

Evolutionary ecology of bird-parasite associations

Daniel Michael Tompkins

Wolfson College

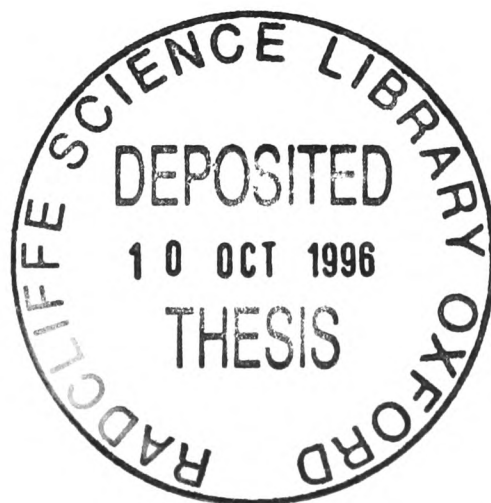
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Abstract

This thesis investigates the ecological determinants of chewing louse (Insecta: Phthiraptera) host-specificity on four species of Malaysian swiftlets (Aves: Apodidae). Influences of host coloniality on louse ecology were also demonstrated, illustrating the dependence which these permanent ectoparasites have on their hosts.

Louse collections were made to look for incidences of host-specific lice occurring on the "wrong" host ("straggling"). Straggling was observed, implying that lice disperse among host species. Thus, opportunity for louse dispersal (or lack thereof) does not govern the host-specificity of chewing lice on swiftlets.

Experimental transfers of lice between hosts were conducted. Louse survival was reduced on foreign host species. This implies adaptation to specific host characters, suggesting that specialisation governs chewing louse host-specificity on swiftlets.

There was no evidence for reciprocal adaptation of swiftlets to their normal louse species. Lice had no impact on the fitness of either swiftlets or the related common swift. Furthermore, neither swiftlet nor swift lice were transmitting pathogenic endoparasites. This implies that chewing lice and Malaysian swiftlets have not "coevolved".

Survival of transferred lice was determined by the relatedness of donor and recipient hosts. Closer related swiftlet species are more similar in body size and feather dimensions. When the feather dimensions of the microhabitat distributions of the same louse species on different hosts were compared the results suggested that lice keep the dimensions of barb and barbule diameter, at which they occur, "constant" through microhabitat shifts. This suggests that feather dimensions are the host characters which determine the survival (and host-specificity) of chewing lice on birds.

The ability of chewing lice to survive on hosts with similar feather morphology implies that "host-switching", between distantly related hosts with similar morphological characters (due to parallel or convergent host evolution), may have been an important factor in the evolution of bird-lice associations.

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Chapter 1 General introduction

1.1 Evolutionary ecology

Using molecular data and parsimony analysis, accurate phylogenies can now be constructed for almost any group of organisms (Brooks & McLennan 1993).

However, although phylogenies reveal patterns of evolution, they allow only limited understanding of the processes (speciation and extinction) underlying them (Brooks & McLennan 1991; Roderick 1996). These processes are governed by environmental pressures over evolutionary time-scales (Otte & Endler 1989; Thompson 1994).

"Evolutionary ecology" is the study of the environmental pressures which have determined the evolutionary patterns of biological organisms. As such, it investigates the ecological reasons for speciation and extinction in phylogenetic lineages, increased understanding of which are important for predicting future changes in the biodiversity of ecosystems (Thompson 1996).

The principle goal of this thesis is to identify the environmental factors that govern the host-specificity of ectoparasitic chewing lice (Insecta: Phthiraptera) on Malaysian swiftlets (Aves: Apodidae). Host-parasite systems are powerful arenas for the study of evolutionary patterns because the environment of many parasites is delimited chiefly by the host (Price 1980; Adamson & Caira 1994). For this reason, factors governing species diversity and degree of specialisation may be easier to identify for parasites than for free-living organisms (Rohde 1994). Furthermore, since chewing lice are permanent ectoparasites which complete their entire lifecycle on the host (see below), the proximate factors governing their host-specificity may be easier to identify than those for parasites with more complex lifecycles (Dogiel 1964). An understanding of the environmental factors which govern host-specificity of parasitic organisms in ecological time can shed light on the mechanisms that have governed the evolution of both parasitic and free-living organisms.

There are a number of areas of research that inform evolutionary ecology. However, one of the most powerful tools for evolutionary ecologists is manipulations of an organisms environment to test explicit predictions of micro-evolutionary theory (Alstad & Edmunds 1983; Futyma *et al.* 1994; Hanks & Denno 1994; Mopper *et al.* 1995; Joshi & Thompson 1996). This is the main approach used in my thesis work.

1.2 The hosts: Malaysian swiftlets (Aves: Apodidae)

The swiftlets (Collocaliini) are distributed from the Indian Ocean, through South-East Asia and Northern Australia, to the Pacific (Pratt 1986; Chantler & Driessens 1995). They are a group of tiny → small aerial insectivores (Hails & Amirrudin 1981; Waugh & Hails 1983), ranging in body mass from 8-39g (Chantler & Driessens 1995). Since insect abundance in tropical climes is relatively constant (Hails 1982), swiftlets are non-migratory.

As a group, the swiftlets show a high degree of morphological similarity (Sims 1961; Medway 1966) making them notoriously difficult to classify: "Every author who has ever worked with these small swiftlets of the Indo-Australian region will contend that their classification presents the most difficult problem in the taxonomy of birds...most of the species are of practically the same dull sooty grey coloration with almost the same development of structural characters..." (Mayr 1937). The paucity of distinguishing morphological characters among swiftlets has led to a reliance on behavioural characters, especially nest structure, in their classification.

Most swiftlet species nest in caves and have the ability to echolocate, using buccal clicks, for navigational purposes (Medway 1959; Brock Fenton 1975; Coles *et al.* 1987; Collins & Murphy 1993; Fullard *et al.* 1993). This characteristic has been used to split the swiftlets into two distinct genera: swiftlets which can echolocate (*Aerodramus* spp.), and those which cannot (*Collocalia* spp.) (Brooke 1972; Medway

& Pye 1977; Holyoak & Thibault 1978). A recent molecular phylogeny shows that *Aerodramus* and *Collocalia* are each a monophyletic group (Lee *et al.* in press). However, the tribe Collocaliini (swiftlets) is not monophyletic; *Aerodramus* and *Collocalia* have independent origins.

Four species of Malaysian swiftlets were involved in my study: the glossy swiftlet (*Collocalia esculenta*), mossy-nest swiftlet (*Aerodramus salanganus*), white-nest swiftlet (*Aerodramus fuciphagus*), and black-nest swiftlet (*Aerodramus maximus*) (Table 1.1; Figures 1.1 & 1.2). DNA sequencing has confirmed that all four swiftlets are distinct species (Lee *et al.* in press). The nests of white-nest and black-nest swiftlets are harvested for use in Chinese medicine and cuisine ("edible bird's nests"; Sims 1959; Medway 1969; Francis 1987a) (Figure 1.3). Nests are formed from hardened salival secretions (mixed with body feathers in the case of black-nests) (Wang 1921; Marshall & Folley 1956; Medway 1962a).

Malaysian swiftlets are ideal subjects for the study of host-parasite evolutionary ecology. They are colonial, enabling the efficient study of large samples. Furthermore, swiftlet nestlings can be taken into temporary captivity for collection of parasites since they do not require brooding for warmth or frequent feeding (adults feed young with boluses of aerial insects which take a long time to collect). At my main study site (Gomantong Caves, Sabah (Northern Borneo)) all four swiftlet species nest in the same caves, with overlapping breeding dates (generally February - September; Francis 1987a). This enabled the experimental transfer of lice between host species (manipulation of parasite environment) to be carried out (see below).

1.3 The parasites: chewing lice (Insecta: Phthiraptera)

Lice are ectoparasitic insects of birds and mammals which have evolved from a common ancestor with the free-living Psocoptera (booklice, barklice) (Lyal 1985;

Table 1.1 Morphological, behavioural and reproductive characteristics of Malaysian swiftlets.

Taxonomic name	Common name	Body mass	Wing chord	Echolocate ?	Nest sites	Nest structure	Clutch size	Incubation period	Nestling period
<i>Collocalia esculenta</i>	Glossy swiftlet	7-8 g	95-105 mm	No	Caves, houses, culverts, etc.	Vegetation	2	22 days	35-40 days
<i>Aerodramus salanganus</i>	Mossy-nest swiftlet	10-15 g	115-127 mm	Yes	Interior of caves	Vegetation	2	23 days	50 days
<i>Aerodramus fuciphagus</i>	White-nest swiftlet	10-15 g	114-123 mm	Yes	Interior of caves	Saliva	2	23 days	43 days*
<i>Aerodramus maximus</i>	Black-nest swiftlet	14-21 g	128-137 mm	Yes	Interior of caves	Saliva and feathers	1	28 days	59 days

Sources: Medway 1962b; Langham 1980; Francis 1987a,b.
* measured from an unmatured colony maintained, for the purpose of nest-harvesting, inside a house.

Figure 1.1 a) Adult glossy swiftlet (*Collocalia esculenta*). b) Cluster of black-nest swiftlets (*Aerodramus maximus*).



Figure 1.2 Mossy nests (of *Aerodramus salanganus*) containing a) two eggs and b) young nestlings (approximately 1 week old).

a)



b)



Figure 1.3 Harvesting of "edible birds nests".



Barker 1994). They are obligate permanent ectoparasites that complete their entire lifecycle on the body of the host (Figure 1.4) (Marshall 1981a; Lee & Clayton 1995). It is this dependence on the host which has most likely led to the high levels of host-specificity (Hopkins 1949; Clay 1957; Price 1980; Clayton 1990) and cospeciation (i.e. parasite phylogeny mirrors host phylogeny; Hafner & Nadler 1988; Paterson *et al.* 1993; Hafner & Page 1995) observed in host-louse associations.

The chewing lice I studied are of the genus *Dennyus* (Amblycera: Menoponidae) (Clayton *et al.* in review) (Figure 1.5). Amblyceran lice on birds feed on a variety of substances, including feather parts, dermal debris, host eye secretions, blood, other lice, and feather mites (Marshall 1981a), but generally show microhabitat preferences on the body of their hosts (Nelson & Murray 1971; Eveleigh & Threlfall 1976; Choe & Kim 1988; Stock & Hunt 1989). *Dennyus hirundinis*, on the common swift (*Apus apus*), feeds on dermal debris, blood, and host eye-fluid (Rothschild & Clay 1952; Bromhall 1980; Lee & Clayton 1995).

Chewing louse populations are relatively easy to quantify on avian hosts (Clayton & Walther in press). *Dennyus* spp. are no exception (Lee & Clayton 1995). The eggs, which are glued to the hosts feathers with a glandular cement, are large enough to see with the naked eye (0.5-1 mm long) and are white, making them easy to detect in the dark plumage of swifts and swiftlets. The post-hatching stages, consisting of three nymphal instars and the adult, are also relatively large and easy to see. Male *Dennyus* are 15-20% smaller than females (Clayton *et al.* in review).

Due to the close host association, Amblyceran lice generally require direct contact to transmit among individual hosts, although transmission via shared nest sites, dust-bathing sites etc. may also occur (Marshall 1981a). The main route of dispersal for *Dennyus hirundinis* on the common swift is vertical transmission from adult birds to their offspring in the nest (Lee & Clayton 1995), especially since adult birds spend all of their time flying when away from the nest (Lack 1956a), although transmission is

Figure 1.4 Chewing louse lifecycle.

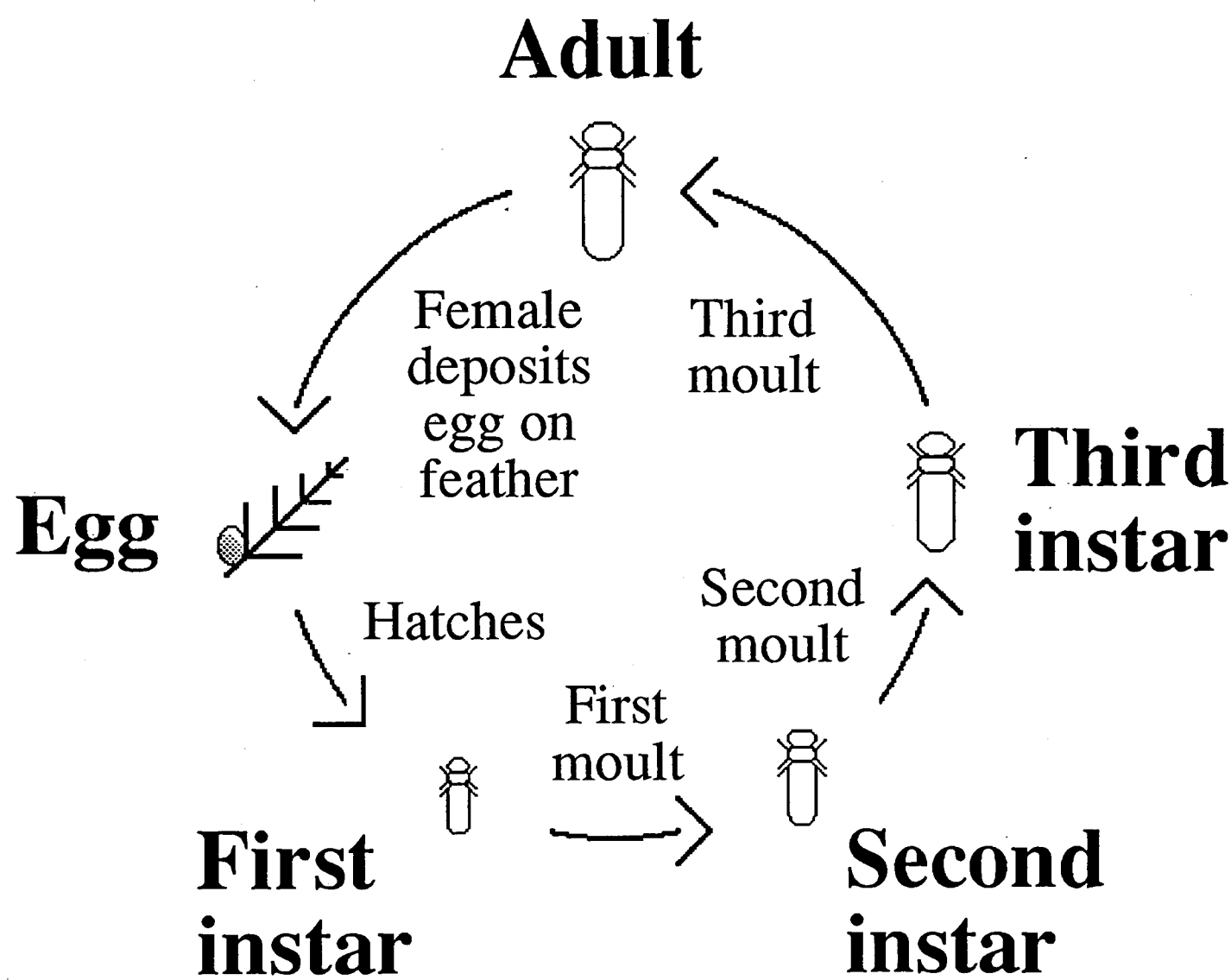
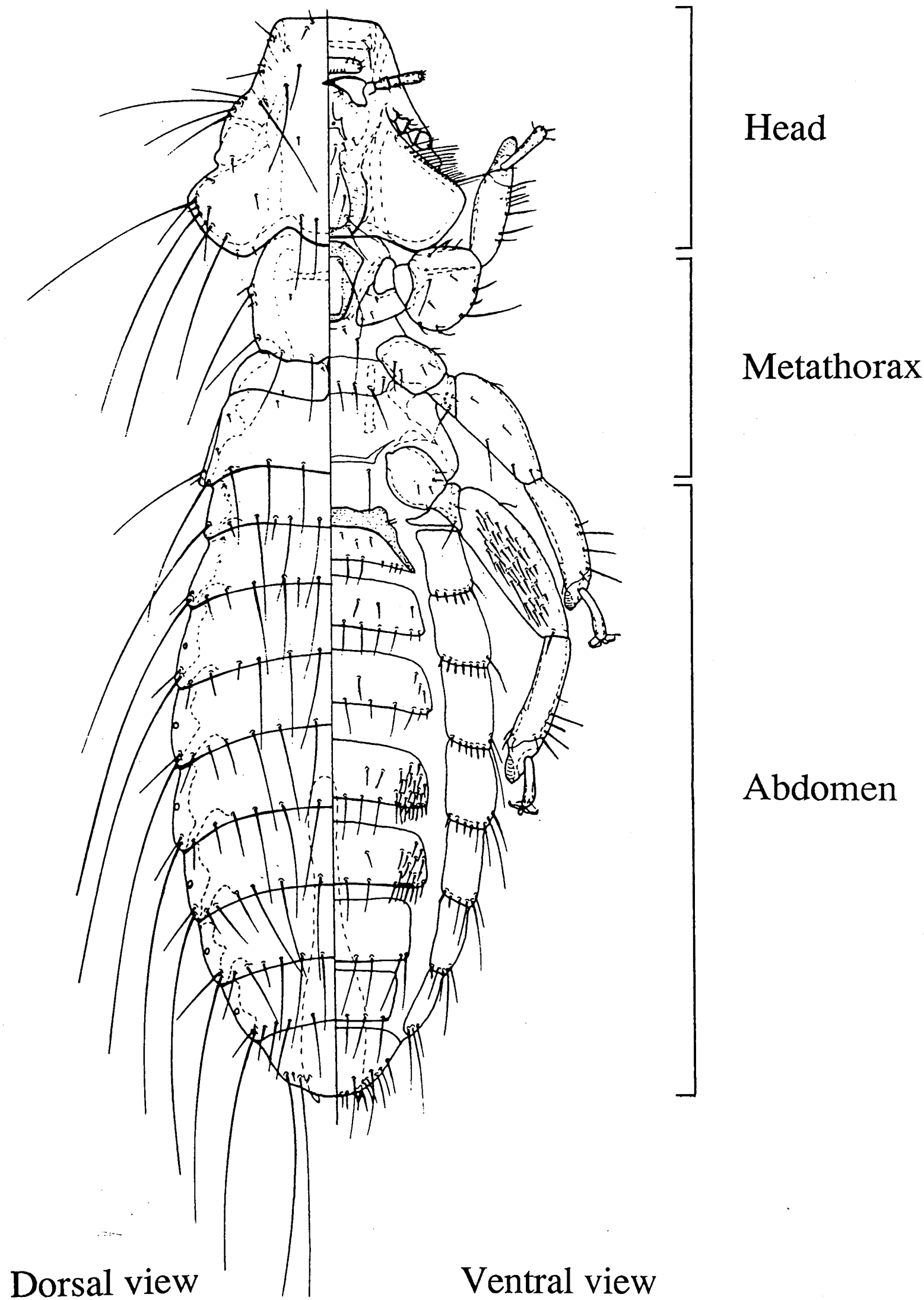


Figure 1.5 Male *Dennyus distinctus* from the glossy swiftlet (*Collocalia esculenta*) .

Total length $\cong$ 2.1mm. (after Clayton *et al.* in review)



likely to occur between mated adults at the nest. This is, presumably, also the case for lice on swiftlets. Since horizontal transmission of lice among host group members does occur (Durden 1983), some lice may also transmit between swiftlets nesting in close proximity. Furthermore, swiftlet adults often fight with conspecifics (personal observation), during which louse transfer may occur.

The number of lice present on birds has been shown to fluctuate seasonally, usually with a maximum occurring just before the hatching of the birds' eggs (Foster 1969; Eveleigh & Threlfall 1976; Clark *et al.* 1994). It is proposed that the timing of louse breeding coincides with the timing of host breeding, resulting in sufficient numbers of lice for transfer to newly available juvenile hosts.

Chewing lice are ideal subjects for the study of host-parasite evolutionary ecology. Their close host association limits the environmental factors which could govern their host-specificity (Price 1980) and, since they are ectoparasites, they are relatively easy to quantify and conducive to experimental manipulation. Furthermore, *Dennyus* spp. on Malaysian swiftlets vary in their degree of host-specificity, from monoxenous (only one host species) to oligoxenous (two or more host species in the same genera) (Clayton *et al.* in review).

1.4 Historical context

The evolutionary study of host-parasite associations originated with the work of von Ihring (1902). His main interest was in the argument over the relative importance of natural selection versus isolation in evolution and speciation. He believed isolation to be the more important factor, interpreting taxonomic and distributional evidence for parasitic helminths of fish, birds and mammals as evidence for a former land connection linking the southern continents. To von Ihring, the three concepts of

speciation by isolation, relative age of lineages, and the correlation between host and parasite phylogenies combined to form an important biogeographic tool.

Host-louse relationships were brought into an evolutionary context by two independent researchers in the early 1900's. Kellogg (1913), in agreement with von Ihring, considered isolation between parasite populations on different hosts to be the most important factor governing louse speciation, resulting in a high degree of host-specificity and congruence between host and louse phylogenies (cospeciation). However, whereas von Ihring considered natural selection to play practically no role in evolution and speciation, Kellogg believed it to complement isolation. Kellogg believed that not only isolation but also parasite adaptation to host characters (specialisation), preventing successful louse transfer between distantly related host species ("host-switching"), maintained high levels of louse host-specificity and led to congruent phylogenies. However, Kellogg did not consider specialisation to have a role in the host-specificity and speciation of lice on closely related hosts. He believed that the host characters to which lice adapt (feathers and skin) change at a slower rate than the environmental characters to which hosts adapt and speciate in response to, resulting in a slower rate of parasite divergence.

Like von Ihring, Fahrenholz (1913) considered host-specificity of lice to be governed by isolation between parasite populations on different hosts. However, like Kellogg, he also believed that lice adapt to specific host characters but, unlike Kellogg, he believed that not only will lice cospeciate with their hosts but they will also diverge to the same extent as their hosts through continual adaptation (since, to a louse, the host is the entire environment). Due to the supposed identical nature of host and parasite phylogenies, Fahrenholz concluded that parasite relations could be used as a tool to elicit host relations. This theory was summarised by Eichler (1942) as "Fahrenholz's Rule": "Among numerous (mainly permanent) parasites, the historical development and splitting of the hosts is paralleled by a corresponding development and splitting of the parasites. Therefore, the resulting phylogenetic relationships of the parasites

can be used to draw conclusions about the (often obscured) phylogenetic relationships of the hosts".

The use of louse distributions to deduce host relatedness ("comparative parasitology") was first suggested by Jardine (1841; cited in Hopkins 1951). Utilising the framework created by Fahrenholz and Eichler, this was attempted by many parasitologists in the mid-1900's (e.g. Hopkins 1942). However, problems with this method soon became apparent (Paterson *et al.* 1995). Although louse relationships do generally reflect those of their hosts (through cospeciation) it became evident that, in many cases, the basic relationships have become confused leading to anomalous louse distributions which would be misinformative if used as guides to host relatedness (e.g. Sibly *et al.* 1969). Clay (1949) was one of the first to question the validity of the "comparative parasitology" approach, listing possible causes of anomalous louse distributions as "...discontinuous distribution, excessive convergent and parallel evolution making a reliable evaluation of phylogenetic relationships difficult, and secondary interspecific infestations."

Current theory maintains that host-louse associations have evolved through a combination of cospeciation (Page 1993a), host-switching events (Lyal 1986; Barker 1991), and sorting events such as the presence of multiple lineages of parasites coupled with parasite extinction (Page 1993b) or failure of parasites to colonise both descendants of a host speciation event (Paterson & Gray in press). However, there is still much contention over the relative importance of each factor (Barker 1994; Page *et al.* 1996; Barker 1996). In investigating the ecological determinants of louse host-specificity, this thesis has the potential to shed light on the mechanisms behind the evolution of host-louse associations, clarifying this situation.

1.5 Chapter contents.

Chapter 2 documents the basic ecology of chewing lice on the glossy swiftlet, including population structure, microhabitat use and population dynamics. The influence which host ecology (specifically host density) has on louse ecology is assessed to illustrate the high degree of chewing louse dependence on the host.

In Chapter 3, the blood parasites of Malaysian swiftlets are surveyed. Ectoparasites are known to transmit endoparasites between hosts (Balashov 1984). If blood parasites are present, their possible transmission by ectoparasites may have greatly effected the evolution of the host-parasite associations (swiftlets and their chewing lice) under investigation.

Chapter 4 investigates the determinants of chewing louse host-specificity on Malaysian swiftlets. The principle question asked is whether louse host-specificity is governed by restricted gene flow between louse populations on different host populations due to lack of dispersal opportunity and/or louse adaptation to specific host characters (specialisation). The possibility of reciprocal host adaptation to their normal species of chewing lice is also investigated. Degree of adaptation, of both parasites and hosts, is assessed through controlled transfer experiments. Lice are transferred between swiftlet nestlings (of all four species involved in the study) to monitor the effect of environmental manipulation on louse survival, and the effect of parasite manipulation on nestling growth and fledging success.

In Chapter 5, host characters which may determine the survival of chewing lice on Malaysian swiftlets (host characters to which lice may have adapted), and thus govern louse host-specificity, are identified.

One reason why swiftlet hosts may not have adapted to their normal chewing lice is that *Dennyus* spp. may have no impact on host fitness. This possibility is investigated in Chapter 6, by looking for effects of *Dennyus hirundinis* on the growth and fledging

of common swift nestlings. The impact of another ectoparasite, the hippoboscid fly *Crataerina pallida* (Diptera: Hippoboscidae), on the growth and fledging of swift nestlings is also investigated.

Finally, as an independent project, Chapters 7 & 8 document work carried out for the Sabah Wildlife Department (Malaysia), investigating the impact of nest-harvesting on the reproductive success of "edible-nest" swiftlets.

In the conclusion, the implications of the results from Chapters 2-6 on host-parasite evolutionary theory (specifically the evolution of host-chewing louse associations) are discussed. Future experiments are suggested.

Chapter 2 Influence of host density on the ecology of avian lice

2.1 Introduction

Increased infection by contagious parasites (those transmitted by close proximity among hosts or via infected faeces) has long been considered a universal cost of group-living in animals (Alexander 1974; Freeland 1976). Groups members have a higher chance of acquiring and accumulating parasites than solitary individuals due to increased parasite transmission through closer proximity and greater number of physical contacts between hosts (Møller, Dufva & Allander 1993). In a recent meta-analysis of published studies, Cote & Poulin (1995) demonstrated a strong positive correlation between both the prevalence and intensity of contagious parasites and host group size across a variety of taxa.

Rozsa *et al.* (1996) demonstrated a relationship between the degree of host coloniality and the population ecology of (contact transmitted) avian lice. They compared the louse populations on two congeneric host species, the territorial hooded crow (*Corvus corone cornix*) and the colonial rook (*Corvus frugilegus*). The frequency distribution of lice on rooks was less aggregated than that of lice on crows, and prevalence was higher. Horizontal transmission of lice among group members is known to occur (Durden 1983), making it likely that the above results were due to increased parasite transmission among host individuals at greater density. Degree of parasite aggregation depends on many factors including transmission rates (Anderson & May 1985).

Rozsa *et al.* also showed that louse sex ratios were less female-biased on colonial rooks than on territorial crows. They interpreted their result as louse adaptation to different levels of local mate competition. Skewed sex ratios are a common phenomena in avian lice, usually showing a preponderance of females (Marshall 1981b; Wheeler & Threlfall 1986; Clayton *et al.* 1992; but see Lee & Clayton 1995). Female-biased ratios should evolve in isolated populations subject to inbreeding, when

relatively few males are required to fertilise all the females (local mate competition) (Fisher 1930). Under such conditions, the production of females is selectively favoured because eggs are more limited than sperm (Charnov 1982). This should be the case in permanent parasites, such as lice, where populations on host individuals generally show high degrees of genetic isolation (Nadler *et al.* 1990). However, if horizontal transmission of lice among hosts is greater in denser populations, mating among lice would be more random, and a less-biased sex ratio would be selected for.

As stated by Rozsa *et al.*, both intra- and inter-specific comparisons of avian social systems might be used to investigate the influence of host coloniality on parasite population ecology. They considered intra-specific comparisons (of, for example, facultatively colonial bird species) to be hindered in that hosts would not be limited to the density in which they were observed, but may easily switch between density levels (see Sasvari & Hegyi 1994). Thus, parasites on these hosts would not be able to "develop their adaptive features to a particular social system".

Here I argue that, when interpreting results, intra-specific comparisons are preferable to inter-specific comparisons. Rozsa *et al.* interpreted their results as louse adaptations to host ecology. However, their results may also have been non-adaptive, resulting solely from changes in the environmental variables which influence louse population dynamics. In contrast, results from intra-specific comparisons, where host density was variable over time (precluding parasite adaptations to specific levels of host density), could be confidently interpreted as having ecological, and not evolutionary, causes.

Other inter-specific correlations between the prevalence and intensity of contagious ectoparasites versus host density have been previously reported for parasites of both birds and mammals (but see Poiani 1992). Poulin (1991) reported higher prevalences of feather mites on colonial versus solitary passerines when analysed at the level of both host species and host family. Ubelaker (1970) reported that bat species roosting in large aggregations have more streblid flies than species found in smaller roosts.

Hoogland (1979) reported higher prevalences and intensities of fleas, lice, mites and ticks on a densely colonial species of prairie dog (the black-tailed prairie dog, *Cynomys ludovicianus*) than on a loosely colonial species (the white-tailed prairie dog, *Cynomys leucurus*).

Many intra-specific correlations between the prevalence and intensity of contagious ectoparasites versus host density have also been previously reported for parasites of both birds and mammals. Hoogland & Sherman (1976) reported increasing prevalence and intensity of fleas on bank swallows (*Riparia riparia*) with increasing colony size. Brown & Brown (1986) reported increasing intensity of both fleas and swallow bugs on cliff swallows (*Hirundo pyrrhonota*) with increasing colony size. Møller (1987) reported increasing prevalence of swallow bugs on barn swallows (*Hirundo rustica*) with increasing colony size. Kunz (1976) reported increasing intensity of streblid flies on bats (*Plecotus townsendii*) with increasing colony size, and Hoogland (1979) reported increasing prevalence and intensity of fleas in prairie dog burrows with increasing colony size for both black- and white-tailed species. However, all of the above correlations are among colonies, where parasite adaptation to local host density may have occurred. Thus, they may be due to either adaptive or non-adaptive reasons.

The purpose of the work described in this chapter is to investigate the influence of host density on avian louse ecology, through an intra-specific comparison of lice on hosts at different densities within a single colony of the glossy swiftlet (see Chapter 1).

Since morphological (Lone 1991) and genetic (Nadler *et al.* 1990) differences among louse subpopulations have been demonstrated, restricting the comparison between host densities to intra-colony variation was necessary to rule out adaptive components.

Breeding adult swiftlets are nest-site specific, rebuilding nests on the same exact sites both within and between seasons (Francis 1987a; Kang *et al.* 1991). However, the pattern of nest density within the glossy swiftlet colony studied alters from year to year (personal observation), presumably due to adult mortality and juvenile

recruitment. This would, most likely, preclude parasite adaptations to specific levels of host density within the colony.

The first goal of this work was to document the basic ecology of chewing lice on the glossy swiftlet, including louse population structure, microhabitat use, and population dynamics. Second, the influence of nest density on louse ecology (specifically parasite loads and sex ratios) was investigated.

2.2 Study site

The study was carried out, April - July 1994, on a colony of glossy swiftlets in a rural area 16km west of the town of Sandakan, Sabah (Northern Borneo) (5°52'N, 117°59'E). The colony, which has been present at this site for over 30 years, was located under a house raised 3m off the ground on stilts. Wooden support beams, to which swiftlet nests were attached, ran the width of the house at 40cm intervals and ran the depth of the house at 400cm intervals, effectively dividing the colony into a number of separate cells (Figure 2.1) (for more details see Francis 1987b). During the 1994 breeding season, the colony consisted of 769 nests (in 20 colony cells).

Within cells, contact between the feathers of birds sitting on neighbouring nests was observed. Adult birds nesting within the same cell also made contact in flight more often (going to and from nests) than with birds nesting in different cells (personal observation). Also, several adult swiftlets were observed alighting on the "wrong" nest within a cell, during which fighting with the resident adult would ensue. Fighting consisted of "grappling" with claws. Thus, the opportunity for horizontal transmission of lice was far greater within cells than between cells. Glossy swiftlets at the study site were host to two species of Amblyceran lice, *Dennyus distinctus* and *Dennyus somadikartai* (Clayton *et al.* in review) (see Chapter 1 for life-history details). The two species could not be reliably identified in the field.

Figure 2.1 Glossy swiftlet colony located beneath the study house. The division of the colony into a number of separate "cells" can be seen clearly.



2.3 Materials and methods

All glossy swiftlet nests at the study site were removed in December 1993. This forced the birds into the synchronous building of nests at the start of the 1994 breeding season (February/March). The density of nests within each colony cell was measured early in the breeding season as soon as all nest building was completed.

All 769 nests present at the colony could be reached with the aid of a 1.5m stepladder, and were numbered using pencil marks on the wooden support beams. Of these nests, three hundred (evenly distributed spatially throughout the colony) were monitored every 4-5 days. Nests were checked, and nestlings examined for lice, between 08:00 and 14:00. During this period adults were normally away foraging. Nestling wing-chord and length of the greater primary underwing coverts were measured with a 15cm chord stick. Nestling body mass was measured to the nearest 0.1g using pesola spring balances (5g and 10g). Nestlings discovered after hatching were aged at first weighing by reference to a sample of precisely aged young (Bryant & Hails 1983). Nestmates were distinguished by clipping the tip of one toe-nail.

Prior to feather emergence young nestlings were examined for lice by visually searching the entire surface of each nestling, which took about 30 seconds. For older nestlings, with emerged feathers, the following protocol was used. First, wing and tail feathers were spread, and the underwing coverts lifted with a pair of forceps. Next the body of the bird was examined, starting with the head and neck region and moving down both the dorsal and ventral surfaces. Feathers were again lifted with a pair of forceps. The whole procedure took approximately 3 minutes. Whenever lice were encountered, their position and sex were noted. This was relatively easy since birds were host to a mean of only 0.93 lice (range = 1 to 6).

As a test of the visual counting technique, 25 pre-fledging glossy swiftlets were examined for lice, all of which were removed with forceps. The birds were then killed

and left over-night in individual sealed paper bags. Amblyceran lice normally leave the body of a dead bird as it cools down (Marshall 1981a), and *Dennyus* spp. are known to do this (Dale Clayton, personal communication). Bags and birds were carefully checked the next day for lice that had been missed.

2.4 Statistical analyses

The following measures of parasite load are used: "Mean intensity" is the number of a particular parasite in a sample of a host species divided by the total number of host individuals in that sample. "Prevalence" is the number of individuals of a host species infected with a particular parasite divided by the number of hosts examined (expressed as a percentage). "Intensity" is the number of individuals of a particular parasite on each infected host.

Local mean nest densities (per square metre) were calculated by dividing the number of nests within each colony cell by 1.6 (the area of each cell = 4m x 0.4m). Analyses of louse loads were performed using the total number of lice on nestlings per nest. To investigate possible confounding trends associated with differing louse loads on different aged nestlings, an age-profile was created using the loads of nestlings in 46 nests monitored on the same day (June 1st). Loads were compared to the emergence of the greater primary underwing coverts, as lice are often found beneath these feathers (personal observation).

To investigate the influence of host density on louse ecology the lice seen within each nest on nestlings aged 28-31 days, over the course of the study, were analysed with respect to local nest density (mean density per colony cell). Negative binomial models (Bliss 1953) were used to characterise observed louse frequency distributions, with the degree of aggregation quantified as k , the positive exponent of the model (Rozsa *et al.* 1996). As aggregation decreases (the variance of the negative binomial approaches

the mean) $k \rightarrow \infty$. Conversely, as aggregation increases, $k \rightarrow 0$ (Shaw & Dobson 1996).

To investigate possible confounding trends associated with differing louse loads in nests with one versus two nestlings, the prevalence (per nest) and intensity (per infested nest) of lice on 28-31 day old nestlings within these two groups were compared. The positions of lice observed on glossy swiftlet nestlings were categorised into four locations on the body of the host (see Figure 2.2): tail, primary, secondary and body feathers. The time period over which 28-31 day old nestlings were monitored was divided into 12 weeks (beginning May 24th).

2.5 Results

TEST OF COUNTING TECHNIQUE. On visual examination of the 25 nestlings involved in the test of the counting technique, 21 lice (16 adult and 5 immature) were seen and removed. After the birds were killed, two more immature lice were found. Thus, as a rough guide, the visual examination picked up 100% of adult lice and 71% of immature lice on 28-31 day old glossy swiftlet nestlings.

LOUSE LOAD VERSUS NESTLING AGE. Louse infestation of nestlings occurred as a step-function at approximately 25 days of age (Figure 2.3), just after the emergence of the greater primary underwing coverts. The relationship between number of lice per nest and mean nestling age was highly significant (Spearman $r_s = 0.55$, $p < 0.001$). Number of lice per nest was also correlated with mean covert length ($r_s = 0.55$, $p < 0.001$). However, when the analysis was restricted to old nestlings only (after the first age of infestation), there was no significant relationship between louse load and either nestling age ($r_s = 0.22$, $p = 0.24$) or covert length ($r_s = 0.21$, $p = 0.27$).

LOUSE POPULATION STRUCTURE. Over the course of the study, 165 lice were observed on 28-31 day old glossy swiftlet nestlings. Of these, 24.85% were immature

Figure 2.2 Glossy swiftlet topography showing the four louse microhabitat regions
(after Chantler & Driessens 1995).

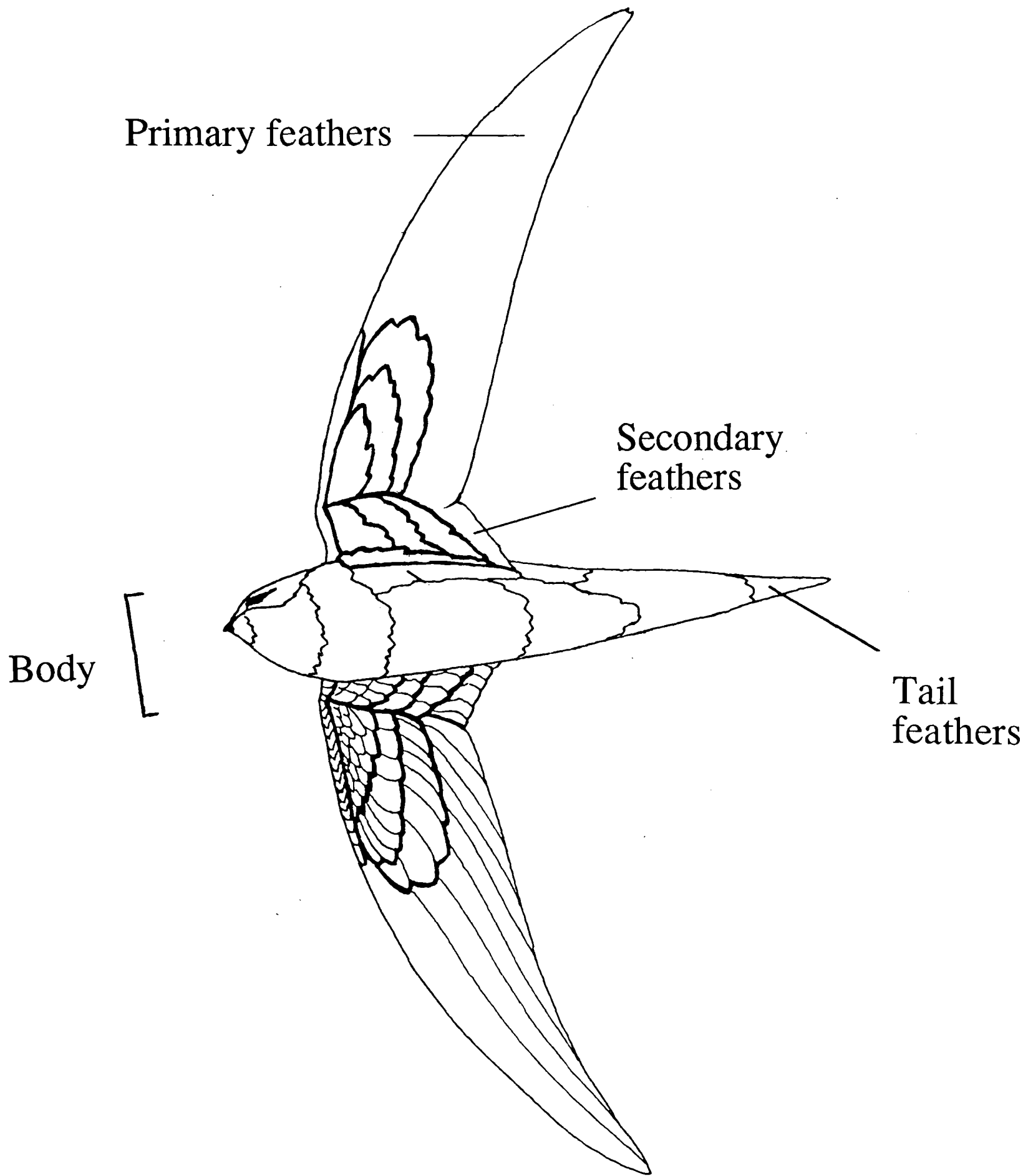
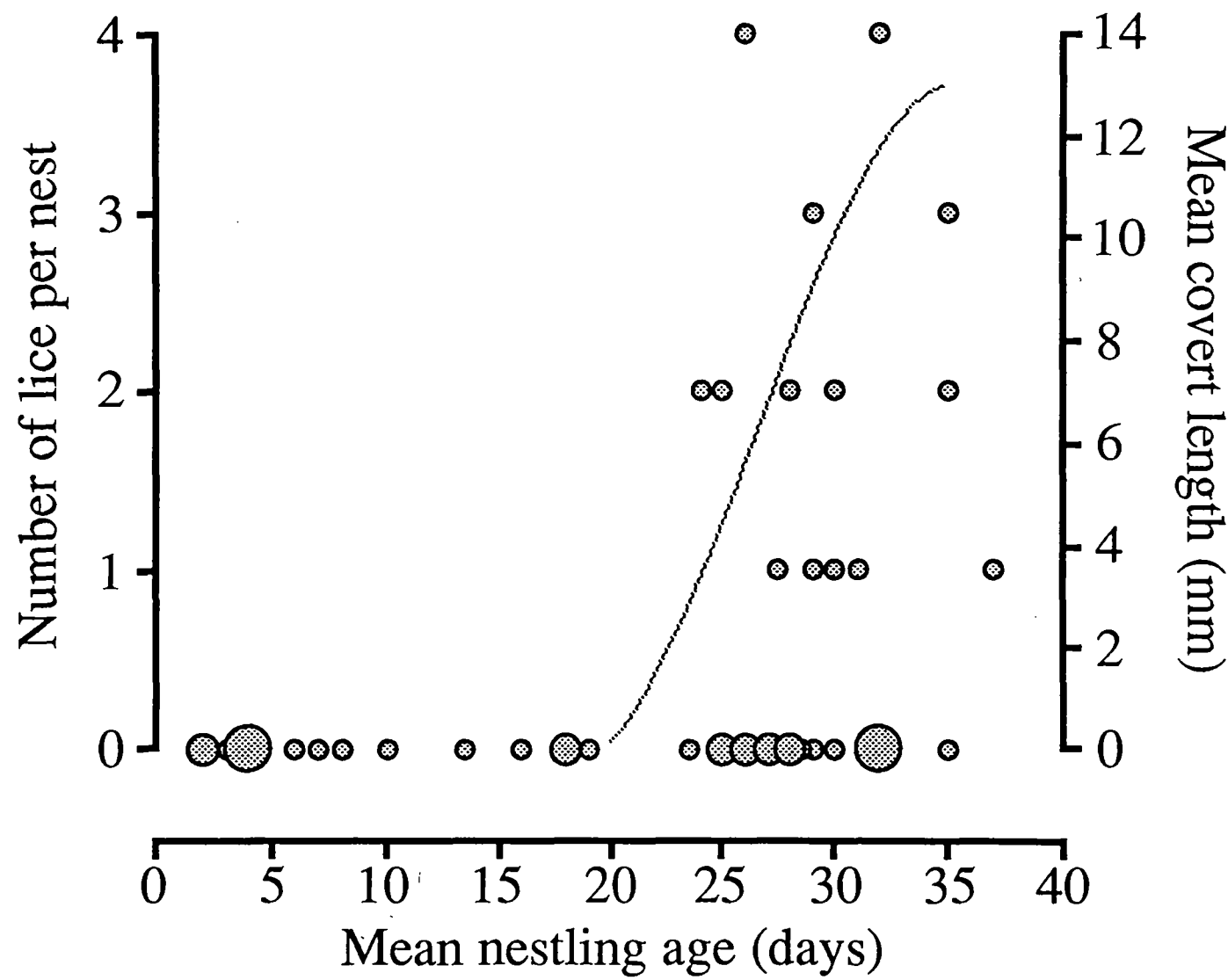


Figure 2.3 Relationship of nestling louse load and emergence of the greater primary underwing coverts to age (n = 46 nests). Number of lice per nest is indicated by points, covert emergence by a best-fit curve. Diameter of points is proportional to the number of nests (1-3).



and 75.15% were adult. Of the adult lice, 45.16% were male and 54.84% were female (not significantly different from a 1:1 ratio; $\chi^2 = 0.58$, d.f. = 1, $p = 0.45$). Lice were seen mainly on the primary feathers, but also on the tail, secondary and body feathers (Figure 2.4). There was a significant difference in the microhabitat distributions of immature versus adult lice. Immature lice were found more often on body feathers (Fisher exact $p = 0.01$) and less often on tail feathers (Fisher exact $p = 0.05$) than adult lice. There was a non-significant trend for female lice to be found less often on secondary feathers than male lice ($\chi^2 = 2.94$, d.f. = 1, $p = 0.09$).

Of the 300 nests monitored, 177 (in 18 colony cells) contained nestlings aged 28-31 days old over the course of the study. No nest had more than one brood of 28-31 day old nestlings during this time. The incidence of lice among host nests, on nestlings of this age, followed an aggregated distribution (Figure 2.5) that was not significantly different from a negative binomial with an arithmetic mean of 0.93 and a positive exponent of 1.76 ($\chi^2 = 3.02$, d.f. = 4 [counts for nests with more than 3 lice were lumped], $p = 0.56$). Louse prevalence (per nest) was 53.7%, and intensity (per infested nest) was 1.74 ± 1.12 (s.d.). There was no difference in the louse infestation of nests with one ($n = 125$ nests) versus two ($n = 52$ nests) nestlings. Prevalence was 52% in nests with one nestling versus 57.7% in nests with two nestlings (Mann-Whitney $U = 3065$, $p = 0.55$). Intensity was 1.74 ± 1.16 in nests with one nestling versus 1.73 ± 1.05 in nests with two nestlings ($U = 970.5$, $p = 0.97$).

LOUSE POPULATION DYNAMICS. Neither prevalence ($r_s = 0.18$, $p = 0.56$), nor intensity ($r_s = 0.01$, $p = 0.97$) of lice, on 28-31 day old glossy swiftlet nestlings, showed any consistent trend over time (Figure 2.6). There was no significant change in either the ratio of immature to adult lice ($r_s = -0.25$, $p = 0.41$) or the sex ratio of adult lice ($r_s = -0.19$, $p = 0.54$), on 28-31 day old glossy swiftlet nestlings, over the course of the study (Figure 2.7).

Figure 2.4 Microhabitat distribution of immature, male and female lice on 28-31 day old glossy swiftlet nestlings (n = 165 lice).

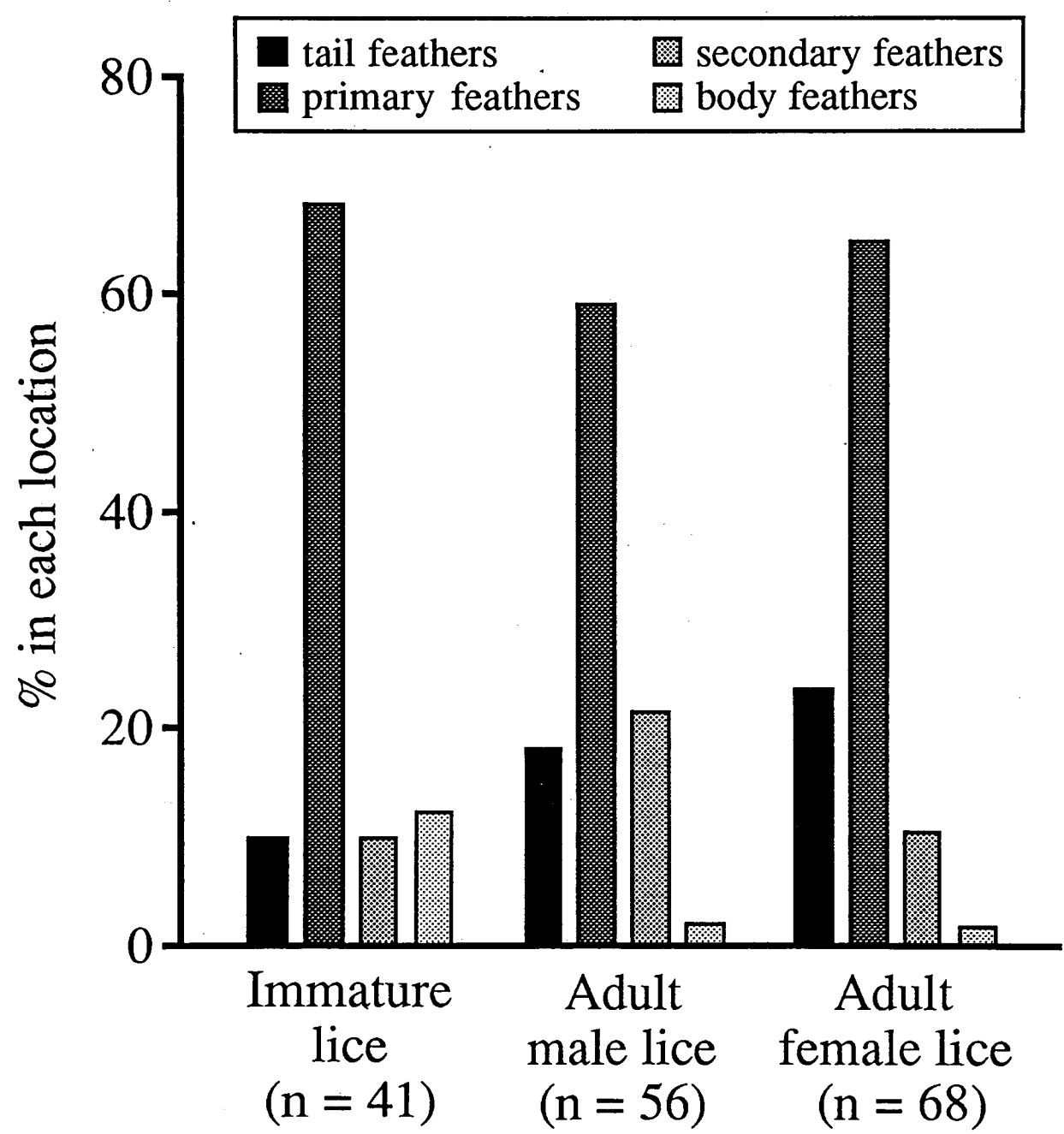


Figure 2.5 Frequency distribution of lice per nest, on 28-31 day old glossy swiftlet nestlings (n = 177 nests). Asterisks indicate predicted values from a negative binomial model with an arithmetic mean of 0.93 and a positive exponent of 1.76.

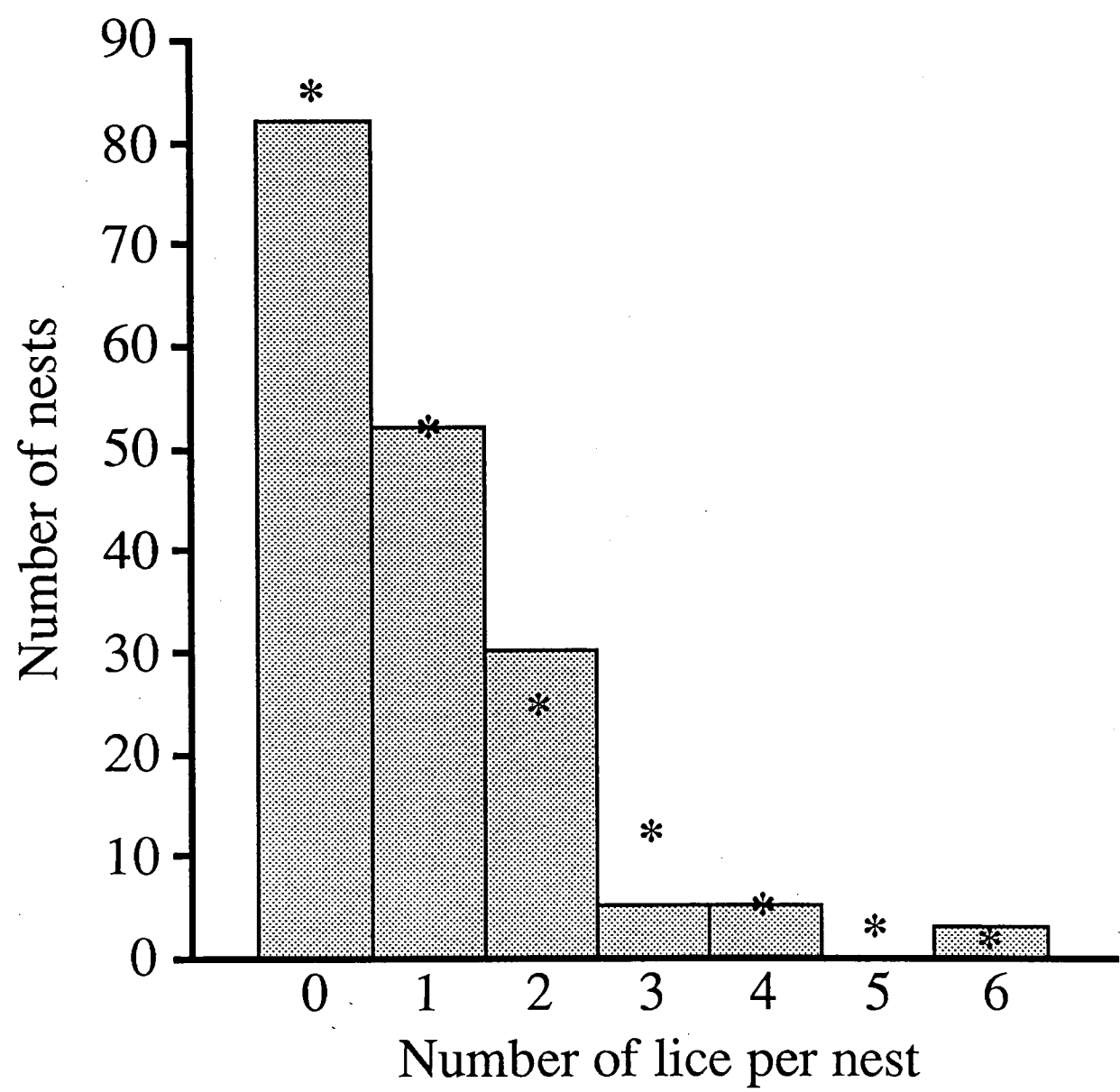


Figure 2.6 a) Prevalence (per nest) and b) intensity (per infested nest) of lice, on 28-31 day old nestlings, over the course of the study (n = 12 weeks).

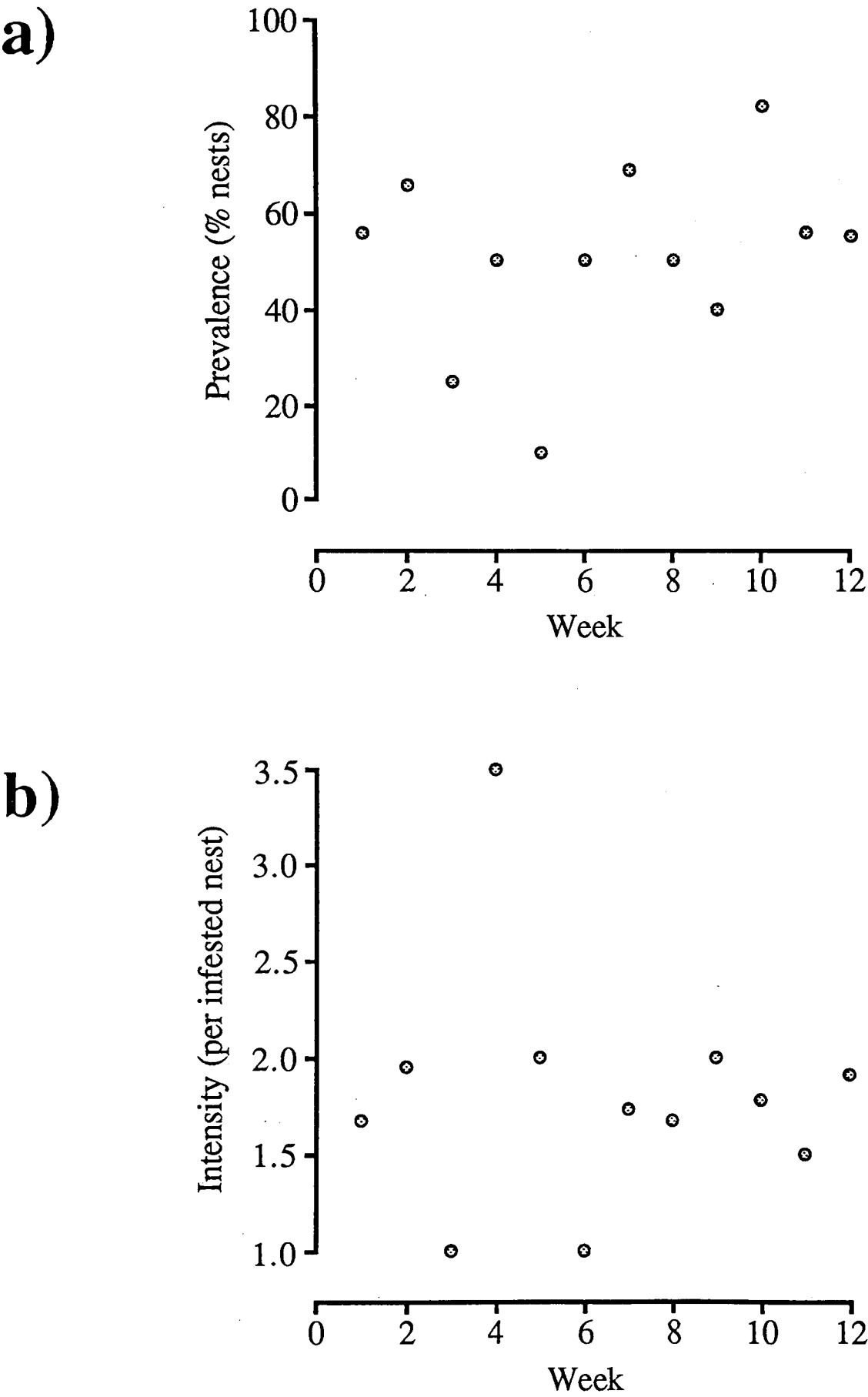
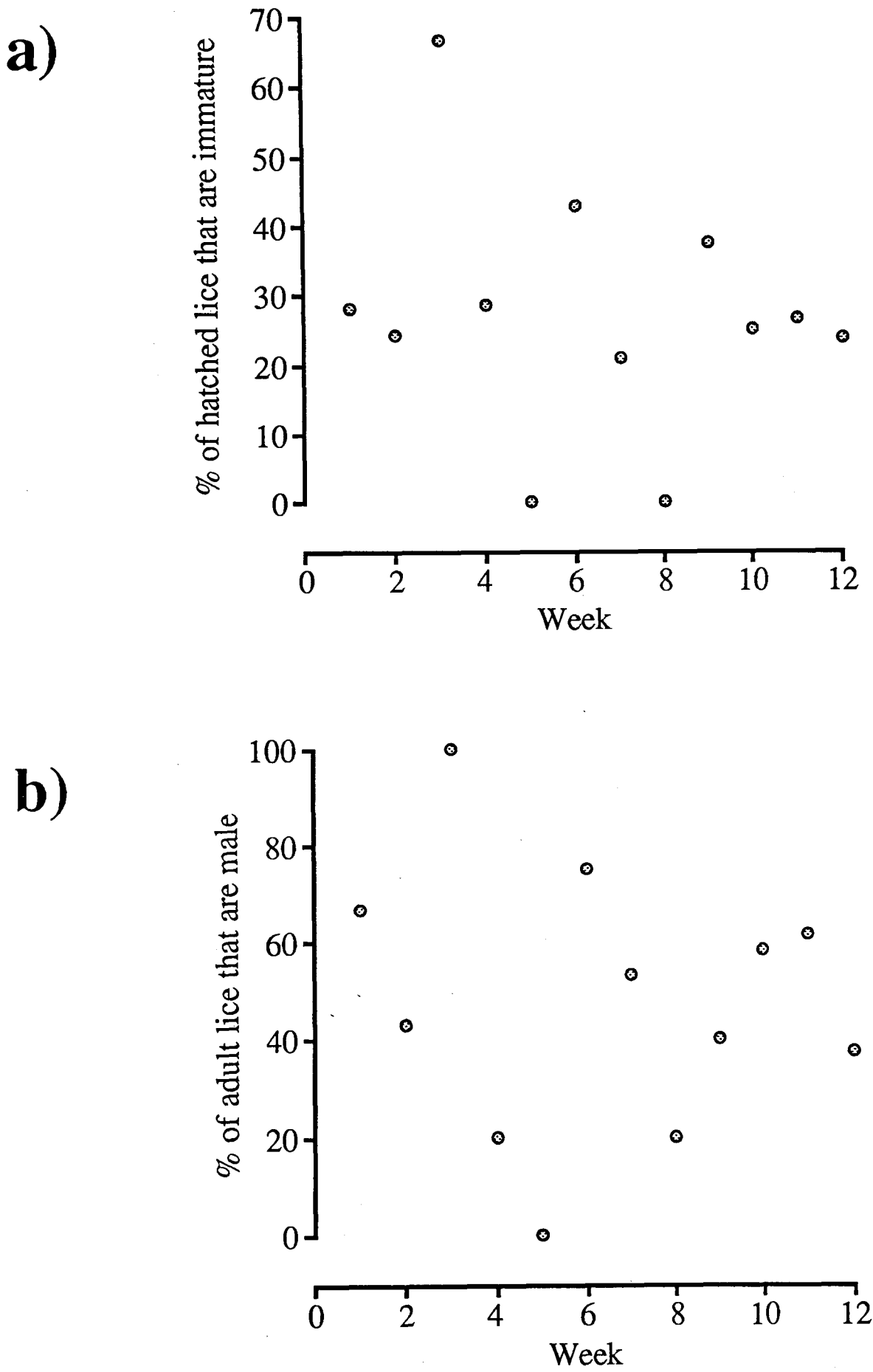


Figure 2.7 a) The ratio of immature to adult lice and b) the sex ratio of adult lice, on 28-31 day old glossy swiftlet nestlings, over the course of the study (n = 12 weeks).



INFLUENCE OF HOST DENSITY. Within the 18 colony cells there was an approximately ten-fold range of nest densities (from 6.25 to 61.25 nests per square metre). The distribution of lice among host nests in the nine denser colony cells (mean density of 36.17 nests per square metre) had a lower degree of aggregation than that among nests in the nine less dense cells (mean density of 19.76 nests per square metre) (Figure 2.8). The best-fit negative binomial for the high-density distribution had an arithmetic mean of 1.16 and a positive exponent of 3.42, compared to an arithmetic mean of 0.57 and a positive exponent of 0.68 for the low-density distribution ($\chi^2 = 16.74$, d.f. = 3 [counts for nests with more than 2 lice were lumped], $p < 0.001$). Across the 18 colony cells, local nest density was highly correlated with louse prevalence ($r_s = 0.74$, $p = 0.002$) (Figure 2.9a). However, there was no significant relationship between louse intensity and local nest density ($r_s = 0.25$, $p = 0.34$) (Figure 2.9b).

Louse population structure in the nine denser colony cells was equivalent to that in the nine less dense cells (Figure 2.10). There was no significant difference in the ratio of immature to adult lice ($\chi^2 = 0.45$, d.f. = 1, $p = 0.48$), or the sex ratio of adult lice ($\chi^2 = 0.01$, d.f. = 1, $p = 0.98$). Likewise, across the 18 colony cells, there was no relationship between either the ratio of immature to adult lice ($r_s = 0.03$, $p = 0.92$), or the sex ratio of adult lice ($r_s = 0.32$, $p = 0.22$), and local nest density (Figure 2.11). However, there was a significant increase in the percentage of adult lice that were male with increasing mean louse intensity per nest (arithmetic mean) ($r_s = 0.62$, $p = 0.02$), although the ratio of immature to adult lice was unaffected ($r_s = -0.24$, $p = 0.36$) (Figure 2.12).

Figure 2.8 Frequency distribution of lice per nest, on 28-31 day old glossy swiftlet nestlings, in a) the nine denser colony cells (n = 110 nests) and b) the nine less dense colony cells (n = 67 nests). Asterisks indicate predicted values from negative binomial models.

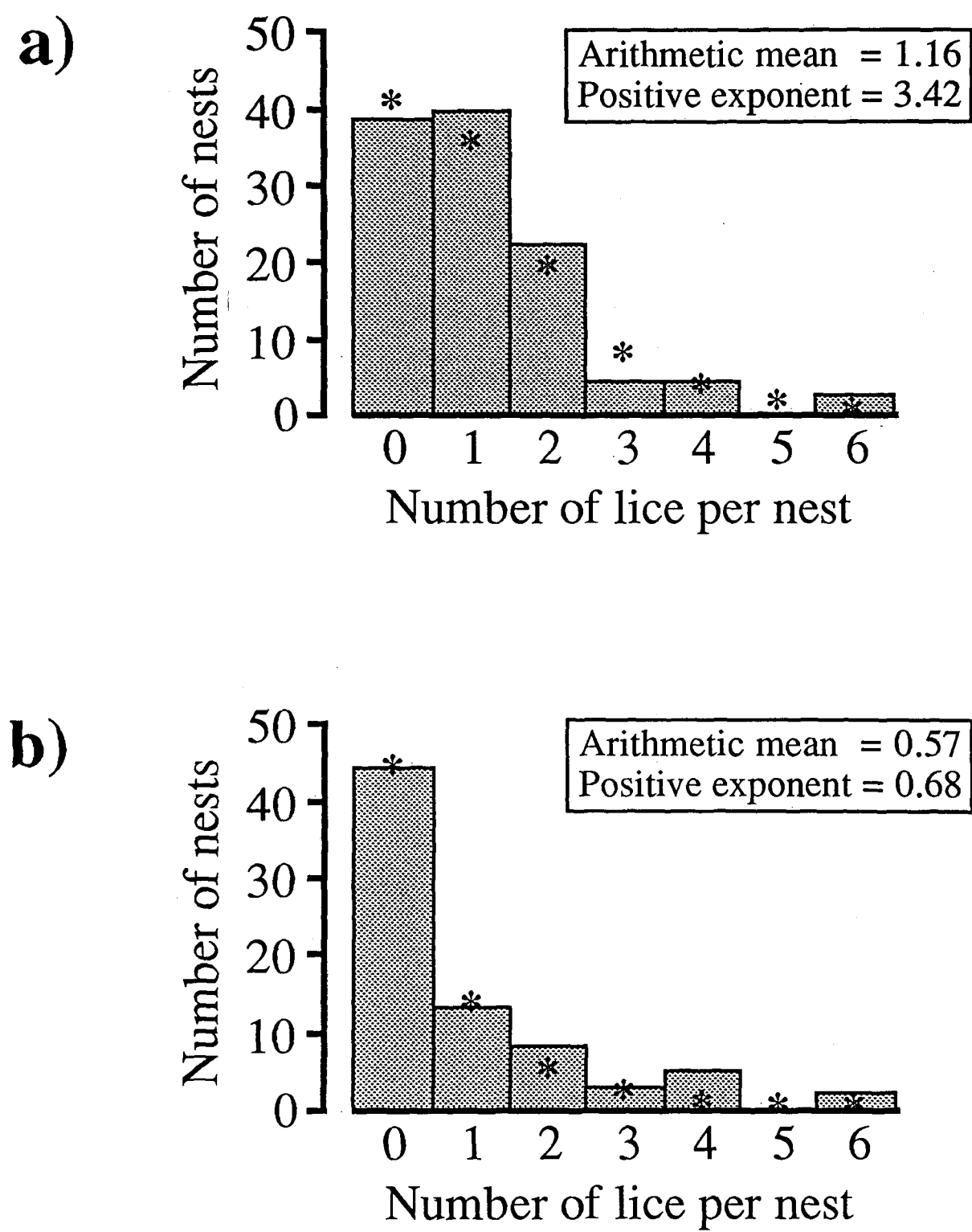


Figure 2.9 Relationship between a) prevalence (per nest) and b) intensity (per infested nest) of lice, on 28-31 day old glossy swiftlet nestlings, versus local nest density (n = 18 colony cells).

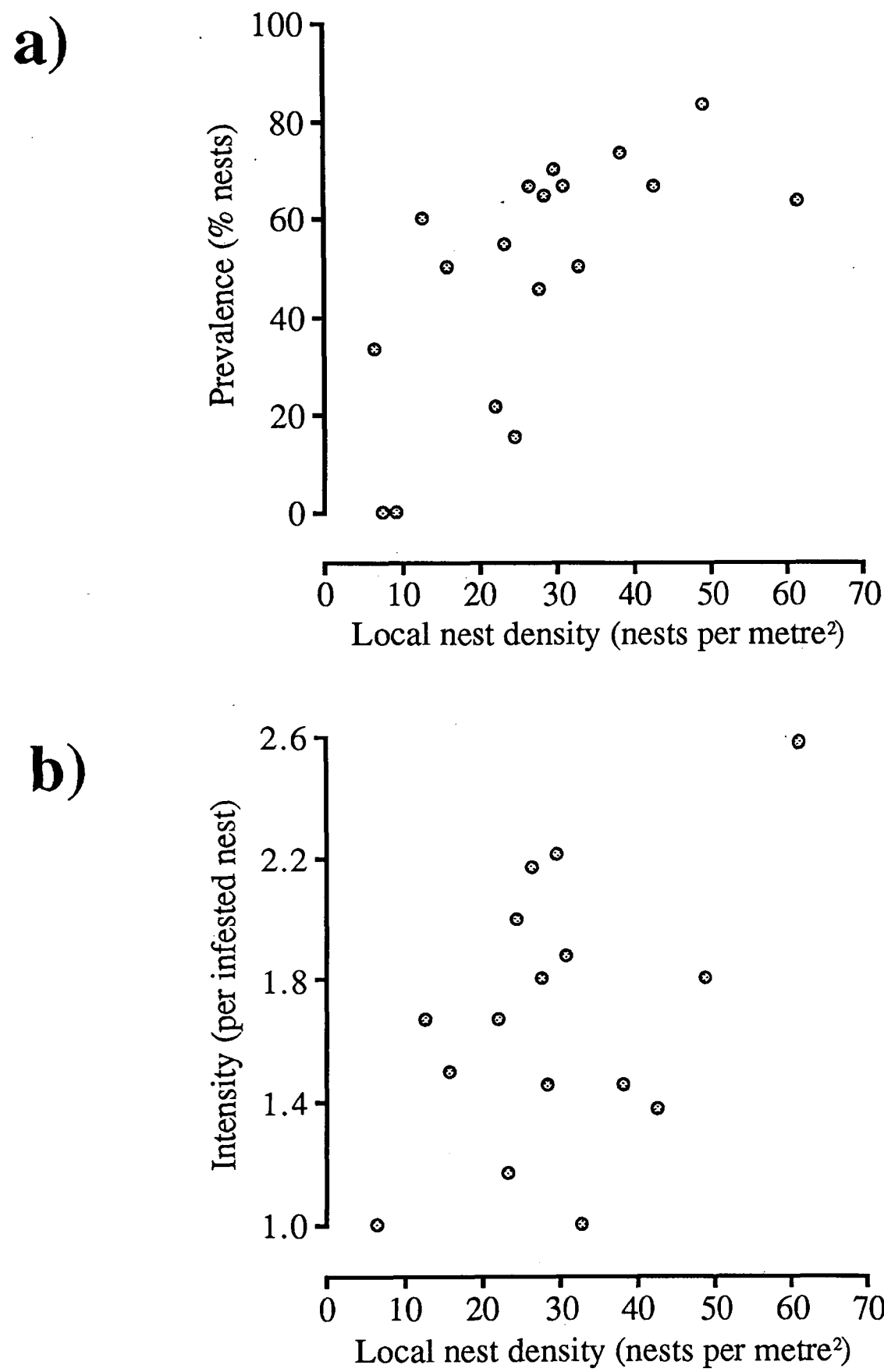


Figure 2.10 Louse population structure, on 28-31 day old glossy swiftlet nestlings, in a) the nine denser colony cells (n = 127 lice in 110 nests) and b) the nine less dense colony cells (n = 38 lice in 67 nests).

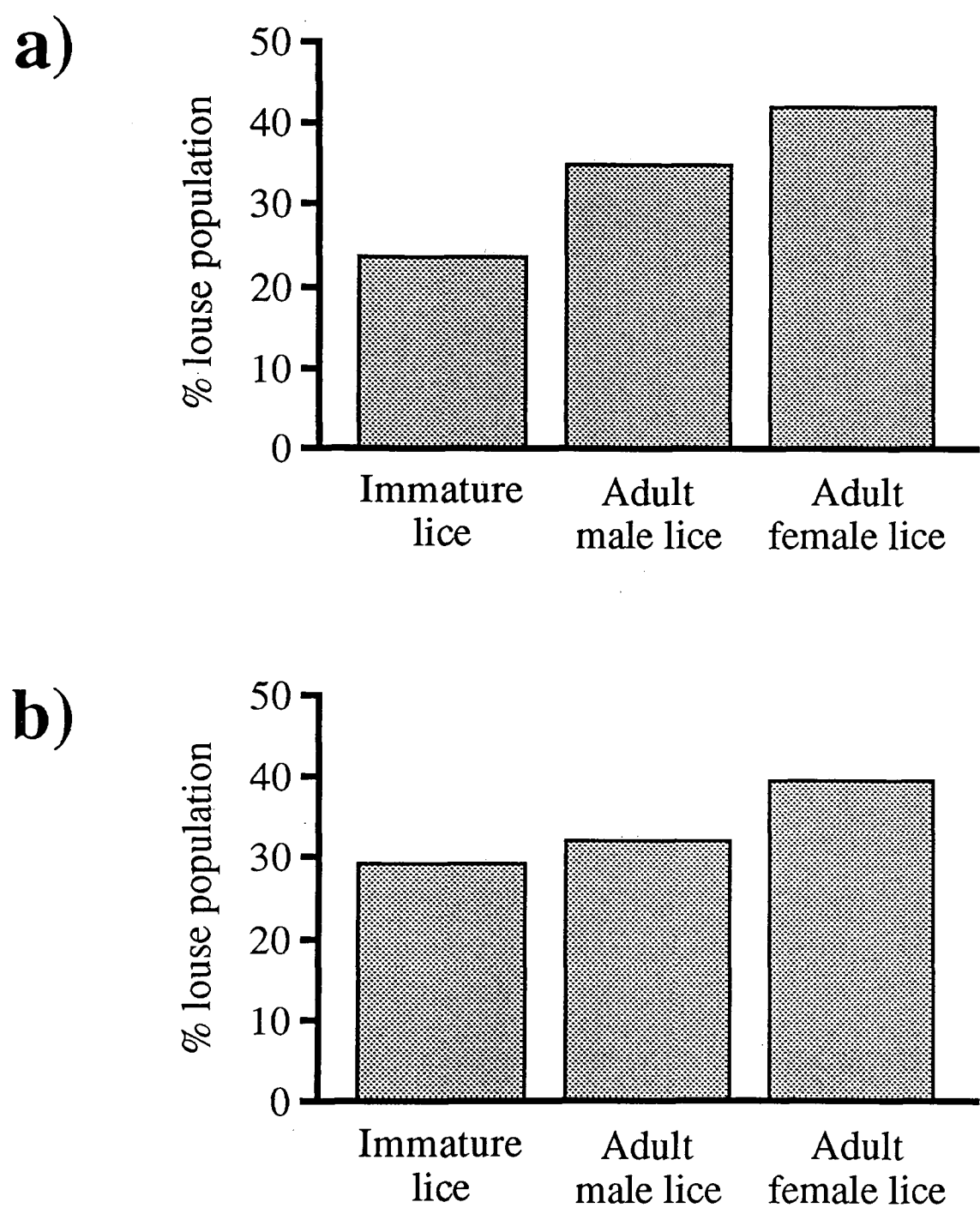


Figure 2.11 Relationship between a) the ratio of immature to adult lice and b) the sex ratio of adult lice, on 28-31 day old nestlings, versus local nest density (n = 16 colony cells (2 cells contained no lice)).

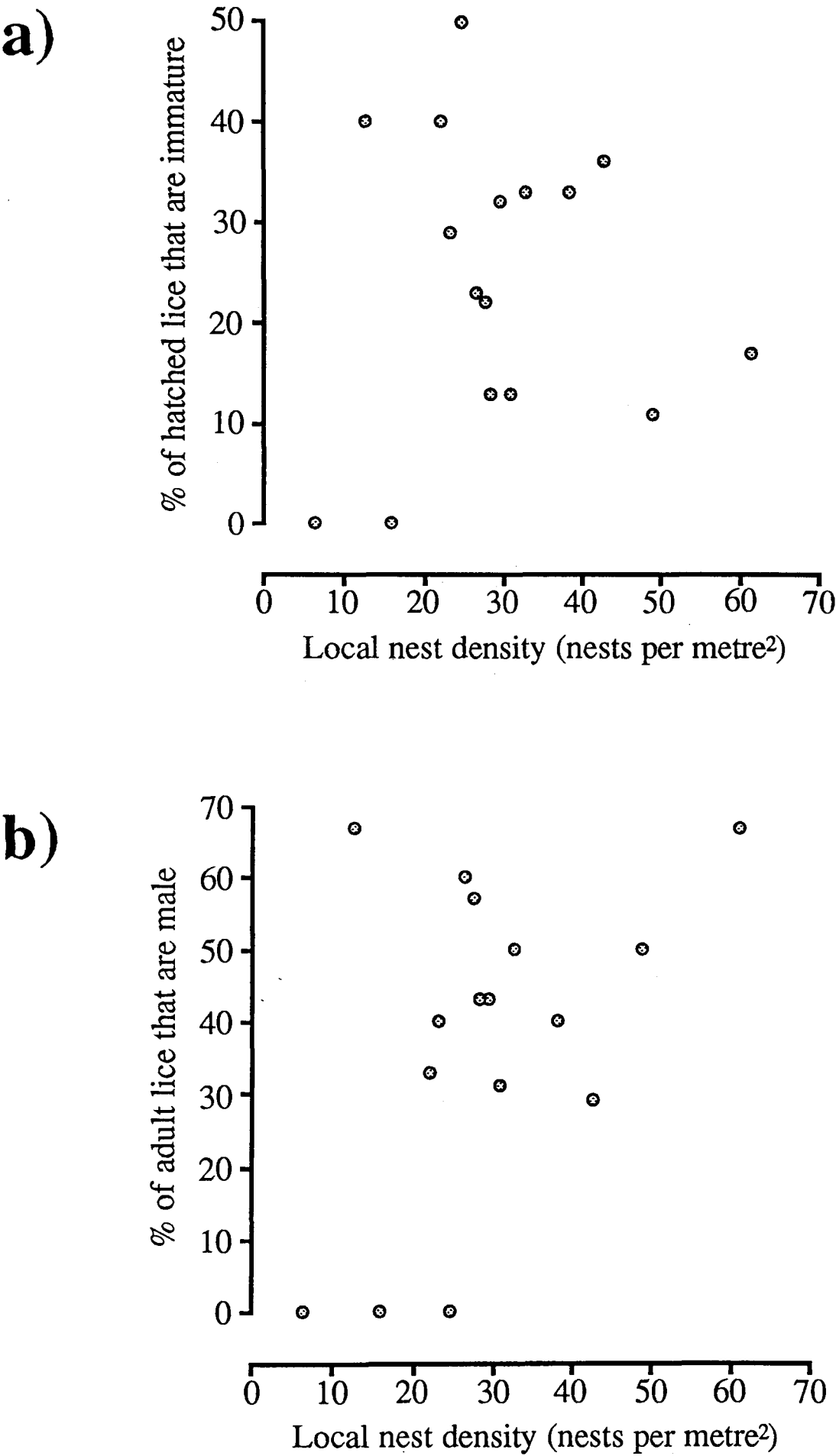
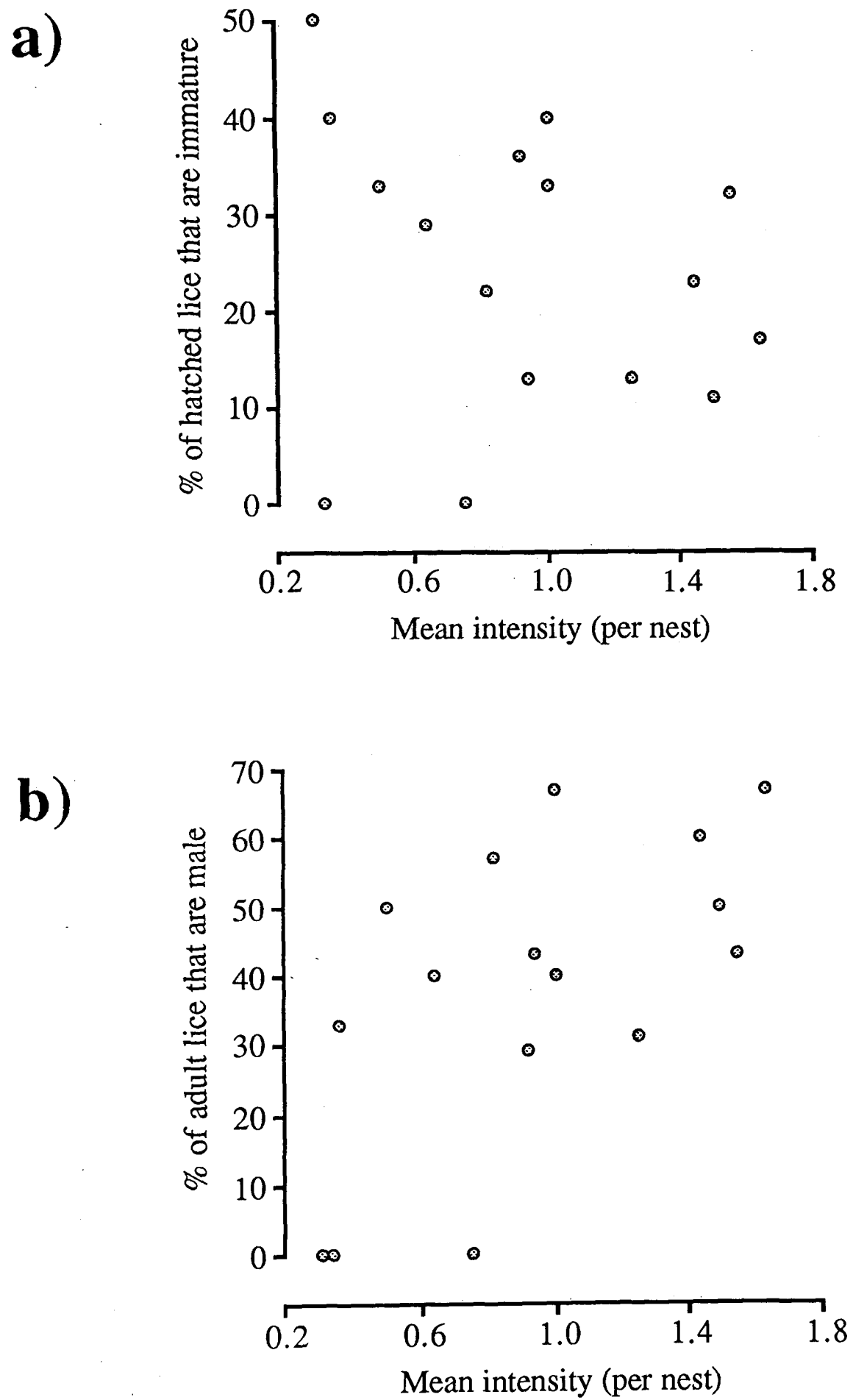


Figure 2.12 Relationship between a) the ratio of immature to adult lice and b) the sex ratio of adult lice, on 28-31 day old nestlings, versus mean intensity of louse infestation (n = 16 colony cells (2 cells contained no lice)).



2.6 Discussion

The visual method for counting lice on 28-31 day old glossy swiftlet nestlings was accurate for counts of adult lice but probably underestimated the loads of immature lice by at least 30%. This was perhaps due to the difference in microhabitats of immature and adult lice (Figure 2.4). Adult lice on nestling swiftlets were usually found on the wings and tail, with a distinct preference for primary feathers, where it is relatively easy to see the lice. Immature lice were more likely to be found among the denser body feathers where lice, especially small immatures, are more likely to be missed when counting. However, since the same counting technique was used throughout the study, the bias in its accuracy would not have effected the comparative results.

The vertical transmission of lice from parents to nestling swiftlets did not occur until the greater underwing primary coverts on the wings of nestlings had begun to emerge at approximately 25 days of age (Figure 2.3). At this stage the primary feathers are approximately half emerged from their protective sheaths (personal observation). Lice on swiftlets may be dependent on the emergence of underwing coverts for protection; they are usually found hidden beneath these feathers (whilst sitting on the primaries). Although swiftlet nestlings do not fly, they vigorously exercise their wings prior to fledging, which may threaten lice with dislodgement (personal observation). After the age of first infestation there was no relation between nestling age (or covert length) and louse load. Restricting all comparative analyses to louse load data for 28-31 day old nestlings avoided a confounding influence of nestling age.

The overall incidence of lice among host nests followed an aggregated distribution (Figure 2.5), as reported for many other species of lice (Eveleigh & Threlfall 1976; Fowler & Williams 1985; Fowler & Hodson 1991; Clark *et al.* 1994; Clayton & Tompkins 1995), including *Dennyus hirundinis* on the common swift (*Apus apus*) (Lee & Clayton 1995). The aggregated distribution of lice on glossy swiftlet nestlings

was not significantly different from a negative binomial. This is typical of contagious parasites where the distribution is determined by a combination of factors including heterogeneity of host behaviour, effective immunity of hosts, direct reproduction within hosts, and spatial heterogeneity in sources of transmission (Anderson & Gordon 1982). Any of these factors may have contributed to the aggregated distribution of lice in this study.

Both the prevalence (per nest) and intensity (per infested nest) of lice were independent of the number of nestlings present. This is not surprising since lice were being transmitted to nestlings from two breeding adult swiftlets at each nest. If parasite loads had been calculated as the mean on nestlings there would have been confounding negative relationships between load and number of nestlings per nest. There was no consistent trend in either the prevalence or intensity of lice on glossy swiftlet nestlings over the course of the study (Figure 2.6). Since there was also no significant change in louse population structure over the course of the study (Figure 2.7), there was no possibility of temporal trends confounding comparisons of lice at different host densities.

Lice on nestlings in denser colony cells occurred in a less aggregated frequency distribution than that of lice on nestlings in less dense colony cells (Figure 2.8). This was due to an increase in louse prevalence with increasing nest density (Figure 2.9a). However, there was no significant relationship between louse intensity and nest density (Figure 2.9b). These results imply a direct effect of host density on louse transmission. If time taken for successful transmission between hosts influences louse survival (i.e. through depletion of stored resources), a higher rate of contact among swiftlets nesting closer together may reduce louse mortality during transmission. This would result in lice being present in more nests (increased prevalence) without necessarily increasing louse intensity (per infested nest).

Louse population structure in denser colony cells was no different to that of lice in less dense colony cells (Figure 2.10). There was no significant relationship between either the ratio of immature to adult lice, or the sex ratio of adult lice, and local nest density (Figure 2.11). However, the sex ratio of adult lice was significantly correlated with mean louse intensity (per nest) within colony cells (Figure 2.12). Sex ratios became less female biased with increasing mean louse intensity. This result may also be due to louse transmission. If male lice are the dispersing sex (with the cue for dispersal being absence of female lice to mate with), then male lice in colony cells where overall mean louse intensity is low would be more likely to transmit between individual hosts, searching for female lice, than male lice in colony cells where overall mean louse intensity is higher. The associated male mortality during transmission would cause the increase in the female-bias of observed sex ratios. Host density may exacerbate this effect since low mean louse intensity is associated with low glossy swiftlet nest density (Figure 2.8), where greater louse mortality during transmission may occur (see above).

This study, of the influence host density has on louse ecology, was carried out with one colony of one host species, where louse adaptations to specific levels of host density were most likely precluded by the dynamic nature of the colony. Observed patterns were most likely due to variation in host density influencing the parameters of louse population dynamics. Thus, inter-specific comparisons of host density on louse population ecology, where lice on denser host populations are less aggregated, more prevalent, and less female-biased (such as Rozsa *et al.* (1996)), cannot be taken as evidence for "louse adaptations to particular social systems". The same results were observed here for solely non-adaptive reasons.

Chapter 3 Blood parasites of Malaysian swiftlets, including a new species of *Hepatozoon* (Apicomplexa: Haemogregarinidae)

[Tompkins, D.M., Anwar, M. & Clayton, D.H. Submitted. Blood parasites of Malaysian swiftlets, including a new species of *Hepatozoon* (Apicomplexa: Haemogregarinidae). *Journal of Parasitology*.]

3.1 Summary

Blood smears were taken from four species of adult and nestling Malaysian swiftlets (Aves: Apodidae): the glossy swiftlet (*Collocalia esculenta cyanoptila* Oberholser 1906), the black-nest swiftlet (*Aerodramus maximus lowi* Hume 1878), the white-nest swiftlet (*Aerodramus fuciphagus vestitus* Thunberg 1812) and the mossy-nest swiftlet (*Aerodramus salanganus natunae* Streubel 1848). No parasites were detected in smears from white-nest or mossy-nest swiftlets. Glossy swiftlet adults at Ampang Reservoir, Kuala Lumpur (Peninsular Malaysia) were infected with *Lankesterella* Labbe 1899 (Apicomplexa: Lankesterellidae) (22% prevalence) and *Hepatozoon* n. sp. (Apicomplexa: Haemogregarinidae) (11% prevalence). Black-nest swiftlet adults at Gomantong Caves in Sabah (Northern Borneo) were infected with microfilaria (7% prevalence). No blood parasites were observed in smears from nestling swiftlets. We speculate that ectoparasites may have been vectoring the blood parasites among host individuals. There was an apparent relation between host nesting density and blood parasite prevalence.

Keywords: Apodidae, haematozoa, *Lankesterella*, microfilaria, sporozoa.

3.2 Introduction

Birds are host to a wide variety of blood parasites, including intra-cellular sporozoa, filarial nematodes, and flagellated trypanosomes (Atkinson & van Riper III 1991; Janovy in press). Intra-cellular sporozoa complete part of their life-cycle within circulating blood cells. Filarial nematodes live in the tissues or peritoneal cavity of their hosts and release larval microfilaria into the blood circulation. Flagellated trypanosomes divide and circulate widely in the bloodstream.

The purpose of this paper is to present the results of a survey of blood parasites of Malaysian swiftlets (Aves: Apodidae). The swiftlets (Collocaliini) are small aerial insectivorous birds that are distributed from the Indian Ocean, through South East Asia and Northern Australia, to the Pacific (Chantler & Driessens 1995). Most species of swiftlets breed in caves and have the ability to echolocate for navigational purposes (Medway 1966). The nests of some species are highly prized for Chinese birds' nest soup (Lee *et al.* in press). As part of a long term study of swiftlet parasites, we examined the following four species for blood parasites: glossy swiftlet (*Collocalia esculenta cyanoptila* Oberholser 1906), black-nest swiftlet (*Aerodramus maximus lowi* Hume 1878), white-nest swiftlet (*Aerodramus fuciphagus vestitus* Thunberg 1812) and mossy-nest swiftlet (*Aerodramus salanganus natunae* Streubel 1848).

Intra-cellular sporozoa have been reported previously in Malaysia from the glossy swiftlet. Baker *et al.* (1972) reported, in two out of 55 blood films collected, the presence of intra-erythrocytic '*Lankesterella* Labbe 1899 -like' haemogregarines (Apicomplexa: Lankesterellidae; see Desser 1993), as well as the more typical pale-staining intra-leucocytic stages of *Atoxoplasma* Garnham 1950 (Apicomplexa: Atoxoplasmatidae; see Levine 1982). McClure *et al.* (1978) examined 240 blood smears from 11 species of Apodidae. They found intra-cellular sporozoa, identified as *Atoxoplasma* sp., in two glossy swiftlets only. Neither of the above reports contain precise taxonomic descriptions of the parasites collected.

3.3 Materials and methods

Smears were made from blood collected from adult and nestling swiftlets at three locations in Malaysia during January 1994 and May-August 1994 and 1995. Adults were caught with a hand-net and nestlings were taken directly off the nest. Thin blood smears were prepared from small drops of peripheral blood removed from a clipped toenail, then air dried, fixed in 100% ethanol for one minute, and stained with conventional Giemsa's solution (Bennett 1970). Entire smears were examined for microfilaria and flagellated trypanosomes under a phase-contrast microscope at x40 magnification. Smears were then examined for 15 minutes each under x100 magnification (oil immersion) to search for intra-cellular sporozoa.

Smears from black-nest, white-nest, and mossy-nest swiftlets were collected at Gomantong Caves, Sabah (see Francis 1987a). Smears from glossy swiftlets were collected from adult birds at Ampang Reservoir, Kuala Lumpur (see Medway 1969) as well as from adult and nestling birds living under a house near Sandakan, Sabah (see Francis 1987b).

3.4 Results

Overall, blood parasites were detected in only three (1.6%) of 185 blood smears (Table 3.1). Two (22%) of the nine adult glossy swiftlets from Ampang Reservoir were infected with intra-cellular sporozoa that were intra-erythrocytic '*Lankesterella*-like' haemogregarines (Figure 3.1). We follow Baker *et al.* (1972) in identifying these parasites as *Lankesterella* sp.

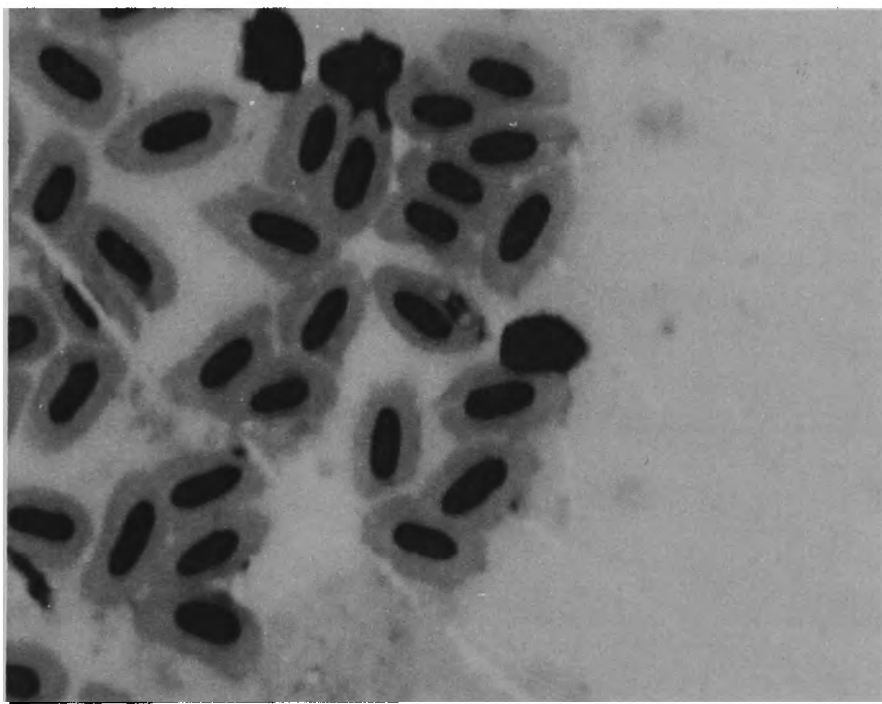
One of the two infected swiftlets from Ampang Reservoir was also infected with intra-leucocytic sporozoa (11% prevalence in adults at this site). Unlike in Baker *et al.*

Table 3.1 Parasites occurring in blood smears from four species of Malaysian swiftlet. Each smear was made from a different individual bird.

Swiftlet species		Number of smears examined	Number of smears containing:		
			Intracellular sporozoa	Filarial nematodes	Flagellated trypanosomes
<i>Collocalia esculenta</i>	adult ^{a,b}	9,15	2,0	0,0	0,0
	nestling ^b	37	0	0	0
<i>Aerodramus maximus</i>	adult ^c	15	0	1	0
	nestling ^c	25	0	0	0
<i>Aerodramus fuciphagus</i>	adult ^c	9	0	0	0
	nestling ^c	25	0	0	0
<i>Aerodramus salanganus</i>	adult ^c	15	0	0	0
	nestling ^c	35	0	0	0

Collection localities: a - Ampang Reservoir, b - Sandakan, c - Gomantong Caves.

Figure 3.1 Photomicrograph of *Lankesterella* sp. from the glossy swiftlet (*Collocalia esculenta cyanoptila*). Magnification x1000.



(1972), where *Atoxoplasma* sp. was reported from the glossy swiftlet, the intra-leucocytic parasite we collected is a *Hepatozoon* Miller 1908 (Apicomplexa: Haemogregarinidae). *Hepatozoon* in birds is generally host specific at the level of the host family or sub-family (Bennett *et al.* 1992). As there is no previous report of *Hepatozoon* from members of the Apodidae, we suspected this parasite to be a new species. This proved to indeed be the case upon closer examination (see below).

One adult black-nest swiftlet from Gomantong Caves was infected with microfilaria (7% prevalence in adults) (Figure 3.2). As we have not seen the adult worm we are unable to identify the species. Microfilaria have never been described from swiftlets. Amongst the Apodidae, microfilaria have been recorded from the common swift (*Apus apus*) (Peirce 1984) and from the little swift (*Apus affinis*) (Bartlett & Anderson 1987).

Flagellated trypanosomes were not observed in any of the blood smears collected.

Lankesterella sp. (Figure 3.1)

The parasite appears in the cytoplasm of host red blood cells as a crescentic body, blunt at one end and pointed at the other, with pale cytoplasm and a belt shaped nucleus spanning the width of the parasite. The sporozoa measured (mean $\pm$ standard deviation) $5.47 \pm 0.39\mu\text{m}$ (range 4.8 - 6.2 μm) by $2.18 \pm 0.22\mu\text{m}$ (range 1.6 - 2.8 μm) (n = 40 parasites).

Hepatozoon n. sp. (Figure 3.3)

The parasite appears in the cytoplasm of host monocytes as an oval-shaped body lying alongside the host cell nucleus (the host cell cytoplasm is rarely seen) with a relatively large ribbon-shaped nucleus occupying the central region of the parasite. The

Figure 3.2 Photomicrograph of microfilaria from the black-nest swiftlet (*Aerodramus maximus lowi*). Magnification x1000.

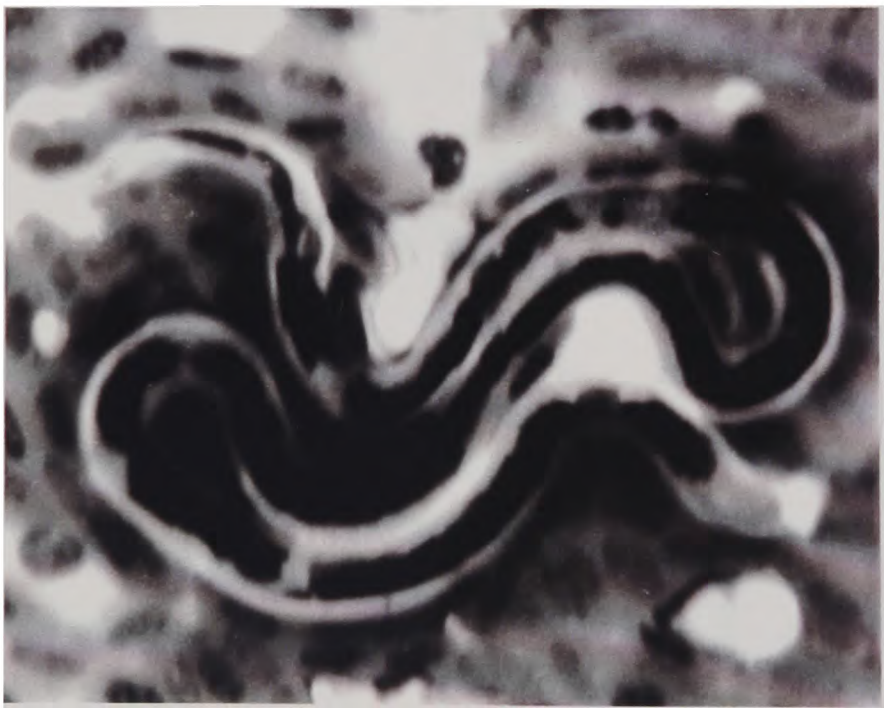
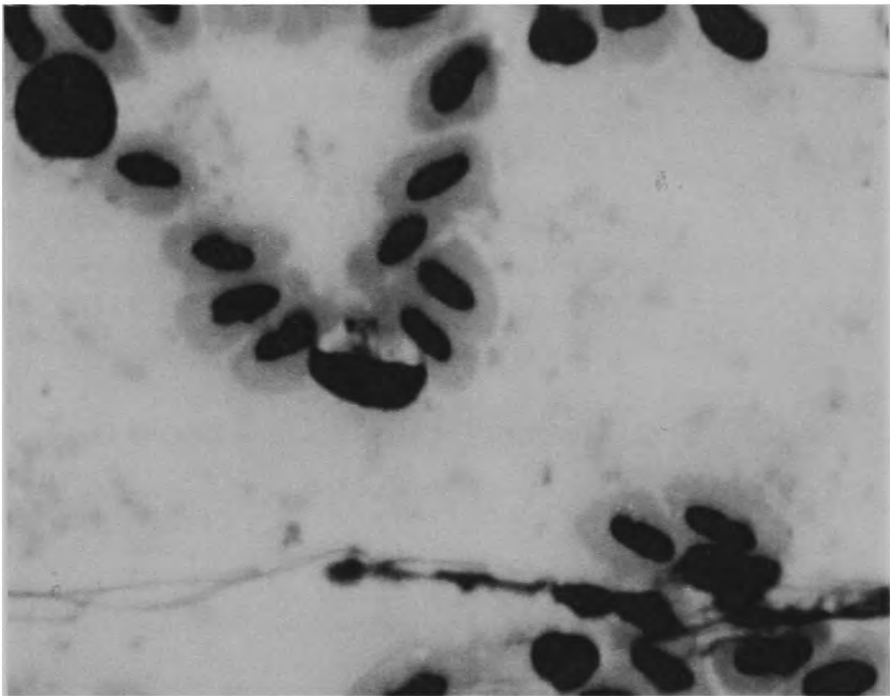


Figure 3.3 Photomicrograph of *Hepatozoon* n. sp. from the glossy swiftlet (*Collocalia esculenta cyanoptila*). Magnification x1000.



sporozoa measured $8.65 \pm 0.96\mu\text{m}$ (range 6.0 - 9.2 μm) by $4.20 \pm 0.55\mu\text{m}$ (range 3.2 - 5.0) (n = 15 parasites).

Taxonomic summary

Type host: *Collocalia esculenta cyanoptila* Oberholser 1906.

Type locality: South-East Asia, Malaysia, Kuala Lumpur, Ampang Reservoir, 3°7'N, 101°42'E.

Type specimens: Thin peripheral blood smears, air dried, fixed in 100% ethanol, and stained with conventional Giemsa's solution.

Prevalence: Two (22%) of nine adult hosts.

Material deposited: Two blood smears in the Natural History Museum, London U.K., no.s xxxx/xxxx.

3.5 Discussion

Malaysian swiftlets are host to two blood-feeding ectoparasites that may act as vectors of the blood parasites reported in this paper (Balashov 1984). The true bug *Paracimex borneensis* Usinger 1959 (Hemiptera: Cimicidae) is common in nests of all four swiftlet species studied (unpublished data). *Crataerina* spp. louse-flies (Diptera: Hippoboscidae) occur in relatively low numbers on all four of the swiftlet species (unpublished data).

Assuming the probability of being infected with blood parasites increases over time, owing to cumulative exposure to vectors, older birds will be more likely to be infected than young birds. This could explain the higher prevalence of blood parasites in adult

versus nestling swiftlets (Table 3.1). With the prevalence in adult birds being so low, the lack of infection in nestlings is not surprising.

High parasite prevalence and intensity in dense host aggregations is a common phenomenon, apparently due to an increase in the efficiency of parasite transmission (Cote & Poulin 1995). We found sporozoa in swiftlets only at the location (Ampang Reservoir) with the greatest nesting density of any of the four swiftlet species studied (personal observation). *Microfilaria* were collected only from the black-nest swiftlet, which has the greatest nesting density of any of the three *Aerodramus* spp. at Gomantong Caves (Francis 1987a). These data suggest that swiftlet blood parasite prevalence is correlated with host nest density. Similarly, both the prevalence and intensity of chewing lice (Phthiraptera: Menoponidae) on nestlings at the glossy swiftlet house colony were significantly correlated with nest density (Tompkins 1996).

Chapter 4 Determinants of host-specificity in bird-louse associations: neutral genetic change or adaptation?

4.1 Introduction

"...without a reasonable understanding of the taxonomic distribution of species diversity and the degree of specialisation among species, our musings over the organisation of communities, the evolution of interspecific interactions, the outcomes of coevolution, and priorities for conservation all have a hollow ring." (Thompson 1994).

The purpose of the work described in this chapter is to identify factors governing the host-specificity of three species of chewing lice on the four species of Malaysian swiftlets (Figure 4.1). The evolution and maintenance of parasite host-specificity is presumably due to restricted gene flow among parasite populations on different hosts (Figure 4.2). Restricted gene flow in this case has two possible major components: parasites may be physically incapable of dispersing to foreign hosts, or parasites may be adapted to specific host characters (specialisation) which prevents them from maintaining viable populations after dispersing to foreign hosts (Lyal 1986).

The first goal of this work was to quantify the degree of host-specificity of swiftlet lice. To do this, a general collection of adult chewing lice was made from individuals of all four swiftlet species to look for incidences of occurrence on the "wrong" host (straggling) (Hopkins 1939; Rozsa 1993). The incidence of straggling lice is relatively common in other studies of bird/louse associations (Kettle 1977; Horning *et al.* 1980; Zonfrillo 1991; Furness & Palma 1992). Cases of straggling imply that lice do occasionally disperse, either passively or actively, to foreign host species.

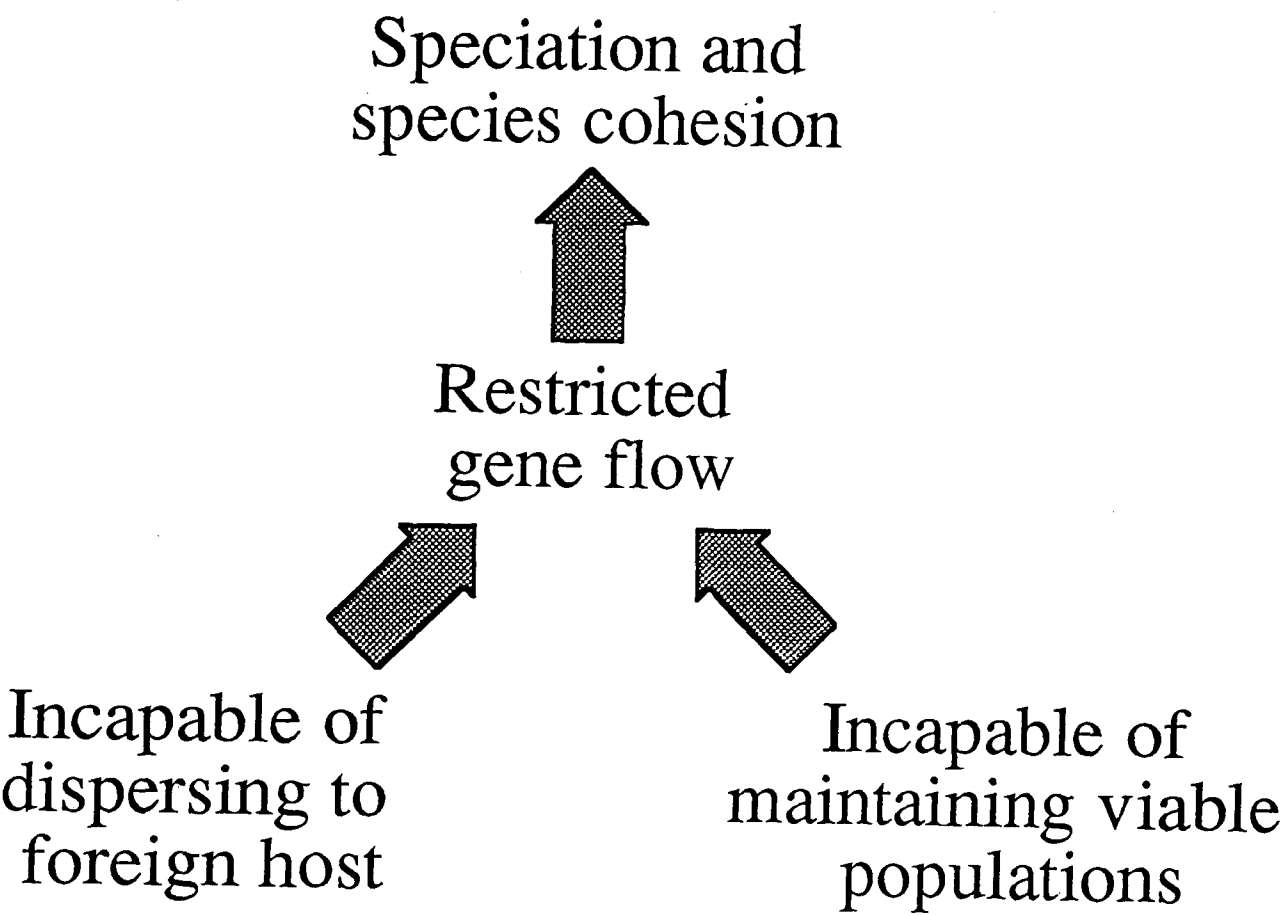
Opportunities exist for both kinds of dispersal at my field site (Gomantong Caves, Sabah; see Francis 1987a). Lice may actively disperse between swiftlet species nesting in close proximity (so close that feathers overlap), or may be passively dispersed as individual birds collide whilst flying to and from nests. Collisions are often observed,

Figure 4.1 Host-specificity, on Malaysian swiftlets, of three species of chewing lice.

Ticks/crosses represent presence/absence of louse species on host species. Louse names are from Clayton *et al.* (in review).

Louse species	Host species			
	<i>Collocalia esculenta</i>	<i>Aerodramus salanganus</i>	<i>Aerodramus fuciphagus</i>	<i>Aerodramus maximus</i>
<i>Dennyus distinctus</i>	√	X	X	X
<i>Dennyus somadikartai</i>	√	X	X	X
<i>Dennyus carljonesi</i>	X	√	√	√

Figure 4.2 General factors contributing to parasite host-specificity and speciation.



especially at dawn and dusk when all 1.5 million swiftlets which live at Gomantong Caves leave and re-enter the complex within short time periods.

The second goal of this work was to test the null hypothesis that host-specific parasites have equal survival on normal and foreign hosts. Experimental transfers of louse species between hosts were carried out to monitor louse survival on foreign, relative to normal, swiftlet species. Transfer experiments have been used widely as a tool to investigate local adaptation of i) plants and animals to their environment (Stalter & Batson 1969; Davies & Snaydon 1976; Turkington & Harper 1979; Lovett Doust 1981; Antonovics & Primack 1982; McGraw & Antonovics 1983; Ayre 1985; Etter 1988; Ayre 1995), ii) insect herbivores to their plant hosts (Edmunds & Alstad 1978; Rice 1983; Wainhouse & Howell 1983; Unruh & Luck 1987; Karban 1989; Cobb & Whitham 1993; Futyma *et al.* 1994; Hanks & Denno 1994; Mopper *et al.* 1995) and iii) parasites, parasitoids, and pathogens to their hosts (Files & Cram 1949; Dawson 1967; Kettle 1977; Parker 1985; Leuchtman & Clay 1989; Lively 1989; Jarosz & Burdon 1991; Ballabeni & Ward 1993; Kraaijeveld & van Alphen 1993; Ebert 1994; Kraaijeveld & van der Wel 1994; Reed 1994; Manning *et al.* 1995; Morand *et al.* 1996). One noteworthy parasite transfer experiment was that conducted by Barbara Downes (1990) in which unionicolan mite species were transplanted between freshwater mussel hosts. The surprising result was that nymphs transplanted to the 'wrong' host metamorphosed into the 'wrong' parasite, demonstrating that the morphological characters used to differentiate these mite species are plastic and host determined. This phenomenon is unlikely to occur in lice, which have a direct lifecycle. However, to safeguard against this possibility, all transfer experiments in this study were carried out using adult lice, which do not moult and therefore do not change in size or shape over time.

The third goal of this study was to investigate whether populations of the same louse species on different host species form "adaptive demes". The "deme formation" hypothesis states that when gene flow is limited, populations within species should

adapt to local environmental characters to optimise fitness, and thus have reduced fitness when transferred into novel environments (Edmunds & Alstad 1978; Karban 1989; Alstad & Corbin 1990; Hanks & Denno 1994; Mopper *et al.* 1995).

Experimental transfers of louse populations between host species were carried out to monitor louse survival on different swiftlet hosts, relative to survival on the host species from which they were transferred.

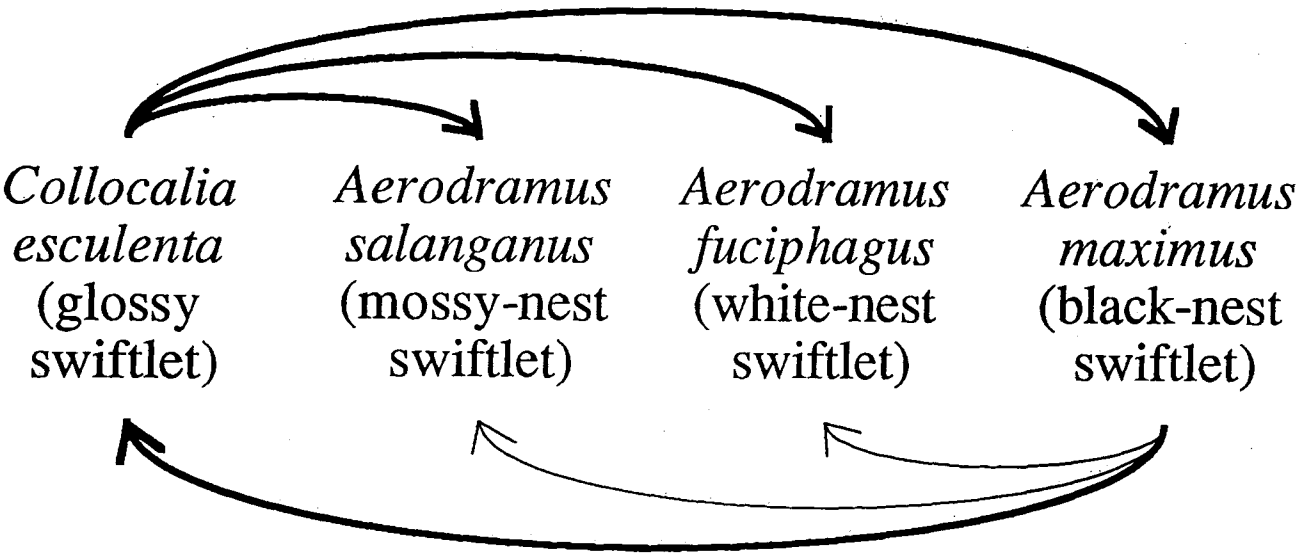
The results of all transfer experiments were compared to host taxonomy (Figure 4.3), to identify the level at which chewing lice are showing adaptations to their hosts (i.e. are there adaptive differences between louse populations and/or louse species and, if so, at what level of host taxonomy are these adaptations occurring).

Parasites transferred to foreign hosts may have a reciprocal effect on host fitness that differs significantly from the effect of a host's resident parasites. For example, foreign parasites - and/or the pathogens they vector - might be more virulent than resident parasites, assuming residents have coevolved some degree of avirulence (Toft & Karter 1990). If this is the case, then foreign parasites conceivably could reinforce reproductive isolation between host species (Price *et al.* 1986). Although controversial (Coyne 1992), reinforcement deserves study because of its potentially important implications as a mechanism of speciation (Wheatley 1980). Few, if any, experimental studies have tested for parasite-mediated reinforcement in natural populations. Thus, the final goal of this study was to compare the impact of normal versus foreign parasites on host fitness.

4.2 Study sites

The study was carried out, March - August 1994 and 1995, at two sites. The first site was the glossy swiftlet colony, located beneath a house near the town of Sandakan, Sabah (Northern Borneo), described in Chapter 2. The second site was Gomantong

Figure 4.3 Planned experimental transfers of chewing lice from two donor swiftlet species (*Collocalia esculenta* and *Aerodramus maximus*) to other host species. Thick arrows indicate transfers of lice to foreign host species, thin arrows indicate transfers of generalist lice between normal host species.



Caves, located 30km south of Sandakan. Gomantong Caves is a large limestone cave complex where all four swiftlet species shown in Figure 4.1 breed (Francis 1987a). The glossy swiftlet nests high (25m+) on the walls and ceiling in well-lit regions near the entrances to the cave complex. The mossy-nest, white-nest and black-nest swiftlets nest in the dark interior. Mossy-nest swiftlets nest at any height, whilst white-nest and black-nest swiftlets only nest high (15m+) on the cave walls and ceiling. All four species tend to nest and roost in mono-specific clusters (Francis 1987a), although some mixing between glossy and black-nest swiftlets, and between white-nest and black-nest swiftlets occurs (personal observation).

4.3 Materials and methods

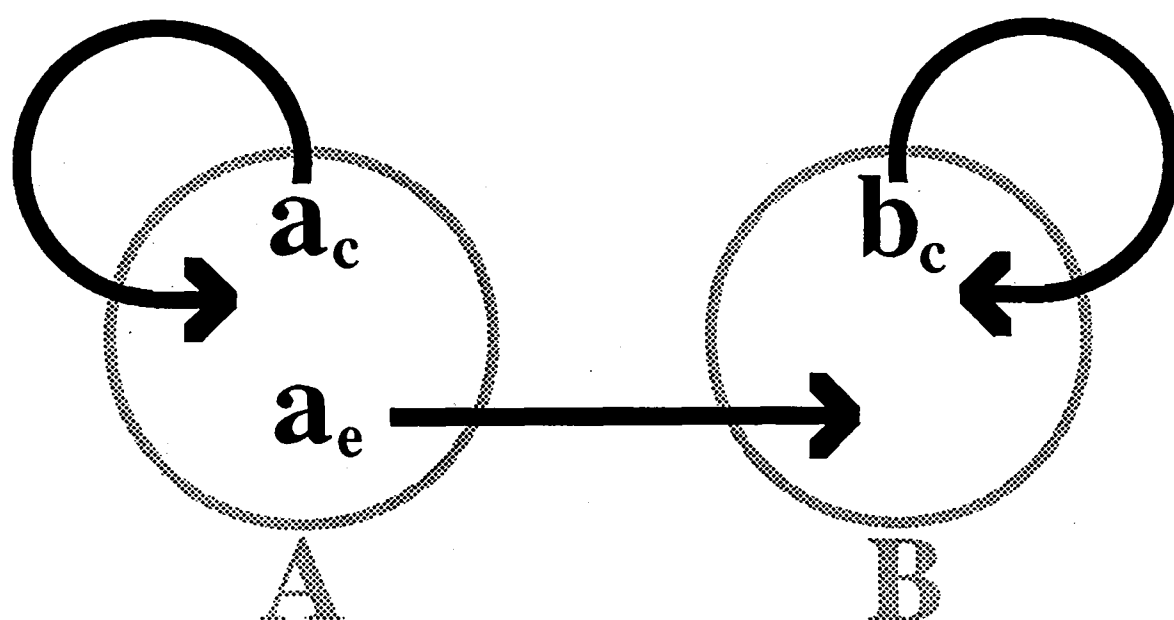
MEASUREMENT OF HOST-SPECIFICITY. Lice were collected from both nestlings and adults of all four swiftlet species involved in the study. Nestlings were removed from nests by hand, adult swiftlets roosting on nests were caught with a hand net. The net measured 40cm in diameter and was lashed to a series of bamboo poles measuring 1-5m in length. At the cave location the nests of mossy-nest, white-nest and black-nest swiftlets were reached with the aid of a 6m surveyor's ladder, three 15m extension ladders and SRT (single rope technique) climbing equipment. Headlamps were used to provide illumination. The surveyor's ladder could be disassembled into four 1.5m pieces, allowing for easy transportation between collecting sites, and was used to collect lice from mossy-nest swiftlets only. The extension ladders, weighing 90 kg each, were used to collect lice from white-nest and black-nest swiftlets. Once an extension ladder was installed at a collecting site it remained there throughout the season (retracted and locked when not in use). The SRT kit was used to abseil through a hole in the ceiling of one of the caves to net black-nest swiftlet adults from nests 60m above the cave floor. Unfortunately, only a few glossy swiftlet nests could be reached at the cave location (with an extension ladder). The majority of collecting from this

species took place at the house location, where hundreds of nests could be reached using a 1.5m step-ladder. Birds were searched for lice as in Chapter 2 (adults were examined using the same method as for feathered nestlings). Hands and nets were carefully checked to prevent accidental transferring of lice between birds.

THE TRANSFER EXPERIMENT. Transfer of lice between hosts was carried out as illustrated in Figure 4.4, with subsequent louse survival and host body mass and survival being monitored. The null hypothesis for effects on parasite survival is $H_0: S_a = S_c$, where S is mean louse survival, the component of parasite fitness measured. Failure to reject this hypothesis would indicate that the parasite is equally adapted to survive on both the normal host (control transfer) and the host to which it was experimentally transferred. The alternative outcome $S_a < S_c$ would imply local adaptation of the parasite to its normal host. The outcome $S_a > S_c$ would imply that the parasite is escaping from resistance mechanisms of its normal host or that it is pre-adapted to some feature of the new host. The null hypothesis for effects on body mass, one component of host fitness measured, is $H_0: MB_a = MB_b$ where MB_x is the mean body mass of host B with parasite x. Failure to reject this hypothesis would indicate that the parasite plays no current role in the reinforcement of reproductive isolation between host species. The alternative outcome $MB_a < MB_b$ would imply a potential role for the parasite in reinforcement of host reproductive isolation. The outcome $MB_a > MB_b$ would imply a potential role for the parasite in facilitating host introgression, which seems unlikely. The null hypothesis for effects of lice on host survival, and related possible outcomes, is parallel to that for effects of lice on host body mass.

TRANSFER OF HOST-SPECIFIC LICE TO FOREIGN HOST SPECIES. The host-specific lice *Dennyus distinctus* and *Dennyus somadikartai* were experimentally transferred from nestlings of their normal host species (glossy swiftlet) to nestlings of the mossy-nest and black-nest swiftlets (foreign host species). Experimental transfers to white-nest swiftlet nestlings were not carried out because not enough nests were accessible. Control transfers back to young nestlings in different glossy swiftlet nests

Figure 4.4 Design of experiment for estimating the effects of parasite transfers on components of the fitness of parasite **a** and host **B**, where **A** and **B** are host species (or populations) and **a** and **b** are the respective parasites normally found on those hosts. Experimental transfers are subscripted with **e** and controls with **c**.



were also carried out. All nests involved in the experiment occurred in mono-specific clusters. *D. distinctus* and *D. somadikartai* were transferred together, each in a proportion relative to their natural loads, as they are indistinguishable in the field. Recaptured lice were identified in the lab upon completion of the experiment (see below).

Marking of transferred lice was attempted using acrylic paint, oil paint, powdered dye (Southwood 1978) and setal clipping (Durden 1983). The use of paint and powder reduced louse survival to near zero. Setal clipping was inaccurate due to high natural variation in louse setal length, and accidental breakage of setae during handling and storage of specimens. Thus, transfer experiments had to be carried out with unmarked lice.

Four adult lice (two males and two females) were moved to singleton nestlings in 25 nests for each group of transfers. Four was the maximum number of lice seen on glossy swiftlet nestlings (of any age) before the start of the experiment. Recipient nests were haphazardly chosen from all the available nests containing singleton nestlings. Lice were transferred to nestlings at the age at which transmission of lice from adults to nestlings normally occurs. Transmission is dependent on nestling feather emergence (see Chapter 2). Natural transmission occurs at approximately 25 days of age for glossy swiftlet nestlings, 30 days of age for mossy-nest swiftlet nestlings, and 35 days of age for black-nest swiftlet nestlings.

Lice were removed from donor nestlings, at the house location, as detailed in Chapter 2. Approximately three hours later lice were placed onto the primary feathers of recipient nestlings (at the cave location for the experimental transfers and at the house location for the control transfers). One male and one female louse were placed on each wing. Ten days later, just before nestlings fledged, all recipient nestlings were examined for lice, as detailed in Chapter 2, and all lice observed were removed and stored in 70% alcohol for subsequent identification. At the same time, singleton

nestlings at 25 unmanipulated glossy swiftlet nests (haphazardly distributed at the house location) were examined for lice to give a measurement of the natural "background" loads of the donor species. These lice were also removed and stored in alcohol. The proportion of each louse species (*D. distinctus* versus *D. somadikartai*) moved in the experimental transfers was assumed to be equal to their relative abundance in the background load collection. The survival of lice in control transfers was estimated as the number of each species recovered minus their relative background loads.

Estimates of survival of transferred lice could conceivably be confounded by back-transmission of lice from nestlings to adults. To check for this possibility, parents of the recipient mossy-nest swiftlet nestlings were caught at the nest and examined for lice at the same time as their nestlings. To investigate whether any horizontal transmission of lice was occurring from recipient nestlings to nestlings in adjacent nests, nestlings at all surrounding nests were also examined for lice over the course of the study. These lice were also removed and stored in alcohol. To measure the ability of lice to survive off any host, 25 adult glossy swiftlet lice (11 females and 14 males) were removed from old nestlings at the house location and kept individually in eppendorf tubes in a closed container at air temperature (26-32°C). Tubes were opened and louse survival was checked every 12 hours until all of the lice died.

IMPACT OF NORMAL VERSUS FOREIGN LICE ON NESTLING GROWTH AND SURVIVAL. Some nestlings disappeared before the ten day period prior to louse recapture was over. These disappearances were of nestlings younger than the minimum fledging age for each swiftlet species (see Chapter 1). They were thus assumed to have died (either fallen out of nests or taken by predators, such as cats or snakes; see Francis 1987a). The impact of normal versus foreign parasites on nestling survival to fledging was assessed by comparing the rate of disappearance of nestlings with normal versus foreign lice.

Body mass of all nestlings involved in the transfer experiments was also measured, at four day intervals, to the nearest 0.1g with a pesola spring balance. All nestlings were ringed prior to fledging with numbered 4mm aluminium split-rings. A modified Richards sigmoid growth model (Brisbin *et al.* 1986) was fitted to the body mass measurements of each nestling:

$$M_{(x+i)} - M_x = \frac{2i(m+1)(a^{(1-m)} M_x^m - M_x)}{T(1-m)} + e$$

where M_x = nestling body mass at time x ,

i = time interval between measurements,

a = asymptotic mass,

T = growing period (until asymptote),

m = Richards shape parameter,

e = stochastic error.

The three parameters (a , m and T) were predicted for each nestling. Near $m = 2.0$, 0.67, 0.0 or when $m \rightarrow 1.0$, the Richards model becomes the logistic, von Bertalanffy, monomolecular or Gompertz models respectively (Richards 1959). A fourth parameter, mean growth rate, was calculated by dividing asymptotic mass by growing period. Measurements of fledging mass and age were taken from the raw data. Parameter values for nestlings with normal versus foreign lice transferred to them were compared using "one-way analysis of variance". Only nestlings that survived to fledge were included in the analyses.

ENDOPARASITE SURVEY. Blood smears and faecal samples were also taken from recipient nestlings of the three host species involved in the transfer experiment, as well as from 25 white-nest swiftlet nestlings. Thin blood smears were prepared from small

drops of peripheral blood taken from a clipped toe-nail, air dried and fixed in 100% ethanol for 1 minute and stained with conventional Giemsa's solution. Faecal samples were collected whenever defaecation was observed, stored in 2.5% potassium dichromate and treated with saturated NaCl solution to float off coccidia. Entire blood smears were examined for microfilaria and flagellated trypanosomes under a phase-contrast microscope at x40 magnification. Smears were then examined for 15 minutes each under x100 magnification (oil immersion) to search for intra-cellular sporozoans. Faecal samples were examined for 5 minutes, under x40 magnification, for coccidia.

ENVIRONMENTAL CONTROL. Although the body of the host represents the environment for chewing lice (Hopkins 1949; Kettle 1977) it is possible that, in transferring lice from donors at the house location to recipients in the cave, louse survival could have been influenced by the new host's environment as well as the new host itself. Both temperature and relative humidity are known to influence chewing louse survival (Marshall 1981a). To investigate whether there was a host environmental component to louse survival, *D. distinctus* and *D. somadikartai* were also experimentally transferred from normal host nestlings at the house (glossy swiftlets) to mossy-nest swiftlet nestlings cross-fostered into glossy swiftlet nests at the house. Control transfers back to young glossy swiftlet nestlings, cross-fostered into different glossy swiftlet nests were also carried out. Transfers were made to singleton nestlings in 15 nests for each group. Fifteen singleton glossy swiftlet nestlings were also cross-fostered between nests, where the louse loads remained unmanipulated, to assess natural "background" loads.

The cross-fostering of nestlings was carried out five days before the transfer of lice, at approximately 20 days of age for glossy swiftlet nestlings and 25 days of age for mossy-nest swiftlet nestlings. Nests containing singleton nestlings to be transferred were removed intact and kept in a closed container at air temperature. Nestlings were transferred into recipient nests two hours later, displacing singleton resident nestlings at the same developmental stage (the latter were removed). Subsequent transfer and

recapture of lice, and measurements of nestling body mass, were carried out as described in the above section.

TRANSFER OF LICE AMONG NORMAL HOST SPECIES. The non-specific louse *Dennysus carljonesi* was experimentally transferred from adult black-nest swiftlets (at the cave location) to young nestlings of the mossy-nest and white-nest swiftlets (all normal host species at the cave). Experimental transfers to glossy swiftlet nestlings (foreign host species) at the house were also carried out, as were control transfers back to young nestlings at different black-nest swiftlet nests. All nests involved in the experiment occurred in mono-specific nest clusters.

Donor lice were removed from adult black-nest swiftlets caught at the nest with a hand-net. Transfer of lice to nestlings was carried out as in the previous sections, at the age of natural transmission of lice from adults to nestlings (approximately 30 days of age for the white-nest swiftlet). Transfers were carried out to singleton nestlings in 15 nests for each group. Subsequent recapture of lice and measurements of nestling body mass were carried out as previously described. Louse survival on each host was estimated as the number of *D. carljonesi* recovered minus the background load on each host species.

4.4 Results

MEASUREMENT OF HOST-SPECIFICITY. A total of 567 adult lice were collected from 1381 host individuals, of which 440 were of the louse species involved in this study (*Dennysus distinctus*, *D. somadikartai*, and *D. carljonesi*) (Table 4.1). Five other louse species were also present on the four species of Malaysian swiftlets involved in this study (a total of 127 lice collected from 1381 host individuals) (see Clayton *et al.* in review). Their low prevalence precluded them from use in any transfer experiments.

Table 4.1 Number of *Dennyus distinctus*, *D. somadikartai* and *D. carljonesi* collected from Malaysian cave swiftlets. Percentages, which are a measure of host-specificity, are the proportion of each louse species that was present on each host species (the mean intensity of each louse species on each host divided by the sum of the mean intensities (of that louse) across all four host species).

Louse species	Host species			
	<i>C. esculenta</i> (N = 240)	<i>A. salanganus</i> (N = 398)	<i>A. fuciphagus</i> (N = 207)	<i>A. maximus</i> (N = 536)
<i>D. distinctus</i> *	35 (98.7%)	—	—	1 (1.3%)
<i>D. somadikartai</i> *	99 (99.5%)	—	—	1 (0.5%)
<i>D. carljonesi</i>	—	23 (5.3%)	177 (77.2%)	104 (17.5%)

* Number of female lice only (male *D. distinctus* and male *D. somadikartai* are indistinguishable; Clayton *et al.* in review). 143 male *D. distinctus*/*D. somadikartai* were also collected from *C. esculenta*.

Only 3% of lice collected from the glossy swiftlet were collected at the cave location. However, the same louse species were present on this host at both collecting locations (house and cave). Single specimens of both *D. distinctus* and *D. somadikartai* were taken from a 'wrong' host (the black-nest swiftlet). Together they accounted for less than 2% of the lice collected from that species (Table 4.1).

LICE TRANSFERRED TO FOREIGN HOST SPECIES. The background louse load on glossy swiftlet nestlings (the normal host) was a mean ($\pm$ s.d.) of 0.56 ± 0.87 (0.28 ± 0.45 females and 0.28 ± 0.53 males). Of the females, 0.08 ± 0.27 were *D. distinctus* and 0.20 ± 0.40 were *D. somadikartai*.

Overall (without reference to particular species of lice), lice transferred from normal to foreign host nestlings had lower recapture rates than lice transferred back onto normal host nestlings (Figure 4.5). Significantly fewer lice were recovered from the mossy-nest swiftlet (foreign host) than from the normal host ($\chi^2 = 14.86$, d.f. = 1, $p < 0.001$). Also, significantly fewer lice were recovered from the black-nest swiftlet (foreign host) than from the mossy-nest swiftlet ($\chi^2 = 21.67$, d.f. = 1, $p < 0.001$). Recapture rates from the black-nest swiftlet were near zero.

There was a significant difference in the recapture of male versus female lice transferred from normal host nestlings to mossy-nest nestlings; fewer males were recovered (Figure 4.6; $\chi^2 = 8.39$, d.f. = 1, $p < 0.005$). Significantly fewer male lice were recovered from foreign hosts than from normal hosts ($\chi^2 = 14.86$, d.f. = 1, $p < 0.001$). There was a non-significant trend for lower recapture of female lice from foreign hosts than from normal hosts ($\chi^2 = 3.30$, d.f. = 1, $p = 0.07$).

Examined at the level of particular species, fewer female *D. distinctus*, than female *D. somadikartai*, were recaptured when transferred from normal host nestlings to mossy-nest nestlings (although the trend was non-significant; Figure 4.7; Fisher exact $p = 0.06$). Significantly fewer female *D. distinctus* were recaptured from the foreign host

Figure 4.5 Transfer of lice from normal (glossy swiftlet) to foreign (mossy-nest and black-nest swiftlet) nestlings. Numbers of transferred lice recaptured are shown in bold. (In this figure percentages and raw numbers are identical because a total of 100 lice were transferred to each host species).

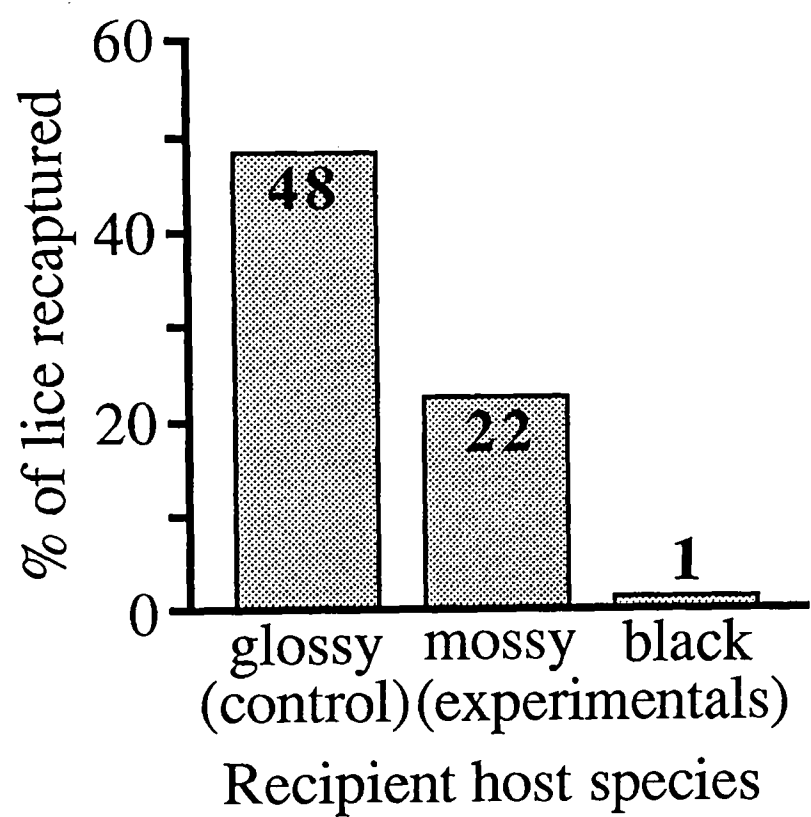


Figure 4.6 Transfer of a) male and b) female lice from normal (glossy swiftlet) to foreign (mossy-nest swiftlet) nestlings. Numbers of transferred lice recaptured are shown in bold.

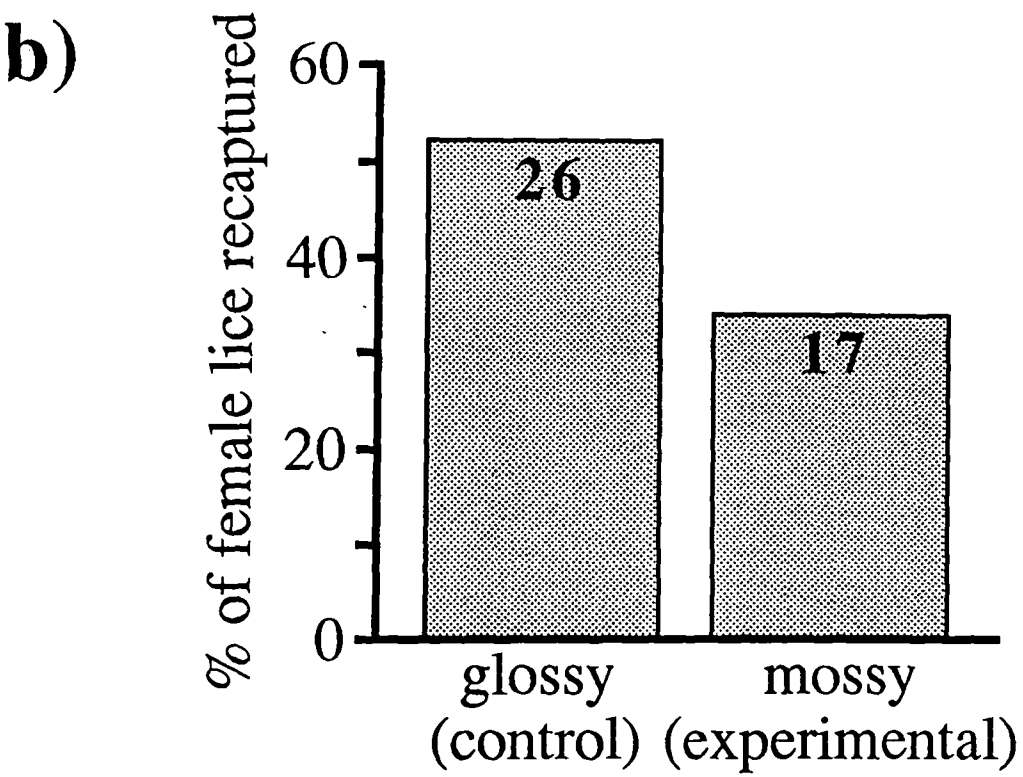
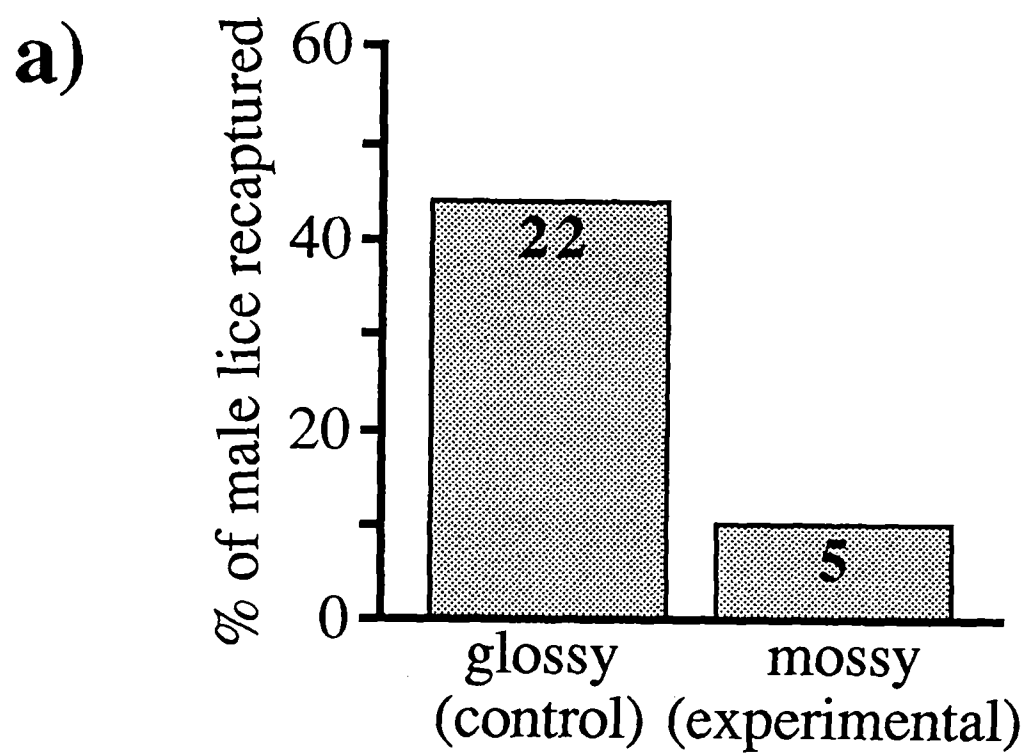
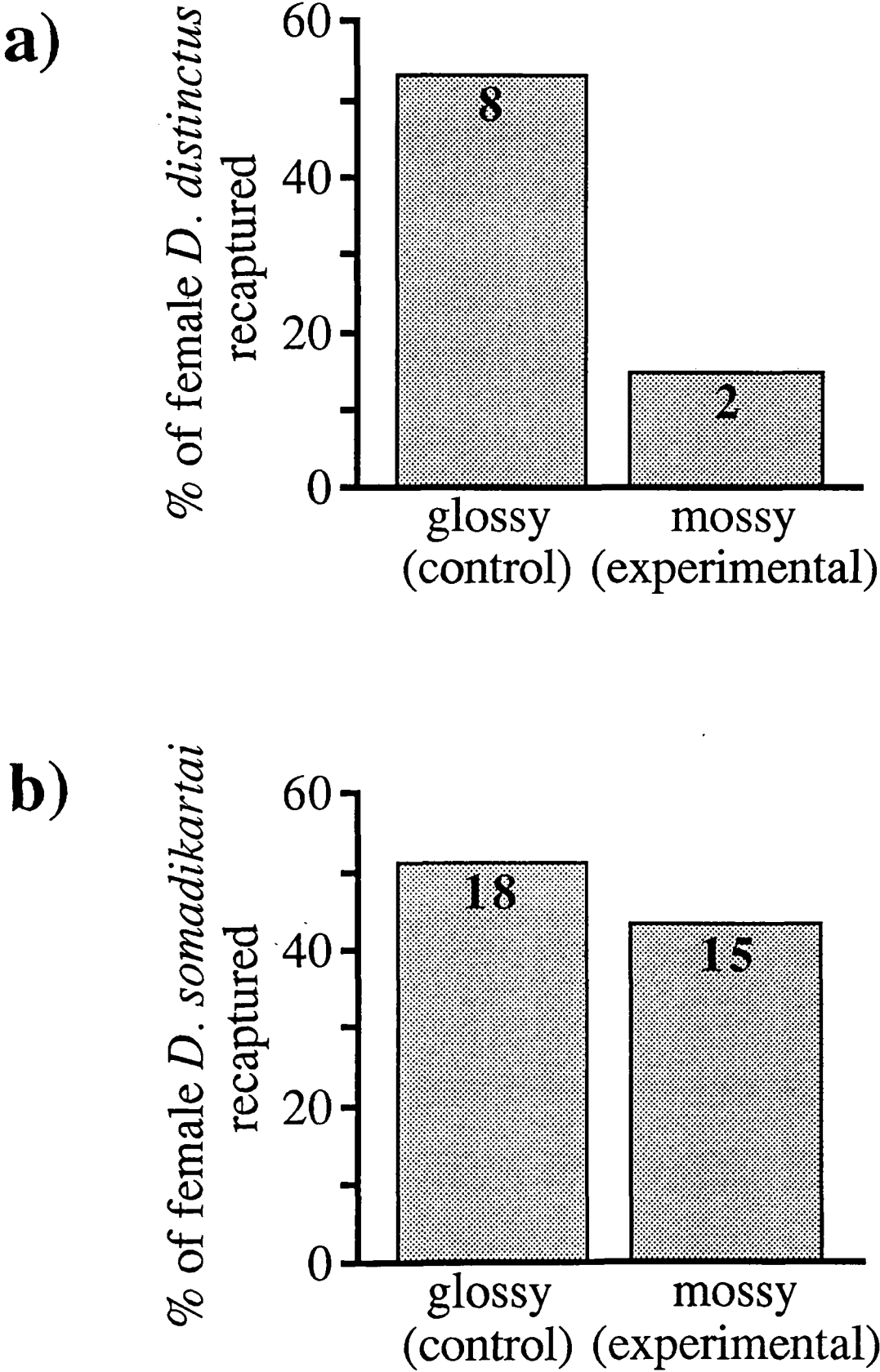


Figure 4.7 Transfer of a) female *Dennyus distinctus* and b) female *Dennyus somadikartai* from normal (glossy swiftlet) to foreign (mossy-nest swiftlet) nestlings. Numbers of transferred lice recaptured are shown in bold.



than from the normal host ($\chi^2 = 5.40$, d.f. = 1, $p = 0.02$), whereas there was no significant difference in the recapture of female *D. somadikartai* ($\chi^2 = 0.52$, d.f. = 1, $p = 0.53$).

To check for back-transmission of lice, parents at 18 of the recipient mossy-nest swiftlet nests were caught and examined for lice at the same time as their nestlings. No glossy swiftlet lice were found on them. To check for horizontal transmission of lice from recipient nestlings to nestlings in adjacent nests, nestlings at 108 mossy-nests and 57 black-nests (surrounding the nests receiving foreign lice) were also examined for lice. No foreign lice were found on any of them.

When measuring the ability of lice to survive off any host, no louse survived for more than 72 hours (Figure 4.8). Only 28% of lice kept off any host were still alive after 36 hours. There was no difference in the survival of male and female lice (Mann-Whitney $U = 59.5$, $p = 0.34$).

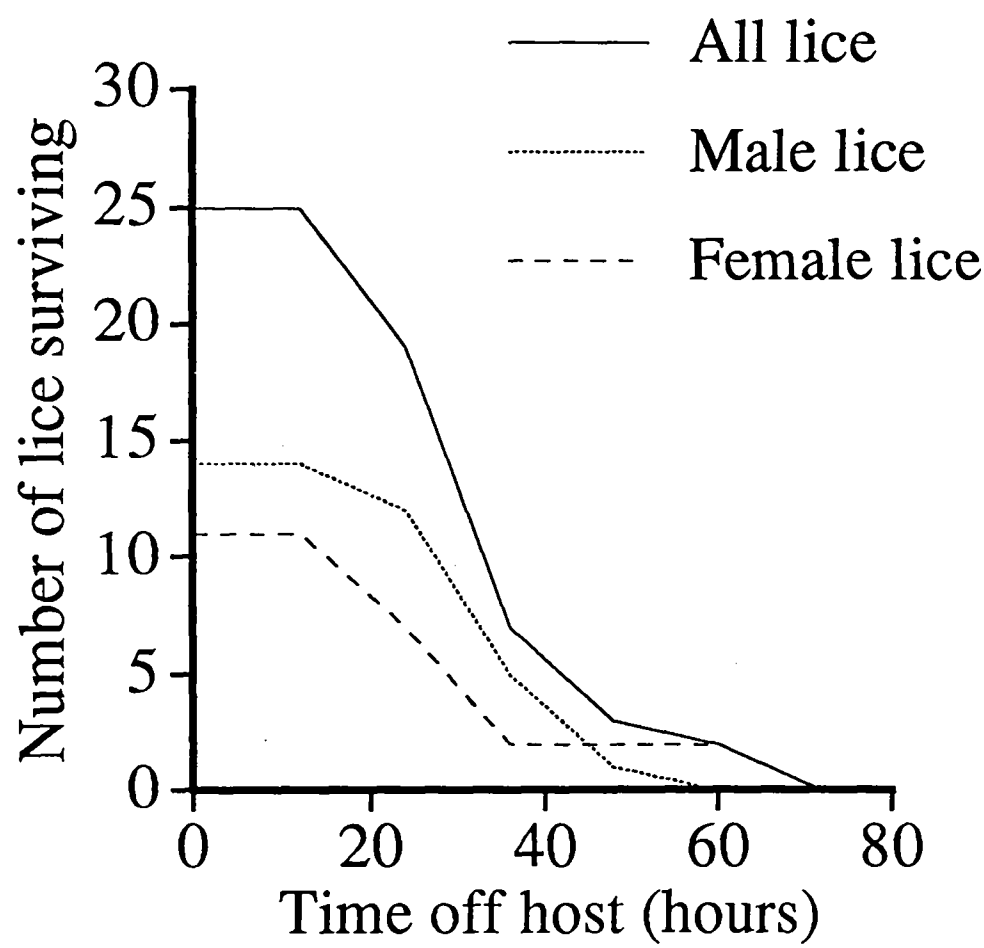
ENDOPARASITE SURVEY. No haematozoa were detected in any of the blood samples examined ($n = 100$). Likewise, no coccidia were detected in the faecal samples examined ($n = 137$).

ENVIRONMENTAL CONTROL. Lice transferred from glossy swiftlets at the house to mossy-nest nestlings, cross-fostered into glossy swiftlet nests, had similar survival as lice transferred to mossy-nest nestlings at the cave (see below).

The background louse load on singleton glossy swiftlet nestlings (the donor host), cross-fostered between glossy swiftlet nests ($N = 15$), was a mean of 0.60 ± 0.74 (0.33 ± 0.47 females and 0.27 ± 0.44 males). Of the females, 0.13 ± 0.34 were *D. distinctus* and 0.20 ± 0.40 were *D. somadikartai*.

Lice transferred from normal host nestlings to mossy-nest nestlings (cross-fostered into the normal host environment) again had significantly lower recapture rates than lice

Figure 4.8 Survival of glossy swiftlet lice kept off any host.



transferred back onto normal host nestlings (Figure 4.9; $\chi^2 = 8.04$, d.f. = 1, $p < 0.005$). As before, there was a significant difference in the recapture of male versus female lice transferred to foreign nestlings; fewer males were recovered (Figure 4.10; $\chi^2 = 12.00$, d.f. = 1, $p < 0.001$). Significantly fewer male lice were recovered from foreign hosts than from normal hosts ($\chi^2 = 12.00$, d.f. = 1, $p < 0.001$), whereas there was no significant difference in the recapture of female lice from normal and foreign host nestlings ($\chi^2 = 0.63$, d.f. = 1, $p = 0.43$).

As before, fewer female *D. distinctus*, than female *D. somadikartai*, were recaptured when transferred from normal host nestlings to mossy-nest nestlings (Figure 4.11; Fisher exact $p = 0.02$). However, there was no significant difference in the recapture of either female *D. distinctus* (Fisher exact $p = 0.15$) or female *D. somadikartai* ($\chi^2 = 0.00$, d.f. = 1, $p = 1.00$) from normal or foreign host nestlings.

Comparing the recapture of lice transferred to mossy-nest swiftlet nestlings in the foreign environment, to that of those transferred to mossy-nest swiftlet nestlings in the normal environment, there was no significant difference in the overall percentage recaptured ($\chi^2 = 0.67$, d.f. = 1, $p = 0.41$), the percentage of males recaptured (Fisher exact $p = 0.09$), the overall percentage of females recaptured ($\chi^2 = 0.00$, d.f. = 1, $p = 1.00$), the percentage of female *D. distinctus* recaptured (Fisher exact $p = 0.61$), nor the percentage of female *D. somadikartai* recaptured ($\chi^2 = 0.34$, d.f. = 1, $p = 0.56$).

LICE TRANSFERRED AMONG NORMAL HOST SPECIES. The background loads of *D. carljonesi* on nestlings of its normal host species were 0.20 ± 0.40 for the black-nest swiftlet (0.13 ± 0.34 females and 0.07 ± 0.25 males), 0.87 ± 0.88 for the white-nest swiftlet (0.40 ± 0.61 females and 0.47 ± 0.62 males), and 0.13 ± 0.34 for the mossy-nest swiftlet (0.07 ± 0.25 females and 0.07 ± 0.25 males).

There was no significant difference in the recapture of *D. carljonesi* transferred from the black-nest swiftlet to either mossy-nest nestlings, white-nest nestlings, or back onto

Figure 4.9 Transfer of lice from normal (glossy swiftlet) to foreign (mossy-nest swiftlet) nestlings, cross-fostered into glossy swiftlet nests. Numbers of transferred lice recaptured are shown in bold.

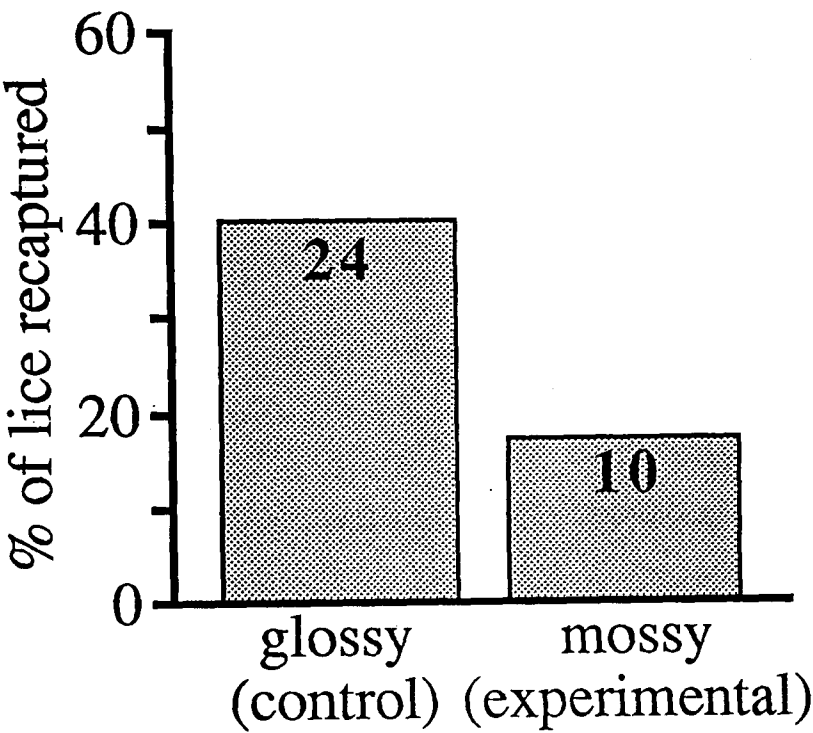


Figure 4.10 Transfer of a) male and b) female lice from normal (glossy swiftlet) to foreign (mossy-nest swiftlets) nestlings, cross-fostered into glossy swiftlet nests. Numbers of transferred lice recaptured are shown in bold.

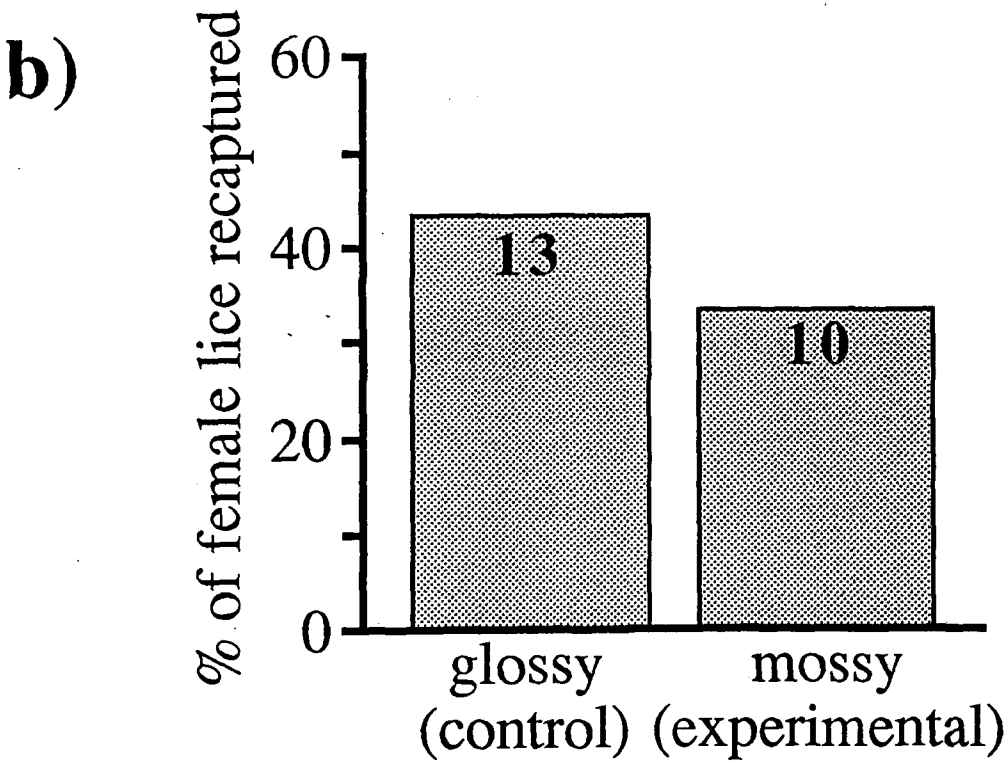
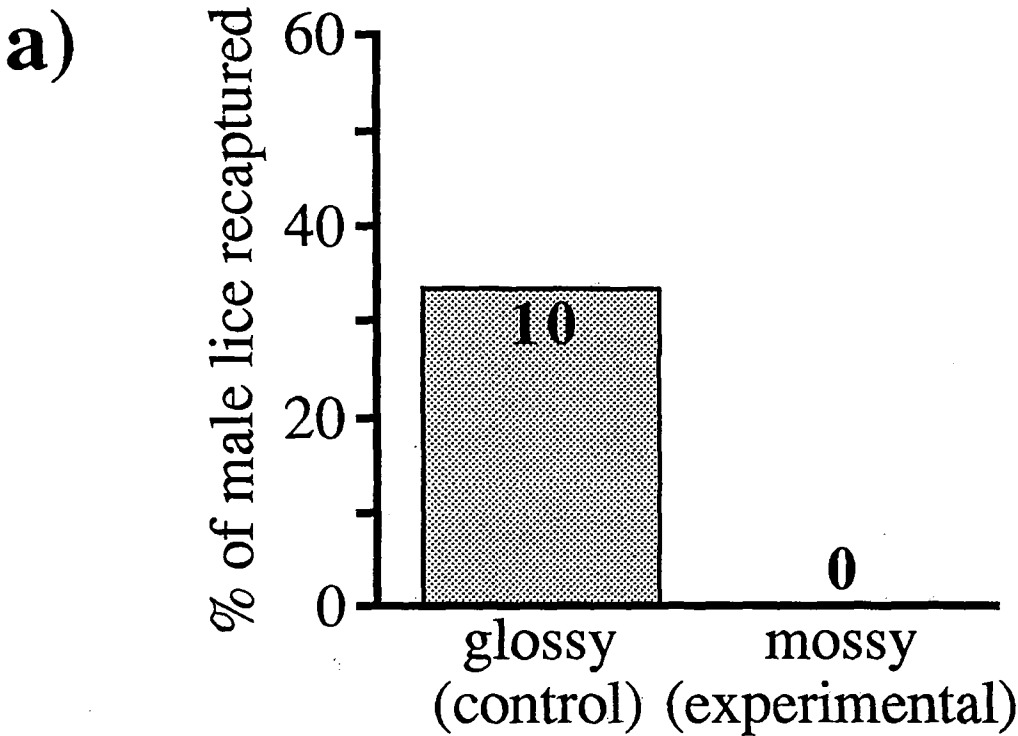
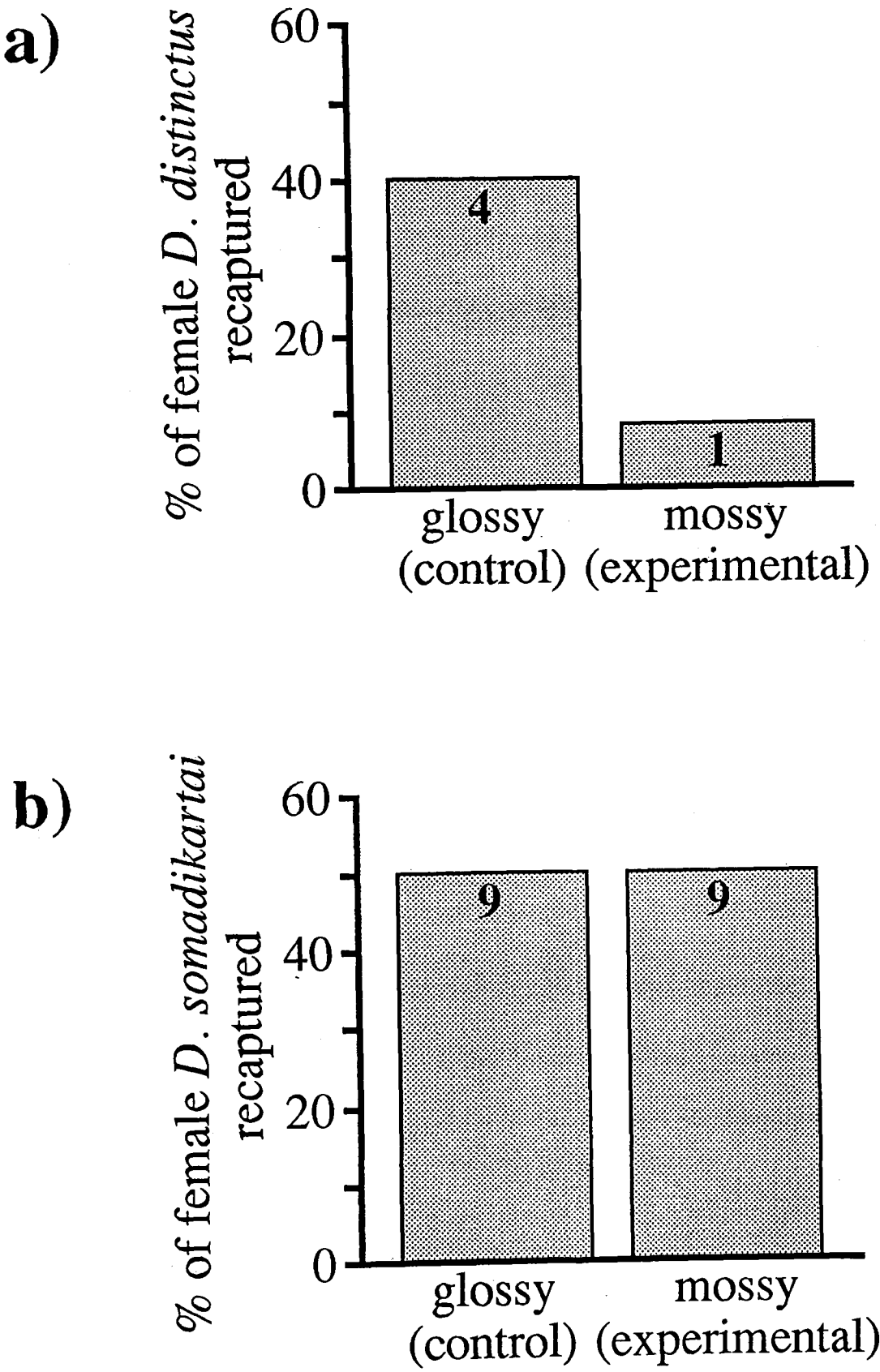


Figure 4.11 Transfer of a) female *Dennyus distinctus* and b) female *Dennyus somadikartai* from normal (glossy swiftlet) to foreign (mossy-nest swiftlet) nestlings, cross-fostered into glossy swiftlet nests. Numbers of transferred lice recaptured are shown in bold.



black-nest nestlings (controls) (Figure 4.12; $\chi^2 = 0.44$, d.f. = 2, $p = 0.80$). However, significantly fewer *D. carljonesi*, transferred from the black-nest swiftlet, were recaptured from glossy nestlings (foreign host species) than from controls (Figure 4.12; $\chi^2 = 21.67$, d.f. = 1, $p < 0.001$). Recapture rates from the glossy swiftlet were near zero.

IMPACT OF NORMAL VERSUS FOREIGN LICE ON NESTLING GROWTH AND SURVIVAL. Through the transfers carried out, the impact of normal versus foreign lice on nestling growth and survival can be compared for both the mossy-nest and black-nest swiftlets. There was no significant difference in survival to fledging of either mossy-nest ($\chi^2 = 0.40$, d.f. = 1, $p = 0.53$) or black-nest (Fisher exact $p = 1.00$) nestlings with normal versus foreign lice transferred to them (Figure 4.13).

There was no significant difference in the growth of mossy-nest nestlings with normal versus foreign lice transferred to them (Figure 4.14). Growth of these two groups was similar before the louse transfers (no significant difference in asymptotic mass ($F = 0.36$, $p = 0.55$), growing period ($F = 0.00$, $p = 1.00$), mean growth rate ($F = 0.08$, $p = 0.78$) or Richards shape parameter ($F = 0.18$, $p = 0.68$)) and remained similar after the louse transfers occurred (no significant difference in fledging mass ($F = 0.52$, $p = 0.48$) or age at fledging ($F = 0.00$, $p = 0.97$)).

There was no significant difference in the growth of black-nest nestlings with normal versus foreign lice transferred to them (Figure 4.14). Growth was again similar both before the louse transfers (no significant difference in asymptotic mass ($F = 0.62$, $p = 0.44$), growing period ($F = 0.17$, $p = 0.68$), mean growth rate ($F = 0.00$, $p = 0.95$) or Richards shape parameter ($F = 0.09$, $p = 0.76$)) and after the louse transfers occurred (no significant difference in fledging mass ($F = 0.16$, $p = 0.69$) or age at fledging ($F = 0.40$, $p = 0.53$)).

Figure 4.12 Transfer of *Dennyus carljonesi* from one normal host species (black-nest swiftlet) to nestlings of two other normal hosts (mossy-nest and white-nest swiftlets), and to nestlings of a foreign host species (glossy swiftlet). Numbers of transferred lice recaptured are shown in bold.

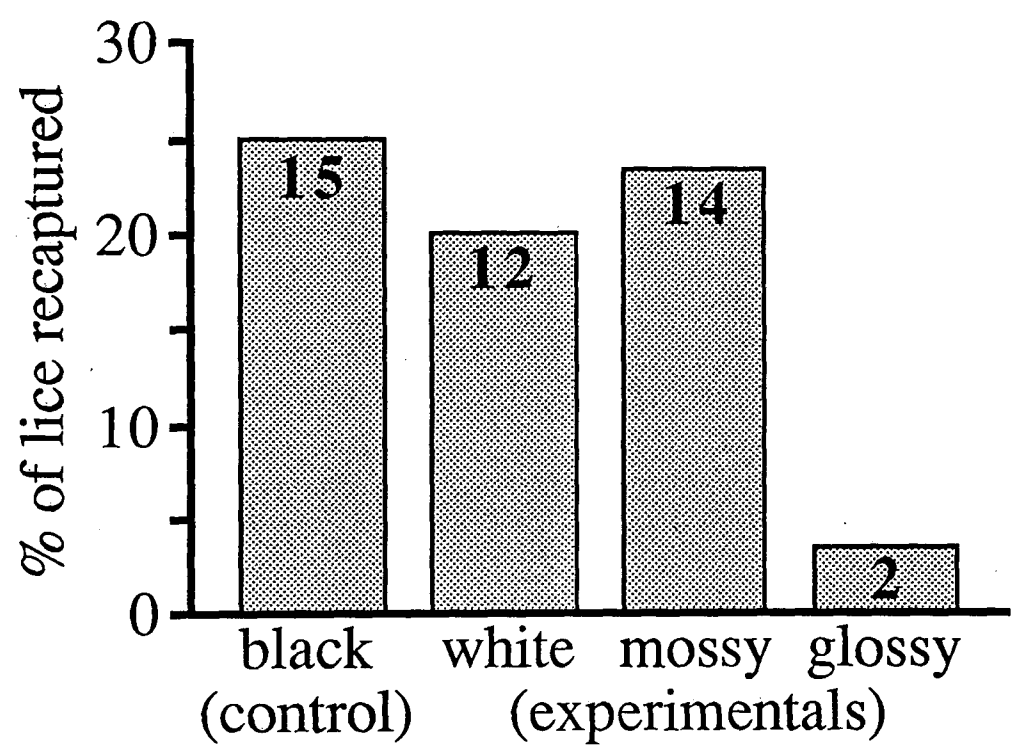


Figure 4.13 Survival to fledging of a) mossy-nest, and b) black-nest nestlings with normal versus foreign lice transferred to them.

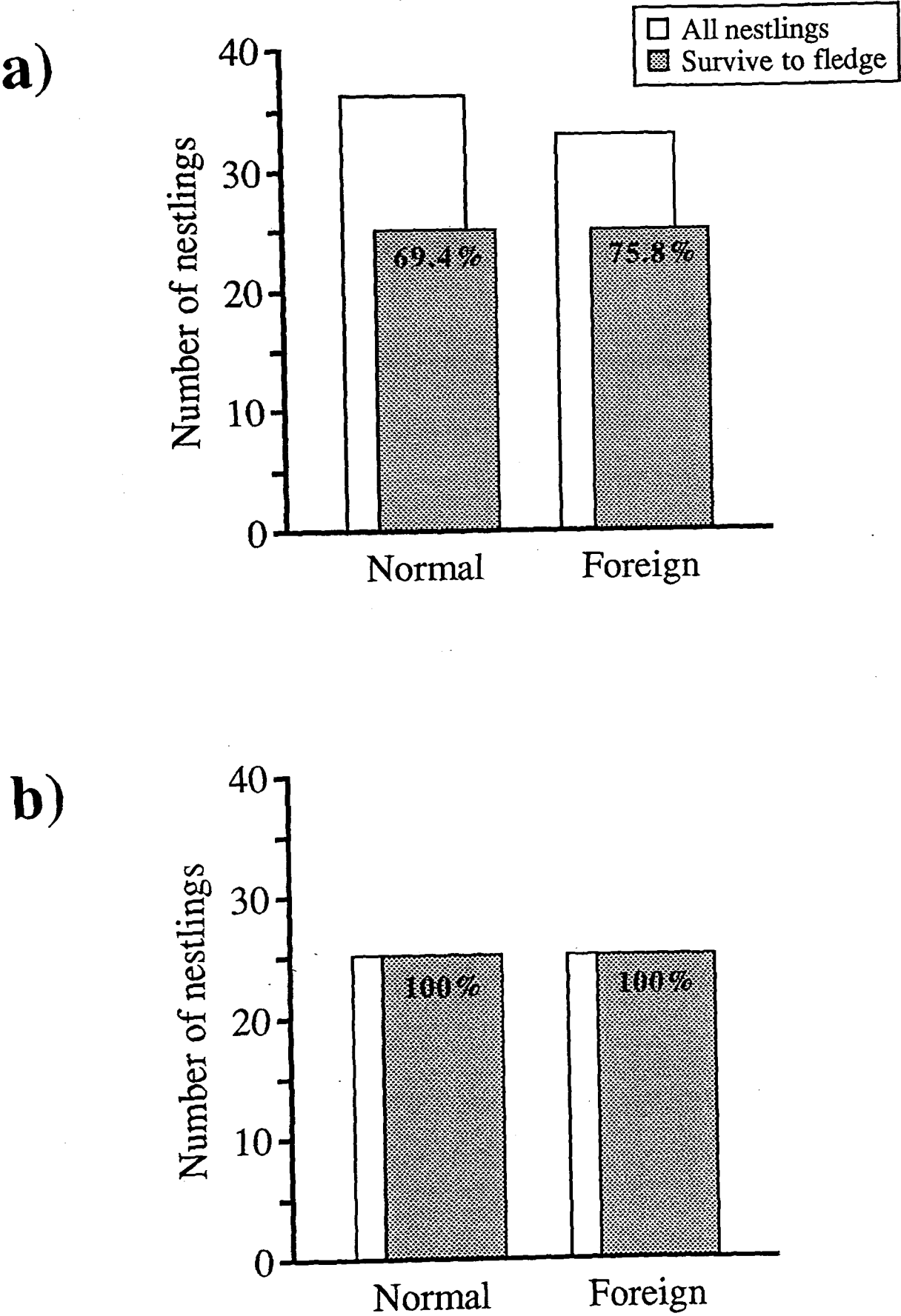
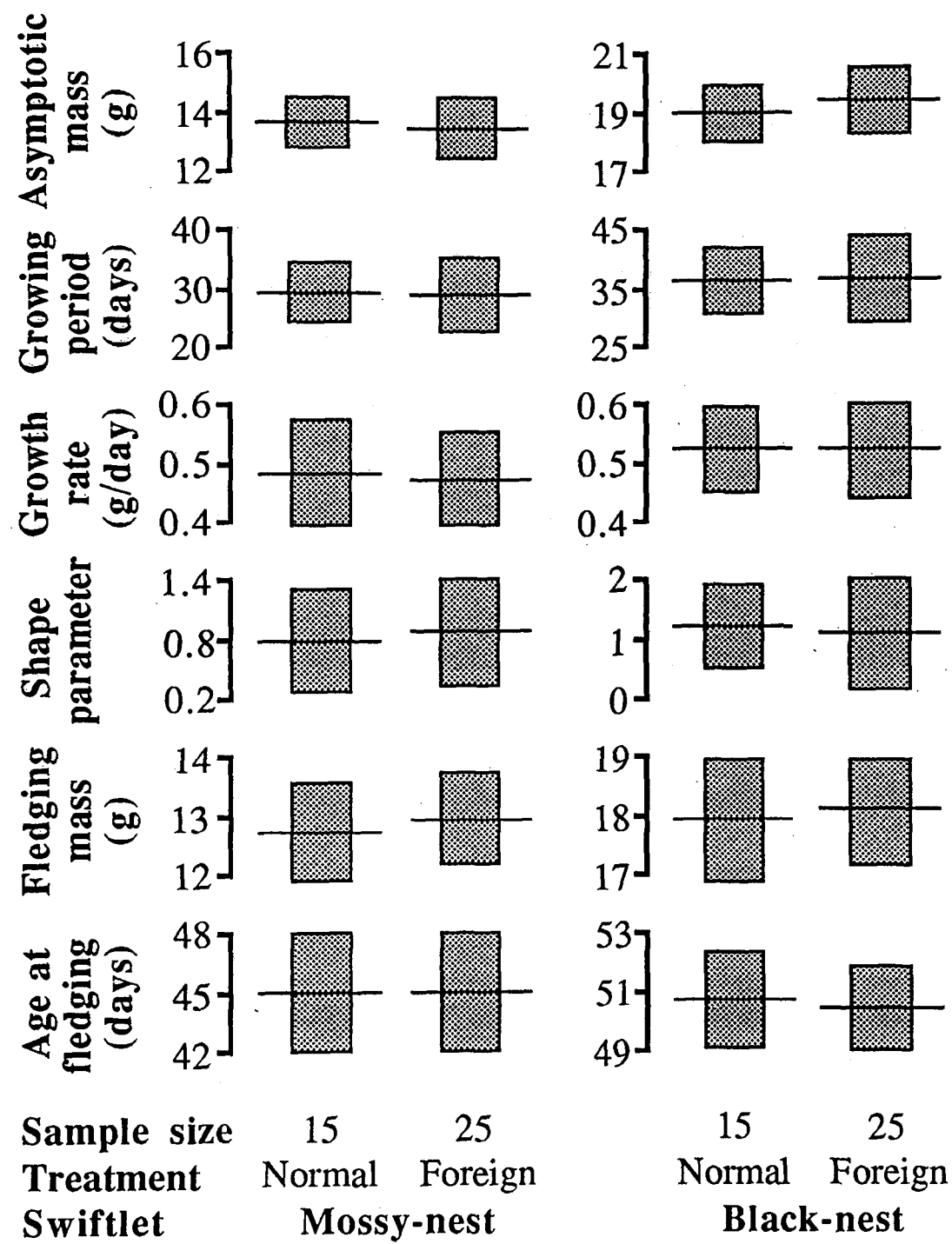


Figure 4.14 Growth components for nestlings with normal versus foreign lice transferred to them, for mossy-nest and black-nest swiftlets. Values are means ($\pm$ 1s.d.) as fitted to the data by "General Linear Model" analyses.



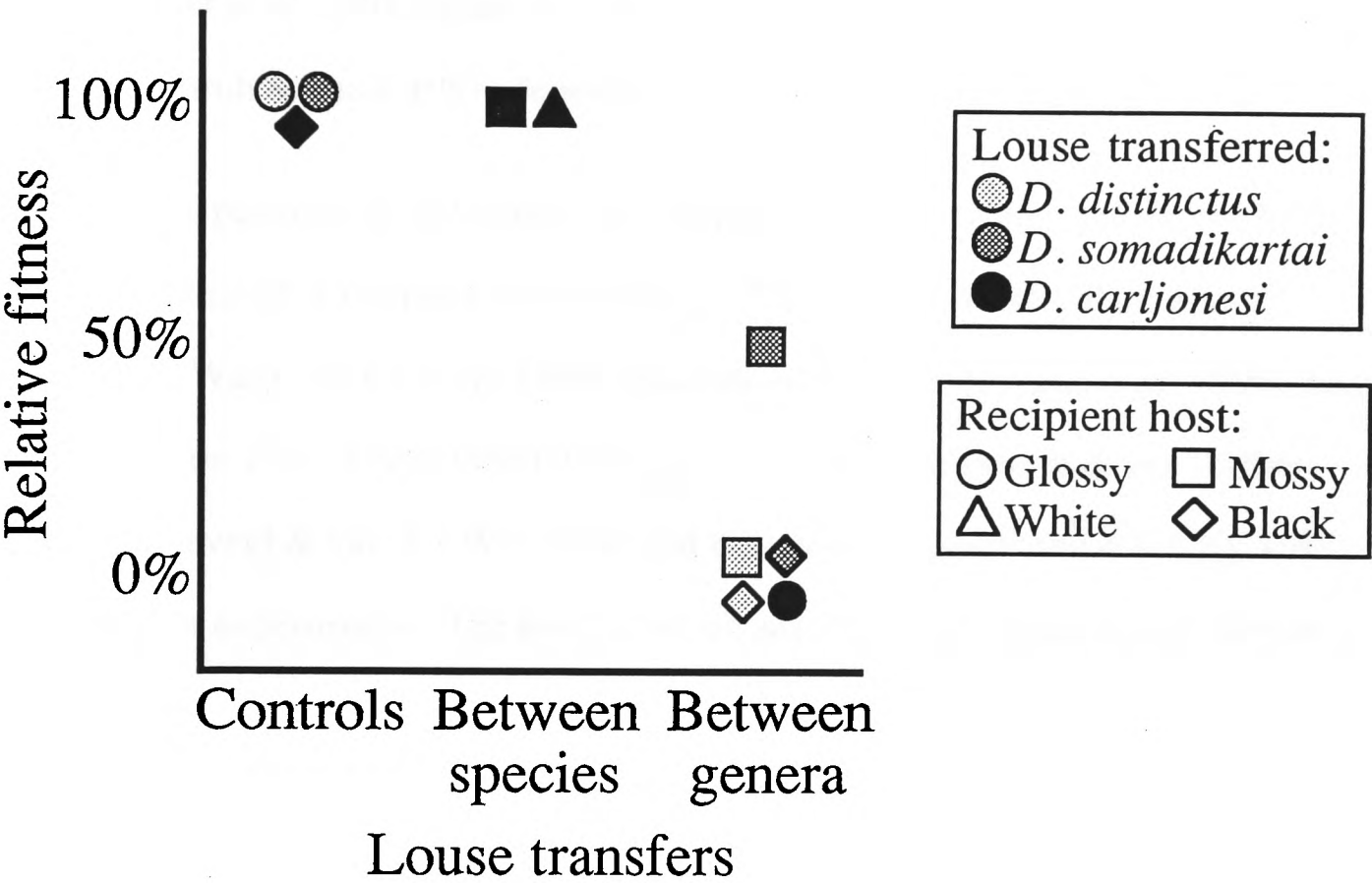
LOUSE SURVIVAL RELATIVE TO HOST TAXONOMY. There was a negative correlation between louse survival and host taxonomy (Figure 4.15). The survival of lice transferred between host species was similar to controls, but was significantly reduced (to near zero in most cases) when lice were transferred between host genera.

4.5 Discussion

Opportunity for louse dispersal (or lack thereof) does not govern the host specificity of chewing lice on Malaysian swiftlets (Figures 4.1 & 4.2). One individual each of *D. distinctus* and *D. somadikartai* were found on the "wrong" host (the black-nest swiftlet; Table 4.1), implying that passive or active dispersal of these lice to individuals of a foreign host species occurs. Glossy and black-nest swiftlets (between which the straggling of lice occurred) are often found nesting in close proximity at my field site (Gomantong Caves). The straggling of lice between host species living in close ecological proximity is a common phenomena (Eveleigh & Threlfall 1976; Horning *et al.* 1980; Furness & Palma 1992). No straggling of lice from glossy swiftlets to the other two species was detected in this study. This is not surprising since glossy, mossy-nest and white-nest swiftlets rarely nest in close proximity (personal observation). The dispersal of lice to glossy swiftlets from other host species (especially the black-nest swiftlet nesting in close proximity) might have been observed if a large sample of lice had been collected from glossy swiftlets at Gomantong Caves. However, 97% of the lice collected from glossy swiftlets in this study were taken at the house location where no other swiftlet species were present (Table 4.1).

There was no evidence for adaptive differences between demes of *D. carljonesi* on its three normal host species (the mossy-nest, white-nest and black-nest swiftlets). Similar numbers of lice were recaptured after transfer from black-nest swiftlet adults to nestlings of all three hosts (Figure 4.12). This implies that lice were not adapting to

Figure 4.15 Relation between louse survival and host taxonomy, i.e. whether lice are transferred between host individuals (control transfers), host species or host genera.



local host characters. The formation of adaptive demes is only possible if gene flow between populations is somewhat restricted (Edmunds & Alstad 1978; Karban 1989; Alstad & Corbin 1990; Hanks & Denno 1994; Mopper *et al.* 1995). For example, deme formation in herbivorous insects can be at the level of the plant clone (Karbon 1989), individual plant (Edmunds & Alstad 1978; Rice 1983; Wainhouse & Howell 1983; Mopper *et al.* 1995), plant locality (Cobb & Whitham 1993; Hanks & Denno 1994; Mopper *et al.* 1995), plant species (Futyma *et al.* 1994; Mopper *et al.* 1995) or not at all (Unruh & Luck 1987) depending on patterns of dispersal.

For parasites, parasitoids and pathogens, adaptive differences between demes have been demonstrated at the level of host locality (Files & Cram 1949; Parker 1985; Ballabeni & Ward 1993; Lively 1989; Manning *et al.* 1995; Morand *et al.* 1996), host strain (Dawson 1967; Ebert 1994), host species group (Kraaijeveld & van Alphen 1993; Kraaijeveld & van der Wel 1994) and host genera (Leuchtman & Clay 1989) using transfer experiments. The level at which adaptive deme formation by lice on swiftlet populations would occur would thus be determined by the level at which louse dispersal is restricted. The observed straggling of host-specific lice onto foreign host species (above) makes it very likely that dispersal of *D. carljonesi* between its three normal host species also occurs (particularly between closely nesting black-nest and white-nest swiftlets). Thus, adaptive differences between demes of *D. carljonesi* may be prevented by too much gene flow between louse populations.

Adaptive differences between demes of *D. carljonesi* may also be lacking because there are no selective differences on the different hosts. This is unlikely in light of the results of the experimental transfers of foreign lice to mossy-nest and black-nest swiftlets (both normal hosts of *D. carljonesi*). Louse survival was not equivalent on the two hosts (see below) implying that there are differences in the environment presented by the different hosts. Thus, the selective pressure for adaptive deme formation by *D. carljonesi* on different host species may well be present. However, as no deme

formation is detected here, the selective pressures cannot be great enough to overcome the stabilising effect of gene flow between louse populations (Alstad & Edmunds 1983). In surviving concurrently on three host species, which differ in the characters important to louse survival, *D. carljonesi* is exhibiting a degree of tolerance to environmental variation.

Although there is no evidence for adaptive deme formation at the louse population level, there is evidence for adaptations to normal host characters by louse species. When the host specific *D. distinctus* and *D. somadikartai* were transferred from glossy swiftlets (normal host species) to mossy-nest and black-nest swiftlets (foreign host species) subsequent louse recapture was reduced relative to controls (Figure 4.5). The null hypothesis that host-specific parasites have equal survival on normal and foreign hosts is thus rejected. This result is in agreement with most previous attempts by researchers to rear lice on unnatural hosts (Hopkins 1942, 1949; Ash 1960; Kettle 1977), where viable populations could not be maintained. The only lice in my study which did not have reduced recapture rates on a foreign host were female *D. somadikartai* transferred to mossy-nest swiftlets (Figures 4.6 & 4.7). However, *D. somadikartai* would have been unable to maintain a viable population on the mossy-nest swiftlet due to the reduction in survival of males (Figure 4.6). Thus, adaptation of lice to specific host characters presumably governs the host-specificity of chewing lice on Malaysian swiftlets.

Reduced recapture of transferred lice from foreign nestlings may have been due to either reduced louse survival, which was the variable under investigation, or dispersal of lice away from foreign nestlings. The vertical back-transmission of lice from nestlings to adults can be ruled out as no foreign lice were ever observed on the adults of mossy-nest nestlings which received experimental transfers of foreign lice. This is not surprising since documented patterns of louse transmission at the nest are from adults to nestling birds, and not vica-versa (Clayton & Tompkins 1994; Lee & Clayton 1995; Chapter 2). The horizontal transmission of lice to birds at surrounding nests can

also be ruled out as no foreign lice were ever observed on nestlings at nests adjacent to experimental nests (receiving transfers of foreign lice). This is not surprising since chewing lice are highly dependent on the host integument (Hopkins 1949; Kettle 1977), restricting dispersal off the host. Thus, the small numbers of transferred lice that were recaptured from foreign hosts was almost certainly a result of louse mortality.

The ten day window, for which transferred lice were left on hosts before recapture, was sufficient to detect whether the recipient host was a suitable environment for louse survival. When kept off any host, glossy swiftlet lice could not survive for more than 72 hours (Figure 4.8). Thus, for transferred lice to be recaptured, hosts must have exhibited specific characters which permitted louse survival. Note, however, that I have not collected any data pertaining to the ability of lice to reproduce on foreign versus normal hosts.

Reduced survival of lice transferred from glossy swiftlets at the house to foreign hosts at the cave may have been due either to a lack of adaptedness to the new host (which again was the variable under investigation) or lack of adaptedness to the new host environment. Inside the caves the temperature was lower (mean midday temperature of 25.2°C versus 31.8°C) and the humidity was higher (mean midday relative humidity of 85.1% versus 65.4%) than at the house. However, any influence of changing host environment on louse survival can be ruled out in light of experimental transfers conducted. The same pattern of louse survival was evident when lice were transferred from glossy swiftlets to mossy-nest swiftlets at either the cave or the house (Figures 4.9, 4.10 & 4.11). This is not surprising since the host integument provides a relatively stable environment for ectoparasites, regardless of external conditions (Kettle 1977; Marshall 1981a).

Rozsa (1993) suggested that the formation of viable populations on foreign hosts by dispersing lice (stragglers) is likely to be prevented by competition with the foreign hosts normal lice. Therefore, in this study, the survival of lice experimentally

transferred to new hosts may have been reduced through competition with local louse populations. However, all recipient nestlings in the transfer experiments had very low background louse loads (range of 0-4 lice). Furthermore, in perhaps the only thorough study to date, Choe & Kim (1988) found no evidence for competition between lice coexisting on several species of seabirds. Thus, local lice most likely had minimal impact on transferred louse survival over the ten day window.

There was a negative relationship between transferred louse fitness and host taxonomy (Figure 4.15). Louse survival was equivalent to controls when transferred between host species within the same genus (*Aerodramus*; see Figure 4.3), but was significantly reduced when transferred between host genera. This result is in agreement with a recent study of trichodectid lice on pocket-gophers (Reed 1994). Laboratory transfers determined that lice were able to establish colonies when transferred between pocket-gopher sub-species and species (respective transfer success rates of 100% and 75%), but this ability was severely reduced when lice were transferred between host genera (7% success rate). By design, this experiment did not allow for competition of transferred lice with the recipient hosts normal lice. Reed concluded that the existence of distinct louse species on sub-species and species of their pocket gopher hosts was due to the inability of lice to disperse between host populations, and that the existence of distinct louse species on different host genera was due, not just to restricted dispersal, but also to adaptation of lice to genera-specific host characters (specialisation).

The survival of lice transferred from glossy to mossy-nest swiftlets (from *Collocalia* to *Aerodramus*) was not equivalent to that of those transferred from glossy to black-nest swiftlets (also from *Collocalia* to *Aerodramus*), or from black-nest to glossy swiftlets (from *Aerodramus* to *Collocalia*), even though all transfers were between the same two host genera. The survival of female *D. somadikartai* (transferred from the glossy swiftlet) on the mossy-nest swiftlet was similar to controls (Figure 4.7), whereas louse survival in all other host taxonomically equivalent transfers was near zero (Figures 4.5

& 4.12). Female *D. somadikartai* thus exhibited greater environmental tolerance than other lice, allowing it to survive on a host in a different genus to its normal host. However, as mentioned earlier, *D. somadikartai* would presumably be unable to maintain a viable population on the mossy-nest swiftlet due to the reduction in survival of males.

The results of this study, and that of Reed (1994), can be combined to model the respective roles of lack of dispersal opportunity and adaptation to specific host characters in the evolution and maintenance of chewing louse host-specificity (Figure 4.2): lice have sufficient tolerance to environmental variation to enable the maintenance of viable populations on closely related hosts (hosts within one genera for both this and Reed's (1994) study). However, if louse dispersal is restricted, populations diverge through adaptively neutral genetic change. This is seen in lice on different sub-species and species of pocket gophers in Reed's (1994) study. Here, lice may also be slightly adapted to specific host characters (transfer success rate of only 75% between pocket-gopher species, compared to 100% between pocket-gopher sub-species), but not to the extent that it prevents survival on hosts within the same genus. If louse dispersal is not restricted, gene flow prevents louse populations from diverging. This explains the presence of *Dennyus carljonesi* on all three species of *Aerodramus* swiftlets at Gomantong caves. Gene flow between louse populations also prevents the formation of adaptive demes to specific characters of hosts within the same genus.

Louse populations on less related hosts may be formed and maintained as distinct species by local adaptation to host characters, regardless of the presence (this study) or absence (Reed 1994) of louse dispersal. Louse species are not sufficiently tolerant to allow concurrent survival on hosts with differences in characters as extreme as is normally seen between host genera (both this study, and Reed (1994)). (This theory assumes that host taxonomy reflects host phylogeny, and that closely related hosts are more similar, than more distantly related hosts, in the characters to which lice adapt)

Whilst there was evidence for louse adaptation to specific host characters, there was no evidence for reciprocal adaptation of swiftlet hosts to resident parasite populations. There was no difference in the growth (Figure 4.14) or survival to fledging (Figure 4.13) of either mossy-nest or black-nest nestlings with normal versus foreign lice transferred to them. This may be for two reasons. First, birds have only generalised defence mechanisms against louse infestation (mainly preening; Clayton 1991). These defences are unlikely to adapt to a specific louse species. (Lice have specialised adaptations to their avian hosts as the host, to a louse, is the entire environment upon which its fitness depends (Hopkins 1949; Kettle 1977), whereas chewing lice are just one component of a bird's environment). The lack of specialisation versus louse infestation implies that any host defences will be just as effective against foreign lice as they are against normal lice. This, in turn, implies that both normal and foreign lice will have an equivalent impact on host fitness. The situation would be different if lice were vectoring host-specific pathogens. This is unlikely in this study as no endoparasites were detected in the blood or faecal samples of any nestling which had lice transferred to it.

There may also be no difference in impact of foreign versus normal lice since chewing lice may be benign parasites with no impact whatsoever on nestling growth. Even though effects of avian lice on the long-term survival of their adult hosts have been documented (Booth *et al.* 1993; Brown *et al.* 1995), chewing lice have been reported to have no impact on nestlings (Clayton & Tompkins 1995; Lee & Clayton 1995). The impact of chewing lice on the growth of nestling common swifts (*Apus apus*) is investigated further in Chapter 6.

Chapter 5 Ecological determinants of chewing louse survival on Malaysian swiftlets

5.1 Introduction

In Chapter 4, I showed that *Dennyus* lice on Malaysian swiftlets are able to survive only on closely related host species. This may have been due to closely related host species being more similar in the characters which lice are adapted to (and thus depend on for survival). The aim of this chapter is to identify the host characters which may be the proximate determinants of louse survival on Malaysian swiftlets.

The morphology of lice appears to be adapted to the physical environment presented by the avian host. There is, in particular, a general correlation between louse size and shape and feather form (Hopkins 1942; Clay 1949). This is related to the selective pressure of host preening (Nelson & Murray 1971; Stock & Hunt 1989; Clayton 1991). On regions of the host where the efficacy of host preening is relatively low (i.e. the head and neck regions) lice tend to be relatively sedentary with short bodies, broad heads, short legs and well developed claws. On regions where host preening is more efficient (i.e. wings and back) lice tend to be more mobile with flattened, elongate bodies, narrow heads, longer legs and weaker claws. The tendency for lice on birds to have greater variation in body shape than lice on mammals is often attributed to the greater structural variation of feathers, compared to that of hair (Marshall 1981a).

Adaptation of lice to detailed host structure has been invoked to explain the positive correlation between parasite and host body size that is often observed in host/lice associations (Ward 1957; Clay 1962; Marshall 1981a; Kirk 1991). This relationship was first formalised, for lice on birds, as "Harrison's Rule" (Harrison 1915). A positive correlation between parasite and host body size is observed for chewing lice

on pocket gophers, even when phylogenetic relatedness is controlled for (Harvey & Keymer 1991).

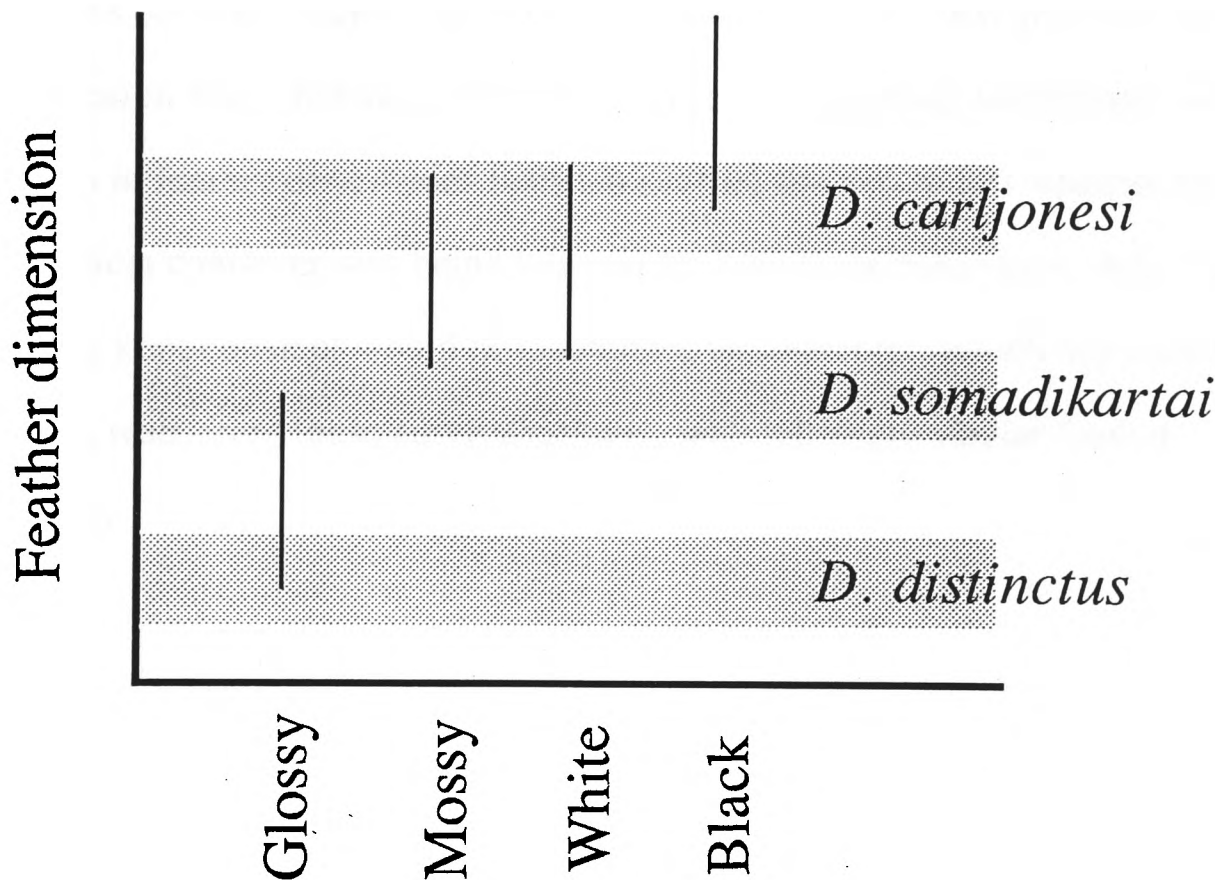
When experimentally transferred among host species, adult chewing lice demonstrated the ability to survive on swiftlets with different body sizes (Figure 5.1). Female *Dennyus somadikartai* had equivalent survival when transferred onto nestlings of a foreign host species (mossy-nest swiftlet) as they did when (control) transferred to nestlings of their normal host species (glossy swiftlet), even though the mossy-nest swiftlet has a wing-chord 20% greater than that of the glossy swiftlet. Both male and female *Dennyus carljonesi* had equivalent survival when transferred from adult black-nest swiftlets onto nestlings of their three normal host species (mossy-nest, white-nest, and back onto black-nest swiftlets), even though the largest of these species (black-nest swiftlet) has a wing chord 10% greater than that of the other two.

Feather dimensions are a likely candidate for what constrains the ability of lice to survive on different sized hosts (Clay 1957). There is a high level of variation in detailed feather structure both within and between individual feathers and feather tracts of a single bird species (see below). Lice may be able to survive on hosts where the variation in feather morphology overlaps (Figure 5.2). This would explain the pattern of survival of *Dennyus* spp. on Malaysian swiftlets, where more closely related hosts are more similar in body size (Figure 5.1) and are therefore, presumably, more similar in detailed feather structure. The overlap between host feather dimensions and louse requirements, theoretically depicted in Figure 5.2, correlates with the results from Chapter 4. *Dennyus distinctus* cannot survive when transferred from the glossy swiftlet (its normal host) to any other host. *D. somadikartai*, normally found on the glossy swiftlet, shows partial survival when transferred to the mossy-nest swiftlet and no survival when transferred to the black-nest swiftlet. *D. carljonesi* is normally found on the mossy-nest, white-nest and black-nest swiftlets (where it has equivalent survival on each) but cannot survive when transferred to the

Figure 5.1 The survival of *Dennyus* lice, transferred between swiftlet host species in Chapter 4, in relation to host wing-chord. Percentages are approximate survival, relative to controls. Numbers in parentheses are mean wing-chords (from Chantler & Driessens 1995).

DONOR HOST	RECIPIENT HOST (wing chord)			
	<i>Collocalia esculenta</i> (98.5mm)	<i>Aerodramus salanganus</i> (118mm)	<i>Aerodramus fuciphagus</i> (118.5mm)	<i>Aerodramus maximus</i> (131.5mm)
<i>Collocalia esculenta</i>	100%	25%	----	0%
<i>Aerodramus maximus</i>	0%	100%	100%	100%

Figure 5.2 Hypothetical relationship between swiftlet feather dimensions (for the glossy, mossy-nest, white-nest and black-nest swiftlet) and the range of those dimensions over which different species of *Dennyus* lice can survive. Vertical bars represent variation in feather dimensions for each swiftlet species (spaced proportionally to the mean wing-chord of each species). Shaded regions represent ranges over which louse species can survive (spaced proportionally to the mean body length of adult female lice of each species, from Clayton *et al.* in review).



glossy swiftlet. If *Dennyus* lice are utilising variation in feather dimensions to allow survival on swiftlet species of different sizes, and if feather dimensions vary among feather tracts of an individual bird, one would expect to see the same louse species occupying different microhabitats on different host species.

The first goal of this chapter was to test the null hypothesis that the dimensions of detailed feather structure are not significantly correlated with swiftlet body size. The null hypothesis that feather dimensions do not vary significantly among feather tracts of swiftlet species was also tested. The second goal was to test the null hypothesis that the same species of louse does not occupy significantly different microhabitat distributions on host species with different body sizes. The final goal was to combine the information about behavioural shifts in louse microhabitat distribution, and differences in feather dimensions between swiftlet species, to test whether any particular host character was being kept constant by louse behaviour. Any feather dimensions kept constant would be candidates for characters which lice may be adapted to, restricting louse survival to hosts with relatively similar feather morphology.

5.2 Materials and methods

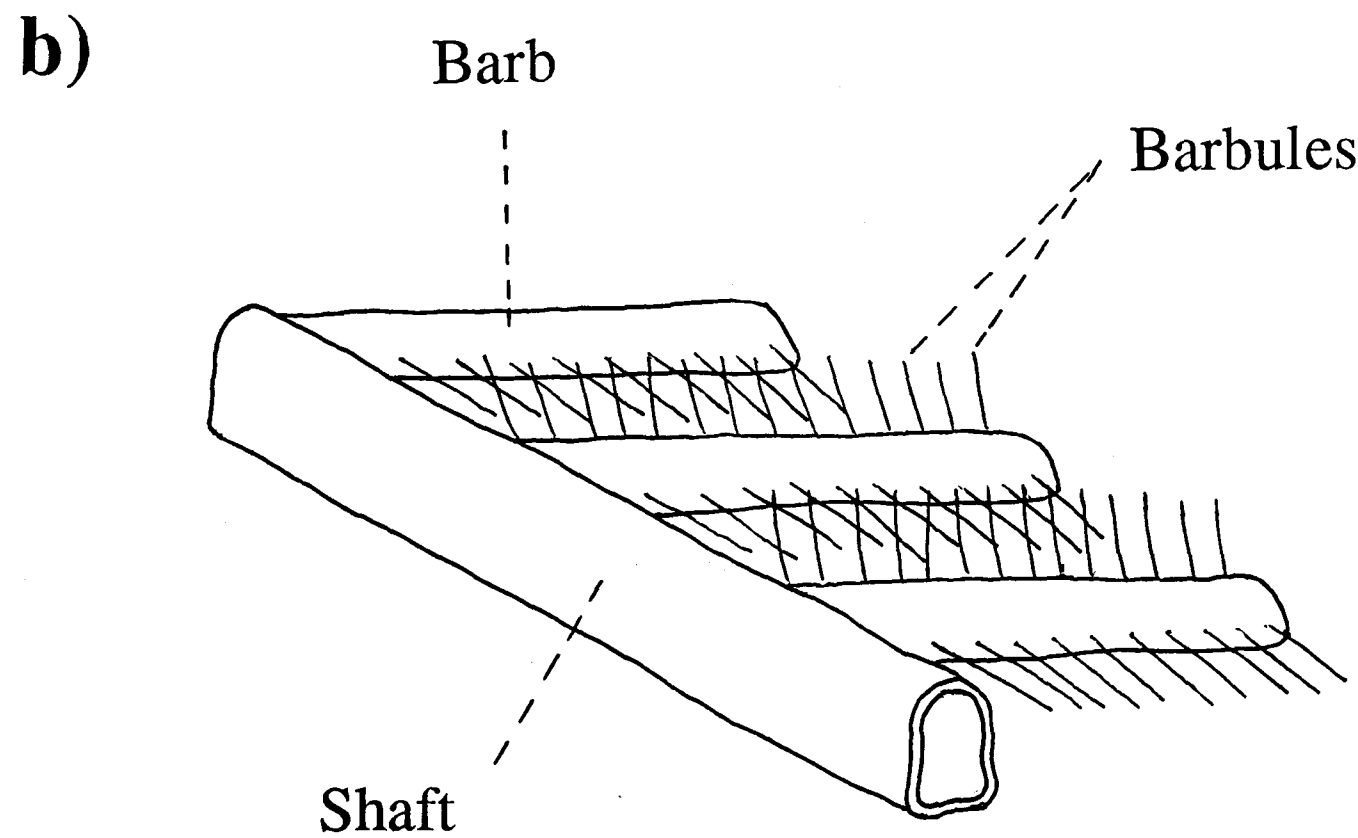
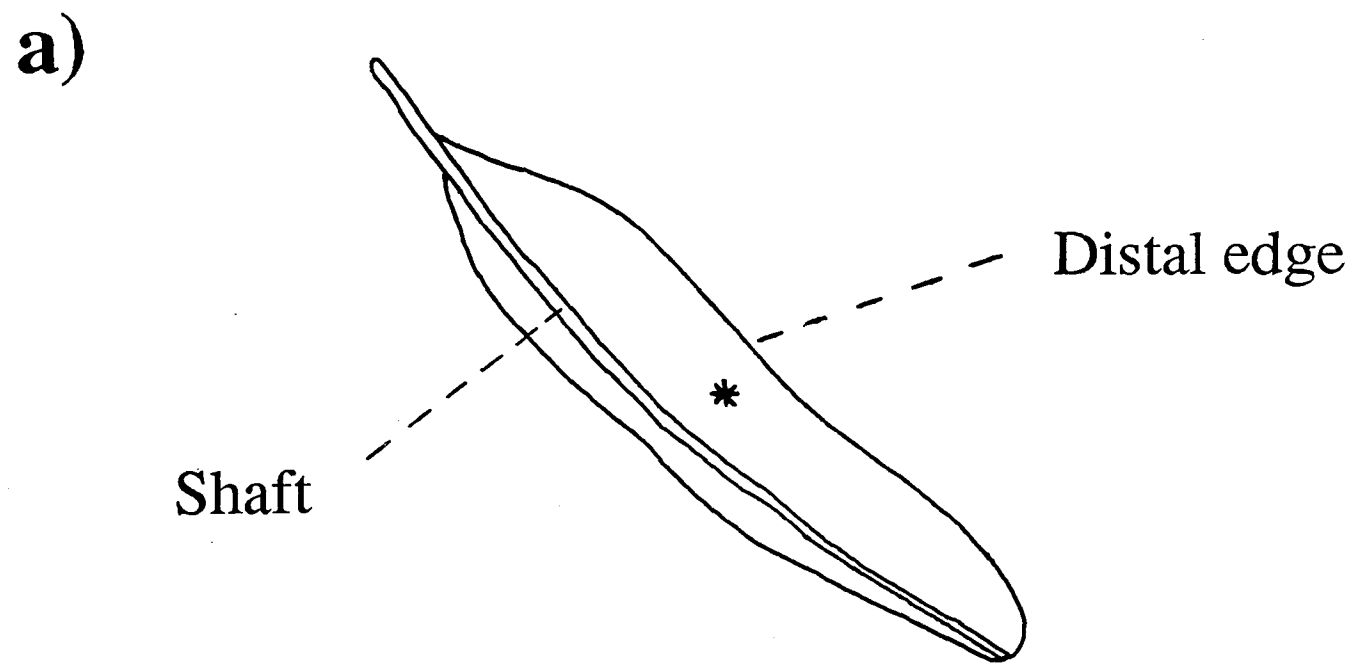
HOST FEATHER DIMENSIONS. Feathers were collected from adults of each swiftlet species involved in the transfer experiments (Chapter 4): the glossy, mossy-nest, white-nest and black-nest swiftlets. The following were taken from each of five individuals of all four species: primary feathers 2, 4, 6 and 8, secondary feathers 2, 3, 4 and 5, and tail feathers 1, 2, 3, and 4 (see Figure 2.2). In Chapter 2, 98% of adult lice on the glossy swiftlet occurred on flight feathers (primary, secondary and tail feathers). The same relative point on the underside of each feather (mid-way along the length, and mid-way between the feather shaft and distal edge (i.e. middle of the

trailing vane for primary feathers)) was examined under x10 and x40 magnification of a phase-contrast microscope (Figure 5.3a). The following dimensions were measured: barb diameter, distance between barbs, barbule diameter, and distance between barbules (Figure 5.3b). Each dimension recorded, for each feather, was the mean of five measurements. Feather dimensions were compared using the mean dimensions for each feather tract on each swiftlet individual. The wing-chord of each bird from which feathers were taken was also measured, using a 15cm chord stick.

LOUSE MICROHABITAT DISTRIBUTIONS. When transferred lice were recaptured in the experiments described in Chapter 4, their location on the host was recorded (i.e. primary, secondary, tail or body feathers; see Figure 2.2). The survival of female *Dennyus somadikartai* on the mossy-nest swiftlet, after being transferred from the glossy swiftlet, was equivalent to controls. Thus, the microhabitat distributions of female *D. somadikartai* on the glossy and mossy-nest swiftlet could be compared. The survival of *Dennyus carljonesi* on both the white-nest and mossy-nest swiftlet, after being transferred from the black-nest swiftlet, was equivalent to controls. Thus, the microhabitat distributions of *D. carljonesi* on the black-nest, white-nest and mossy-nest swiftlet could also be compared.

EFFECT OF LOUSE BEHAVIOUR ON THE FEATHER DIMENSIONS AT WHICH THEY OCCUR. The effect of louse microhabitat shifts on the feather dimensions at which lice occurred on different host species was investigated for both *D. somadikartai* and *D. carljonesi*. This was carried out in two stages, for each louse species. First, I measured how different the microhabitat dimensions for lice on different host species would have been if transferred lice had not altered their distribution patterns. This was accomplished by assuming that the number of transferred lice recaptured from each host species was equal to the number recaptured from controls, and occurred in the same distribution pattern. The control distribution on each host species was characterised, with regards to each of the four feather dimensions measured, by the corresponding arrays of mean dimensions (mean

Figure 5.3 a) Flight feather. The asterisk marks the relative point on the underside where dimensions were measured for each feather. b) Close up of flight feather underside, indicating relevant structures (both figures after Choe & Kim 1988).



structural dimensions for each feather tract on each host species). For example, if nine female *D. somadikartai* were recaptured from the glossy swiftlet (having been transferred as controls), three of which occurred on tail feathers (where the mean barb diameter of glossy swiftlet tail feathers was p), three on primary feathers (where the mean barb diameter was q) and three on secondary feathers (where the mean barb diameter was r), the dimension array of barb diameters at which female *D.*

somadikartai occurred on the glossy swiftlet would be " $p, p, p, q, q, q, r, r, r$ ".

Microhabitat dimension arrays (for control louse distributions) were then compared among host species, using non-parametric statistics, for each feather dimension measured. Significant differences were interpreted as instances where louse microhabitat dimensions on different host species would be different if lice did not alter their distribution patterns.

Second, I measured how (if at all) the microhabitat dimensions for lice on different host species were made more similar by lice altering their distribution patterns. The same procedure as above was carried out, for each louse species, using the actual numbers of transferred lice recaptured from each host species and the actual distribution patterns in which they occurred. Microhabitat dimension arrays were again compared among host species, using non-parametric statistics, for each feather dimension measured. Significant differences were interpreted as instances where louse microhabitat dimensions on different host species would still be different, even with lice altering their distribution patterns. However, if microhabitat dimension arrays, which were significantly different among swiftlet hosts for control louse distributions, were not significantly different for actual louse distributions, this would imply that lice may have been shifting their microhabitat distribution to keep feather dimensions "constant".

5.3 Results

FEATHER DIMENSIONS. There was a high degree of overlap between swiftlet species in feather dimensions (Figures 5.4 & 5.5). However, all four dimensions measured were significantly correlated with wing chord (analysing measurements from all feather tracts together). Barb diameter (Spearman $r_s = 0.39$, $p = 0.003$), distance between barbs ($r_s = 0.32$, $p = 0.01$), barbule diameter ($r_s = 0.34$, $p = 0.01$), and distance between barbules ($r_s = 0.36$, $p = 0.005$) all increased with increasing wing chord (all four dimensions were highly correlated with each other ($r_s \geq 0.67$, $p < 0.001$ for all paired combinations)). Furthermore, there were significant differences among feather tracts in all dimensions measured (analysing measurements from all swiftlet species together). Barb diameter (Kruskal-Wallis $H = 40.64$, $p < 0.001$), distance between barbs ($H = 37.48$, $p < 0.001$), barbule diameter ($H = 23.71$, $p < 0.001$), and distance between barbules ($H = 38.55$, $p < 0.001$) all increased from secondary $\rightarrow$ primary $\rightarrow$ tail feathers.

MICROHABITAT DISTRIBUTIONS OF GLOSSY SWIFTLET LICE. Female *Dennyus somadikartai* occurred in significantly different microhabitat distributions on the glossy and mossy-nest swiftlet (Figure 5.6). Whilst no lice occurred on body feathers of either host species, proportionally more lice occurred on secondary feathers of the mossy-nest swiftlet than on secondary feathers of the glossy swiftlet ($\chi^2 = 9.87$, d.f. = 1, $p < 0.005$).

EFFECT OF LOUSE BEHAVIOUR ON THE FEATHER DIMENSIONS AT WHICH GLOSSY SWIFTLET LICE OCCUR. When microhabitat dimensions were compared for female *D. somadikartai* on the glossy and mossy-nest swiftlet, under the assumption that the number of transferred lice recaptured from the mossy-nest swiftlet was equal to the number recaptured from the glossy swiftlet (control host for this set of transfers) and occurred in the same distribution pattern, significant differences were seen (Figure 5.7). If the lice had not altered their distribution pattern

Figure 5.4 Measurements of a) barb diameter and b) distance between barbs for the secondary, primary and tail feathers of the glossy, mossy-nest, white-nest and black-nest swiftlet. Diameter of points is proportional to number of individual birds (1-4). Numbers in parentheses are the mean wing-chord (mm) for the five individuals of each swiftlet species for which feather dimensions were measured.

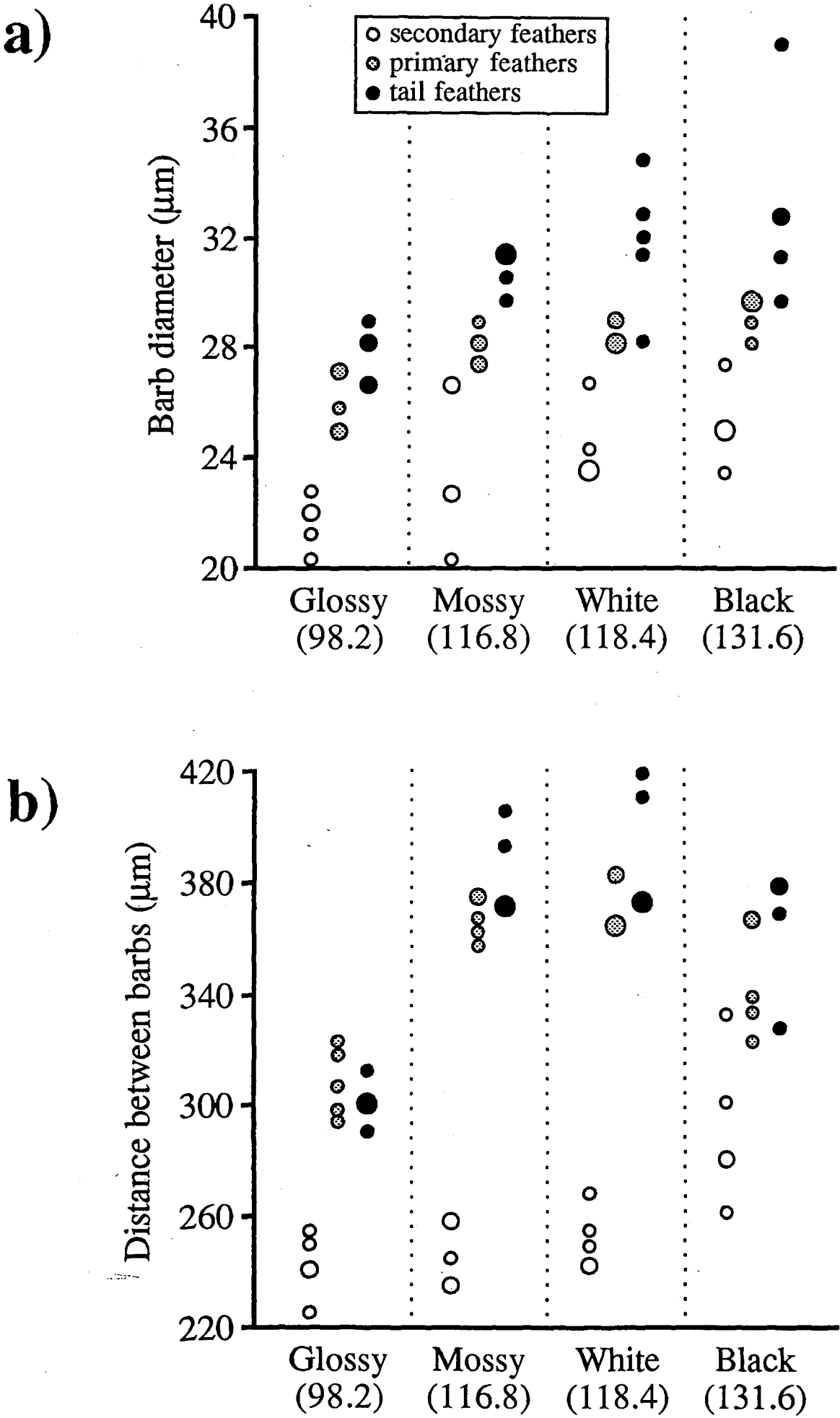


Figure 5.5 Measurements of a) barbule diameter and b) distance between barbules for the secondary, primary and tail feathers of the glossy, mossy-nest, white-nest and black-nest swiftlet. Diameter of points is proportional to number of individual birds (1-4). Numbers in parentheses are the mean wing-chord (mm) for the five individuals of each swiftlet species for which feather dimensions were measured.

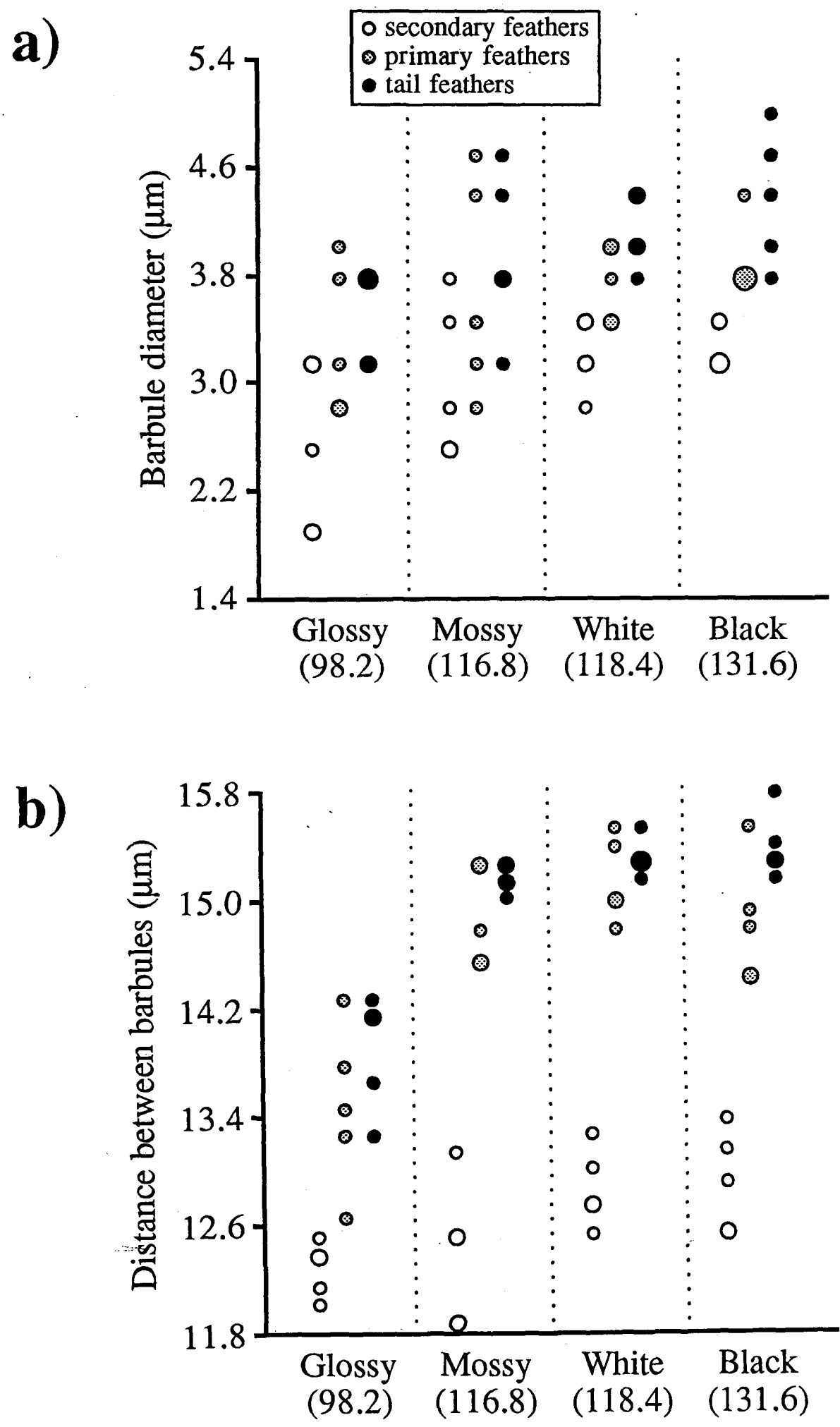


Figure 5.6 Microhabitat distributions of transferred female *Dennyus somadikartai* recaptured from the glossy and mossy-nest swiftlet.

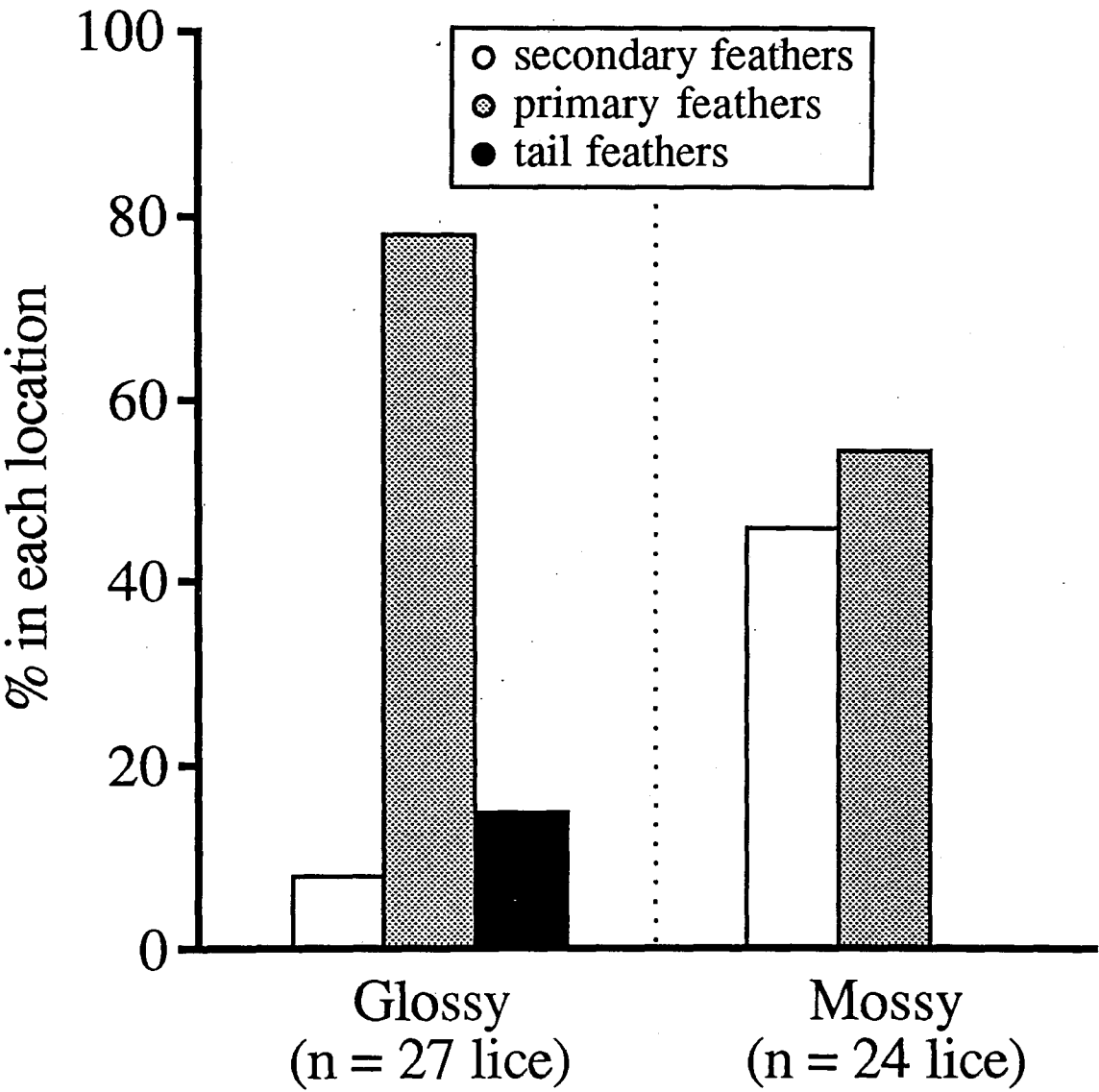
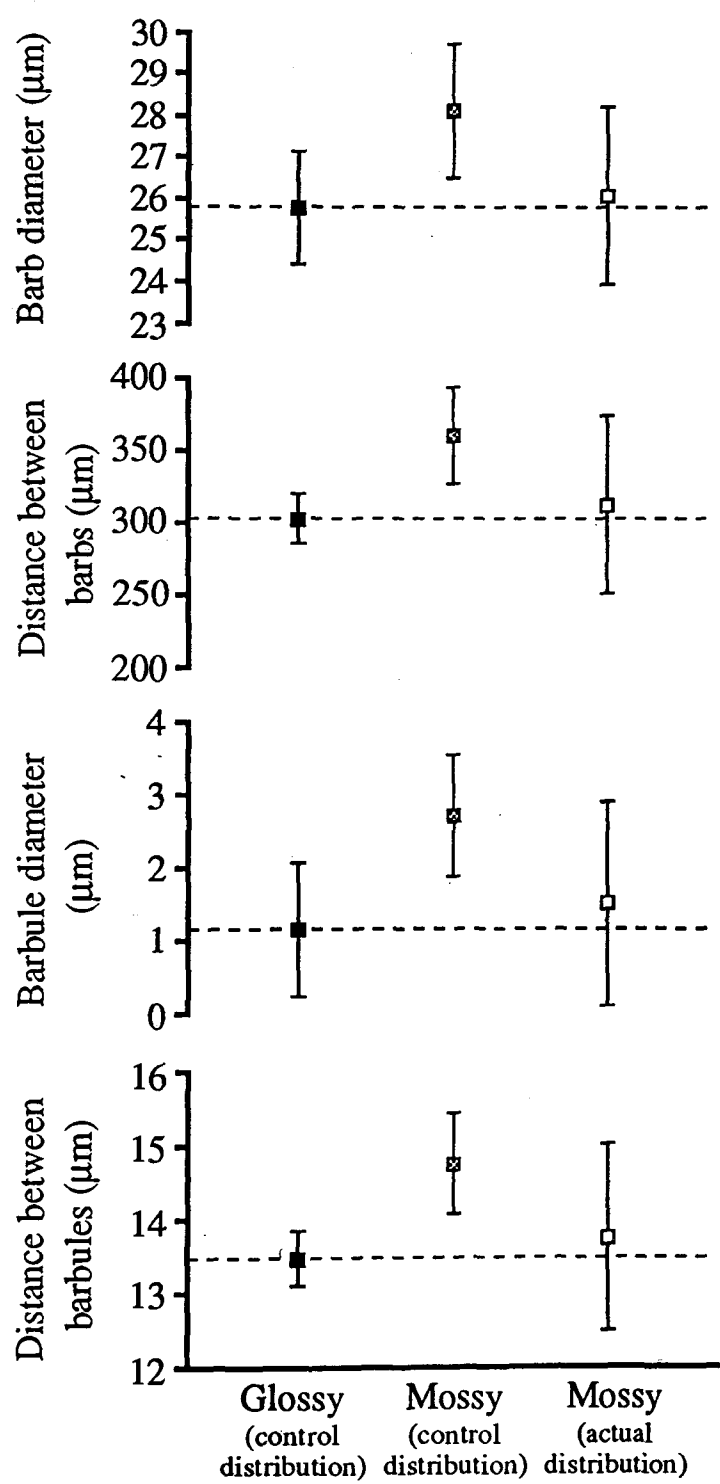


Figure 5.7 Mean ($\pm$ s.d.) microhabitat dimensions of female *Dennyus somadikartai* on the glossy and mossy-nest swiftlet. Glossy swiftlet feather dimensions, for the microhabitat distribution in which lice occurred on the glossy swiftlet (control distribution), are compared to mossy-nest swiftlet feather dimensions, first for the control distribution (i.e. if the lice had not shifted microhabitat), and second for the distribution in which lice actually occurred on the mossy-nest swiftlet. Microhabitat shifting caused the feather dimensions at which lice occurred to become more similar on both host species. (n = 27 lice on the glossy swiftlet, 24 on the mossy-nest swiftlet)



the microhabitat dimensions, of barb diameter (Mann-Whitney $U = 50$, $p < 0.001$), distance between barbs ($U = 50$, $p < 0.001$), barbule diameter ($U = 50$, $p < 0.001$) and distance between barbules ($U = 50$, $p < 0.001$), at which they would have occurred would have been greater on the mossy-nest than on the glossy swiftlet (U -values were identical due to the four feather dimensions being highly correlated with each other).

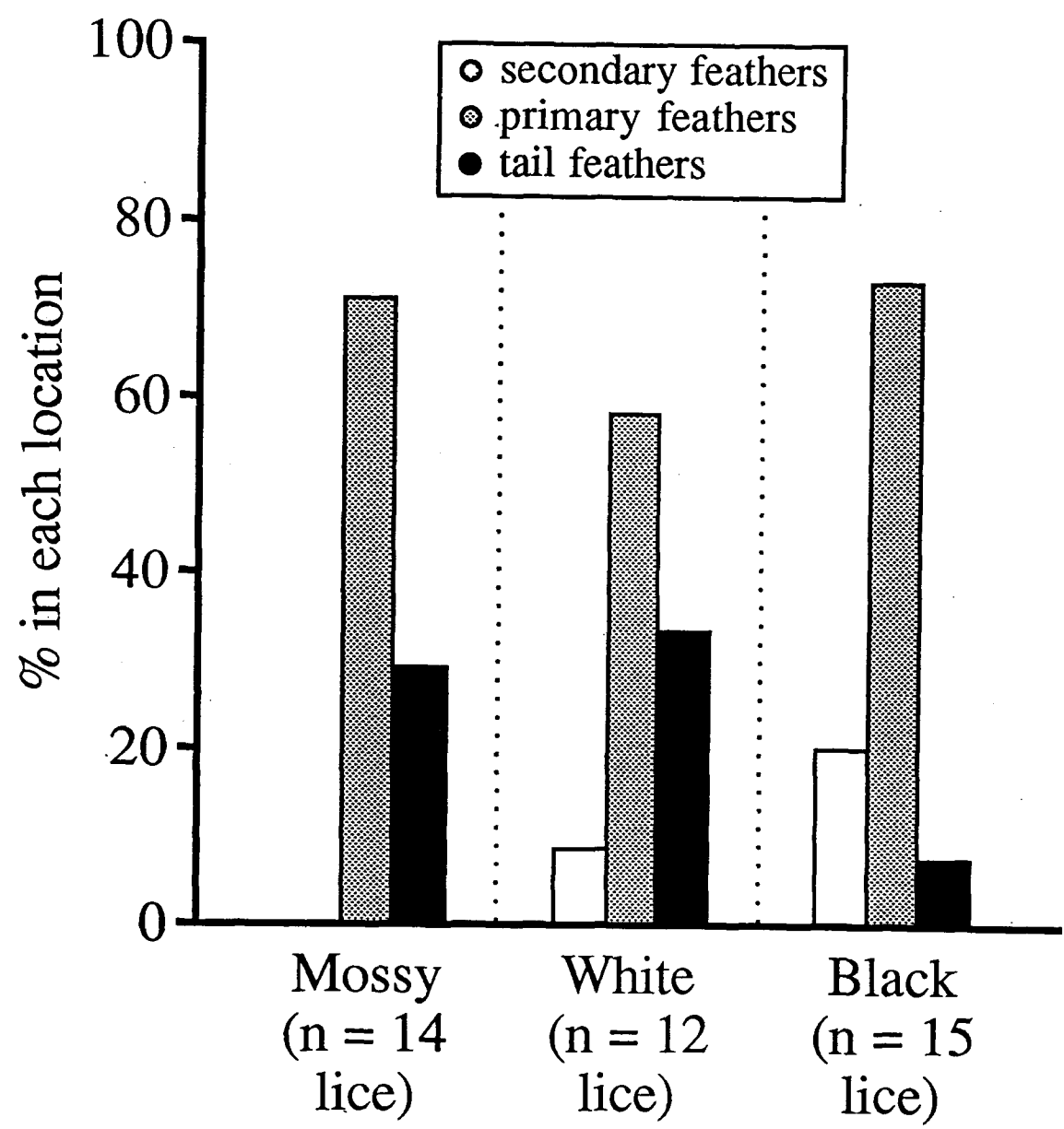
However, when microhabitat dimensions were compared for female *D. somadikartai* on the glossy and mossy-nest swiftlet, using the actual number of transferred lice recaptured from each host species and the actual distribution patterns in which they occurred, no significant differences were seen (Figure 5.7). The microhabitat dimensions, of barb diameter ($U = 275$, $p = 0.36$), distance between barbs ($U = 275$, $p = 0.36$), barbule diameter ($U = 275$, $p = 0.36$) and distance between barbules ($U = 275$, $p = 0.36$), at which the lice actually occurred were similar on both host species. This implies that female *D. somadikartai* may have been shifting their microhabitat distribution to keep feather dimensions "constant".

MICROHABITAT DISTRIBUTIONS OF BLACK-NEST SWIFTLET LICE.

Dennyus carljonesi occurred in (non-significantly) different microhabitat distributions on the black-nest, white-nest and mossy-nest swiftlet (Figure 5.8). Whilst no lice occurred on body feathers of any host species, proportionally fewer lice occurred on tail feathers of the black-nest swiftlet than on tail feathers of the white-nest and mossy-nest swiftlets (Fisher exact $p = 0.08$). Furthermore, proportionally more lice occurred on secondary feathers of the black-nest swiftlet than on the secondary feathers of the mossy-nest swiftlet (Fisher exact $p = 0.12$).

EFFECT OF LOUSE BEHAVIOUR ON THE FEATHER DIMENSIONS AT WHICH BLACK-NEST SWIFTLET LICE OCCUR. When microhabitat dimensions were compared for *D. carljonesi* on the black-nest, white-nest and mossy-nest swiftlet, under the assumption that the number of transferred lice recaptured from each species was equal to the number recaptured from the black-nest swiftlet (control

Figure 5.8 Microhabitat distributions of transferred *Dennyus carljonesi* recaptured from the mossy-nest, white-nest and black-nest swiftlet.



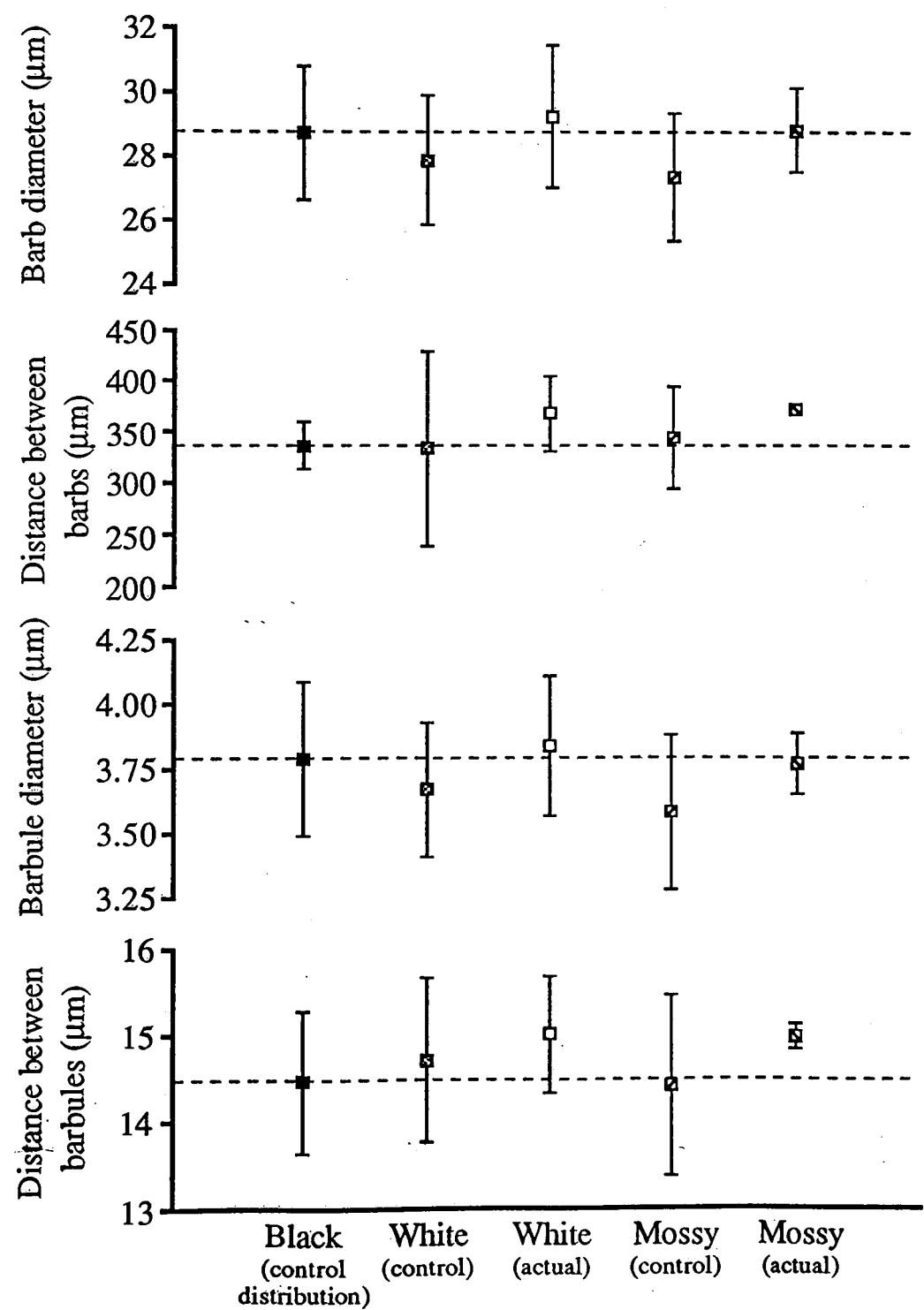
host for this set of transfers) and occurred in the same distribution pattern, significant differences were seen (Figure 5.9). If the lice had not altered their distribution pattern the microhabitat dimensions, of barb diameter (Kruskall-Wallis $H = 13.27$, $p = 0.001$), distance between barbs ($H = 13.59$, $p = 0.001$), barbule diameter ($H = 13.27$, $p = 0.001$) and distance between barbules ($H = 10.46$, $p = 0.005$), at which they would have occurred would have been different on the three host species.

When microhabitat dimensions were compared for *D. carljonesi* on the black-nest, white-nest and mossy-nest swiftlet, using the actual number of transferred lice recaptured from each host species and the actual distribution patterns in which they occurred, significant differences were still seen (Figure 5.9). The microhabitat dimensions of distance between barbs ($H = 24.98$, $p < 0.001$) and distance between barbules ($H = 18.88$, $p < 0.001$), at which the lice actually occurred, remained different on the three host species. However, the microhabitat dimensions of barb diameter ($H = 2.82$, $p = 0.24$) and barbule diameter ($H = 2.82$, $p = 0.24$), at which the lice actually occurred, were similar on all three host species. This implies that *D. carljonesi* may have been shifting their microhabitat distribution to keep the feather dimensions of barb diameter and barbule diameter, but not distance between barbs or distance between barbules, "constant".

5.4 Discussion

Adults of both *Dennyus somadikartai* and *Dennyus carljonesi* occurred exclusively on the flight feathers (primary, secondary and tail feathers) of their swiftlet hosts (Figures 5.6 & 5.8). Thus, examination of these feathers provided an accurate picture for the structural dimensions of the microhabitat of these lice. It was hypothesised that for *Dennyus* spp. to be able to survive on different sized swiftlet hosts there must be variation in feather dimensions which overlaps between swiftlet species. This is

Figure 5.9 Mean ($\pm$ s.d.) microhabitat dimensions of *Dennyus carljonesi* on the black-nest, white-nest and mossy-nest swiftlet. Black-nest swiftlet feather dimensions, for the microhabitat distribution in which lice occurred on the black-nest swiftlet (control distribution), are compared to feather dimensions of the other host species, first for the control distribution (i.e. if the lice had not shifted microhabitat), and second for the distributions in which lice actually occurred on the other host species. Microhabitat shifting caused the feather dimensions of barb diameter and barbule diameter, at which lice occurred, to become more similar on all host species. (n = 15 lice on the black-nest swiftlet, 12 on the white-nest swiftlet, 14 on the mossy-nest swiftlet)



the pattern observed (Figures 5.4 & 5.5). The null hypothesis that feather dimensions do not vary among feather tracts of swiftlet species is rejected. The dimensions of detailed feather structure increase from secondary → primary → tail feathers.

Furthermore, there is a high degree of overlap in flight feather dimensions among all four swiftlet species involved in this study.

The null hypothesis that the dimensions of detailed feather structure are not significantly correlated with swiftlet body size is rejected. All four feather dimensions measured (barb diameter, distance between barbs, barbule diameter and distance between barbules) increase with increasing swiftlet wing-chord (Figures 5.4 & 5.5). Due to this positive correlation, overlap in flight feather dimensions is greater for swiftlets with more similar body sizes. Since lice, experimentally transferred among swiftlet species in Chapter 4, were able to survive on host species with similar body sizes but not on host species with dissimilar body sizes (Figure 5.1), degree of overlap in feather dimensions may have been determining louse survival (as hypothesised in Figure 5.2).

To utilise similar microhabitat dimensions on swiftlet hosts of different size, it was hypothesised that the same louse species must occupy different microhabitats on different host species. This is the pattern observed (Figures 5.6 & 5.8). The null hypothesis, that the same species of louse does not occupy different microhabitat distributions on host species with different body sizes, is rejected for both louse species involved in this study. With increasing host body size, *D. somadikartai* shifted its microhabitat away from tail and primary feathers, towards secondary feathers. With increasing host body size, *D. carljonesi* shifted its microhabitat from tail → primary → secondary feathers. Thus, on larger hosts, both louse species shifted their microhabitats towards flight feathers with smaller structural dimensions. This implies that lice may have been altering their microhabitat to keep certain feather dimensions "constant".

When the observed behavioural shifts in louse microhabitat distribution were combined with the measurements of host feather dimensions, it was seen that the microhabitat dimensions of barb and barbule diameter, at which lice occurred, were being kept "constant" on different swiftlet species (Figures 5.7 & 5.9). If either louse species had not altered their distribution patterns the microhabitat dimensions of barb and barbule diameter, at which they occurred, would have been significantly different on different host species. Thus barb and/or barbule diameter may be host characters important for louse survival on swiftlets. The larger louse, *D. carljonesi*, occurred at greater microhabitat dimensions, on swiftlet hosts, than the smaller louse, *D. somadikartai* (female *D. carljonesi* are 8% larger than female *D. somadikartai*; Clayton *et al.* in review). This implies that components of louse morphology, which are proportional to overall body size, may be adapted to host feather dimensions.

This study supports the idea that lice are able to survive on any of several hosts, where variation in microhabitat structure overlaps, by shifting their microhabitat. For *Dennyus* lice on Malaysian swiftlets, where feather dimensions are positively correlated with wing chord, the observations here are in keeping with the patterns of louse survival on different hosts seen in Chapter 4 (Figure 5.1). *D. somadikartai* had approximately 25% survival (relative to controls) when transferred from the glossy swiftlet (its normal host species) to the mossy-nest swiftlet (where there is some overlap with the glossy swiftlet in the barb and barbule diameters of flight feathers; see Figures 5.4 & 5.5). In contrast, there was approximately 0% survival when this louse was transferred to the black-nest swiftlet, where there is less overlap. *D. carljonesi* had survival equivalent to controls when transferred from the black-nest swiftlet to either of its other normal host species: white-nest or mossy-nest swiftlets (where there is a high degree of overlap between host species in barb and barbule diameters). *D. distinctus* had approximately 0% survival when transferred from the glossy swiftlet (its normal host species) to the mossy-nest swiftlet (where, again, there is some overlap between host species in barb and barbule diameters).

The difference in survival of *D. distinctus* and *D. somadikartai*, when transferred from their normal host species (the glossy swiftlet) to a larger host species (the mossy-nest swiftlet) was most likely caused by less suitable microhabitat being available to *D. distinctus* on the larger host. In Chapter 2, adult lice were seen on body feathers of the glossy swiftlet. Since, in this study, no *D. somadikartai* occurred on body feathers of the glossy swiftlet, the lice on body feathers in Chapter 2 were most likely the other species present, *D. distinctus*. Since swiftlet body feathers have smaller structural dimensions than flight feathers (personal observation), *D. distinctus* may require smaller microhabitat dimensions for survival than *D. somadikartai*. Also, female *D. distinctus* are 10% smaller than female *D. somadikartai* (Clayton *et al.* in review). Thus, a larger host (with larger flight feather dimensions) would have less suitable microhabitat available for *D. distinctus* than for *D. somadikartai*, resulting in reduced survival.

The results of this study imply that the dimensions of barb and barbule diameter, of the flight feathers on which these lice occur, are proximate determinants of the survival of *Dennyus* lice on their Malaysian swiftlet hosts. For mites on birds, adaptations to detailed feather structure are important for attachment to the host, feeding from the host, and reproducing on the host (Kethley 1971; Kethley & Johnston 1975; Choe & Kim 1991; Fain 1994). For chewing lice on pocket gophers, adaptations to detailed hair structure are important for attachment to the host (Reed 1994). *Dennyus* spp. on swiftlets may be adapting to host feather barb and/or barbule diameter for any of the above functions. Lice may also be adapted to occur in a species-specific microhabitat distribution (with feather barb and/or barbule diameter as a cue) to ensure that lice meet to reproduce on hosts where prevalence and intensity of louse infestation is often low (Rohde 1979). However, this is unlikely to effect louse survival.

Louse species may need to stay in a particular microhabitat of their host's feathers in case they need to grip onto a feather barbule of a particular size with their mouthparts to ensure they are not dislodged from their hosts. Lice on pocket gophers grip onto host hairs with particular dimensions using a rostral groove located on the head of the louse (Reed 1994). Chewing lice on pigeons have been observed gripping onto host feather barbules with their mouthparts (Nelson & Murray 1971). Ability to grip the host may be important for the survival of *Dennyus* lice experimentally transferred to nestling swiftlets (Chapter 4), as nestlings flap their wings vigorously in preparation for flight whilst still in the nest (personal observation). Host preening is unlikely to be involved, since nestling swiftlets do not preen (personal observation).

The mouthparts of lice on swiftlets may also be adapted for feeding on feather barbules of a particular size range. Species of amblyceran lice have been observed feeding on feather particles (Marshall 1981a), although this is unlikely here since feather particles were never seen in the gut contents of *Dennyus* lice removed from swiftlets (Roger Price, personal communication).

I concluded in Chapter 4 that chewing lice on Malaysian swiftlets have sufficient tolerance to environmental variation to survive on closely related hosts, but not on distantly related hosts. This was based on the results of transfer experiments carried out in both Chapter 4 and Reed (1994), where chewing lice also had reduced survival when transferred between more distantly related pocket gophers. The results here suggest that the relationship between swiftlet body size (and thus feather dimensions) and relatedness is the proximate cause of the relationship between transferred louse survival and host relatedness. Furthermore, they suggest that louse behaviour (microhabitat shifts) enables survival on hosts with similar feather dimensions.

Chapter 6 Effect of vertically transmitted ectoparasites on the reproductive success of swifts (*Apus apus*)

[Tompkins, D.M., Jones, T. & Clayton, D.H. In press. Effect of vertically transmitted ectoparasites on the reproductive success of swifts (*Apus apus*). *Functional Ecology*.]

6.1 Summary

1. Parasites that are transmitted vertically from parent hosts to offspring are expected to be relatively benign since their fitness depends on successful host reproduction. We tested the effects of two species of vertically transmitted ectoparasites on the reproductive success of swifts (*Apus apus*). We experimentally manipulated populations of the chewing louse *Dennyus hirundinis* (Phthiraptera: Menoponidae) and the flightless louse fly *Crataerina pallida* (Diptera: Hippoboscidae), effectively converting the natural aggregated frequency distribution of each species into a bimodal distribution of high and low loads.
2. Neither parasite had any effect on nestling growth or fledging success, even though parasite loads were boosted above natural levels, and host environmental conditions were poor during part of the study, thus increasing the chances of detecting an effect of the parasites.
3. In contrast to parasite load, year, brood size and hatch date were all significantly related to components of nestling growth. Year and brood size were also significantly related to fledging success.
4. Our results are consistent with theoretical models suggesting that vertically-transmitted parasites evolve reduced virulence because they depend on host reproduction for dispersal to new hosts.

Keywords: Chewing lice, Hippoboscidae, louse fly, Phthiraptera, virulence.

6.2 Introduction

Ectoparasites are known to reduce several components of avian fitness (Lehman 1993; Brown, Brown & Rannala 1995) and influence a range of host life history variables (Møller in press). Some ectoparasites, however, appear to have little or no effect on the host (Clayton & Tompkins 1994, 1995). Observational studies of chewing lice and louse flies on swifts (*Apus apus*) and of louse flies on alpine swifts (*Apus melba*) showed no correlation between parasite load and host condition, survival or reproductive success (Hutson 1981; Lee & Clayton 1995; Tella *et al.* 1995). One possible reason for the apparent avirulence of swift lice and flies is that observational studies lack the inferential power required to detect subtle effects. Parasites generally show an aggregated frequency distribution among hosts with most individuals having few parasites and a few individuals having many parasites (Anderson & Gordon 1982). Subtle effects of such parasites may be overlooked unless very large samples are studied (Booth *et al.* 1993).

An alternative explanation for avirulence is that swift lice and flightless louse flies, both of which are vertically transmitted from parent hosts to their offspring, have evolved reduced virulence. All else being equal, vertically transmitted parasites are expected to be less virulent than parasites capable of horizontal transmission to unrelated hosts because the fitness of vertically transmitted parasites is tightly linked to the reproductive success of the host (Anderson & May 1982; Ewald 1983; Clayton & Tompkins 1994). In reducing host fitness, vertically transmitted parasites reduce their own fitness, thus selecting for a reduction in virulence.

We conducted an experimental field study to test whether vertically transmitted ectoparasites of swifts have an impact on host reproductive success. We transferred lice and louse flies among nests to convert the aggregated distributions of these parasites (Lee & Clayton 1995) into bimodal distributions of high- and low-load nests. We then compared the growth and fledging success of nestlings in high- and low-load

nest. Nestling body mass is known to be influenced by environmental factors such as weather (Bryant 1978), and is often correlated with post-fledging size and survival in birds (Boag 1987; Magrath 1991). Body mass is therefore a component of fitness that parasites might easily affect (Møller 1994). For example, Johnson & Albrecht (1993) reported a slight impact of haematophagous ectoparasites on the body mass of nestling house wrens, even though they detected no impact of ectoparasites on haematocrit, tarsal growth or feather growth.

We also examined the effect of several non-parasite factors (year, brood size and hatching date) on nestling growth and fledging success. These variables are known to have strong effects on the reproductive success of swifts (Lack & Lack 1951; Lack 1956b), and are also known to interact with parasite effects on avian hosts (de Lope *et al.* 1993; Møller 1993). Finally, we took blood and faecal samples to check nestlings for endoparasites. Both chewing lice and louse flies act as vectors for avian endoparasites (Baker 1967; Balashov 1984), transmission of which may be one of the fitness costs of ectoparasite infestation of swifts (Dutton 1905).

6.3 Background

Swifts (*Apus apus* (Linnaeus)) are aerial, insectivorous birds (Cucco *et al.* 1993) that breed in Eurasia and overwinter in sub-Saharan Africa (small numbers winter in northern India and Arabia) (Chantler & Driessens 1995). The swifts in this study were breeding in nest-boxes in the tower of the Oxford University Museum of Science. Swifts have used these boxes annually since 1948 (Lack 1956a). Birds in the museum colony normally arrive from the South African wintering grounds during the first week of May. A clutch of two to three eggs is laid several weeks later and is incubated for three weeks. Young birds fledge in late July or early August at a mean age of 41 days post-hatching (range = 37 - 51, $n = 96$; Lee & Clayton 1995). Soon

after fledging they begin the long migratory flight to Africa. Swifts do not breed until four years of age (Perrins 1971), and they often use the same nest site each year.

The most common species of chewing louse on swifts is *Dennyus hirundinis* (Linnaeus). It is a "permanent" ectoparasite that completes its entire life cycle on the body of the host (Lee & Clayton 1995). *D. hirundinis* feeds on dermal debris, blood and host eye-fluid (Rothschild & Clay 1952; Bromhall 1980; Lee & Clayton 1995).

The flightless louse fly *Crataerina pallida* (Latreille) (which has vestigial wings) is a nest-based parasite that feeds on swift blood (Bequaert 1953). Adults feed about every five days and take up to 25mg of blood per feeding (Kemper 1951), which is nearly 5.0% of the total blood volume of an adult swift (Lee & Clayton 1995). The lifecycle of *C. pallida* is attuned to that of its host, with adult flies emerging during early summer from pupae that have over-wintered in the nest (Marshall 1981a). There are no records of adult flies on wintering swifts in Africa (Zumpt 1966).

D. hirundinis populations are relatively easy to quantify (Lee & Clayton 1995). The eggs, which are glued to the feathers with a glandular cement, are large enough to see with the naked eye (1 mm long) and are white, making them easy to detect in the host's dark plumage. The post-hatching stages, consisting of three nymphal instars and the adult, are also relatively large and easy to see. Populations of *D. hirundinis* are usually fairly small and are therefore tractable (< 12 adults per host; Lee & Clayton 1995).

C. pallida populations are also easy to quantify (Lee & Clayton 1995). Adult flies are large (7 mm) and easy to observe on the host or in its nest. The pupae are large (4 mm) and black, making them easy to see in the nest - particularly in the Museum Tower, where each nest is in a solidly constructed nest box. Like swift lice, louse flies occur in small, tractable populations (< 5 adults per nest; Lee & Clayton 1995).

Transmission of *D. hirundinis*, which requires direct contact among individual swifts, is constrained by the fact that swifts spend all of their time flying when away from the nest (Lack 1956a). The main route of dispersal for swift lice is vertical transmission from adult birds to their offspring in the nest (Lee & Clayton 1995). Lice are presumably also exchanged between mated adults. Some horizontal transmission may occur between unrelated males during prolonged fights over nest-boxes at the start of the breeding season (Lack 1956a). DNA fingerprinting reveals little extra-pair paternity in our swift colony (<5.0%; Jeremy Blakey, unpublished data), making extra-pair copulations an unlikely route of louse transmission.

Louse flies, unlike lice, are capable of efficient locomotion away from the body of the host (Marshall 1981a). Nevertheless, there is little horizontal transmission of the flightless *C. pallida* among nests in our colony (Lee & Clayton 1995; personal observation). The present study was carried out over two host breeding seasons; there was a strong correlation between number of pupae in nests at the end of the first breeding season and number of emerged flies in nests at the beginning of the second breeding season (Spearman $r_s = 0.84$, $n = 36$, $p < 0.001$). Also, nests with high numbers of flies at the beginning of a breeding season (natural loads or experimentally increased; see below) maintained high numbers through to the end of that season. Nests with low numbers of flies at the beginning of a breeding season (natural loads or experimentally decreased) maintained low numbers through to the end of that season. Even under conditions of high density, flies did not transmit horizontally between nests. The nearest neighbour distance for each nest in this study was 0.23m, but most nests were much further away (nest-boxes in the Museum tower are arranged in pairs). An independent mark-recapture experiment (Summers 1975) showed little horizontal transmission of the congeneric species *Crataerina hirundinis*, which lives in the nests of house martins (*Delichon urbica*). Only six of 96 flies (6.25%) dispersed between different nests in Summer's (1975) study which had a nearest neighbour distance of 0.13m. Thus flightless louse flies, like lice, are mainly transmitted vertically.

6.4 Materials and methods

The study was carried out May-August of 1993 and 1994. We manipulated lice and louse flies by manually transferring them from donor (low-load) to recipient (high-load) nests. Nests were assigned treatments using a randomised block design. Nests containing at least two nestlings were blocked into groups of three nests based on brood size (two vs. three nestlings) and hatching synchrony (eggs hatching within a span of 4 days). One nest in each block was randomly assigned as a recipient. The other two nests were assigned as donors. Nests with brood sizes of one were also used as donors, but were excluded from all analyses.

The goal of the transfers was to boost the parasite loads of recipient nests above the natural loads observed in the swift colony during May-August 1992 (see Lee & Clayton 1995). Several donor nests were required to create each high-load recipient nest. Over the course of two field seasons we were able to create a total of 13 high-lice nests (8 in '93, 5 in '94), and 13 high-fly nests (6 in '93, 7 in '94). This left 44 nests with low loads of both parasites (23 in '93, 21 in '94). Three nests in 1993 were given high loads of both parasites (these nests are already included in the sample sizes for high-load nests given above). Nests with high parasite loads in the first year of the study were assigned as low-load nests in the second year.

Nests were checked between the hours of 08:00 and 12:00 every day in 1993 and every other day in 1994. Adults were normally away foraging at this time of day, except for a one week period of non-stop brooding immediately after the eggs hatched. At each visit nestling mass was measured to the nearest 0.1g with a pesola spring balance. Nest mates were distinguished by clipping the tip of one toenail.

Lice were moved from adult birds in donor nests to nestlings in recipient nests when the nestlings were 16-18 days old. This is the age at which transmission of lice, from adults to nestlings, begins (Lee & Clayton 1995). Lice are never seen on nestlings

before this age (Lee & Clayton 1995; personal observation). Swifts in donor nests were examined for periods of 10 minutes under a head lamp and any lice or louse flies observed were removed with a pooter or forceps and placed in a 1.5 ml microcentrifuge tube. Within several hours the parasites were transferred to nestlings in recipient nests. Lice not captured during the initial transfer were moved from donor nestlings during a second bout of transfers when recipient nestlings were 25-27 days of age. When nestlings reached pre-fledging age (35-37 days) their louse populations were quantified as described in Lee & Clayton (1995).

Flies were moved from donor to recipient nests when both contained 1-3 day old nestlings. Throughout the study nests and nestlings were searched for flies every 1-2 days for a period of 1 minute with illumination from a head lamp. Any flies encountered in donor nests after the initial transfer were distributed evenly among recipient nests. At the end of the breeding season (two weeks after the departure of all birds) the contents of each nest box were thoroughly examined for newly deposited fly pupae to check for evidence of flies which had been missed previously.

When nestlings were 25-27 days of age a blood smear was taken to check for blood parasites. Smears were fixed in 100% ethanol and stained in Giemsa's solution. Faecal samples were also taken from 25-37 day old nestlings to check for coccidian parasites. Samples were collected whenever defaecation was observed (08:00 - 12:00). Faecal samples were stored in 2.5% potassium dichromate and treated with saturated NaCl solution to float off coccidia. Blood and faecal samples were searched for parasites using 10x and 40x objectives of a phase-contrast microscope.

6.5 Statistical analyses

A modified Richards sigmoid growth model (Brisbin *et al.* 1986) was fitted to the body mass measurements of each nestling. As body mass recession occurs in swift

nestlings prior to fledging (Lack & Lack 1951) growth curves were fitted from hatching through two days after the maximum recorded mass was attained (approximately 25 days of age). Three parameters for each growth curve were predicted: asymptotic mass, growing period (hatching through age at asymptote) and the Richards shape parameter (m). Near $m = 2.0$, 0.67 , 0.0 or when $m \rightarrow 1.0$, the Richards model becomes the logistic, von Bertalanffy, monomolecular or Gompertz models respectively (Richards 1959). A fourth parameter, mean growth rate, was calculated by dividing asymptotic mass by growing period. Measurements of fledging mass and age were taken from the raw data. Mean parameter values per nest were analysed using "General Linear Models", incorporating year, brood-size, hatch-date, louse-load and fly-load as separate factors. All non-significant interaction factors were discarded. Only nestlings that survived to fledge were included in the analyses. Mean hatching dates were assigned to one of three weeks in order to test for temporal trends in reproductive success (all nests hatched between June 14th and July 4th in 1993, and between June 21st and July 11th in 1994).

Fledging success was analysed non-parametrically using contingency tests. Nestlings that disappeared prior to the minimum fledging age (37 days; Lack 1956a) were presumed to have fallen through the hole in the floor which forms the entrance to each nest box (Lack 1956a). Dead nestlings were occasionally recovered at the base of the tower.

To investigate the possibility of Type II errors in the analyses of parasite effects on components of nestling growth, a statistical power test was carried out (Lipsey 1990). Johnson & Albrecht (1993) reported a slight impact of haematophagous ectoparasites on nestling body mass that had an "effect size" of approximately 0.75. "Effect size" is the difference in the mean of the variable under consideration in two experimental groups divided by the common standard deviation (Cohen 1988). We used an "effect size" of 0.75 to calculate the power of our study to detect effects of ectoparasites on swift nestlings that were equal in magnitude to those detected in the Johnson &

Albrecht study (equivalent to a difference in swift nestling growth rate of approximately 0.04 g/day).

6.6 Results

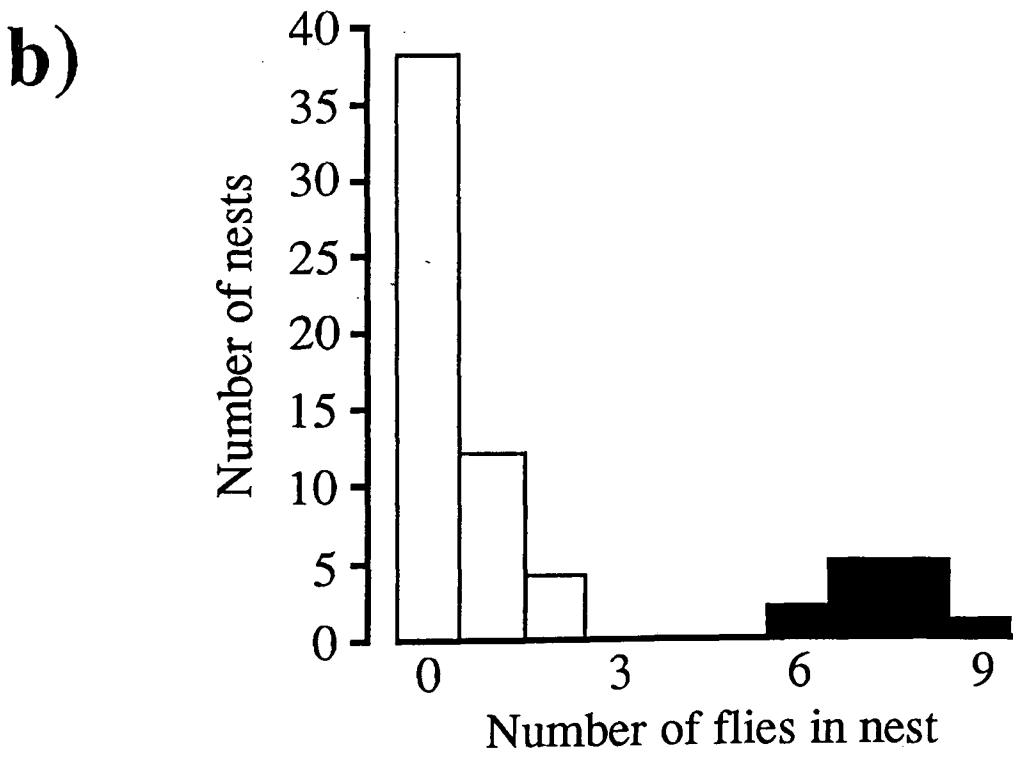
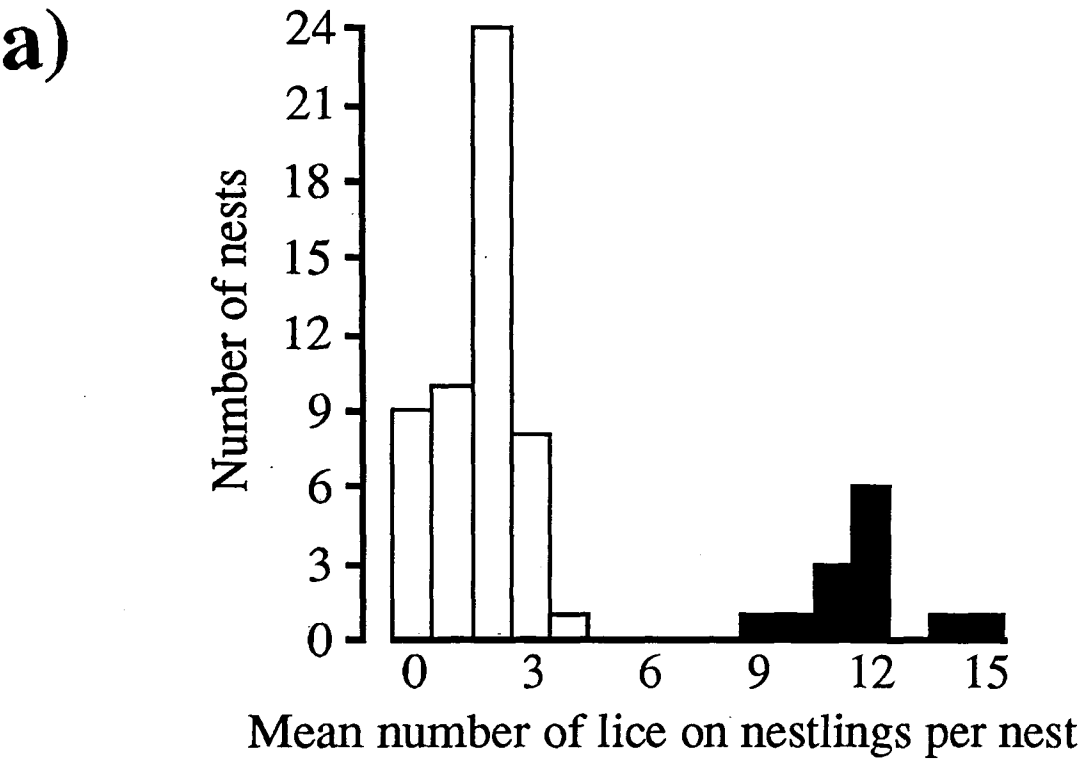
PARASITE LOADS. High-louse, high-fly and low-load nests had similar parasite loads prior to manipulation: mean ($\pm$ s.d.) louse loads on 16-18 day old nestlings were 0.44 ± 0.61 lice per nestling in low-louse nests, compared to 0.46 ± 0.97 lice per nestling in high-louse nests (Mann-Whitney $U = 303$, $p = 0.57$). Mean fly loads in nests of 1-3 day old nestlings were 1.20 ± 2.55 flies in low-fly nests, compared to 1.00 ± 1.53 flies in high-fly nests ($U = 316$, $p = 0.58$).

Experimental transfer of lice between nests had the desired effect. At the 25-27 day old census low-louse nests had a mean of 0.48 ± 0.58 lice per nestling compared to 3.46 ± 0.88 lice per nestling in high-louse nests ($U = 1$, $p < 0.001$). At the 35-37 day old census low-louse nests had a mean of 1.65 ± 1.01 lice per nestling compared to 11.69 ± 1.44 lice per nestling in high-louse nests (Figure 6.1a; $U = 0$, $p < 0.001$).

In comparing louse fly loads we used the maximum number of flies observed at any one count. This minimised the chance of missing flies temporarily away from the nest attached to foraging adult hosts. As *C. pallida* has but one generation per year, with flies emerging more or less synchronously in the spring (Lee & Clayton 1995), this approach would not have been confounded by short-term increases in louse fly populations. Over the course of the study a mean maximum of 0.37 ± 0.62 flies was observed in low-fly nests compared to 7.39 ± 0.87 flies in high-fly nests (Figure 6.1b; $U = 0$, $p < 0.001$). At the end of the breeding season low-fly nests contained a mean of 0.09 ± 0.35 new pupae compared to 9.54 ± 3.28 new pupae in high-fly nests ($U = 0$, $p < 0.001$).

Figure 6.1 (a) Distribution of chewing lice on 35-37 day old nestlings. (b)

Distribution of louse flies (maximum observed) in nests. Open bars are donor (low-load) nests; shaded bars are recipient (high-load) nests.



No haematozoa were detected in any of the blood samples examined (N = 89).

Likewise, no coccidia were detected in the faecal samples examined (N = 182).

NESTLING SURVIVAL. Neither parasite had a significant effect on fledging success (Figure 6.2). Forty-four of 54 low-louse nests fledged their entire brood, compared to 10 of 13 high-louse nests (Fisher exact $p = 0.49$). Forty-three of 54 low-fly nests fledged their entire brood, compared to 11 of 13 high-fly nests (Fisher exact $p = 0.78$). In contrast, year did have a significant effect on fledging success (Figure 6.2c): 24 of 34 nests fledged their entire brood in 1993, compared to 30 of 33 nests in 1994 ($\chi^2 = 4.41$, d.f. = 1, $p = 0.04$). Brood size also had an effect on fledging success (Figure 6.2d): 34 of 38 nests with broods of two fledged their entire brood compared to 20 of 29 nests with broods of three ($\chi^2 = 4.41$, d.f. = 1, $p = 0.04$). Hatch week had no significant effect on fledging success: 14 of 18 nests (78%) with eggs hatching in Week 1 fledged their entire brood, compared to 15 of 20 nests (75%) with eggs hatching in Week 3 (Fisher Exact $p = 0.57$).

NESTLING GROWTH. Neither parasite had a significant effect on any component of nestling growth (Figures 6.3 & 6.4). Nestlings in low-louse and high-louse nests did not differ significantly in asymptotic mass ($F = 0.10$, $p = 0.75$), growing period ($F = 0.30$, $p = 0.58$), growth rate ($F = 0.53$, $p = 0.47$), Richards shape parameter ($F = 0.44$, $p = 0.51$), fledging mass ($F = 0.62$, $p = 0.44$) or fledging age ($F = 0.02$, $p = 0.90$). Likewise, nestlings in low-fly and high-fly nests did not differ significantly in asymptotic mass ($F = 0.25$, $p = 0.62$), growing period ($F = 0.43$, $p = 0.51$), growth rate ($F = 0.24$, $p = 0.63$), Richards shape parameter ($F = 0.04$, $p = 0.85$), fledging mass ($F = 0.00$, $p = 1.00$) or fledging age ($F = 0.14$, $p = 0.71$).

On the other hand, year, brood size and hatch week all had significant effects on components of nestling growth (Figures 6.3 & 6.4). Nestlings in 1993 had lower asymptotic mass ($F = 59.86$, $p < 0.001$), slower growth rates ($F = 13.26$, $p = 0.001$), higher Richards shape parameters ($F = 4.65$, $p = 0.04$), and fledged at a later age ($F =$

Figure 6.2 Impact on fledging success of (a) louse load, (b) fly load, (c) year and (d) brood size. Values in bars are the percentages of nests that fledged their entire brood.

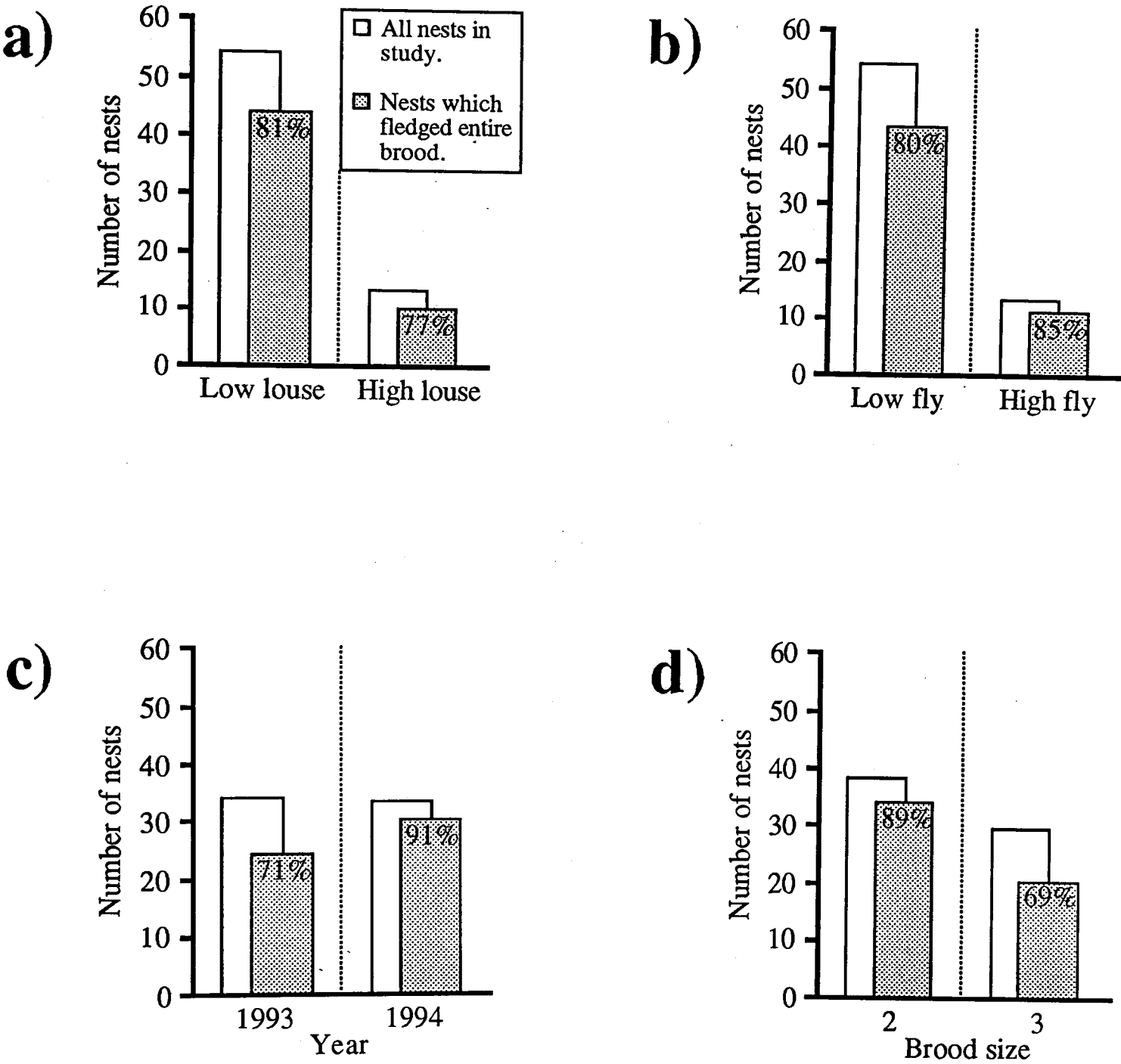


Figure 6.3 Impact of various factors on components on nestling growth. Values are means ($\pm$ 1s.d.) adjusted for all other factors (as fitted to the data by "General Linear Model" analyses). For parasite loads L = low load nests, H = high load nests. * $p < 0.05$, ** $p \leq 0.001$.

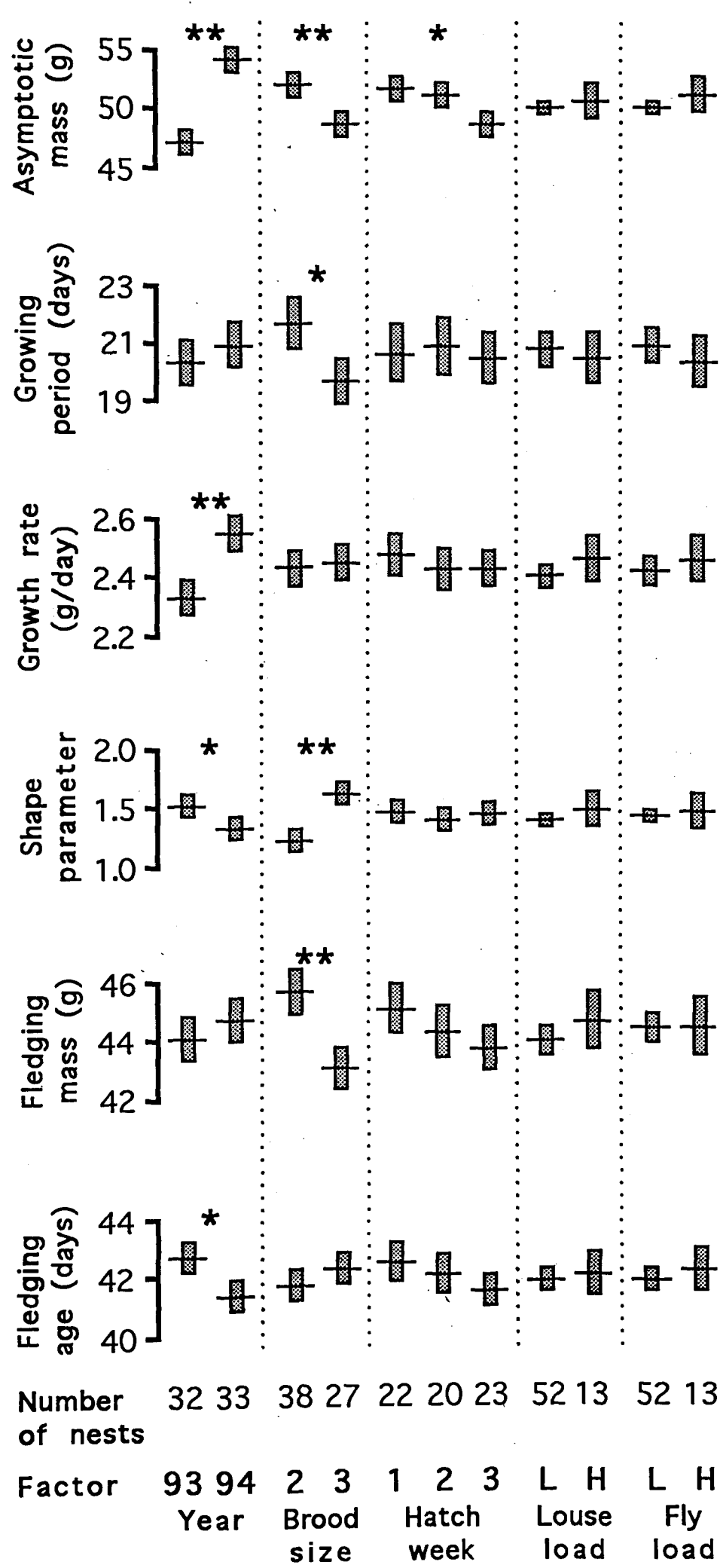
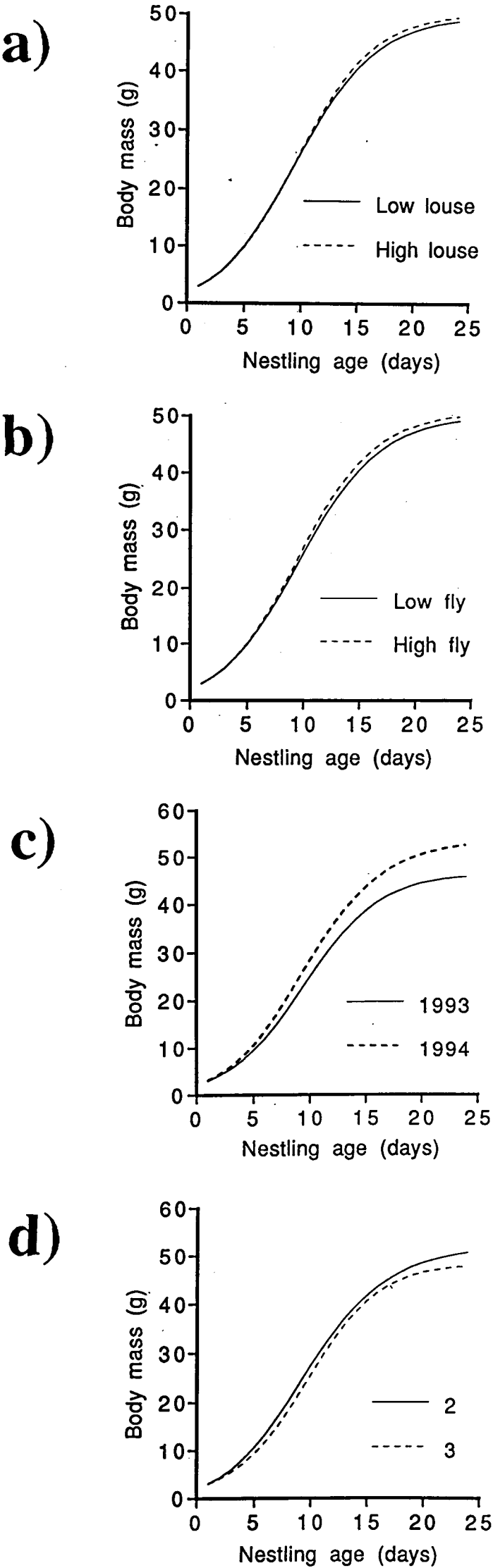


Figure 6.4 Impact on nestling growth (to asymptotic body mass) of (a) louse load, (b) fly load, (c) year, and (d) brood size. Curves were generated by fitting mean parameters for each group of nests back into the modified Richards growth model used.



4.75, $p = 0.03$) than did nestlings in 1994. However, there was no significant difference in growing period ($F = 0.49$, $p = 0.49$) or fledging mass ($F = 0.53$, $p = 0.47$) between the two years.

Nestlings from broods of two had higher asymptotic mass ($F = 16.19$, $p < 0.001$), longer growing periods ($F = 4.78$, $p = 0.03$), lower Richards shape parameters ($F = 16.07$, $p < 0.001$), and higher fledging mass ($F = 11.84$, $p = 0.001$) than nestlings from broods of three (with no difference in growth rate ($F = 0.03$, $p = 0.86$) or fledging age ($F = 0.89$, $p = 0.35$)). There was also a marginally significant year $\times$ brood size interaction with regard to asymptotic mass. The difference between asymptotic mass for nestlings in different brood sizes was greater in 1993 (49.63 ± 1.10 (s.d.) for broods of 2 compared to 43.96 ± 1.17 for broods of 3) than in 1994 (55.01 ± 1.11 for broods of 2 compared to 53.12 ± 1.09 for broods of 3) ($F = 4.07$, $p = 0.05$).

Nestlings hatched in weeks one and two had higher asymptotic mass ($F = 4.43$, $p = 0.02$) than nestlings hatched in week three (Figure 6.3), but there was no difference in growing period ($F = 0.10$, $p = 0.91$), growth rate ($F = 0.43$, $p = 0.65$), Richards shape parameter ($F = 0.15$, $p = 0.86$), fledging mass ($F = 1.25$, $p = 0.30$), or fledging age ($F = 1.10$, $p = 0.34$).

The power of the analyses to detect a 0.75 "effect size" of either parasite on any component of nestling growth was 70% at $\alpha = 0.05$ (two-tailed). If the type I error probability is increased to $\alpha = 0.10$ (which can be done for parasite effects in this study without shifting any result to significance) power increases to 80%, which is the standard level of statistical power recommended for experimental research (Cohen 1988). Thus, the power of the high-load versus low-load comparisons was adequate.

6.7 Discussion

Our manipulations of ectoparasite load were effective. High-load nests had significantly more lice and flies than low-load nests (Figure 6.1). The number of parasites in high-load nests exceeded natural loads by a considerable margin. The median number of lice on 25-27 day old high-louse nestlings was six-fold greater than the natural median observed on same age birds in the tower colony in 1992 (3.0 versus 0.5; Lee, personal communication). The median number of lice on 35-37 day old high-louse nestlings was more than twice the natural median observed on same aged birds in 1992 (12 versus 5). The median number of flies in high-fly nests was seven-fold the natural median observed in the tower colony in 1992 (7 versus 1).

There was no significant effect of either ectoparasite on any component of nestling survival (Figure 6.2) or growth (Figures 6.3 & 6.4), despite the fact that the experimental manipulations of both parasites were of sufficient power to detect even slight effects. This result is striking in light of the fact that 1993 was a very bad year for swift reproduction in Oxford owing to heavy rainfall (see below).

Nestlings in 1993 suffered higher mortality, had lower asymptotic mass, slower growth rates, higher Richards shape parameters, and fledged at a later age than those in 1994 (Figures 6.2, 6.3 & 6.4). Yearly differences in the condition of swift nestlings have been documented previously at the tower colony (1947-56; Lack 1956b), with rainfall being the major causal factor, owing to reduced food abundance in wetter conditions (Koskimies 1950) (greater rainfall during the nestling period led to lower nestling body mass). The proportion of days on which rain fell during the six week period after the first nestlings hatched in 1993 was much greater than in 1994 (28 of 42 days (67%) in 1993, 14 of 42 days (33%) in 1994; $\chi^2 = 9.33$, d.f. = 1, $p = 0.002$). In fact, 1993 was wetter than the worse year recorded (1953) in Lack's (1956b) ten year study, when rain fell on only 27 of the 42 days (64%).

Several non-parasite factors, in addition to year, were also significantly related to components on nestling survival (Figure 6.2) and growth (Figures 6.3 & 6.4).

Nestlings from broods of two suffered lower mortality, had higher asymptotic mass, longer growing periods, lower Richards shape parameters and higher fledging mass than nestlings from broods of three. An effect of brood size on nestling growth and mortality has been documented previously in altricial birds (Klomp 1970), including swifts at the tower colony (Lack & Lack 1951; Lack 1956b). The effect is due to reduced food provisioning per capita in larger broods, even though overall food delivery by adults increases (Martins & Wright 1993a). The effect of brood size on asymptotic mass was greater in 1993 than in 1994, presumably because adults were less able to provision larger broods in the poorer weather (Martins & Wright 1993b,c).

Time of hatching within a season also had a significant effect on one component of swift reproduction. Earlier hatched nestlings had significantly higher asymptotic mass than later hatched nestlings (Figure 6.3). Effects of later hatching on swift nestlings have been documented previously at the tower colony (Lack & Lack 1951) and may be due to decreased food abundance later in the season (Koskimies 1950). The abundance of aerial insects in Southern England has been shown to decrease from July through August (Bryant 1975).

A recently proposed alternative hypothesis for the lack of detectable effects of parasites on nestlings is that adults compensate young with high parasite loads through increased provisioning of food (Johnson & Albrecht 1993; Møller 1994). Adults will be better able to compensate in "good" years (warm and dry) than in "bad" years (cold and wet) (de Lope *et al.* 1993). In our study, 1993 was an extremely bad year in which adults would have had difficulty compensating for any effects of parasites. The fact that we detected no effect or trend of an effect of parasites on any component of nestling survival or growth strongly suggests that parental compensation is not the explanation for the avirulence of swift lice and louse flies. The lack of any interaction

between parasite loads and year in their effects on nestling growth reinforces this point.

It is clear from our experiments that swift lice and louse flies are avirulent. Even when boosted to unnaturally high loads no effect of either parasite could be detected. The results of our study are consistent with the theoretical prediction that vertically transmitted ectoparasites evolve to become relatively avirulent since they depend on successful host reproduction for direct transmission to host offspring (Clayton & Tompkins 1994). Our conclusion could be strengthened by comparing the effects of the vertically transmitted parasites in this study to the effects of horizontally transmitted ectoparasites, such as Dermanyssid mites (Clayton & Tompkins 1994, 1995), on the same species of host. Unfortunately, the birds in our study colony were not host to these or other horizontally transmitted ectoparasites.

Chapter 7 Impact of nest-harvesting on swiftlet reproduction

[Tompkins, D.M. In press. Impact of nest-harvesting on swiftlet reproduction. *Journal of Wildlife Management and Research, Sabah.*]

7.1 Summary

1. Certain species of Malaysian swiftlets (Aves: Apodidae) produce the "edible birds' nests" prized in Chinese medicine and cuisine. Population sizes of these birds are known to be declining at many locations in SE Asia. One possible reason is decreased reproductive success due to over-harvesting of nests.
2. At Gomantong Caves (Sabah) nest-harvesting is controlled by the Sabah Wildlife Department. Nests are taken only at the beginning and end of the birds breeding season (February - September); the birds have a 4 month undisturbed period in which to breed.
3. The impact, on swiftlet reproductive success, of the harvesting regime currently used at Gomantong Caves was compared to a regime where nests were harvested only at the end of the breeding season. This was carried out using the glossy swiftlet (*Collocalia esculenta*) as a model species.
4. Number of clutches laid, number of eggs laid, and number of nestlings fledged were greater for nests harvested only at the end of the breeding season. However, there was no effect of harvesting treatment on any component of nestling growth.
5. The reproductive rate of swiftlets at nests harvested at both the beginning and end of the breeding season was theoretically insufficient to maintain population sizes at Gomantong Caves, whereas the reproductive rate of swiftlets at nests harvested only at the end of the breeding season was sufficient.

Keywords: Apodidae, *Collocalia esculenta*, edible-nest, glossy swiftlet.

7.2 Introduction

Certain species of Malaysian swiftlet (Aves: Apodidae) produce the "edible birds' nests" prized in Chinese medicine and cuisine (Medway 1969). These are the white-nest swiftlet (*Aerodramus fuciphagus*) and the black-nest swiftlet (*Aerodramus maximus*), whose nests are formed from hardened salival secretions (Wang 1921; Marshall & Folley 1956; Medway 1962a). With edible birds nests fetching high prices (1kg of the highest quality nests can retail at up to M\$4,000), the pressure to attain the maximum possible yearly harvest from the various colonies of these birds is great. However, over-harvesting of nests can affect swiftlet reproduction (Kang *et al.* 1991) resulting in decreasing population sizes (Francis 1987a). In extreme cases nests are always removed immediately after being built, and the adult birds are never allowed to breed (Sims 1959). A recent report funded by the WWF on the "International Trade In Swiftlet Nests" noted how, over the South-East Asian range of these birds, there is already evidence of over-exploitation, population decline and even local extinction (Lau & Melville 1994).

The major site for the birds nest industry in Sabah (Malaysia) is Gomantong Caves (5°31'N, 118°04'E), home to 1.5 million swiftlets. Here approximately 5,000kg of nests are harvested yearly, although yield data show an approximately 50% fall in size of harvest over the last 50 years (Francis 1987a). This may reflect a decrease in swiftlet population size. As a control measure the Sabah Wildlife Department limit the collection of nests at Gomantong Caves. Only two harvests are allowed during the birds breeding season (February - September), with a few nests also being taken outside the breeding season (Francis 1987a). The first harvest occurs during April; nests are taken immediately after being built before more than 10% contain eggs. The second occurs during August after nestlings have fledged from 90% of nests. This regime allows the birds to rear one clutch of young - one complete breeding cycle of nest building, egg laying and incubation, and nestling growth to fledging takes

approximately 4 months. This harvesting regime is similar to that recommended by Kang *et al.* (1991). They suggested that commercially exploited swiftlet colonies be left unharvested during the middle part of their breeding season to allow a period of successful reproduction (approximately 135 days).

I compared the impact, on swiftlet reproduction, of the harvesting regime currently in use at Gomantong Caves to one where nests are removed in August only (at the end of the breeding season). I also examined the effect of brood size on nestling growth and fledging success. Brood size is known to influence these parameters in many altricial birds (Klomp 1970), including glossy swiftlets (Bryant & Hails 1983). The experiment was carried out on a non edible-nest swiftlet, the glossy swiftlet (*Collocalia esculenta*). The life history of this bird is similar to that of the edible-nest species (Francis 1987a,b), making it an ideal model species for studying the impact of nest-harvesting on swiftlet reproduction.

7.3 Study site and species

The study was carried out, April - August 1994, in a rural area 16km west of the town of Sandakan, Sabah (5°52'N, 117°59'E), approximately 30km north of Gomantong Caves. The swiftlet colony was located underneath a house raised about 3m off the ground on stilts. Wooden support beams ran the length of the house, to which swiftlet nests were attached (see Francis 1987b). There was considerable human activity around the colony which was, most likely, a major source of disturbance to the breeding birds.

The glossy swiftlet is a small (ca 7.5g) aerial insectivore (Hails & Amirrudin 1981) which nests in colonies in caves, rock shelters, man-made culverts and buildings in South-East Asia and the South-West Pacific (Chantler & Driessens 1995). Glossy swiftlets at Sandakan share the same February - September breeding season as the

edible-nest swiftlets in Gomantong Caves (Francis 1987a,b). Normal clutch size is two eggs, laid a mean of 3.4 days apart (Francis 1987b), although clutches of one are common. Mean incubation period is 22 days, with the first egg hatching a mean of 2.1 days before the second. Glossy swiftlets fledge at approximately 36 days post-hatching (Bryant & Hails 1983). Breeding adult swiftlets are nest-site specific, rebuilding nests on the same exact sites both within and between seasons (Francis 1987a; Kang *et al.* 1991).

7.4 Experimental protocol

In preparation for the harvesting experiment, all glossy swiftlet nests at the house site were removed in December 1993 by the owner. In April 1994 experimental treatments were haphazardly assigned to 36 nests within the same 2m colony section; 18 nests were assigned to be "harvested" on both April 15th and August 15th, 18 were assigned to be harvested only on August 15th. These dates coincided with the harvesting of edible-nests from swiftlets at Gomantong Caves in 1994.

Nests were monitored every 2 days; contents were noted and any nestlings present weighed using a 10g pesola spring balance. Siblings were identified by the clipping of one toe-nail on either foot. At 30 days post-hatching, all nestlings were ringed with numbered aluminium wrap-around bands. By this method, all nests and nestlings were treated equally. This is desirable as human disturbance causes "egg-dumping" in the glossy swiftlet, and the handling of nestlings has an adverse effect on nestling growth (Francis 1987b).

Number of clutches laid, number of eggs laid, laying interval, hatching success, hatching interval, and fledging success per nest were compared using non-parametric analyses. Nestlings that disappeared prior to 35 days post-hatching were presumed to have fallen out of nests. A modified Richards sigmoid growth model (Brisbin *et al.*

1986) was fitted to the body mass measurements of each nestling. Nestling body mass is known to be influenced by environmental factors (Bryant 1978), and is correlated with post-fledging size and survival (Boag 1987; Magrath 1991). Three parameters for each growth curve were predicted: asymptotic mass, growing period (hatching through age at asymptote) and the Richards shape parameter (m). Near $m = 2.0$, 0.67 , 0.0 or when $m \rightarrow 1.0$, the Richards model becomes the logistic, von Bertalanffy, monomolecular or Gompertz models respectively (Richards 1959). A fourth parameter, mean growth rate, was calculated by dividing asymptotic mass by growing period. Measurements of fledging mass and age were taken from the raw data. Mean parameter values per nest were analysed using "General Linear Models", incorporating harvesting regime and brood size as separate factors. All non-significant interaction factors were discarded. Only nestlings that survived to fledge were included in the analyses.

7.5 Results

EFFECT OF TREATMENT. By the April 15th "nest-harvest" all 36 nests involved in the study were fully formed. There was no difference in the number of eggs per nest, on April 15th, for nests to be harvested (mean ($\pm$ s.d.) of 1.22 ± 0.89) versus those to be left unharvested (1.17 ± 0.81 ; Mann-Whitney $U = 153.5$, $p = 0.79$). All 18 harvested nests were subsequently rebuilt. The date of the first egg hatching was significantly later for the 18 harvested nests than the 18 unharvested nests (Figure 7.1; $U = 13$, $p < 0.001$).

IMPACT ON REPRODUCTION. Up until August 15th, when all nests in the study were harvested, reproduction in nests left unharvested on April 15th was more successful than that in those which were harvested (Figure 7.2). Adults at unharvested nests laid significantly more clutches than those at harvested nests (mean of 1.50 ± 0.71 versus 1.11 ± 0.47 clutches per nest; $U = 101.5$, $p = 0.03$), laying 50%

Figure 7.1 Date of first egg hatching for nests harvested on April 15th (n = 17), versus those left unharvested (n = 16).

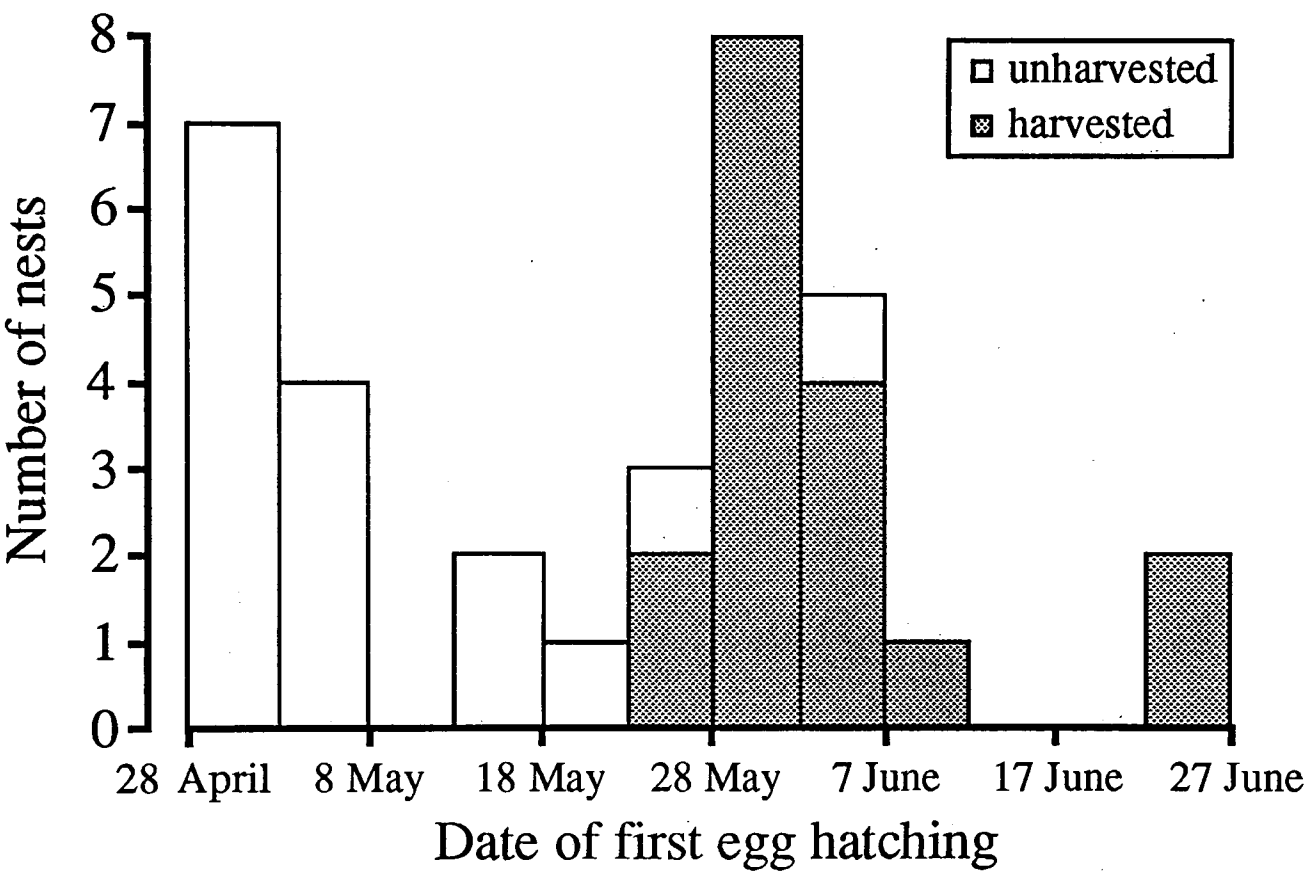
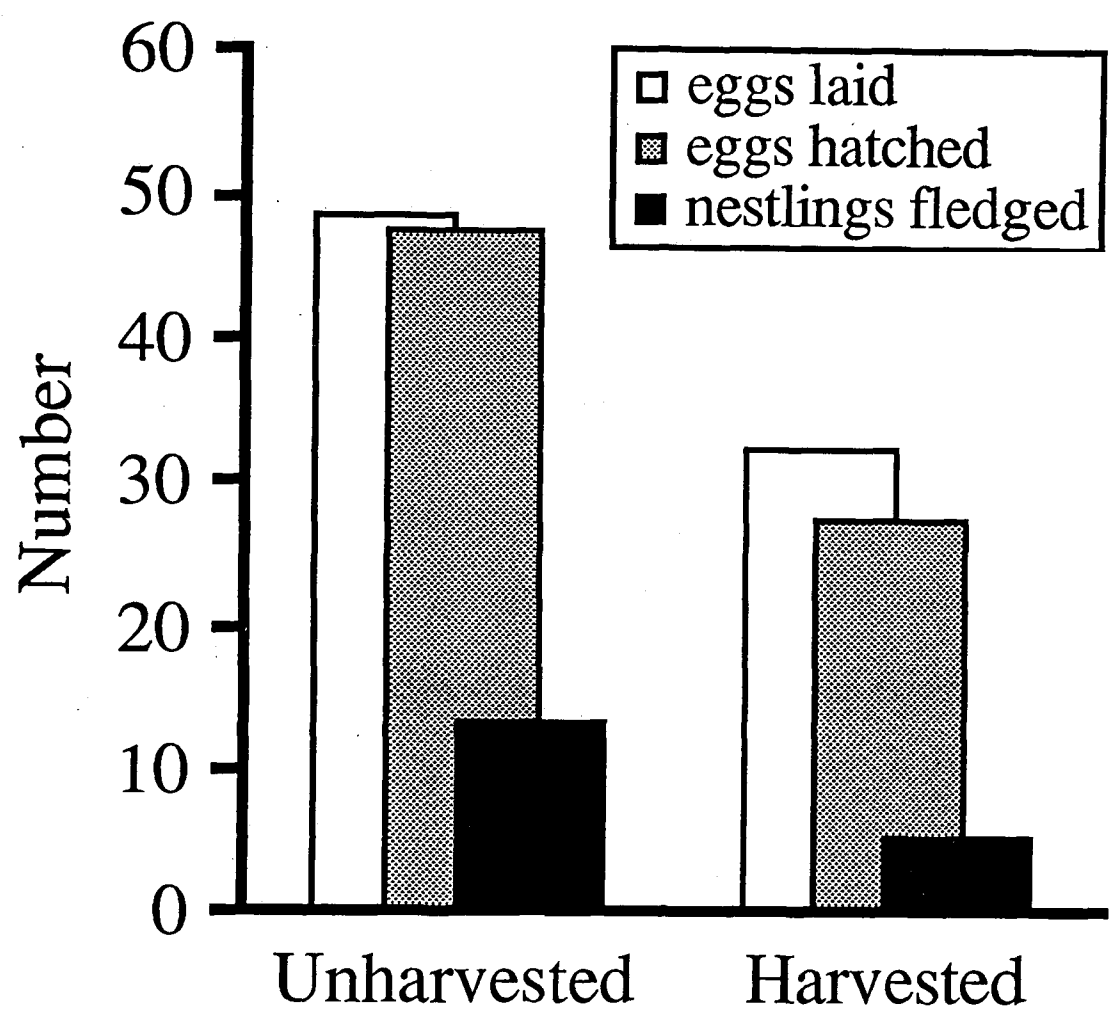


Figure 7.2 Eggs laid, eggs hatched, and nestlings fledged in nests harvested on April 15th (n = 18), versus those left unharvested (n = 18).



more eggs per nest ($U = 95.5$, $p = 0.03$). The laying interval between first and second eggs was shorter at unharvested than at harvested nests (mean of 3.29 ± 0.91 versus 4.18 ± 0.98 days per nest; $U = 46$, $p = 0.05$). Hatching success of eggs was (non-significantly) greater in unharvested than in harvested eggs. Fifteen of 16 (94%) unharvested nests hatched all eggs, compared to 12 of 17 (71%) harvested nests (Fisher exact $p = 0.10$). Laying interval and hatching success were negatively correlated. Nests where all eggs hatched had a mean laying interval of 3.37 ± 0.83 days versus 4.67 ± 1.03 days in nests where not all eggs hatched ($U = 22$, $p = 0.01$). Subsequently, brood sizes were (non-significantly) greater in unharvested than in harvested nests (1.72 ± 0.36 versus 1.41 ± 0.48 nestlings per nest; $U = 87$, $p = 0.06$). The hatching interval between first and second eggs was similar under both harvesting regimes (2.57 ± 0.83 versus 2.56 ± 0.73 in unharvested versus harvested nests; $U = 62.5$, $p = 0.97$). Fledging success was also similar under both harvesting regimes. Only one of 18 (6%) unharvested nests fledged all young, compared to one of 17 (6%) harvested nests (Fisher exact $p = 0.74$). However, overall, more nestlings fledged from unharvested than from harvested nests (0.72 ± 0.75 versus 0.28 ± 0.46 nestlings per nest; $U = 105.5$, $p = 0.04$).

Fledging success was greater for broods of one than for broods of two. All nestlings fledged from seven of 16 (44%) broods of one, compared to only one of 29 (3%) broods of two (Fisher exact $p = 0.002$). No nest fledged young from more than one brood. Overall, similar numbers of nestlings fledged from nests with broods of one and two (0.44 ± 0.34 versus 0.38 ± 0.30 nestlings per nest respectively; $U = 214$, $p = 0.67$).

Nestling growth was similar under both harvesting regimes (Figures 7.3 & 7.4a). There was no significant difference in asymptotic mass ($F = 0.04$, $p = 0.84$), growing period ($F = 0.41$, $p = 0.53$), growth rate ($F = 0.25$, $p = 0.62$), Richards shape parameter ($F = 0.00$, $p = 0.97$), fledging mass ($F = 0.29$, $p = 0.60$), or age at fledging ($F = 0.45$, $p = 0.50$).

Figure 7.3 Impact of harvesting regime and brood size on components on nestling growth. Values are means ($\pm$ 1s.d.) adjusted for all other factors (as fitted to the data by "General Linear Model" analyses). * $p < 0.05$, ** $p < 0.01$.

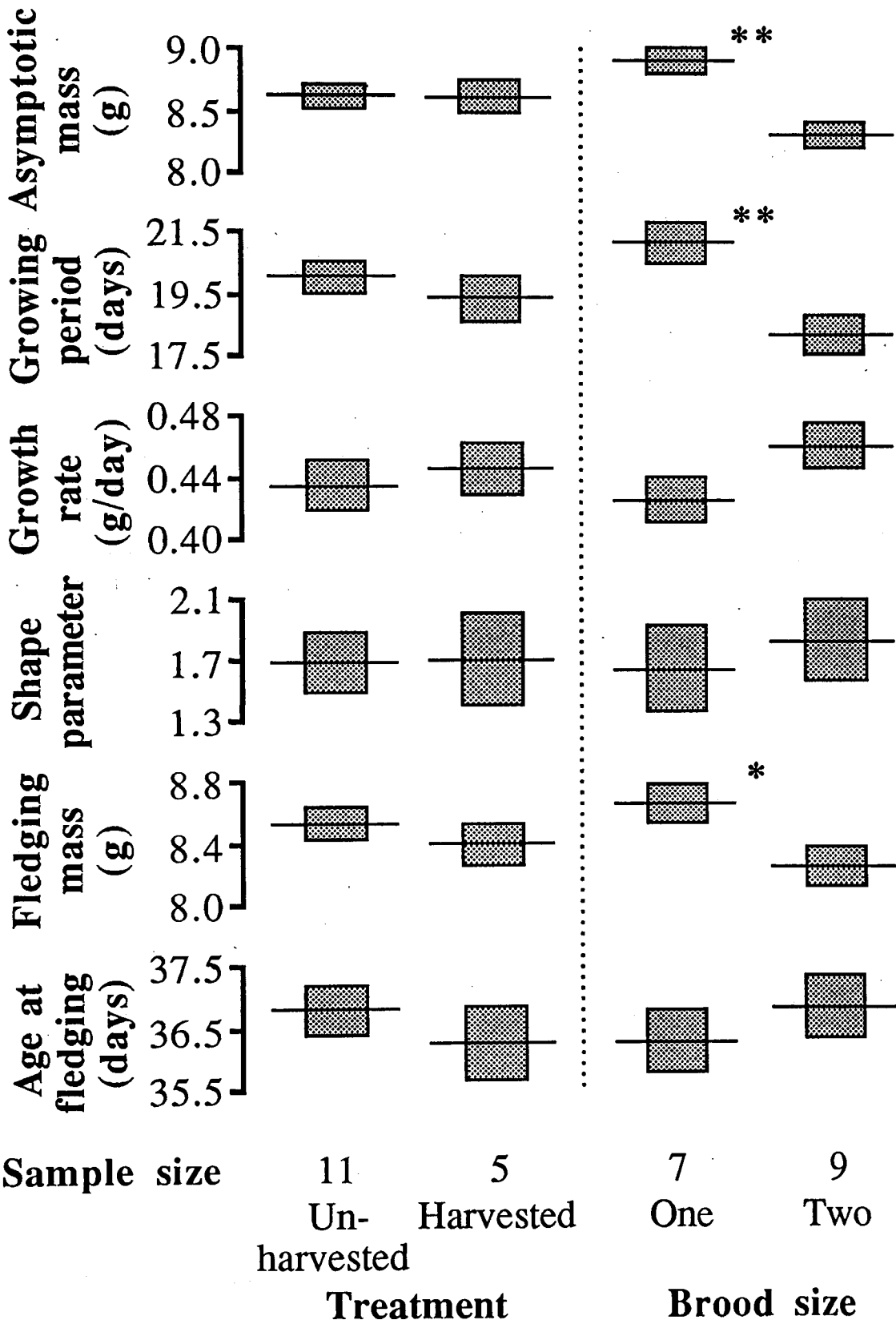
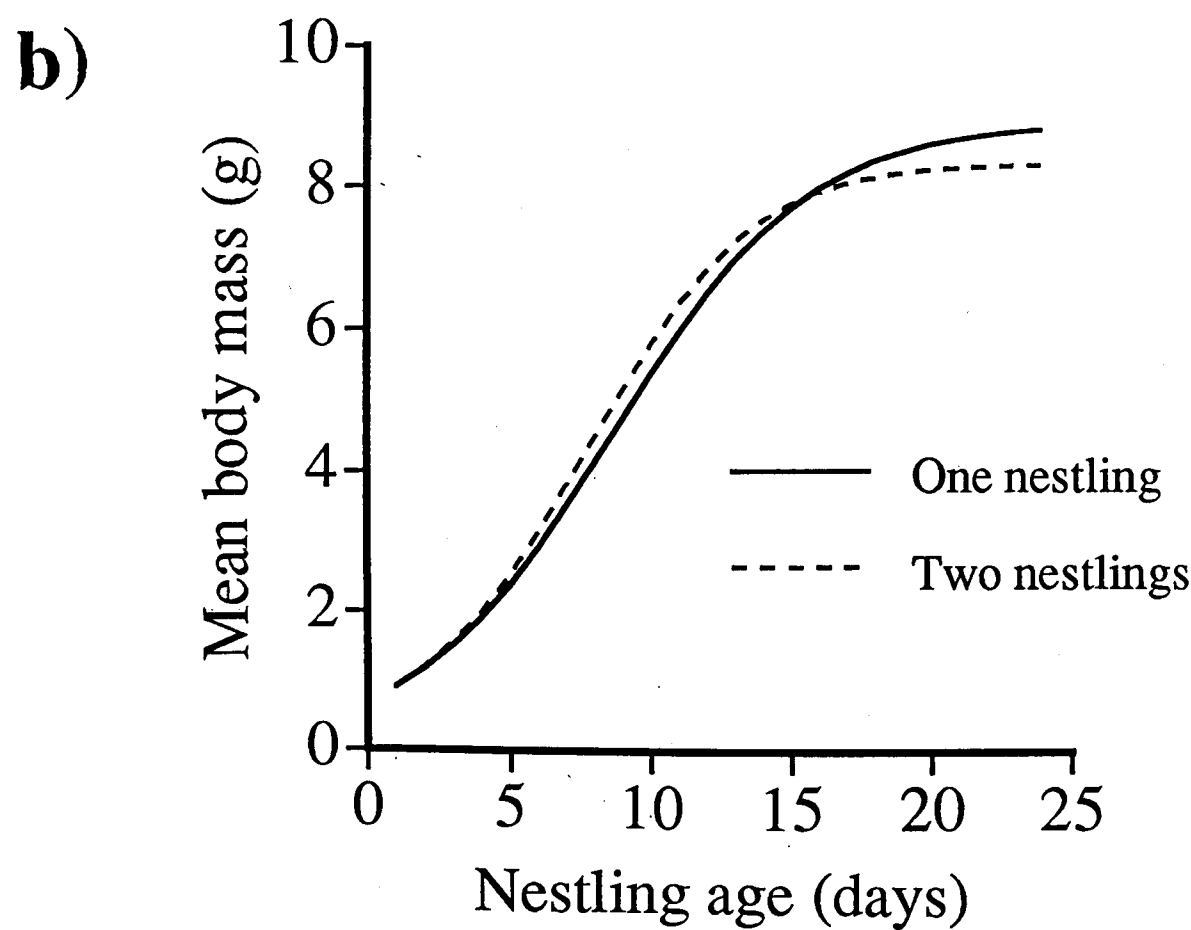
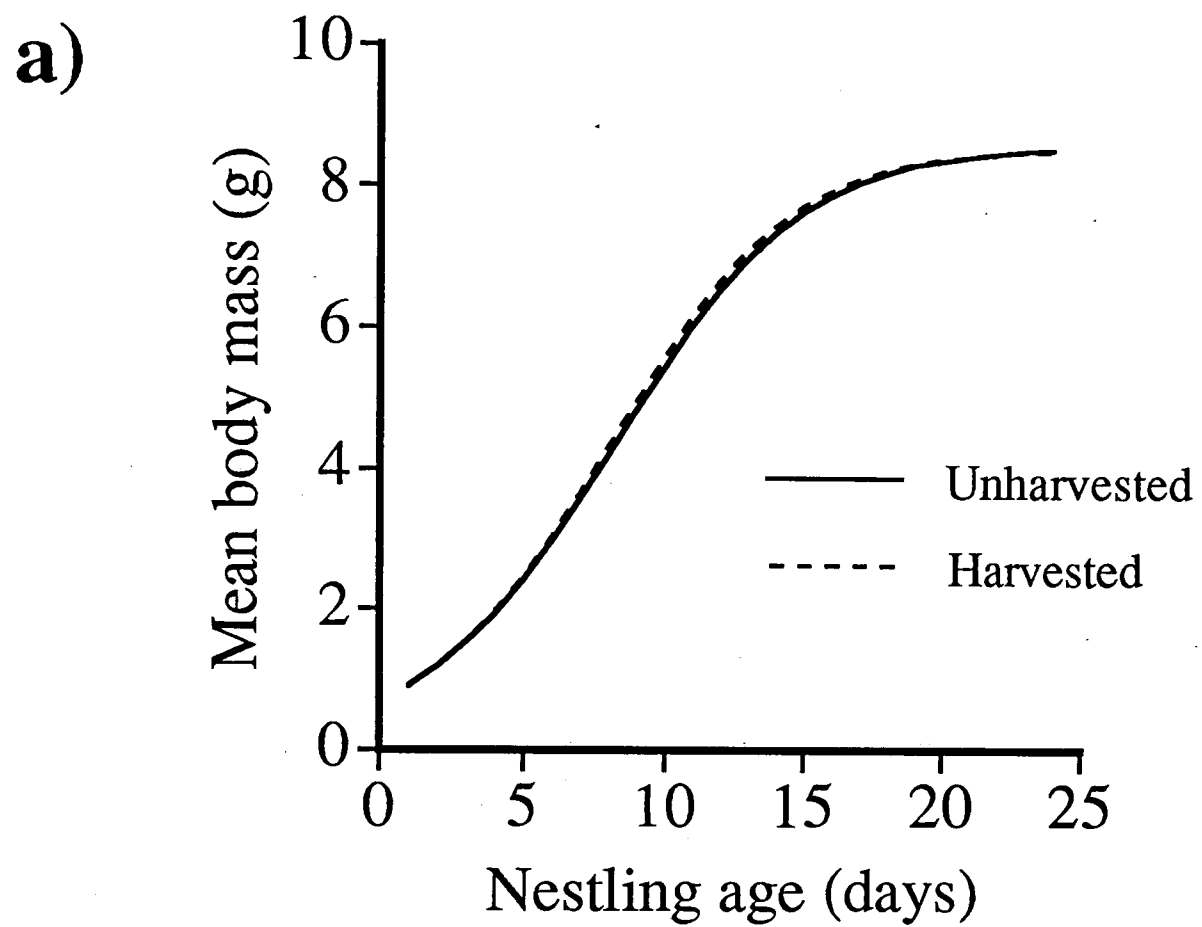


Figure 7.4 Impact on nestling growth of (a) harvesting regime, and (b) brood size.

Curves were generated by fitting mean parameters for each group of nests back into the modified Richards growth model used.



= 0.51). However, brood size did have a significant effect on nestling growth (Figures 7.3 & 7.4b). Nestlings in broods of one had higher asymptotic mass ($F = 14.22$, $p = 0.002$), longer growing periods ($F = 11.24$; $p = 0.005$), and higher fledging mass ($F = 5.31$, $p = 0.04$) than nestlings in broods of two (although there were no differences in growth rate ($F = 3.35$, $p = 0.09$), Richards shape parameter ($F = 0.29$, $p = 0.60$), or age at fledging ($F = 0.52$, $p = 0.48$)).

7.6 Discussion

Glossy swiftlets at nests harvested under the current harvesting regime used in Gomantong Caves (at the beginning and end of the breeding season) had significantly lower reproductive success than those harvested only at the end of the breeding season. Date of first egg hatching was significantly later for nests harvested on April 15th than those not (Figure 7.1). This resulted in swiftlets at unharvested nests being able to lay significantly more clutches (and thus significantly more eggs) than those at harvested nests (Figure 7.2). Over 2.5 times more young fledged from unharvested nests than from harvested nests. This was most likely due to many more nests in the harvested group having insufficient time to relay clutches (and rear nestlings to fledging before the second harvest) after total brood failure. There was no difference in any component of nestling growth for nestlings in harvested versus unharvested nests (Figures 7.3 & 7.4a).

Nestlings in broods of one had higher asymptotic mass, longer growing periods, higher fledging mass, and greater fledging success than nestlings in broods of two (Figures 7.3 & 7.4b). An effect of brood size on nestling growth and mortality has been documented previously in the Apodidae (swifts) (Lack & Lack 1951; Lack 1956b). The effect is due to reduced food provisioning per capita in larger broods, even though overall food delivery by adults increases (Martins & Wright 1993a). Since similar

numbers of nestlings fledged from nests with broods of one and two, and nestlings from broods of one most likely had higher post-fledging survival than nestlings from broods of two (due to higher fledging mass), brood reduction increased the reproductive success of glossy swiftlets in this study. Lack (1947) proposed that, if the food supply for the young is unpredictable at egg-laying, female birds will lay a number of eggs equal to the number of young that could be raised if breeding conditions were good, but will reduce the brood size if conditions are poor by selectively starving the last young to hatch (O'Connor 1978). The high level of human activity at the house colony may have disturbed the feeding of nestlings by adult swiftlets.

In their harvesting experiment on edible-nest swiftlets, Kang *et al.* (1991) reported a decrease in re-nesting success, and longer laying intervals between first and second eggs in nests previously harvested. In this study re-nesting success was not compromised, but a longer laying interval between first and second eggs in nests previously harvested was observed. Kang *et al.* believed this phenomena to be due to a shortage of energy or depletion of stored lipids in adults who have had to rebuild nests. Hails & Turner (1985) showed that breeding glossy swiftlet adults may be foraging near their maximum limit, and nutrients may be in short supply during egg laying. This shortage would be exacerbated by nest-harvesting, forcing birds to invest more energy and nutrients into nest building, and may lead to the increased laying interval seen. The effect would be worse if, as in this study, eggs are also lost at harvesting. The correlation between laying interval and hatching success seen indicates that increased laying intervals, caused by nest harvesting, decrease hatching success of eggs. This may have been due to breeding adults ejecting second eggs, in clutches of two, which failed to hatch soon after first eggs hatched (Francis 1978b). Thus, nest-harvesting most likely had the following effect on glossy swiftlet reproduction: breeding adults were placed under higher energetic stress, decreasing the

hatching success of eggs, thus increasing the probability of total brood failure in circumstances where there was insufficient time for a second breeding attempt.

The impact of nest-harvesting may be reducing the recruitment rate of swiftlets at Gomantong Caves below that which is necessary to maintain the population. Using approximate swiftlet mortality data (adult yearly mortality rate of 10%, juvenile yearly mortality rate of 50%), Francis (1987a) estimated that 40% of swiftlet nests must fledge at least one young each year if a population is to be maintained. In this study swiftlet nests, which were harvested at both beginning and end of the breeding season (the current harvesting regime at Gomantong Caves), fledged a mean of 0.28 young per nest. This is equivalent to only 28% of nests fledging one young per year. Thus, if the above estimates of swiftlet mortality are reasonably correct, and the glossy swiftlet is an accurate model for edible-nest swiftlets, then swiftlet reproduction under this harvesting regime would be insufficient to maintain population sizes in Gomantong Caves. However, nests which were harvested only at the end of the breeding season fledged a mean of 0.72 young per nest. This is equivalent to 72% of nests fledging one young per year, and would (theoretically) be sufficient to maintain population sizes in Gomantong Caves.

The lack of time for re-laying of clutches (and rearing of nestlings to fledging) after total brood failure may be worse for harvested edible-nest swiftlets (especially black-nest swiftlets) than for the glossy swiftlets in this study. The breeding cycles for the white- and black-nest swiftlet are approximately 2 and 4 weeks longer than for the glossy swiftlet. Edible-nest swiftlets would have less time remaining in the breeding season, after harvesting of nests and total brood failure of the first clutch, for successful laying and rearing of subsequent clutches. This would reduce the reproductive success at nests, harvested at the beginning of the breeding season, below that seen in this study.

This study recommends that the current harvesting regime of edible-nests at Gomantong Caves be modified to allow the swiftlets a longer undisturbed window for successful reproduction during the breeding season. This would not be accomplished by delaying the August harvest (at the end of the breeding season) as reproduction is naturally declining by this time (personal observation). Rather, the April nest-harvest (at the beginning of the breeding season) should be brought forward at least to a point before any nests contain eggs. This would allow the swiftlets more time during the breeding season to re-lay clutches when total brood failure occurred. This would also reduce the energetic impact of nest-harvesting on the breeding adults, as less nest material and no eggs would be lost during the early harvest, reducing the occurrence of total brood failure of first clutches. This recommended regime would reduce the yield of edible-nests at the early harvest by a small fraction, but may enable the breeding swiftlets to fledge sufficient young each year to maintain population sizes at Gomantong Caves. However, for more accurate information concerning this problem, a larger scale harvesting experiment needs to be carried out at Gomantong Caves, on the edible-nest swiftlets themselves, to observe their reproductive success under a range of harvesting regimes.

Chapter 8 Influence of nest harvesting on the black-nest swiftlet

(*Aerodramus maximus*) at Gomantong Caves, Sabah

8.1 Introduction

Certain species of Malaysian swiftlet produce the "edible birds' nest" prized in Chinese medicine and cuisine. In Chapter 7, a non-edible nest swiftlet (*Collocalia esculenta*, the glossy swiftlet) was used as a model species to investigate the impact of nest-harvesting on swiftlet reproduction. The major site for the birds nest industry in Sabah (Malaysia) is Gomantong Caves. The impact of the nest-harvesting regime currently in force at Gomantong Caves (harvesting at both the beginning and end of the birds breeding season) was compared to a regime of one harvest (at the end of the breeding season) only. It was concluded that, although an impact of nest-harvesting on swiftlet reproduction was apparent, a study of actual 'edible-nest' swiftlet species was required to determine the applicability of this result (see Chapter 7 for further details).

The goal of this study was to investigate the influence of nest-harvesting on the black-nest swiftlet (*Aerodramus maximus*) at Gomantong Caves. The nest of the black-nest swiftlet is constructed from hardened salival secretions and body feathers (Wang 1921; Marshall & Folley 1956; Medway 1962a). At Gomantong Caves, the nests of black-nest swiftlets are harvested twice during their breeding season (February-September). The first harvest occurs during April; nests are taken immediately after being built before more than 10% contain eggs. The second occurs during August after nestlings have fledged from 90% of nests. Ostensibly, this regime allows the birds to rear one clutch of young - one complete breeding cycle of nest building, egg laying and incubation, and nestling growth to fledging takes approximately four months. Cleaned black-nests retail at up to M\$1,000 per kg (Francis 1987a).

8.2 Study site and species

The study was carried out, April - August 1995, at Gomantong Caves, Sabah (5°31'N, 118°04'E), a large limestone cave complex home to approximately 1.5 million swiftlets (see Chapter 7 for further details).

The black-nest swiftlet is the largest (ca 17.5g) of the Malaysian swiftlets (Chantler & Driessens 1995). It is a true "cave swiftlet" in that it nests in the total darkness of cave interiors, using echolocation to navigate (see Table 1.1). At Gomantong Caves, the black-nest swiftlet nests high on the cave walls and ceiling (Francis 1987a). Nest building takes 3-4 weeks. Clutch size is one egg only. Mean incubation period is 28 days. Nestlings fledge at approximately 59 days of age (Medway 1962b). Breeding adult black-nest swiftlets are nest-site specific, rebuilding nests on the same exact sites both within and between seasons (Francis 1987a; Kang *et al.* 1991).

8.3 Experimental protocol

Reproduction at two mono-specific nest clusters of the black-nest swiftlet was observed under different harvesting regimes. Both clusters contained approximately 200 nests each, at similar densities (personal observation). One cluster was harvested under the regime currently in use at Gomantong Caves (harvested on April 7th and August 13th in 1995) whilst the second cluster was left undisturbed for the entire breeding season.

Due to the inaccessibility of black-nest swiftlet clusters, it was only possible to observe the breeding activity in nests using 10x40 binoculars and strong torch-light. The contents of 50 nests in each cluster were monitored over the breeding season. Nests were first observed at 2.00pm on March 25th. Positions of nests to be monitored were noted in sketch maps. Nests were subsequently monitored every 14 days (at

2.00pm), and nest contents noted as either empty, egg, naked nestling ($\cong$ 0-2 weeks old), nestling with emerging feathers ($\cong$ 2-4 weeks old), or fully feathered nestling ($\cong$ 4+ weeks old). Age categories were determined by reference to a subset of precisely aged young. If nestlings reached the fully feathered stage they were assumed to have fledged when they disappeared from the nest.

8.4 Results

On April 7th, prior to the first harvest, nests to be harvested and those to be left unharvested contained similar numbers of eggs. Twelve (24%) of the 50 "harvested" nests contained eggs, compared to 14 (28%) of the 50 "unharvested" nests ($\chi^2 = 0.21$, d.f. = 1, $p = 0.65$). After harvesting, all 50 harvested nests were rebuilt at the exact same locations.

The harvesting of nests had significant effects on black-nest swiftlet reproduction (Figure 8.1; Table 8.1). Fewer harvested nests (subsequently rebuilt) showed any sign of breeding activity, compared to those left unharvested. Of the re-built harvested nests, 24% were subsequently left empty (no egg laid), compared to only 8% of unharvested nests ($\chi^2 = 4.76$, d.f. = 1, $p = 0.03$). Laying date for breeding adults at harvested nests (after the April 7th harvest) was approximately four weeks later than the mean at unharvested nests (Mann-Whitney $U = 25$, $p = 0.001$).

The incubation period was longer at harvested, than at unharvested, nests ($U = 759$, $p = 0.001$). Also, nestlings in harvested nests fledged at a later age than those in unharvested nests (although the difference was non-significant; $U = 711$, $p = 0.06$). Lack of significance was due to nestlings in both harvested and unharvested nests spending equal lengths of time as naked nestlings ($U = 863$, $p = 0.90$), and as nestlings with emerging feathers ($U = 804$, $p = 0.26$). However, fully feathered nestlings did spend significantly longer in harvested, than in unharvested nests ($U = 636$, $p = 0.005$).

Figure 8.1 Reproductive activity at black-nest swiftlet nests either a) harvested on April 7th (n = 50), or (b) left unharvested (n = 50).

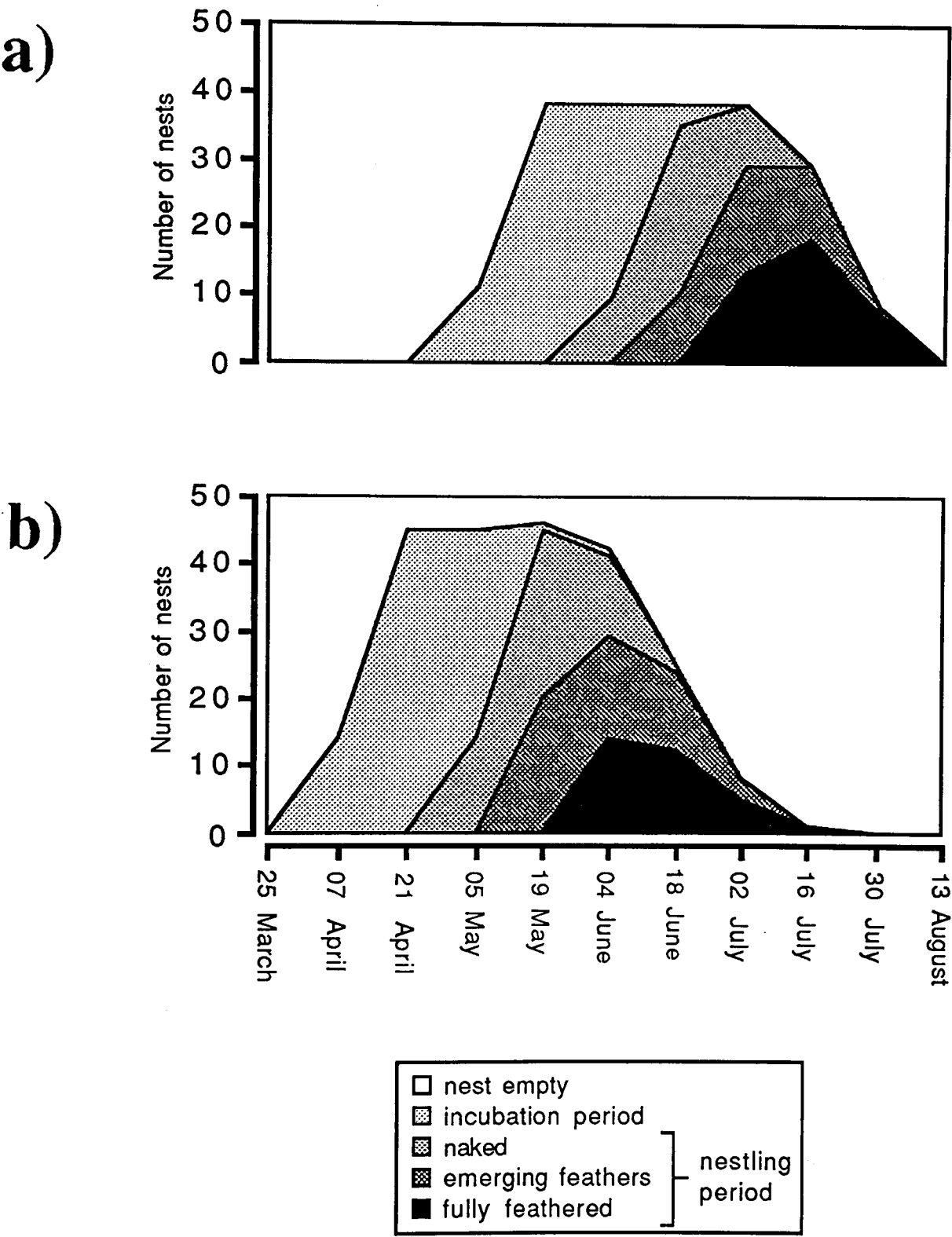


Table 8.1 Components of reproductive success for black-nest swiftlets at nests either harvested on April 7th, or left unharvested. Mean values are accompanied by standard deviations.

	Harvested (n = 50 nests)	Unharvested (n = 50 nests)		
Number of active nests	38	46	$\chi^2 = 4.76$	$p = 0.03$
Mean laying day	65.9 ± 6.4	38.3 ± 8.0	U = 25	$p = 0.001$
Incubation period (days)	29.8 ± 4.8	28.0 ± 0.0	U = 759	$p = 0.01$
Nestling period (days)				
- naked	15.8 ± 5.8	15.8 ± 8.7	U = 863	p = 0.90
- emerging feathers	14.0 ± 5.6	15.2 ± 4.0	U = 804	p = 0.26
- fully feathered	14.0 ± 6.5	9.7 ± 6.5	U = 636	$p = 0.005$
- total	43.8 ± 4.8	40.8 ± 8.8	U = 711	p = 0.06
Mean number fledged	0.76 ± 0.06	0.92 ± 0.04	U = 1050	$p = 0.03$

Overall, the number of nestlings fledged from harvested nests was lower than the number fledged from unharvested nests ($U = 1050$, $p = 0.03$). All eggs laid at black-nests in either the harvested or unharvested nests (after April 7th for the harvested nests) hatched. All nestlings survived to fledging.

8.5 Discussion

Nest-harvesting had a significant influence on the reproduction of black-nest swiftlets at Gomantong Caves (Figure 8.1; Table 8.1). All nests harvested at the beginning of the breeding season were subsequently rebuilt. However, three times more harvested, than unharvested, nests showed no subsequent sign of breeding activity (no egg laid). The harvest delayed the subsequent laying of eggs by approximately four weeks, compared to unharvested controls, although hatching success was similar in both groups.

Fully feathered nestlings spent longer in harvested nests (prior to fledging) than they did in unharvested controls, although nestlings fledged with similar success from both groups. Overall, the mean number of nestlings fledged from harvested nests was 17% lower than the mean number fledged from unharvested controls. This was solely due to the increased number of inactive nests after harvesting.

Previous studies have indicated two ways in which nest-harvesting depresses swiftlet reproduction. First, by forcing swiftlets to build two nests at the beginning of the breeding season (because the first is removed), nest-harvesting places an energetic strain on breeding adults which is greater if, as in this study, eggs are also lost at harvesting. This energetic strain may lead to a decrease in re-nesting success (Kang *et al.* 1991) and longer laying intervals between first and second eggs (Kang *et al.* 1991; Chapter 7). Second, by reducing the length of breeding season available, harvested swiftlets may have insufficient time to relay clutches (and rear nestlings to fledging)

after total brood failure (Chapter 7). Nest-harvesting at Gomantong Caves effectively reduces the breeding season for black-nest swiftlets from eight months (February - September) to four months (April - July). However, in the nest-harvesting experiment carried out here, no brood failure occurred and no swiftlets attempted to breed more than once. In all nests where an egg was laid (after the first harvest for the experimental group), that egg hatched successfully, and all nestlings fledged. Thus, four months per breeding season is sufficient time for black-nest swiftlet reproduction at Gomantong Caves. The high levels of hatching and fledging success may be due to the single egg clutch, resulting in a low probability of eggs and nestlings falling out of nests (which is common in swiftlet species which lay a clutch of two eggs; personal observation), and no brood reduction by adult swiftlets (see Chapter 7). Also, when not being harvested, there is no human disturbance to black-nest swiftlets nesting high on the walls and ceiling at Gomantong Caves, unlike the situation for glossy swiftlets in Chapter 7.

An energetic cost of nest-harvesting was apparent in this study. Although there was no decrease in re-nesting success following harvesting, fewer nests were reproductively active (no egg laid), compared to nests left unharvested. This was the main effect of nest-harvesting on black-nest swiftlet reproduction in this study. Fully-feathered nestlings also spent longer (prior to fledging) in nests which had been previously harvested. This may reflect a lower rate of provisioning of the young by breeding adults energetically stressed through being forced to build a second nest.

As mentioned above, when black-nest swiftlets were allowed a full breeding season all active nests successfully fledged a nestling from the first egg laid (the first one fledged by mid-June) but none had a second attempt at breeding, even though sufficient time was available. Interestingly, the congeneric swiftlet *Aerodramus salanganus* (the mossy-nest swiftlet), which also nests in Gomantong Caves but builds an inedible nest, has multiple successful breeding attempts during the February-September breeding season (personal observation). One explanation is that, over centuries of nest-

harvesting (Francis 1987a), black-nest swiftlets have been selected to breed only once during each season, since any other attempts would have been unsuccessful due to nests being harvested before nestlings could fledge.

It is obvious that nest-harvesting had been a major selective influence on the evolution of edible-nest swiftlet reproductive behaviour at Gomantong Caves. For example, mossy-nest swiftlets nest low down on cave walls, whereas edible-nest swiftlets only nest in higher regions (Francis 1987a). A likely explanation for this is that edible-nest swiftlets have been selected to nest in more inaccessible locations, through centuries of nest-harvesting, as the lowest (most accessible) nests were subjected to greater harvesting pressure and, subsequently, had the lowest reproductive success. Thus, nest-harvesting may have led to a behavioural change in nest-site selection.

Nest-harvesting may have also had a selective effect on the nestling period of black-nest swiftlets at Gomantong Caves. Medway (1962b) reported a nestling period of approximately eight weeks for the black-nest swiftlet in Malaysia. However, in this study, the black-nest swiftlet had a nestling period of approximately 6 weeks. It is likely that the shorter time swiftlet nestlings take to fledge, the less likely they are to be killed during nest-harvesting. The shorter nestling period may thus have been selected for by nest-harvesting at Gomantong Caves.

When the glossy swiftlet was nest-harvested under the Gomantong Caves regime, nests fledged 61% less young than unharvested controls (Chapter 7). In contrast, black-nest swiftlets, nest-harvested under the Gomantong Caves regime in this study, fledged only 17% less young than unharvested controls. However, this reduction may still be sufficient to reduce the recruitment rate of black-nest swiftlets at Gomantong Caves below that which is necessary to maintain the population. Using approximate swiftlet mortality data (adult yearly mortality rate of 10%, juvenile yearly mortality rate of 50%), Francis (1987a) estimated that 40% of swiftlet nests must fledge at least one young each year if a population is to be maintained. In this study, black-nest

swiftlet nests, harvested under the Gomantong Caves regime, fledged a mean of 0.76 young per nest. This is equivalent to 76% of nests fledging one young per year, and is (theoretically) sufficient to maintain (and possibly even increase) their population size.

If an increase in the reproductive success of black-nest swiftlets at Gomantong Caves was required, the harvesting regime could be modified in two ways. First, the nest-harvest at the beginning of the breeding season could be brought forward. This would not be to give the swiftlets a longer breeding period (to allow time for successful reproduction if total brood failure occurred, as recommended in Chapter 7), since brood failure is uncommon and the birds only have one breeding attempt per season (which only requires four undisturbed months). Rather, it would be to decrease the energetic stress that the harvest places on breeding adults, by reducing egg loss. At present, energetic stress caused by nest-harvesting is most likely responsible for some adult black-nest swiftlets being unable to attempt to breed even once during each season. Even though the harvesting regime currently employed at Gomantong Caves does attempt to prevent mass egg loss at the beginning of the breeding season, by initiating the first harvest when only 10% of swiftlet nests contain eggs, many more eggs are often lost since the harvest takes approximately two weeks to complete (for example, during the harvesting of experimental nests in this study, eggs were lost from 24% of nests). The one drawback to bringing the first harvest forward is that it would decrease the yield of edible-nests at the first harvest, since not all birds would have completed nest-building.

A second, more radical (and theoretically more effective) modification of the nest-harvesting regime, to increase the reproductive success of black-nest swiftlets at Gomantong Caves, would be to delay the first harvest of the breeding season by approximately three months. This would allow the birds to breed early in the season, under conditions of minimal energetic stress. After harvesting (in July, when nestlings had fledged), the birds would most likely rebuild nests (since at least two months of the breeding season would still be available). The second nests could be harvested

upon completion (in September). This regime would allow two complete nest-harvests to take place during each breeding season at Gomantong Caves, with minimal influence on black-nest swiftlet reproductive success, and thus warrants further investigation. Experiments should also be carried out to see if nest-harvesting has the same influence on the reproductive success of the white-nest swiftlet (the other, more valuable, edible-nest species at Gomantong Caves) as it does on the black-nest swiftlet. The reproductive biology of the white-nest swiftlet differs from that of the black-nest swiftlet in that it lays a clutch of two eggs (versus one for the black-nest swiftlet) and has shorter incubation and nestling periods (see Table 1.1).

Chapter 9 Conclusion

9.1 Coevolution in host-louse associations

Host-louse associations (specifically chewing lice) have recently been used as textbook examples of coevolution (Ridley 1993), due to the high degree of cospeciation (parallel cladogenesis) seen between hosts and their chewing lice (Hafner & Nadler 1988; Paterson *et al.* 1993; Hafner & Page 1995). However, coevolution is strictly defined as "an evolutionary change in a trait of the individuals in one population in response to a trait of the individuals of a second population, followed by an evolutionary response by the second population to the changes in the first" (Janzen 1980). Thus, to imply that coevolution has occurred, evidence of cospeciation is not sufficient since coevolution is just one of many mechanisms which may result in a pattern of cospeciation between interacting populations. To imply coevolution, evidence for reciprocal adaptation between interacting species is required (Thompson 1994). Since past adaptation cannot be experimentally manipulated, we can only test whether the forces involved in reciprocal adaptation are currently operating between interacting species (see Appendix 3).

Reciprocal adaptation between interacting species has two components: reciprocal selection (changes in the phenotypic distribution, within a generation, of interacting traits; Brodie *et al.* 1995), and evolutionary response to that selection (changes in the distribution of heritable traits across generations). Without reciprocal selection, coevolution cannot occur (Thompson 1994).

Clayton *et al.* (unpublished manuscript; see Appendix 3) demonstrated reciprocal selection between feral pigeons (*Columba livia*) and their chewing lice. They showed that bill deformities in feral pigeons impaired preening, resulting in higher louse loads than pigeons with normal bills (pigeons control their louse loads through efficient preening; Clayton 1991). High louse loads significantly reduced the survival of

pigeons, reducing the fitness of birds with bill deformities. Reciprocally, preening with a normal bill selected for small body size in lice, which may facilitate their escape from preening. Reciprocal selection between interacting populations was thus demonstrated. However, this example does not imply coevolution (in the strict, Janzen (1980), sense) between feral pigeons and their lice since the interaction investigated is not specific. There is no evidence that preening in pigeons evolved in response to the selective pressure of one louse species. It is more likely to be a general adaptation, evolved in birds in response to the selective pressure imposed by ectoparasites as a whole. This is "diffuse coevolution", defined as "an evolutionary change in a trait of the individuals in one array of populations (i.e. birds) in response to a trait of the individuals in another array of populations (i.e. ectoparasites), followed by an evolutionary response by that array (ectoparasites) to the changes in the first (birds)" (Janzen 1980; 1985).

In this study, chewing lice transferred to foreign species of their swiftlet hosts had reduced survival, whereas there was no difference in the impact of normal versus foreign louse species on swiftlet fitness (Chapter 4). This implies that although lice are adapted to their normal host species, swiftlets are not adapted to their normal louse species. This is perhaps not surprising since the host, to a louse, is the entire environment upon which its fitness depends (Hopkins 1949; Kettle 1977), whereas chewing lice are just one component of a bird's environment. Louse dependence on the swiftlet host is evident in the influence of nest density on louse population structure and dynamics (Chapter 2). In comparison, lice on swiftlets may impose no selective pressure whatsoever on their hosts since lice had no impact on the fitness of either swiftlets (personal observation) or the related common swift (Chapter 6). Furthermore, neither swiftlet nor swift lice were transmitting pathogenic endoparasites (Chapters 3, 4 & 6). Thus, this study implies that associations between hosts and their chewing lice may not be examples of coevolution: lice have adapted in

response to selection pressures imposed by their hosts, but hosts may not have adapted in response to selection pressures imposed by their lice.

9.2 Cospeciation in host-lice associations

Cospeciation between host and chewing louse phylogenies (Page 1993a) is fostered by the high level of host-specificity and the clear dependence of these obligate permanent parasites on the host integument (Hopkins 1949; Kettle 1977), resulting in congruent phylogenies (Fahrenholz's Rule). However, as mentioned in Chapter 1, the relative importance of cospeciation in the evolution of host-lice associations is a point of debate (Paterson *et al.* 1995). Although cospeciation is the prevailing pattern (Page *et al.* 1996), complete congruence between host and louse phylogenies is seldom, if ever, seen (Barker 1994). Incongruence may be due to host-switching of lice between unrelated hosts ("straggling" to, and successful colonisation of, new hosts) (Lyal 1986; Barker 1996) or sorting events such as the presence of multiple lineages of parasites coupled with parasite extinction (Page 1993b) or failure of parasites to colonise both descendants of a host speciation event (Paterson & Gray in press).

Lyal (1986) states that straggling of lice is likely to occur only between host species living sympatrically and syntopically (shared habitat), where hosts come into direct physical contact. Furthermore, he states that successful colonisation must involve some overlap between the environmental tolerance of the parasite and the environmental conditions provided by the host. This study, of chewing lice on swiftlets, corroborates these points. Suspected straggling of lice was only detected between swiftlet species nesting sympatrically and syntopically in close proximity (Chapter 4). Also, lice experimentally transferred between host species survived only where there was a high degree of overlap in the environmental conditions (feather

dimensions) provided by donor and recipient hosts (within the parasites range of behavioural plasticity) (Chapter 5).

For the four species of swiftlet involved in this study, the host characters which appeared to be important determinants of louse survival (flight feather dimensions, proportional to host body size) were directly related to host taxonomy. Furthermore, for these four species, host taxonomy is an accurate reflection of host phylogeny (Lee *et al.* in press). Thus, other factors related to host phylogeny, and not host body size components per se, may have been determining transferred louse survival. This is an important distinction with regards to the parameters governing host-switching, and the relative importance of host-switching in the evolution of host-louse associations. If louse survival is restricted to closely related hosts (Chapter 4), the likelihood of host-switching being a major cause of incongruence between host and louse phylogenies would be relatively low. However, if louse survival on different hosts is governed by similarity in host morphological characters (Chapter 5) then host-switching, between more distantly related hosts with similar morphological characters (due to parallel or convergent evolution), is likely to be a much greater cause of incongruency between host and louse phylogenies.

One detail of the results presented in Chapter 4 suggests that similarity of morphology, rather than relatedness, may be governing louse survival on different host species. The louse, *Dennyus somadikartai*, did not have similar survival when transferred from its normal host (the glossy swiftlet) to mossy-nest and black-nest swiftlets (both foreign host species), even though the transfers were phylogenetically equivalent (both transfers of lice were from the host genus *Collocalia* to the host genus *Aerodramus*). Rather, louse survival was higher on the recipient host (mossy-nest swiftlet) more similar in body size (and, thus, feather dimensions) to the donor host (glossy swiftlet).

In light of the results of this study, two simple experiments involving the transfer of Malaysian swiftlet lice can be suggested to clarify the role of host-switching as a cause of incongruency between avian host and chewing louse phylogenies. The first would be to test whether or not flight feather characteristics are the host traits determining louse survival. Transfer of lice between swiftlet species, where louse survival is reduced (for example from nestlings of the glossy swiftlet to nestlings of the mossy-nest swiftlet; see Chapter 4) could be carried out where the flight feathers of recipient nestlings were replaced (through cutting and "imping") with the flight feathers of donor nestlings ("imping" is the splicing together of feathers - a technique commonly used in falconry to repair damaged flight feathers). If louse survival on recipient hosts with the flight feathers of donors was greater than controls (lice transferred to recipient nestlings where flight feathers were replaced with the flight feathers of other nestlings of the recipient species), this would strongly suggest that feather characteristics are important determinants of louse survival.

The second experiment would be to test the relative roles of host phylogenetic relatedness and morphological similarity in determining louse survival. Lice could be transferred from the glossy swiftlet (*Collocalia*) to a host species that is of similar body size, but more distantly related to, the glossy swiftlet than *Aerodramus* spp. (where transferred louse survival was reduced; see Chapter 4). An ideal candidate for the recipient host is the Asian palm swift (*Cypsiurus balasiensis*) (in the Apodini, a sister-tribe to the swiftlets). The Asian palm swift also nests in Malaysia (Chantler & Driessens 1995), shares the same breeding season as the glossy swiftlet (Hails & Turner 1984), is of very similar body size to the glossy swiftlet (adult body mass of 8.6g and wing chord of 104mm for the Asian palm swift, versus 7-8g and 95-105mm for the glossy swiftlet; Francis 1987a), and is more distantly related to the glossy swiftlet than *Aerodramus* spp. (Lee *et al.* in press). If the survival of lice transferred from nestlings of the glossy swiftlet to nestlings of the Asian palm swift was greater than that of lice transferred to nestlings of *Aerodramus* spp., this would again strongly

suggest that morphological similarity is a more important determinant of louse survival on different hosts than phylogenetic relatedness. This would imply that host-switching is a likely source of incongruence between host and louse phylogenies, specifically where parallel or convergent evolution in host morphology occurs.

In this study, controlled transfer experiments have proven to be a very useful tool for investigating host-parasite evolutionary ecology. By testing explicit predictions from micro-evolutionary theory, they can be used to reveal the underlying ecological mechanisms responsible for observed patterns of evolution. A similar approach to this thesis could be used to inform the evolutionary ecology of not just other host-parasite associations, but also that of free-living organisms.

References

- Adamson, M.L. & Caira, J.N. 1994. Evolutionary factors influencing the nature of parasite specificity. *Parasitology*, **109**: S85-S95.
- Alexander, R.D. 1974. The evolution of social behaviour. *Annual Review of Ecology and Systematics*, **5**: 325-383.
- Alstad, D.N. & Corbin, K.W. 1990. Scale insect allozyme differentiation within and between host trees. *Evolutionary Ecology*, **4**: 43-56.
- Alstad, D.N. & Edmunds, G.F. 1983. Adaptation, host specificity, and gene flow in the black pineleaf scale. Pp 413-426 in *Variable Plants and Herbivores in Natural and Managed Systems* (Denno, R.F. & McClure, M.S. eds). Academic Press, New York.
- Anderson, R.M. & Gordon, D.M. 1982. Processes influencing the distribution of parasite numbers within host populations with special emphasis on parasite-induced host mortalities. *Parasitology*, **85**: 373-398.
- Anderson, R.M. & May, R.M. 1982. Coevolution of hosts and parasites. *Parasitology*, **85**: 411-426.
- Anderson, R.M. & May, R.M. 1985. Helminth infections of humans: mathematical models, population dynamics and control. *Advances in Parasitology*, **24**: 1-101.
- Antonovics, J. & Primack, R. 1982. Experimental ecological genetics in *Plantago*. VI. The demography of seedling transplants of *P. lanceolata*. *Journal of Ecology*, **70**: 55-75.
- Ash, J.S. 1960. A study of the Mallophaga of birds with particular reference to their ecology. *Ibis*, **102**: 93-110.

- Atkinson, C.T. & van Riper III, C. 1991. Pathogenicity and epizootiology of avian haematozoa. Pp 19-48 in *Bird-parasite interactions: Ecology, evolution and behaviour* (Loye, J.E. & Zuk, M. eds). Oxford University Press, Oxford.
- Ayre, D.J. 1985. Localized adaptation of clones of the sea anemone *Actina tenebrosa*. *Evolution*, **39**: 1250-1260.
- Ayre, D.J. 1995. Localized adaptation of sea-anemone clones - evidence from transplantation over two spatial scales. *Journal of Animal Ecology*, **64**(2): 186-196.
- Baker, J.R. 1967. A review of the role played by the Hippoboscidae (Diptera) as vectors of endoparasites. *Journal of Parasitology*, **53**: 412-418.
- Baker, J.R., Bennett, G.F., Clark, G.W. & Laird, M. 1972. Avian blood coccidians. *Advances in Parasitology*, **10**: 1-30.
- Balashov, Y.S. 1984. Interaction between blood-sucking arthropods and their hosts, and its influence on vector potential. *Annual Review of Entomology*, **29**: 137-156.
- Ballabeni, W. & Ward, P.I. 1993. Local adaptation of a trematode, *Diplostomum phoxini* to the European minnow *Phoxinus phoxinus*, its second intermediate host. *Functional Ecology*, **7**: 84-90.
- Barker, S.C. 1991. Evolution of host-parasite associations among species of lice and rock-wallabies: coevolution? *International Journal for Parasitology*, **21**: 497-501.
- Barker, S.C. 1994. Phylogeny and classification, origins, and evolution of host associations of lice. *International Journal for Parasitology*, **24**: 1285-1291.
- Barker, S.C. 1996. Lice, cospeciation and parasitism. *International Journal for Parasitology*, **26**: 219-222.

- Bartlett, C.M. & Anderson, R.C. 1987. Additional comments on species of *Pelecitus* (Nematoda: Filarioidea) from birds. *Canadian Journal of Zoology*, **65**: 2813-2814.
- Bennett, G.F. 1970. Simple techniques for making avian blood smears. *Canadian Journal of Zoology*, **48**: 585-586.
- Bennett, G.F., Earle, R.A. & Peirce, M.A. 1992. New species of avian *Hepatozoon* (Apicomplexa: Haemogregarinidae) and re-description of *Hepatozoon neophronitis* (Todd & Wohlbach, 1912) Wenyon, 1926. *Systematic Parasitology*, **23**: 183-193.
- Bequaert, J.C. 1953. The Hippoboscidae or louse-flies (Diptera) of mammals and birds. Part 1. Structure, physiology and natural history. *Entomologica*, **32**: 1-209.
- Bliss, C.I. 1953. Fitting the negative binomial distribution to biological data. *Biometrics*, **9**: 176-200.
- Boag, P.T. 1987. Effects of nestling diet on growth and adult size of zebra finches (*Poephila guttata*). *Auk*, **104**: 155-166.
- Booth, D.T., Clayton, D.H. & Block, B.A. 1993. Experimental demonstration of the energetic cost of parasitism in free-ranging hosts. *Proceedings of the Royal Society of London series B*, **253**: 125-129.
- Brisbin, I.L.Jr., White, G.C. & Bush, P.B. 1986. PCB intake and the growth of waterfowl: multivariate analysis based on a reparameterized Richards sigmoid model. *Growth*, **50**: 1-11.

- Brock Fenton, M. 1975. Acuity of echolocation in *Collocalia hirundinacea* (Aves: Apodidae), with comments on the distributions of echolocating swiftlets and Molossid bats. *Biotropica*, **7**(1): 1-7.
- Brodie III, E.D., Moore, A.J. & Janzen, F.J. 1995. Visualizing and quantifying natural selection. *Trends in Ecology and Evolution*, **10**: 313-318.
- Bromhall, D. 1980. *Devil birds: the life of the swift*. Hutchinson, London.
- Brooke, R. K. 1972. Generic limits in old world Apodidae and Hirundinidae. *Bulletin of the British Ornithology Club*, **92**: 53-57.
- Brooks, D.R. & McLennan, D.A. 1991. *Phylogeny, ecology, and behaviour: A research program in comparative biology*. The University of Chicago Press, Chicago.
- Brooks, D.R. & McLennan, D.A. 1993. *Parascript: Parasites and the language of evolution*. Smithsonian Institution Press, Washington.
- Brown, C.R. & Brown, M.B. 1986. Ectoparasitism as a cost of coloniality in cliff swallows (*Hirundo pyrrhonota*). *Ecology*, **67**: 1206-1218.
- Brown, C.R., Brown, M.B. & Rannala, B. 1995. Ectoparasites reduce long-term survival of their avian hosts. *Proceedings of the Royal Society of London series B*, **262**: 313-319.
- Bryant, D.M. 1975. Breeding biology of house martins *Delichon urbica* in relation to aerial insect abundance. *Ibis*, **117**: 180-216.
- Bryant, D. M. 1978. Environmental influences on growth and survival of nestling house martins *Delichon urbica*. *Ibis*, **120**: 271-283.

- Bryant, D.M. & Hails, C.J. 1983. Energetics and growth patterns of three tropical bird species. *Auk*, **100**: 425-439.
- Chantler, P. & Driessens, G. 1995. *Swifts. A guide to the Swifts and Treeswifts of the World*. Pica Press, Mountfield.
- Charnov, E.L. 1982. *The theory of sex allocation*. Princeton University Press, Princeton.
- Choe, J.C. & Kim, K.C. 1988. Microhabitat preference and coexistence of ectoparasitic arthropods on Alaskan seabirds. *Canadian Journal of Zoology*, **66**: 987-997.
- Choe, J.C. & Kim, K.C. 1991. Microhabitat selection and adaptation of feather mites (Acari: Analgoidea) on murres and kittiwakes. *Canadian Journal of Zoology*, **69**: 817-821.
- Clark, F., Farrell, J. & Hill, L.A. 1994. A study of a population of the House Martin (*Delichon urbica* (L.)) feather louse *Bruelia gracilis* Nitzsch (Mallophaga: Ischnocera) in Lincolnshire, U.K. *The Entomologist*, **113**(3&4): 198-206.
- Clay, T. 1949. Some problems in the evolution of a group of ectoparasites. *Evolution*, **3**(4): 279-299.
- Clay, T. 1957. The Mallophaga of birds. Pp 120-158 in *First symposium on Host Specificity among Parasites of Vertebrates* (Baer, J.G. ed). University de Neuchatel, Neuchatel.
- Clay, T. 1962. A key to the species of *Actornithophilus* Ferris with notes and descriptions of new species. *Bulletin of the British Museum (Natural History) Entomology*, **11**: 189-244.

- Clayton, D.H. 1990. Host specificity of *Strigiphilus* Owl Lice (Ischnocera: Philopteridae), with the Description of New Species and Host Associations. *Journal of Medical Entomology*, **27**(3): 257-265.
- Clayton, D.H. 1991. Coevolution of avian grooming and ectoparasite avoidance. Pp 258-289 in *Bird-parasite interactions: Ecology, evolution and behaviour* (Loye, J.E. & Zuk, M. eds). Oxford University Press, Oxford.
- Clayton, D.H. & Tompkins, D.M. 1994. Ectoparasite virulence is linked to mode of transmission. *Proceedings of the Royal Society of London series B*, **256**: 211-217.
- Clayton, D.H. & Tompkins, D.M. 1995. Comparative effects of mites and lice on the reproductive success of rock doves (*Columba livia*). *Parasitology*, **110**: 195-206.
- Clayton, D.H. & Walther, B.A. In press. Collection and quantification of arthropod parasites of birds. In *Host-parasite evolution: General principles and avian models* (Clayton, D.H. & Moore, J. eds). Oxford University Press, Oxford.
- Clayton, D.H., Gregory, R.D. & Price, R.D. 1992. Comparative ecology of Neotropical bird lice (Insecta: Phthiraptera). *Journal of Animal Ecology*, **61**: 781-795.
- Clayton, D.H., Price, R.D. & Page, R.D.M. In review. Revision of *Dennyus* (*Collodennyus*) lice (Phthiraptera: Menoponidae) from swiftlets, with descriptions of 16 new species and subspecies. *Systematic Entomology*.
- Clayton, D.H., Lee, P.L.M., Tompkins, D.M. & Brodie III, E.D. Unpublished manuscript. Reciprocal natural selection on host-parasite phenotypes.
- Cobb, N.S. & Whitham, T.G. 1993. Herivore deme formation on individual trees: a test case. *Oecologia*, **94**: 496-502.

- Cohen, J. 1988. *Statistical power analysis for the behavioural sciences* (2nd edition). Academic Press, New York.
- Coles, R.B., Konishi, M. & Pettigrew, J.D. 1987. Hearing and echolocation in the Australian grey swiftlet, *Collocalia spodiopygia*. *Journal of Experimental Biology*, **129**: 365-371.
- Collins, C.T. & Murphy, R. 1993. Echolocation acuity of the Palawan swiftlet (*Aerodramus palawensis*). *Avocetta*, **2**(17): 157-162.
- Cote, I.M. & Poulin, R. 1995. Parasitism and group size in social animals: a meta-analysis. *Behavioural Ecology*, **6**(2): 159-165.
- Coyne, J.A. 1992. Genetics and speciation. *Nature*, **355**: 511-515.
- Cucco, M., Bryant, D.M. & Malacarne, G. 1993. Differences in diet of common (*Apus apus*) and pallid (*A. pallidus*) swifts. *Avocetta*, **2**(17): 131-138.
- Davies, M.S. & Snaydon, R.W. 1976. Rapid population differentiation in a mosaic environment. III. Measures of selection pressures. *Heredity*, **36**: 59-66.
- Dawson, J.R.O. 1967. The adaptation of tomato mosaic virus to resistant tomato plants. *Annals of Applied Biology*, **60**: 209-214.
- de Lope, F., Gonzalez, G., Perez, J.J. & Møller, A.P. 1993. Increased detrimental effects of ectoparasites on their bird hosts during adverse environmental conditions. *Oecologia*, **95**: 234-240.
- Desser, S.S. 1993. The Haemogregarinidae and Lankesterrellidae. Pp 247-272 in *Parasitic protozoa* (Kreier, J.P. ed). Academic Press, London.

- Dogiel, V.A. 1964. *General Parasitology (3rd edition)*. (Revised and enlarged by Polyanski, Yu.I. & Kheisin, E.M.). English translation. Oliver & Boyd, Edinburgh.
- Downes, B.J. 1990. Host-induced morphology in mites: implications for host-parasite coevolution. *Systematic Zoology*, **39**: 162-168.
- Durden, L.A. 1983. Sucking louse (*Hoplopleura erratica*: Insecta, Anoplura) exchange between individuals of a wild population of Eastern chipmunks, *Tamias striatus*, in central Tennessee, U.S.A. *Journal of Zoology*, **201**: 117-123.
- Dutton, E.J. 1905. The intermediary host of *Filaria cypseli* (Annett, Dutton, Elliot); the filaria of the African swift *Cypselus affinis*. *Thompson Yates (and Johnston) Laboratory Report, Liverpool School of Tropical Medicine*, **6**: 139-147.
- Ebert, D. 1994. Virulence and local adaptation of a horizontally transmitted parasite. *Science*, **265**: 1084-1086.
- Edmunds, G.F. & Alstad, D.N. 1978. Coevolution in insect herbivores and conifers. *Science*, **199**: 941-945.
- Eichler, W. 1942. Die Entfaltungsregel und andere Gesetzmässigkeiten in den parasitogenetischen Beziehungen der Mallophagan und anderer standiger Parasiten zu ihren Wirten. *Zoologischer Anzeiger*, **137**: 77-83.
- Etter, R.J. 1988. Asymmetrical developmental plasticity in an intertidal snail. *Evolution*, **42**: 322-334.
- Eveleigh, E.S. & Threlfall, W. 1976. Population dynamics of lice (Mallophaga) on auks (Alcidae) from Newfoundland. *Canadian Journal of Zoology*, **54**: 1694-1711.

- Ewald, P.W. 1983. Host-parasite relations, vectors, and the evolution of disease severity. *Annual Review of Ecology and Systematics*, **14**: 465-485.
- Fahrenholz, H. 1913. Ectoparasiten und Abstammungslehre. *Zoologischer Anzeiger*, **41**: 371-374.
- Fain, A. 1994. Adaptation, specificity and host-parasite coevolution in mites (Acari). *International Journal for Parasitology*, **24**(8): 1273-1283.
- Files, V.S. & Cram, E.B. 1949. A study of the comparative susceptibility of snail vectors to strains of *Schistosoma mansoni*. *Journal of Parasitology*, **35**: 555-560.
- Fisher, R.A. 1930. *The Genetical Theory of Natural Selection*. Clarendon Press, Oxford.
- Foster, M.S. 1969. Synchronised life cycles in the orange-crowned warbler and its mallophagan parasites. *Ecology*, **50**: 315-323.
- Fowler, J.A. & Hodson, D.P. 1991. Feather lice (Mallophaga) of the Dipper *Cinclus cinclus* in Central Wales. *Ringing & Migration*, **12**: 43-45.
- Fowler, J.A. & Williams, L.R. 1985. Population dynamics of Mallophaga and Acari on reed buntings occupying a communal winter roost. *Ecological Entomology*, **10**: 377-383.
- Francis, C.M. 1987a. *The management of edible bird's nest caves in Sabah*. Sabah Forest Department, Sandakan.
- Francis, C.M. 1987b. Hatching asynchrony and egg-size variation in white-bellied swiftlets (*Collocalia esculenta*). M.Sc. thesis, Queen's University, Kingston.
- Freeland, W.J. 1976. Pathogens and the evolution of primate sociality. *Biotropica*, **8**: 12-24.

- Fullard, J.H., Barclay, R.M.R. & Thomas, D.W. 1993. Echolocation in free-flying Atiu swiftlets (*Aerodramus sawtelli*). *Biotropica*, **25**(3): 334-339.
- Furness, R.W. & Palma, R.L. 1992. Phthiraptera of petrels and skuas from Gough Island, South Atlantic Ocean. *Seabird*, **14**: 33-42.
- Futyma, D.J., Walsh, J.S.Jr., Morton, T., Funk, D.J., & Keese, M.C. 1994. Genetic variation in a phylogenetic context: Responses of two specialized leaf beetles (Coleoptera: Chrysomelidae) to host plants of their congeners. *Journal of Evolutionary Biology*, **7**: 127-146.
- Hafner, M.S. & Nadler, S.A. 1988. Phylogenetic trees support the coevolution of parasites and their hosts. *Nature*, **332**: 258-259.
- Hafner M.S. & Page, R.D.M. 1995. Molecular phylogenies and host-parasite cospeciation - gophers and lice as a model system. *Philosophical Transactions of the Royal Society of London series B*, **349**: 77-83.
- Hails, C.J. 1982. A comparison of tropical and temperate insect abundance. *Biotropica*, **14**: 310-313.
- Hails, C.J. & Amirrudin, A. 1981. Food samples and selectivity of white-bellied swiftlets *Collocalia esculenta*. *Ibis*, **123**: 328-333.
- Hails, C.J. & Turner, A.K. 1984. The breeding biology of the Asian Palm Swift *Cypsiurus balasiensis*. *Ibis*, **126**: 74-81.
- Hails, C.J. & Turner, A.K. 1985. The role of fat and protein during breeding in the White-bellied swiftlet (*Collocalia esculenta*). *Journal of Zoology*, **206**: 469-484.
- Hanks, L.M. & Denno, R.F. 1994. Evidence for local adaptation in the armoured scale insect *Pseudaulacaspis pentagona* (Homoptera: Diaspididae). *Ecology*, **75**: 2301-2310.

- Harrison, L. 1915. Mallophaga from *Apteryx* and their significance; with a note on the genus *Rallicola*. *Parasitology*, **8**: 88-100.
- Harvey, P.H. & Keymer, A.E. 1991. Comparing live histories using phylogenies. *Philosophical Transactions of the Royal Society of London series B*, **332**: 31-39.
- Holyoak, D.T. & Thibault, J.C. 1978. Notes on the biology and systematics of Polynesian swiftlets, *Aerodramus*. *Bulletin of the British Ornithology Club*, **98**: 59-65.
- Hoogland, J.L. 1979. Aggression, ectoparasitism, and other possible costs of prairie dog (*Sciuridae*, *Cynomys* spp.) coloniality. *Behaviour*, **69**: 1-35.
- Hoogland, J.L. & Sherman, P.W. 1976. Advantages and disadvantages of bank swallow (*Riparia riparia*) coloniality. *Ecological Monographs*, **46**: 33-58.
- Hopkins, G.H.E. 1939. Straggling in the Mallophaga. *Entomologist*, **72**: 75-77.
- Hopkins, G.H.E. 1942. The Mallophaga as an aid to the classification of birds. *Ibis*, **14**(6): 94-106.
- Hopkins, G.H.E. 1949. The host-associations of the lice of mammals. *Proceedings of the Zoological Society of London*, **119**: 387-604.
- Hopkins, G.H.E. 1951. Stray notes on Mallophaga, XI. *Annals and Magazine of Natural History*, **4**: 371-377.
- Horning, D.S., Palma, R.L. & Pilgrim, R.L.C. 1980. *The lice (Insecta, Phthiraptera) from the Snares Islands, New Zealand*. National Museum of New Zealand miscellaneous series 6.

- Hutson, A.M. 1981. The population of the louse-fly, *Crataerina pallida* (Diptera, Hippoboscidae) on the European swift *Apus apus* (Aves, Apodidae). *Journal of Zoology*, **194**: 305-316.
- Janovy, J. Jr. In press. Protists, helminths and arthropods. In *Host-parasite evolution: General principles and avian models* (Clayton, D.H. & Moore, J. eds). Oxford University Press, Oxford.
- Janzen, D.H. 1980. When is it coevolution? *Evolution*, **34**: 611-612.
- Janzen, D.H. 1985. Coevolution as a process. Pp 83-99 in *Coevolution of parasitic arthropods and mammals* (Kim K.C. ed). John Wiley & Sons, New York.
- Jarosz, A.M. & Burdon, J.J. 1991. Host-pathogen interaction in natural populations of *Linum marginale* and *Melampsora lini*: II. Local and regional variation in patterns of resistance and racial structure. *Evolution*, **45**: 1618-1627.
- Johnson, L.S. & Albrecht, D.J. 1993. Effects of haematophagous ectoparasites on nestling house wrens, *Troglodytes aedon*: who pays the cost of parasitism? *Oikos*, **66**: 255-262.
- Joshi, A. & Thompson, J.N. 1996. Evolution of broad and specific competitive ability in novel versus familiar environments in *Drosophila* species. *Evolution*, **50**(1): 188-194.
- Kang, N., Hails, C.J. & Sigurdsson, J.B. 1991. Nest construction and egg-laying in Edible-nest Swiftlets *Aerodramus* spp. and the implications for harvesting. *Ibis*, **133**: 170-177.
- Karban, R. 1989. Fine-scale adaptation of herbivorous thrips to individual host plants. *Nature*, **340**: 60-61.

- Kellogg, V.L. 1913. Distribution and species-forming of ectoparasites. *American Naturalist*, **47**: 129-158.
- Kemper, H. 1951. Beobachtungen an *Crataerina pallida* Latr. und *Melphagus ovinus* L. *Zeitschrift für Hygiene (Zoologie)*, **39**: 225-259.
- Kethley, J.B. 1971. Population regulation in quill mites (Acarina: Syringophilidae). *Ecology*, **52**(6): 1113-1118.
- Kethley, J.B. & Johnston, D.E. 1975. Resource tracking in bird and mammal ectoparasites. *Miscellaneous Publications of the Entomological Society of America*, **9**: 231-236.
- Kettle, P.R. 1977. *A study of the Phthiraptera* (chiefly Amblycera and Ischnocera) with particular reference to the evolution and host-parasite relationships of the order. D.Phil. thesis. Kings College, London.
- Kirk, W.J. 1991. The size relationship between insects and their hosts. *Ecological Entomology*, **16**: 351-359.
- Klomp, H. 1970. The determination of clutch size in birds: a review. *Ardea*, **58**: 1-124.
- Koskimies, J. 1950. The life of the swift, *Microapus apus* (L.), in relation to the weather. *Annales Academiae Scientiarum Fennicae A, IV Biologica*: 1-151.
- Kraaijeveld, A.R. & van Alphen, J.J.M. 1993. Successful invasion of North America by two species of *Drosophila* species (Diptera, Drosophilidae) - a matter of immunity to local parasitoids. *Netherlands Journal of Zoology*, **43**(3-4): 235-241.
- Kraaijeveld, A.R. & van der Wel, N.N. 1994. Geographic variation in reproductive success of the parasitoid *Asobara tabida* in larvae of several *Drosophila* species. *Ecological Entomology*, **19**(3): 221-229.

- Kunz, T.H. 1976. Observations of the winter ecology of the bat fly *Trichobius corynorhini* Cockerell (Diptera: Strebelidae). *Journal of Medical Entomology*, **12**: 631-636.
- Lack, D. 1947. The significance of clutch size. Parts I and II. *Ibis*, **89**: 302-352.
- Lack, D. 1956a. *Swifts in a tower*. Methuen, London.
- Lack, D. 1956b. Further notes on the breeding biology of the swift (*Apus apus*). *Ibis*, **98**: 606-619.
- Lack, D. & Lack, E. 1951. The breeding biology of the swift (*Apus apus*). *Ibis*, **93**: 501-546.
- Langham, N. 1980. Breeding biology of the edible-nest swiftlet *Aerodramus fuciphagus*. *Ibis*, **122**: 477-461.
- Lau, A.S.M. & Melville, D.S. 1994. *International trade in swiftlet nests with special reference to Hong Kong*. TRAFFIC International, Cambridge.
- Lee, P.L.M. & Clayton, D.H. 1995. Population biology of swift (*Apus apus*) ectoparasites in relation to host reproductive success. *Ecological Entomology*, **20**: 43-50.
- Lee, P.L.M., Clayton, D.H., Griffiths, R. & Page, R.D.M. In press. Does behaviour reflect phylogeny in cave swiftlets (Aves: Apodidae)? A test using cytochrome *b* mitochondrial DNA sequences. *Proceedings of the National Academy of Sciences, U.S.A.*
- Lehman, T. 1993. Ectoparasites: Direct impact on host fitness. *Parasitology Today*, **9**: 8-13.

- Leuchtman, A. & Clay, K. 1989. Experimental evidence for genetic variation in compatibility between the fungus *Atkinsonella hypoxylon* and its three host grasses. *Evolution*, **43**: 825-834.
- Levine, N.D. 1982. The genus *Atoxoplasma* (Protozoa, Apicomplexa). *Journal of Parasitology*, **68**: 719-723.
- Lipsey, M.W. 1990. *Design sensitivity: statistical power for experimental research*. Sage Publications, London.
- Lively, C.M. 1989. Adaptation by a parasitic trematode to local populations of its snail host. *Evolution*, **43**(8): 1663-1671.
- Lone, E. 1991. Intraspecific morphometric variability of *Gonoicotes chrysocephalus* (Phthiraptera: Ischnocera). *Wiadomości Parazytologiczne*, **37**: 71-75.
- Lovett Doust, L. 1981. Population dynamics and local specialisation in a clonal perennial *Ranunculus repens*. II. The dynamics of leaves and a reciprocal transplant experiments. *Journal of Ecology*, **69**: 757-768.
- Lyal, C.H.C. 1985. Phylogeny and classification of the Psocodea, with particular reference to the lice (Psocodea: Phthiraptera). *Systematic Entomology*, **10**: 145-165.
- Lyal, C.H.C. 1986. Coevolutionary relationships of lice and their hosts: a test of Fahrenholtz's Rule. Pp 77-93 in *Coevolution and systematics* (Stone, A.R. & Hawksworth, D.L. eds). Clarendon Press, Oxford.
- Magrath, R.D. 1991. Nestling weight and juvenile survival in the blackbird, *Turdus merula*. *Journal of Animal Ecology*, **60**: 335-351.

- Manning, S.D., Woolhouse, M.E.J., & Ndamba, J. 1995. Geographic compatability of the freshwater snail *Bulinus globosus* and schistosomes from the Zimbabwe highveld. *International Journal of Parasitology*, **25**(1): 37-42.
- Marshall, A.G. 1981a. *The ecology of ectoparasitic insects*. Academic Press, London.
- Marshall, A.G. 1981b. The sex ratio in ectoparasitic insects. *Ecological Entomology*, **6**: 155-174.
- Marshall, A.J. & Folley, S.J. 1956. The origin of nest-cement in edible-nest swiftlets (*Collocalia* sp.). *Proceedings of the Zoological Society of London*, **126**: 383-389.
- Martins, T.L.F. & Wright, J. 1993a. Cost of reproduction and allocation of food between parent and young in the swift (*Apus apus*). *Behavioural Ecology*, **4**: 213-223.
- Martins, T.L.F. & Wright, J. 1993b. Brood reduction in response to manipulated brood sizes in the common swift (*Apus apus*). *Behavioural Ecology and Sociobiology*, **32**: 61-70.
- Martins, T.L.F. & Wright, J. 1993c. Patterns of food allocation between parent and young under differing weather conditions in the common swift (*Apus apus*). *Avocetta*, **2**(17): 147-156.
- Mayr, E. 1937. Notes on New Guinea birds, I, *Collocalia*. *Am. Mus. Novit.*, **915**: 1-19.
- McClure, H.E., Poonswad, P., Greiner, E.C. & Laird, M. 1978. *Haematozoa in the birds of eastern and southern Asia*. Memorial University of Newfoundland Press, St. John's, Newfoundland.

- McGraw, J.B. & Antonovics, J. 1983. Experimental ecology of *Dryas octopetala* ecotypes. I. Ecotypic differentiation and lifecycle stages of selection. *Journal of Ecology*, **71**: 879-897.
- Medway, Lord. 1959. Echolocation among *Collocalia*. *Nature*, **184**: 1352-1353.
- Medway, Lord. 1962a. The relationship between the reproductive cycle, moult and changes in sublingual salivary glands of the swiftlet *Collocalia maxima* Hume. *Proceedings of the Zoological Society of London*, **138**: 305-315.
- Medway, Lord. 1962b. The swiftlets (*Collocalia*) of Niah Cave, Sarawak. Part 1. Breeding biology. *Ibis*, **104**: 45-66.
- Medway, Lord. 1966. Field characters as a guide to the specific relations of swiftlets. *Proceedings of the Linnean Society of London*, **177**: 151-172.
- Medway, Lord. 1969. Studies on the biology of the edible-nest swiftlets of South-east Asia. *Malaysian Nature Journal*, **22**(2): 57-63.
- Medway, Lord. & Pye, J.D. 1977. Echolocation and the systematics of swiftlets. Pp 225-238 in *Evolutionary ecology* (Stonehouse, B. & Perrins, C. eds). Macmillan Press, London.
- Møller, A.P. 1987. Advantages and disadvantages of coloniality in the swallow, *Hirundo rustica*. *Animal Behaviour*, **35**: 819-832.
- Møller, A.P. 1993. Ectoparasites increase the cost of reproduction in their hosts. *Journal of Animal Ecology*, **62**: 309-322.
- Møller, A.P. 1994. Parasites as an environmental component of reproduction in birds as exemplified by the swallow *Hirundo rustica*. *Ardea*, **82**: 161-172.

- Møller, A.P. In press. Parasitism and the evolution of host life history. In *Host-parasite evolution: General principles and avian models* (Clayton, D.H. & Moore, J. eds). Oxford University Press, Oxford.
- Møller, A.P., Dufva, R. & Allander, K. 1993. Parasites and the evolution of host social behaviour. *Advances in the Study of Behaviour*, **22**: 65-102.
- Mopper, S., Beck, M., Simberloff, D. & Stiling, P. 1995. Local adaptation and agents of selection in a mobile insect. *Evolution*, **49**(5): 810-815.
- Morand, S., Manning, S.D. & Woolhouse, M.E.J. 1996. Parasite-host coevolution and geographic patterns of parasite infectivity and host susceptibility. *Proceedings of the Royal Society of London series B*, **263**: 119-128.
- Nadler, S.A., Hafner, M.S., Hafner, J.C. & Hafner, D.J. 1990. Genetic differentiation among chewing lice populations (Mallophaga: Trichodectidae) in a pocket gopher contact zone (Rodentia: Geomyidae). *Evolution*, **44**: 942-951.
- Nelson, B.C. & Murray, M.D. 1971. The distribution of Mallophaga on the domestic pigeon (*Columba livia*). *International Journal for Parasitology*, **1**: 21-29.
- O'Connor, R.J. 1978. Brood reduction in birds: selection for fratricide, infanticide and suicide? *Animal Behaviour*, **26**: 79-96.
- Otte, D. & Endler, J.A. 1989. *Speciation and its consequences*. Sinauer Associates, Sunderland.
- Page, R.D.M. 1993a. Parasites, phylogeny and cospeciation. *International Journal for Parasitology*, **23**: 499-506.
- Page, R.D.M. 1993b. Genes, organisms, and area: the problem of multiple lineages. *Systematic Biology*, **42**: 77-84.

- Page, R.D.M., Clayton, D.H. & Paterson, A.M. 1996. Lice and cospeciation: A response to Barker. *International Journal for Parasitology*, **26**(2): 213-218.
- Parker, M.A. 1985. Local population differentiation for compatibility in an annual legume and its host-specific fungal pathogen. *Evolution*, **39**: 713-723.
- Paterson, A.M. & Gray R.D. In press. Host parasite cospeciation, host switching and missing the boat. In *Host-parasite evolution: General principles and avian models* (Clayton, D.H. & Moore, J. eds). Oxford University Press, Oxford.
- Paterson, A.M., Gray, R.D. & Wallis, G.P. 1993. Parasites, petrels and penguins: does louse presence reflect seabird phylogeny? *International Journal for Parasitology*, **23**(4): 515-526.
- Paterson, A.M., Gray, R.D. & Wallis G.P. 1995. Of lice and men: the return of the "comparative parasitology" debate. *Parasitology Today*, **11**: 158-160.
- Peirce, M.A. 1984. Haematozoa of British birds. IX. Blood parasites of birds from Sark and miscellaneous findings from other areas. *Journal of Natural History*, **18**: 793-795.
- Perrins, C. 1971. Age of first breeding and adult survival rates in the swift. *Bird Study*, **18**: 61-70.
- Poiani, A. 1992. Ectoparasitism as a possible cost of social life: a comparative analysis using Australian passerines (Passeriformes). *Oecologia*, **92**: 429-441.
- Poulin, R. 1991. Group living and infestation by ectoparasites in passerines. *Condor*, **93**: 418-423.
- Pratt, H.D. 1986. A review of the English and scientific nomenclature of cave swiftlets (*Aerodramus*). *'Elepaio*, **46**(11): 119-125.

- Price, P.W. 1980. *Evolutionary biology of parasites*. Princeton University Press, Princeton.
- Price, P.W., Westoby, M., Rice, B., Atsatt, P.R., Fritz, R.S., Thompson, J.N. & Mobley, K. 1986. Parasite mediation in ecological interactions. *Annual Review of Ecology and Systematics*, **17**: 487-505.
- Reed, D.L. 1994. *Possible mechanism for cospeciation in pocket gophers and lice, and evidence of resource partitioning among coexisting genera of lice*. M.Sc. Thesis. Louisiana State University and Agricultural and Mechanical College.
- Rice, W.R. 1983. Sexual reproduction: an adaptation reducing parent-offspring contagion. *Evolution*, **46**: 1317-1320.
- Richards, F.J. 1959. A flexible growth function for empirical use. *Journal of Experimental Botany*, **10**: 290-300.
- Ridley, M. 1993. *Evolution*. Blackwell Scientific Publications, Boston.
- Roderick, G.K. 1996. Geographic structure of insect populations: Gene flow, phylogeography, and their uses. *Annual Review of Entomology*, **41**: 325-352.
- Rohde, K. 1979. A critical evaluation of intrinsic and extrinsic factors responsible for restricting niches. *American Naturalist*, **114**: 648-671.
- Rohde, K. 1994. Niche restriction in parasites: proximate and ultimate causes. *Parasitology*, **109**: S69-S84.
- Rothschild, M. & Clay, T. 1952. *Fleas, flukes and cuckoos*. Collins, London.
- Rozsa, L. 1993. Speciation patterns of ectoparasites and "straggling" lice. *International Journal for Parasitology*, **23**(7): 859-864.

- Rozsa, L., Rekasi, J. & Reiczigel, J. 1996. Relationship of host coloniality to the population ecology of avian lice (Insecta: Phthiraptera). *Journal of Animal Ecology*, **65**: 242-248.
- Sasvari, L. & Hegyi, Z. 1994. Colonial and solitary nesting choice as alternative breeding tactics in tree sparrow *Passer montanus*. *Journal of Animal Ecology*, **63**: 265-274.
- Shaw, D.J. & Dobson, A.P. 1996. Patterns of macroparasite abundance and aggregation in wildlife populations: a quantitative review. *Parasitology*, **111**: S111-S133.
- Sibly, C.G., Corbin, K.W. & Haavie, J.H. 1969. The relationships of the flamingos as indicated by the egg-white proteins and hemoglobins. *Condor*, **71**: 155-179.
- Sims, R.W. 1959. Edible bird's nests. *New Biology*, **30**: 47-58.
- Sims, R.W. 1961. The identification of Malaysian species of swiftlets. *Ibis*, **103a**: 205-211.
- Southwood, T.R.E. 1978. *Ecological Methods (2nd edition)*. Chapman & Hall, London.
- Stalter, R. & Bateson, W.T. 1969. Transplantation of salt marsh vegetation, Georgetown, South Carolina. *Ecology*, **50**: 1087-1089.
- Stock, T.M. & Hunt, L.E. 1989. Site specificity of three species of lice, Mallophaga, on the Willow Ptarmigan, *Lagopus lagopus*, from Chikot Pass, British Columbia. *Canadian Field-Naturalist*, **103**(4): 584-588.
- Summers, R.W. 1975. On the ecology on *Crataerina hirundinis* (Diptera: Hippoboscidae) in Scotland. *Journal of Zoology*, **175**: 557-570.

- Tella, J.L., Gortazar, C., Gajon, A. & Osacar, J.J. 1995. Apparent lack of effects of a high louse-fly infestation (Diptera, Hippoboscidae) on adult colonial alpine swifts. *Ardea*, **83**: 435-439.
- Thompson, J.N. 1994. *The coevolutionary process*. The University of Chicago Press, Chicago.
- Thompson, J.N. 1996. Evolutionary ecology and the conservation of biodiversity. *Trends in Ecology and Evolution*, **11**(7): 300-303.
- Toft, C.A. & Karter, A.J. 1990. Parasite-host coevolution. *Trends in Ecology and Evolution*, **5**: 326-329.
- Tompkins, D. M. 1996. *Evolutionary ecology of bird-parasite associations*. D.Phil. Thesis. University of Oxford, Oxford.
- Turkington, R. & Harper, J.L. 1979. The growth, distribution and neighbour relationships of *Trifolium repens* in a permanent pasture. IV. Fine-scale biotic differentiation. *Journal of Ecology*, **67**: 245-254.
- Ubelaker, J.E. 1970. Some observations on ecto- and endoparasites of Chiroptera. In *About bats* (Slaughter, B.E. & Walton, D.W. eds). Southern Methodist University Press, Dallas.
- Unruh, T.R. & Luck, R.F. 1987. Deme formation in scale insects: a test with the pinyon needle scale and a review of other evidence. *Ecological Entomology*, **12**: 439-449.
- von Ihring, H. 1902. Die Helminthen als Hilfsmittel der zoogeographischen Forschung. *Zoologischer Anzeiger*, **26**: 42-51.

- Wainhouse, D. & Howell, R.S. 1983. Intraspecific variation in beech scale populations and susceptibility of their host *Fagus sylvatica*. *Ecological Entomology*, **8**: 351-359.
- Wang, C.C. 1921. The composition of Chinese edible birds' nests and the nature of their proteins. *Journal of Biological Chemistry*, **49**: 429-438.
- Ward, R.A. 1957. A study of the host distribution and some relationships of biting lice (Mallophaga) parasitic on birds on the order Tinamiformes. Part II. *Ant. ent. Soc. Am.*, **50**: 452-459.
- Waugh, D.R. & Hails, C.J. 1983. Foraging ecology of a tropical aerial feeding bird guild. *Ibis*, **125**: 200-217.
- Wheatley, B.P. 1980. Malaria as a possible selective factor in the speciation of macaques. *Journal of Mammology*, **61**: 307-311.
- Wheeler, T.A. & Threlfall, W. 1986. Observations on the ectoparasites of some Newfoundland passerines (Aves: Passeriformes). *Canadian Journal of Zoology*, **64**: 630-636.
- Zonfrillo, B. 1991. The feather louse *Halipeurus gravis gravis* Timmermann, 1961 (Mallophaga: Philopteridae) from a sooty shearwater *Puffinus griseus* in Scotland. *Seabird*, **11**: 17-18.
- Zumpt, F. 1966. The arthropod parasites of vertebrates in Africa south of the Sahara. 3. Insecta excl. Phthiraptera. *Publications of the South African Institute for Medical Research*, **13**: 1-283.

Appendix C Reciprocal natural selection on host-parasite phenotypes

[Clayton, D.H., Lee, P.L.M., Tompkins, D.M. & Brodie III, E.D.

Unpublished manuscript. Reciprocal natural selection on host-parasite phenotypes]

Coevolution is one of the most important processes shaping adaptation of interacting species (1-7). However, the unique mechanism thought to drive coevolution - reciprocal selection on interacting phenotypic traits - has not been demonstrated in natural populations (7). We tested for reciprocal selection in a simple host-parasite system consisting of feral pigeons (*Columba livia*) and their chewing lice (Phthiraptera: Insecta). Previous work showed that pigeons control their louse loads through efficient preening, while lice try to escape from preening with complex avoidance behaviour (8). Here we show that feral pigeons with impaired preening, owing to slight bill deformities, have higher louse loads than pigeons with normal bills. High louse loads significantly reduce the survival of pigeons, thus reducing the fitness of birds with bill deformities. Preening with a normal bill reciprocally selects for small body size in two species of lice, which may facilitate their escape from preening. These results verify a crucial element of coevolutionary theory by demonstrating that reciprocal selection on interacting phenotypes can occur in nature.

Coevolution is a generally accepted process even though much of the evidence for it remains circumstantial (5-7). Coevolution has often been inferred from patterns of coadaptation like that between species of coumarin-containing plants and the caterpillars with counter-adaptations for feeding on them (9). Another example is the pattern of coadaptation between brood-parasitic cuckoos, which lay mimetic eggs, and their host species, which attempt to eject those eggs (10, 11). Although such patterns are

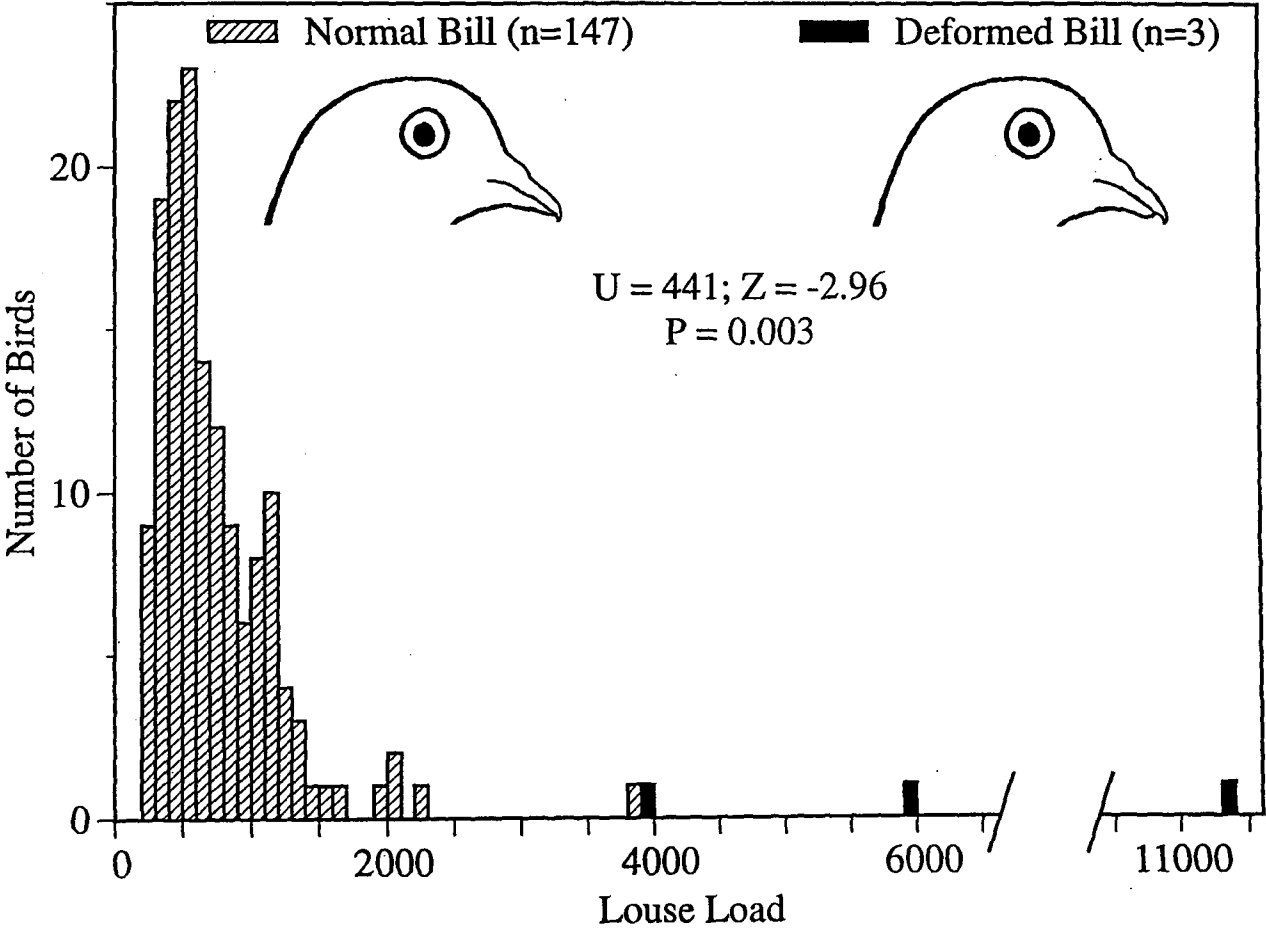
compelling (but see (12)), they are not robust evidence for coevolution because traits that appear to be coadapted could originally have evolved in other contexts (2-4, 6, 7, 13, 14). More convincing evidence requires a demonstration of reciprocal selection currently operating on interacting traits, analogous to the recent demonstration of character displacement operating on the phenotypic traits of competing species (15).

In keeping with formal evolutionary theory (16-21), we distinguish phenotypic selection, which is a change in the phenotypic distribution of a trait *within* a generation, from evolutionary response, which is a change in the distribution of a heritable trait *across* generations. Coevolution cannot occur in the absence of reciprocal phenotypic selection. The feasibility of reciprocal selection has long been one of the most pressing questions in coevolution research (2, 6, 7, 22). Our goal was to carry out controlled experiments to test for reciprocal selection on components of host defence and parasite escape.

Chewing lice are obligate parasites that occur on most, if not all, species of birds (23, 24). They feed largely on feathers and dermal debris and complete their entire life cycle on the body of the host, even gluing their eggs to feathers with a glandular cement. Birds attempt to remove lice by preening them off with the bill (8). In natural populations louse loads are highest on birds with minor bill deformities that prevent the full occlusion of the mandibles necessary for efficient preening (8, 23-26). In our study population the highest louse loads were on birds with such deformities (Figure C.1).

If high louse loads reduce host fitness, lice will generate selection against these bill deformities. Although feather damage from lice is known to have energetic costs (27), no resultant impact on host reproductive success has been demonstrated (28). We tested for an effect of lice on mortality, which is another important component of avian fitness (20). We experimentally manipulated the louse loads of the host population in Figure C.1, then monitored their long-term survival. Bill deformities might reduce survival

Figure C.1 Distribution of lice on adult pigeons nesting under a bridge over a stream near Manteno, Illinois, U.S.A.. The highest louse loads were on birds with minor bill deformities, such as several mm missing from the tip of the lower mandible (illustrated). The frequency of deformities in the study population (2.0%) was similar to that in other populations of pigeons (8) as well as in other species of birds (26). The three deformed birds were all in good condition and one individual was incubating eggs at the time of capture. All birds were live-trapped within a three week period (21 May - 11 June 1988). Louse loads were estimated from regression models that predict total load from timed counts of lice on various body regions ($r^2 \geq 0.82$) (8, 27, 28).



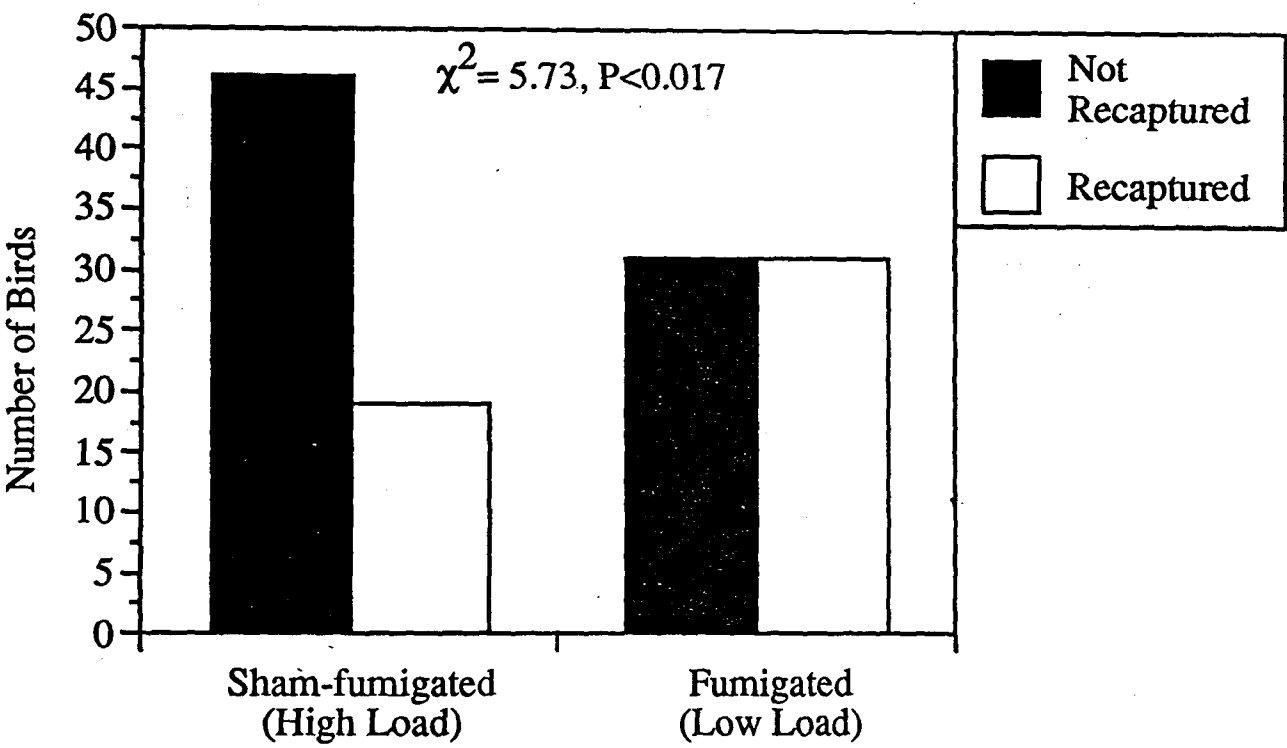
directly, as well as through their effects on louse populations, so it was necessary to manipulate louse load independently of bill deformities to test for an impact of lice on host fitness. To do this we impaired the preening ability of birds in our study population by placing "bits" between the mandibles of *every* bird's bill (29). We then fumigated "low-load" birds to prevent increases in their lice, while sham-fumigating "high-load" birds with water. The combination of biting and fumigation resulted in high-load birds having significantly more lice than low-load birds (30). High-load birds also had significantly more feather damage, but significantly lower body mass, than low-load birds (30). High louse loads did not affect short-term host survival; three months into the study the recapture rates of high- and low-load birds were 80% and 85%, respectively ($\chi^2 = 0.17$, $p = 0.68$).

The second recapture, one year into the study (31), revealed a significant long-term effect of high louse load on host survival. Only 29% of high-load birds were recaptured, compared to 50% of low-load birds (Figure C.2). Birds not recaptured were presumed to have died as opportunities for dispersal to new roost sites were severely limited. Pigeons require man-made structures for roosting (32) and the bridge in our study was surrounded by large expanses of open farmland. Birds from our study population were never captured at another bridge being monitored five miles away.

It is likely that the high-load birds in our study died as a result of thermoregulatory stress. Feather damage from pigeon lice causes an increase in whole-body thermal conductance in response to the reduced insulative effectiveness of the plumage (27). Damaged birds maintain a constant body temperature, despite the increase in thermal conductance, by elevating their metabolic rates an average of 8.5%. This elevated metabolic rate is maintained by drawing on fat reserves, leading to a reduction in body mass (27) like that shown in the current study (30). This is the first demonstration of an effect of lice on the survival of pigeons or any other free-ranging species of bird.

Figure C.2 Reduced survival of pigeons with experimentally increased louse loads.

The number of birds recaptured in June 1989 was compared to the number released after the initial treatment the previous June. Significantly more low-load than high-load birds were recaptured. Louse loads were manipulated using a two-step procedure applied to the normal-billed birds in Figure C.1 (29).



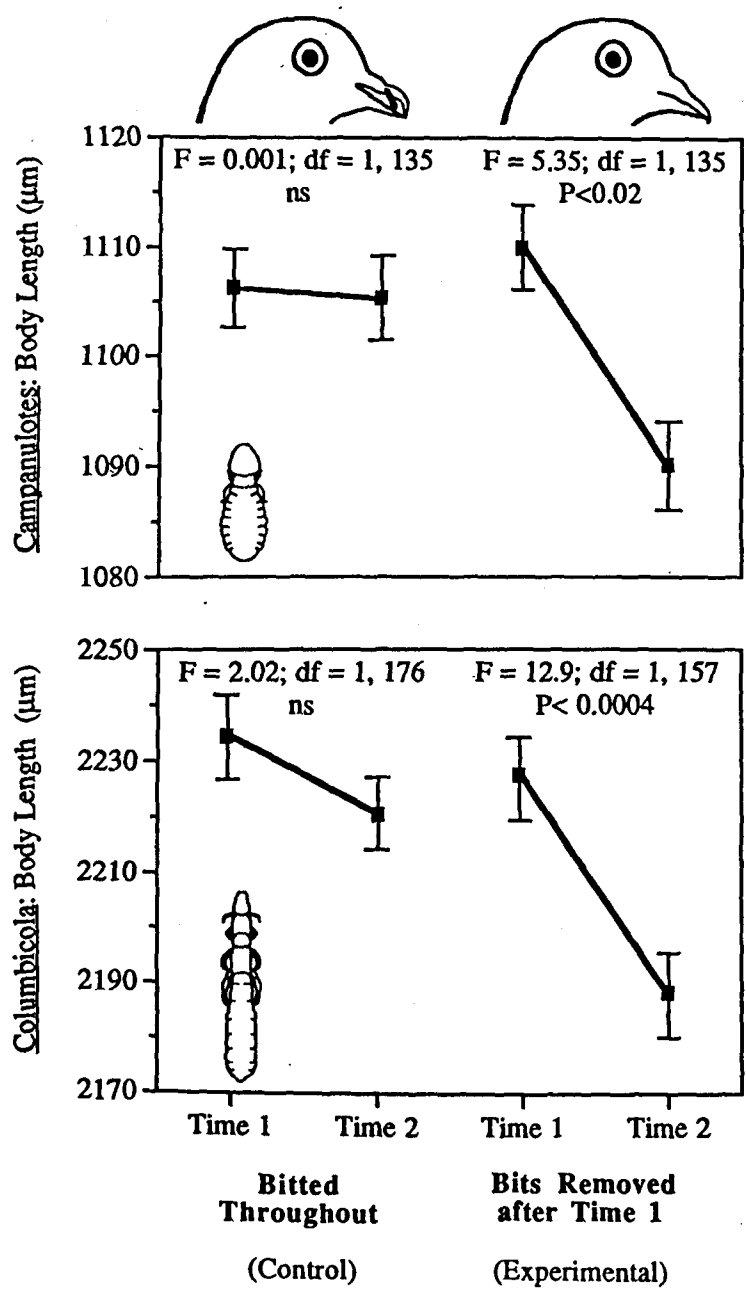
The reciprocal test for a selective effect of pigeons on lice used captive pigeons bred from wild stock (33). Two species of lice were studied, neither of which was capable of leaving the body of the host since both have appendages that are specialised for locomotion only on feathers (8). Transmission among hosts requires physical contact between the feathers of different individual birds (34).

Preening selected for smaller body size in both lice. The size of lice on control birds did not change significantly between T1 and T2 for either species, whereas there was a dramatic reduction in the size of both species of lice on experimental birds (Figure C.3). *Campanulotes* showed a mean reduction of 20 microns on experimentals, compared to a reduction of <1 micron on controls. *Columbicola* showed a mean reduction of 39 microns on experimentals, compared to a (nonsignificant) reduction of 13 microns on controls (Figure C.3).

The experimental design allowed us to confirm preening as the causative agent of selection on louse body size (35). Treating each experimental-control pair of birds (33) as a replicate selection line, we directly compared the change in lice on experimentals to the change in lice on controls. *Campanulotes* changed significantly more on experimentals than on controls (paired-t = 2.98, d.f. = 5, $p < 0.03$, 2-tailed). *Columbicola* showed a similar trend, albeit nonsignificant (paired-t = 2.09, d.f. = 7, $p < 0.07$, 2-tailed). Preening appears to be an effective generalised defense since *Campanulotes* and *Columbicola* differ greatly in morphology, behaviour and microhabitat distribution on the host (36). Generalised defences are considered excellent candidates for diffuse coevolution between a host and its parasite community (6).

Small body size may facilitate the ability of lice to escape from preening. *Campanulotes* spends most of its time on the abdomen and escapes by running through the downy regions of contour feathers (8). Small size may improve its ability to manoeuvre through the downy matrix. *Columbicola* spends most of its time on the underside of the

Figure C.3 Selective effect of preening on the morphology of two species of pigeon lice: *Campanulotes bidentatus* and *Columbicola columbae* (illustrated to the same scale). Preening-imposed selection was relaxed for several louse generations by biting all birds prior to the experiment (33). Samples of adult lice were collected at Time 1, then the preening ability of experimental birds was restored by removing their bits. Post-selection samples of adult lice were collected at Time 2. An average of 26.4 days (s.d. = 1.04) was allowed to pass between the T1 and T2 samples, thus maximising the episode of selection without exceeding the time to first reproduction of the lice (= 27 days (23)). Phenotypic selection is best measured within a generation to avoid confounding it with the evolutionary response to selection (44). Body lengths (mean $\pm$ 1 s.e.) of both species of lice decreased significantly on birds with restored preening, but did not change significantly on birds that wore bits throughout the trial (33).



wings and escapes by insertion between the rigid barbs of flight feathers. Small size may improve the fit of this species between the barbs (37).

The results of our first experiment (Figure C.2) show that high louse loads reduce host survival, thus selecting against bill deformities that result in high louse loads (Figure C.1). Lice may also have a selective effect on normal bills, analogous to the effect of diet on bill morphology (38), but we have not yet tested this possibility. Our second experiment (Figure C.3) shows that preening with a normal bill reciprocally selects for small body size in lice, which may facilitate their escape from preening. Preening could also have a selective effect on other phenotypic traits of lice such as microhabitat distribution and oviposition site, as suggested by Nelson and Murray (36).

For reciprocal selection to result in coevolution, both phenotypic targets of selection must have heritable components (6, 39). If phenotypic variation is purely environmental, there can be no evolution of the trait. It is extremely likely that both of the traits we studied have heritable components. Heritable variation in bill morphology, including congenital deformities, have been documented for a variety of bird species (reviewed in (40) and (26)) and heritable deformities of the bill are common in pigeons (41, 42). It also seems likely that predisposition for accidental deformities, such as bill tip breakage, would have some heritable component. We are unaware of any studies of the heritability of body size in bird lice; however, body size is heritable in most other insect groups that have been tested (43).

The selective effect of lice on pigeon morphology, and the tandem effect of pigeons on louse morphology, is an example of reciprocal selection on interacting phenotypic traits. Reciprocal selection is the unique mechanism that drives coevolution, and is the critical parameter distinguishing coevolution from all other modes of evolution (2, 3, 7). The results of our experiments confirm for the first time that reciprocal phenotypic selection can operate in natural populations of hosts and parasites.

1. P. R. Ehrlich, P. H. Raven, *Evol.* **18**, 586-608 (1964).
2. D. H. Janzen, *Evol.* **34**, 611-612 (1980).
3. D. J. Futuyma, M. Slatkin, *Coevolution* (Sinauer, Sunderland, MA, 1983).
4. M. H. Nitecki, *Coevolution* (University of Chicago Press, Chicago, 1983).
5. D. J. Futuyma, R. M. May, in *Genes in Ecology*, R. J. Berry, T. J. Crawford, G. M. Hewitt, Eds. (Blackwell Scientific, London, 1992), pp. 139-166.
6. M. D. Rausher, in *Insect Chemical Ecology: an Evolutionary Approach*, B. D. Roitberg, M. B. Isman, Eds. (Routledge, Chapman and Hall, New York, 1992), pp. 20-88.
7. J. N. Thompson, *The Coevolutionary Process* (The University of Chicago Press, Chicago, 1994).
8. D. H. Clayton, in *Bird-parasite Interactions: Ecology, Evolution, and Behaviour* J. E. Loye, M. Zuk, Eds. (Oxford University Press, Oxford, 1991), pp. 258-289.
9. M. Berenbaum, *Evol.* **37**, 163-179 (1983).
10. N. B. Davies, M. de L. Brooke, *J. Anim. Ecol.* **58**, 207-224 (1989).
11. N. B. Davies, M. de L. Brooke, *J. Anim. Ecol.* **58**, 225-236 (1989).
12. J. N. Thompson, in *Coevolution and Systematics*, A. R. Stone, D. L. Hawksworth, Eds. (Clarendon Press, Oxford, 1986), pp. 119-143.
13. D. R. Strong, J. H. Lawton, T. R. E. Southwood, *Insects on Plants: Community Patterns and Mechanisms* (Harvard University Press, Cambridge, 1984).
14. M. Ridley, *Evolution* (Blackwell Scientific, Oxford, 1993).

15. D. Schluter, *Science* **266**, 798-801 (1994).
16. R. A. Fisher, *The Genetical Theory of Natural Selection* (Oxford University Press, Oxford, 1930).
17. J. B. S. Haldane, *Proc. IX Intl. Cong. Genet. (Caryologica supplement)* **1**, 480-487 (1954).
18. R. Lande, S. J. Arnold, *Evol.* **37**, 1210-1226 (1983).
19. S. J. Arnold, M. J. Wade, *Evol.* **38**, 720-734 (1984).
20. T. D. Price, P. T. Boag, in *Avian Genetics*, F. Cooke, P. A. Buckley, Eds. (Academic Press, London, 1987), pp. 257-288.
21. E. D. Brodie III, A. J. Moore, F. J. Janzen, *Trends in Ecology and Evolution* **10**, 313-318 (1995).
22. D. W. Schemske, in *Coevolution*, M. H. Nitecki, Eds. (University of Chicago Press, Chicago, 1983), pp. 67-110.
23. A. G. Marshall, *The Ecology of Ectoparasitic Insects* (Academic Press, London, 1981).
24. M. Rothschild, T. Clay, *Fleas, Fukes and Cuckoos* (Collins, London, 1952).
25. J. S. Ash, *Ibis* **102**, 93-110 (1960).
26. D. E. Pomeroy, *Br. Birds* **55**, 49-72 (1962).
27. D. T. Booth, D. H. Clayton, B. A. Block, *Proc. R. Soc. Lond. B* **253**, 125-129 (1993).
28. D. H. Clayton, D. M. Tompkins, *Parasitol.* **110**, 195-206 (1995).

29. Bits are small (< 0.8 g) C-shaped pieces of plated steel inserted between the mandibles and crimped slightly in the nostrils to prevent dislodging, but without piercing the tissue. Bits create a 1.0 - 3.0 mm gap between the mandibles that impairs preening with no detectable side effects (8, 27, 28; D. H. Clayton, *Am. Zool.* 30, 251-262 (1990)). To control for any undetected effects, *all* birds in the population were bitted. Unless fumigated, bitted birds experience an increase in louse load that mimics *but does not exceed* the increased loads of deformed birds (8, 28). Half of the birds, selected at random, were designated "low-load" and fumigated with 1.0% pyrethrum, which has been shown to have no side effects on the host (28; J. A. Jackson, *Sialia* 7, 17-25 (1985)). The remaining "high-load" birds were sham-fumigated with water. All birds were then released at the capture site.
30. In September, three months following initial treatment, all birds were recaptured and their louse loads estimated with a six category scoring system that greatly reduces the amount of time birds must be held captive (cf. Figure C.1 procedure). Categorical scoring proved reliable in an earlier study (27). Sixty-five high-load birds had a mean louse score of 4.2 (s.d. = 1.3) compared with a score of 2.8 (s.d. = 1.4) for 62 low-load birds (Mann-Whitney $U = 945$, $p = 0.0001$). Feather samples taken from 54 of the high-load birds (ref 27 for methods) weighed a mean of 0.60 mg (s.d. = 0.08) compared with a mean of 0.74 mg (s.d. = 0.09) from 56 low-load birds ($t = 8.50$, d.f. = 108, $p = 0.0001$, 1-tailed). The mean overall body mass of high-load birds was 337.5 g (s.d. = 25.6) compared with 345.8 g (s.d. = 22.8) for low-load birds ($t = 1.92$, d.f. = 125, $p = 0.029$, 1-tailed). Low-load birds were refumigated and high-load birds sham-fumigated, then all birds were released at the capture site.
31. In June 1989 the entire population under the bridge was recaptured at night by sealing off all escape routes with large nets, which made the capture method

independent of experimental treatment. Bits were removed from all birds prior to final release.

32. R. F. Johnston, in *The Birds of North America, No. 13*, A. Poole, P. Stettenheim, F. Gill, Eds. (Philadelphia, The Academy of Natural Sciences, 1992), pp. 1-16.
33. The experiment used captive birds bred from individuals captured about 100 miles from the field site (8). Eighteen birds, bitted several months prior to the experiment, were randomly isolated in 18 cages arranged in pairs with one experimental and one control to a pair. This paired design controlled for possible side effects of correlated preening rates between neighbouring pigeons (D. H. Clayton, *Am. Zool.* **30**, 251-262 (1990)). The cages were well separated to prevent contact between feathers, which could allow transmission of lice between birds. At Time 1, 100 lice of each species were collected from each bird and preserved in 70% alcohol. Bits were then removed from the nine experimental birds but were left in place on the nine controls. At Time 2 all of the lice remaining on each bird were collected and preserved. Five male and five female lice of each species from each bird were randomly selected from the T1 samples and mounted on microscope slides. An equal number of lice from the T2 samples were mounted. All lice were measured with a phase-contrast microscope by one of us (PLML) who was unaware of host treatment. The body length measurement was highly repeatable for both species (*Campanulotes* $r = 0.88$; *Columbicola* $r = 0.98$; $p = 0.0001$ for both; $n = 25$ males of each species; repeatabilities calculated from two measurements of each specimen taken on different days (C. M. Lessells, P. T. Boag, *Auk* **104**, 116-121 (1987)). Birds from which at least five lice of each sex could not be recovered at Time 2 were omitted from the analysis so that T1 and T2 means would be estimated using equal numbers of observations. All measurements were of adult lice, which do not moult or undergo other developmental changes in body size. Female lice of both species are

approximately 15% larger than males, so female means were adjusted for pooling of the sexes (Figure C.3; variation within each sex was similar). One-way ANOVA's were used to compare the body lengths of lice at T1 and T2.

34. D. H. Clayton, D. M. Tompkins, *Proc. Roy. Soc. Lond. B.* **256**, 211-217 (1994).
35. M. J. Wade, S. Kalisz, *Evol.* **44**, 1947-1955 (1990).
36. B. C. Nelson, M. D. Murray, *Intnl. J. Parasitol.* **1**, 21-29 (1971).
37. *Columbicola* body width, which is presumably the most important determinant of interbarb fit, is significantly correlated with body length (unpublished data).
38. P. T. Boag, P. R. Grant, *Science* **214**, 82-85 (1981); T. B. Smith, *Evol.* **44**, 832-842 (1990).
39. R. E. Lenski and B. R. Levin, *Amer. Natur.* **125**, 585-602 (1985).
40. P. R. Grant, *Ecology and Evolution of Darwin's Finches* (Princeton University Press, Princeton, 1986).
41. E. L. Hanebrick, *Am. Pigeon J.* **59**, 105 (1970).
42. D. C. Tudor, *Pigeon Health and Disease* (Iowa State Univ. Press, Ames, 1991).
43. T. A. Mousseau, D. A. Roff, *Heredity* **59**, 181-197 (1987).
44. B. F. J. Manly, *The Statistics of Natural Selection on Animal Populations* (Chapman and Hall, London, 1985).
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Appendix C

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