it was contaminated onto a host via inadequate collection methods, or made the host switch under its own power but while it or its host was under duress upon collection). Discriminating between these two alternative explanations (natural vs. human mediated) is the critical first step to understanding host–parasite specificity. Furthermore, determination of whether nonprimary host associations are natural or a result of human intervention can identify the cases when parasites are inclined to switch hosts naturally (see Sect. 6.3.4) and direct future studies of the ecology and evolution of host–parasite associations.

In a recent survey specially designed to eliminate cross-host contamination of parasites, Dick and Gettinger (2005) determined that >99 % of nearly 2,500 fly individuals sampled in Paraguay were associated with primary host species. Using information on primary and nonprimary host captures per day, and assuming flies could not become contaminated on days they were not collected, all but one of the

15 mismatches were shown to have resulted from human error upon sampling contamination (Dick 2007). In other words, naturally caused host-switching appeared to be rare indeed. Accounting for these cases of human error, the level of host specificity observed for the Paraguay collection was higher than that in any other study published to date on bat flies. In that study, 27 of the 31 bat fly species (87 %) were strictly monoxenous. Had the nonprimary associations observed during the Paraguay study been mistakenly reported as primary associations, the observed degree of monoxeny would have been only 71 %.

In addition to sampling contamination, another cause of nonspecific associations is disturbance transfer. Disturbance transfers are difficult to control, avoid, and assess. The vast majority of bat captures involves the use of harp traps, mist nets, and handheld loop nets (Kunz et al. 2009). Typically, nets are deployed along trails, streams, and other flyways frequented by bats (Kunz and Kurta 1988). When a bat is captured, it experiences some degree of physiological stress and will often struggle to escape. Moreover, bats of different species are often captured simultaneously (particularly in harp traps) and will be in close proximity for a period of time. Bat flies are sensitive to the stress experienced by the bat, and often become agitated and leave the host during this time (R. L. Wenzel, personal communication; personal observation). Particularly among winged bat fly species (~75 % of New World streblid species are capable of flight; Whitaker et al. 2009), there is an increased likelihood of disturbance transfers from one bat species to another. Finally, when field workers handle and remove bats from nets or traps, the bats become further stressed and the ectoparasites further disturbed. Flies commonly move onto the hands of bat handlers, or fly toward the handler's light source. As it is with highly host-specific gopher lice (Hafner et al. 2003), dispersal capability was long held to be an important factor in the degree of host specificity, in bat flies with flightless species being more host specific, and winged species being less specific (Jobling 1949). In general, flightless species only able to crawl from host to host should be more host limited than volant species that may easily move from host to host over great distances. However, nycteribiids (which are all wingless) appear to be no more host specific than streblids (of which ~25 % of the species are incapable of flight) (Marshall 1980). Marshall (1976) regarded both families of bat flies as being "host specific" with species overwhelmingly monoxenous but occasionally stenoxenous. Recently, ter Hofstede et al. (2004) suggested that mobility has no effect on the degree of host specificity among Belizean bat flies. All said, these dynamics serve to further obscure collections-based assessments of host specificity because they serve to increase the likelihood of unnatural transfers of what otherwise may be host-specific parasites. Frequent or even constant monitoring of mist nets and harp traps may reduce the likelihood of disturbance transfers but cannot eliminate them entirely.

Recent collections from Ecuador (unpublished data) provide some additional evidence that host specificity among bat flies is high, particularly when assessed under controlled conditions. Based on information collected from >2,000 bats of 77 species and >6,000 bat flies of 97 species, criteria for bat fly sample size (20 or more per fly species) resulted in 42 fly species of sufficient number. For the multiyear and multi-investigator collections countrywide, 17 fly species were strictly monoxenous, 22 were stenoxenous, and three were polyxenous. In other words, ~93 % of well-sampled Ecuadorian bat flies parasitize a single host species or no more than a few closely related host species. To illustrate the importance of standardization and strict control of collection protocol, the Ecuadorian data was divided into two sets. One set comprised all data known from collecting efforts in Ecuador, which included all samples from multiple collection events over multiple years in Ecuador, some of which are old and employed little if any control over sampling. These data were collected by numerous individuals, who lacked parasitological training, and who were less likely to be aware of the necessity for sampling protocols to ensure true primary host associations were determined. The second set is a subset of the total data that included only samples from one large but recent collection (Sowell Expedition) where all of the sampling protocols were known, several appropriate measures were taken to avoid sampling error, and all samples were collected by only two people (CWD and a student under direct supervision). This subset of the data includes more than ½ of the total dataset. Using the same sampling parameters described above, 30 bat fly species met the criteria, and a picture of even greater host specificity emerges. It was found that out of 30 bat fly species; 17 were strictly monoxenous, while 13 were stenoxenous. None were observed to be oligo- or polyxenous. These results strongly indicate that Ecuadorian bat fly species are quite specific to individual species or genera of bats and are consistent with emerging consensus of high host specificity among bat flies.

Clearly, well-executed collections of ectoparasites that minimize interhost contamination, and the statistical evaluation of nonprimary host associations, both contribute to the increased understanding of high host specificity among bat flies. Typically, realistically high levels of specificity are observed only when (1) large enough samples of hosts and parasites are available, (2) sampling protocols strictly control for interhost sampling contamination, and (3) instances of nonprimary host associations are evaluated statistically.

6.3.2 Experimental and Molecular Assessments

Although survey and collection-based assessments of specificity reveal high host specificity among bat flies, molecular and experimental assessments have the potential to obscure and complicate such an understanding. In one of the few published experimental studies of streblid bat flies and their choice of hosts, cafeteria-choice arenas were established to allow flies to choose among alternative host options (Dick and Dick 2006). Results of this study revealed strong preference of flies for host bats that varied in quantity and quality of previous infestation. Similar results were obtained in an unpublished study by Dittmar (in 2009) for the nycteribiid *Leptocyclopodia* sp. in a host choice experiment between the bat species *Cynopterus brachyotis* and *Pteronotus jagori* in the Philippines. In a more natural setting, specificity of bat flies may also depend in part on whether alternative host species live allopatrically or sympatrically. In eight caves in Sri Lanka, Seneviratne et al. (2009) investigated specificity of bat flies parasitizing *Rousettus leschenaulti*, *Rhinolophus rouxi*, and *Hipposideros speoris*. In two caves, the bat species were sympatric, but in six other caves (2, 2, and 2, respectively) the bat species roosted alone. Generally, local monoxeny was reported for seven of eight species of nycteribiid, and for five of six species of streblid. However, certain bat fly species were five times more likely to show strong host preferences when host species were roosting together, whereas less specificity was evinced on host species roosting alone (Seneviratne et al. 2009). These preferences were evident in both nycteribiids and streblids and may reflect strong selection for fly fitness (higher fitness on specific hosts, but lower fitness on less-preferred hosts when only that host is available). Such selection may be driven by competition among parasite species or by differential nutritive properties of the host blood. Laboratory experiments with parasite transfers, however, highlight the distinct possibility of parasite establishment on novel hosts in the absence of the various ecological and evolutionary dynamics typically preventing encounter and establishment (Perlman and Jaenike 2003; Bush and Clayton 2006). This trend is important in light of bat flies as pathogen vectors, and the likelihood at which new emerging infectious diseases may spillover to novel hosts; to the degree to which environmental disturbance and/or novel host–host contact occurs, the potential with which parasites may spill over and establish on novel hosts increases.

While experimental studies, particularly those done in the laboratory, suggest that host specificity is not evolutionarily “fixed”, in some cases molecular scrutiny attests to even higher specificity than is seen through field survey data alone (Poulin and Morand 2005). In fact, the existence of morphologically cryptic species diversity is being increasingly discovered in what were previously understood to be less-than-monoxenous parasites (Whiteman et al. 2006; Smith et al. 2006; Sect. 6.3.4). Such assessment has not been done to date with bat flies, but the existence of widespread cryptic species speaks to the potential for currently understood steno-, oligo-, or polyxenous bat fly species to actually comprise multiple monoxenous species that are currently indistinguishable morphologically. Such a situation, if it comes to be known, would greatly increase the operational host specificity of bat flies and further decrease their likelihood to transmit pathogens across host species boundaries.

6.3.3 Maintenance of Specificity

Many factors are at play with respect to the cause of host specificity, which includes both the evolution and maintenance of specificity in parasites. Various proximate factors have been identified, such as nutritive and immunological compatibility of blood biochemistry, sensory cues that allow a parasite to recognize a specific host (e.g., olfactory and thermal cues emanating from the host and the greater environment), or the morphological adaptations of parasites that facilitate physical establishment and maintenance on the host's body landscape. Ultimate causes of host specificity include the ability to find and utilize food and mates, evolutionary constraints upon vertically transmitted parasite taxa that have cospeciated with their hosts, or any other suite of factors that increase the reproductive fitness of the parasite over evolutionary time.

The evolution of specificity in parasites centers on the dynamics of parasite dispersal, host proximity availing parasite establishment, and adaptation of the parasites themselves (Timms and Read 1999). As a lineage of mammals, bats harbor an incredible array of arthropod parasite diversity (e.g., Acari [mites and ticks], Diptera, Dermaptera, Hemiptera, and Siphonaptera) and particularly for parasite diversity of higher taxonomic rank (e.g., order and family) appear to harbor more parasite diversity than any other group of mammals (Wenzel and Tipton 1966). As mentioned above, the roosting environment of bats plays a profound role in shaping dynamics of parasitism. Throughout the evolutionary history of bats and their potential parasites, surely the utilization of relatively stable and long-lived roosting sites has facilitated many independent transitions to parasitism among arthropods associated with bats. Long-lived and dependable roost sites, coupled with copious and steady supplies of organic matter in the form of guano and dead carcasses, as well as presence of the bats themselves, has facilitated the evolution of parasitism among roost-associated arthropod groups (e.g., Siphonaptera). Such roosting dynamics would have facilitated arthropod dispersal to bats and increased encounter rates, and the duration of these roosting sites would have allowed strong selection to shape the myriad adaptations among bat parasites that we observe today.

As a general rule, host-specific parasites typically are limited in their dispersal capacity (e.g., lice on solitary fossorial mammals; Hafner et al. 2003) or are morphologically, behaviorally, or physiologically adapted to their host to such a degree that survival on a novel host may be severely diminished (Tompkins and Clayton 1999; Bush and Clayton 2006). Thus, the evolution and maintenance of host specificity among bat flies is probably fueled by selective compromise. On the one hand, additional potential host species represent additional resources for population establishment, and exploiting them should increase both abundance and overall fitness of the parasite (Poulin 1998a). On the other hand, broad exploitation strategies would expose such a parasite to a larger array of physical and immunological challenges as well as potential competitors for niche space, in turn diminishing such a parasite's ability to fine tune its exploitation to the ecology and behavior of any one primary host species. Given the dispersal capabilities of bat flies and the social structure of their bat hosts, specificity in bat flies is likely adaptive rather than being produced by intrinsic dispersal limitations (Dick and Patterson 2007).

The evolution of parasite life cycles and scenarios for why a particular parasite species might be present or absent on a particular host species was outlined by Combes (1991). This framework later became known as the filter concept (Poulin 1998b). The paradigm has direct application to the evolution and maintenance of host specificity in bat flies. Combes (1991) described two filters that independently constrain potential parasites from establishing on a host species. First, the encounter filter serves to exclude potential hosts the parasite cannot locate, encounter, or colonize for behavioral or ecological reasons. For bat parasites, for example, reliable and long-lived roosting environments would serve to make the encounter filter porous by facilitating likelihood of encounter and colonization of bats by potential parasites. The encounter filter embodies parasite dispersal limitations described by Timms and Read (1999). The second filter, the compatibility filter, represents another threshold in the evolution of parasitism, by excluding all host individuals on which the parasite cannot survive for morphological, physiological, or immunological reasons (Combes 1991). The compatibility filter embodies parasite adaptive limitations (Timms and Read 1999). Together, the encounter and compatibility filters would each have to be passed before parasite populations were established, and before any degree of host specificity can be reached (Combes 1991). The filters constrain the pool of potential parasite taxa.

As such, the filter concept is largely host centric (e.g., the host must be encountered, and the host must be compatible) and may not account for characteristics intrinsic to the parasite or potential parasite. In any parasite, one factor of crucial importance to evolutionary fitness is the efficacy with which they can encounter and successfully mate with other members of the same species. To incorporate this aspect of the evolution and maintenance of host specificity, the reproductive filter was proposed (Dick and Patterson 2006). This filter serves to exclude all host individuals on which the parasite cannot find mates, or upon which they are subject to decreased reproductive fitness. On the basis of mate availability and reproductive potential, high host specificity of bat flies should reinforce selection for continued specificity. If potential mates are absent from nonprimary hosts, the reproductive potential of colonizing flies would be zero, despite having passed the encounter and compatibility filters with respect to nonprimary hosts. Flies that transferred to nonprimary hosts are more likely to perish without reproducing, unless they move back onto a primary host. If indeed bat flies, or certain lineages of bat flies, are cospeciated with their hosts (Patterson et al. 1998), host speciation is likely to have been allopatric and would have forced reproductive isolation of fly populations isolated on new host populations. So while potential cospeciation may in some cases cause reproductive isolation, the reproductive filter could serve to maintain such isolation. Indeed, even transient movement of flies onto nonprimary hosts appears in general to be exceedingly rare in ecological time (Dick 2007). In other words, flies may be constrained to specificity because their primary host is the only place where they consistently and successfully find their mates. This would suggest that a reproductive filter may act as an additional threshold to the successful establishment and maintenance of parasite–host specificity.

It is possible that bat flies may also have developed immunocompatibility with specific host species. This has been demonstrated in other bloodfeeding parasites (e.g., fleas), where more generalist parasites infested host birds with weakened immune systems, while more host-specific flea species were able to feed on certain hosts regardless of the status of their immune systems (Møller et al. 2004). This points to the potential existence of shared antigenic epitopes between host and parasite, where each associate may use the same or similar immune-signaling molecules to avoid aggressive immunosurveillance. In bat flies, it has long been held that irritation from the mechanical damage of bites and potentially from introduced saliva may provoke grooming responses, which represent a source of ectoparasite mortality (Marshall 1981). Risk of mortality from host grooming may be higher for relatively large bat flies (ca. 1–5 mm), given that the hosts of many are insectivorous in their feeding habits. For permanent, host-specific parasites, selection should work to reduce irritation and thus grooming response from parasite feeding bouts. Although such studies have not been undertaken with bat flies, the presence of nested immunological compatibility in diverging host clades may go far to explain the well-known phylogenetic pattern of host specificity and the maintenance thereof.

6.3.4 Breakdown of Specificity

Although the general trend among streblid and nycteribiid flies appears to be one of high host specificity, there appear to be certain cases where bat flies depart from the overall pattern. As discussed above, field survey and natural history collections data reveal repeated (although limited) instances of steno-, oligo-, and polyxenous bat fly species. For example, 3 of 31 streblid fly species surveyed in Paraguay were less specific than strictly monoxenous (Dick and Gettinger 2005). Looking more closely at these species, they include *Trichobius jubatus*, *Aspidoptera phyllostomatis*, and *Megistopoda aranea*. Each of these species appears to be a special case. First, *T. jubatus* are rarely encountered flies in the *Trichobius dunni* species group (sensu Wenzel 1976) that parasitize several genera and species of New World molossid bat. *Trichobius dunni* group species are the only bat flies of new world molossid bats and interestingly, molossids are the only New World bats that host another group of bloodfeeding parasites, the bat bugs (Hemiptera: Polyctenidae; Whitaker et al. 2009). In Paraguay, *T. dunni* parasitized *Molossus molossus*, *M. rufus*, *Eumops patagonicus*, *E. glaucinus*, and *Molossops temminckii*. Prevalence rates on these bat species were typically low, ranging from <1 % to 15.8 %. On at least three of these host species, there is no evidence that records could possibly be due to sampling contamination. However, whether *T. jubatus* actually comprises several cryptic species, or whether the species truly is oligoxenous remains unknown at this time. But among the ca. 210 species of western hemisphere bat flies, *T. jubatus* is among the least specific species, parasitizing at least three genera of molossid bats. Its associations certainly are exceptional. As for the species *Aspidoptera phyllostomatis* and *Megistopoda aranea*, these stenoxenous species tend to co-occur on the same host species, which are the phyllostomid bats *Artibeus fimbriatus* and *A. planirostris*. A closely related species, *Megistopoda proxima*, occurs on numerous species of *Sturnira* and is known to be a complex of closely related species (Wenzel 1976) that are separable morphometrically, but species boundaries are not diagnosable (CW Dick, unpublished data). Sequence data from quickly evolving markers should serve to illuminate these units of biodiversity, and it is predicted that these stenoxenous species (like others, see Poulin and Keeney 2007) will in fact turn out to be closely related monoxenous species.

Another relatively rare situation where host specificity appears to truly break down occurs in "hot roost caves" of Puerto Rico and elsewhere in the American Antilles (Rodríguez-Durán 1998). These particular cave roosts are unique and characterized by temperatures between 28 and 40 °C and relative humidity exceeding 90 %. The high temperatures are understood to result from the metabolic activity of numerous bats and a cave topology that allows entrapment of heat (Rodríguez-Durán 1998). Within these caves, bats are parasitized by species of the streblid genus *Nycterophilia*, which are associated with bats belonging to the families Mormoopidae, Natalidae, and Phyllostomidae. *Nycterophilia* flies appear to exclusively parasitize obligate cave-roosting bats and often parasitize bats inhabiting "hot roosts." Recently, fly ecology and host associations of flies and their endosymbiotic *Gammaproteobacteria* in Puerto Rican hot caves were evaluated in the context of host specificity and roost specificity (Morse et al. 2012a). Likelihood-based ancestral character mapping provided evidence that symbionts were originally associated with bat flies that facultatively used both hot cave main chambers and cooler cave foyers. Later, the *Nycterophilia* and their symbionts evolved to an association exclusively with hot caves and bat species therein. In addition, this transition appeared to be accompanied by flies switching from being more host and less habitat specific to flies being more habitat specific and less host specific (Morse et al. 2012a). If these transitions represent the true evolutionary history of these bats, flies, and endosymbionts, it suggests selective pressure on the environmental tolerance of particular life history stages of bat flies. This situation may be a rare case of an evolutionary "retrogression" away from high host specificity among bat flies.

6.3.5 Potential as Vectors

As obligate bloodfeeding parasites, bat flies would appear to be excellent candidates for vectors of zoonotic agents. Moreover, bats are numerically abundant in the environment, evolutionarily diverse, and geographically widespread and as such are reservoirs to a stunning variety of viruses and other potential pathogens (Calisher et al. 2006). Even compared to the more speciose Rodentia, bat species on average harbor more pathogens than those of any other mammal group (Luis et al. 2013). The biology, ecology, and behavior of bats (e.g., dietary breadth, colonial sociality, mobility and migration, roost choice amplitude, etc.) make them ideal reservoir hosts for viruses and other potential disease agents. That bats are subject to higher rates and diversity of pathogen infestation than other mammal groups may owe to their high rates of sympatry, which should encourage cross-reservoir spillover of pathogens among species. Bat flies and other bloodfeeding arthropods are often suspected as playing a role in such spillover. To anyone who is experienced with handling bats, bat flies are perhaps the most consistently noticeable of the bloodfeeding arthropod parasites. They are typically large (1–5 mm), active, and stand out against bat fur and patagia. But for their size, bloodfeeding habits, and often high rates prevalence and intensity, it is currently unclear if bat flies are well positioned to moving pathogens across host species, let alone in transference to humans. Though there appears to be ample evidence of lineage sorting among bat and bat fly associations over evolutionary time, the rate at which flies naturally switch hosts in ecological time appears to be miniscule, although this is not a well-researched topic. If this is true, however, this would greatly diminish the potential for flies to serve as interhost species vectors of pathogens.

However, some of the characteristics owing to why bats have so many pathogens in the first place may speak to the likelihood of increased host switching under bat duress and disturbance. In cases of habitat or roost disturbance, bats that are otherwise ecologically separated may increase the frequency and intensity of contact with novel species. This disturbance in turn may cause an increase of accidental or transitory associations of bat flies with nonprimary or nonnatural hosts. In contrast to otherwise normal ecological conditions, such dynamics may in fact increase likelihood of host switching and hence, of movement of pathogens to novel host species.

Bats often host complex ectoparasite assemblages, and there is anecdotal evidence that some of these parasites harbor and transmit bacteria and other pathogens. For example, the bat tick *Carios kelleyi* is known to vector a number of pathogenic bacteria (*Rickettsia*, *Borrelia*, and *Bartonella*) (Loftis et al. 2005); cimicid ectoparasites of bats are vectors of *Trypanosoma* (Paterson and Woo 1984); and nycteribiids are known to transmit *Polychromophilus*, a protozoan parasite, to bats (Gardner and Molyneux 1988).

Given host specificity of bat flies, what does seem abundantly probable is that bat flies may be implicated in intrahost reservoir transference and maintenance of pathogens. For example, species of both nycteribiids and streblids are known to infest *Hypsignathus monstrosus, Epomops franqueti*, and *Myonycteris torquata*, three species of Old World fruit bats (Pteropodidae) that were reported to harbor Ebola virus (Leroy et al. 2005). Within a host population, bat flies readily move from bat to bat, feeding on multiple individuals during the course of days and weeks (Overal 1980). If flies can actually uptake viral pathogens and those pathogens remain viable within bat flies, it is probable that even host-specific flies transfer such viruses among host bats within a population.

The burgeoning literature on arthropod endosymbionts points to a multitude of bacteria and other agents in bat flies that may be mutualistic or pathogenic (Trowbridge et al. 2006; Nováková et al. 2009; Hosokawa et al. 2012; Morse et al. 2012a, b; Billeter et al. 2012). Specifically, recent research has identified *Bartonella* spp. in bat flies and bats. Bartonellae have been implicated in zoonoses in humans (Harms and Dehio 2012). The role of *Bartonella* spp. for bat flies and bats is unclear at the time, but their function as symbionts or pathogens (for bat flies) and pathogens (for bats) are being considered. There is also strong evidence that among bat fly symbionts, at least some are capable of being transmitted vertically from mother to offspring, rather than being passed laterally by uptake from the environment at large (Hosokawa et al. 2012; Morse et al. 2013). Moreover, phylogenies of some of the endosymbionts (e.g., Candidatus *Aschnera chinzeii*) are highly congruent with that of their nycteribiid bat fly hosts (Hosokawa et al. 2012) lending credence to the hypothesis that the associates are cospeciated, and that transmission of the bacteria is indeed vertical rather than horizontal. Although these results do not rule out the possibility of horizontal transmission of pathogenic agents by bat flies, they do suggest tightly coevolved linkages between bat flies, and associated symbionts and pathogens.

References

Autino AG, Claps GL, Barquez RM (1999) Insectos ectoparasitos de murciélagos de las Yungas de la Argentina. Acta Zool Mex 78:119–169

Billeter SA, Hayman DTS, Peel AJ, Baker K, Wood JLN, Cunningham A, Suu-Ire R, Dittmar K, Kosoy MY (2012) *Bartonella* species in bat flies (Diptera: Nycteribiidae) from western Africa. Parasitology 139:324–329

Brooks DR, McLennan DA (1993) Parascript: parasites and the language of evolution. Smithsonian Institution, Washington, DC, 429 pp

Bush SE, Clayton DH (2006) The role of body size in host specificity: reciprocal transfer experiments with feather lice. Evolution 60:2158–2167

Caire W, Hornuff L (1986) Overwintering population dynamics of the bat fly *Trichobius major* (Diptera: Streblidae). Southwest Nat 31:126–129

Calisher CH, Childs JE, Field HE, Holmes KV, Shountz T (2006) Bats: important reservoir hosts of emerging viruses. Clin Microbiol Rev 19:531–545

Ching LM, Marshall AG (1968) The breeding biology of the bat-fly *Eucampsipoda sundaicum* Theodor, 1955 (Diptera: Nycteribiidae). Malayan Nat J 21:171–180

Clayton DH, Bush SE, Johnson KP (2004) Ecology of congruence: past meets present. Syst Biol 53:165–173

Combes C (1991) Evolution of parasite life cycles. In: Toft CA, Aeschlimann A, Bolis L (eds) Parasite-host associations: coexistence or conflict? Oxford University Press, Oxford, pp 62–82

Dick CW (2007) High host specificity of obligate ectoparasites. Ecol Entomol 32:446–450

Dick CW, Dick SC (2006) Effects of prior infestation on host choice of bat flies (Diptera: Streblidae). J Med Entomol 43:433–436

Dick CW, Gettinger D (2005) A faunal survey of streblid bat flies (Diptera: Streblidae) associated with bats in Paraguay. J Parasitol 91:1015–1024

Dick CW, Miller JA (2010) Streblidae. In: Brown BV, Borkent A, Cumming JM, Wood DM, Woodley NE, Zumbado M (eds) Manual of central American diptera, vol II. National Research Council, Ottawa, pp 1249–1260

Dick CW, Patterson BD (2006) Bat flies: obligate ectoparasites of bats. In: Morand S, Krasnov B, Poulin R (eds) Micromammals and macroparasites: from evolutionary ecology to management. Springer, Berlin, pp 179–194

Dick CW, Patterson BD (2007) Against all odds: explaining high host specificity in dispersal-prone parasites. Int J Parasitol 37:871–876

Dittmar K, Porter ML, Murray S, Whiting MF (2006) Molecular phylogenetic analysis of nycteribiid and streblid bat flies (Diptera: Brachycera, Calyptratae): implications for host associations and phylogeographic origins. Mol Phylogenet Evol 38:155–170

Dittmar K, Dick CW, Patterson BD, Whiting MF, Gruwell ME (2009) Pupal deposition and ecology of bat flies (Diptera: Streblidae): *Trichobius* sp. (caecus group) in a Mexican cave habitat. J Parasitol 95:308–314

Dittmar K, Morse S, Gruwell M, Mayberry J, DiBlasi E (2011) Spatial and temporal complexities of reproductive behavior and sex ratios: a case from parasitic insects. PLoS One 6(5):e19438. doi:10.1371/journal.pone.0019438

Eichler WD (1963) Arthropoda. Insecta. Phpthiraptera. I. Mallophaga. In: Bronns HG (Ed) Klassen und Ordnungen des Tierreichs. III. Insecta. 7b. Pthiraptera. Akademische Verlagsanstalt Geest und Portig, Leipzig, pp 158–179

Fritz GN (1983) Biology and ecology of the bat flies (Diptera: Streblidae) on bats of the genus *Carollia*. J Med Entomol 20:1–10

Gardner RA, Molyneux DH (1988) *Polychromophilus murinus* – a malarial parasite of bats – life history and ultrastructural studies. Parasitology 96:591–605

Graciolli G, de Carvalho CJB (2001) Moscas ectoparásitas (Diptera, Hippoboscoidea) de morcegos (Mammalia, Chiroptera) do Estado do Paraná. II. Streblidae. Chave pictórica para gêneros e espécies. Rev Bras Zool 18:907–960

Guerrero R (1997) Catálogo de los Streblidae (Diptera: Pupipara) parásitos de murciélagos (Mammalia: Chiroptera) del Nuevo Mundo. VII. Lista de especies, hospedadores y paises. Acta Biol Venez 19:9–24

Hafner MS, Page RDM (1995) Molecular phylogenies and host parasite coevolution: gophers and lice as a model system. Philos Trans R Soc Lond B Biol Sci 349:77–83

Hafner MS, Demastes JW, Hafner DJ, Spradling TA, Sudman PD, Nadler SA (1998) Age and movement of a hybrid zone: implications for dispersal distance in pocket gophers and their chewing lice. Evolution 52:278–282

Hafner MS, Demastes JW, Spradling TA, Reed DL (2003) Cophylogeny between pocket gophers and chewing lice. In: Page RDM (ed) Tangled trees: phylogeny, cospeciation and coevolution. University of Chicago Press, Chicago, pp 195–220

Hagan HR (1951) Embryology of the viviparous insects. Ronald, New York

Harms A, Dehio C (2012) Intruders below the radar: molecular pathogenesis of *Bartonella* spp. Clin Microbiol Rev 25:42–78

Heeb P, Kölliker M, Richner H (2000) Bird-ectoparasite interactions, nest humidity, and ectoparasite community structure. Ecology 81:958–968

Hosokawa T, Nikoh N, Koga R, Satô M, Tanahashi M, Meng X-Y, Fukatsu T (2012) Reductive genome evolution, host–symbiont co-speciation and uterine transmission of endosymbiotic bacteria in bat flies. ISME J 6:577–587

Hutson AM (1984) Keds, flat-flies and bat-flies (diptera, hippoboscidae and nycteribiidae). Handbook for the identification of British insects. Royal Entomological Society of London, London, 40 pp

Jobling B (1949) Host-parasite relationship between the American Streblidae and the bats, with a new key to the American genera and a record of the Streblidae from Trinidad, British West Indies (Diptera). Parasitology 39:315–329

Klompen JSH, Black WC, Keirans JE, Oliver JH (1996) Evolution of ticks. Annu Rev Entomol 41:141–161

Komeno CA, Linhares AX (1999) Batflies parasitic on some phyllostomid bats in southeastern Brazil: parasitism rates and host-parasite relationships. Mem Inst Oswaldo Cruz 94:151–156

Krasnov BR, Shenbrot GI, Medvedev SG, Vatschenok VS, Khokhlova IS (1997) Host–habitat relations as an important determinant of spatial distribution of flea assemblages (Siphonaptera) on rodents in the Negev Desert. Parasitology 114:159–173

Krasnov BR, Shenbrot GI, Khokhlova IS, Medvedev SG, Vatschenok VS (1998) Habitat dependence of a parasite-host relationship: flea (Siphonaptera) assemblages in two gerbil species of the Negev Desert. J Med Entomol 35:303–313

Krasnov BR, Stanka M, Miklisova D, Morand S (2006) Habitat variation in species composition of flea assemblages on small mammals in central Europe. Ecol Res 21:460–469

Kunz TH (1982) Roosting ecology. In: Kunz TH (ed) Ecology of bats. Plenum, New York, pp 1–55

Kunz TH, Kurta A (1988) Capture methods and holding devices. In: Kunz TH (ed) Ecological and behavioral methods for the study of bats. Smithsonian Institution, Washington, DC, pp 1–29

Kunz TH, Lumsden LF (2003) Ecology of cavity and foliage roosting bats. In: Kunz TH, Fenton MB (eds) Bat ecology. University of Chicago Press, Chicago, pp 3–89

Kunz TH, Hodgkison R, Weise CD (2009) Methods of capturing and handling bats. In: Kunz TH, Parsons S (eds) Ecological and behavioral methods for the study of bats. Johns Hopkins University Press, Baltimore, pp 3–35

Leroy EM, Kumulungui B, Pourrut X, Rouquet P, Hassanin A, Yaba P, Delicat A, Paweska JT, Gonzalez JP, Swanepoel R (2005) Fruit bats as reservoirs of Ebola virus. Nature 438:575–576

Lewis SE (1995) Roost fidelity of bats: a review. J Mammal 76:481–496

Light JE, Reed DL (2009) Multigene analysis of phylogenetic relationships and divergence times of primate sucking lice (Phthiraptera: Anoplura). Mol Phylogenet Evol 50:376–390

Loftis AD, Gill JS, Schriefer ME, Levin ML, Eremeeva MW, Gilchrist MJR, Dasch GA (2005) Detection of *Rickettsia, Borrelia*, and *Bartonella* in *Carios kelleyi* (Acari: Argasidae). J Med Entomol 42:473–480

Luis AD, Hayman DT, O'Shea TJ, Cryan PM, Gilbert AT, Pulliam JR, Mills JN, Timonin ME, Willis CK, Cunningham AA, Fooks AR, Rupprecht CE, Wood JL, Webb CT (2013) A comparison of bats and rodents as reservoirs of zoonotic viruses: are bats special? Proc R Soc B 280:20122753. doi:10.1098/rspb.2012.2753

Marshall AG (1970) The life cycle of *Basilia hispida* Theodor 1957 (Diptera: Nycteribiidae) in Malaysia. Parasitology 61:1–18

Marshall AG (1976) Host-specificity amongst arthropods ectoparasitic upon mammals and birds in the New Hebrides. Ecol Entomol 1:189–199

Marshall AG (1980) The comparative ecology of insects ectoparasitic upon bats in west Malaysia. In: Wilson DE, Gardner AL (eds) Proceedings of the 5th international bat research conference. Texas Tech, Lubbock, TX, pp 135–142

Marshall AG (1981) The ecology of ectoparasitic insects. Academic, London, p 459

Medvedev SG, Krasnov BR (2006) Fleas: permanent satellites of small mammals. In: Morand S, Krasnov BR, Poulin R (eds) Micromammals and macroparasites: from evolutionary ecology to management. Springer, Tokyo, pp 161–177

Miles VIA, Kinney R, Stark HE (1957) Flea-host relationships of associated *Rattus* and native wild rodents in the San Francisco Bay area of California, with special reference to plague. Am J Trop Med Hyg 6:752–760

Møller AP, Marti M, Vivaldi N, Soler JJ (2004) Parasitism, host immune defence and dispersal. J Evol Biol 17:603–612

Morse S, Dick CW, Patterson BD, Dittmar K (2012a) Some like it hot: evolution and ecology of novel endosymbionts in bat flies of cave-roosting bats (Hippoboscoidea, Nycterophiliinae, *Nycterophilia*). Appl Environ Microbiol 78:8639–8649

Morse S, Olival K, Kosoy M, Billeter S, Patterson BD, Dick CW, Dittmar K (2012b) Global distribution and genetic diversity of *Bartonella* in bat flies (Hippoboscoidea, Streblidae, Nycteribiidae). Infect Genet Evol 12:1717–1723

Morse SF, Bush S, Patterson BD, Dick CW, Gruwell M, Dittmar K (2013) Evolution, multiple acquisition, and localization of endosymbionts in bat flies (Hippoboscoidea, Streblidae, Nycteribiidae). Appl Environ Microbiol 79:2952–2961. doi:10.1128/AEM.03814-12

Nováková E, Hypsa V, Moran NA (2009) *Arsenophonus*, an emerging clade of intracellular symbionts with a broad host distribution. BMC Microbiol 9:143. doi:10.1186/1471-2180-9-143

Overal WL (1980) Host-relations of the batfly *Megistopoda aranea* (Diptera: Streblidae) in Panama. Univ Kansas Sci Bull 52:1–20

Paterson WB, Woo PTK (1984) The development of the culture and blood-stream forms of 3 *Trypanosoma* (*Schizotrypanum*) spp. (Protista, Zoomastigophorea) from bats in *Cimex lectularius* (Hemiptera, Cimicidae). Can J Zool 62:1581–1587

Patterson BD, Ballard JWO, Wenzel RL (1998) Distributional evidence for cospeciation between Neotropical bats and their bat fly ectoparasites. Stud Neotrop Fauna Environ 33:76–84

Patterson BD, Dick CW, Dittmar K (2007) Roosting habits of bats affect their parasitism by bat flies (Diptera: Streblidae). J Trop Ecol 23:177–189

Patterson BD, Dick CW, Dittmar K (2008a) Parasitism by bat flies (Diptera: Streblidae) on Neotropical bats: effects of host body size, distribution and abundance. Parasitol Res 103:1091–1100

Patterson BD, Dick CW, Dittmar K (2008b) Sex biases in parasitism of neotropical bats by bat flies (Diptera: Streblidae). J Trop Ecol 24:387–396

Perlman SJ, Jaenike J (2003) Infection success in novel hosts: an experimental and phylogenetic study of *Drosophila*-parasitic nematodes. Evolution 57:544–557

Petersen FT, Meier R, Kutty SN, Wiegmann BM (2007) The phylogeny and evolution of host choice in the Hippoboscoidea (Diptera) as reconstructed using four molecular markers. Mol Phylogenet Evol 45:111–122

Peterson BV, Wenzel RL (1987) Nycteribiidae. In: McAlpine JF, Peterson BV, Shewell GE, Teskey HJ, Vockeroth JR, Wood DM (eds) Manual of nearctic Diptera. Research Branch, Agriculture Canada, Ottawa, pp 1283–1291

Pilosof S, Dick CW, Corine C, Patterson BD, Krasnov B (2012) Effects of anthropogenic disturbance and climate on patterns of bat fly parasitism on bats. PLoS One 7:1–7. doi:10.1371/journal.pone.0041487

Poulin R (1998a) Evolutionary ecology of parasites: from individuals to communities. Chapman and Hall, London, 212 pp

Poulin R (1998b) Host and environmental correlates of body size in ticks (Acari: Argasidae and Ixodidae). Can J Zool 76:925–930

Poulin R, Keeney DB (2007) Host specificity under molecular and experimental scrutiny. Trends Parasitol 24:24–28

Poulin R, Morand S (2005) Parasite Biodiversity. Smithsonian Institution, Washington, 224 pp

Poulin R, Mouillot D (2003) Parasite specialization from a phylogenetic perspective: a new index of host specificity. Parasitology 126:473–480

Presley SJ (2004) Ectoparasitic assemblages of Paraguayan bats: ecological and evolutionary perspectives. Ph.D. Dissertation, Texas Tech University, Lubbock, TX, 326 pp

Price PW (1980) Evolutionary biology of parasites. Monogr Popul Biol 15:1–237

Reckardt K, Kerth G (2006) The reproductive success of the parasitic bat fly *Basilia nana* (Diptera: Nycteribiidae) is affected by the low roost fidelity of its host, the Bechstein's bat (*Myotis bechsteinii*). Parasitol Res 98:237–243

Reed DL, Light JE, Allen JM, Kirchman JJ (2007) Pair of lice lost or parasites regained: the evolutionary history of anthropoid primate lice. BMC Biol 5:1–11

Reisen WK, Kennedy ML, Reisen NT (1976) Winter ecology of ectoparasites collected from hibernating *Myotis velifer* (Allen) in southwestern Oklahoma (Chiroptera: Vespertilionidae). J Parasitol 62:628–635

Rodríguez-Durán A (1998) Nonrandom aggregations and distribution of cave-dwelling bats in Puerto Rico. J Mammal 79:141–146

Rohde K (1980) Host specificity indices of parasites and their application. Experientia 36:1369–1371

Ross A (1960) Notes on *Trichobius corynorhini* on hibernating bats (Diptera: Streblidae). Wasmann J Biol 18:271–272

Seneviratne SS, Fernando HC, Udagama-Randeniya PV (2009) Host specificity in bat ectoparasites: a natural experiment. Int J Parasitol 39:995–1002

Smith MA, Woodley NE, Janzen DH, Hallwachs W, Hebert PDN (2006) DNA barcodes reveal cryptic host-specificity within the presumed polyphagous members of a genus of parasitoid flies (Diptera: Tachinidae). Proc Natl Acad Sci USA 103:3657–3662

Stark HE, Kinney AR (1969) Abundance of rodents and fleas as related to plague in Lava Beds National Monument, California. Pan-Pac Entomol 38:249–251

Tello JS, Stevens RD, Dick CW (2008) Patterns of species co-occurrence and density compensation: a test for interspecific competition in bat ectoparasite infracommunities. Oikos 117:693–702

ter Hofstede HM, Fenton MB (2005) Relationships between roost preferences, ectoparasite density, and grooming behaviour of Neotropical bats. J Zool 266:333–340

ter Hofstede HM, Fenton MB, Whitaker JO Jr (2004) Host and host-site specificity of bat flies (Diptera: Streblidae and Nycteribiidae) on Neotropical bats (Chiroptera). Can J Zool 82:616–626

Theodor O (1957) Parasitic adaptation and host-parasite specificity in the pupiparous Diptera. In: Mayr E (ed) First symposium on host specificity among parasites of vertebrates. Institut de Zoologie, Université de Neuchâtel, Switzerland, pp 50–63

Timm RM (1987) Tent construction by bats of the genera *Artibeus* and *Uroderma*. In: Patterson BD, Timm RM (eds) Studies in neotropical mammalogy: essays in honor of Philip Hershkovitz, vol 39, Fieldiana: zoology, new series . Field Museum of Natural History, Chicago, pp 187–212

Timms R, Read AF (1999) What makes a specialist special? Trends Ecol Evol 14:333–334

Tompkins DM, Clayton DH (1999) Host resources govern the specificity of swiftlet lice: size matters. J Anim Ecol 68:489–500

Trowbridge RE, Dittmar K, Whiting MF (2006) Identification and phylogenetic analysis of *Arsenophonus*- and *Photorhabdus*-type bacteria from adult Hippoboscidae and Streblidae (Hippoboscoidea). J Invertebr Pathol 91:64–68

Wenzel RL (1976) The streblid batflies of Venezuela (Diptera: Streblidae). Brigham Young Univ Sci Bull 20:1–177

Wenzel RL, Tipton VJ (1966) Some relationships between mammal hosts and their ectoparasites. In: Wenzel RL, Tipton VJ (eds) Ectoparasites of Panama. Field Museum of Natural History, Chicago, pp 677–723

Wenzel RL, Tipton VJ, Kiewlicz A (1966) The streblid batflies of Panama (Diptera: Calypterae: Streblidae). In: Wenzel RL, Tipton VJ (eds) Ectoparasites of Panama. Field Museum of Natural History, Chicago, pp 405–675

Whitaker JO Jr, Ritzi CM, Dick CW (2009) Collecting and preserving ectoparasites for ecological study. In: Kunz TH, Parsons S (eds) Ecological and behavioral methods for the study of bats. Johns Hopkins University Press, Baltimore, pp 806–827

Whiteman NK, Sánchez P, Merkel J, Klompen H, Parker PG (2006) Cryptic host specificity of an avian skin mite (Epidermoptidae) vectored by louseflies (Hippoboscidae) associated with two endemic Galapagos bird species. J Parasitol 92:1218–1228

Whiting MF, Whiting AS, Hastriter MW, Dittmar K (2008) A molecular phylogeny of fleas (Insecta: Siphonaptera): origins and host associations. Cladistics 24:677–707