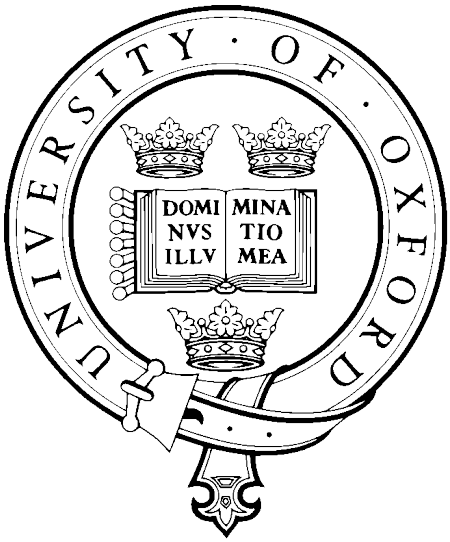
Avian Malaria in the Montane Tropics



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Abstract

Understanding the unequal distribution of life on earth is a fundamental goal of ecology and evolutionary biology. Past efforts to explain large-scale patterns in diversity have tended to focus on two broad classes of explanation, one invoking the importance of abiotic factors (i.e. climate and vegetation) and the other biotic (i.e. competition); but neither has proven entirely adequate. Parasites are a major but poorly understood component of life that may offer some answers. Yet despite widespread theoretical support and some empirical evidence, the role of parasites in explaining patterns in the diversity, distribution, and abundance of species remains largely untested in natural communities. In this thesis I use a mega-diverse elevation gradient of birds as a model system to study the role of avian malaria in explaining these macroecological patterns.

In the first data chapter I tested the extent to which patterns of infection across species is predictable. I found that the effects of host ecology and environment were weakly related to infection prevalence and were not consistent across different malaria lineages. Instead, I show that hosts coexisting with many close phylogenetic relatives consistently experience higher infection than evolutionarily distinct host species. In the second chapter I tested if parasite sharing may help explain these observed relationships and show that parasite sharing among host pairs declines with the time since divergence. Spatial contiguity between host pairs was also positively associated with parasite sharing. In the third chapter I tested how infection prevalence varies across species ranges in accordance with expected variation in host abundance. I show that birds are more likely to be infected at the centre of their elevation range, where host abundance is expected to be highest. Intriguingly, I also found that the incidence of host infection is unrelated to the position within the geographic range of the parasite. In the fourth data chapter, I tested whether parasites may regulate diversity by limiting geographic ranges of their hosts through 'apparent competition' in which a non-lethal parasite in a primary host, may be lethal in a secondary host. In support of this, I found that more observed bird ranges end at parasite infection zones than would be expected by chance.

Taken together, my results suggest that parasites may play a major role in shaping patterns in the distribution and diversity of species, over both ecological and evolutionary scales. This is likely to arise and be maintained by host parasite interactions in which distantly related hosts are less likely to be infected by local parasites than close relatives, thus promoting the build up of diversity locally. On the basis of my analyses, I conclude that across montane elevation gradients in birds, and across diversity gradients more generally, parasites are likely to play a crucial role in the origin and maintenance of high biological diversity.

Declaration

The work presented in this thesis is my own with the following acknowledgements.

The empirical data used throughout this thesis is a combination of avian blood samples collected during my field season and through the generous contribution of additional samples from Chris Merkord (University of South Dakota), Jill Jankowski (University of British Columbia) and Christopher Trisos (University of Oxford), who surveyed birds along the same elevation transect as part of their own research projects.

The methods for calculating evolutionary uniqueness and morphological uniqueness (Chapters 2 & 3) were developed in collaboration with Christopher Trisos. In addition, Alex Pigot helped develop the range simulation approach used in Chapter 5.

My supervisors, Dr Nathalie Seddon, Dr Joe Tobias (both University of Oxford) and Olof Hellgren (University of Lund) helped develop ideas and provided comments on all of my chapters and Professor Robert Ricklefs (University of Missouri St Louis) provided editorial input and helped develop the ideas presented in Chapters 4 and 5.

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I'd also like to thank Dr Chris Merkord and Dr Jill Jankowski for their generous contribution of avain blood samples collected along the same study transect. These almost doubled my sample size and helped crystallize the results presented herein. I am very grateful to Professor Robert Ricklefs for offering excellent insights on many aspects of this research. The two separate visits I had to the Ricklefs lab were crucial in developing the ideas present herein, and I thank Bob and his entire lab group for their critical but constructive feedback, and for providing such a welcoming social atmosphere. To Dr Alex Pigot, thank you for your constant encouragement, critical appraisal, and willingness to answer an endless array of questions. I benefited greatly from research discussions with Alex and his excellent ability to synthesise current knowledge across a wide range of topics.

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Chapter 1

General Introduction

What explains the diversity of ecological communities?

Life is unevenly distributed across the surface of the Earth. While some areas support extremely high densities of species others are largely depauperate. For instance, a single 110 x 110 km quadrat in the Peruvian Andes may contain over 900 bird species (Hawkins et al. 2007), whereas a similarly sized area toward the poles may support only 40 species (Orme et al. 2005). Such disparities in diversity are repeated across taxa, on land and in the ocean, across latitude, depth and elevation (Gaston 2000). Although the existence of these diversity gradients has been recognized for centuries (Von Humboldt 1850, Darwin 1859, Wallace 1869), and have been increasingly well documented (Adams 2009, Hawkins et al. 2003, McCain and Grytnes 2010), the processes regulating species diversity continue to elude general explanation (Scheiner and Willig 2005, Rosenzweig 1992, Mittelbach et al. 2007, Currie et al. 2004).

Efforts to explain the factors regulating species diversity have focused largely on the role of climate and evolutionary history (Scheiner and Willig 2005, Gaston and Blackburn 2000, Willig et al. 2003, Jablonski et al. 2006). Many studies have reported strong correlations between community richness and aspects of the abiotic environment such as temperature, rainfall and productivity: at regional to global

scales these correlations typically explain >80% of the variance in species richness (Hawkins et al. 2003, Evans et al. 2005). The consistent relationships between the climate and richness have been taken as evidence that community diversity is regulated by the ecological capacity of the environment to support diversity (Francis and Currie 2003). However, the ecological mechanisms setting any limit to diversity remain poorly understood. Even in the absence of local limits to diversity, variation in the number of species co-occurring in any given place may arise due to larger-scale evolutionary and biogeographic processes (Ricklefs 1987). For instance, higher diversity in the tropics may be driven by faster rates of species diversification or because the greater antiquity of tropical climates has provided more time for communities to evolve (Wallace 1891). Studies of the fossil record and molecular phylogenies have revealed that over evolutionary timeframes the diversity of clades and regions may exist at or near a steady state (Ricklefs 2007). These results imply that the short-term ecological process regulating the diversity of local communities may 'feed-up' to determine these macroevolutionary dynamics (Rabosky et al. 2012). As a result, it is now widely recognized that a complete understanding of diversity gradients and community assembly will require approaches that bridge these highly disparate scales of both space and time.

The ecological mechanisms regulating diversity

Explanations for processes regulating the diversity of communities have largely focused on the role of competition in limiting species coexistence (Diamond 1978, MacArthur 1972, MacArthur 1969). For instance, at geographic scales species richness tends to increase with ecosystem productivity (Currie 1991). One prominent

explanation for this pattern is that more productive sites enable species to become specialized on a narrower range of resources and thus be 'packed' more tightly within communities (Klopfer and MacArthur 1961, Klopfer and MacArthur 1960). Alternatively, productive tropical communities may be more diverse because they contain a wider variety of resources allowing greater niche partitioning. Although the particular mechanism may vary, the key point under these traditional 'niche based' models is that diversity is limited because species that compete for the same resources cannot coexist (Gause 1934, Hutchinson 1961). In other words, the maintenance of diversity is regulated by the potential for ecological differentiation amongst species (Chesson 2000, Adler et al. 2007).

While ecological competition has dominated much thinking in community ecology, an alternative, and more controversial, theory has also been proposed in which natural enemies, and in particular parasites dictate broad-scale patterns in the distribution and diversity of species (Ricklefs 2010a, Ricklefs 2010b, Briers 2003, Hochberg and Ives 1999). Through reducing their hosts' intrinsic rate of increase, density, or condition parasites may potentially impact upon almost any aspect of their hosts' ecology and evolution (Holt and Lawton 1993). Recent studies have shown that as many as 40% of species in a habitat or ecosystem are parasitic on the remaining 60% that are free living (Dobson et al. 2008, Lafferty et al. 2006) and worldwide estimates suggest that parasitism may in fact be the most common animal lifestyle (Lafferty et al. 2006, De Meeûs and Renaud 2002). As an illustration of this, insects show high levels of parasitism and it is estimated that an average of 6 parasite species infect every insect host (Hawkins 2005) and that parasites are the dominant form of insect mortality (Cornell et al. 1998). These basic facts along with the global distribution of parasites

in all environments on earth, suggest that the role of parasites in regulating the structure and diversity of ecological communities may have been underestimated.

The role of parasites in species diversity

The possibility that parasites may hold the key to the problem of species diversity first gained widespread attention as an explanation for the extreme richness of trees in the tropical rainforests. Here, hundreds of species may co-occur on a single hectare plot but all these species compete for the same few limiting resources, something that is seemingly incompatible with niche based explanations for diversity. Any process that allows species to increase when rare, in what is termed the 'rare species advantage', can potentially lead to the maintenance of high species diversity by reducing the chance of population extinction. Janzen (1970) and Connell (1971) independently proposed that density dependent predation on seeds and seedlings (a form of parasitism) may explain the astounding diversity of tropical trees. According to the Janzen-Connell hypothesis, species are prevented from becoming so dominant that they exclude others because of specialized natural enemies. Although developed for plants and their pathogens, these effects may equally apply to herbivores and seed predators (Freckleton and Lewis 2006). In fact, the underlying assumption that specialized natural enemies cause density dependent mortality in their hosts, applies more broadly to host-parasite associations. If the abundance and degree of specialization in parasites is higher in the tropics then this may provide a general explanation for large-scale gradients in host-species richness, but begs the question of why parasite pressure itself varies across space.

In addition to promoting diversity by reducing species extinction, interactions between hosts and their parasites may also act to maintain higher richness via their effects on rates of speciation. New species are often formed following the chance colonization of a new island (either a true island or habitat island e.g. mountain top, lake etc) but many speciation events are likely to be prematurely terminated because low abundances following colonization increase the chance of population extinction. If population abundance is often regulated by parasites then the colonization of novel region where these parasites are absent can result in what has been termed 'enemy release', allowing populations of the host to rapidly increase and attain higher densities thus avoiding extinction (Keane and Crawley 2002). Another possibility is that parasites may regulate rates of speciation more indirectly via their effects on the evolution of traits that promote population divergence. For instance, Hamilton and Zuk (1982) showed that avian species with a higher parasite burden tended to have more striking sexual displays (i.e. 'male brightness', 'female brightness' and 'male song'). The mechanism behind this association is thought to reflect an honest signal of parasite burden, in which birds choose mates based on these condition-dependent traits. Although this theory remains controversial, stronger sexual selection may accelerate rates speciation providing a possible indirect effect of parasites on diversity (West-Eberhard 1989).

The Janzen-Connell and speciation rate hypothesis predict that parasites can promote diversity and in fact present no theoretical upper limit to the number of species that can be maintained. However, one other theory posits that parasites can also play a fundamental role in limiting diversity. Wilson (1961) developed the Taxon cycle model to describe the inferred pattern for species on archipelagoes to undergo

sequential phases of geographic expansion and contraction through time (Ricklefs and Bermingham 2002, Wilson 1961). He argued that new species arriving on an island would tend to drive the existing resident species to extinction, before themselves falling victims to a new wave of colonists, a process that would ultimately cap local diversity. The mechanisms driving the Taxon cycle remain obscure, but for birds occurring on islands in the West Indies, it has been speculated that new colonists may outcompete established residents due to their escape from fitness depressing parasites (Ricklefs and Cox 1972). This process thus includes elements of the enemy release hypothesis, but here acts to limit rather than enhance diversity. An alternative way in which the arrival of a new species may drive a resident extinct is if it carries a novel weapon in the form of a parasite that the resident cannot tolerate *sensu* 'apparent competition' (Holt 1977, Holt and Lawton 1994).

Under each of these scenarios, parasite may limit diversity by driving the geographic contraction and ultimately the extinction of resident species. However, these same processes may also act in the opposite direction. In particular, species may be prevented from expanding their range into new environments because of the presence of a competitor that is superior either because it is free from parasites (Colautti et al. 2004, Torchin et al. 2003), or because it carries a novel weapon (Callaway and Ridenour 2004, Prenter et al. 2004).

Unresolved questions

Current theory therefore predicts that parasites may act as a 'double-edged' sword, both promoting and limiting the origin and maintenance of species diversity. However, despite the long heritage of these ideas and countless studies on host-

parasite interactions, many of the most fundamental ingredients required to understand the role of parasites in regulating diversity remain unresolved. Here I highlight two broad research areas where our understanding is particularly limited.

First, the incidence of parasites among individuals and prevalence of diseases among species show widespread variation (Poulin 2011). For instance, among co-occurring bird species subject to avian malaria, the majority of species typically have few or no infections, whilst only a few harbour parasites at high prevalence (Scheuerlein and Ricklefs 2004, Hellgren et al. 2011). The factors responsible for this variation remain largely unknown. Indeed, it is not even clear whether patterns of parasite infection are generally predictable or whether they are instead largely idiosyncratic and context dependent. Explaining why some individuals or species suffer higher levels of infection than others has important consequences for models of how parasites either promote or depress richness in multi-species assemblages. For instance, if infection probability is largely dependent on population density then this would support models in which parasites promote coexistence by providing a rare species advantage. In contrast, if parasite infection simply reflects differences in host ecology then parasites may have a limited role beyond traditional niche-based explanations (e.g. resource competition) in structuring communities. Finally, because not all parasites are specialized on a single host species, the probability of infection may also depend on how parasites are shared amongst species and the composition of the wider community. Addressing how parasites are shared amongst hosts is therefore important in understanding how population density may affect parasite prevalence and how local host-parasite interactions may interact with broader-scale macro-evolutionary dynamics. Few studies have however considered the possible roles of chance,

ecology, environment and evolution in a single framework and so their consequences for parasite infection remain unclear.

Second, the role of parasites in limiting host geographic ranges remains mired in debate. Each species has a limited geographic distribution and understanding the factors that prevent species from expanding their ranges is therefore a key step in determining what regulates species diversity within any local community. A wide range of factors have been hypothesized to cause range limits (Hengeveld and Haeck 1982, Brown and Lomolino 1998, Lawton 1996) including physiological constraints, a reduction in resource abundance or quality (Andrewartha and Birch 1954), increased competition (Hall et al. 1992), predation (Holt et al. 2011), or parasitism (Hochberg and Ives 1999, Briers 2003). Of these, the role of parasites is particularly controversial. Theory suggests that parasites can prevent the expansion of species distributions but evidence of this from natural systems is scarce. A major reason for this is that most studies of parasite infection have been conducted over inappropriate geographical scales with data compiled from disparate studies; or through inadequate sampling of hosts and parasites throughout their entire geographic distribution. Thus while it is clear that parasites can suppress local host abundance the biogeographic consequences of this have been less easily determined.

The introduction of novel diseases by humans has provided a natural experiment to study the role of pathogens in setting range boundaries. For instance, following the arrival of avian malaria to Hawaii in the last century many native species, with no previous exposure to the disease, contracted their distributions to higher elevations where malaria is unable to persist (Atkinson et al. 2000, van Riper III et al. 1986).

Numerous other animals and plants have been shown to have similar effects when they have been introduced outside their native ranges. For example, the eastern grey squirrel (*Sciurus carolinensis*) introduced to the United Kingdom from North America habours a viral pathogen that is lethal in the native red squirrel (*Sciurus vulgaris*) (Tompkins et al. 2003). Similarly, signal crayfish (*Pacifastacus leniusculus*) also introduced into the United Kingdom from North America, are carriers of a water mould (*Aphanomyces astaci*) that is the causative agent of crayfish plague, which they can effectively resist, but which is highly lethal in the native white–clawed crayfish (*Austropotamobius pallipes*) (Alderman et al. 1990). Furthermore, worldwide introductions of chytrid fungus (*Batrachochytrium dendrobatidis*) have decimated many native frog populations (Kilpatrick et al. 2010), though some act as reservoirs for the disease and suffer little to no effect (Woodhams et al. 2007). However, whether these findings extend to natural communities with a potentially long history of host-parasite coevolution and counter-adaptation is unclear. Addressing this question in natural communities is therefore essential in order to understand the role of parasites in governing the composition and diversity of local communities more generally. Furthermore, given the prospect of rapid global climate change, understanding what limits species distributions is also a major conservation and management concern (Parmesan 2006, Adler and HilleRisLambers 2008, Parmesan and Yohe 2003).

Avian malaria along elevation gradients: a model system

Avian malaria

Haemosporidian (phylum Apicomplexa: Plasmodiidae) blood parasites offer an ideal system within which to explore host parasite interactions as they are an ancient and diverse group that infect a range of vertebrate hosts including mammals, birds and reptiles (Perkins and Schall 2002, Valkiunas 2005). Among the most well studied members of this group are those that cause malaria in humans (*Plasmodium falciparum* and *P. vivax*), which in 2010 were responsible for an estimated 219 million clinical cases and approximately 660 000 deaths (World Health Organization 2012). Avian malaria parasites have a long pedigree of use as model systems. Indeed, in 1902 Sir Ronald Ross received a Nobel Prize in Medicine for discovering the mosquito transmission of malaria using avian malaria as a model system (Marzal 2012). Increasingly these parasites are being used as a model system in evolutionary ecology and biogeography, and have formed the bases for a wide range of studies on community composition (Ricklefs et al. 2005, Loiseau et al. 2012), host-parasite coevolution (Ricklefs 2010a, Jenkins et al. 2012), life history theory (Knowles et al. 2010b), and patterns in the geographical distribution of species (Fallon et al. 2005).

Avian malaria parasites comprise an as yet unknown diversity of evolutionary distinct entities. Traditional microscopy methods of classification have recorded over 200 species belonging to four distinct genera, *Plasmodium, Haemoproteus, Leucocytozoon* and *Fallisia* (Braga et al. 2011), yet modern PCR based techniques are revealing a wealth of previously unrecorded lineages (Pérez-Tris et al. 2005, Fallon et al. 2003),

with some suggestions that there may be as many parasite species as there are host bird species (Bensch et al. 2004).

These parasites have a two-host life cycle in which blood-feeding arthropod vectors transmit the parasite to hosts. Recent work suggests that host compatibility rather than vector host encounter rates determine the host range of avian malaria (Medeiros et al. 2013). The primary vectors of *Plasmodium* are mosquitoes belonging to the genera *Culex* and *Aedes*, *Haemoproteus* is transmitted by biting midges (genera *Culicoides*) and louse files (genera *Ornithomyia*), and vectors of *Leucocytozoon* are blackflies of the genera *Simulium.* All haemosporidian parasites undergo a two-staged life cycle with sexual stages and sporogony occurring in the definitive host (vector) and merogony and development of gametocytes occurring in vertebrate host (Figure 1). Given the intimate association of avian malaria blood parasites with their hosts across evolutionary time periods (Anderson and May 1982, Hafner and Nadler 1988, Ricklefs 2010a, Jenkins et al. 2012), sensitivity of insect vectors to environmental conditions (Wilson 2001, Freed et al. 2005, Githeko et al. 2000), and varying degrees of pathogenicity (Atkinson et al. 2000, Martinez-de la Puente et al. 2010, Marzal et al. 2005); this group of haemosporidians provides an ideal system within which to address a range of ecological and evolutionary questions pertaining to the importance of biotic interactions in shaping community composition.

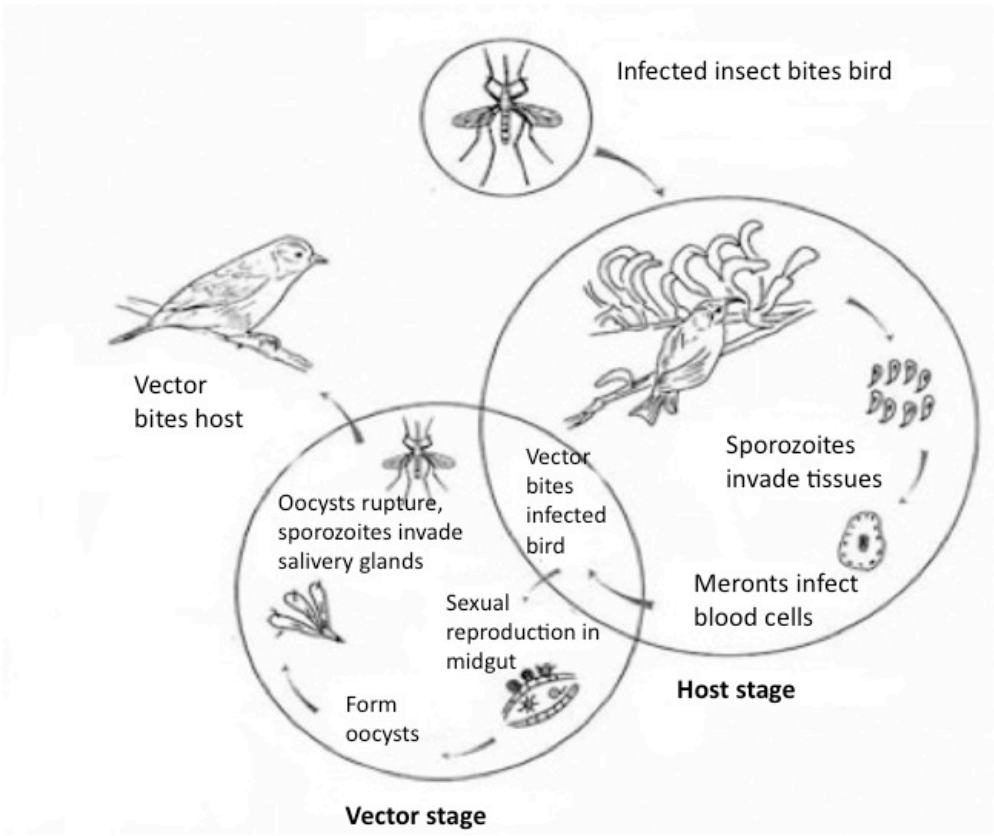


Figure 1.1 The complex general lifecycle of avian haemosporidian parasites begins with a sexual phase within the midgut of the insect vector where male microgametes and female macrogametes meet and fertilization occurs, forming a diploid ookinete which then migrates to the gut wall and undergoes asexual reproduction, forming oocysts that rupture and release sporozoites that enter the salivary glands via the haemocoele. When an infected insect vector bites an avian host, sporozoites enter the host's blood stream during feeding, and invade various host tissues such as parenchymal cells in the liver, endothelial cells in the kidneys or macrophages. Merogony takes place in tissue cells for *Haemoproteus* and *Leucocytozoon* genera, and within erythrocytes in *Plasmodium.* Within host blood cells, gametocytes of all three genera grow and develop and can displace the host cell's nucleus, in the case of *Leucocytozoon* infections often giving the cell a 'fusiform' appearance. At this stage the parasite are infective and the cycle continues when an insect vector bites an infected host, repeating the cycle. Figure adapted from (Friend and Franson 1999) Page 193.

Tropical elevation gradients

Tropical elevation gradients provide a natural laboratory in which to test the role of avian malaria, and indeed parasites more generally, in driving patterns in species diversity. Steep gradients in environmental conditions (e.g. temperature decreases by an average of 0.6°C per 100 m increase (Barry 2008)), result in dramatic differences in species diversity over relatively small spatial scales, allowing hosts and their parasites to be sampled under strongly contrasting biotic and abiotic conditions. Moreover, many host species have extremely narrow elevation ranges often only a few 100's of m wide (Walker et al. 2006, Schulenberg et al. 2010). This provides a unique opportunity to study how the incidence of parasite infection varies across the entire geographic distribution of the host, a task that would be prohibitive under normal conditions where host ranges may be 1000's of km in extent. Finally, the extremely high diversity of tropical gradients, while presenting a challenge, also ensures sufficient power for large-scale comparative analysis of the predictors of parasite infection (Ricklefs et al. 2005, Fecchio et al. 2011).

The study system

The study was conducted in the Kosñipata valley of southeastern Peru, in the department of Cusco (Figure 1.2). All sites were located along an elevation transect traversing the eastern slope of the Andes in a region of mostly undisturbed primary forest (Figure 1.3). Sites along this transect were accessed via the Manu road, an unsealed dirt road linking the Andean highlands to the Amazonian lowlands. The elevation transect was bound at the top by Wayqecha Research Station at 3120 m asl and at the bottom by Tono, a site within Manu National Park at 879 m asl. Along this gradient, five key sites centered around elevations of 930 m, 1130 m, 1325 m, 1965 m, and 2908 m were used as bases for bird surveys (Table 1.1). Apart from the lowland area surrounding Tono, the terrain in this region is extremely precipitous and surveys were therefore limited to locations that could be accessed at least a short distance off the main road. This region has one of the highest levels of bird diversity recorded for any area with over 1000 species between 250 and 4000 m (Walker et al. 2006), yet no previous study has examined parasite prevalence in this avifauna or the impact of parasites on patterns of species richness.

Figure 1.2 Map of Peru showing location of Wayqecha research station (listed as cloud forest programs (a), and the location of the study transect along Manu national Park (b). Image credits: World Atlas of Panoramic Aerial Images and Amazon Conservation Association (ACA); http://www.biosci.missouri.edu/avianecology/kosnipata/

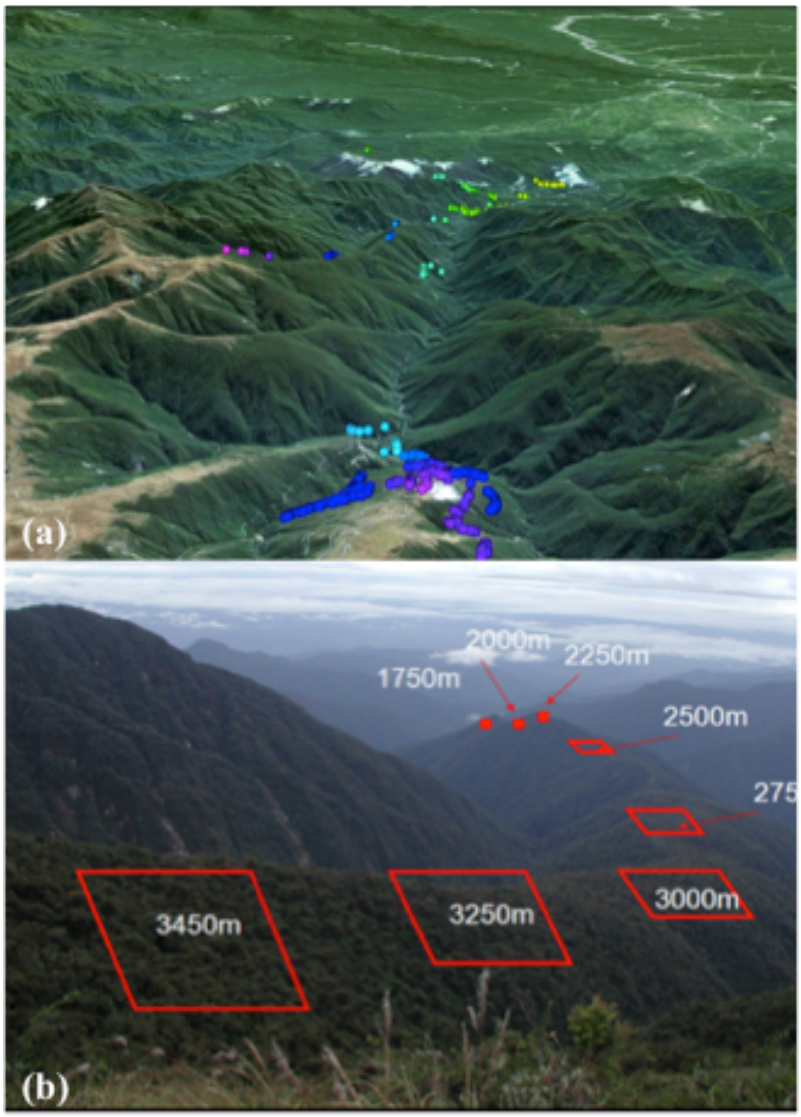


Figure 1.3 Aerial view of mist netting locations along the Kosñipata Valley, Peru (a) and the locations of remote weather stations along the Kosnipata Transect (b). Image credits: Google Earth and photo by Kenneth J. Feeley on http://news.mongabay.com/2009/0923-hance_feeley.html

Table 1.1 Description of key study sites in the Kosñipata Valley, Peru spanning 800 m – 3500 m. In addition to mistnetting birds at these locations, surveys were also conducted at elevations above and below these points to provide fine-scale continuous sampling from along the entire gradient.

Site	Base Camp elevation; Co-ordinates	Elevational band	Annual precipitation (mm yr^{-1})	Mean annual temp (°C)	Forest type [a]	Vegetation description [a]
Tono	930 m S12 57.431 W71 34.010	879 m - 961 m	3087	20.7 ± 0.02	Submontane rainforest	Tall canopy; well-structured forest; Moraceae, Fabaceae and Rubiaceae dominant tree families; tree ferns and bamboo (*Guadua* and *Chusquea* spp.) abundant; moderate amounts of moss on trees and forest floor
Quitacalzon	1130 m S13 02.123 W71 30.737	1122 m - 1155 m				
San Pedro	1325 m S13 03.252 W71 32.292	1302 m - 1402 m	2631	18.8 ± 0.02	Cloud forest	Moderate canopy height; Euphorbiaceae, Fabaceae and Lauraceae dominant tree families; aboreal and ground ferns abundant; moderate amounts of moss on trees and forest floor
Rocotal	1965 m S13 06.070 W71 34.127	1908 m - 1986 m				
Wayqecha (incl. river)	2908 m S13 10.549 W71 35.159	2171 m – 3120 m	1706	12.5 ± 0.05	Treeline cloud forest	Medium-low canopy; *Weinmannia* and *Clusia* dominant tree genera; arboreal bromeliads > 5 m, arboreal and ground ferns, and moss on trees and floor abundant; moderate amount of bamboo (*Chusquea* spp.) in middle story

[a] Data from Palin et al. (2011)

General methodology

Bird surveys

Following previous studies birds were sampled using mist nets (Terborgh 1971). These were set up at ground level and used to sample birds flying through the undergrowth of undisturbed mature forest and edge habitats bordering landslides, path or road edges. At each site a maximum of 15 nets were set approximately 40 m apart and within a 50 m wide elevational band. Individual nets were placed along narrow trails or along lines cut into the surrounding undergrowth at locations aimed at capturing the local diversity of the site. Nets were typically opened a few minutes before sunrise and closed at sunset. Whilst open, nets were checked at least every 20

minutes and captured birds brought were brought back to a central ringing station. Captured birds had the following measurements recorded: mass, wing length, kips distance, tarsus length, bill depth, bill width, and bill length. Approximately 100 – 150 µl of blood was also collected by wing venipuncture of the brachial (ulnar) vein for parasite analysis and stored in 100% ethanol before birds were finally banded with a unique coded ring and released at the site of capture.

Parasite screening and sequencing

Total DNA was extracted from whole blood using a standard ammonium acetate procedure with ethanol precipitation (Bruford et al. 1998). The final extraction products were diluted in low TE Buffer (10 mM Tris, 0.1 mM EDTA). Quantifications of DNA from host blood samples (parasite and host DNA) were performed with an 8-well NanoDrop (Thermo Scientific ND-8000). Quantifications were based on overall amount of host and parasite DNA, the larger concentrations of which increase the likely amount of parasite DNA within the sample. Previous studies have found that that as little as 2 ng μl^{-1} of host genomic DNA is sufficient to detect infections of *Plasmodium circumflexum* (Knowles et al. 2010b), so this was used as the minimum concentration for subsequent PCR. The average concentration of DNA in each blood sample was 110 ng/µl (±SE 5 ng/µl).

Primers used during this investigation are those described by Hellgren et al. (2004) and were designed using published sequences of avian *Plasmodium, Haemoproteus* and *Leucocytozoon* mitochondrial DNA (mtDNA) by Perkins and Schall (2002) within the 5' end of the cytochrome *b* gene of the parasite's mitochondrial genome. The initial primers used for PCR amplification of parasite mtDNA of *Haemoproteus,*

Plasmodium and *Leucocytozoon* lineages were HaemNFI [5'-CATATATTAAGAGAAITATGGAG-3'] and HaemNR3 [5'-ATAGAAAGATAAGAAATACCATTC-3'] where I represents the universal base inosine. For the second round of PCR reactions HaemF [5'-ATGGTGCTTTCGATATGCATG-3'] and HeamR2 [5'-GCATTATCTGGATGTGATAATGGT-3'] were used for *Plasmodium* and *Haemoproteus* lineages and HaemFL [5'-ATGGTGTTTTAGATACTTACATT-3'] and HaemR2L [5'-CATTATCTGGATGAGATAATGGIGC-3'] for *Leucocytozoon* lineages. Using these primers I amplified a 416 bp region of the parasite mitochondrial cytochrome *b* gene.

PCR reactions were performed with 2 µl of each sample extraction in an 8 µl nested PCR that comprised of 1µl ddH_20, 1 µl primer mix (NFI, NR3), 2 µl of extraction, and 4 µl of Qiagen Mastermix. This first PCR with primers HaemNF1 and HaemNR3 was run under the following conditions: initial incubation for 15 minutes at 95°C, and then 20 cycles beginning with 30 s at 94°C, 30 s at 50°C, and 45 s at 72°C with a final 10 m at 72°C for annealing. For the second round PCR reactions, the round 1 PCR product was diluted with 8 µl of ddH_20, and 2 µl of this diluted product was used of for each successive reaction amplifying either *Leucocytozoon* (HaemFL and HaemR2L primers) or *Plasmodium/Haemoproteus* (HaemF and HaemR2 primers) lineages. These reactions were performed under the same conditions as the first round PCR.

To determine if PCR products of parasite DNA were amplified, round 2 PCR products were diluted with 8 µl of ddH_20 and 4 µl of this was added to 6 µl Orange G

(2x) for loading into a 1.5% agarose gel containing 25 µl SyberSafe. A 1kBP ladder was loaded into each gel and standard positives for both *Plasmodium/Haemoproteus* and *Leucocytozoon* from a previous study of avian malaria infection conducted on blue tits in Bagley wood, Oxfordshire by Knowles et al. (2010b). Any samples that had clear bands were taken as positives and the absence of band as negatives. PCR products were sent away for single-direction sequencing at LGC Genomics Berlin, Germany and if the initial sequence didn't work or was unclear, the sample was re-sequenced using the reverse primer.

Evaluation

The obtained sequences were edited and aligned in Sequencher® version 4.2. Samples with positive amplification tended to produce DNA fragments of approximately 480 bp in size. The aligned and edited sequences (416 bp) were exported to BioEdit, version 7.0.9.0 for Windows (Hall, 1999), file containing all known sequences of malaria lineages (MalAvi database 21.3.11). MalAvi is a public database of malaria parasites and related haemosporidians in avian hosts based on mitochondrial cytochrome *b* lineages produced by Lund University (Bensch et al. 2009).

All closest identified sequences were checked by aligning each sequence individually in BioEdit (Hall 1999) to assess changes in base pairs. These novel lineages were named after the host species from which they were found according to the standard British Trust for Ornithology nomenclature. For example, (P MIOST 1) would denote the first new *Plasmodium* infection found in *Mionectes striaticollis.*

Designation of parasite lineages

Parasite lineages were defined based on any variation in the 416 bp region of the cytochrome b gene. Previous studies have revealed that lineages differing by <1 % sequence divergence often have unique host associations (Bensch et al. 2000, Ricklefs and Fallon 2002, Bensch et al. 2004). For instance, Reullier et al. (2006) found that two avian malaria lineages which differ by 0.2% at the cytochrome b gene, are almost entirely restricted to one bird species each, even though transmission between them is possible. In this study, parasite lineages belonging to either *Plasmodium* or *Leucocytozoon* were readily identifiable, however within the genus *Haemoproteus* (Haemosporida, Haemoproteidae), subgenera may include both *Haemoproteus* and *Parahaemoproteus*, which are both generally accepted within the broader genus (Krizanauskiene et al. 2013).

For samples that contained mixed lineage infections identifiable as 'double base callings', if possible, the dominant lineage in the sample was resolved (Hellgren et al. 2008) otherwise the sequence was discarded (Figure 1.4). All avian malaria sequences recorded in this study have been deposited in GenBank (Accession numbers KF874666 – KF874818, Appendix 2 Table 5.7).

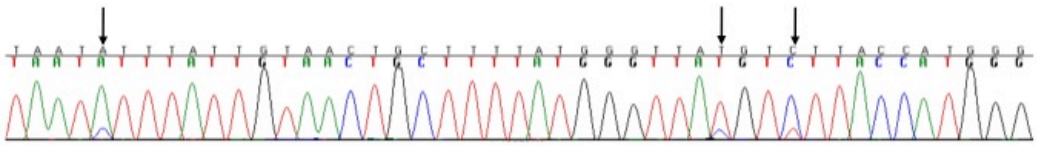


Figure 1.4 Chromatogram of mixed lineage infections with arrows showing the 'double base callings'. In this instance, the dominate bases from left to right are 'A', 'T' and 'C' although at these same positions 'C', 'A' and 'T' can also be detected as smaller peaks.

Bird Phylogeny

The incidence of parasite infections in host bird species may result from phylogenetically non-independent processes such as co-speciation in which closely related hosts share a parasite inherited from a common ancestor, greater host switching among close relatives, or through greater extinction of parasites from more distant hosts (Clayton and Moore 1997). To control for phylogenetic relatedness among the resident birds species I used a recently published time calibrated avian tree based on the Hackett backbone phylogeny (for full details see Jetz et. al (2012)). Prior to analysis the tree was pruned to contain only the 245 bird species that were sampled and tested for avian malaria in this study. To account for uncertainty in phylogenetic relationships I repeated all analysis (except where indicated in the text) across 100 trees randomly selected from across the posterior distribution of trees (Jetz et al. 2012).

Parasite phylogeny

I used sequence data from the 416 bp region of parasite mitochondrial cytochrome b gene for all parasite lineages recorded from resident birds to create a parasite phylogeny. I used a lognormal relaxed clock (uncorrelated) model in BEAST (Drummond and Rambaut 2007), and conducted two runs each using a GTR+G+I model of sequence evolution, with estimated base frequencies and a Yule prior on branch lengths. Each chain was run for 20 million generations, sampling every 2000 with the first 10% (i.e. 9000) trees removed as burn-in. The two separate runs were combined with TreeAnnotator version 1.7.2. (Rambaut and Drummond 2012). Trees were rooted with other vertebrate malaria parasites downloaded from genbank (Figure 1.5).

Figure 1.5 Evolutionary tree of 144 avian malaria parasite lineages made up of 19 *Plasmodium* spp., 34 *Haemoproteus* spp. and 91 *Leucocytozoon* spp. recorded from 245 resident bird species in the Kosñipata Valley SE Peru, and including common outgroup lineages recorded from birds and mammals, downloaded from genbank.

Chapter 2

The influence of host and environment on disease prevalence in montane tropical birds

Abstract

Understanding what determines variation in the distribution of pathogenic organisms among individuals and species is a core aim of ecology and evolutionary biology. However, there is great uncertainty about the relative importance of the host's intrinsic ecology, environment and evolutionary history. Here I use data on haemosporidian infections in birds from a hyperdiverse elevation gradient in the Peruvian Andes to test the relative importance of these factors. First I show that variation in prevalence amongst species and across elevations is non-random, and second, find support for a model in which infection depends on the combined effects of intrinsic host characteristics, environment and evolutionary uniqueness. Body mass was positively correlated with overall and *Leucocytozoon* infections, and capture elevation was negatively correlated with *Haemoproteus* infections but positively correlated with *Leucocytozoon* infections. The most consistent finding is that among co-occurring bird species, overall haemosporidian infections, as well as *Plasmodium* and *Leucocytozoon* infections, decline with increasing evolutionary uniqueness, controlling for differences in host ecology and environment. For individuals as well,

infection prevalence was best predicted by evolutionary uniqueness relative to all other co-occurring species. This study provides the first evidence that host ecology, environment, and in particular evolutionary history determine infection prevalence in avian communities, with implication for understanding the current and predicted future distributions of diseases.

Introduction

Parasites exhibit heterogeneous distributions among hosts (Poulin 2011) and among co-occurring species, the majority of species often have few or no infections, whilst only a few harbour parasites at high prevalence (Scheuerlein and Ricklefs 2004, Hellgren et al. 2011). Identifying the factors responsible for this variation is important for a range of topics in biology, from understanding mechanisms of species co-existence and geographical range limits, to determining the current and future distributions of disease organisms. However, while multiple factors have been invoked (Scheuerlein and Ricklefs 2004, Garvin and Remsen 1997, Ricklefs et al. 2005, Loiseau et al. 2012, Krasnov et al. 2010), their relative importance is not well understood (Grenfell and Dobson 1995, Nunn and Altizer 2006, Schrag and Wiener 1995).

A key hypothesis for variation in host infection identifies the importance of environmental conditions. Previous authors have found that environmental differences are important predictors of the similarity of parasite faunas among hosts (Krasnov et al. 2010). Temperature in particular is known to play a key role in influencing exposure to parasites, particularly for diseases transmitted between hosts by arthropod

vectors (Zamora-Vilchis et al. 2012, Keyghobadi et al. 2006, LaPointe et al. 2010). This is because temperature is a key determinant of both vector abundance (Lindsay and Birley 1996) and parasite developmental rates (LaPointe et al. 2010, Wilson 2001).

Intrinsic host characteristics such as foraging strata, diet, body mass and range size offer a complimentary set of explanations for differences in prevalence among species. These factors are generally thought to influence prevalence by altering the probability that an infective parasite will contact a susceptible host. At geographical scales parasite prevalence is generally linked to the distribution and abundance of vectors (Scheuerlein and Ricklefs 2004, Greiner et al. 1975) and at local scales, habitat features such as vertical stratification of nesting sites (Garvin and Remsen 1997) or distance to permanent water bodies where vector larval stages occur (Wood et al. 2007) are also known to be important. Accordingly, in birds, foraging data and diet may influence exposure to parasites. Body size has also been identified as a potential correlate of parasite prevalence, as it adequately identifies the host as an insular habitat in which larger hosts may be more susceptible to infection (Kuris et al. 1980). Large geographic range size of a species is also expected to promote infection if more widely distributed species have more exposure to parasites (Gregory 1990).

In addition to intrinsic traits, the chance of infection may depend on local community composition. In particular, the evolutionary (phylogenetic) uniqueness of a focal species relative to all other species it co-occurs with may be important. This is because a host species that is closely related to other community members is more likely to share characteristics that influence parasite development, infection and

transmission (Combes 2004), leading to amplification effects in prevalence in certain host species (Dobson 2004). Conversely, distantly related host species are more likely to have different ecological, behavioural and physiological characteristics that may act as encounter and compatibility filters to parasite host range (Combes 2004). Most previous studies investigating a role of host evolutionary uniqueness have been aimed at predicting the likelihood of different host species sharing parasites. These have often been carried out at local scales which may reflect site specific patterns and be limited by too few host parasite associations (Gilbert and Webb 2007), or at regional scales which are based on less well resolved host parasite associations due to infection data being collated from non-standardized surveys (Krasnov et al. 2010, Davies and Pedersen 2008, Cooper et al. 2012). Additional experimental studies often use novel host parasite associations that are unlikely to occur under normal circumstances (Gilbert and Webb 2007, Perlman and Jaenike 2003, Longdon et al. 2011). Therefore, although previous research supports a role of host evolutionary uniqueness in predicting the likelihood of parasite sharing among hosts, its importance for predicting a host's overall incidence of infection and likelihood of sharing among naturally co-occurring species has not previously been well tested.

Difficulties in adequately sampling ecologically and evolutionarily distinct hosts from across a range of environmental conditions have made it difficult to resolve the importance of each of these characteristics toward influencing infections among co-occurring species. To overcome this, I collected blood samples from resident bird species along an elevation gradient on the eastern slope of the Andes Mountains in Peru and tested these for haemosporidian (malaria) infections (*Plasmodium, Haemoproteus, Leucocytozoon*) to investigate the importance of host ecology,

environment, and evolutionary uniqueness in influencing infection. Haemosporidian parasites are a suitable system within which to investigate hypotheses about what determines infection risk, as they occur across a wide geographical range and have been recorded from almost all bird families (Valkiunas 2005). These parasites have a two-host life cycle in which blood-feeding arthropod vectors transmit the parasite to vertebrate hosts, within which the parasites exhibit specialist or generalist host distributions depending on the extent of host parasite co-evolution and virulence resistance. Specifically I test the importance of intrinsic host characteristics including: foraging strata, diet, elevation range size, body mass; environmental characteristics: using elevation as a proxy for changes in temperature and moisture (Barry 2008); and evolutionary characteristics: using evolutionary uniqueness; in explaining the observed incidence of infection.

Material and Methods

Host characteristics

For each species sampled, I quantified variation in four key host characteristics hypothesised to influence likelihood of infection: (i) body mass, (ii) range size, (iii) foraging strata, and (iv) primary diet. Body mass (g) was obtained from Dunning (2008) and is based on average species values; ecological data (i-iv) were collated from Stotz et al. (1996) with updates and amendments from recent literature (Belmaker et al. 2012). I classified foraging strata into three categories: (1) Forest understory, (2) Shrub/grassland and (3) Forest canopy. Species were assigned to one of three dietary categories representing their primary adult food source: (1) Omnivore (more than one type of major food source, e.g., fruit and insects), (2) Insectivore

specialist (although this category includes a small number of carnivores (*Glaucidium jardinii*) and piscivores (*Chloroceryle aenea*) and (3) Plant specialist (including granivores, frugivores and nectarivores). Elevation limits of bird species are those presented in Walker et al. (2006) and are based on detailed site, sounds or sign (i.e. nest) records along the study gradient and in surrounding areas. Temperature has previously been identified as key environmental variable influencing the prevalence of avian malaria (Zamora-Vilchis et al. 2012), and because temperature changes predictably with elevation according to the adiabatic lapse rate (Barry 2008) by including capture elevation in the model I indirectly tested the importance of temperature in shaping observed patterns.

Evolutionary uniqueness

To calculate the evolutionary uniqueness of the 245 resident bird species recorded in this study, I used a recently published tree [for full details see Jetz et. al.(2012)]. 100 trees from the posterior distribution were downloaded from (www.birdtree.com) and pruned to contain all the 1002 resident bird species recorded along the study gradient based on Walker (2006). For each tree I calculated each of the 245 sampled bird species evolutionary uniqueness based on the average patristic branch length distance between these and all other species that overlapped their elevation range. Haemosporidian parasite lineages have the potential to occur across a broad host range with numerous lineages having been recorded from hosts belonging to different orders (for a summary see: http://mbio-serv2.mbioekol.lu.se/Malavi/), so by calculating host evolutionary uniqueness relative to all other co-occur bird species, this takes into account the potential of parasites to infect diverse hosts.

Phylogenetic signal

I tested for phylogenetic signal in binary host traits including diet (Insectivore, Omnivore, Plant specialist) and foraging strata (Canopy, Scrub/grassland, Forest understory) using the methods described in Fritz & Purvis (2010). To test for phylogenetic signal in continuous host traits including body mass and range size, I estimated Blomberg et al.'s K (Blomberg et al. 2003). I ran each of these estimates on the same 100 phylogenetic trees used in the evolutionary uniqueness analyses and report averages from all these runs.

Statistical analyses

To determine whether the observed variation in parasite prevalence could be explained by chance alone I tested whether the distribution of prevalence across species departed from a null model in which infection occurred independently of host species. To do this I maintained the observed number of infections and randomly shuffled these across hosts. I then recorded the number of species within prevalence quartiles (i.e. >0-25%, >25-50%, >50-75%, >75-100%) and repeated this procedure 100 times to generate expected distribution of prevalence (median, 95% confidence intervals) (Figure 2.1). I repeated this for each of the four types of infection classification: Overall Infection, *Plasmodium*, *Haemoproteus* or *Leucocytozoon* infections.

To evaluate the relative importance of intrinsic, extrinsic and evolutionary factors on the incidence of infection I used Markov Chain Monte Carlo estimation implemented with "MCMCglmm" (Hadfield 2010). I ran all models across the same 100 separate bird phylogenies used to calculate evolutionary uniqueness to control for phylogenetic

signal. I entered the infection status of individual birds as a binary response variable with family specified as "categorical" and included birds elevation range size, primary diet, foraging strata, body mass, capture elevation, and evolutionary uniqueness as explanatory variables. Four separate models were run for each category of infection. All analyses were conducted on a restricted dataset of only resident bird species, as these are more likely to have been infected close to their point of capture than species classified as vagrant, boreal migrant, or austral migrant, which by definition exhibit greater movement within their ranges. All analyses were run across each of the 100 phylogenetic trees with the following parameters: burnin = 10000, thin = 100, iterations = 110000; and with priors specified as: V = 1, nu = 0.002, and fix = 1 (because the variance cannot exceed 1 with binomial data) see Hadfield (2010) for further details. Because minimum adequate models can inflate Type I errors, I report the average results of the full models and include the change in the deviance information criterion (DIC) between the full model and the model excluding the predictor as an additional measure to help assess the relative importance of predictors.

I had no *a priori* reason to choose a particular parametric form to describe the relationship between capture elevation and prevalence, so to graphically show the fit of a simple logistic model of infection explained by capture elevation, I fitted a generalized additive model with capture elevation entered as a smoothed term and infection status as a binary response variable (Figure 2.2).

Results

Form and distribution of infection

A total of 2188 resident birds were captured along the elevation gradient and among these I recorded 570 (26%) overall infections (i.e. any of the three parasite genera), 109 (4.9%) *Plasmodium* infections, 236 (10.8%) *Haemoproteus* infections, and 311 (14.2%) *Leucocytozoon* infections. The observed number of species with infection prevalence within each of the four arbitrarily defined categories differed significantly from that expect by chance, as shown by the empirical number of species in each prevalence category falling outside the 95% confidence intervals when infections were randomised (Figure 2.1).

The number of species with overall and *Leucocytozoon* infections in the 0-25% prevalence category was significantly more than expected by chance, whereas in the 25-50% prevalence category the number of species with overall, *Haemoproteus* and *Leucocytozoon* infections was significantly less than expected by chance. Only for overall infections were there significantly more species infected in the 75-100% prevalence category. These results show that infections are not randomly distributed among species.

The diversity of lineages within each of the parasite genera also showed widespread variation. In total I recorded 19 *Plasmodium* lineages, 34 *Haemoproteus* lineages and 91 *Leucocytozoon* lineages from the 245 resident bird species recorded in this study.

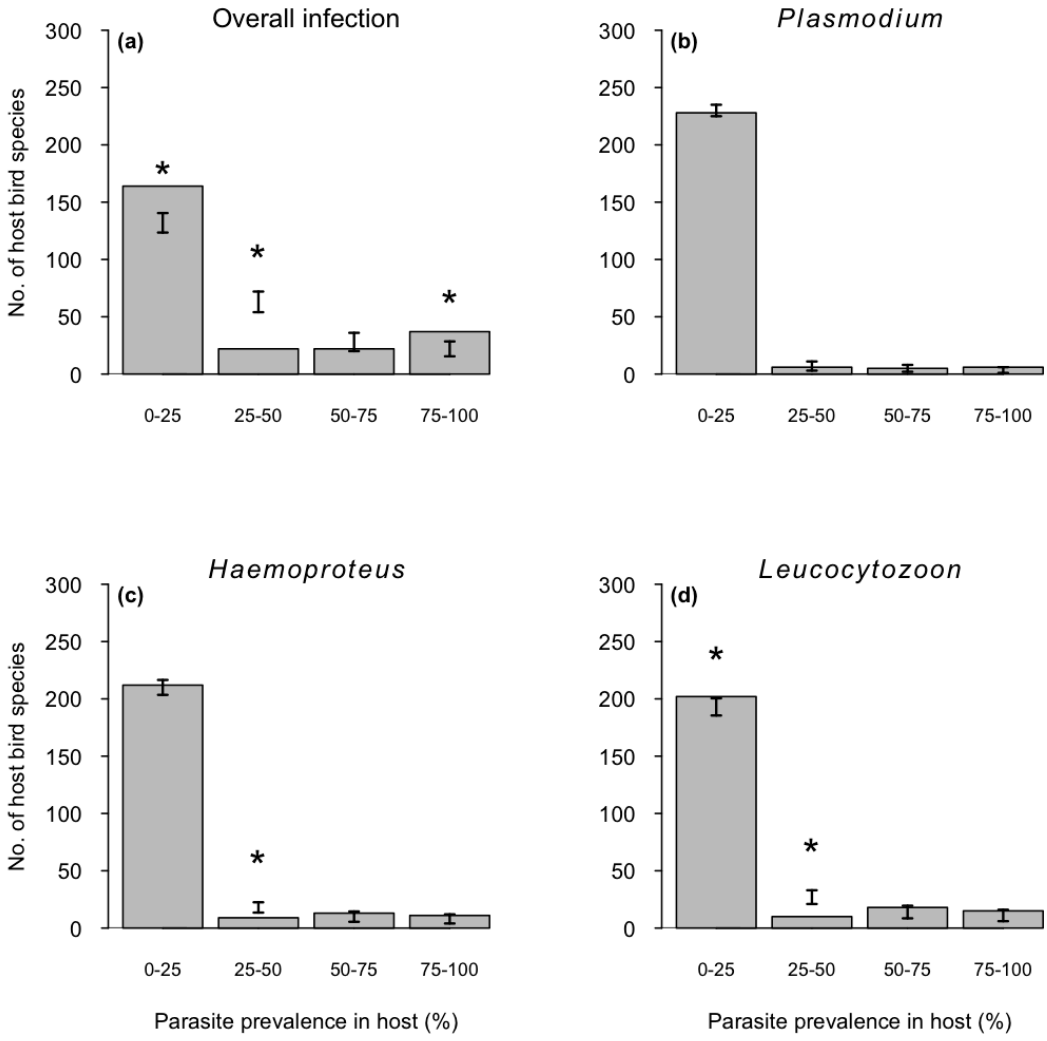


Figure 2.1 The number of infected bird species with avian malaria prevalence within each of four arbitrarily defined categories, for (a) Overall infections (any of the three parasite genera), (b) *Plasmodium* infections (c) *Haemoproteus* infections and (d) *Leucocytozoon* infections. The lines show the 95% confidence intervals around the expected number of species in each prevalence category based on 100 random simulations of the empirically recorded number of infections of each infection type. Bars represent the empirical number of species in each prevalence category and asterisks (*) represent a significant difference between the empirical and random number of species in each category.

Multi-predictor models of infection

I found that body mass was significantly associated with an increased incidence of overall (P_{MCMC} = 0.013) and *Leucocytozoon* infections (P_{MCMC} = 0.038), but was not an important predictor of *Plasmodium* or *Haemoproteus* infections ($P > 0.05$; Table 2.1). In other words, as species body mass increased, so did the incidence of overall and *Leucocytozoon* infections. Of the additional intrinsic host characteristics investigate here (i.e. range size, primary diet and foraging strata), none of these were found to be associated with the overall, or genera-specific incidence of infections among species ($P > 0.05$, Table 2.1).

The importance of elevation in explaining the incidence of infection showed considerable variation across each of the four infection classifications. There was no significant effect of elevation on the incidence of overall or *Plasmodium* infections ($P > 0.05$). However, the incidence of *Haemoproteus* infections declined significantly (P_{MCMC} = 0.003) with elevation, whereas *Leucocytozoon* infections increased significantly (P_{MCMC} = 0.001, Table 2.1).

Finally, I found that a bird species' evolutionary uniqueness from co-occurring bird species was strongly and significantly associated with overall haemosporidian infections (P_{MCMC} = 0.001), *Plasmodium* infections (P_{MCMC} = 0.006) and *Leucocytooon* infections (P_{MCMC} = 0.001), but not *Haemoproteus* infections ($P > 0.05$). In all significant models, the slope was negative, indicating that the more evolutionarily distinct a species was, relative to all other co-occurring species, the lower its level of infection (Table 2.1). When evolutionary uniqueness was calculated for individual bird species relative to all other co-occurring species at their capture

elevation, greater phylogenic uniqueness was still the most significant variable associated with reduced prevalence of overall haemosporidian (P_{MCMC} = 0.010) and *Plasmodium* infections (P_{MCMC} = 0.008), but not for *Haemoproteus* or *Leucocytozoon* infections (both P_{MCMC} = 0.05), Table 2.2.

Table 2.1 Effects of host species ecology, environment and evolutionary uniqueness from co-occuring species on overall and genera-level infections.

Overall Infection	**Slope**	**Lower-CI**	**Upper-CI**	**ess**	***P*-value**		**Δ_{DIC}**
Intercept	19.938	7.724	32.585	190	0.002	*	
Elevation range	0.000	-0.001	0.001	959	0.827		-0.79
Omnivore	0.707	-0.489	1.890	1000	0.246		-0.65
Plant specialist	1.024	-0.281	2.328	994	0.132		-0.65
Shrub/Grassland	-0.810	-1.930	0.304	986	0.157		-0.59
Forest understory	-0.003	-0.796	0.789	987	0.899		-0.59
Body mass	0.015	0.004	0.027	959	0.013	*	-3.06
Capture elevation	0.000	0.000	0.000	804	0.624		0.05
Evolutionary uniqueness	-0.160	-0.243	-0.080	285	0.001	*	12.03
Plasmodium							
Intercept	16.959	-0.819	35.507	122	0.061		
Elevation range	0.0002	-0.001	0.001	583	0.737		-1.71
Omnivore	1.099	-0.743	2.944	485	0.235		1.08
Plant specialist	0.791	-1.205	2.872	443	0.454		1.08
Shrub/Grassland	-0.368	-2.084	1.341	639	0.673		0.92
Forest understory	-0.100	-1.343	1.144	718	0.860		0.92
Body mass	0.015	-0.001	0.030	820	0.070		-0.96
Capture Elevation	-0.0005	-0.001	0.0001	612	0.105		-0.10
Evolutionary uniqueness	-0.165	-0.295	-0.042	157	0.006	*	2.48
Haemoproteus							
Intercept	8.083	-9.573	26.187	115	0.378		
Elevation range	0.000	-0.001	0.001	678	0.908		0.16
Omnivore	1.049	-0.744	2.836	715	0.253		-1.09
Plant specialist	1.562	-0.387	3.504	723	0.118		-1.09
Shrub/Grassland	-0.291	-2.156	1.582	607	0.766		-0.21
Forest understory	0.400	-0.836	1.675	777	0.544		-0.21
Body mass	0.004	-0.014	0.022	853	0.639		2.25
Capture elevation	-0.001	-0.002	-0.0003	480	0.003	*	9.66
Evolutionary uniqueness	-0.091	-0.210	0.025	186	0.130		3.09
Leucocytozoon							
Intercept	25.231	6.364	44.558	118	0.006	*	
Elevation range	0.001	0.000	0.001	753	0.184		2.75
Omnivore	0.509	-1.110	2.111	830	0.536		1.57
Plant specialist	1.215	-0.492	2.954	834	0.165		1.57
Shrub/Grassland	-1.406	-2.832	0.008	932	0.052		1.11
Forest understory	0.044	-0.990	1.086	934	0.874		1.11
Body mass	0.016	0.002	0.030	912	0.038	*	-3.88
Capture elevation	0.001	0.001	0.002	732	0.001	*	-0.79
Evolutionary uniqueness	-0.241	-0.378	-0.113	166	0.001	*	16.31

Overall infection DIC=1840.17, Plasmodium DIC=696.63, Haemoproteus DIC=950.89 and Leucocytozoon DIC=1179.02. Values refer to the average model output from models run on 100 phylogenies of the 245 bird species. ΔDIC is the change in DIC when the term is removed from the full model run on a single phylogenetic tree. * P < 0.05. ess= effective sample size, CI=confidence interval

Table 2.2 Effects of an individual bird species ecology, environment and evolutionary uniqueness from co-occuring species on overall and genera-level infections.

Overall Infection	**Slope**	**Lower-CI**	**Upper-CI**	**ess**	***P*-value**		**Δ_{DIC}**
Intercept	9.411	-0.971	19.788	205	0.081		
Elevation range	0.00002	-0.001	0.001	979	0.937		-2
Omnivore	0.581	-0.581	1.752	1016	0.329		-1
Plant specialist	0.915	-0.365	2.176	991	0.165		-1
Shrub/Grassland	-0.794	-1.889	0.312	996	0.162		-1
Forest understory	-0.098	-0.872	0.677	1003	0.804		-1
Body mass	0.015	0.003	0.026	997	0.015	*	-3
Capture Elevation	0.00015	-0.0002	0.0005	986	0.376		-3
Evolutionary uniqueness	-0.091	-0.158	-0.023	332	0.010	*	2
Plasmodium							
Intercept	12.985	-1.918	28.342	153	0.094		
Elevation range	0.0001	-0.001	0.001	648	0.821		0
Omnivore	0.988	-0.747	2.778	529	0.264		1
Plant specialist	0.656	-1.247	2.629	490	0.517		1
Shrub/Grassland	-0.455	-2.133	1.185	649	0.592		0
Forest understory	-0.185	-1.391	0.991	765	0.753		0
Body mass	0.015	0.001	0.030	867	0.050	*	-3
Capture Elevation	-0.0002	-0.001	0.0003	746	0.508		0
Evolutionary uniqueness	-0.139	-0.246	-0.036	216	0.008	*	2
Haemoproteus							
Intercept	3.227	-12.502	18.934	117	0.696		
Elevation range	-0.0001	-0.001	0.001	683	0.877		0
Omnivore	0.958	-0.816	2.744	727	0.289		1
Plant specialist	1.489	-0.441	3.416	716	0.132		1
Shrub/Grassland	-0.280	-2.151	1.559	603	0.770		2
Forest understory	0.310	-0.925	1.575	790	0.637		2
Body mass	0.004	-0.014	0.021	847	0.631		1
Capture Elevation	-0.001	-0.001	-0.0002	540	0.006	*	7
Evolutionary uniqueness	-0.058	-0.161	0.043	204	0.268		0
Leucocytozoon							
Intercept	3.653	-11.465	18.943	146	0.650		
Elevation range	0.0004	-0.0004	0.001	855	0.362		1
Omnivore	0.438	-1.085	1.975	857	0.572		2
Plant specialist	1.109	-0.509	2.737	863	0.180		2
Shrub/Grassland	-1.426	-2.802	-0.080	946	0.038	*	2
Forest understory	-0.126	-1.115	0.850	949	0.804		2
Body mass	0.015	0.001	0.028	948	0.042	*	-3
Capture Elevation	0.001	0.001	0.002	831	0.001	*	2
Evolutionary uniqueness	-0.091	-0.195	0.010	240	0.082		4

Overall infection DIC=1850, *Plasmodium* DIC=700, *Haemoproteus* DIC=953 and *Leucocytozoon* DIC=1193. Values refer to the average model output from models run on 100 phylogenies of the 245 bird species. Δ_{DIC} is the change in DIC when the term is removed from the full model run on a single phylogenetic tree. * $P < 0.05$. ess=effective sample size, CI=confidence interval.

Results of significance tests analysing the strength of phylogenetic signal in binary host traits associated with foraging strata, guild and the incidence of infection from each parasite genus showed considerable variation. The majority of these traits supported a Brownian motion model of trait evolution, however the incidence of *Plasmodium*, *Leucocytozoon* and overall infections supported neither a random or Brownian motion model of trait evolution, and instead showed significant phylogenetic signal (Table 2.3). In the analyses of phylogenetic signal in continuous host traits, body mass exhibited strong phylogenetic signal (Z = -2.102, p = 0.014, mean of the random values = 303.236), whereas elevation range size did not (Z = 0.079, p = 0.584, mean of the random values = 42502.580).

Table 2.3 Results from tests of phylogenetic signal in categorical host traits including foraging strata, diet (foraging guild), and the incidence of infection among host bird species.

Trait	Estimated D	Probability of E(D) = E(1) resulting from random phylogenetic structure	Probability of E(D) = E(0) resulting from Brownian phylogenetic structure	
Foraging strata				
Canopy	0.187	0	0.236	
Scrub/Grassland	0.498	0.002	0.071	
Forest understory	0.189	0	0.238	
Guild				
Insectivore	-0.453	0	0.958	
Omnivore	-0.356	0	0.914	
Plant specialist	-0.527	0	0.970	
Infection type				
Plasmodium	0.671	0.001	0.002	*
Haemoproteus	0.375	0	0.057	
Leucocytozoon	0.423	0	0.033	*
Overall infection	0.626	0	0.001	*

* $P < 0.05$. Results are the mean from Fritz and Purvis' tests run on 100 separate bird phylogenetic trees.

In summary, I found little support for the hypothesis that life-history characteristics influenced malarial prevalence in our sample of birds. Instead, I found that evolutionary uniqueness, body mass and elevation (and hence temperature) were important determinants, with prevalence decreasing with increasing evolutionary uniqueness and increasing with body size for overall parasite infections. The relationship with elevation was more variable, with *Leucocytozoon* infection increasing with elevation but declining at the very highest elevations, and *Haemoproteus* infection showing idiosyncratic variation (Figure 2.2).

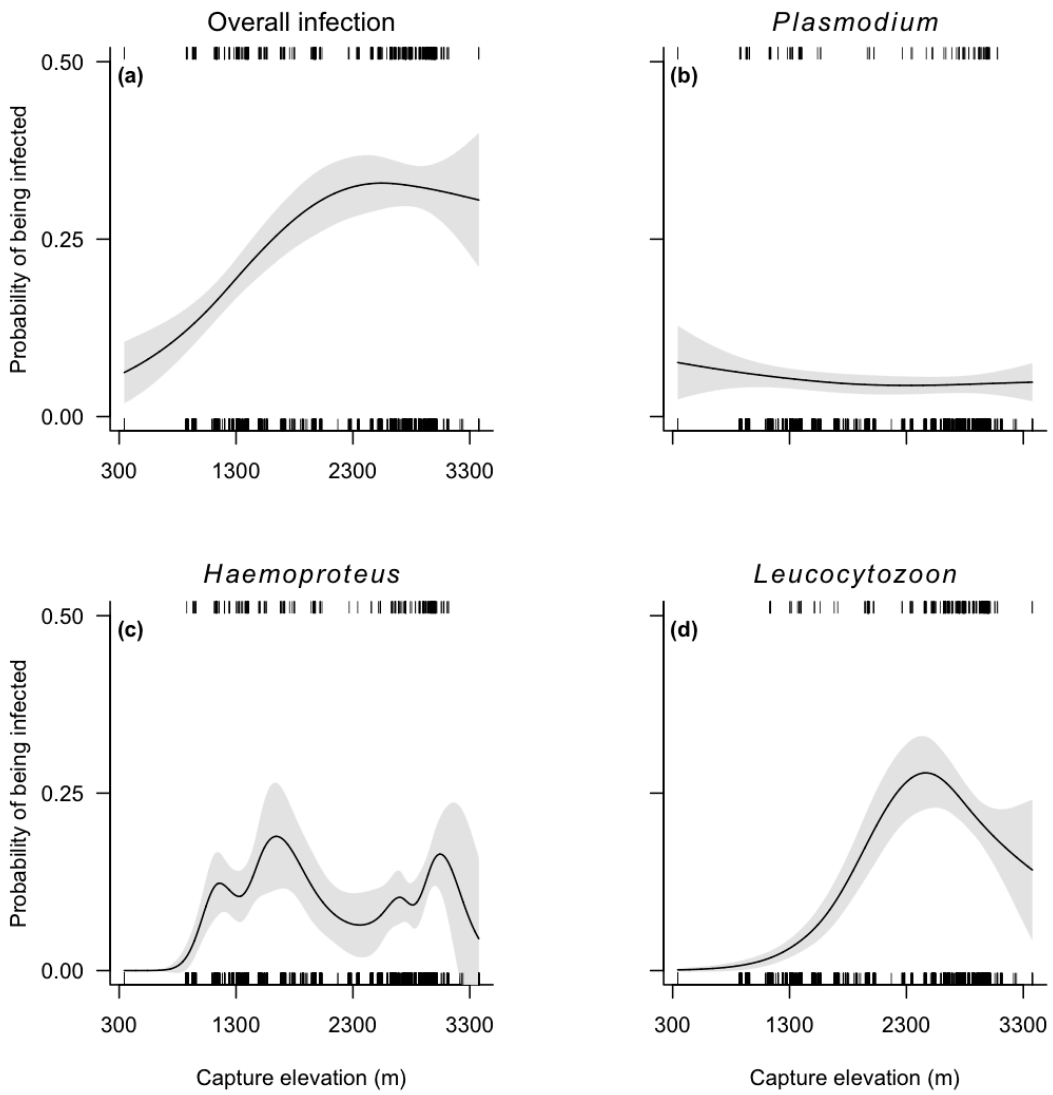


Figure 2.2 Probability of being infected with avian malaria parasites along the elevation gradient for (a) Overall infections (any of the three parasite genera), (b) *Plasmodium* infections, (c) *Haemoproteus* infections and (d) *Leucocytozoon* infections. Vertical dashed lines at the top and bottom of the plots show the location of infected (top) and uninfected (bottom) birds; solid lines show the smoothed linear fit of the predicted probability of infection across the entire elevation gradient, and dotted lines represent the 95 % confidence intervals around these.

Discussion

By quantifying the prevalence of avian malaria in a broad sample of bird species along a continuous elevation gradient, I tested the role of ecological versus evolutionary (i.e. intrinsic versus extrinsic) factors as determinants of disease prevalence. My analyses revealed that the incidence of infections is not a consequence of random infections among hosts, but rather that characteristics of the host and environmental are crucial. Specifically I found that body mass, elevation and evolutionary uniqueness were significant predictors of infection, while other factors thought to be important (elevation range size, foraging strata and diet) played no role.

Of all predictors, evolutionary uniqueness was the most important determinant of host infection. I found that the more evolutionarily distinct a species is within its community, the less likely it was to be infected with haemosporidian parasites in general, as well as for *Plasmodium* and *Leucocytozoon* infections in particular, and controlling for the effects of capture elevation, elevation range size, foraging strata and diet. This finding, though novel, is consistent with other studies showing reduced phytophagy on phylogenetically isolated trees (Yguel et al. 2011), reduced likelihood that a fungal pathogen can infect two plant species with increasing evolutionary distance between them (Gilbert and Webb 2007), and studies on *Drosophila* showing that infection success is significantly reduced if novel hosts are distantly related from the native host (Perlman and Jaenike 2003).

Although a number of mechanisms may contribute to the likelihood that a species becomes infected with avian malaria, these results show that when the roles of host

ecology, environment and phylogeny are evaluated simultaneously, host phylogeny is the best predictor. For overall haemosporidian infections and *Plasmodium* and *Leucocytozoon* infections, increasing evolutionary distance from co-occurring species was associated with reduced prevalence. This is consistent with a recent study showing that host compatibility rather than vector-host feeding patterns, explain the host distribution of *Plasmodium* parasites (Medeiros et al. 2013). These results support a role of internal host chemistry as the major factor influencing whether a parasite is able to successfully invade a host and provide a benchmark for future investigations to resolve the general importance of internal host body chemistry including the roles of host MHC genes (Bonneaud et al. 2006, Westerdahl et al. 2005, Loiseau et al. 2011) and cell surface proteins (Cowman and Crabb 2006) in influencing the invasion of red blood cells by malaria parasites. However, as an alternative explanation, it is also possible that more evolutionarily unique species within a community occur at lower abundance, which may also reduce their likelihood of infection (Mi et al. 2012).

Key aspects of host ecology (foraging strata and diet) were included in the overall model because they have been proposed to influence the likelihood that a host encounters a parasite (Ricklefs et al. 2005). For example, previous studies have shown strong spatial heterogeneity in the prevalence of avian malaria at a local scale (Wood et al. 2007) and suggest that habitat characteristics such as the suitability of potential vector larval habitats may be important determinants of prevalence (Valera et al. 2003, Little and Earlé 1995). Additional studies have recorded variation in the vertical distribution of hematophagous Diptera and suggest that forest structure may influence the height of host searching (Swanson and Adler 2010). By including foraging strata

as a potential explanatory variable influencing the incidence of infections among individuals I tested the assumption that vector-host encounter rates may differ between forest canopy, forest understory and shrub/grassland habitats. Dietary categories were also included in the model as these may also influence the exposure of hosts to parasites if invertebrate vectors of parasites comprise or are associated with other invertebrate food sources of birds. However, these predictions about the importance of host foraging strata and diet were not supported by any of the analyses. This may reflect vectors having much broader habitat associations, than the foraging and strata level associations measured here. In support of this, and contrary to a role of evolutionary uniqueness simply overshadowing the importance of these alternate variables, when evolutionary uniqueness was removed from the full model these later characteristics were still non-significant. These results are consistent with results from the aforementioned studies showing that although at broad geographical scales vector distribution and the incidence of avian malaria are strongly tied (Valera et al. 2003, Little and Earlé 1995), at local scales different bird species microhabitat associations don't appear to influence their disease prevalence (Ricklefs et al. 2005).

A key intrinsic host characteristic that was positively correlated with increased prevalence was body mass. This finding is consistent with previous studies showing a significant but weak positive effect of body mass on avian malaria prevalence in European (Scheuerlein and Ricklefs 2004) and North American birds (Ricklefs et al. 2005). The potential mechanism underlying increased infection risk for larger birds relates to larger birds producing more carbon dioxide, which arthropod vectors use as a signal to detect hosts (Gillies 1980), and also because larger birds present more surface area for infection (Atkinson and van Ripper III 1991). Another possibility is

that because larger birds are longer lived, they may have more chance to accumulate infection.

In spite of the strong association of increasing overall infection with elevation along the gradient (Figure 2.2), I found this to be non-significant in explaining overall and *Plasmodium* infections in the full multi-predictor model. However, capture elevation significantly predicted the prevalence of the other two genera and was positively correlated with *Leucocytozoon* infections, and negatively correlated with *Haemoproteus* infections. A likely mechanism that may explain this is that the abundance of vectors transmitting each of the parasite genera varies uniquely with changing environmental conditions along the gradient. This has previously been shown for *Leucocytozoon* in North America (Greiner et al. 1975) and Europe (Scheuerlein and Ricklefs 2004) where the prevalence of parasites is generally linked to the abundance of simuliid flies. In Hawaii as well, decreasing prevalence of introduced *Plasmodium relictum* is associated with a decline in the abundance of *Culex* mosquitoes, which are the main vector of introduced avian *Plasmodium* (Woodworth et al. 2005, Benning et al. 2002). The reported pattern of increasing *Leucocytozoon* prevalence with elevation may also be due to an increase in the abundance of arthropod vectors, although at present these are still unknown for each of the unique parasite lineages recorded here. Family level associations of haemosporidians with unique arthropod vectors suggests that the observed variation in prevalence is likely to be due to each genera's vector composition and geographic distribution. For instance, *Plasmodium* complete their development in the family Culicidae; *Haemoproteus* in the families Ceratopogonidae and Hippoboscidae; and

Leucocytozoon in representatives from Simuliidae and Ceratopogonidae; each of which have broadly different environmental requirements (Valkiunas 2005).

The increase in prevalence of *Leucocytozoon* toward higher elevations is consistent with previous studies that have shown it to reach similarly high prevalence (21.3 %) in the high Andes of Columbia (Rodriguez et al. 2009), and from studies that show increasing prevalence with latitude and decreasing environmental temperature in Chile (Merino et al. 2008). The two additional patterns of relatively consistent *Plasmodium* prevalence with elevation, and a mid-elevation peak in *Haemoproteus* prevalence, illustrate how each of these genera responds uniquely to changing host and environmental characteristics along the gradient.

Taken together, these results show that when the incidence of avian malaria infections are evaluated in relation to intrinsic, extrinsic and evolutionary host characteristics, host evolutionary characteristics explain the majority of the variation in infection prevalence among species. While larger body mass and capture elevation were significant predictors of increased overall and *Leucocytozoon* infections, the other intrinsic and extrinsic host characteristics considered here were found to be non-significant. Specifically I found no support for a role of host foraging strata, diet or elevation range size in influencing the incidence of infections among species. Instead, these results show that a host's evolutionary uniqueness relative to the community it occurs in is the best predictor of its infection prevalence. I found that the more evolutionarily distinct a host species was from the community it co-occurs with, the lower its overall, *Plasmodium* and *Leucocytozoon* infection prevalence. One plausible mechanism that may explain these results and that deserves further attention is that

evolutionary uniqueness reflects aspects of internal host chemistry and that this is the primary determinant of haemosporidian infection likelihood or ability to persist in a host. These results are consistent with a role of host physiology as a major driver of infection prevalence and predict that reduced infection prevalence with increasing phylogenetic distance may be driven by a reduced likelihood of parasite sharing among more distantly related hosts. Given that host ecology was unrelated to the incidence of infections among species, future study should focus on why evolutionary uniqueness is so important in predicting the incidence of infections, and whether or not it may also explain patterns of host sharing among parasite lineages.

Chapter 3

Host evolutionary uniqueness predicts the prevalence of avian malaria

Abstract

Interactions between hosts and their natural enemies show widespread variation, but the factors responsible for this remain poorly understood. Previous studies have highlighted the importance of host evolutionary characteristics in explaining observed patterns but the roles of community diversity and parasite sharing remain untested. These factors may potentially influence susceptibility to disease via either dilution or amplification effects in the first instance, or through more closely related hosts sharing internal or external compatibility filters in the second instance. Here I address this by firstly analyzing the prevalence of avian malaria across a wide range of host species and testing the importance of community diversity as well as host evolutionary and morphological uniqueness in explaining the observed variation. Secondly, I tested parasite sharing in relation to host ecological, environmental and evolutionary similarity as possible mechanism underlying the observed results. I find that the species richness of a community does not explain infection prevalence, whereas community evolutionary relatedness does, controlling for host evolutionary and ecological characteristics. Moreover, parasite sharing was strongly predicted by

host evolutionary differences, such that the probability of parasite sharing declined as the evolutionary distance between hosts increased. The amount of elevation range overlap between species was also a significant predictor of parasite sharing, but common diet, foraging strata or differences in body mass were each non-significant. Taken together, these findings support host internal body chemistry as the primary determinant of avian malaria prevalence among bird species and suggest that parasite sharing may underlie overall patterns.

Introduction

Interactions between hosts and their natural enemies are recognized as among the most important biotic interactions regulating populations, but vary considerably across species (Poulin 2011). For example, within communities the number of infected host species varies widely, and among these infections may occur in just a few individuals or at very high prevalence (Loiseau et al. 2013). Although previous studies have highlighted an important role of host evolutionary uniqueness in explaining variation in infection prevalence among species, potential mechanisms underlying this remains unknown. An outstanding question therefore is what determines the reduced infection prevalence of evolutionarily unique species in a community? Two themes that may help answer this question are: (1) understanding the potential importance of community composition and (2) parasite sharing.

Firstly, reduced infection prevalence among more evolutionarily unique species within a community may be due to the broader network of interactions within which an individual is embedded. For instance, increasing community diversity may reduce

infection risk through the 'dilution effect' which describes an overall net effect of species diversity reducing disease risk via incompatible hosts reducing the transmission success among compatible hosts in the same community (Keesing et al. 2006). Alternatively, greater host diversity may in fact have the opposite 'amplification effect' on prevalence if parasites have broad host associations in which different host species act as reservoirs for disease (Power and Mitchell 2004). Secondly, and embedded within these themes, parasite sharing among hosts may underlie observed relationships. Invoking a role for parasite sharing, higher infection prevalence of more evolutionarily similar hosts may be driven by more closely related species sharing physiological, environmental or ecological characteristics that make them more susceptible to infection by the same natural enemies (Bertheau et al. 2010, Combes 2004, Perlman and Jaenike 2003).

Previous studies have highlighted the importance of evolutionary uniqueness from co-occurring species as a primary determinant of infection prevalence but potential correlates between evolutionary uniqueness and ecological uniqueness remain untested. For instance, evolutionary uniqueness and morphological uniqueness may be correlated if morphological characteristics are phylogenetically conserved (i.e. more closely related hosts may have similar diets and habitat associations). However, by simultaneously testing the roles of evolutionary and morphological uniqueness in explaining infection risk among co-occurring species, it is possible to resolve their relative importance and indirectly assess whether infections are likely to be limited by internal (host) or external (i.e. ecological) compatibility filters (Combes 2004).

Recent empirical work on the importance of host evolutionary uniqueness in determining susceptibility to attack from natural enemies has focused on plants and shown that within a single species, increasing phylogenetic isolation from neighbours can correspond to a 10-fold decline in phytophagy (Yguel et al. 2011), and that among forest trees infected with fungal pathogens there is significant phylogenetic signal in plant pathogen-host range (Gilbert and Webb 2007). Additional studies show that the fitness of phytophages is reduced by feeding on novel host species compared to ancient host species (Bertheau et al. 2010). Among the few studies on animals, the potential host range of parasitic nematodes on *Drosophila* hosts is much larger than their actual range, and the likelihood of these parasites establishing in novel hosts decreases with increasing phylogenetic distance between the native and novel host (Perlman and Jaenike 2003). A further geographic study on primates shows that host phylogenetic relatedness is an important predictor of parasite sharing, but fails to fully explain observed associations (Cooper et al. 2012).

Although each of these aforementioned studies have investigated the likelihood that parasite sharing declines with increasing evolutionary divergence between host pairs, none have investigated the extent to which this may influence observed levels of parasite prevalence for individual species within diverse natural communities. Moreover, previous results have been based on either broad scale geographical studies with variable sampling effort of hosts and parasites (Cooper et al. 2012), artificial infections in the laboratory (Perlman and Jaenike 2003) or the field (Gilbert and Webb 2007), or based on just a small subset of host parasite associations (Yguel et al. 2011). Therefore the potential importance of community diversity and parasite sharing underlying the observed reduced infection prevalence of evolutionarily unique

species remains completely untested among diverse assemblages of locally co-occuring species. Parasites have been put forward as a major factor explaining vertebrate, and in particular bird, distribution and diversity so evaluating their importance remains an important theme in ecology and evolutionary biology (Atkinson and van Ripper III 1991, Ricklefs 2010a, Ricklefs and Bermingham 2002, Hochberg and Ives 1999).

Avian haemosporidian (malaria) infections offer an ideal system within which to investigate the importance of community diversity and parasite sharing toward influencing infection prevalence because they are widespread among bird species and show heterogeneous distributions across hosts (Valkiunas 2005, Bensch et al. 2009). Previous studies have revealed instances of within-host speciation of avian malaria parasites (Pérez-Tris et al. 2007) and broad scale analysis of diversification and host switching show significant cospeciation between hosts and their avian malaria parasites (Ricklefs and Fallon 2002). These results suggest that sharing among close relatives may be an important determinant of infection prevalence. Additional studies show that host environment can influence avian malaria prevalence at both local (Wood et al. 2007, Garvin and Remsen 1997) and regional scales (Zamora-Vilchis et al. 2012, Loiseau et al. 2013), supporting a role for species ecological traits in determining the likelihood of infection. Here I test the extent to which prevalence is predicted by community diversity, and the extent to which parasite sharing may explain observed results.

Materials and Methods [For details on the study site, sampling, and how evolutionary uniqueness was calculated see methods in Chapter 2]

Functional uniqueness

I quantified functional uniqueness of each species using ecologically important host traits based on the following measurements (1) Bill length (2) Bill width (3) Bill depth (4) Tarsus length (5) Wing length and (6) Tail length. I used principal component analyses to reduce the correlation among these traits and chose the PC axes that explained more of the variation in traits than when each trait was randomly shuffled among species. Functional uniqueness for a species was then calculated as the average Euclidean distance in trait space between that species and all species that overlapped its elevation range. Morphometric data were only available for passerine bird species so functional uniqueness measurements are relative to other passerine species

Parasite Phylogeny [see general methods in Chapter 1]

Statistical analysis

I tested the importance of host evolutionary and morphological uniqueness using Markov Chain Monte Carlo estimation implemented with "MCMCglmm". The response variable was host species infection status entered as a multinomial response using the number of infected vs. number of uninfected captures of each species and with family specified as "multinomial2". Each bird species' elevation range midpoint was also entered as a predictor variable in the model to control for variation in the community composition across the elevation gradient. All analyses were conducted

on the restricted dataset of 219 resident Passerine bird species as these were the ones for which I had both morphological and evolutionary information, and because resident birds are more likely to have been infected near their point of capture than migrants or vagrants which by definition exhibit much greater movements within their ranges. I investigated the drivers of both overall infection and infection among the three major parasite genera found within the study (*Plasmodium*, *Haemoproteus* and *Leucocytozoon*). All analyses were run across the each of the 100 phylogenetic trees used to calculate host evolutionary uniqueness with the following parameters: burnin=10000, thin=100, iterations = 220000; and with priors specified as: V=1, nu=0.002, see Hadfield (2010) for further details. Because minimum adequate models can inflate Type I errors, I report the average results of the full models and include the change in the deviance information criterion (DIC) between a the full model and the model excluding the predictor as an additional measure to help assess the relative importance of predictors.

Parasite sharing

I tested the extent to which two bird species were likely to share a parasite lineage based on their pairwise phylogenetic distance, amount of elevation range overlap, body mass difference, and whether or not they had the same foraging guild or foraging strata. Elevation range overlap was defined as the average overlap between two species. This was calculated as the amount of species 1 elevation range overlapping with species 2's elevation range, divided by species 1 total elevation range size; plus, the amount of species 2's elevation range overlapping with species 1's elevation range, divided by species 2's total elevation range size. Therefore, species pairs can have any value from 0 – 2, 0 indicating no elevation range overlap

and 2 indicating that species 1 shares 100% of its elevation with species 2 and that species 2 shares 100% of its elevation range with species 1. Body mass difference was the only predictor variable that was log transformed prior to analysis. Although generalized linear models make no assumptions about the normality of predictor variables, by doing this smaller differences in species pairs' body mass were more heavily weighted than large differences, which I feel is ecologically sound given parasite sharing is likely to decrease with increasing body mass differences between the pair. This analysis was restricted to 41 well-sampled bird species (i.e. represented by 15 or more captures).

First, I created a dataset of all possible pairwise combinations (n = 820) of these species and then only selected species pairs with unique most recent common ancestors to avoid pseudoreplication in divergence times. To account for variation in the species pairs that could be chosen sharing each of these unique nodes, I randomly sampled unique species pairs, across 100 phylogenetic trees. I then calculate the pairwise phylogenetic distance between pairs, resulting in 100 datasets of unique species pairs. Although this dataset contains pseudoreplication in the form of host species appearing multiple times among species pairs, because the analysis is based on unique pairwise distance values between pairs across 100 phylogenetic trees, and because each pair produces a unique value of parasite similarity, in accordance with Cooper et al. (2012) I don't feel this is a major issue.

To evaluate the extent to which each of the predictor variables explained parasite sharing among species pairs, I fitted a general linear model (glm) with the response variable entered as the number of shared lineages versus the number of unshared

lineages between pairs, with family specified as binomial and with the model weighted by the total number of infections. To assess the importance of each of the predictor variables I followed the model averaging approach described in Grueber et al. (2011), with the exception that I used the entire model set to determine average importance values rather than only including models based on an arbitrary cutoff. As a further illustration of the significance of each of the predictor variables, I also included the average model output of the global glm run across all 1000 datasets of species pairs.

Results

Overall infection

Contrary to expectations for community diversity having either a dilution or amplification effect on parasite prevalence, I found no evidence for this in any of the analyses (i.e. using all recorded infections, *Plasmodium* infections, *Haemoproteus* infections, or *Leucocytozoon* infections; all $P > 0.05$) (Table 3.1).

Table 3.1 The importance of host evolutionary and morphological uniqueness as well as species midpoint elevation range and community diversity, in influencing the incidence of infections among all 219 sampled bird species.

Overall Infection	**Slope**	**Lower-CI**	**Upper-CI**	**ess**	***P* - value**		**Δ_{DIC}**
Intercept	12.931	-0.485	26.386	567	0.059		
Evolutionary uniqueness	-0.120	-0.222	-0.019	695	0.019	*	3.077
Morphological uniqueness	0.742	-0.074	1.565	789	0.075		-1.723
Community diversity	0.000	-0.003	0.003	799	0.753		-1.201
Elevation midpoint	0.000	-0.001	0.001	838	0.894		-0.797
Plasmodium							
Intercept	18.625	-3.966	41.460	464	0.103		
Evolutionary uniqueness	-0.207	-0.386	-0.035	465	0.016	*	1.559
Morphological uniqueness	1.003	-0.062	2.038	846	0.072		-1.604
Community diversity	0.003	-0.002	0.008	525	0.209		4.855
Elevation midpoint	0.000	-0.001	0.002	578	0.769		3.046
Haemoproteus							
Intercept	2.024	-19.265	23.510	468	0.850		
Evolutionary uniqueness	-0.028	-0.195	0.135	504	0.750		0.065
Morphological uniqueness	-0.959	-2.482	0.510	430	0.200		1.233
Community diversity	-0.002	-0.007	0.004	543	0.560		-0.104
Elevation midpoint	-0.001	-0.002	0.001	634	0.350		-1.953
Leucocytozoon							
Intercept	15.470	-5.629	36.899	567	0.151		
Evolutionary uniqueness	-0.177	-0.341	-0.016	626	0.029	*	3.206
Morphological uniqueness	0.752	-0.338	1.814	990	0.174		-0.661
Community diversity	0.001	-0.004	0.005	852	0.656		0.422
Elevation midpoint	0.002	0.000	0.003	748	0.012	*	2.668

Overall infection DIC=1830.150, *Plasmodium* DIC=700.468, *Haemoproteus infection* DIC=965.336 and *Leucocytozoon* DIC=1182.954. Values refer to the average model output from models run on 100 phylogenies of the 219 bird species. ΔDIC is the change in DIC when the term is removed from the full model run on a single phylogenetic tree. * $P < 0.05$. ess=effective sample size,CI=confidence interval.

Consistent with previous studies, increasing evolutionary distance from co-occurring birds was significantly associated with reduced overall haemosporidian infections (any of the three genera) ($P_{MCMC} = 0.019$) (Figure 3.1 & 3.2), *Plasmodium* infections ($P_{MCMC} = 0.016$) and *Leucocytozoon* infections ($P_{MCMC} = 0.029$), but not *Haemoproteus* infections ($P_{MCMC} > 0.05$) (Table 3.1).

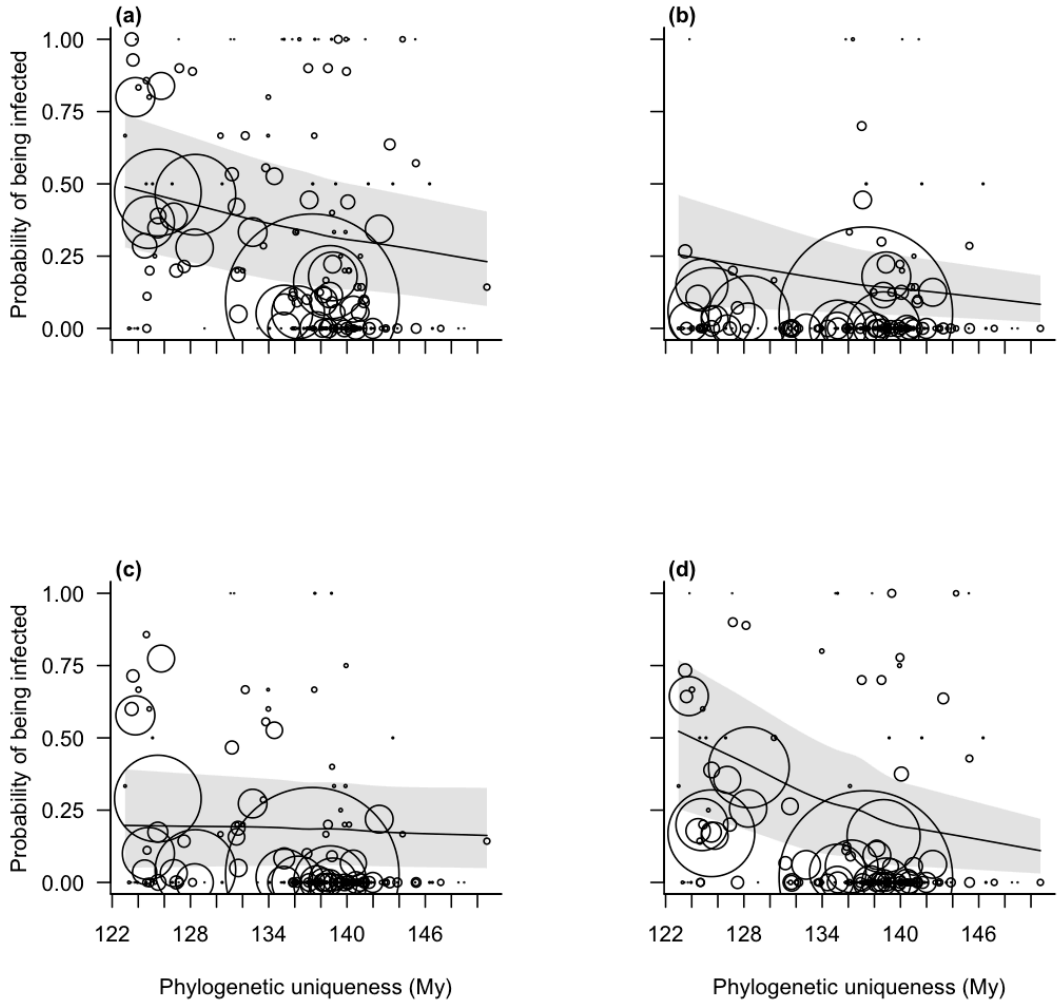


Figure 3.1 Relationship between evolutionary (phylogenetic) uniqueness and prevalence for (a) Overall infections (any of the three parasite genera), (b) *Plasmodium* infections (c) *Haemoproteus* infections and (d) *Leucocytozoon* infections. The boxes include the mean (solid line) and upper and lower credible intervals (shaded) of the fitted relationship between the variables. The size of the circles indicate the number of captures of each of the 219 passerine bird species. Phylogenetic uniqueness is the average path length distance between the focal bird host species and all other co-occuring bird species within its elevation range.

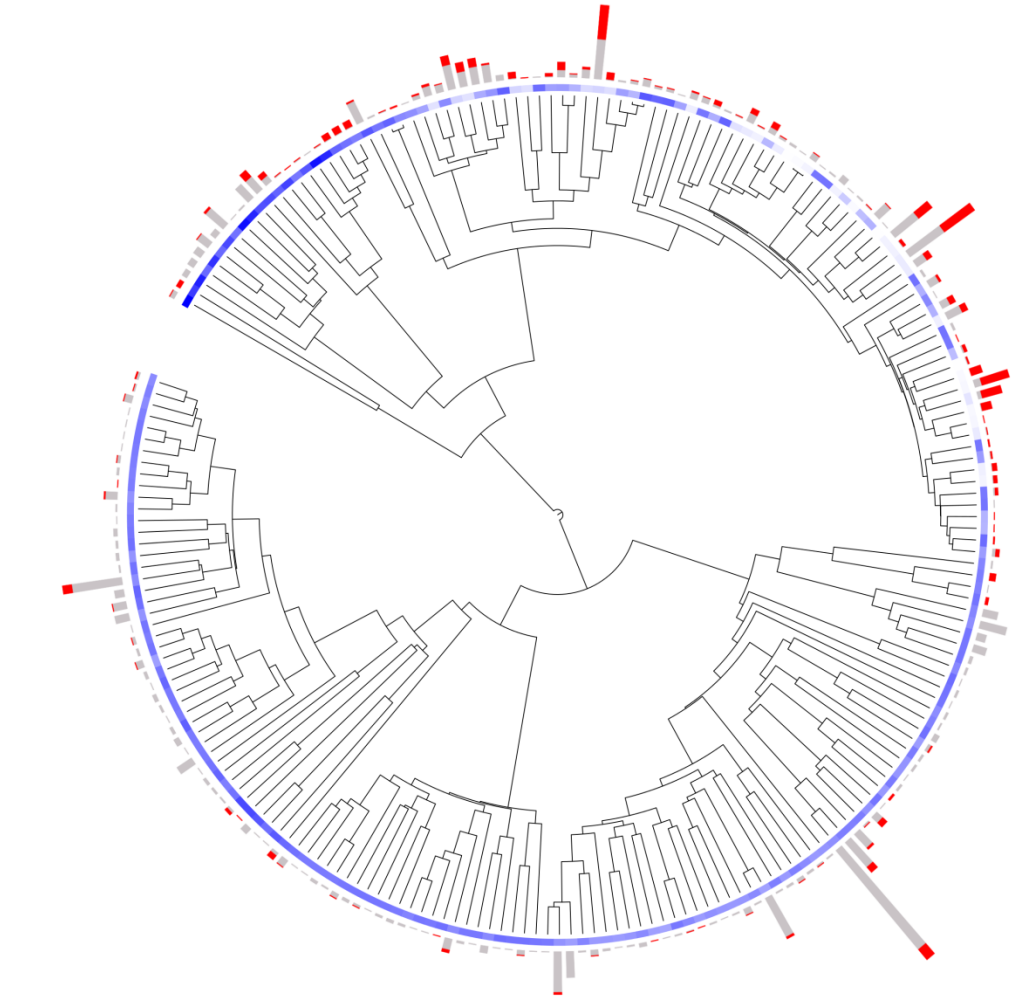


Figure 3.2 Relationship between evolutionary uniqueness and infection prevalence for 219 passerine species. Grey bars represent the number of uninfected birds, red bars the number of infected birds, and the intensity of blue squares increases with the relative phylogenetic uniqueness of each species. Closely related species can have different evolutionary uniqueness values because this is calculated relative to the bird community they co-occur with, which may be significantly different for closely related species (particularly if they do not co-occur due to competitive exclusion). Figure created online (http://itol. embl. de) (Letunic and Bork 2011).

Results from other terms in the model revealed that there was no significant effect of host morphological uniqueness in explaining the prevalence of infections among species for any of the infection classifications, although for overall and *Plasmodium* infections there was a slight positive relationship between prevalence and increasing morphological uniqueness. Investigating whether elevation may be an important predictor of infection prevalence, only for *Leucocytozoon* infections was there a significant positive effect of species midpoint elevation range on infection prevalence ($P = 0.012$).

Parasite sharing

Testing parasite sharing as a potential mechanism behind the importance of evolutionary uniqueness in predicting the incidence of infection among species, I found that the likelihood of two bird species sharing a parasite lineage declined with increasing evolutionary distance between the species pair (importance = 0.939, $P = 0.031$) (see results Table 3.2 & 3.3, Figure 3.3 & 3.4). Investigating a role of environmental and ecological characteristics, the amount of elevation range overlap between species pairs was the next most significant predictor of parasite sharing (importance = 0.939, $P = 0.031$); whereas shared foraging strata, guild or differences in body mass were all non-significant predictors of parasite sharing (all $P > 0.05$) (Table 3.3).

Table 3.2 Summary Multimodel inference outputs testing the importance of species pair ecological and evolutionary differences in influencing the incidence of shared infections between them.

Species pair characteristics	Coefficient	SE	Adjusted SE	Lower-CI	Upper-CI	Importance
Intercept	-2.61	0.093	0.097	-2.802	-2.420	
Pairwise phylogenetic distance	-1.79	0.293	0.303	-2.385	-1.200	0.939
Elevation range overlap	0.66	0.113	0.117	0.433	0.890	0.972
Body mass difference	0.34	0.119	0.124	0.432	0.920	0.854
Shared guild	0.34	0.166	0.172	0.0004	0.670	0.737
Shared strata	0.37	0.146	0.152	0.070	0.660	0.712

SE = standard error, CI = confidence interval.

Table 3.3 The importance of species pair ecological and evolutionary differences in influencing the incidence of shared infections between them.

Species pair characteristics	Estimate	SE	Z -Value	Pr (>|Z|)
Intercept	-2.482	0.246	-10.4	0.002
Pairwise phylogenetic distance	-0.031	0.005	-5.7	0.031*
Elevation range overlap	0.508	0.087	5.9	0.012*
Body mass difference	0.655	0.113	6.2	0.082
Shared guild	0.331	0.167	1.7	0.148
Shared strata	0.363	0.148	2.4	0.172

Results are the average model outputs of the global glm run across 1000 datasets of species pairs. SE = standard error.

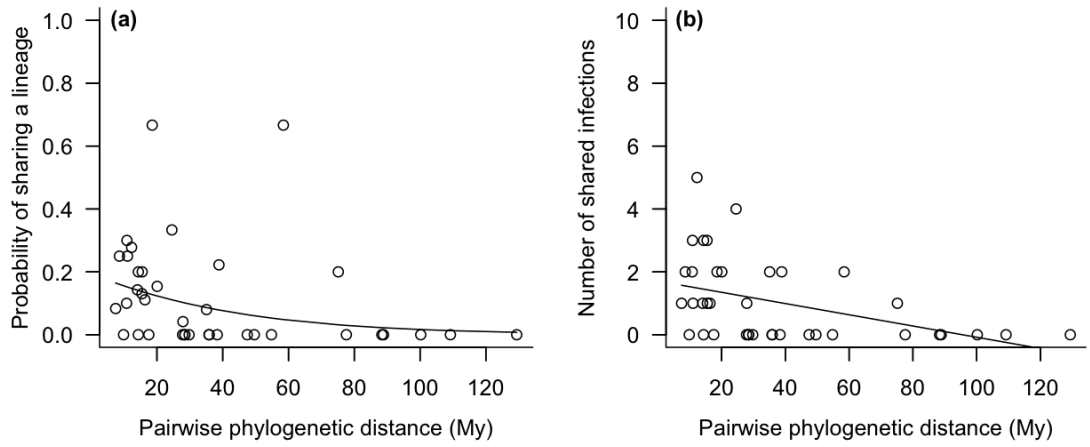


Figure 3.3 The relationship between pairwise phylogenetic distance between 41 bird host species pair combinations and (a) the probability of sharing a parasite lineage, (b) the number of shared parasite lineages. Each individual bird species had ≥15 captures to control for the reduced likelihood of detecting infections among less well sampled species.

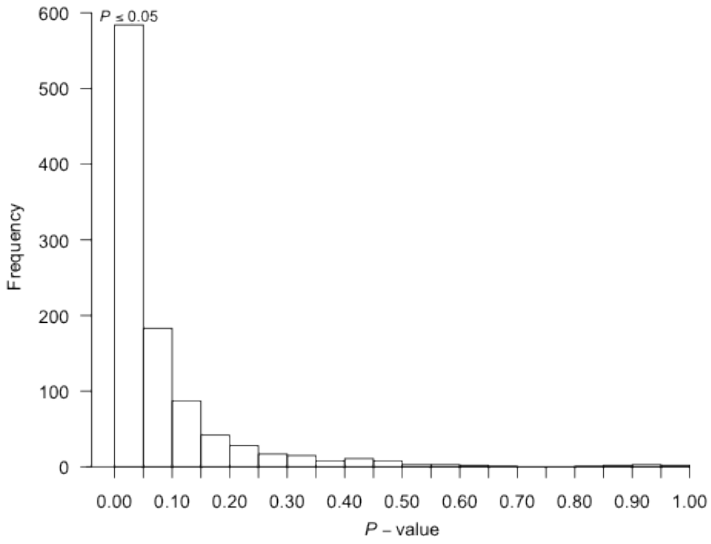


Figure 3.4 Histogram of *P*-values from general linear model testing the relationship between the proportion of lineages that were shared by species pairs and their pairwise phylogenetic distance. Mean = 0.082, Median = 0.038.

In summary, these results show that the number of species within a community is not an important predictor of avian malaria prevalence within passerine bird assemblages but the evolutionary diversity of the community is. Specifically, I found that more evolutionary similar community members suffer higher infection prevalence compared to more evolutionarily distinct host species.

Discussion

In this study I tested if community diversity may be an important predictor of variation in the prevalence of avian malaria among a diverse assemblage of resident bird species. Contrary to expectations from hypotheses about the potential for community diversity to either amplify or dilute infection prevalence among species, I found no evidence to support this based on analyses of the overall incidence of avian malaria infections, or from any of the three parasite genera (*Plasmodium*, *Haemoproteus*, and *Leucocytozoon*). Instead, I found strong support for a principle role of mean evolutionary distance from co-occurring species in shaping the observed variation – indicating that it is not community diversity per say, but rather community relatedness that best explains the widespread variation in avian malaria among evolutionarily and ecologically distinct hosts.

Consistent with previous studies, my results show that increasing evolutionary uniqueness of a bird host from other species it co-occurs with strongly predicts reduced prevalence of avian malaria, controlling for host ecological (morphological) characteristics. For overall haemosporidian infections as well as *Plasmodium* and *Leucocytozoon* infections, increasing evolutionary uniqueness was consistently

associated with reduced infection. As a possible mechanism underlying this result I tested whether parasite sharing may be important. I found that greater evolutionary divergence time between species pairs corresponded to a reduced likelihood of their sharing a parasite lineage. Taken together, these results suggest that the observed lower infection prevalence of more evolutionarily distinct host species within a community may be driven by their reduced likelihood of sharing parasites with other community members. Both results are consistent with the hypotheses that evolutionarily distinct host species within a community are likely to have differing physiological, chemical or other characteristics from other host species that make them less susceptible to infection by shared natural enemies (Bertheau et al. 2010, Combes 2004, Perlman and Jaenike 2003).

One interpretation of increasing evolutionary uniqueness corresponding to reduced infection prevalence is that because community composition varies strongly with elevation I have simply detected the effect of altitudinal differences in the prevalence of parasites (Zamora-Vilchis et al. 2012). However, in addition to finding no support for overall community diversity predicting infection prevalence, the significant effects of host species evolutionary uniqueness remain intact when altitude is included as a covariate in the analyses. A further possibility for the importance of greater evolutionary uniqueness corresponding to reduced infection prevalence is that phylogeny approximates ecology, *sensu* Phylogenetic niche conservatism. Under this scenario, evolutionarily distinct species may have particular habitat associations that make them less prone to infection. However, I didn't find any effect of morphological (i.e. ecological) uniqueness on infection prevalence so I consider this to be unlikely. A further possibility is that more evolutionarily unique species are less susceptible to

infection due to lower abundance. Although the relationship between evolutionary uniqueness and abundance in birds remains unknown, in a study on the contribution of rare plant species to community phylogenetic diversity across a global network of forest plots, rare species were found to be distantly related to common species (Mi et al. 2012) – suggesting abundance and evolutionary uniqueness may be negatively correlated. Abundance is a major determinant of prevalence for directly transmitted parasites that have density dependent transmission, but its importance for vector-transmitted parasites, which are thought to primarily have frequency dependent transmission, is less well understood. Empirical work on a wide range of vertebrates and invertebrates suggests that most pathogens fall between these two extremes (Hudson et al. 2002), so it is possible that the haemosporidian parasites studied here also have some element of density dependent transmission. If this is the case, the hypothesized lower abundance of more evolutionarily unique species, may be the reason for their lower infection prevalence. However, in this study I was unable to estimate abundance for the 219 bird species sampled for avian malaria parasites, so could not directly test this hypothesis.

An alternative explanation that I consider to be the most plausible based on these results, is that internal host body chemistry shapes the observed relationships due to differing individual and species-level immune system adaptations that are phylogenetically conserved. Studies on humans have revealed that genetic factors are important in predicting the susceptibility and outcome of malaria infections (Fortin et al. 2002) and research on birds suggests that individuals with high MHC diversity or with specific MHC alleles are associated with malaria resistance (Bonneaud et al. 2006, Westerdahl et al. 2005, Loiseau et al. 2011, Sepil et al. 2013). Furthermore, a

recent study by Medeiros et al. (2013) has shown that the feeding patterns of the dominant vectors of *Plasmodium* parasites are similar across a range of avian hosts but do not explain the distribution of these parasites among hosts. Both these results support the hypothesis that internal host compatibly filters are primary determinants of infection prevalence.

A major implication of increasing evolutionary uniqueness corresponding to reduced infection prevalence within a community is consistent with previous work suggesting mechanisms through which natural enemies may play a role in the accumulation of species diversity locally. Plants provide some of the most well studied examples and numerous studies support a potential role for host specific plant pathogens in regulating forest diversity (Janzen 1970, Gilbert and Webb 2007), although the exact nature of these effects still remain largely unknown (Freckleton and Lewis 2006). My results offer further support for a pathogen mediated mechanism through which species diversity could accumulate locally because through having lower infection prevalence, evolutionarily distinct species are hypothesized to be at a comparative advantage within their community. This result is consistent with the 'enemy release' hypothesis which in its broadest sense, states that species introduced to an exotic region should benefit from a decrease in regulation by their natural enemies (Keane and Crawley 2002, Torchin et al. 2003, Colautti et al. 2004).

Taken together, these results also have important implications for understanding variation in disease prevalence and species coexistence. The importance of a host community's evolutionary diversity, but not overall diversity (i.e. species richness), along with increased parasite sharing among more closely related species, suggests

that host internal compatibility filters (i.e. internal body chemistry) are more important than external compatibility filters (i.e. associated with host ecology or environment) in explaining infection prevalence within my study system. These results are based upon analyses from a wide range of evolutionarily and morphologically distinct host species so may be indicative of more widespread patterns among birds and their avian malaria parasites.

Chapter 4

Prevalence of avian malaria increases toward the centre of hosts' elevation ranges

Abstract

Theory predicts that population densities should be highest at the centres of geographical ranges, where conditions are optimal for a species, and decline toward the edges. Although some studies support this prediction for animals, plants, and for parasites with direct transmission between hosts, little is known about populations of vector-distributed parasites. Here I investigate the prevalence of avian malaria parasites (*Plasmodium, Haemoproteus,* and *Leucocytozoon* [Apicomplexa: Plasmodiidae]) to determine the relationship between prevalence and position within range. I do this in two distinct ways: first within host species elevation ranges and second across the parasites' entire elevation range. Firstly, I found that bird species are more infected with parasites toward the centre of their elevation ranges. When parasite genera were analysed separately, the incidence of *Plasmodium* infections increased toward the centre of their hosts' ranges, whereas *Haemoproteus* and *Leucocytozoon* showed no such pattern. I also show that the prevalence of a parasite lineage within a host does not predict the location of infections within the host's elevation range. Secondly, across their own elevation ranges lineages did not show

any significant difference in prevalence. I conclude that although birds are more infected toward the centre of their elevation range – possibly reflecting variation in local host density – the broad host and environmental associations of avian malaria parasites do not appear to result in a concentrated abundance at the centre of their elevation range.

Introduction

A key hypothesis concerning species distributions is that population densities are highest at the centres of geographical ranges, and decline toward the edges (Brown 1984, Gaston 2003, Andrewartha and Birch 1954, Hengeveld and Haeck 1982). Causal factors thought to shape such patterns include a variety of limiting abiotic and biotic factors. For instance, abiotic factors may limit species ranges due to physiological constraints associated with unsuitable physical conditions, whereas biotic limits may arise from competition, predation, disease or parasitism (Sexton et al. 2009, Chown and Gaston 1999). Empirical studies of a variety of free-living terrestrial organisms and directly transmitted parasites reveal distributions where abundance is highest at the range centre and significantly reduced toward the range edges, lending support to the generality of this hypothesis (Whittaker 1960, Whittaker 1965, Bystrak 1981, Terborgh 1971, Brown 1984, Hengeveld and Haeck 1982). However, Sagarin and Gaines (2002) found only mixed support for the 'abundant centre' distribution in a meta-analysis investigating species abundances across geographical ranges of free-living organisms. Moreover, almost nothing is known about the population distributions of vector-dispersed parasites, in which abundance is

influenced not only by hosts, whose abundance influences probabilities of infection, but also by vectors, which are sensitive to physical conditions of the environment.

Here, I investigate the prevalence of avian malaria parasites across the elevation range of host bird species to firstly explore host susceptibility to infection in relation to the location of individuals within their species range, and secondly test the abundant-centre hypothesis for specific lineages (=species) of parasites. Because lineages are able to infect multiple host species, I investigate parasite abundance within individual host bird species elevation ranges, and also across the parasites entire elevation range. I examine whether avian malaria parasites are, in general, more abundant in the centre of their host ranges owing to generalized responses of the parasites or their vectors to physical conditions of the environment or the abundance of their host bird species. Alternatively, the opposite pattern might arise if particularly pathological parasites endemic to one species limit the geographical distribution of a second species, as has previously been suggested (Briers 2003, Hochberg and Ives 1999, Ricklefs 2010a, Hoffmann and Blows 1994, Holt and Lawton 1994).

Parasites present ideal systems within which to test hypotheses about patterns of abundance, as they exhibit a variety of generalist to specialist infection patterns with wide variation in prevalence among hosts (Hellgren et al. 2009, Daszak et al. 2000, Dobson 2004), which lend themselves to testable predictions about where parasites are transmitted to hosts within the host's range. For vector-borne parasites in particular, transmission could remain relatively constant over host range, and also across changes in host density, when the rate at which susceptible hosts encounter infective vectors is determined by the proportion of infected hosts within a population, rather than overall density of hosts (Thrall et al. 1995, McCallum et al.

2001, Antonovics et al. 1995). However, even when vector distribution is uniform across a host's geographical range, parasites may reach their greatest prevalence in host individuals where host abundance is greatest due to an increased likelihood of transmission (Arneberg et al. 1998, Grenfell and Dobson 1995, Anderson and May 1978, May and Anderson 1978). Furthermore, a centred-abundance pattern within a host's range would be more likely for parasites with restricted or more specialized host distributions, as these will be more sensitive than generalist parasites to changes in the abundance of a suitable host across its range. These considerations lead to two competing predictions about the distribution of parasite prevalence across a host's range. Firstly, from the host's perspective, if ranges are limited by parasites that directly or indirectly reduce fitness, then I expect the highest prevalence of parasites to occur at the edges of a host's range where these depressing effects are most apparent. Moreover, such parasites are likely to be maintained as infection reservoirs in other susceptible host species. Alternatively, from the parasite's perspective, if parasite populations are limited by transmission between hosts, prevalence should increase toward the centre of a host's range, or wherever host abundance is highest. Additionally, the prevalence of all parasites together might reflect favourable physical conditions for the parasites or for their vectors, and be abundance-centred with respect to ecological gradients, rather than host distributions. Aside from studies of human malaria prevalence with latitude (Dunn et al. 2010, Snow et al. 2005) few studies have examined spatial variation in the prevalence of vector-transmitted parasites within their hosts' range, and of the few that have, these have often only investigated pattern within a single host species (Briers 2003).

I tested these predictions by studying the prevalence of avian malaria parasites (Pérez-Tris et al. 2005) belonging to the genera *Plasmodium, Haemoproteus,* and *Leucocytozoon* (Apicomplexa: Plasmodiidae), over the ranges of host bird species along an elevation gradient on the eastern slope of the Andes Mountains in Peru. Avian malaria parasites comprise an ideal system within which to investigate parasite prevalence across host ranges, as they occur across a wide geographical range and have been recorded from almost all bird families (Valkiunas 2005). Meanwhile, elevation transects offer a tractable system within which to investigate parasite prevalence across host distribution because high beta turnover and changing environmental conditions allows for patterns to be investigated across a wide range of species over relatively short distances. I begin by analysing parasite prevalence within individual host bird species elevation ranges for each of the three parasite genera to test for overall abundance centring within host elevation ranges. In addition, I examine parasite prevalence across the entire elevation gradient to test for abundance centring across the overall ecological gradient. Then, I use an alternative approach focused on independent lineages (i.e. species) of parasites to test for abundance centring within specific host bird ranges, and across the parasite lineage's entire elevation range which may contain multiple host species. The second approach additionally tests whether more abundant parasite lineages, defined as those that reach higher prevalence within their hosts, are more range-centred than lineages recorded at lower prevalence.

Materials and Methods

Parasite prevalence within host elevation ranges

Logistical constraints precluded identical sampling effort at all survey sites along the elevation gradient. Because of this, overall prevalence estimates are based on the total number of infected birds divided by the total number of examined birds in accordance with Margolis et al. (1982). Elevation limits of bird species recorded in the study are those presented in Walker et al. (2006) and are based upon detailed site, sound or sign (i.e. nest) records along the gradient and in surrounding areas. I calculated a proportional distance from the nearest elevation limit of a host species as the distance between an individual's capture location and its nearest elevation range limit, divided by half of its total elevation range. For birds captured above or below their recorded elevation range limits I assigned proportional distances from edge equal to 0. To ensure these results were not biased by unequal sampling effort across the elevation ranges of individual host species, in which more intense sampling at a particular capture elevation may have corresponded to a specific proportional distance from near limit (i.e. as occurs at the limits of the gradient where species ranges end) I tested for an interaction between sampling elevation and the proportional distance from the host species near elevation limit for all captures and found no significant effect.

Lineages prevalence within lineage elevation range

I defined the elevation range limits of parasite lineages as the maximum and minimum elevations at which they were recorded. Within parasite lineage ranges I calculated their prevalence within the inner 50% of their range defined as 25% of the range above and below the elevation range midpoint; and outer 50% of their range,

defined as 25% of the range above the lower limit and 25% of the range below the upper range limit. Due to the overall low prevalence of lineages throughout their ranges, this provided the best estimate of lineage prevalence relative to range position.

Data analyses

To test whether individuals of each host species had greater overall prevalence of avian malaria infections closer to the centre of their elevation ranges, I fitted a linear mixed regression model using the R (R Development Core Team 2010) package MCMCglmm (Hadfield 2010) with the infection status of individual birds entered as a binary response variable, and with Proportion Distance from Near Elevation Limit, Capture Elevation and Elevation Range Size included as continuous explanatory variables, and with Bird Species entered as a random effect. This accounted for different bird species as a source of random variation and therefore tested whether the observed result was independent of host species. Four separate models were run, one for each category of infection, i.e. (i) all parasite genera, (ii) *Plasmodium*, (iii) *Haemoproteus*, and (iv) *Leucocytozoon* infections individually. All analyses were conducted on a restricted dataset of only resident bird species as these are more likely to have been infected close to their point of capture than species classified as vagrant, boreal migrant, or austral migrant. All analyses were run across 100 phylogenetic trees with the following parameters: burnin = 10000, thin = 100, iterations = 220000; and with priors specified as: fix = 1, V = 1, nu = 0.002, (following Hadfield (2010)). Because minimum adequate models can inflate Type I errors, I follow Salisbury (2012) and report the average results of the full models and include the change in the deviance information criterion (DIC) between a the full model and the model excluding the predictor as an additional measure to help assess the relative importance

of predictors. Using this method, a change in DIC > 2 indicates a significant effect of the predictor.

Parasite prevalence within host species elevation range

To visualize an individual bird's probability of being infected in relation to its elevation range position, I plotted infection prevalence within four sections of species ranges representing 0-25%, 25-50%, 50-75% and 75-100% of the distance between the centre and the edge of the range (Figure 4.2).

I also tested whether lineages that reached high prevalence within their hosts (≥ 15 % prevalence) were more range centred than those recorded at lower prevalence (< 15 % Prevalence within the host) using Fisher's Exact Tests on the counts of each of these infection types in their hosts inner and outer ranges, relative to the number of uninfected hosts. This analysis was restricted to hosts with a minimum of 3 captures in each of their inner and out range.

I also investigated the pattern of host abundance within their elevation range for 8 well sampled bird species for which there were a minimum of 50 captures by calculating the proportion of host species captures relative to all species captures in 200 m wide elevation bands.

Lineage prevalence within lineage elevation range

Finally, I tested if parasite lineages were recorded at higher prevalence toward the centre of their elevation ranges, using a linear mixed regression model using the R (Team 2008) package MCMCglmm (Hadfield 2010). Infection records of individual

lineages, and the captures location of the uninfected host species it was recorded in, were entered as a binary response variable, with explanatory variables of range location entered as a categorical variable (inner or outer range), and Elevation Range Size included as continuous explanatory variables. Three separate models were run for all lineages with 5 or more records belonging to *Plasmodium*, *Haemoproteus*, and *Leucocytozoon* genera of parasites.

Results

I tested a total of 2188 resident birds representing 245 species for avian malaria parasites. Of these, 109 individuals from 42 species were infected with 19 lineages of *Plasmodium*; 236 individuals from 59 species were infected with 34 lineages of *Haemoproteus*; and 311 individuals from 65 species were infected with 91 lineages of *Leucocytozoon.*

Parasite prevalence within host's elevation range

Plotting the average proportion distance from range edge for infected and uninfected birds in each 200 m elevation band (Figure 4.1) showed that birds caught toward the centres of their elevation ranges were more likely to be infected with any of the three parasite genera, classified as 'Overall infection', or just infected with *Plasmodium*, than uninfected individuals. Based on the frequency of captures in different sections of species ranges, it is also evident that the capture rate of species is greater toward the centre of their elevation range (Figure 4.2)

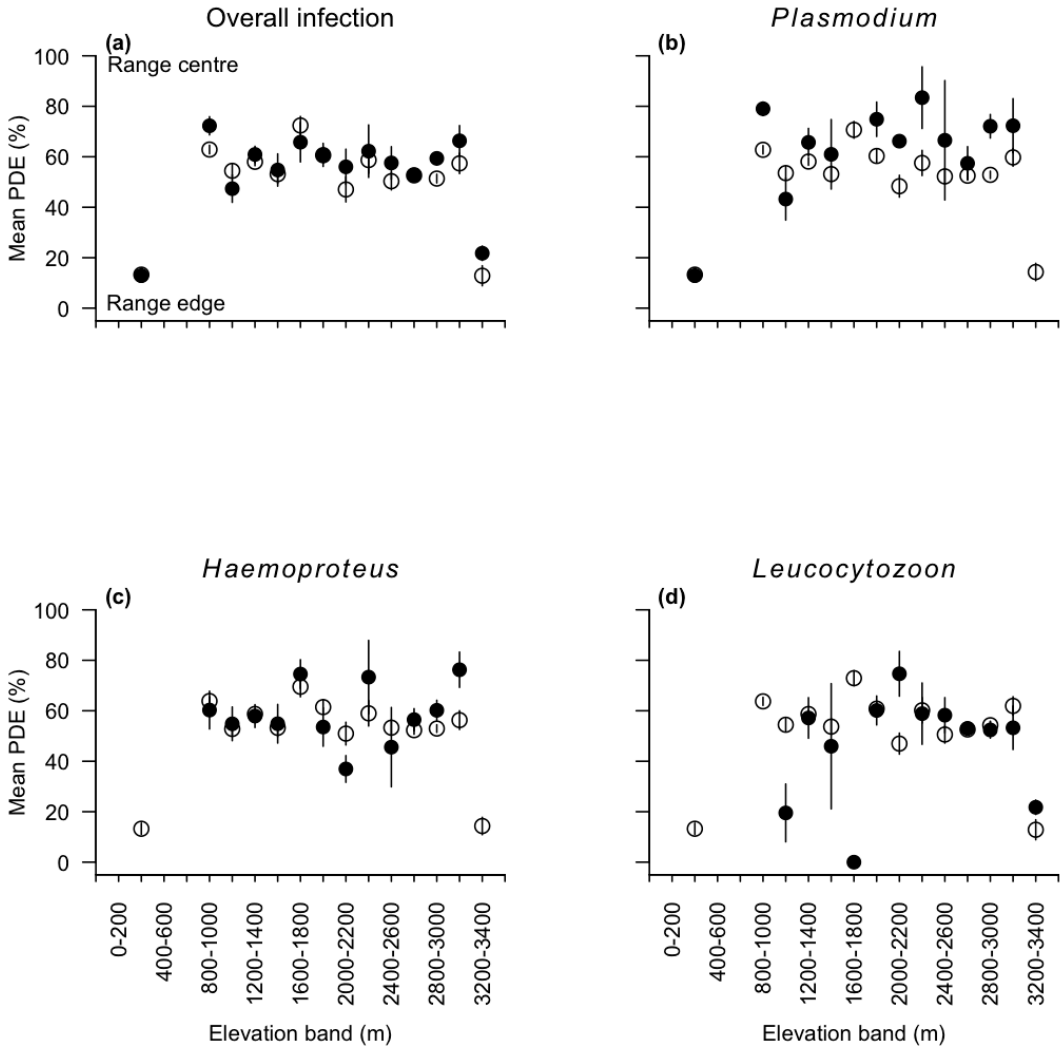


Figure 4.1 The mean proportional distance from edge for all infected (filled circles) and uninfected (empty circles) birds captured within 200 m wide elevation bands across the gradient; separately shown for (a) Overall infections (any of the three parasite genera), (b) *Plasmodium* infections, (c) *Haemoproteus* infections, and (d) *Leucocytozoon* infections. The low values of the lowest and highest range limits reflect the lack of samples at the lowest and highest elevations. Vertical bars are standard errors of the estimates

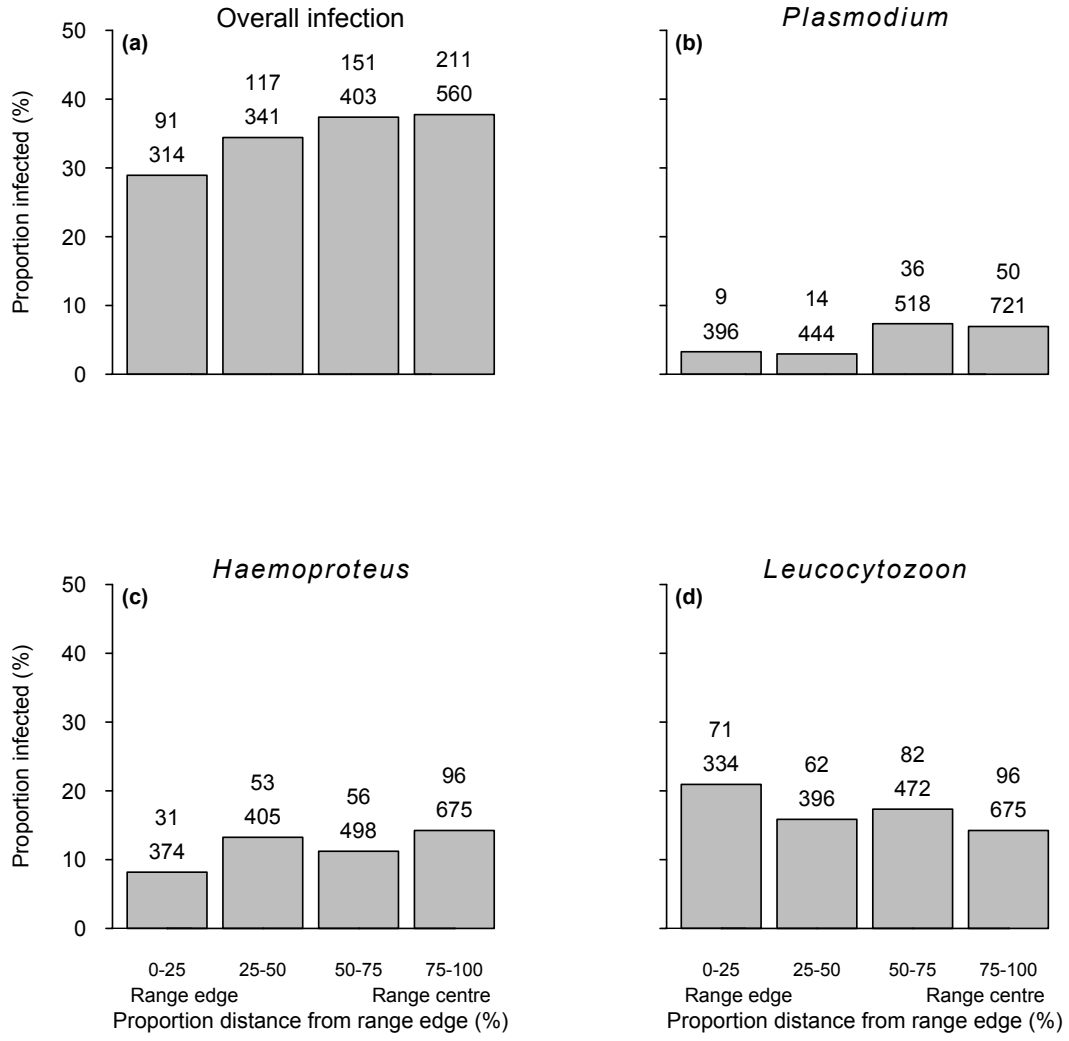


Figure 4.2 Proportion of infected birds in each 25% section of species' elevation ranges, for (a) Overall infections (any of the three parasite genera), (b) *Plasmodium* infections, (c) *Haemoproteus* infections, and (d) *Leucocytozoon* infections. Stacked numbers above each bar show the number of infected (top) and uninfected (bottom) birds in each category.

The MCMCglmm analyses showed that controlling for host species phylogenetic relatedness, individual birds were significantly more infected toward the centre of their elevation range than toward the edge when infections were classified as Overall Infection (infected with any of the three parasite genera) ($P = 0.026$) or *Plasmodium* infection ($P = 0.002$; Table 4.1). *Haemoproteus* and *Leucocytozoon* infections were

not significantly associated with elevation range position ($P > 0.05$). Capture elevation and elevation range size were not significant predictors of overall or *Plasmodium* infections in the full model, but capture elevation was a significant predictor of *Haemoproteus* and *Leucocytozoon* infections. *Leucocytozoon* infections increased with capture elevation ($P = 0.001$, slope = 0.001), whereas *Haemoproteus* infection declined with capture elevation ($P = 0.005$, slope = -0.0005).

Table 4.1 The effect of range position, capture elevation, and elevation range size on infection with avain malaria parasites for resident bird species (n = 245).

Overall infection	**Slope**	**Lower-CI**	**Upper-CI**	**ess**	***P*-value**		**Δ_{DIC}**
Intercept	-2.275	-4.171	-0.414	291	0.017	*	
Proportion distance from range edge	0.004	0.001	0.008	998	0.026	*	5.065
Capture elevation	0.0002	-0.0001	0.0004	1000	0.193		-1.878
Elevation range	0.00004	-0.0004	0.0005	997	0.851		0.325
Plasmodium							
Intercept	-4.704	-7.160	-2.381	167	0.001	*	
Proportion distance from range edge	0.010	0.004	0.016	809	0.002	*	7.888
Capture elevation	-0.000002	-0.0003	0.0003	771	0.919		0.832
Elevation range	0.0002	-0.0004	0.001	688	0.583		2.380
Haemoproteus							
Intercept	-2.996	-5.584	-0.512	165	0.016	*	
Proportion distance from range edge	0.003	-0.003	0.008	906	0.324		-3.473
Capture elevation	-0.0005	-0.001	-0.0001	681	0.005	*	2.737
Elevation range	0.00001	-0.001	0.001	692	0.927		-1.406
Leucocytozoon							
Intercept	-8.650	-12.545	-4.914	83	0.001	*	
Proportion distance from range edge	0.002	-0.006	0.009	932	0.630		2
Capture elevation	0.001	0.001	0.002	847	0.001	*	4
Elevation range	0.000	-0.0004	0.001	918	0.408		1

Overall infection DIC=1849.13, *Plasmodium* DIC=700.754, *Haemoproteus* DIC=954.646 and *Leucocytozoon* DIC=1199. Values refer to the average model output from models run on 100 phylogenies of the 245 bird species. Δ_{DIC} is the change in DIC when the term is removed from the full model run on a single phylogenetic tree. * $P < 0.05$. ess=effective sample size, CI=confidence interval.

I also tested whether more prevalent parasite lineages, defined as those that reach ≥ 15% prevalence within a host, were more range centred than less prevalent lineages. However, the incidence of each of these infection types did not differ significantly in the inner vs. outer portion of individual bird host ranges (Fisher's exact tests, all $P > 0.05$).

Lineage prevalence within lineage range

I found no significant difference in prevalence of parasite lineages in the inner vs. the outer proportion of their elevation range relative to all captures of their host species, controlling for the phylogenetic relatedness among lineages and their elevation range size (Table 4.2). Although I didn't detect a statistically significant difference, the general pattern was for a slightly higher prevalence toward the centre of their range for all lineages analysed together, and for lineages belonging to each of the three parasite genera (Figure 4.3).

Table 4.2 Summary of MCMCglmm model outputs investigating whether the prevalence of lineages varies between the inner and outer proportion of their elevation ranges. Analyses are based on lineages with 5 or more records and their prevalence within their resident host bird speices in the inner vs. outer proportions of the lineage's range.

All lineages	**Slope**	**Lower-CI**	**Upper-CI**	**ess**	***P*-value**	Δ_{DIC}
Intercept	-1.143	-3.133	0.745	1000	0.234	
Proportion distance from range edge	-0.307	-0.751	0.081	1000	0.134	3
Elevation range	-0.0004	-0.001	0.00001	1145	0.056	2
Elevation midpoint	0.0001	-0.001	0.001	911	0.768	1
Plasmodium						
Intercept	-1.404	-5.812	2.882	805.7	0.394	
Proportion distance from range edge	-0.325	-1.33	0.81	1000	0.418	-1
Elevation range	0.001	-0.002	0.003	880	0.678	-1
Elevation midpoint	-0.001	-0.004	0.003	873	0.746	-1
Haemoproteus						
Intercept	-0.825	-2.864	1.092	1000	0.41	
Proportion distance from range edge	-0.213	-1.043	0.526	1000	0.62	0
Elevation range	-0.001	-0.001	0.0004	1000	0.222	0
Elevation midpoint	0.0002	-0.001	0.001	1000	0.57	0
Leucocytozoon						
(Intercept)	0.901	-4.301	5.260	647	0.65	
Proportion distance from range edge	0.315	-0.219	1.048	715	0.312	0
Elevation range	-0.001	-0.002	-0.00003	700	0.022	2
Elevation midpoint	-0.001	-0.002	0.001	756	0.212	1

All lineages (DIC=2996), Plasmodium (DIC=597), Haemoproteus (DIC=927), Leucocytozoon (DIC=1477). Values refer to the model output from models run on a parasite phylogeny of 38 lineages. Δ_{DIC} is the change in DIC when the term is removed from the full model. ess=effective sample size, CI=confidence interval.

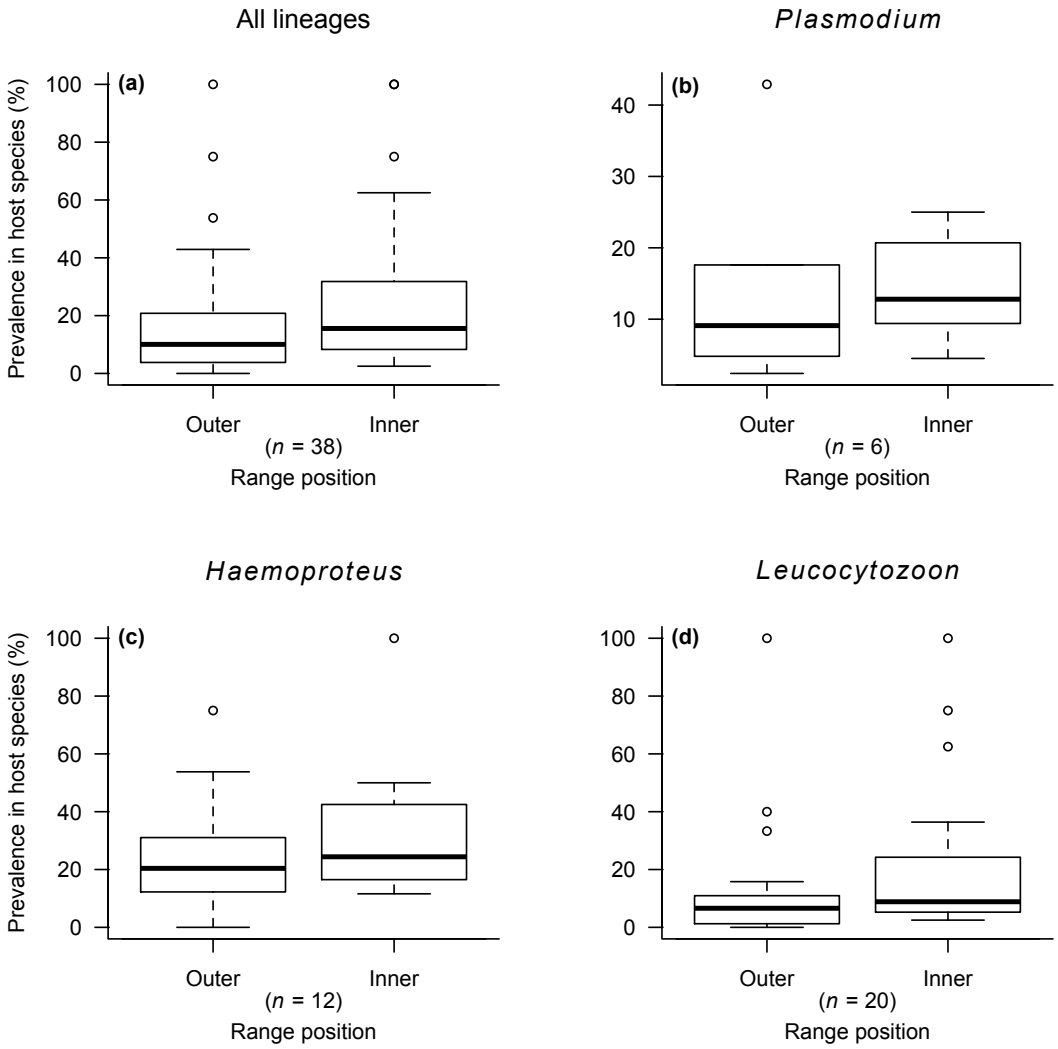


Figure 4.3 Prevalence of individual parasite lineages recorded from all captures of their host species (i.e every host they were recorded from) in the outer and inner 50 % proportions of their range; for (a) All lineages (any of the three parasite genera), (b) *Plasmodium* lineages, (c) *Haemoproteus* lineages, and (d) *Leucocytozoon* lineages. There was no significant difference in any of the analyses. Plots are Tukey box-plots, whiskers show the lowest and highest data points within 1.5 x interquartile range, sample sizes are number of lineages.

Plotting the pattern of host abundance within elevation range for the 8 most well sampled bird species, 5 of the 8 species supported a pattern of increasing abundance toward their range centre (Figure 4.4). Parasite lineages show a more varied pattern of

abundance within range consistent with the overall non-significant interaction between prevalence and range position (inner vs. outer) (Appendix Figure 4.1).

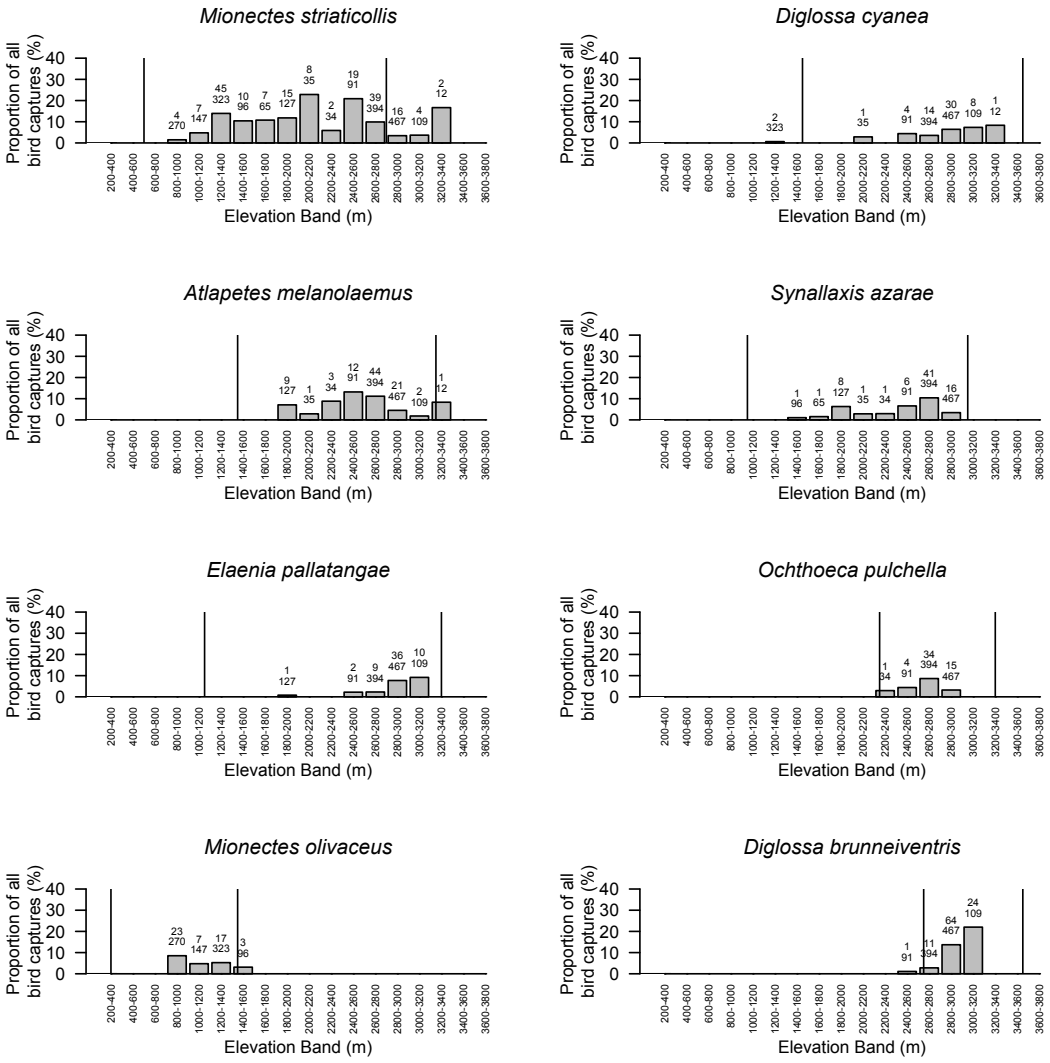


Figure 4.4 The relative proportion of species captures within their elevation ranges for 8 bird species with >= 50 captures. The number on top of each bar represents the number of focal birds species captures in that elevation band and the number below is the total number of bird species captures. The vertical lines represent the focal species lower and upper elevation range limit as recorded in Walker *et al.* 2006, with bars above or below these representing captures outside the recorded range.

Discussion

Using a continuous elevation gradient with high turnover in birds occupying restricted ranges, I recorded the prevalence of avian malaria parasites to explore patterns in parasite prevalence across host elevation ranges. I found a significant relationship between the position of a host individual within the host elevation range and infection status, in which infected birds were significantly more range-centred than uninfected birds. The key finding that overall infection prevalence and *Plasmodium* prevalence was highest toward the centre of bird species' elevation ranges is consistent with epidemiological theory for directly transmitted parasites, which predicts that host population density positively increases the probability that a parasite transmission stage will contact a susceptible host (Anderson and May 1978, Dobson 1990).

For vector transmitted parasites (that have frequency dependent transmission) it is generally assumed that prevalence should be relatively unaffected by host density and if so, that the transmission rate of vectors will be dependent upon the likelihood that a vector has previously been in contact with an infected host (McCallum et al. 2001). However, at very low density too few interactions between infected vectors and susceptible hosts may reduce the chances for successful transmission to occur (May and Anderson 1978, Anderson and May 1978). Furthermore, when vectors are generalised and carry the parasites of multiple host species, as is the case for temperate *Plasmodium* (Medeiros *et al.* 2013), host compatibility rather than vector-host-encounter rate determines the host range of parasites. If vectors are generalized with respect to blood parasites, then vectors towards the centre of a host's range are more likely to carry that particular species' parasites because they will encounter

more of the same host species within their area of activity. At the edge of host's range, generalist vectors will only contact that particular host in one direction and not the other, reducing the changes of carrying that particular species' parasites.

My analyses did not reveal any overall patterns of more prevalent lineages being more abundant within the centre of their host's ranges. This was in spite of the analyses showing greater prevalence of parasites belonging to any of the three parasite genera or *Plasmodium* genera being more range-centred, controlling for elevation range size and capture elevation. One potential explanation for lineages showing no significant relationship between their prevalence within a host species and the location of their infections within the host's range, is that the broad host associations recorded here preclude patterns of centred abundance within any particular host species. Under this scenario, the abundance of any single bird host species would not approximate the actual transmission potential of a lineage because of differences in the elevation ranges and centres of abundance of other bird species infected by the same parasite lineage. This may also explain why I did not detect an overall effect of lineages reaching greater prevalence toward the centre of their own elevation ranges. However without greater sampling of specific lineages to refine their host and environmental associations, it is difficult to conclusively determine whether lineages are abundance-centred within their own ranges. The non-significant trend of increasing prevalence of overall, *Plasmodium, Haemoproteus,* and *Leucocytozoon* lineages toward the centre of their elevation ranges is consistent with a lineage infecting multiple host species that occur across different elevations, yet over the lineage's entire recorded host range, these hosts are still range-centred.

In the genera-level analyses I detected an increase in the prevalence of *Leucocytozoon* infections toward higher elevations along the study transect. *Leucocytozoon* lineages of avian malaria are primarily transmitted by simuliid files (Valkiunas 2005), which tend to prefer cool fast flowing streams (Dalmat 1955, Hernández et al. 2009) that occur at high elevations toward the mountain top in the study site. Few bird species have their range centre toward this upper elevation limit, which may explain why even though high elevation birds are more susceptible to infection with *Leucocytozoon*, these infections are not range-centred within their hosts. These analyses also tested whether infections were located at the edge of host ranges, but this was also non-significant when elevation was included in the full model. In contrast, the prevalence of *Haemoproteus* parasites showed a negative relationship with increasing elevation when other variables were included in the full model, and *Plasmodium* parasites showed no significant variation, which may be why in the analyses with all recorded infections I didn't detect an overall effect of elevation, and by association environmental conditions, on prevalence.

The core finding that overall malaria prevalence as well as *Plasmodium* prevalence is highest at the centre of hosts' elevation ranges is consistent with epidemiological theory that predicts host population density positively increases the probability that a parasite transmission stage will contact a susceptible host (Anderson and May 1978, May and Anderson 1978). Numerous hypotheses predict that population density of hosts should be highest at the range centre due to a range of ecological and evolution hypotheses, such as central populations being sources and edge populations sinks, central populations occupying more habitats, or competition being more important at the range edge (for a detailed list and evaluation of each see Sagarin and Gaines

(2002)). Based on these scenarios, low density populations of hosts at the range edge are hypothesised to have too few interactions with parasites for successful transmission to occur (Lafferty and Holt 2003). Although this theory was developed for directly transmitted parasites, the same process should operate for vector-transmitted parasites if vector distribution is constant across a host's range. This is because if vectors in more abundant host populations have increased encounter rates with susceptible hosts, this would also increase parasite abundance through increased transmission.

Although I did not estimate host density across elevation range for the 245 bird species recorded in this study, I did investigate the pattern of host abundance within elevation range for 8 well sampled bird species for which there were a minimum of 50 captures by calculating the proportion of host species captures relative to all species captures in 200 m wide elevation bands along the gradient. When sampling effort was standardised in this way, 5 of the 8 species supported a pattern of increasing abundance toward their range centre. Moreover, one of the most comprehensive studies of tropical bird communities along a similarly sized gradient in the Peruvian Andes recorded the greatest abundance of a number of the species recorded here to occur at their elevation range centre (Terborgh 1971). The result of increased *Plasmodium* infections toward a host's range centre is consistent with hypothesised greater transmission opportunities where host abundance is highest.

This study demonstrates that an individual bird's position within a population and local environment leads to predicable variation in the prevalence of avian malaria. The recorded differences in infection prevalence within a host's range suggest that a

rarity of compatible hosts at the range edge leads to reduced parasite transmission relative to the range centre. From the perspective of parasite lineages, broad host associations or vector transmission rates appear to be independent of a range-centred peak in prevalence.

Chapter 5

Parasites and geographical range limitation in birds

Abstract

The importance of biotic interactions in limiting species geographic ranges is a fundamental, yet unanswered, question. One controversial theory is that parasites may prevent the expansion of species geographic distributions, but this has rarely been tested in natural communities. Here, I analyse the range-wide incidence of avian malaria parasites across multiple hosts, using the high species turnover of a tropical elevation gradient as a model system. Specifically I test for associations between the elevational range limits of montane birds and the opposing presence of malarial parasite lineages, and compare these patterns to those expected under null models of geographic range expansion. Based on a null model simulation approach, 16 % of the empirically recorded parasite lineages had elevation range distributions that were consistent with a potential role in limiting bird elevation ranges. These findings are robust to the definition of parasite lineages and uncertainty in elevation ranges. Taken together, these results are consistent with the hypothesis that parasites limit host distributions and suggest that biogeographic patterns associated with ecology or competition may be driven by host-pathogen dynamics.

Introduction

Understanding what limits species from occurring beyond their geographic ranges is a central theme in ecology and is essential for understanding current patterns in the distribution and diversity of species (Holt 2003, MacArthur 1972, Krebs 1994, Brown et al. 1996). Where species ranges do not end at clearly defined landscape or environmental features, but instead end along otherwise continuous environmental gradients, the mechanisms limiting species from occurring beyond their geographic ranges are not well understood (Sexton et al. 2009). Ecological constraints thought to shape such patterns arise from a variety of abiotic and biotic factors. For instance, abiotic factors may limit species ranges due to physiological constraints associated with unsuitable temperature or moisture (Kearney and Porter 2009) whereas biotic limits may arise from competition, predation, or parasitism (Chown and Gaston 1999, Sexton et al. 2009). Although these factors are not mutually exclusive, they have different implications for understanding species distributions and how these change over time.

Numerous empirical studies support roles of abiotic factors in limiting species distributions (Crozier 2004, Mott 2010, Bateman et al. 2012, Root 1988). For instance, Root (1988) showed that the northern range limits of Nearctic birds tended to coincide with temperature isotherms that were predictable according to basal metabolic rate. Recent meta-analyses across ectotherms also provide evidence that geographic range size may be broadly correlated with the breadth of species' physiological niches (Bozinovic et al. 2011). In contrast, the importance of biotic factors is less well known. Yet, a number of studies have also highlighted that closely

related species, with presumably similar environmental niches, often have abutting but non-overlapping geographic distributions (Diamond 1978, Diamond 1975, Terborgh 1971, Terborgh and Weske 1975, Tanner 1952). Comparison of these patterns to those expected under null models of random range positioning can potentially provide strong evidence for the role of negative biotic interactions in constraining range overlap (Pigot and Tobias 2013). However, in most studies, any evidence of biotic interactions is automatically attributed to ecological competition for resources (Terborgh 1971, Terborgh and Weske 1975, Diamond 1978). The possibility that parasites may be responsible for preventing geographic expansion of hosts has only rarely been properly considered (Ricklefs 2010a, Ricklefs 2010b).

Mathematical models show that in theory parasites may limit host distributions (Holt and Lawton 1993). For instance, when multiple host species are attacked by shared pathogens, the potential exists for apparent competition to limit range overlap between hosts (Williamson 1957). This has been supported by laboratory studies demonstrating that apparent competition between two hosts sharing a common parasitoid can result in the elimination of one of the host species (Bonsall and Hassell 1997). Some of the best evidence for parasites limiting host geographic distributions in the wild comes from studies examining the effects of introduced pathogens on native species that have evolved in isolation. For instance, declines of Hawaiian forest birds have occurred following the introduction of avian malaria (*Plasmodium relictum*), and many species ranges are now limited by the disease at lower elevations where the mosquito vector *Cuculoides quinquefasciatus* can survive (Atkinson and LaPointe 2009). However, the extent to which parasites may limit species ranges

among naturally occurring host-parasite assemblages, particularly where hosts and parasites have a long history of co-evolution is poorly known (Ricklefs 2010a).

Here I address this, and investigate the role of avian malaria parasites in limiting the elevation ranges of bird species along an altitudinal transect in Peru. If parasites are important in setting host range limits, then I predict that bird species and pathogens will tend to have abutting elevation ranges (Figure 5.1).

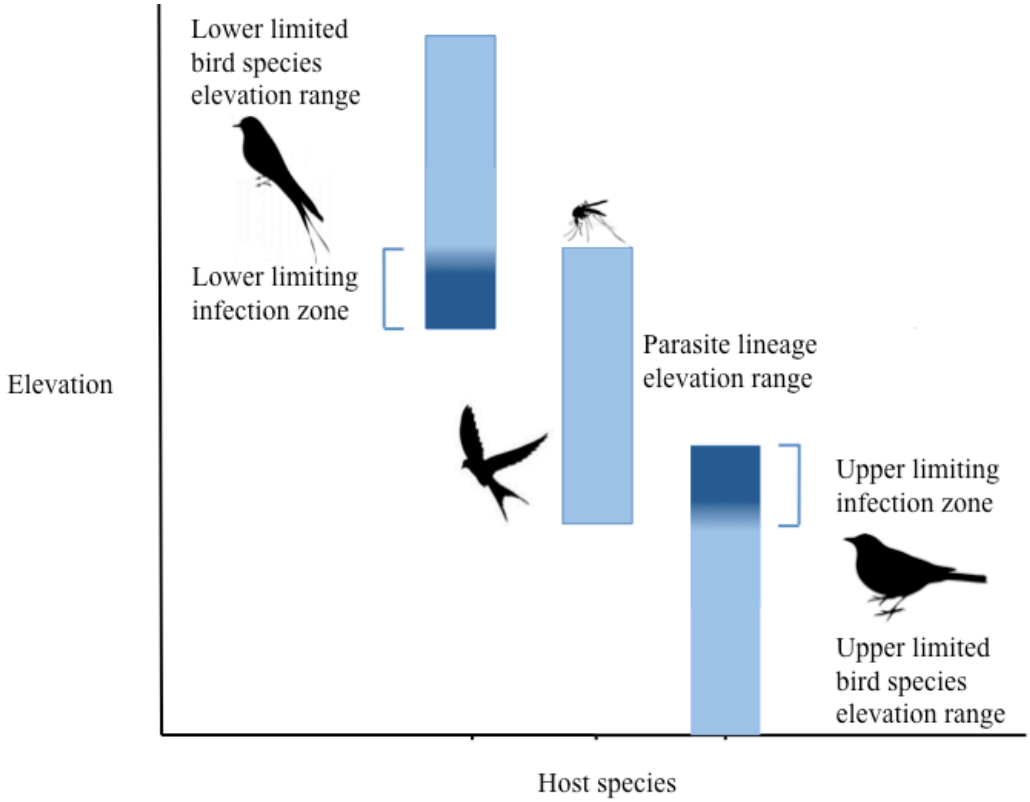


Figure 5.1 Schematic of the potential for a parasite lineage to limit the upper or lower elevation ranges of certain host species.

Specifically, I expect that if a bird species is unable to tolerate a particular parasite lineage then its elevation range should end where the distribution of the parasite starts. Cases where a bird species reaches its range limit at, or within a short distance

of, the parasite's elevation range edge thus potentially provide evidence for parasite induced range limits. In contrast, bird species that can tolerate a particular parasite should be able to penetrate further into the parasite range. In cases where a parasite is specialized on a particular (tolerant) avian host species, then the host and parasite may have coincident range limits. This latter scenario would not, however, count as range abutment: range abutment only includes cases where opposing range limits meet i.e. the upper limit of the bird coincides with the lower limit of the parasite. Bird ranges may abut the distribution of a parasite lineage simply by chance. I therefore develop a novel null-model of geographic range placement and for each parasite lineage test the evidence for non-random range abutment.

If parasites are important in setting host range limits in natural systems, then susceptible bird species should have range boundaries that coincide with the distribution limit of the 'novel' parasite lineage to which they are not adapted. Depending on how lethal the parasite is, this may lead to contrasting patterns of infection at the bird range edge. If the progression from infection to death is a protracted process, then individuals occurring at the range edge will tend to be infected. On the other hand, if infections are highly pathogenic, then infected individuals might be difficult to detect close to the range boundary (i.e. any infected individuals will be dead and so will not be recorded). To provide an initial assessment of this, for those birds whose geographic ranges abut a parasite lineage I investigated patterns of infection.

Materials and Methods

Designation of parasite lineages and elevation range abutment

I define parasite lineages based on any variation in the 416 bp region of the cytochrome b gene. Previous studies have revealed that lineages differing by <1 % sequence divergence often have unique host associations (Bensch et al. 2000, Ricklefs and Fallon 2002, Bensch et al. 2004). For instance, two avian malaria lineages which differ by 0.2% at the cytochrome b gene are almost entirely restricted to one bird species each, even though transmission between them is possible (Reullier et al. 2006).

I used the recorded distribution of parasite lineages from captured birds along the elevation gradient to define the position of parasite upper and lower range limits. Defining lineage range boundaries is challenging (Fortin et al. 2005). In particular, the position of parasite range boundaries may be obscured by low infection prevalence and the ability of birds to move parasites beyond transmission zones. To ensure that my results are robust, I used a number of different criteria to define range boundaries. First, upper and lower limits were simply defined as the maximum and minimum elevation record of each lineage. Second, for each lineage I used the upper and lower boundary of the 95% confidence interval of elevation records. This later method is sensitive to the normality of occurrences, which improves with the number of samples. To explore the extent to which parasite elevation ranges may be determined by sampling effort I quantified the number of bird captures and parasite lineages with range edges within 50 m wide elevation bands along the gradient.

Range abutment was defined as cases when the upper range limit of a bird coincided with the lower range limit of the parasite lineage. Instances where the parasite and bird share their upper (or lower) range boundaries does not count as range abutment: in this case the parasite and bird would have partially overlapping ranges. While the estimated position of parasite range boundaries can take any value, published bird range limits are available at a resolution of 50 m (Walker et al. 2006). To incorporate the uncertainty in the position of avian range limits and accuracy in recording bird capture elevations (from which parasite range boundaries were calculated) I used a buffer zone around the parasite range boundary to determine range abutment. Specifically, I assigned a positive case of range abutment if the upper limit of the bird range fell within a 100 m band centred on the lower limit of the parasite. To assess the influence of this arbitrary cut-off, I repeated the analysis using a 200 m buffer zone. As a final test, I used an internal buffer zone i.e. a 100 m above and below the parasite lower and upper range limit respectively. The rational behind this final measure is that vectors may spread the influence of pathogens beyond their distribution, although if this is occurring then this could already be underlying the maximum and minimum parasite records. Because the effects of parasites may differ across elevation I separately tested evidence of range abutment for parasite upper and lower range boundaries.

Null model simulations

To test whether the frequency of range abutment differs from that expected due to chance I generated artificial bird ranges using a null simulation approach. Specifically, for each bird species I selected at random two 50 m elevation bands from across empirically sampled sections of the gradient (300-3400 m), assigning the

higher value as the upper range boundary and the smaller value as the lower range boundary. Under this approach a bird may occasionally occur within only a single elevation band. This is justified because some real species ranges on the gradient also only encompass a single elevation band (Walker et al. 2006). Many empirical species ranges extend beyond the lower (300 m) or upper (3400 m) limit to the gradient and randomizing the position of these range edges would artificially inflate the number of range boundaries found at intervening elevations, thus biasing the analysis against detecting significant range abutment. For species with truncated ranges, I therefore only selected a single range boundary position (i.e. corresponding to the non-truncated edge). Having generated the artificial bird ranges I counted the number of range boundaries abutting the upper and lower range boundary for each parasite lineage. I repeated this whole procedure 10000 times to generate a null expectation for the number of cases of range abutment. I identified 'range limiting parasites' (RLP's) as those lineages with upper or lower range boundaries that had a higher frequency of avian range abutment than expected by chance (defined as exceeding the 95 % CI of the simulated data).

Range boundaries of hosts and parasites may show significant range abutment if both are limited by an extrinsic environmental boundary (e.g. vegetation zone transition). To determine whether autecological factors may be important in influencing patterns of range abutment, I compared the overall frequency of observed and simulated avian and parasite range limits along the gradient. Results of this analysis may identify locations on the gradient where more birds reach their range limits than expected by chance, irrespective of the presence of parasite lineages.

If parasite lineages have been 'over split' then this could lead to either the number of 'range limiting parasites' being over- or under-estimated, depending on which lineages have been split. To explore this possibility I tested whether RLP's were phylogenetically clustered on the parasite phylogeny by treating RLP status as a binary trait and estimating the D statistic, which provides a measure of phylogenetic signal (Fritz and Purvis 2010). D indicates the standardized sum of the estimated nodal change along the edges of a phylogeny, with higher values thus indicating more labile traits. Specifically, a value of D = 0 indicates a trait that is phylogenetically conserved (as expected under a Brownian threshold model) while a value of 1 indicates a trait that is random with respect to phylogenetic position (random). A value of D smaller or greater than 1 indicates traits that are more highly conserved and more overdispersed respectively.

The null model approach that I employ here differs from standard models of stochastic range placement in which species ranges are randomly shuffled within a bounded domain (e.g. upper and lower limit of the elevation transect) (Colwell and Lees 2000). This latter approach, often termed the 'mid-domain model' (MDE) has been most widely applied to study patterns of species richness (see Appendix Paper on MDE in Birds from the Manu Biosphere Reserve) and typically treats the empirical distribution of species range sizes as fixed (Hawkins et al. 2005). However, because here I am interested in whether the position of range boundaries, and not species ranges *per se*, depart from random expectation, the more appropriate null model is one in which range boundaries are randomized and range size is allowed to vary.

These methods were used to test the following predictions:

Prediction 1: Susceptible hosts' ranges abut the limiting parasite range

Prediction 2: Bird species' ranges end at or within a short distance of a parasite lineage range and are also infected in these zones

Results

I captured a total of 2188 birds representing 245 species. Among these I recorded 570 overall infections (any of the three parasite genera), which consisted of 109 *Plasmodium* infections, representing 19 lineages; 236 *Haemoproteus* infections, representing 34 lineages; and 311 *Leucocytozoon* infections representing 91 lineages. 86 birds had multiple infections. Of these, there were 9 *Plasmodium*, 21 *Haemoproteus* and 38 *Leucocytozoon* unique lineages that were available for analysis i.e. with a minimum of two infection records.

Prediction 1: Susceptible hosts' ranges abut the limiting parasite range

Based on the most restrictive of the range simulation analyses, in which range limited bird species were classified as having their ranges ending within 100 m of parasite elevation zones (defined as the parasite lineages maximum and minimum elevation records), I found support for 7 parasite lineages limiting bird upper elevation ranges and 5 lineages limiting birds lower ranges (Table 5.1). Among these, 4 (19 %) of the recorded *Haemoproteus* lineages, and 3 (8 %) of the *Leucocytozoon* lineages had more bird upper elevation ranges ending than would be expected by chance (Figure 5.2a). When I tested the potential importance of lineages in limiting the lower elevation ranges of bird species a separate combination of 2 *Haemoproteus* lineages,

and 3 *Leucocytozoon* lineages had more bird lower elevation ranges ending than would be expected by change (Figure 5.2b). Using the 95 % confidence interval of all lineage records and varying the size of the infection zone within which birds with their elevation range edges were classified as limited increased the number of range limiting lineages and demonstrates how the classification is sensitive to the size of range limitation zones (Table 5.2, Appendix Figure 5.1).

Table 5.1 Range limiting lineages with two or more records and defined as those with more empirical bird ranges ending at parasite infection zones than randomly simulated bird ranges.

	Plasmodium		*Haemoproteus*		*Leucocytozoon*	
Infection zone	**No. (%)**	**Lineages**	**No.**	**Lineages**	**No.**	**Lineages**
Upper range limiting lineages						
100 m into parasite minimum recorded elevation	0		4 (19.0%)	H_BASSI21, H_CHLCA16, H_EUPXA10, H_TANCH25	3 (7.9%)	L_AULCO3, L_CONAR57, L_PIPIN120
100 m into parasite lower elevation limit (using 95% CI's)	2 (22.2%)	P_CATUS1, P_TURNI2	6 (28.6%)	H_BASCO20, H_BASSI21, H_CHLFL37, H_DIGBR17, H_EUPXA10, H_TANCH25	9 (23.7%)	L_ATLME80, LBASLU104, L_CHLRI43, L_AULCO3, L_DIGBR35, L_MYARA32, L_MECST6, L_CONAR57, L_PIPIN120
200 m into parasite minimum recorded elevation	7 (77.8%)	P_MYARA13, P_AUTRU15, P_DYSME16, P_CATUS1, P_MIOOL14, P_MYCHI5, P_PHYOP7	14 (66.7%)	H_IRIAN3, H_BASCO20, H_BASSI21, H_CHLCA22, H_CHLCA24, H_CHLCA16, H_CHLFL37, H_DIGBR17, H_DIGGL4, H_EUPXA10, H_MIOST8, H_TANCH25, H_TANAR27, H_THREP30	12 (31.6%)	L_ATLME80, L_BUTMO90, L_HEMAT126, L_BASLU104, L_CHLRI43, L_IRIJE30, L_MYIME127, L_AULCO3, L_MYARA32, L_CONAR57, L_DIGSI116, L_PIPIN120
200 m into parasite lower elevation limit (using 95% CI's)	7 (77.8%)	P_AUTRU15, P_DYSME16, P_CATUS1, P_MIOOL14, P_MYCHI5, P_PHYOP7, P_TURNI2	13 (61.9%)	H_IRIAN3, H_BASCO20, H_BASSI21, H_CHLCA22, H_CHLCA24, H_CHLFL37, H_DIGBR17, H_DIGGL4, H_EUPXA10, H_MIOST8, H_TANCH25, H_TANAR27, H_THREP30	11 (28.9%)	L_ATLME80, L_HEMAT126, L_ATLME49,L_BASLU104, L_CHLRI43, L_MYIME127, L_AULCO3, L_DIGBR35, L_MYARA32, L_MECST6, L_CONAR57
100 m above and below minimum recorded elevation	2 (22.2%)	P_MYARA13, P_CATUS1	6 (28.6%)	H_BASSI21, H_CHLCA24, H_CHLFL16, H_CHLFL37, H_EUPXA10, H_TANCH25	6 (15.8%)	L_CHLRI43, L_IRIJE30, L_AULCO3, L_MYARA32, L_CONAR57, L_PIPIN120
100 m above and below lower elevation limit (using 95% CI's)	2 (22.2%)	P_CATUS1, P_TURNI2	8 (38.1%)	H_BASCO20, H_BASSI21, H_CHLCA24, H_CHLCA16, H_CHLFL37, H_DIGBR17, H_EUPXA10, H_TANCH25	12 (31.6%)	L_ATLME80, L_ATLME49, L_BASLU104, L_CHLRI43, L_IRIJE30, L_AULCO3, L_DIGBR35, L_MYARA32, L_CONAR57, L_DIGSI116, L_PIPIN120, L_TURCH70
Lower range limiting lineages						
100 m into parasite maximum recorded elevation	0		2 (9.5%)	H_CHLCA22, H_CHLCA24	3 (7.9%)	L_ARRTO113, L_ATLME41, L_TURSE67
100 m into parasite upper elevation limit (using 95% CI's)	0		4 (19.0%)	H_ATLME35, H_CHLCA22, H_CHLCA24, H_MIOST8	3 (7.9%)	L_ARRTO113, L_ATLME41, L_TURSE67
200 m into parasite maximum recorded elevation	3 (33.3%)	P_AUTRU15, P_DYSME16, P_CATUS1	4 (19.0%)	H_ATLME35, H_MYARA9, H_CHLCA22, H_CHLCA24	4 (10.5%)	L_ARRTO113, L_ATLME41, L_TURSE67
200 m into parasite upper elevation limit (using 95% CI's)	3 (33.3%)	P_AUTRU15, P_DYSME16, P_CATUS1	4 (19.0%)	H_ATLME35, H_CHLCA22, H_CHLCA24, H_MIOST8	3 (7.9%)	L_ARRTO113, L_ATLME41, L_TURSE67
100 m above and below maximum recorded elevation	2 (22.2%)	P_AUTRU15, P_DYSME16,	3 (14.3%)	H_BASSI21, H_CHLCA24, H_CHLFL37	2 (5.3%)	L_ARRTO113, L_ATLME41
100 m above and below upper elevation limit (using 95% CI's)	3 (33.3%)	P_AUTRU15, P_DYSME16, P_MYCHI5	3 (14.3%)	H_ATLME35, H_CHLCA24, H_CHLFL37	2 (5.3%)	L_ARRTO113, L_ATLME41

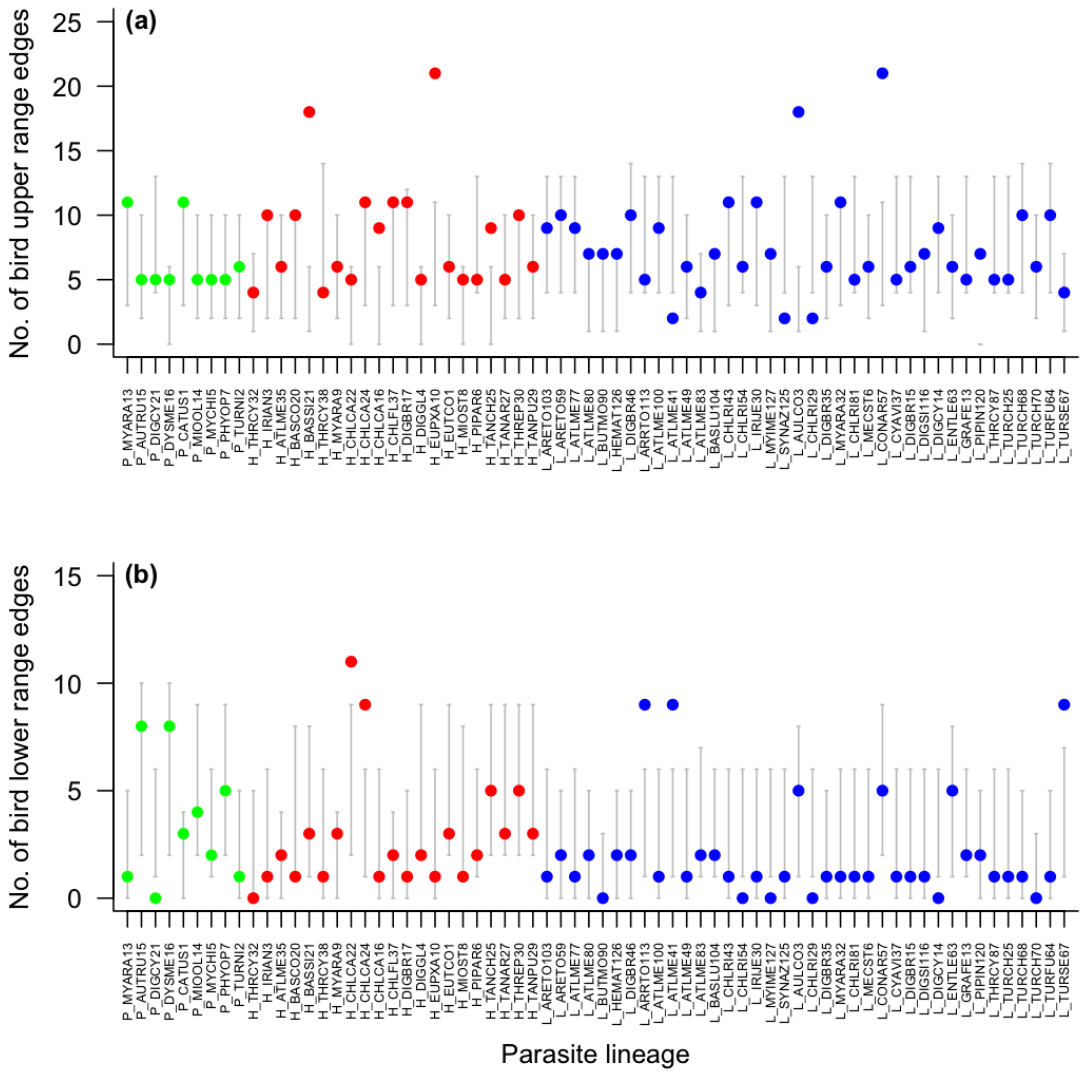


Figure 5.2 The number of empirical (filled circles) and expected (95% CI vertical bars) bird species' ranges abutting parasite lineage ranges. Circles falling outside the estimated range of expected values show parasite lineages that abutted significantly more (above) or fewer (below) bird species' than expected by change. Results are shown for (a) upper range limiting parasite lineages, and (b) lower range limiting parasite lineages for each of the three major parasite genera (*Plasmodium* [green], *Heamoproteus* [red], and *Leucocytozoon* [blue]). Parasite infection zones were defined as 100 m into the maximum and minimum elevation of all recorded infections.

To assess whether autecological boundaries may be important in influencing these results, I compared the distribution of avian richness and range limits across the elevation gradient to those expected under the null model (Figure 5.3) (results for parasite lineages are provided in Appendix Figure 5.2). The null model is able to produce the major trend in avian richness along the gradient (Figure 5.3b). In the majority of 50 m elevation bands, the number of birds reaching either their upper or lower range limit is no greater than expected by chance (Figure 5.3c & 5.3d). I note that there is a zone, centered on an elevation of 1500 m, where the number of bird species reaching their upper range limits is greater than expected by chance (Figure 5.3d).

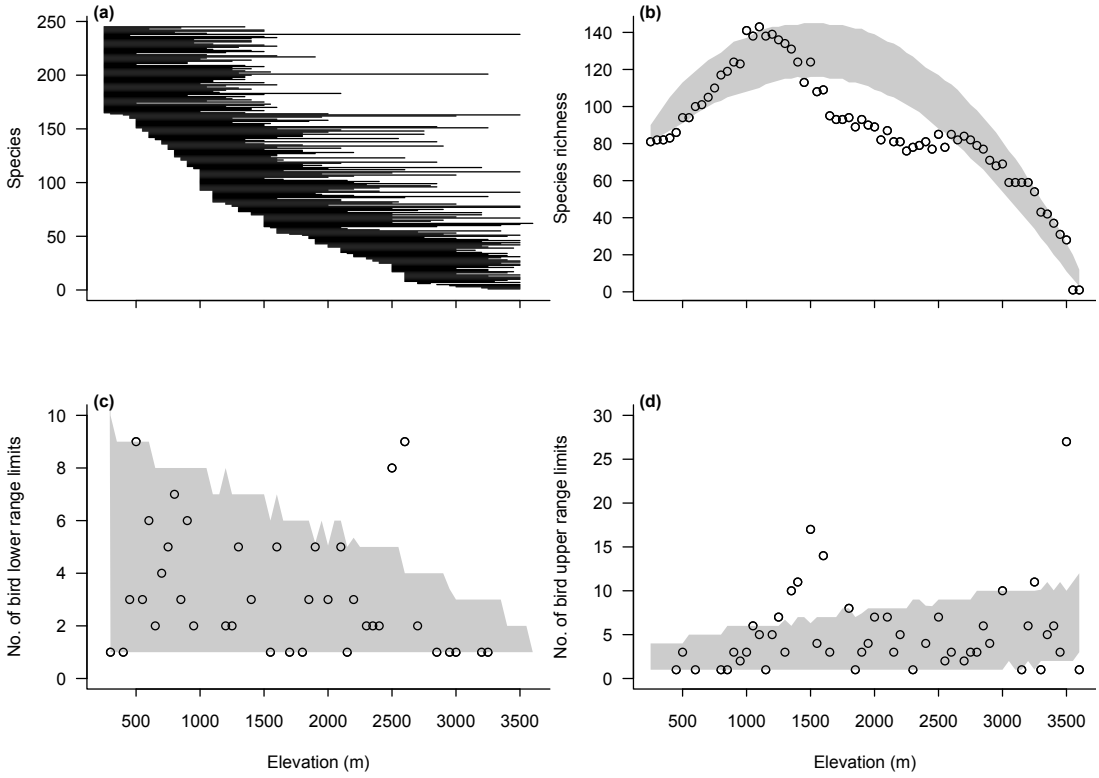


Figure 5.3 Elevation ranges (black lines) of the 245 bird species used to test if parasites may be limiting host ranges (a); the relationship between species richness and elevation for empirical (open circles) and simulated (grey shading, 95% CI) data (b); the observed (open circles) and simulated (grey shading) number of lower (c) and upper (d) bird range limits in each elevation band.

Prediction 2: Bird species' ranges end at or within a short distance of a parasite lineage range and are also infected in these zones

In total 13 of the 68 parasite lineages (i.e. 19 %) had elevation range distributions and the occurrence of infections in birds that were consistent with any one of the three classification of range limitation investigated here (Table 5.2). Among these, I recorded 6 lineages that may be limiting the upper elevation of bird species, and 7 lineages that may be limiting the lower elevation range limits of birds. In other words, I found limited support for parasites limiting the elevation ranges of birds based on the most conservative criteria. Under this classification, range limiting lineages were made up entirely of *Haemoproteus* and *Leucocytozoon* genera, whereas there was no support for a role of *Plasmodium* lineages in limiting bird elevation ranges. Furthermore, there was little phylogenetic signal in the distribution of RLP's and a random model could often not be rejected (Table 5.3).

Table 5.2 The number and identity of range limiting lineages in each of the three investigated infection zones using a more conservative range limiting criterion i.e. range abutment and cases of infection were recorded within infection zones.

	Plasmodium		Haemoproteus		Leucocytozoon	
Infection zone	**No. (%)**	**Lineages**	**No. (%)**	**Lineages**	**No. (%)**	**Lineages**
Upper range limiting lineages						
100 m into parasite minimum recorded elevation	0		0		1 (2.6%)	L_AULCO3
100 m into parasite lower elevation limit (using 95% CI's)	0		0		0	
200 m into parasite minimum recorded elevation	0		3 (14.3%)	H_MIOST8, H_TANCH25, H_THRCY38	2 (5.3%)	L_ARRTO59, L_AULCO3
200 m into parasite lower elevation limit (using 95% CI's)	0		1 (4.8%)	H_THRCY38	1 (2.6%)	L_ARRTO59
100 m above and below minimum recorded elevation	0		0		1 (2.6%)	L_AULCO3
100 m above and below lower elevation limit (using 95% CI's)	0		0		1 (2.6%)	L_AULCO3
Lower range limiting lineages						
100 m into parasite maximum recorded elevation	0		0		1 (2.6%)	L_HEMAT126
100 m into parasite upper elevation limit (using 95% CI's)	0		0		1 (2.6%)	L_HEMAT126
200 m into parasite maximum recorded elevation	0		2 (9.5%)	H_CHLCA22, H_PIPAR6	4 (10.5%)	L_HEMAT126, L_ARRTO113, L_ATLME83, L_GRAFE13
200 m into parasite upper elevation limit (using 95% CI's)	0		2 (9.5%)	H_CHLCA22, H_PIPAR6	3 (7.9%)	L_HEMAT126, L_MYIME127, L_GRAFE13
100 m above and below maximum recorded elevation	0		0		1 (2.6%)	L_HEMAT126
100 m above and below upper elevation limit (using 95% CI's)	0		0		1 (2.6%)	L_HEMAT126

Table 5.3 Results of tests measuring phylogenetic signal (Estimated D) in range limiting lineages across a range of infection zones.

Infection zone	Estimated D	Probability of E(D) resulting from random structure	Probability of E(D) resulting from Brownian phylogenetic structure
UpLim100mCI	0.9	0.4	0.0
UpLim100mMaxMin	0.8	0.3	0.2
UpLim200mCI	0.9	0.2	0.0
UpLim200mMaxMin	0.9	0.3	0.0
LowLim100mCI	1.3	0.9	0.0
LowLim100mMaxMin	1.6	0.9	0.0
LowLim200mCI	1.3	0.9	0.0
LowLim200mMaxMin	1.2	0.8	0.0

The extent to which parasite elevation ranges may be determined by sampling effort

Plotting the elevation ranges of parasite lineages shows that the distribution of parasite ranges are clustered along the gradient and show considerable variation in extent (Appendix Figure 5.2).

Although sampling effort in each of the 50 m wide elevation bands showed considerable variation, elevations with greater bird species captures did not correspond to areas of greater bird species turnover (Appendix Figure 5.3). For parasite lineages however there was a significant relationship between sampling effort and lineage turnover (Appendix Figure 5.4).

Based on estimates of species richness of parasites lineages recovered from sampled avian hosts, the recovered lineages provided a close match to the predicted number of lineages within the community (Appendix Table 5.1 & Appendix Figure 5.5).

Evidence for range limitation based on all lineage records and their position within bird species' elevation ranges

This analysis used all records of infections within hosts and recorded whether birds had infections coincident with a parasite lineage at the birds range edge, even if this was a unique parasite only recorded once in the study.

When parasite range limits were defined as their maximum and minimum elevation records and infection zones were classified as 100 m into these, 5 bird species had lower elevation ranges that ended within parasites' upper elevation infection zone and also had infection occurring within these. Four bird species had their upper ranges ending in parasites' lower infection zones and also had infection recorded within these (Appendix Table 5.2). When parasite limits were based on the maximum and minimum elevation records and infections zones were classified as 200 m into these, 14 bird species had lower elevation ranges that ended within parasites' upper elevation infection zone and also had infection occurring within these. A separate 14 bird species had their upper range sending in parasites' lower infection zones and also had infection recorded within these (Appendix Table 5.3).

Using 95% CI's to define parasite elevation ranges limits and an infection zone 100 m into parasite elevation range limits, only 5 species had their lower ranges ending at parasite upper elevation infection zones and also had infection occurring within these. Three bird species had their upper ranges ending in parasite lower elevation infection zones and also had infections recorded within these (Table 5.4). Using a 200 m wide infection zone, 13 bird species had their lower elevation ranges occurring within parasite upper elevation infection zones and also had infections occurring within

these. Eleven bird species had their upper elevation range ending within parasites' lower elevation infection zones and were also infected within these (Appendix Table 5.5).

Finally, when parasite infection zones were classified as 100 m above and below the maximum and minimum elevation recorded for each lineage, 7 bird species had lower elevation ranges that ended within parasites' upper elevation infection zone and were also infected in this zone. Four bird species had their upper range ending in parasite lower infection zones and also had infection recorded within these. (Appendix Table 5.6).

Discussion

That parasites may play an important role in limiting the geographic distributions of host species remains controversial and broad-scale tests of this idea in the wild are rare. Here, I used a null model based approach to explore whether avian malaria parasites may be responsible for limiting host geographic ranges along a continuous elevation gradient. I found partial support for this hypothesis, and identified a number of cases where the number of bird species abutting the elevation range of a parasite lineage was greater than expected by chance. Although the hypothesis that susceptible birds will have their ranges ending and be infected at the edge of parasite ranges was not widely supported from the empirical data, there was nevertheless widespread support for the hypothesis that parasites may limit species ranges but that infected individuals are unlikely to be detected.

Across the elevation gradient there were numerous cases where the upper or lower range limit of a bird species abutted the range of a parasite lineage. By comparing the frequency of range abutment to that expected under a stochastic model of random range edge placement I was able to show that these patterns cannot be explained simply by chance. Instead, for particular parasite lineages, significantly more avian species have ranges ending at the boundary of the parasites range than predicted by the null model. Although the frequency of range abutments was sensitive to the particular way in which parasite range edges were defined, the finding of non-random range abutment was robust to these different definitions. Under the most conservative criteria (recording bird species with ranges ending within 100 m of parasite maximum and minimum elevation ranges) 15% of *Leucocytozoon* lineages, and 28.5 % of *Haemoproteus* lineages had range boundaries coinciding with more avian range limits than expected by chance. Although these results have an obvious weakness in that defining parasite ranges based on as few as two records may not accurately reflect actual host ranges, based on the empirical records reported here and from widely reported instances of parasites occurring across a broad host range elsewhere in the literature, even these lineages with as few as two records, may accurately reflect their elevation distributions given the overall extensive sampling of birds across the elevation gradient. However, the only way to conclusively determine parasite ranges is through greater sampling over a longer period of time.

One explanation for the high frequency of range abutment between avian species and particular parasite lineages is that these parasites directly limit range expansion via their effects on host fitness or death rates. Parasites occurring within a primary host may occur at a relatively benign virulence due to a long history of host adaptation and

parasite counter adaptations. However, the same parasite may be highly virulent at the range edge of a secondary host where there is contact (Ricklefs 2010a). This may be most likely where the secondary host is closely related enough to be infected but hasn't had the history of exposure to develop virulence resistance, or simply if the host is unable to mount an immune response. Ricklefs & Bermingham (2007) suggested that this mechanism may be responsible for the low rates of secondary sympatry inferred amongst Lesser Antillean birds. The results of the current analysis provide evidence that the effects of parasites in limiting avian ranges may be more general and are also present in highly diverse continental systems.

Classic studies along tropical montane gradients revealed that relatively few avian species have range limits coinciding with ecotones and that species instead appear to drop out idiosyncratically across space (Terborgh and Weske 1975). Here I statistically confirm this pattern, showing that with a single exception, at around 1500 m, the number of birds reaching their range limits at any given elevation is no greater than expected by chance. This apparent idiosyncrasy in the location of avian range limits was interpreted as evidence that diffuse competition is the primary factor limiting range expansion. Indeed, almost two thirds of the elevation limits of Andean birds were attributed to competition (Terborgh and Weske 1975). The results of the current analysis provide an alternative explanation and suggest that range limits may instead be driven by host-pathogen dynamics. Thus while overall patterns of range limits may appear largely random, this disguises the high concentration of range abutments where species come into contact with novel pathogens.

The possibility that a host species may harbor parasites that are lethal or have a strongly deleterious effects on the fitness of a close relative is supported by examples showing effects of introducing novel host or parasites on native populations. For example, the eastern grey squirrel (*Sciurus carolinensis*) introduced to the United Kingdom harbours a viral pathogen that is lethal in the native red squirrel (*Sciurus vulgaris*) (Tompkins et al. 2003). Similarly, signal crayfish (*Pacifastacus leniusculus*) also introduced from North America into the United Kingdom, is a reservoir host of a water mould (*Aphanomyces astaci*) that is the causative agent of crayfish plague, which is highly lethal in the native white–clawed crayfish (*Austropotamobius pallipes*) (Alderman et al. 1990). Furthermore, worldwide introductions of chytrid fungus (*Batrachochytrium dendrobatidis*) have decimated many native frog populations (Kilpatrick et al. 2010), although some species are known to experience little effect and instead act as reservoirs for the disease (Woodhams et al. 2007). Among naturally occurring host and parasite associations, there is also limited support for a role of parasites limiting host geographic ranges. For instance, a study of trematode infections in a gastropod snail, *Lymnaea stagnalis* showed that infections increased as the species approached one of its geographical range limits (Briers 2003). However, in this case, it is not know if this was due to contracting infections from a neighbouring species or whether this just reflected changes in the hosts own unique parasite fauna within its range. Although these studies haven't directly tested the parasite range limitation hypothesis as outlined here, they nevertheless suggest that parasites have the potential to limit host geographic ranges.

While the patterns of non-random range abutment indicate a role for parasites in limiting host ranges, there was little evidence that susceptible hosts were more infected at their range edge as might be expected. A possible explanation for this, is that range limiting parasites are lethal or impose such strong fitness consequences to hosts that infected individuals are unlikely to be active and thus captured (Valkiunas 2005, Asghar et al. 2011). Even if dead hosts were detected in the field, attributing the cause of death unequivocally to parasites would be extremely difficult to demonstrate (McCallum and Dobson 1995). A further possibility for not detecting many infected individuals at the range edge is simply that host density is greatly reduced in these areas. More intensive sampling at range edges is required to exclude this sampling artefact (see Chapter 3 & 4).

Although reduced host fitness is a key assumption of parasites limiting host distributions, few studies have addressed whether hosts may suffer increased impacts of parasites at their range edges. In this study I was unable to test whether birds that were infected at their range edge were more immunocompromised or had reduced fitness, compared to birds captured within other parts of their range. However medication experiments on wild birds have shown that *Haemoproteus* spp. can induce considerable fitness consequences to the host (Marzal et al. 2005, Merino et al. 2000). Furthermore, on Hawaii introduced avian malaria is implicated in setting the range boundaries of many native species (Warner 1968, Vanriper et al. 1986) and results from experimental infections reveal that mortality may be as high as 65% following exposure to a single infective mosquito bite (Atkinson et al. 2000).

Within the present study site the only conclusive way to determine virulence would also be to undertake experimental infections and record host responses to different pathogens, from within and immediately outside their elevation ranges. However, given the wide diversity of hosts, parasites, and their elevation range distributions, this is neither ethical nor practical (Ricklefs 2010a). A more tractable approach would be to undertake aviary transplant experiments in which hosts are housed in areas just outside their species current distributions, and then monitored for infection and possible fitness consequences (Ricklefs 2010a). Under such an experimental manipulation, control birds may even be housed in the same aviaries but treated with antimalarial drugs such as primaquine (Marzal et al. 2005, Merino et al. 2000, Martinez-de la Puente et al. 2010) or Malarone^{TM} (Knowles et al. 2010a) which have previously been successful in clearing certain strains of avian malaria in wild birds. Results from hosts that contract infections, including the fitness consequences associated with these, could then be combined with information on host infections within their range, providing the most accurate insight into whether a parasite may be limiting a host from occurring beyond its geographical range.

Another method for inferring the impact of parasites on hosts, and one that is the logical next step to the current study, is to recorded parasitemia levels (number of infected blood cells) in host bird species. If parasites play a role in limiting hosts then the expectation would be that parasite load should increase sharply toward their elevation range limits as they come into contact with limiting parasites. One caveat however, is that parasitemia is highly variable both within and between host species, showing idiosyncratic variation with time since infection (Atkinson et al. 1995) and seasonality (Bensch et al. 2007).

Parasites have been implicated as a major factor limiting the geographic expansion of host species but broad-scale evidence from natural systems is largely lacking. Using a novel simulation procedure I have shown that across an elevational gradient, more avian species have ranges ending at parasite infection zones than would be expected by chance. This pattern of non-random range abutment between parasites and hosts is consistent with a model in which novel parasites are responsible for preventing host range expansion. Furthermore, in a number of cases of range abutment, birds at the edge of their range were infected with the novel parasite. Taken together, the results suggest that parasites may have a much more important role in limiting host geographic ranges than is commonly assumed.

Chapter 6

General Discussion

6.1 Avian malaria in the montane tropics

The extent to which parasites may play a role in structuring ecological communities has been debated for decades. In this thesis I set out to address this by recording patterns in the distribution and diversity of hosts and their parasites and the mechanisms underpinning these relationships. The research was carried out along a pristine elevation gradient in the Peruvian Andes, spanning 250 - 3500 m and harbouring among the highest diversities of birds in the world, with ~1002 species recorded from within the Kosñipata Valley, of which I was able to sample 245 species (~24% of all species). In the General Introduction (Chapter 1) I gave a broad overview of past research into explaining patterns of diversity and highlighted that although much research has been carried out on the role of abiotic factors in influencing observed patterns, relatively little has been done on the potential importance of biotic interactions. I then introduced parasites as an often-invoked but poorly understood potential driver of observed relationships. I highlighted how despite having strong theoretical support, few empirical studies have directly investigated the role of parasites in limiting species ranges and shaping patterns in diversity. I then provided a brief overview of the ways in which parasites may be important in structuring ecological communities, and highlighted two key areas where

further information was needed. Firstly, understanding the relative importance of hosts ecological, evolutionary, or environmental characteristics toward predicting infections, and embedded within this theme, understanding how parasite distributions are structured in relation to host distributions; and secondly, testing whether parasites may play a role in limiting their hosts' geographic ranges.

6.2 The influence of host and environmental characteristics on disease prevalence

In Chapter 2 I showed that avian malaria infections show non-random variation in prevalence among hosts and used this as the basis to explore the potential importance of host intrinsic and extrinsic characteristics in explaining patterns of infection. I found that contrary to predictions that host ecology or extrinsic environmental conditions are the major drivers of infection prevalence, these factors could only partially explain the patterns of infection. For instance, variation in host ecological traits including foraging strata, diet, and elevation range size were all non-significant predictors of infection prevalence. While I detected significant effects of both body mass and elevation (which is a proxy for temperature and rainfall), these effects were specific to particular parasite genera and thus do not appear to be general predictors of parasite prevalence. This lack of generality in predictors of prevalence may arise because of the widespread variation in the host and vector distribution of each of three parasite genera, which alters the relative importance of predictor variables toward explaining infection. Contrary to the idiosyncratic importance of host ecological and environmental conditions, I found that the most consistent and significant predictor of infection prevalence was host phylogenetic uniqueness: hosts that were more distantly

related to other co-occurring species exhibited reduced infection prevalence. This novel finding formed the basis of the following chapter, which provided an in-depth analysis of the role of host evolutionary relationships in shaping patterns of host parasite infection and parasite sharing.

A number of studies have previously investigated the roles of ecology (Ricklefs et al. 2005, Fecchio et al. 2011), environment (Freed et al. 2005, Hay et al. 2002, Garamszegi 2011), and evolution (Clayton and Moore 1997, Ricklefs 2010b) in explaining parasite infection. However, any one of these studies has typically only investigated a single or small subset of possible explanatory factors. One important implication of my results is that they highlight the importance of simultaneously considering multiple predictors. For instance, while the prevalence of *Leucocytozoon* blood parasites varies with body mass, I found that this effect was superseded by environmental (elevation) and evolutionary (host evolutionary uniqueness) factors. In order to evaluate the importance of host environment, ecology or evolution, future studies should aim to include at least one measure from each of these categories rather than just investigating any single one in isolation. Furthermore, while studies focused on single host-parasite interactions may reveal detailed insights into the mechanisms shaping infection in a particular instance, the generality of any conclusions remain unclear. By analyzing patterns of infection across a large number of hosts that exhibit substantial variation in ecology, environment and evolutionary history, my analysis offers novel insights into the general predictors of prevalence in avian malaria.

6.3 The importance of host evolutionary uniqueness in predicting infection prevalence

In Chapter 3 I explored potential mechanisms through which host evolutionary uniqueness may lead to reduced infection prevalence. In doing so, I investigated if potential correlates of host evolutionary uniqueness such as morphological uniqueness (which I used as a surrogate for numerous aspects of ecology), or overall community diversity, may shape observed results. For instance, community diversity may reduce infection prevalence by a 'dilution effect' (Schmidt and Ostfeld 2001) or alternatively amplify infection if heterospecifics are susceptible to the same diseases, and act as additional reservoirs (Power and Mitchell 2004). I found that neither host morphological characteristics, nor the overall diversity of co-occurring species within a community could explain variation in parasite prevalence among species. Instead, host phylogenetic uniqueness remained the sole best predictor of infection among species, consistent with results from Chapter 2 – but controlling for the aforementioned potential correlates.

I then investigated parasite sharing as a potential mechanism shaping the overriding importance of host evolutionary uniqueness in explaining infection prevalence. I found that as the evolutionary distance between any pair of species increased the probability of sharing parasite lineages declined. This suggests that evolutionarily unique hosts may exhibit lower prevalence because they are less likely to share parasites with other syntopic species. A possible mechanism through which this may occur is if more closely related host species share internal body chemistry characteristics including cellular and molecular features (e.g. cell surface receptors,

enzymes, or MHC genes) that make them more susceptible to infection by the same parasites (Pfennig 2000, Medeiros et al. 2013, Perlman and Jaenike 2003). I also found that increased elevation range overlap between species led to more parasite sharing. These results show that although species occurring in the same geographical areas are more likely to share parasites than those with less or no overlap, even among species with overlapping ranges, the evolutionary distance between species is a major determinant of parasite sharing. The finding that avian malaria lineages are more often shared among closely related bird species is consistent with results from a global analyses of parasite sharing in primates by Davies & Pedersen (2008). However, in contrast to the results reported here, the magnitude of range overlap and parasite sharing were unrelated across primates. In addition to the explanations put forward by Davies and Pederson (2008) about range overlap potentially not corresponding to contact rates, host internal compatibility being more important than interspecific encounters, or microhabitat associations precluding hosts from coming into contact; other possibilities exist. One further explanation might be related to the fact that range overlap in primates is extremely limited and when it does occur, this is most often in species that diverged a long time ago (Pigot and Tobias *in review*). In birds however, there is considerable range overlap, even among much younger species (Pigot and Tobias 2013). I argue therefore that birds offer a much better system for studying the importance of range overlap in determining parasite sharing, as within diverse communities birds of all ages coexist. In primates, only relatively old species tend to co-exist, so any effect of overlap may be lost by that stage.

A major implication of these results is that although environmental conditions are likely to be the most important factor limiting vector distribution and abundance at

broad ecological scales (i.e. hence the importance of host elevation range overlap toward predicting sharing); within areas of suitable vector larval habitats, the most susceptible host species are those that have similar evolutionary histories to other community members. One explanation for the decline in parasite sharing with phylogenetic distance is that closely related species occupy similar ecological niches, linked with microhabitat associations (Schoener 1968, Schoener 1974, Cody 1981) or activity periods (Kronfeld-Schor and Dayan 2003) that may influence parasite transmission. However, I found no relationship between parasite sharing and similarity in host foraging strata, diet, or body mass. This is consistent with the finding that host morphological uniqueness does not predict infection and also supports the results from Chapter 2 indicating a weak effect of host ecology on parasite prevalence. Instead, I contend that closely related species share similar internal body chemistry parameters that parasites are able to exploit. This result is consistent with a study by Longdon et al. (2011) that found that host phylogeny explained most of the observed variation in viral persistence and replication across a range of 51 *Drosophila* host species, and concluded that this was likely to be driven by underlying differences in the cellular environment.

6.4 Infection prevalence and host range position

In Chapter 4 I investigated the prevalence of avian malaria infection in two distinct ways, firstly within hosts' elevation ranges, and secondly, across the parasites' entire elevation range. The first part of this analysis builds upon the previous findings that evolutionarily rare host species have reduced infection prevalence, and tests the

hypothesis that in areas of low abundance, hosts may also have reduced infection due to reduced transmission opportunities.

When all recorded infections were analysed together, I found that parasite prevalence increased toward the centre of host's ranges. Separately investigating each of the parasite genera I found this pattern to be driven by *Plasmodium* infections only, whereas *Haemoproteus* and *Leucocytozoon* genera showed no such pattern. Previous studies have recorded species abundances to be highest at their range centre (Whittaker 1960, Whittaker 1965, Bystrak 1981, Terborgh 1971, Brown 1984, Hengeveld and Haeck 1982) and I provided further empirical evidence to support this. Therefore, for *Plasmodium* lineages these results support conventional wisdom for directly transmitted parasites that infection prevalence should increase with host abundance, due to great transmission opportunities. However, for vector-transmitted parasites, transmission is hypothesised to be driven by frequency dependent disease transmission, which increases with the fraction of the host population that is infectious, but not with overall host density per se. These finding are nevertheless consistent with empirical work across a range of animals and plants that shows that most disease transmission is likely to be intermediate between these two extremes (Hudson et al. 2002).

Results from the second part of this investigation revealed that within their own elevation ranges, parasite lineages showed no significant variation in prevalence. A possible explanation for this is that the broad host and environmental associations of avian malaria parasites facilitate high abundance wherever the parasites come into contact with hosts – regardless of where this occurs within their broader ranges. For

instance, if a parasite is distributed across two hosts that have non-overlapping ranges, then abundance could be high in each of these areas and reduced toward the centre of the parasite's range. Few previous studies on wildlife diseases have investigated infection prevalence within host geographic ranges and few, if any, have examined patterns throughout parasites' geographic ranges. However, a study by Briers (2003) on trematode infection in snails (*Lymnaea stagnalis*) showed that prevalence increased as the species approached one portion of its geographic range limit. Similarly, Antonovics et al. (2003) also demonstrated this for plants, showing that the prevalence of anther-smut disease (caused by *Microbotryum violaceum*) in two host species (*Silene virginica* and *S. carolina*) is higher in populations at the range edge. These results reflect the possible importance of parasites in limiting the geographic ranges of their hosts, which formed the topic of the 5th Chapter.

A possible limitation with the analyses undertaken in this chapter was that hosts were unevenly sampled throughout their elevation ranges due to logistical difficulties in sampling every 50 m elevation segment along the gradient. Therefore, it is possible that reduced infection prevalence of species at their range edge is an artefact of reduced sampling effort in these areas. However, this is unlikely to be the case because there was no overall interaction between sampling elevation and the proportional distance from the edge of a species range (i.e. meaning that captures were not biased to certain locations within hosts ranges).

The major implication of this investigation is that it highlights an important link between abundance and evolutionary uniqueness that is a priority for future research to address. Namely, that both evolutionarily rare and numerically rare host species

within a community have reduced likelihood of infection with local parasites. In this study, my measure of evolutionary uniqueness was not abundance weighted because of difficulties in estimating abundances across the distributions of such a large number of species (245 in total). Nevertheless, my results suggest that abundance may be an important factor correlated with a host's evolutionary distance from other community members. For instance it may be that toward the range edge where conspecific density is lower, hosts are more likely to encounter heterospecifics than they are at their range centre. Under this scenario, abundance and evolutionary uniqueness, measured at an individual level, would be negatively correlated. The opposite pattern may be true if at the range edge the host species encounters close relatives that it cannot otherwise co-exist with due to competitive exclusion, which has previously been implicated as a major factor limiting bird distributions in the Peruvian Andes (Terborgh 1971, Terborgh and Weske 1975). Under this scenario, evolutionary uniqueness and abundance may be positively correlated. My results support the first scenario that species are more likely to be infected when they are surrounded by more members of the same or similar species at their range centre. However, conclusively determining how species abundance and evolutionary uniqueness interact to shape parasite prevalence remains an outstanding question that is a priority for future research.

6.5 Parasites and geographical range limitation

In Chapter 5 I used two separate but complimentary approaches to explore the possibility that parasites may limit their hosts elevation ranges – an often invoked but rarely tested idea in ecology and evolutionary biology. I showed that based on a null

model simulation approach, there was widespread support for parasite limiting certain host species' elevation ranges. Furthermore, in a number of the range limiting lineages, birds were recovered with infections at their range edges coincident with the edge of the parasite.

A caveat of this approach is that results are dependent on the particular assumptions of the null model. Most models of stochastic range placement randomly shuffle observed species distributions within a bounded domain (Colwell et al. 2004). However, if parasites are responsible for constraining range expansion then by using the empirical range size distribution, this may inadvertently smuggle the effects of parasites into the null model expectation. To avoid this I developed a new null model in which the position of species lower and upper range edges (rather than range midpoints) were randomized across the elevation gradients

A potential limitation of the null model I used is that it fails to take into account evolutionary history. In Chapter 2, I showed that more evolutionary unique hosts had a reduced likelihood of infection and in Chapter 3, I provided evidence that this may be explained by a reduction in parasite sharing between distantly related hosts. However, the null model I employed ignored phylogenetic relatedness and thus treated all species as identical. In particular the model assumes that parasites may affect hosts regardless of their phylogenetic similarity. Host pathogen specificity is also important because it links with the first, in that the extent to which more evolutionarily similar parasites are restricted to similar host species remains largely unknown – and therefore was unaccounted for. Previous studies have recorded numerous instances of broad host associations of avian malaria parasites (Bensch et

al. 2009) and my own results show numerous instances of lineages appearing across a broad array of host species (Appendix Table 5.3). Therefore I feel that classifying hosts as limited if they occur in a parasite infection zone regardless of their evolutionary history is justified as a first appraisal of whether there is broad support for a role of parasites in limiting host ranges. An important avenue for future research will be to develop more comprehensive null models that take into account host and parasite evolutionary characteristics in testing a role for parasites in limiting host ranges.

A further factor that I was unable to incorporate into my null model was to take into account the abundance and distribution of vectors, which remain completely unstudied within the Kosñipata Valley. For instance, a finer-scale restriction on my null model simulations would be to shift only the lower elevation range limits of high elevation restricted bird species within a threshold elevation, as if their realised lower distributions are limited by parasites, this is only likely to occur within their natural elevation ranges, which for upland birds will be a high elevation restricted range (i.e. just within the top third of the mountain range, or wherever turnover shows a sharp change). The same process could be applied to low elevation bird species, with a threshold restriction placed on their upper limits, but just simulated within a lower bounded domain rather than across the entire gradient. Similarly, the parasite ranges may also be shuffled within similar limitations, i.e. for *Leucocytozoon* lineages ranges would only be shuffled within cooler high elevation, which largely approximate their vector larval requirements. Furthermore, both host and parasite ranges may both be shuffled according to these protocols.

A limitation with the second part of the study where I recorded birds with ranges ending in parasite zones and with infections recorded within these, was not recording parasitemia levels of all infected birds. However, as a potential justification for this, correlations between parasitemia and fitness impacts remain largely idiosyncratic. For instance, even highly susceptible birds that show high mortality from parasite lineages suffer little to no effect from superinfections (Atkinson et al. 2001). Therefore, under the aforementioned scenario, low parasitemia could indicate that the host has recently acquired a parasite that may cause the hosts death relatively quickly, or alternatively, may reflect a chronic infection that the host is able to cope with.

Broader implications

Taken together these results have important implications for future movement of hosts and parasites due to direct or indirect anthropogenic effects. In the first instance, local introductions of novel parasites to naive communities has already been shown to cause devastating effects, through parasites acting as 'novel weapons' (Callaway and Ridenour 2004, Atkinson et al. 2000). Conversely, host introductions without their parasites may have facilitated the expansion of species ranges through 'enemy release' (Marzal et al. 2011, Colautti et al. 2004, Keane and Crawley 2002). The Intergovernmental Panel on Climate Change (IPCC) has projected that by 2100, relative to 1990, mean global surface will increase between 1.4 and 5.8°C, and under this scenario, global water vapour pressure, evaporation and precipitation are all expected to increase (Houghton et al. 2001). Given the temperature sensitivities of insect vectors and larval requirements for free standing water, this is likely to cause widespread shifts in the temporal and spatial distribution of avian malaria parasites.

Indeed this has already been shown in various places throughout the world (Atkinson and LaPointe 2009, Brooks and Hoberg 2007, Chaves and Koenraadt 2010, Garamszegi 2011, Githeko et al. 2000, Hay et al. 2002, Lindsay and Birley 1996, Loiseau et al. 2013, Patz and Olson 2006, Benning et al. 2002). Moreover, given the often reported broad host associations, and variable pathogenicity across these (Atkinson et al. 1995, Bennett et al. 1993), along with the ability of parasites to jump hosts (Woolhouse et al. 2005) understanding the mechanisms underlying which hosts become infected and how parasites are likely to be structured within host populations remains a priority. As such, determining the extent to which these movements may cause geographical range limits in hosts is crucial for predicting potential impacts and remains a priority. In this study, I have provided a range of results that help understand current patterns in the distribution and diversity of host and their parasites which are the necessary first step toward evaluating how climate change may impact upon natural ecosystems.

Overall I tested the role of avian parasites in shaping patterns in bird diversity. I found support for a role of avian malaria parasites in both enhancing and limiting bird diversity. In order to reconcile these apparently opposing findings I suggest that each of these scenarios play out at different spatial scales. For instance, at broad geographic scales, such as within the Kosñipata Valley in Peru, I found support for a role for parasites in promoting the buildup of diversity locally through a rare species advantage. I found that across the whole elevation gradient, species that are evolutionarily rare relative to co-occurring species are at a comparative advantage in avoiding or resisting avian malaria infections compared to those that were more closely related to other species within their elevation ranges. Under this scenario, an

evolutionarily distant species arriving in local community would be less likely to suffer from local parasites, assisting its chances of establishing in the area. However, at finer scales, such as within an individual species elevation range, I found support for a role of parasites in limiting the expansion of species into otherwise suitable habitats beyond their current geographic limits. This suggests that there is a threshold distance to overcome before a species is no longer limited by parasites at its range edge. However, if a bird is able to disperse far enough then at some point it will be sufficiently different from the local avifauna that its establishment is enhanced through the reduced chances of its suffering infection from local parasites. I suggested that the potential mechanisms facilitating each of these scenarios may operate through either host evolutionary uniqueness corresponding to internal body chemistry, or alternatively through the effects of host abundance, altering the chance of infection.

Conclusions

By studying a diverse assemblage of montane tropical birds and their haemosporidian parasites, I have provided evidence that parasites may both enhance and limit species diversity. These effects play out over fine to broad spatial scales and are not closely linked to ecology, but rather reflect host evolutionary history. Thus parasites provide a mechanism explaining the distribution and diversity of species and help reconcile the inability of ecology or environment to fully explain observed variation in the abundance and distribution of species.

Bibliography

Acharya, K. P., O. R. Vetaas, and H. J. B. Birks. 2011. Orchid species richness along Himalayan elevational gradients. Journal of Biogeography 38:1821-1833.

Adams, J. A. 2009. Species richness: patterns in the diversity of life. Springer, Germany.

Adler, P. B., J. HilleRisLambers, and J. M. Levine. 2007. A niche for neutrality. Ecology Letters 10:95-104.

Adler, P. B., J. HilleRisLambers. 2008. The Influence of Climate and Species Composition on the Population Dynamics of Ten Prairie Forbs. Ecology 89:3049-3060.

Alderman, D., D. Holdich, and I. Reeve. 1990. Signal crayfish as vectors in crayfish plague in Britain. Aquaculture 86:3-6.

Anderson, R. M., R. M. May. 1982. Coevolution of Hosts and Parasites. Parasitology 85:411-426.

Anderson, R. M., R. M. May. 1978. Regulation and Stability of Host-Parasite Population Interactions: Regulatory Processes. Journal of Animal Ecology 47:219-247.

Andrewartha, H. G., I. C. Birch. 1954. The Distribution and Abundance of Animals. University of Chicago Press, Chicago.

Antonovics, J., Y. Iwasa, and M. P. Hassell. 1995. A generalized model of parasitoid, venereal, and vector-based transmission processes. The American Naturalist 145:661-675.

Antonovics, J., M. E. Hood, P. H. Thrall, J. Y. Abrams, and G. M. Duthie. 2003. Herbarium studies on the distribution of anther-smut fungus (*Microbotryum*

violaceum) and Silene species (Caryophyllaceae) in the eastern United States. American Journal of Botany 90:1522-1531.

Arneberg, P., A. Skorping, B. Grenfell, and A. F. Read. 1998. Host densities as determinants of abundance in parasite communities. Proceedings of the Royal Society of London. Series B: Biological Sciences 265:1283-1289.

Asghar, M., D. Hasselquist, and S. Bensch. 2011. Are chronic avian haemosporidian infections costly in wild birds? Journal of Avian Biology 42:530-537.

Atkinson, C. T., R. J. Dusek, K. L. Woods, and W. M. Iko. 2000. Pathogenicity of avian malaria in experimentally-infected Hawaii Amakihi. Journal of wildlife diseases 36:197-204.

Atkinson, C., K. Woods, R. Dusek, L. Sileo, and W. Iko. 1995. Wildlife disease and conservation in Hawaii: pathogenicity of avian malaria (*Plasmodium relictum*) in experimentally infected Iiwi (*Vestiaria coccinea*). Parasitology 111:S59—S69.

Atkinson, C. T., D. A. LaPointe. 2009. Introduced Avian Diseases, Climate Change, and the Future of Hawaiian Honeycreepers. Journal of Avian Medicine and Surgery 23:53-63.

Atkinson, C. T., C. van Ripper III. 1991. Pathogenicity and epizootiology of avian haematozoa: *Plasmodium, Leucocytozoon,* and *Haemoproteus*. *In* Loye, J. E. and M. Zuk, editors. Bird-parasite interactions: ecology, evolution and behaviour, Oxford University Press, Oxford.

Atkinson, C. T., R. J. Dusek, and J. K. Lease. 2001. Serological responses and immunity to superinfection with avian malaria in experimentally-infected Hawaii Amakihi. Journal of Wildlife Diseases 37:20-27.

Bachman, S., W. J. Baker, N. Brummitt, J. Dransfield, and J. Moat. 2004. Elevational gradients, area and tropical island diversity: an example from the palms of New Guinea. Ecography 27:299-310.

Barry, R. G. 2008. Mountain Weather and Climate. Cambridge University Press, Cambridge.

Bateman, B. L., S. E. Abell-Davis, and C. N. Johnson. 2012. Climate-driven variation in food availability between the core and range edge of the endangered northern bettong (*Bettongia tropica*). Australian Journal of Zoology 59:177-185.

Belmaker, J., C. H. Sekercioglu, and W. Jetz. 2012. Global patterns of specialization and coexistence in bird assemblages. Journal of Biogeography 39:193-203.

Bennett, G., M. Peirce, and R. Ashford. 1993. Avian haematozoa: mortality and pathogenicity. Journal of Natural History 27:993-1001.

Benning, T. L., D. LaPointe, C. T. Atkinson, and P. M. Vitousek. 2002. Interactions of climate change with biological invasions and land use in the Hawaiian Islands: Modeling the fate of endemic birds using a geographic information system. Proceedings of the National Academy of Sciences of the United States of America 99:14246-14249.

Bensch, S., J. Waldenström, N. Jonzén, H. Westerdahl, B. Hansson, D. Sejberg, and D. Hasselquist. 2007. Temporal dynamics and diversity of avian malaria parasites in a single host species. Journal of Animal Ecology 76:112-122.

Bensch, S., O. Hellgren, and J. Perez-Tris. 2009. MalAvi: a public database of malaria parasites and related haemosporidians in avian hosts based on mitochondrial cytochrome b lineages. Molecular Ecology Resources 9:1353-1358.

Bensch, S., J. Perez-Tris, J. Waldenstrom, and O. Hellgren. 2004. Linkage between nuclear and mitochondrial DNA sequences in avian malaria parasites: Multiple cases of cryptic speciation? Evolution 58:1617-1621.

Bensch, S., M. Stjernman, D. Hasselquist, O. Ostman, B. Hansson, H. Westerdahl, and R. T. Pinheiro. 2000. Host specificity in avian blood parasites: a study of *Plasmodium* and *Haemoproteus* mitochondrial DNA amplified from birds. Proceedings of the Royal Society of London. Series B: Biological Sciences 267:1583-1589.

Bertheau, C., E. G. Brockerhoff, G. Roux-Morabito, F. Lieutier, and H. Jactel. 2010. Novel insect-tree associations resulting from accidental and intentional biological 'invasions': a meta-analysis of effects on insect fitness. Ecology Letters 13:506-515.

Bhattarai, K. R., O. R. Vetaas, and J. A. Grytnes. 2004. Fern species richness along a central Himalayan elevational gradient, Nepal. Journal of Biogeography 31:389-400.

Blomberg, S. P., T. Garland, and A. R. Ives. 2003. Testing for phylogenetic signal in comparative data: behavioral traits are more labile. Evolution 57:717-745.

Bonneaud, C., J. Pérez-Tris, P. Federici, O. Chastel, and G. Sorci. 2006. Major histocompatibility alleles associated with local resistance to malaria in a passerine. Evolution 60:383-389.

Bonsall, M., M. Hassell. 1997. Apparent competition structures ecological assemblages. Nature 388:371-373.

Bozinovic, F., P. Calosi, and J. I. Spicer. 2011. Physiological correlates of geographic range in animals. Annual Review of Ecology, Evolution, and Systematics 42:155-179.

Braga, E. M., P. Silveira, N. O. Belo, and G. Valkiunas. 2011. Recent advances in the study of avian malaria: an overview with an emphasis on the distribution of Plasmodium spp in Brazil. Memorias Instituto Oswaldo Cruz 106(Suppl.I):3-11.

Brehm, G., R. K. Colwell, and J. Kluge. 2007. The role of environment and mid-domain effect on moth species richness along a tropical elevational gradient. Global Ecology and Biogeography 16:205-219.

Briers, R. A. 2003. Range limits and parasite prevalence in a freshwater snail. Proceedings of the Royal Society of London. Series B: Biological Sciences 270:S178-S180.

Brooks, D. R., E. P. Hoberg. 2007. How will global climate change affect parasite-host assemblages? Trends in Parasitology 23:571-574.

Brown, J. H. 1984. On the relationship between abundance and distribution of species. The American Naturalist 124:255-279.

Brown, J. H., M. V. Lomolino. 1998. Biogeography. Sinauer Associates, Sunderland.

Brown, J. H., G. C. Stevens, and D. M. Kaufman. 1996. The geographic range: size, shape, boundaries, and internal structure. Annual Review of Ecology and Systematics 27:597-623.

Brown, V. K., T. R. E. Southwood. 1983. Trophic Diversity, Niche Breadth and Generation Times of Exopterygote Insects in a Secondary Succession. Oecologia 56:220-225.

Bruford, M. W., O. Hanotte, J. F. Y. Brookfield, and T. Burke. 1998. Multilocus and single-locus DNA fingerprinting. Pages 287-336 *In* Hoelzel, A. R., editor. Molecular Genetic Analysis of Populations: A Practical Approach, IRL Press, Oxford.

Bystrak, D. 1981. The North American Breeding Bird Survey. Studies in Avian Biology 6:34-41.

Callaway, R. M., W. M. Ridenour. 2004. Novel weapons: invasive success and the evolution of increased competitive ability. Frontiers in Ecology and the Environment 2:436-443.

Chaves, L. F., C. J. M. Koenraadt. 2010. Climate Change and Highland Malaria: Fresh Air for a Hot Debate. Quarterly Review of Biology 85:27-55.

Chesson, P. 2000. Mechanisms of maintenance of species diversity. Annual Review of Ecology and Systematics 31:343-366.

Chown, S. L., K. J. Gaston. 1999. Exploring links between physiology and ecology at macro-scales: the role of respiratory metabolism in insects. Biological reviews of the Cambridge Philosophical Society 74:87-120.

Clayton, D. H., J. Moore. 1997. Host-parasite evolution: general principles and avian models. Oxford University Press, Oxford.

Cody, M. L. 1981. Habitat selection in birds: the roles of vegetation structure, competitors, and productivity. Bioscience 31:107-113.

Colautti, R. I., A. Ricciardi, I. A. Grigorovich, and H. J. MacIsaac. 2004. Is invasion success explained by the enemy release hypothesis? Ecology Letters 7:721-733.

Colwell, R. K. 1999. RangeModel: a Monte Carlo simulation tool for assessing geometric constraints on species richness, Ver. 2. User's guide and application published at http:/www.viceroy.eeb.uconn.edu/RangeModel.

Colwell, R. K., G. C. Hurtt. 1994. Nonbiological gradients in species richness and a spurious Rapoport effect. The American Naturalist 144:570-595.

Colwell, R. K., D. C. Lees. 2000. The mid-domain effect: geometric constraints on the geography of species richness. Trends in Ecology & Evolution 15:70-76.

Colwell, R. K., C. Rahbek, and N. J. Gotelli. 2004. The mid-domain effect and species richness patterns: What have we learned so far? The American Naturalist 163:E1-E23.

Combes, C. 2004. Parasitism: the ecology and evolution of intimate interactions. University of Chicago Press, Chicago.

Connor, E. F., E. D. McCoy. 1979. Statistics and Biology of the Species-Area Relationship. The American Naturalist 113:791-833.

Cooper, N., R. Griffin, M. Franz, M. Omotayo, and C. L. Nunn. 2012. Phylogenetic host specificity and understanding parasite sharing in primates. Ecology Letters 15:1370-1377.

Cornell, H. V., B. A. Hawkins, and M. E. Hochberg. 1998. Towards an empirically-based theory of herbivore demography. Ecological Entomology 23:340-349.

Cowman, A. F., B. S. Crabb. 2006. Invasion of red blood cells by malaria parasites. Cell 124:755-766.

Crozier, L. G. 2004. Field transplants reveal summer constraints on a butterfly range expansion. Oecologia 141:148-157.

Currie, D. J. 1991. Energy and Large-Scale Patterns of Animal-Species and Plant-Species Richness. The American Naturalist 137:27-49.

Currie, D. J., G. G. Mittelbach, H. V. Cornell, R. Field, J. F. Guegan, B. A. Hawkins, D. M. Kaufman, J. T. Kerr, T. Oberdorff, E. O'Brien, and J. R. G. Turner. 2004. Predictions and tests of climate-based hypotheses of broad-scale variation in taxonomic richness. Ecology Letters 7:1121-1134.

Dalmat, H. T. 1955. The Black Flies (Diptera, Simuliidae) of Guatemala and their Role as Vectors of Onchocerciasis. Smithsonian Institution, Washington.

Darwin, C. 1859. The origin of species by means of natural selection, or, The preservation of favoured races in the struggle for life. John Murray, London.

Daszak, P., A. A. Cunningham, and A. D. Hyatt. 2000. Wildlife ecology - Emerging infectious diseases of wildlife - Threats to biodiversity and human health. Science 287:443-449.

Davies, T. J., A. B. Pedersen. 2008. Phylogeny and geography predict pathogen community similarity in wild primates and humans. Proceedings of the Royal Society of London. Series B: Biological Sciences 275:1695-1701.

De Meeûs, T., F. Renaud. 2002. Parasites within the new phylogeny of eukaryotes. Trends in parasitology 18:247-251.

Diamond, J. M. 1978. Niche Shifts and the Rediscovery of Interspecific Competition: Why did field biologists so long overlook the widespread evidence for interspecific competition that had already impressed Darwin? American Scientist 66:322-331.

Diamond, J. M. 1975. Assembly of species communities. Pages 342-444 *In* Cody, M. L. and Diamond, J. M, editors. Ecology and evolution of communities, Harvard University Press, Cambridge Massachusetts.

Dobson, A. P. 1990. Models for multi-species parasite-host communities. Pages 261-288 *In* Esch, G., A. O. Bush, and J. M. Aho, editors. Parasite communities: patterns and processes, Chapman & Hall, London.

Dobson, A. 2004. Population dynamics of pathogens with multiple host species. The American Naturalist 164:S64-S78.

Dobson, A., K. D. Lafferty, A. M. Kuris, R. F. Hechinger, and W. Jetz. 2008. Homage to Linnaeus: How many parasites? How many hosts? Proceedings of the

National Academy of Sciences of the United States of America 105:11482-11489.

Drummond, A. J., A. Rambaut. 2007. BEAST: Bayesian evolutionary analysis by sampling trees. BMC evolutionary biology 7:214.

Dunn, R. R., T. J. Davies, N. C. Harris, and M. C. Gavin. 2010. Global drivers of human pathogen richness and prevalence. Proceedings of the Royal Society of London. Series B: Biological Sciences 277:2587-2595.

Dunn, R. R., C. M. McCain, and N. J. Sanders. 2007. When does diversity fit null model predictions? Scale and range size mediate the mid-domain effect. Global Ecology and Biogeography 16:305-312.

Dunning, J. 2008. CRC handbook of avian body masses. FL: CRC :.

Evans, K. L., P. H. Warren, and K. J. Gaston. 2005. Species-energy relationships at the macroecological scale: a review of the mechanisms. Biological Reviews 80:1-25.

Fallon, S. M., E. Bermingham, and R. E. Ricklefs. 2005. Host specialization and geographic localization of avian malaria parasites: a regional analysis in the Lesser Antilles. The American Naturalist 165:466-480.

Fallon, S. M., R. E. Ricklefs, B. L. Swanson, and E. Bermingham. 2003. Detecting avian malaria: An improved polymerase chain reaction diagnostic. Journal of Parasitology 89:1044-1047.

Fecchio, A., M. R. Lima, P. Silveira, É M. Braga, and M. Â Marini. 2011. High prevalence of blood parasites in social birds from a neotropical savanna in Brazil. Emu 111:132-138.

Fortin, A., M. Stevenson, and P. Gros. 2002. Susceptibility to malaria as a complex trait: big pressure from a tiny creature. Human molecular genetics 11:2469-2478.

Fortin, M. J., T. H. Keitt, B. A. Maurer, M. L. Taper, D. M. Kaufman, and T. M. Blackburn. 2005. Species' geographic ranges and distributional limits: pattern analysis and statistical issues. Oikos 108:7-17.

Francis, A. P., D. J. Currie. 2003. A globally consistent richness-climate relationship for angiosperms. The American Naturalist 161:523-536.

Freckleton, R. P., O. T. Lewis. 2006. Pathogens, density dependence and the coexistence of tropical trees. Proceedings of the Royal Society of London. Series B: Biological Sciences 273:2909-2916.

Freed, L. A., R. L. Cann, M. L. Goff, W. A. Kuntz, and G. R. Bodner. 2005. Increase in avian malaria at upper elevation in Hawaii. Condor 107:753-764.

Friend, M., J. C. Franson. 1999. Field Manual of Wildlife Diseases. General Field Procedures and Diseases of Birds. United States Department of The Interior, Maryland.

Fritz, S. A., A. Purvis. 2010. Selectivity in mammalian extinction risk and threat types: a new measure of phylogenetic signal strength in binary traits. Conservation Biology 24:1042-1051.

Garamszegi, L. Z. 2011. Climate change increases the risk of malaria in birds. Global Change Biology 17:1751-1759.

Garvin, M. C., J. V. Remsen. 1997. An alternative hypothesis for heavier parasite loads of brightly colored birds: exposure at the nest. The Auk 114:179-191.

Gaston, K. J., T. M. Blackburn. 2000. Pattern and process in macroecology. Blackwell Scientific, Oxford.

Gaston, K. J. 2003. The Structure and Dynamics of Geographic Ranges. Oxford University Press, Oxford.

Gaston, K. J. 2000. Global patterns in biodiversity. Nature 405:220-227.

Gaston, K. J., T. M. Blackburn, and J. I. Spicer. 1998. Rapoport's rule: time for an epitaph? Trends in Ecology & Evolution 13:70-74.

Gause, G. F. 1934. The struggle for existence. Hafner Publishing Company, New York.

Gilbert, G. S., C. O. Webb. 2007. Phylogenetic signal in plant pathogen–host range. Proceedings of the National Academy of Sciences 104:4979-4983.

Gillies, M. 1980. The role of carbon dioxide in host-finding by mosquitoes (Diptera: Culicidae): a review. Bulletin of Entomological Research 70:525-532.

Girardin, C. A. J., Y. Malhi, L. E. O. C. Aragao, M. Mamani, W. H. Huasco, L. Durand, K. J. Feeley, J. Rapp, J. E. Silva-Espejo, M. Silman, N. Salinas, and R. J. Whittaker. 2010. Net primary productivity allocation and cycling of carbon along a tropical forest elevational transect in the Peruvian Andes. Global Change Biology 16:3176-3192.

Githeko, A. K., S. W. Lindsay, U. E. Confalonieri, and J. A. Patz. 2000. Climate change and vector-borne diseases: a regional analysis. Bulletin of the World Health Organization 78:1136-1147.

Gregory, R. 1990. Parasites and host geographic range as illustrated by waterfowl. Functional Ecology 4:645-654.

Greiner, E. C., G. F. Bennett, E. M. White, and R. F. Coombs. 1975. Distribution of the avian hematozoa of North America. Canadian Journal of Zoology 53:1762-1787.

Grenfell, B. T., A. P. Dobson. 1995. Ecology of infectious diseases in natural populations. Cambridge University Press, Cambridge.

Grueber, C., S. Nakagawa, R. Laws, and I. Jamieson. 2011. Multimodel inference in ecology and evolution: challenges and solutions. Journal of Evolutionary Biology 24:699-711.

Grytnes, J. A., J. H. Beaman. 2006. Elevational species richness patterns for vascular plants on Mount Kinabalu, Borneo. Journal of Biogeography 33:1838-1849.

Hadfield, J. D. 2010. MCMC methods for multi-response generalized linear mixed models: the MCMCglmm R package. Journal of Statistical Software 33:1-22.

Hafner, M. S., S. A. Nadler. 1988. Phylogenetic Trees Support the Coevolution of Parasites and their Hosts. Nature 332:258-259.

Hall, C. A., J. A. Stanford, and F. R. Hauer. 1992. The distribution and abundance of organisms as a consequence of energy balances along multiple environmental gradients. Oikos 65:377-390.

Hall, T. A. 1999. BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. Nucleic Acids Symposium Series 41:95-98.

Hamilton, W. D., M. Zuk. 1982. Heritable true fitness and bright birds - a role for parasites. Science 218:384-387.

Hawkins, B. A. 2005. Pattern and process in host-parasitoid interactions. Cambridge University Press, Cambridge.

Hawkins, B. A., J. A. F. Diniz-Filho, and A. E. Weis. 2005. The Mid-Domain Effect and Diversity Gradients: Is There Anything to Learn? The American Naturalist 166:E140-E143.

Hawkins, B. A., J. A. F. Diniz-Filho, C. A. Jaramillo, and S. A. Soeller. 2007. Climate, niche conservatism, and the global bird diversity gradient. The American Naturalist 170:S16-S27.

Hawkins, B. A., R. Field, H. V. Cornell, D. J. Currie, J. F. Guegan, D. M. Kaufman, J. T. Kerr, G. G. Mittelbach, T. Oberdorff, E. M. O'Brien, E. E. Porter, and J. R. G. Turner. 2003. Energy, water, and broad-scale geographic patterns of species richness. Ecology 84:3105-3117.

Hay, S. I., J. Cox, D. J. Rogers, S. E. Randolph, D. I. Stern, G. D. Shanks, M. F. Myers, and R. W. Snow. 2002. Climate change and the resurgence of malaria in the East African highlands. Nature 415:905-909.

Hellgren, O., J. Perez-Tris, and S. Bensch. 2009. A jack-of-all-trades and still a master of some: prevalence and host range in avian malaria and related blood parasites. Ecology 90:2840-2849.

Hellgren, O., S. Bensch, and B. Malmqvist. 2008. Bird hosts, blood parasites and their vectors—associations uncovered by molecular analyses of blackfly blood meals. Molecular Ecology 17:1605-1613.

Hellgren, O., A. Križanauskienė, D. Hasselquist, and S. Bensch. 2011. Low haemosporidian diversity and one key-host species in a bird malaria community on a mid-Atlantic island (São Miguel, Azores). Journal of wildlife diseases 47:849-859.

Hellgren, O., J. Waldenstrom, and S. Bensch. 2004. A new PCR assay for simultaneous studies of Leucocytozoon, Plasmodium, and Haemoproteus from avian blood. Journal of Parasitology 90:797-802.

Hengeveld, R., J. Haeck. 1982. The Distribution of Abundance .1. Measurements. Journal of Biogeography 9:303-316.

Hernández, L. M., F. Montes de Oca, M. Penn, J. Massaferro, A. Garré, and S. J. Brooks. 2009. " Jejenes"(Diptera: Simuliidae) del Parque Nacional Nahuel

Huapi, Patagonia, Argentina: Resultados preliminares. Revista de la Sociedad Entomológica Argentina 68:193-200.

Hochberg, M. E., A. R. Ives. 1999. Can natural enemies enforce geographical range Limits? Ecography 22:268-276.

Hoffmann, A. A., M. W. Blows. 1994. Species Borders - Ecological and Evolutionary Perspectives. Trends in Ecology & Evolution 9:223-227.

Holt, R., J. Lawton. 1994. The ecological consequences of shared natural enemies. Annual Review of Ecology and Systematics 25:495-520.

Holt, R. D. 2003. On the evolutionary ecology of species' ranges. Evolutionary Ecology Research 5:159-178.

Holt, R. D. 1977. Predation, apparent competition, and the structure of prey communities. Theoretical Population Biology 12:197-229.

Holt, R. D., M. Barfield, I. Filin, and S. Forde. 2011. Predation and the evolutionary dynamics of species ranges. The American Naturalist 178:488-500.

Holt, R. D., J. H. Lawton. 1993. Apparent Competition and Enemy-Free Space in Insect Host-Parasitoid Communities. The American Naturalist 142:623-645.

Houghton, J. T., Y. Ding, D. J. Griggs, M. Noguer, van der Linden, Paul J, X. Dai, K. Maskell, and C. Johnson. 2001. Climate change 2001: the scientific basis. Cambridge University Press, Cambridge.

Hudson, P. J., A. R. Rizzoli, B. T. Grenfell, H. Heesterbeek, and A. P. Dobson. 2002. The Ecology of Wildlife Diseases. Oxford University Press, Oxford.

Hutchinson, G. E. 1961. The paradox of the plankton. The American Naturalist 95:137-145.

Jablonski, D., K. Roy, and J. W. Valentine. 2006. Out of the tropics: Evolutionary dynamics of the latitudinal diversity gradient. Science 314:102-106.

Janzen, D. H. 1970. Herbivores and number of tree species in tropical forests. The American Naturalist 104:501-528.

Jenkins, T., G. H. Thomas, O. Hellgren, and I. P. F. Owens. 2012. Migratory behavior of birds affects their coevolutionary relationship with blood parasites. Evolution 66:740-751.

Jetz, W., G. Thomas, J. Joy, K. Hartmann, and A. Mooers. 2012. The global diversity of birds in space and time. Nature 491:444-448.

Karger, D. N., J. Kluge, T. Kroemer, A. Hemp, M. Lehnert, and M. Kessler. 2011. The effect of area on local and regional elevational patterns of species richness. Journal of Biogeography 38:1177-1185.

Kattan, G. H., P. Franco. 2004. Bird diversity along elevational gradients in the Andes of Colombia: area and mass effects. Global Ecology and Biogeography 13:451-458.

Keane, R. M., M. J. Crawley. 2002. Exotic plant invasions and the enemy release hypothesis. Trends in Ecology & Evolution 17:164-170.

Kearney, M., W. Porter. 2009. Mechanistic niche modelling: combining physiological and spatial data to predict species' ranges. Ecology Letters 12:334-350.

Keesing, F., R. D. Holt, and R. S. Ostfeld. 2006. Effects of species diversity on disease risk. Ecology Letters 9:485-498.

Kessler, M. 2000. Upslope-directed mass effect in palms along an Andean elevational gradient: A cause for high diversity at mid-elevations? Biotropica 32:756-759.

Keyghobadi, N., D. LaPointe, R. C. Fleischer, and D. M. Fonseca. 2006. Fine-scale population genetic structure of a wildlife disease vector: the southern house mosquito on the island of Hawaii. Molecular Ecology 15:3919-3930.

Kilpatrick, A. M., C. J. Briggs, and P. Daszak. 2010. The ecology and impact of chytridiomycosis: an emerging disease of amphibians. Trends in Ecology & Evolution 25:109-118.

Klopfer, P. H., R. H. MacArthur. 1961. On the causes of tropical species diversity: niche overlap. The American Naturalist 95:223-226.

Klopfer, P. H., R. H. MacArthur. 1960. Niche size and faunal diversity. The American Naturalist 95:293-300.

Kluge, J., M. Kessler, and R. R. Dunn. 2006. What drives elevational patterns of diversity? A test of geometric constraints, climate and species pool effects for pteridophytes on an elevational gradient in Costa Rica. Global Ecology and Biogeography 15:358-371.

Knowles, S., V. Palinauskas, and B. Sheldon. 2010a. Chronic malaria infections increase family inequalities and reduce parental fitness: experimental evidence from a wild bird population. Journal of Evolutionary Biology 23:557-569.

Knowles, S. C. L., M. J. Wood, and B. C. Sheldon. 2010b. Context-dependent effects of parental effort on malaria infection in a wild bird population, and their role in reproductive trade-offs. Oecologia 164:87-97.

Krasnov, B. R., D. Mouillot, G. I. Shenbrot, I. S. Khokhlova, M. V. Vinarski, N. P. Korallo-Vinarskaya, and R. Poulin. 2010. Similarity in ectoparasite faunas of Palaearctic rodents as a function of host phylogenetic, geographic or environmental distances: Which matters the most? International journal for parasitology 40:807-817.

Krebs, C. J. 1994. Ecology: the experimental analysis of distribution and abundance. Harper Collins College, New York.

Krizanauskiene, A., T. A. Iezhova, R. Sehgal, J. S. Carlson, V. Palinauskas, S. Bensch, and G. Valkiunas. 2013. Molecular characterization of Haemoproteus sacharovi (Haemosporida, Haemoproteidae), a common parasite of columbiform birds, with remarks on classification of haemoproteids of doves and pigeons. Zootaxa 3613:85-94.

Kronfeld-Schor, N., T. Dayan. 2003. Partitioning of time as an ecological resource. Annual Review of Ecology, Evolution, and Systematics 34:153-181.

Kuris, A. M., A. R. Blaustein, and J. J. Alio. 1980. Hosts as islands. The American Naturalist 116:570-586.

Lafferty, K. D., R. D. Holt. 2003. How should environmental stress affect the population dynamics of disease? Ecology Letters 6:654-664.

Lafferty, K. D., A. P. Dobson, and A. M. Kuris. 2006. Parasites dominate food web links. Proceedings of the National Academy of Sciences 103:11211-11216.

LaPointe, D. A., M. L. Goff, and C. T. Atkinson. 2010. Thermal Constraints to the Sporogonic Development and Altitudinal Distribution of Avian Malaria Plasmodium Relictum in Hawaii. Journal of Parasitology 96:318-324.

Lawton, J. H. 1996. Population abundances, geographic ranges and conservation: 1994 Witherby Lecture. Bird Study 43:3-19.

Letunic, I., P. Bork. 2011. Interactive Tree Of Life v2: online annotation and display of phylogenetic trees made easy. Nucleic acids research 39:W475-W478.

Lindsay, S., M. Birley. 1996. Climate change and malaria transmission. Annals of Tropical Medicine and Parasitology 90:573-588.

Little, R., R. Earlé. 1995. Sandgrouse (Pterocleidae) and sociable weavers *Philetarius socius* lack avian haematozoa in semi-arid regions of South Africa. Journal of Arid Environments 30:367-370.

Loiseau, C., R. J. Harrigan, C. Bichet, R. Julliard, S. Garnier, Á Z. Lendvai, O. Chastel, and G. Sorci. 2013. Predictions of avian Plasmodium expansion under climate change. Scientific Reports 3:1126.

Loiseau, C., R. Zoorob, A. Robert, O. Chastel, R. Julliard, and G. Sorci. 2011. *Plasmodium relictum* infection and MHC diversity in the house sparrow (*Passer domesticus*). Proceedings of the Royal Society of London. Series B: Biological Sciences 278:1264-1272.

Loiseau, C., R. J. Harrigan, A. Robert, R. C. Bowie, H. A. Thomassen, T. B. Smith, and R. N. Sehgal. 2012. Host and habitat specialization of avian malaria in Africa. Molecular Ecology 21:431-441.

Lomolino, M. V. 2001. Elevation gradients of species-density: historical and prospective views. Global Ecology and Biogeography 10:3-13.

Longdon, B., J. D. Hadfield, C. L. Webster, D. J. Obbard, and F. M. Jiggins. 2011. Host phylogeny determines viral persistence and replication in novel hosts. PLoS pathogens 7:e1002260.

MacArthur, R. H. 1969. Patterns of communities in the tropics. Biological Journal of the Linnean Society 1:19-30.

MacArthur, R. H. 1972. Geographical ecology: patterns in the distribution of species. Harper & Row, New York.

MacArthur, R. H., J. W. MacArthur, and J. Preer. 1962. On bird species diversity II. Prediction of bird census from habitat measurements. The American Naturalist 98:167-174.

Margolis, L., G. Esch, J. Holmes, A. Kuris, and G. A. Schad. 1982. The use of ecological terms in parasitology (report of an ad hoc committee of the American Society of Parasitologists). The Journal of Parasitology 68:131-133.

Martinez-de la Puente, J., S. Merino, G. Tomas, J. Moreno, J. Morales, E. Lobato, S. Garcia-Fraile, and E. J. Belda. 2010. The blood parasite Haemoproteus reduces survival in a wild bird: a medication experiment. Biology Letters 6:663-665.

Marzal, A. 2012. Recent Advances in Studies on Avian Malaria Parasites. Pages 135-159 *In* Okwa, O. O., editor. Malaria parasites, InTech, www.intechopen.com.

Marzal, A., F. De Lope, C. Navarro, and A. P. Møller. 2005. Malarial parasites decrease reproductive success: an experimental study in a passerine bird. Oecologia 142:541-545.

Marzal, A., R. E. Ricklefs, G. Valkiūnas, T. Albayrak, E. Arriero, C. Bonneaud, G. A. Czirják, J. Ewen, O. Hellgren, and D. Hořáková. 2011. Diversity, loss, and gain of malaria parasites in a globally invasive bird. PLoS one 6:e21905.

May, R. M., R. M. Anderson. 1978. Regulation and stability of host-parasite population interactions: II. Destabilizing processes. The Journal of Animal Ecology 47:249-267.

McCain, C. M., J. A. Grytnes. 2010. Elevational Gradients in Species Richness. Encyclopedia of Life Sciences (ELS). John Wiley & Sons, Ltd, Chichester :.

McCain, C. M. 2009. Global analysis of bird elevational diversity. Global Ecology and Biogeography 18:346-360.

McCain, C. M. 2007a. Could temperature and water availability drive elevational species richness patterns? A global case study for bats. Global Ecology and Biogeography 16:1-13.

McCain, C. M. 2007b. Area and mammalian elevational diversity. Ecology 88:76-86.

McCain, C. M. 2005. Elevational gradients in diversity of small mammals. Ecology 86:366-372.

McCain, C. M. 2004. The mid-domain effect applied to elevational gradients: species richness of small mammals in Costa Rica. Journal of Biogeography 31:19-31.

McCallum, H., A. Dobson. 1995. Detecting disease and parasite threats to endangered species and ecosystems. Trends in Ecology & Evolution 10:190-194.

McCallum, H., N. Barlow, and J. Hone. 2001. How should pathogen transmission be modelled? Trends in Ecology & Evolution 16:295-300.

McCoy, E. D. 1990. The Distribution of Insects Along Elevational Gradients. Oikos 58:313-322.

Medeiros, M. C., G. L. Hamer, and R. E. Ricklefs. 2013. Host compatibility rather than vector–host-encounter rate determines the host range of avian Plasmodium parasites. Proceedings of the Royal Society of London. Series B: Biological Sciences 280:20122947.

Merino, S., J. Moreno, J. J. Sanz, and E. Arriero. 2000. Are avian blood parasites pathogenic in the wild? A medication experiment in blue tits (*Parus caeruleus*). Proceedings of the Royal Society of London. Series B: Biological Sciences 267:2507-2510.

Merino, S., J. Moreno, R. A. Vasquez, J. Martinez, I. Sanchez-Monsalvez, C. F. Estades, S. Ippi, P. Sabat, R. Rozzi, and S. Mcgehee. 2008. Haematozoa in forest birds from southern Chile: Latitudinal gradients in prevalence and parasite lineage richness. Austral Ecology 33:329-340.

Mi, X., N. G. Swenson, R. Valencia, W. J. Kress, D. L. Erickson, A. J. Pérez, H. Ren, S. Su, N. Gunatilleke, and S. Gunatilleke. 2012. The contribution of rare species to community phylogenetic diversity across a global network of forest plots. The American Naturalist 180:E17-E30.

Mittelbach, G. G., D. W. Schemske, H. V. Cornell, A. P. Allen, J. M. Brown, M. B. Bush, S. P. Harrison, A. H. Hurlbert, N. Knowlton, H. A. Lessios, C. M. McCain, A. R. McCune, L. A. McDade, M. A. McPeek, T. J. Near, T. D. Price, R. E. Ricklefs, K. Roy, D. F. Sax, D. Schluter, J. M. Sobel, and M. Turelli. 2007. Evolution and the latitudinal diversity gradient: speciation, extinction and biogeography. Ecology Letters 10:315-331.

Mott, C. 2010. Environmental Constraints to the Geographic Expansion of Plant and Animal Species. Nature Education Knowledge 3:72.

Nunn, C. L., S. Altizer. 2006. Infectious diseases in primates: Behavior, ecology and evolution. Oxford University Press, Oxford.

Orme, C. D. L., R. G. Davies, M. Burgess, F. Eigenbrod, N. Pickup, V. A. Olson, A. J. Webster, T. Ding, P. C. Rasmussen, and R. S. Ridgely. 2005. Global hotspots of species richness are not congruent with endemism or threat. Nature 436:1016-1019.

Palin, O. F., P. Eggleton, Y. Malhi, C. A. J. Girardin, A. Rozas-Davila, and C. L. Parr. 2011. Termite Diversity along an Amazon-Andes Elevation Gradient, Peru. Biotropica 43:100-107.

Parmesan, C. 2006. Ecological and evolutionary responses to recent climate change. Annual Review of Ecology Evolution and Systematics 37:637-669.

Parmesan, C., G. Yohe. 2003. A globally coherent fingerprint of climate change impacts across natural systems. Nature 421:37-42.

Patterson, B. D., V. Pacheco, and S. Solari. 1996. Distributions of bats along an elevational gradient in the Andes of south-eastern Peru. Journal of Zoology 240:637-658.

Patz, J. A., S. H. Olson. 2006. Malaria risk and temperature: Influences from global climate change and local land use practices. Proceedings of the National Academy of Sciences of the United States of America 103:5635-5636.

Pérez-Tris, J., D. Hasselquist, O. Hellgren, A. Krizanauskiene, J. Waldenström, and S. Bensch. 2005. What are malaria parasites? Trends in parasitology 21:209-211.

Pérez-Tris, J., O. Hellgren, A. Križanauskienė, J. Waldenström, J. Secondi, C. Bonneaud, J. Fjeldså, D. Hasselquist, and S. Bensch. 2007. Within-host speciation of malaria parasites. PLoS one 2:e235.

Perkins, S. L., J. J. Schall. 2002. A molecular phylogeny of malarial parasites recovered from cytochrome b gene sequences. Journal of Parasitology 88:972-978.

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

Pfennig, D. W. 2000. Effect of Predator-Prey Phylogenetic Similarity on the Fitness Consequences of Predation: A Trade-off between Nutrition and Disease? The American Naturalist 155:335-345.

Pigot, A. L., J. A. Tobias. 2013. Species interactions constrain geographic range expansion over evolutionary time. Ecology Letters 16:330-338.

Pigot, A. L., J. A. Tobias. *in review*. Dispersal sets the pace of secondary sympatry in vertebrates.

Poulin, R. 1998. Evolutionary Ecology of Parasites: From individuals to Communities. Chapman and Hall, London.

Power, A. G., C. E. Mitchell. 2004. Pathogen spillover in disease epidemics. The American Naturalist 164:S79-S89.

Prenter, J., C. MacNeil, J. T. Dick, and A. M. Dunn. 2004. Roles of parasites in animal invasions. Trends in Ecology & Evolution 19:385-390.

R Develompent Core Team. 2010. R: A language and environment for statistical computing. R Foundation Statistical Computing.

Rabosky, D. L., G. J. Slater, and M. E. Alfaro. 2012. Clade age and species richness are decoupled across the eukaryotic tree of life. PLoS biology 10:e1001381.

Rahbek, C. 1997. The relationship among area, elevation, and regional species richness in neotropical birds. The American Naturalist 149:875-902.

Rahbek, C. 1995. The Elevational Gradient of Species Richness - a Uniform Pattern. Ecography 18:200-205.

Rambaut, A., A. Drummond. 2012. TreeAnnotator version 1.7. 2.

Reullier, J., J. Pėrez-Tris, S. Bensch, and J. Secondi. 2006. Diversity, distribution and exchange of blood parasites meeting at an avian moving contact zone. Molecular Ecology 15:753-763.

Rickart, E. A. 2001. Elevational diversity gradients, biogeography and the structure of montane mammal communities in the intermountain region of North America. Global Ecology and Biogeography 10:77-100.

Ricklefs, R. E. 2007. Estimating diversification rates from phylogenetic information. Trends in Ecology & Evolution 22:601-610.

Ricklefs, R. E. 1987. Community diversity: relative roles of local and regional processes. Science 235:167-171.

Ricklefs, R. E., E. Bermingham. 2007. The causes of evolutionary radiations in archipelagoes: passerine birds in the Lesser Antilles. The American Naturalist 169:285-297.

Ricklefs, R. E., E. Bermingham. 2002. The concept of the taxon cycle in biogeography. Global Ecology and Biogeography 11:353-361.

Ricklefs, R. E., G. W. Cox. 1972. Taxon cycles in the West Indian avifauna. American Naturalist 106:195-219.

Ricklefs, R. E., B. L. Swanson, S. M. Fallon, A. MartÍnez-AbraÍn, A. Scheuerlein, J. Gray, and S. C. Latta. 2005. Community relationships of avian malaria parasites in southern Missouri. Ecological Monographs 75:543-559.

Ricklefs, R. E. 2010a. Host-pathogen coevolution, secondary sympatry and species diversification. Philosophical Transactions of the Royal Society B-Biological Sciences 365:1139-1147.

Ricklefs, R. E. 2010b. Evolutionary diversification, coevolution between populations and their antagonists, and the filling of niche space. Proceedings of the National Academy of Sciences of the United States of America 107:1265-1272.

Ricklefs, R. E., S. M. Fallon. 2002. Diversification and host switching in avian malaria parasites. Proceedings of the Royal Society of London. Series B: Biological Sciences 269:885-892.

Rodriguez, O., A., H. Moya, and N. E. Matta. 2009. Avian blood parasites in the National Natural Park Chingaza: high Andes of Colombia. Hornero 24:1-6.

Rohde, K. 1992. Latitudinal Gradients in Species Diversity: The Search for the Primary Cause. Oikos 65:514-527.

Root, T. 1988. Energy constraints on avian distributions and abundances. Ecology 69:330-339.

Rosenzweig, M. L. 1995. Species diversity in space and time. Cambridge University Press, Cambridge.

Rosenzweig, M. L. 1992. Species Diversity Gradients: We Know More and Less Than We Thought. Journal of Mammalogy 73:715-730.

Rosenzweig, M. L., Y. Ziv. 1999. The echo pattern of species diversity: pattern and processes. Ecography 22:614-628.

Sagarin, R. D., S. D. Gaines. 2002. The 'abundant centre'distribution: to what extent is it a biogeographical rule? Ecology Letters 5:137-147.

Salisbury, C. L., N. Seddon, C. R. Cooney, and J. A. Tobias. 2012. The latitudinal gradient in dispersal constraints: ecological specialisation drives diversification in tropical birds. Ecology Letters 15:847-855.

Sanders, N. J. 2002. Elevational gradients in ant species richness: area, geometry, and Rapoport's rule. Ecography 25:25-32.

Scheiner, S. M., M. R. Willig. 2005. Developing unified theories in ecology as exemplified with diversity gradients. The American Naturalist 166:458-469.

Schemske, D. W. 2009. Biotic interactions and speciation in the tropics. Pages 219-239 *In* Butlin, R. K., J. R. Bridle, and D. Schluter, editors. Speciation and Patterns in Diversity, Cambridge University Press, Cambridge.

Schemske, D. W. 2002. Ecological and evolutionary perspectives on the origins of tropical diversity. Pages 163-173 *In* Chazdon, R. L. and T. C. Whitmore, editors. Foundations of Tropical Forest Biology, University of Chicago Press, Chicago.

Schemske, D. W., G. G. Mittelbach, H. V. Cornell, J. M. Sobel, and K. Roy. 2009. Is There a Latitudinal Gradient in the Importance of Biotic Interactions? Annual Review of Ecology Evolution and Systematics 40:245-269.

Scheuerlein, A., R. E. Ricklefs. 2004. Prevalence of blood parasites in European passeriform birds. Proceedings of the Royal Society of London. Series B: Biological Sciences 271:1363-1370.

Schmidt, K. A., R. S. Ostfeld. 2001. Biodiversity and the dilution effect in disease ecology. Ecology 82:609-619.

Schoener, T. W. 1974. Resource partitioning in ecological communities. Science 185:27-39.

Schoener, T. W. 1968. Sizes of feeding territories among birds. Ecology 49:123-41.

Schrag, S. J., P. Wiener. 1995. Emerging infectious disease: what are the relative roles of ecology and evolution? Trends in Ecology & Evolution 10:319-324.

Schulenberg, T. S., D. F. Stotz, D. F. Lane, J. P. O'Neill, and T. A. Parker III. 2007. Birds of Peru. Princeton University Press, Princeton, New Jersey.

Sepil, I., S. Lachish, A. E. Hinks, and B. C. Sheldon. 2013. Mhc supertypes confer both qualitative and quantitative resistance to avian malaria infections in a wild bird population. Proceedings of the Royal Society of London. Series B: Biological Sciences 278:1025-1033.

Sexton, J. P., P. J. McIntyre, A. L. Angert, and K. J. Rice. 2009. Evolution and ecology of species range limits. Annual Review of Ecology, Evolution, and Systematics 40:415-436.

Snow, R. W., C. A. Guerra, A. M. Noor, H. Y. Myint, and S. I. Hay. 2005. The global distribution of clinical episodes of Plasmodium falciparum malaria. Nature 434:214-217.

Stevens, G. C. 1992. The elevational gradient in altitudinal range: and extension of Rapoport's latitudinal rule to altitude. The American Naturalist 140:893-911.

Stevens, G. C. 1989. The latitudinal gradient in geographical range - how so many species coexist in the tropics. The American Naturalist 133:240-256.

Stotz, D. F., T. A. Parker, and J. W. Fitzpatrick. 1996. Neotropical birds: ecology and conservation. The University of Chicago Press, Chicago.

Swanson, D., P. Adler. 2010. Vertical distribution of haematophagous Diptera in temperate forests of the southeastern USA. Medical and Veterinary entomology 24:182-188.

Tanner, J. T. 1952. Black-capped and Carolina chickadees in the southern Appalachian Mountains. The Auk 69:407-424.

Terborgh, J. 1971. Distribution on environmental gradients: theory and a preliminary interpretation of distributional patterns in the avifauna of the Cordillera Vilcabamba, Peru. Ecology 52:23-40.

Terborgh, J. 1977. Bird Species Diversity on an Andean Elevational Gradient. Ecology 58:1007-1019.

Terborgh, J., J. S. Weske. 1975. Role of Competition in Distribution of Andean Birds. Ecology 56:562-576.

Thrall, P. H., A. Biere, and M. K. Uyenoyama. 1995. Frequency-dependent disease transmission and the dynamics of the Silene-Ustilago host-pathogen system. The American Naturalist 145:43-62.

Tompkins, D., A. White, and M. Boots. 2003. Ecological replacement of native red squirrels by invasive greys driven by disease. Ecology Letters 6:189-196.

Torchin, M. E., K. D. Lafferty, A. P. Dobson, V. J. McKenzie, and A. M. Kuris. 2003. Introduced species and their missing parasites. Nature 421:628-630.

Valera, F., C. M. Carrillo, A. Barbosa, and E. Moreno. 2003. Low prevalence of haematozoa in Trumpeter finches *Bucanetes githagineus* from south-eastern Spain: additional support for a restricted distribution of blood parasites in arid lands. Journal of Arid Environments 55:209-213.

Valkiunas, G. 2005. Avian malaria parasites and other haemosporidia. CRC Press, United States of America.

van Riper III, C., S. G. van Riper, M. L. Goff, and M. Laird. 1986. The epizootiology and ecological significance of malaria in Hawaiian land birds. Ecological Monographs 56:327-344.

Vanriper, C., S. G. Vanriper, M. L. Goff, and M. Laird. 1986. The Epizootiology and Ecological Significance of Malaria in Hawaiian Land Birds. Ecological Monographs 56:327-344.

Vetaas, O. R., J. A. Grytnes. 2002. Distribution of vascular plant species richness and endemic richness along the Himalayan elevation gradient in Nepal. Global Ecology and Biogeography 11:291-301.

Von Humboldt, A. 1850. Aspects of nature, in different lands and in different climates; with scientific elucidations. Lea and Blanchard, Philadelphia.

Walker, B., D. Stotz, T. Pequeno, and J. W. Fitzpatrick. 2006. Birds of the Manu Biosphere Reserve. Fieldiana Zoology 110:23-49.

Wallace, A. 1869. The Malay Archipelago. Macmillan and Company, London.

Wallace, A. R. 1891. Natural selection and tropical nature: Essays on descriptive and theoretical biology. Macmillan, London and New York.

Warner, R. E. 1968. The role of introduced diseases in the extinction of the endemic Hawaiian avifauna. The Condor 70:101-120.

West-Eberhard, M. J. 1989. Phenotypic plasticity and the origins of diversity. Annual Review of Ecology and Systematics 20:249-278.

Westerdahl, H., J. Waldenström, B. Hansson, D. Hasselquist, T. von Schantz, and S. Bensch. 2005. Associations between malaria and MHC genes in a migratory songbird. Proceedings of the Royal Society of London. Series B: Biological Sciences 272:1511-1518.

Whittaker, R. H. 1965. Dominance and Diversity in Land Plant Communities: Numerical relations of species express the importance of competition in community function and evolution. Science 147:250-260.

Whittaker, R. H. 1960. Vegetation of the Siskiyou mountains, Oregon and California. Ecological Monographs 30:279-338.

Williamson, M. 1957. An elementary theory of interspecific competition. Nature 180:422-425.

Willig, M. R., S. K. Lyons. 1998. An analytical model of latitudinal gradients of species richness with an empirical test for marsupials and bats in the New World. Oikos 81:93-98.

Willig, M. R., D. M. Kaufman, and R. D. Stevens. 2003. Latitudinal gradients of biodiversity: Pattern, process, scale, and synthesis. Annual Review of Ecology Evolution and Systematics 34:273-309.

Wilson, E. O. 1961. The nature of the taxon cycle in the Melanesian ant fauna. The American Naturalist 95:169-193.

Wilson, M. L. 2001. Ecology and infectious disease. Pages 283-324 *In* Aron, J. L. and J. A. Patz, editors. Ecosystem Change and Public Health: A Global Perspective, Johns Hopkins University Press, Baltimore, Maryland.

Wolda, H. 1987. Altitude, Habitat and Tropical Insect Diversity. Biological Journal of the Linnean Society 30:313-323.

Wood, M. J., C. L. Cosgrove, T. A. Wilkin, S. C. L. Knowles, K. P. Day, and B. C. Sheldon. 2007. Within-population variation in prevalence and lineage distribution of avian malaria in blue tits, Cyanistes caeruleus. Molecular Ecology 16:3263-3273.

Woodhams, D., K. Ardipradja, R. Alford, G. Marantelli, L. Reinert, and L. Rollins-Smith. 2007. Resistance to chytridiomycosis varies among amphibian species and is correlated with skin peptide defenses. Animal Conservation 10:409-417.

Woodworth, B. L., C. T. Atkinson, D. A. LaPointe, P. J. Hart, C. S. Spiegel, E. J. Tweed, C. Henneman, J. LeBrun, T. Denette, R. DeMots, K. L. Kozar, D. Triglia, D. Lease, A. Gregor, T. Smith, and D. Duffy. 2005. Host population persistence in the face of introduced vector-borne diseases: Hawaii amakihi and avian malaria. Proceedings of the National Academy of Sciences of the United States of America 102:1531-1536.

Woolhouse, M. E., D. T. Haydon, and R. Antia. 2005. Emerging pathogens: the epidemiology and evolution of species jumps. Trends in Ecology & Evolution 20:238-244.

World Health Organization. 2012. World Malaria Report 2012—Fact Sheet.

Yguel, B., R. Bailey, N. D. Tosh, A. Vialatte, C. Vasseur, X. Vitrac, F. Jean, and A. Prinzing. 2011. Phytophagy on phylogenetically isolated trees: why hosts should escape their relatives. Ecology Letters 14:1117-1124.

Zamora-Vilchis, I., S. E. Williams, and C. N. Johnson. 2012. Environmental Temperature Affects Prevalence of Blood Parasites of Birds on an Elevation Gradient: Implications for Disease in a Warming Climate. PLoS one 7:e39208.

Appendix 1

Bird diversity along an Andes-Amazon gradient in Manu Biosphere Reserve, Peru: The role of environmental variables, area, mid-domain effects and Rapoport's rule

Abstract

I studied bird species richness between 200 m and 4000 m along an Andean elevational gradient and tested the roles of temperature, NPP, precipitation, area and mid-domain effects (MDE) in shaping observed patterns. Patterns of species richness with elevation were investigated for all resident bird species and individually for insectivore, carnivore, frugivore, nectarivore and granivore foraging guilds. In addition, I also tested if Rapoport's rule of increasing range amplitude with increasing elevation applies to birds occurring along the study gradient. I recorded three distinct patterns of species richness with elevation: (1) declining, (2) declining with a mid-elevation plateau, and (3) bimodal with peaks separated by a mid-elevation plateau. The pattern of species richness with elevation for all resident bird species and the frugivore guild was declining; carnivore and granivore guilds showed a declining richness pattern with a mid-elevation plateau, and nectarivores birds showed a bimodal pattern with peaks at high and low elevations. The predictions of the mid-domain effect were not supported for all resident bird species nor any of the foraging guilds.

However, for the nectarivore guild, regression of the empirical richness on the null model MDE predictions explained a significant amount of the observed variation (R^2 = 0.11, P = 0.035). Rapoport's rule of increasing range amplitude with elevation was supported for all resident bird species and each of the insectivore, frugivore and nectarivore foraging guilds, but not for birds belonging to the carnivore and granivore guilds. This study shows that among the environmental variables examined, NPP and area were the most important variables for predicting the elevational richness patterns in birds. The mid-domain effect was only supported as a primary driver of species richness patterns with elevation for the nectarivore foraging guild. Rapoport's Rule was supported up to ~2000 m for all resident bird species and each of the foraging guilds, but when considering species range sizes across the entire gradient, the effect of increasing range size with elevation was only detected in all resident bird species and each of the insectivore, frugivore and nectarivore foraging guilds. Results show that a range of biotic and abiotic factors underlie observed richness patterns.

Introduction

Understanding species richness patterns with elevation is a central theme in ecology and has received considerable attention in the past ecological literature. Declining species richness with increasing elevation is one of the most well noted macroecological patterns (Lomolino 2001) and was previously considered to be ubiquitous across taxa, regions and spatial scales (Kattan and Franco 2004). The majority of early studies on elevational gradients of species richness recorded monotonic declines in species richness with increasing elevation and interpreted these patterns as reflecting downscaled versions of latitudinal diversity gradients, which

typically show a decline in species richness with increasing latitude (Brown and Southwood 1983, Rohde 1992). Rahbek (1995) was among the first to critically evaluate this generalization, and showed that although species richness declines with elevation, this decline is not necessarily monotonic. Moreover, Rahbek (1995, 1997) also highlighted the influence sampling effort and area can have on observed relationships, noting that failure to account for these can lead to artefactual results.

Re-evaluation of earlier studies and results from more recent investigations has revealed that a wide range of species richness patterns with elevation are possible, including: monotonic decline, low elevational plateau, low-elevation plateau with a mid-elevation peak and hump-shaped distributions (McCain and Grytnes 2010). Of these, the two most commonly recorded patterns are 1) monotonic declines in species richness with increasing elevation (Lomolino 2001, Terborgh 1977) and 2) hump-shaped species richness patterns centred on the gradients elevational midpoint (Brehm et al. 2007, McCain 2004, Rahbek 1997). The consensus from research into species richness patterns with elevation highlights the great variation that exists in recorded patterns across taxons and gradients, and illustrates that a range of patterns are common. Efforts to explain this wide variation have been the subject of much research seeking to understand the possible mechanistic drives of these patterns (Hawkins et al. 2003).

Because species are responding to variables associated with elevation (not elevation itself) to properly evaluate species richness patterns along elevational gradients it is necessary to base analyses on well-resolved species richness data that takes into account area, environmental variables and appropriate null model distributions.

Developing a detailed understanding of the shape of richness patterns is the first step in investigating possible explanatory variables responsible for driving observed patterns. Assessing the relative importance of these variables is informative for understanding species distributions in general, not just along elevational gradients.

A range of explanatory variables have been proposed as drivers of species richness patterns and among these, climatic variables such as temperature, precipitation, net primary productivity (NPP) and area are each somewhat supported from large-scale studies (Acharya et al. 2011, Brehm et al. 2007, Currie et al. 2004, Karger et al. 2011). Along mountains, temperature is among the most consistent environmental variables that changes predictably with elevation, on average decreasing by 0.6°C per 100 m increase in elevation (Barry 2008). Other important actors that change with elevation but which show much more variation are precipitation, productivity and area. In addition to climatic variable underlying patterns of species richness, abiotic factors such as spatial constraints arising from species ranges naturally overlapping most toward the centre of their distribution in a bounded domain, are also hypothesized to shape observed patterns. The mid-domain effect (MDE) describes the hump-shaped distribution pattern that results from randomly placing species ranges within a geometric null model with hard outer boundaries and has previously been shown to correlate with empirical data for a range of species gradients (Willig and Lyons 1998, Colwell and Hurtt 1994). For certain richness gradients, mid domain null distributions have been shown to explain almost all the variation in the empirical richness patterns with elevation (Bachman et al. 2004, Sanders 2002), yet their overall ubiquity and potential ecological significance remains largely unknown (Colwell et al. 2004, Kessler 2000).

Here I investigate elevational species distributions of birds within the Manu Biosphere Reserve, Peru. The reserve is situated in the departments of Madre de Dios and Cuzco and is Peru's largest national conservation area, covering approximately 18,812 km^2 and encompassing an elevational range extending from 250 m in the Amazonian lowlands to 4000 m in the Andean highlands (Patterson et al. 1996, Walker et al. 2006). The reserve contains one of the highest diversities of birds in the world with over 1000 recorded species. Along the western edge of the park where the reserve boarders the Andes mountain chain, within relatively short horizontal distances one can experience large environmental variation as forest changes from high elevation puna grassland to cloud forest, montane forest and then lowland rainforest. This wide environmental variation is mirrored by biological variation and the limited elevational ranges of many tropical bird species makes this region ideally suited to investigating patterns of species richness with elevation. Birds have previously been used as a model group for investigating patterns of species richness with elevation because they are often abundant, display wide ecological and evolutionary diversity, and show high species turnover with increasing elevation – all of which make them ideally suited to testing a range of possible causal factors influencing their distribution.

The considerable amount of environmental variation captured by elevational gradients over short geographical distances makes them ideally suited to investigating the underlying factors shaping species richness patterns. Unlike latitudinal gradients in diversity, elevational gradients allow for direct measures of climate, diversity and habitat characteristics along continuous transects. Elevational gradients are also less confounded by area, history, biogeography and human interference than latitudinal

gradients and for these reasons, provide an ideal system within which to test hypotheses and predictions concerning proposed drivers of biodiversity.

By analyzing the species richness patterns of all resident bird species recorded from Manu Biosphere Reserve, and separately for a range of distinct foraging guilds (based on primary diets ranging from insects, vertebrates, fruits, nectar and seeds) I examine the generality of bird richness patterns with elevation. If all guilds show similar patterns of richness with elevation then the primary drivers of such relationships can be attributed to abiotic environmental variables, whereas if different pattern are recorded for each of the guilds then this supports the idea that biotic interactions also play an important role in shaping observed relationships, something that requires further investigation (Schemske 2009, Schemske et al. 2009).

To examine the relationship between species distributions and elevation, I also investigate whether Rapoport's Rule of increasing range size with elevation applies to birds recorded along the study gradient. The rule posits that because climate variability increases toward higher elevations, species at higher elevations should be able to tolerate a wider range of environmental conditions and therefore occupy larger elevational ranges. Although Rapoport's rule is generally considered to be at best equivocally supported from latitudinal studies, early studies of elevational gradients have often revealed strong Rapoport effects for a range of organisms including trees, mammals, birds, reptiles, insects and amphibians (Stevens 1992) and further studies from ants (Sanders 2002) and moths (Brehm et al. 2007) provide additional support. However, other studies from a range of species have found no support for Rapoport

effects with elevation (Gaston et al. 1998, Bhattarai et al. 2004), so clearly it's generality requires further investigation from even more species and regions.

The main aims of this study are: (1) to describe the elevational diversity pattern of birds of the Manu biosphere reserve separately for (i) all resident bird species, and (ii) individual foraging guilds including insectivores, nectarivores, frugivores, granivores and carnivores; (2) to investigate the explanatory potential of temperature, precipitation, net primary productivity (NPP) and area in shaping observed patterns; (3) to test the applicability of a mid-domain null model in describing observed relationships between species richness and elevation; and (4) to investigate if there is evidence for Rapoport's rule of increasing range amplitude with increasing elevation for all resident birds and each of the individual foraging guilds.

Materials and Methods

Bird elevational data

Bird diversity data was compiled from 'Birds of the Manu Biosphere Reserve' (Walker et al. 2006), a well-resolved dataset that lists all the species of bird recorded within Manu Biosphere Reserve based on their sight, sound or sign (i.e. discarded feathers, pellets, nests, droppings, and skulls and bones) records as well as the species seasonal status (resident, migrant, vagrant) and maximum and minimum elevation for which records have been collected. Because this dataset represents a regional scale (gamma) species richness estimate based on sight sound and capture records, the observed richness patterns are likely to be highly influenced by area (Lomolino 2001, Rahbek 1997) and may have significant sampling effort biases (McCain 2007a,

Rickart 2001). This source provides maximum and minimum elevational limits for a total 1004 species of birds across an elevational range from 250 m – 4000 m. The gradient was divided into 100 m intervals beginning at 200 m and a species was defined as present in each 100 m band between its lower and upper elevational limit as used in similar studies on other taxa (Bhattarai et al. 2004, Grytnes and Beaman 2006, Patterson et al. 1996, Rahbek 1997). For instance, if a species elevational range was recorded as 250 – 600 m, then it classified as be present in the 200, 300, 400, 500 and 600 m elevational bands. Interpolating species richness in this way gives an estimate of regional species richness (gamma diversity), defined as the total richness within an elevational zone (Bhattarai et al. 2004, Vetaas and Grytnes 2002). I use the term species richness to define the number of bird species recorded, as the term diversity is often used to indicate a measure of species richness and evenness.

Climate and area data

To estimate how climatic variables change with elevation within the study site, I used data from Girardin *et al.*(2010) which lists mean annual air temperature, annual precipitation and net primary productivity (NPP) recorded at weather stations situated within the study region at seven elevations at 210, 1000, 1500, 1855, 2020, 2720, and 3025 m. I plotted each of these environmental variables against elevation and fitted a line of best fit to these data. The climatic variables for each of the missing elevational bands were then interpolated using the function for the line of best fit to calculate missing values.

Area estimates for each of the 100 m elevational bands within Manu National Park were taken from published work by (McCain 2007b) which calculated the area of 100

m elevational bands within the same region as part of a meta-analysis on the relationship between area and mammalian elevational diversity. Area has been shown to have strong effects on species richness at all spatial scales as larger areas contain more individuals and therefore more species (Connor and McCoy 1979). Although species richness generally increases with area, the relationship is not linear so I use log-transformed area as a potential explanatory variable (Rosenzweig and Ziv 1999, Kluge et al. 2006).

Statistical methods

The relationship between species richness and each of the environmental variables including area was calculated for each individual variable using simple linear regression. This analysis tests for linear relationships between the response variable and each of the explanatory variables; the monotonic declines in temperature and precipitation with elevation supported this type of analysis. To explore the relative importance of the different environmental variables and area to explain elevational patterns in bird species richness I used a generalized linear model (GLM) with Poisson errors. Poisson errors were used because the species richness data was categorical (rounded to 50 m intervals and binned into 100 m elevational bands) and with a non-normal distribution that could not be corrected through transformation. Model simplification was undertaken using a stepwise deletion procedure with and without the term of interest, and the difference in model deviance was assessed with an analysis of variance. The final model includes species richness as the response variable, and elevation of sampling locality, temperature, precipitation, net primary productivity (NPP) and log(area) as fixed effects. To consider whether each of the environmental factors and area influenced each of the foraging guilds differently,

analyses were run on all resident bird species and each of the foraging guilds separately. Final model outputs were checked for overdispersion by comparing the residual deviance to the degrees of freedom and if overdispersion was detected the model was re-run with a quasipoisson error distribution to account for this. Prior to analysis, area was the only variable that was transformed (to log(area)), because of the non-linear relationship between species richness area, and because of the highly skewed area elevation relationship in which both high and low elevations have disproportionately large areas in comparison to other elevations (Fig. 4D) (Rosenzweig and Ziv 1999, Kluge et al. 2006).

Mid-domain effect

Here I explore whether species richness patterns for all resident bird species and individual foraging guilds fit mid domain null model predictions, generated from a Monte Carlo simulation procedure using the programme 'Mid-Domain Null' (McCain 2004). This program uses species empirical ranges or range midpoints to simulate species richness curves within a bounded domain based on analytical models developed by Colwell (1999) and Colwell and Hurt (1994). The mid domain null model samples from imported empirical data for either range midpoints or range sizes and by randomizing the placement of these within a bounded domain, describes the null geometric pattern that results from this random placement. The programme "Mid-Domain Null" was run on all empirical richness estimates for all resident birds and each of the foraging guilds separately using each recorded species range size, midpoint and upper and lower elevation limit, and specifying a bounded domain between 250 and 4000 m within 39 elevational bins. 50 000 simulations were run with replacement (because I had no *a priori* expectations of the shape of the curve) and the chosen output was to display empirical species richness graph and 95% confidence limits

graph. Regressions of the empirical values on predicted values gave R^2 estimates of the fit to MDE.

Rapoport effect

To examine the relationship between range size and elevation, I calculated the elevational range size of each species by subtracting the maximum elevation and minimum elevations at which the species was recorded. Species midpoints were calculated as the mean of the highest and lowest elevation at which a species was recorded. Rapoport's rule was investigated following the method of Stevens (1992) which reports the average range size for all species recorded within a particular region along the gradient of interest (i.e. average range size for all species recorded within each 100 m elevational band). Rapoport effects were quantified by Spearman rank correlation analysis between elevation and mean elevation range size. Although *P*-values are presented for these correlations, they should be interpreted with caution as the data points in Stevens plots are not independent as the data is spatially auto correlated. This is because closer sites more are likely to share similar species assemblages, and therefore many of the same species are likely to be sampled and their ranges averaged across multiple sites (Stevens 1992).

Results

Environmental variables

Both temperature and precipitation showed a decreasing linear trend with elevation, whereas NPP showed and overall exponential decreasing pattern (Figure 7.1). Within

the Manu Biosphere area decreases linearly with elevation up to ~ 3000 and then shows an exponential increase due to large amounts of available area at high elevations associated with the Andean mountain range.

Species richness and elevation

In total, maximum and minimum elevational records were available for 915 resident bird species listed in the 'Birds of the Manu Biosphere Reserve' (Walker et al. 2006). Using data on the primary diet of each of these species from field guides and online databases, 542 species were classified as insectivores, 45 as carnivores, 153 as frugivores, 70 as nectarivores and 49 and granivores. These results are based on species with one distinct primary diet source i.e. insects, fruit, vertebrates, nectar or seeds. Species with two or more primary diet sources were excluded from the analyses. Table 7.1 lists the total number of species recorded for all resident bird species and each of the five foraging guilds, as well as the corresponding environmental variables and area estimates for each 100 m wide elevational band.

Plotting species richness by elevation for all resident birds and each of the separate foraging guilds revealed the overall expected decline in richness with elevation, but with considerable variation among each of the slopes. Three distinct patterns of species richness with elevation were recorded: (1) declining species richness with elevation for all resident bird species and birds belonging to the insectivore and frugivore guild; (2) declining species richness with a mid-elevation plateau for birds belonging to carnivore and granivore guilds; and (3) a bimodal pattern with peaks at high and low elevations separated by a mid-elevation plateau was recorded for birds belonging to the nectarivore guild (Figure 7.2).

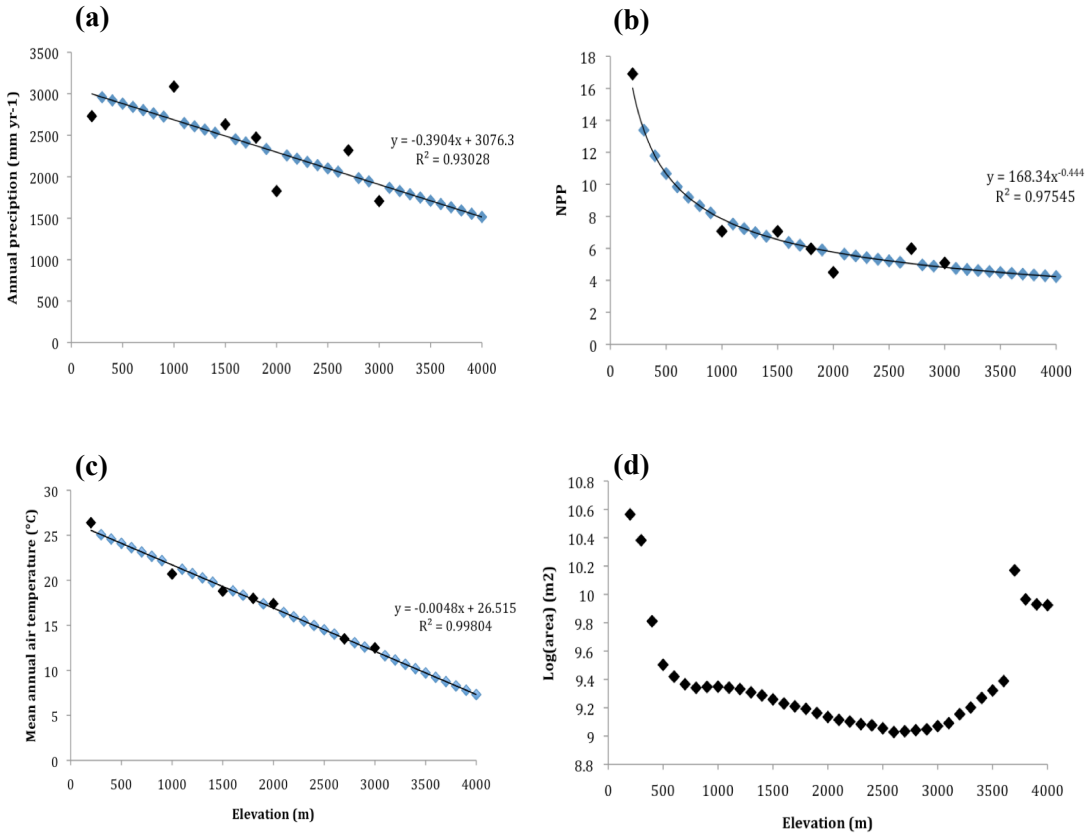


Figure 7.1 The relationship between climate, and elevation with Manu Biosphere Reserve, Peru from seven weather stations at 210, 1000, 1500, 1855, 2020, 2720, and 3025 (black points) with interpolated values (blue points) at each 100 m band occurring between these for (a) precipitation (b) NPP, (c) temperature and (d) area within the study site.

Correlates of species richness

Within the Manu Biosphere Reserve between 200 and 4000 m species richness of all resident birds was strongly negatively correlated with elevation, and positively correlated with temperature, precipitation and NPP (Table 7.2). In other words, sites at lower elevations with higher mean temperature, rainfall and productivity, had the highest number of species. Analyzing each of the guilds separately showed that for all guilds species richness was strongly negatively correlated with elevation. For insectivorous and frugivorous birds, species richness was positively correlated with

temperature, precipitation and NPP; and for carnivorous birds, species richness was strongly positively correlated with both temperature and precipitation. Nectarivorous birds showed strong positive correlation with precipitation and NPP and were also the only guild with strong positive correlations between species richness and area.

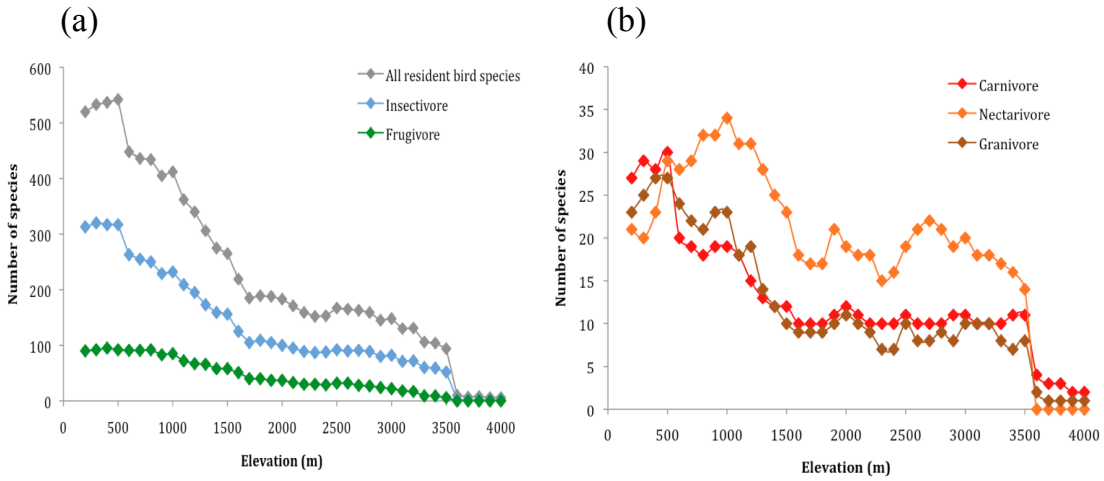


Figure 7.2 Relationship between species richness and elevational band for birds within the Manu Biosphere Reserve, for (a) all resident birds (grey), and birds belonging to insectivorous (blue) and frugivorous foraging guilds, and (b) birds belonging to carnivorous (red), nectarivorous (orange) and granivorous (brown) foraging guilds. Relationships are plotted on separate graphs to illustrate the different richness patterns that are otherwise not visible due to large differences in the total number of species in each of the guilds.

A general linear model with species richness as the response variable and all environmental variables and area as fixed effects explained a large amount of the observed variation in species richness patterns for all resident bird species (Table 7.3). For all resident birds and each of the insectivore, nectarivore and granivore foraging guilds, a significant amount of the observed variation in species richness was explained by NPP and area effects. Species richness of birds belonging to the carnivore foraging guild was best explained by elevation, NPP, area and temperature

variables. The only significant predictor of species richness for the frugivore foraging guild was area. In the final models, the effect of temperature, precipitation and elevation were no longer important in explaining species richness for all resident bird species or any of the guilds (except for carnivores, in which both temperature and elevation remained significant predictors of species richness). Overall results from these analyses show that the effects of temperature, precipitation and elevation mainly drop out as explanatory variables when the effects of NPP and area are included, indicating that these two variables are the best predictors of species richness for all resident birds and each of the foraging guilds within the study site.

Table 7.1 Elevation of 39 elevational bands, listing species richness and corresponding environmental variables including temperature, precipitation, net primary productivity and area. Results are presented for all resident bird species and insectivorous, carnivorous, frugivorous, nectarivorous and granivorous foraging guilds. Results are based on interpolations from maximum and minimum elevational ranges presented in "Birds of Manu Biosphere Reserve" and based on sight, sound, sign and capture records (Walker et al. 2006).

						Foraging guild				
Elevation (m)	Mean annual air temp (°C)	Annual precipitation (mm.yr-1)	Net primary productivity (NPP)	Area (m^2)	All resident bird species	Insecti	Carni	Frugi	Nectari	Grani
200	26.4	2730.0	16.9	36708805944	520	313	27	90	21	23
300	25.1	2959.2	13.4	24082429320	533	320	29	92	20	25
400	24.6	2920.1	11.8	6462754251	537	317	28	95	23	27
500	24.1	2881.1	10.7	3184874239	542	317	30	92	29	27
600	23.6	2842.1	9.8	2627801835	448	263	20	91	28	24
700	23.1	2803.0	9.2	2321156828	436	255	19	91	29	22
800	22.7	2764.0	8.7	2190616197	434	250	18	92	32	21
900	22.2	2724.9	8.2	2222708654	405	229	19	83	32	23
1000	20.7	3087.0	7.1	2230752938	412	232	19	85	34	23
1100	21.2	2646.9	7.5	2196759104	362	209	18	72	31	18
1200	20.7	2607.8	7.2	2146176342	340	195	15	67	31	19
1300	20.3	2568.8	7.0	2034926596	306	173	13	66	28	14
1400	19.8	2529.7	6.8	1936001154	275	159	12	58	25	12
1500	18.8	2631.0	7.1	1815129059	265	156	12	58	23	10
1600	18.8	2451.7	6.4	1697597843	219	125	10	51	18	9
1700	18.3	2412.6	6.2	1621373446	185	105	10	40	17	9
1800	18.0	2472.0	6.0	1556780544	189	109	10	40	17	9
1900	17.4	2334.5	5.9	1453906090	188	105	11	37	21	10
2000	17.4	1827.0	4.5	1362955651	183	100	12	37	19	11
2100	16.4	2256.5	5.6	1301388015	171	95	11	33	18	10
2200	15.9	2217.4	5.5	1265870002	159	89	10	30	18	9
2300	15.5	2178.4	5.4	1213501331	152	87	10	30	15	7
2400	15.0	2139.3	5.3	1191885687	153	88	10	29	16	7
2500	14.5	2100.3	5.2	1134621166	167	92	11	32	19	10
2600	14.0	2061.3	5.1	1067780483	165	90	10	32	21	8
2700	13.5	2318.0	6.0	1082075445	163	91	10	28	22	8
2800	13.1	1983.2	5.0	1100250137	159	89	10	27	21	9
2900	12.6	1944.1	4.9	1116315610	145	80	11	24	19	8
3000	12.5	1706.0	5.1	1175350644	148	82	11	22	20	10
3100	11.6	1866.1	4.7	1233246391	130	71	10	18	18	10
3200	11.1	1827.0	4.7	1427979634	131	72	10	17	18	10
3300	10.7	1788.0	4.6	1590651211	106	60	10	9	17	8
3400	10.2	1748.9	4.6	1858175598	104	59	11	9	16	7
3500	9.7	1709.9	4.5	2097002291	94	52	11	6	14	8
3600	9.2	1670.9	4.4	2444207422	10	2	4	0	0	2
3700	8.7	1631.8	4.4	14777510400	7	2	3	0	0	1
3800	8.3	1592.8	4.3	9224622345	8	2	3	0	0	1
3900	7.8	1553.7	4.3	8504685916	6	2	2	0	0	1
4000	7.3	1514.7	4.2	8399963971	6	2	2	0	0	1

Table 7.2 Univariate analysis of variance showing coefficients of determination (R^2) of diversity recorded for every elevational band of 100 m with explanatory variables for all resident bird species and separated for insectivore, carnivore, frugivore, nectarivore and granivore foraging guilds. Analyses are based on maximum and minimum elevational limits recorded for birds spanning 200 – 4000 m within the Manu Biosphere Reserve (Walker et al. 2006).

Richness variable	Explanatory variable	R^2	Intercept ± se	*T - value*	*P*-value	Slope ± se	*T - value*	*P*-value
All resident birds	Temperature	0.91	-229.665 ± 24.915	-9.218	<0.001	27.956 ± 1.44	19.414	<0.001
d.f. 37	Precipitation	0.85	-493.4 ± 50.116	-9.845	<0.001	0.321 ± 0.022	14.724	<0.001
	NPP	0.79	-112.195 ± 31.34	-3.58	<0.005	51.666 ± 4.378	11.8	<0.001
	log(Area)	0.06	-753.867 ± 625.277	-1.206	0.236	45.571 ± 28.944	1.574	0.124
	Elevation	0.91	512.249 ± 16.264	31.495	<0.001	-0.134 ± 0.007	-19.701	<0.001
Insectivore	Temperature	0.91	-140.987 ± 15.089	-9.343	<0.001	16.595 ± 0.872	19.029	<0.001
d.f. 37	Precipitation	0.85	-296.394 ± 30.578	-9.693	<0.001	0.19 ± 0.013	14.286	<0.001
	NPP	0.81	-74.441 ± 17.633	-4.222	<0.001	31.151 ± 2.463	12.647	<0.001
	log(Area)	0.07	-505.452 ± 369.52	-1.368	0.180	29.521 ± 17.105	1.726	0.093
	Elevation	0.91	299.319 ± 9.917	30.183	<0.001	-0.08 ± 0.004	-19.169	<0.001
Carnivore	Temperature	0.76	-5.367 ± 1.755	-3.058	<0.005	1.11 ± 0.101	11.701	<0.001
d.f. 37	Precipitation	0.68	-15.053 ± 3.228	-4.663	<0.001	0.012 ± 0.001	8.826	<0.001
	NPP	0.79	-1.924 ± 1.368	-1.407	0.168	2.235 ± 0.191	10.939	<0.001
	log(Area)	0.07	-33.556 ± 26.93	-1.246	0.221	2.151 ± 1.247	-10.805	0.093
	Elevation	0.76	24.035 ± 1.172	20.504	<0.001	-0.005 ± 0	1.72	<0.001
Frugivore	Temperature	0.95	-50.513 ± 3.756	-13.449	<0.001	5.699 ± 0.217	26.254	<0.001
d.f. 37	Precipitation	0.91	-106.252 ± 7.673	-13.848	<0.001	0.066 ± 0.003	19.869	<0.001
	NPP	0.70	-21.146 ± 7.477	-2.828	<0.05	9.713 ± 1.044	9.300	<0.001
	log(Area)	0.04	-106.642 ± 126.594	-0.842	0.405	6.94 ± 5.86	1.184	0.244
	Elevation	0.96	100.891 ± 2.285	44.158	<0.001	-0.027 ± 0.001	-28.671	<0.001
Nectarivore	Temperature	0.61	-2.242 ± 2.967	-9.343	0.455	1.306 ± 0.171	19.029	<0.001
d.f. 37	Precipitation	0.64	-16.527 ± 4.486	-9.693	<0.005	0.016 ± 0.002	14.286	<0.001
	NPP	0.23	8.613 ± 3.417	-4.222	<0.05	1.604 ± 0.477	12.647	<0.005
	log(Area)	0.10	91.071 ± 34.91	-1.368	<0.05	-3.328 ± 1.616	1.726	<0.05
	Elevation	0.62	32.562 ± 1.927	30.183	<0.001	-0.006 ± 0.001	-19.169	<0.001
Granivore	Temperature	0.82	-8.619 ± 1.686	-5.112	<0.001	1.259 ± 0.097	12.921	<0.001
d.f. 37	Precipitation	0.77	-20.512 ± 3	-6.837	<0.001	0.014 ± 0.001	11.083	<0.001
	NPP	0.69	-3.082 ± 1.816	-1.697	0.098	2.29 ± 0.254	9.027	<0.001
	log(Area)	0.04	-23.929 ± 30.106	-0.795	0.432	1.668 ± 1.394	1.197	0.239
	Elevation	0.82	24.813 ± 1.103	22.495	<0.001	-0.006 ± 0	-13.01	<0.001

Table 7.3 Generalized linear model (GLM)[1] of diversity recorded for each elevational band of 100 m with explanatory variables for all resident bird species and separated for insectivore, carnivore, frugivore, nectarivore and granivore foraging guilds respectively.

Richness variable	Model Fit R^2	Explanatory variable	Estimate ± se	Test statistic	*P*-value
All resident birds	0.84	Intercept	28.354 ± 9.965	8.094	<0.05
d.f. 33		Temperature	-0.448 ± 0.290	2.381	0.132
		Precipitation	-0.001 ± 0.001	0.927	0.342
		NPP	0.179 ± 0.049	13.381	<0.001
		log(Area)	-0.460 ± 0.098	21.902	<0.001
		Elevation	-0.003 ± 0.002	3.233	0.081
Insectivore	0.84	Intercept	27.668 ± 10.520	6.917	<0.05
d.f. 33		Temperature	-0.441 ± 0.306	2.08	0.159
		Precipitation	-0.001 ± 0.001	0.748	0.394
		NPP	0.186 ± 0.052	12.953	<0.001
		log(Area)	-0.468 ± 0.104	20.25	<0.001
		Elevation	-0.003 ± 0.002	2.846	0.101
Carnivore	0.81	Intercept	37.135 ± 14.439	2.572	<0.05
d.f. 33		Temperature	-0.876 ± 0.427	-2.051	<0.05
		Precipitation	-0.001 ± 0.001	-1.667	0.095
		NPP	0.244 ± 0.068	3.612	<0.001
		log(Area)	-0.41 ± 0.125	-3.282	<0.005
		Elevation	-0.005 ± 0.002	-2.091	<0.05
Frugivore	0.87	Intercept	16.011 ± 10.265	2.4336	0.128
d.f. 33		Temperature	-0.096 ± 0.298	0.103	0.750
		Precipitation	0.000 ± 0.001	0.009	0.926
		NPP	0.079 ± 0.051	2.338	0.136
		log(Area)	-0.001 ± 0.002	14.846	<0.001
		Elevation	-0.001 ± 0.002	0.594	0.446
Nectarivore	0.79	Intercept	31.065 ± 14.089	4.862	<0.05
d.f. 33		Temperature	-0.400 ± 0.412	0.941	0.339
		Precipitation	-0.001 ± 0.001	0.469	0.498
		NPP	0.207 ± 0.073	7.969	<0.05
		log(Area)	-0.792 ± 0.154	26.429	<0.001
		Elevation	-0.002 ± 0.002	1.1025	0.301
Granivore	0.78	Intercept	36.509 ± 14.968	2.439	<0.05
d.f. 33		Temperature	-0.797 ± 0.438	-1.818	0.069
		Precipitation	-0.001 ± 0.001	-1.416	0.157
		NPP	0.21 ± 0.072	2.907	<0.005
		log(Area)	-0.475 ± 0.142	-3.349	<0.001
		Elevation	-0.005 ± 0.002	-1.941	0.052

[1]Model selection involved stepwise linear deletion of non-significant terms and outputs from final model are presented. This model includes species richness as the response variable, and elevation of sampling locality, temperature, precipitation, net primary productivity (NPP) and log(Area) as fixed effects. Analyses are based on maximum and minimum elevational limits recorded for birds spanning 200 – 4000 m within the Manu Biosphere Reserve (Walker et al. 2006). P values are based on z-value estimates where $P = \Pr(>|z|)$.

Mid-domain effect

When species richness was analysed against mid domain null predictions for all resident bird species (R^2 = 0.04, F = 1.388, P = 0.246) and each of the insectivore (R^2 = 0.04, F = 1.667, P = 0.205), carnivore (R^2 = 0.06, F = 2.442, P = 0.127), frugivore (R^2 = 0.01, F = 0.485, P = 0.490) and granivore guilds (R^2 = 0.03, F = 2.66, P = 1.273), none of the observed relationships showed a significant fit to MDE null models (Figure 7.3). However, for the nectarivore guild, regression of the empirical richness on the null model MDE predictions explained a significant amount of the observed variation (R^2 = 0.11, F = 4.811 P = 0.035).

Rapoport's rule

The elevational extent of all species ranges and each of the individual foraging guilds recorded in each 100 m elevational band increased with elevation up to ~ 2000 m (Figure 7.4). Above 2000 m, the average range size of showed much more variable patterns. Using Spearman rank correlation to test for a significant association between elevation and the mean elevational range size of all species occurring at the same elevation revealed significant Rapoport effects for all resident bird species (Spearman's rho = 0.547, d.f. = 37, P = <0.001) and each of the insectivore (Spearman's rho = 0.711, d.f. = 37, P = <0.001), frugivore foraging guilds (Spearman's rho = 0.680, d.f. = 32, P = <0.001) and nectarivore (Spearman's rho = 0.463, d.f. = 32, P = 0.006) foraging guilds. Neither of the carnivore (Spearman's rho = 0.192, d.f. = 37, P = 0.241) or granivore foraging guilds showed significant Rapoport effects (Spearman's rho = -0.087, d.f. = 37, P = 0.600). The corresponding Stevens plots are presented in Figure 7.4.

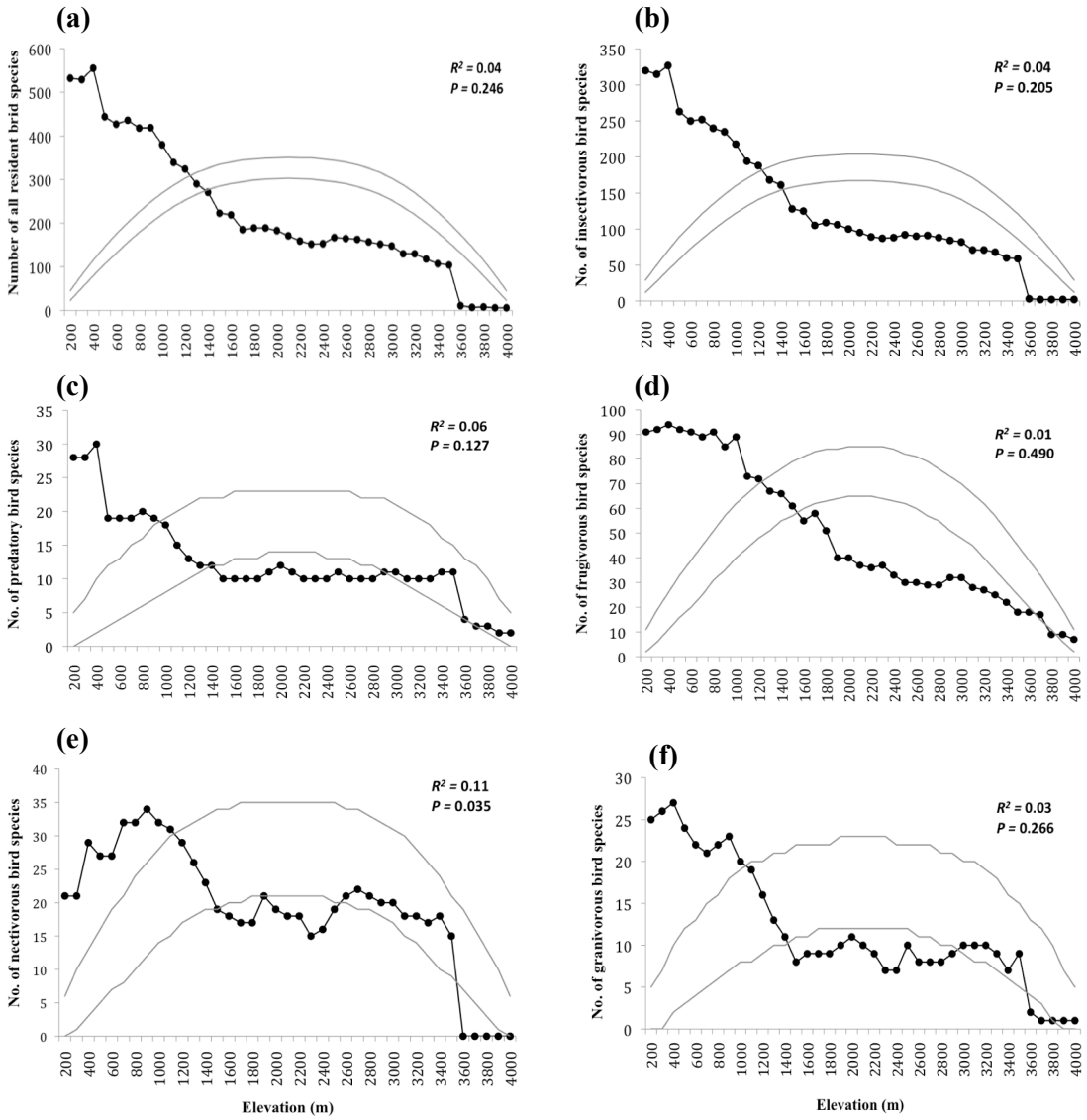


Figure 7.3 Predicted species richness under the assumption of random range placement (MDE) (lines showing upper and lower 95% confidence limits) and interpolated species richness for birds recorded along an elevational gradient spanning 200 – 4000 m in Manu Biosphere Reserve, Peru. Results are shown for (a) all resident bird species, (b) insectivorous bird species, (c) carnivorous bird species, (d) frugivorous bird species, (e) nectarivorous bird species and (f) granivorous bird species. R^2 values indicate the variance in interpolated, recorded species richness that is explained by predicted species richness. *P*-values for R^2 based on 37 d.f.

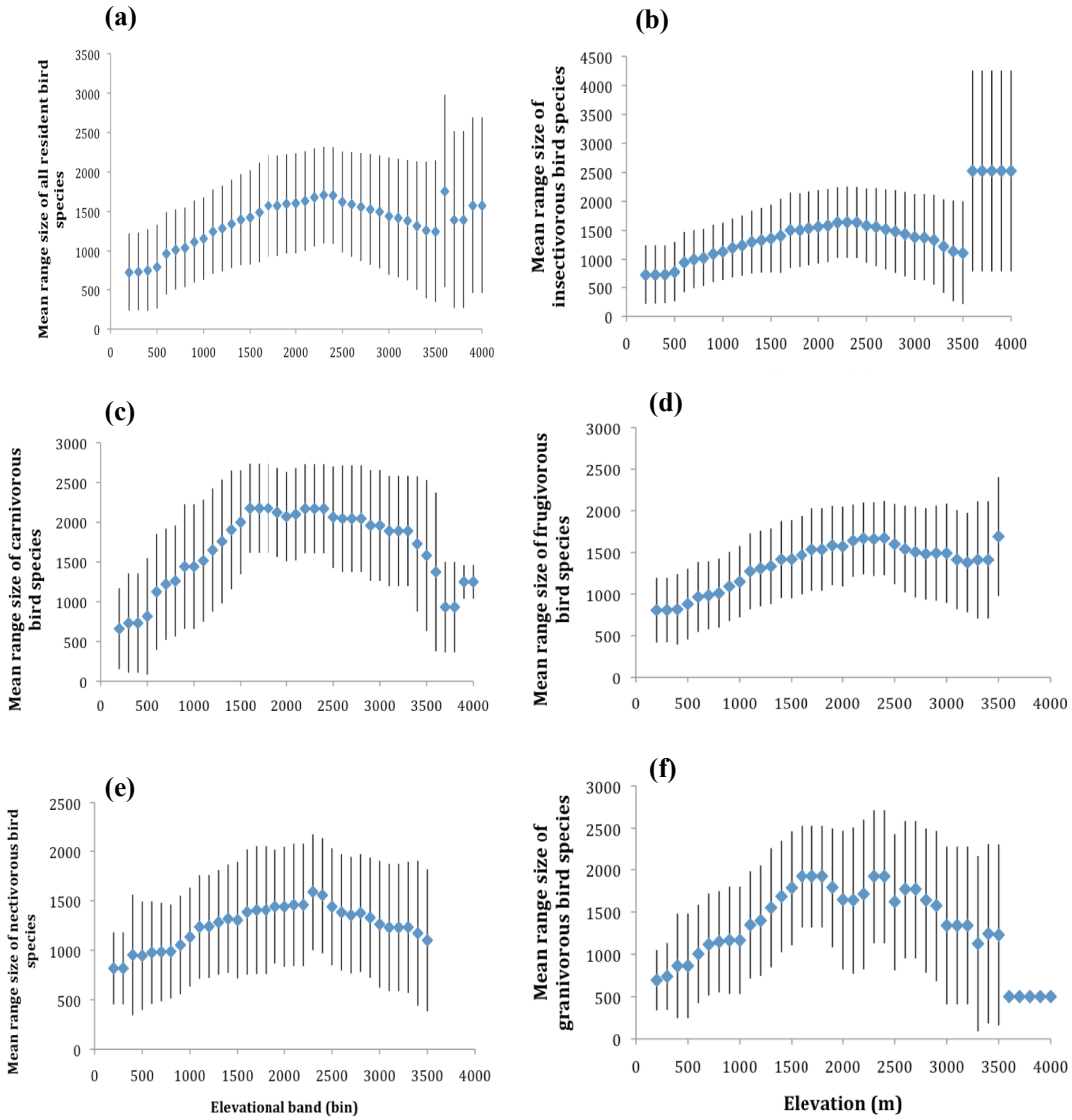


Figure 7.4 Steven's plots of the Rapoport effect in elevational ranges of birds occurring in 100m elevational bands in the Manu Biosphere Reserve, Peru. Lines around points are ± 1 standard deviation of the mean. (a) All resident bird species (Spearman's rho = 0.547, d.f. = 37, P = <0.001), and birds belonging to (b) insectivore (Spearman's rho = 0.711, d.f. = 37, P = <0.001), (c) carnivore (Spearman's rho =0.192, d.f. = 37, P = 0.241), (d) frugivore (Spearman's rho = 0.680, d.f. = 32, P = <0.001), (e) nectarivore (Spearman's rho = 0.463, d.f. = 32, P = 0.006), and (f) granivore foraging guilds (Spearman's rho = -0.087, d.f. = 37, P = 0.600).

Discussion

This study has shown that neotropical birds exhibit a range of species richness patterns with elevation and that these patterns are often differentially influenced by each of the environmental variables (including area) considered here. In total, three distinct patterns of species richness with elevation were recorded: declining species richness with elevation for all resident bird species and the frugivore guild; declining species richness with a mid-elevation plateau for birds belonging to carnivore and granivore guilds; and a bimodal pattern of species richness with peaks at high and low elevations separated by a mid-elevation plateau for birds belonging to the nectarivore guild. Investigating the relationship between species richness and elevation for all resident bird species revealed an overall decreasing pattern of richness with elevation. When all resident bird species and each of the foraging guilds were plotted separately, significant variation around the shape of this relationship were revealed for each gradient (Fig. 5). In the past, patterns of species richness with elevation were thought to ubiquitously show unimodal declines with elevation and any variation around this pattern such as peaks at mid elevations, plateaus, bi-modal patterns etc. were thought to reflect sampling artefacts (McCoy 1990, Wolda 1987). The observed relationship of overall decreasing species richness with elevation for all resident bird species and birds belonging to the insectivore and frugivore guilds is consistent with earlier studies recording similar monotonic declines in species richness with elevation (MacArthur et al. 1962, Terborgh 1977) and more recent work on bird diversity along an elevational gradient in the Andes of Colombia (Kattan and Franco 2004). However, simply plotting the relationship between species richness and elevation is not very informative for investigating underlying mechanisms shaping observed relationships. Early work

by (Rahbek 1997) and more recent meta-analyses on bird species richness with elevation (McCain 2009) show that a range of patterns are possible depending upon whether or not environmental variables and area are taken into account.

Variation in the recorded pattern of species richness such as the peak at low and high elevations for birds belonging to the nectarivore guild, and decreasing species richness with a mid-elevation plateau recorded for carnivore and granivore foraging guilds, is consistent with a more recent appreciation of elevational richness patterns. The overall pattern of declining species richness with elevation was driven primarily by insectivore and frugivore foraging guilds which had proportionally the most species recorded from along the elevational gradient (542 and 153 species respectively). The variation observed for each of the foraging guilds with elevation clearly shows that species richness patterns of each group is differentially influenced by each of the environmental variables and area investigated in this study.

Results from the general linear model show that area was the only consistent explanatory variable for estimating species richness across all resident species and for each of the foraging guilds. Lower elevations along mountain ranges tend to cover larger areas than higher elevations and the observed decline in species richness with elevation is likely to be driven largely by species-area effects (Rosenzweig M.L. 1995). Within the study region, area is greatest at low elevations around 200 m and decline steeply up to 500 m. Above 500 m area shows very gradual declines until ~3000 m where is shows a sharp increase. This pattern is unusual for mountains (where area often decline linearly) and may be partially responsible for the bimodal peak seen in species richness for birds belonging to the nectarivore guild (Fig. 5E).

The overall significance of area in explaining patterns of species richness is consistent with other datasets on birds which have shown area can explain as much as 60-93% of the variation in species richness with elevation (Rahbek 1997), and other work on the influence of area on mammals (McCain 2007b, Rickart 2001) and ants (Sanders 2002).

Visually investigating elevational patterns of species richness for all resident bird species and each of the foraging guilds when area was accounted for by plotting either species richness/area or species richness/log(area) against elevation, failed to produce the hump-shaped pattern other studies have reported when accounting for area in monotonic declining patterns of species (Rahbek 1995). The observed richness pattern with elevation using the species richness/area calibration resulted in slightly more smoothed linear declines, whilst plotting species richness/log(area) against elevation resulted in bimodal peaks similar to the pattern for nectarivores shown in Figure 7.3e. The results of slightly different species richness patterns with elevation for each of the different feeding ecologies is consistent with a recent appreciation of species richness patterns also being influence by biotic interactions in addition to abiotic features of the environment (Schemske 2002). Interestingly though, the result of NPP being an important predictor of frugivore species richness in univariate analysis, but no longer being important when area and other environmental variable are taken into consideration, suggests an overall greater importance of abiotic variable in shaping species richness patterns, at least for this guild. Overall results from the general linear models reveal that NPP and area are the key drivers of the observed relationships when all variable are taken into consideration (both were significant for all analyses except for frugivores, whose richness was only significantly explained by area).

Mid-domain effects

Spatial constraints on species ranges have received considerable attention in previous studies on elevational richness patterns and have strong empirical support (Colwell and Lees 2000, Colwell et al. 2004). Mid-domain null models predict that species should show a unimodal pattern of species richness due to the constraints of hard boundaries within a bounded domain forcing species ranges to overlap more toward the centre of their distribution. Patterns of empirical species richness with elevation for all resident bird species and each of the distinct foraging guilds except nectarivores, did not follow null model distributions i.e. unimodal patterns. Nectarivores were the only forging guild whose pattern of species richness with elevation fitted MDE null predictions ($R^2 = 0.11$, $F = 4.811$ $P = 0.035$).

The lack of an MDE fit (average $R^2 = 0.04$) for all species richness gradients investigated here except for the nectarivore foraging guild is consistent with other elevational studies of species richness showing no or weak fits to MDE for birds (McCain 2009), non-flying small mammals (McCain 2005) and bats (McCain 2007b). Although MDE have been reported from a range of other studies from a diverse array of organisms including ferns (Kluge et al. 2006) and moths (Brehm et al. 2007); for the majority of taxa examined so far, elevational patterns of species richness are not consistent with MDE predictions (Dunn et al. 2007). The variation in MDE fits recorded here between nectarivores and all other bird species may stem for variation around the species area relationships between each of these guilds. Nectarivores were the only guild to show a bimodal pattern (Figure 7.3e) with elevation and were therefore the only group that showed a corresponding increase in species richness that approximated the pattern seen for the amount of available area along the gradient (Fig.

4D). Future tests of MDE fits when area is accounted for will test the potential for area to influence observed patterns.

Rapoport's Rule

Rapoport's rule of increasing range size with latitude has been used to explain the monotonic decline in species richness with latitude and was extended by Stevens (1989) to include monotonic declines recorded along elevational gradients. In the case of elevational gradients, the rule states that with increasing elevation species show increasing range size, as a response to the breadth of climatic conditions experienced toward higher elevations. Support for the rule is mixed with numerous examples of strong Rapoport effects for a wide range of organisms along elevational gradients including trees, insects, mammals, birds, reptiles and amphibians (Brehm et al. 2007, Sanders 2002, Stevens 1992) and other studies along latitudinal gradients showing that its applicability is at best equivocal (Gaston et al. 1998). Results from this study confirm that Rapoport's rule does not have general applicability across elevational gradients, but that it's potential significance needs to be investigated on a case-by-case bases. In this study I found support for Rapoport's rule of increasing range amplitude with elevation for all resident bird species, and each of the insectivore, frugivore and nectarivore bird guilds; but no support for the same effects in each of the carnivore or granivore bird guilds. Interestingly, at elevations up to ~2000 m strong Rapoport effects were apparent for all the gradients studied. Above ~2000 m patterns were declining, with reduced range sizes at higher elevation possibly reflecting the predominance of high elevation specialist bird species in shaping observed relationships.

Conclusions

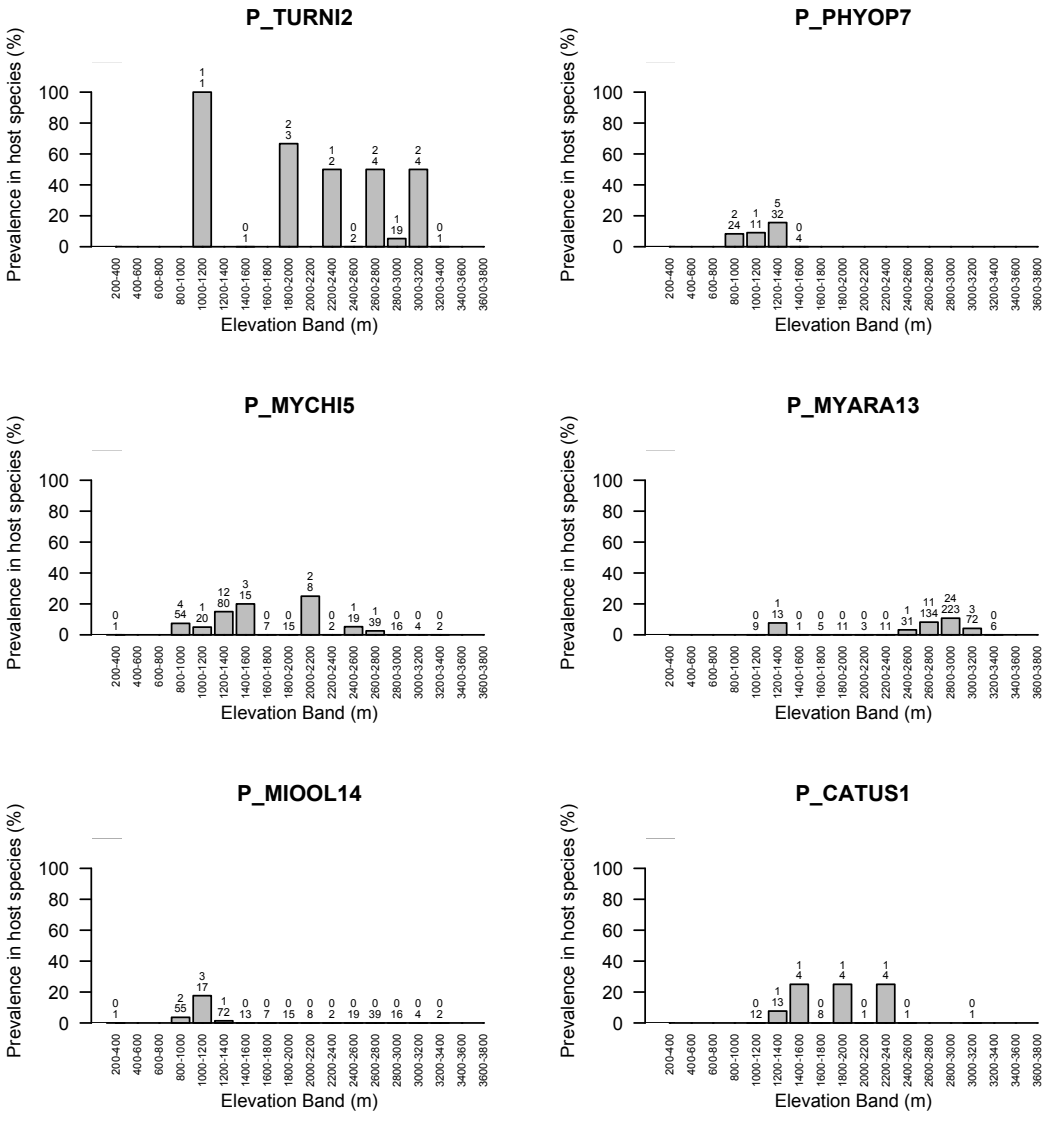
These results suggest that both environmental variables and area are important for shaping species richness patterns with elevation and that the effects of these are unique for individual foraging guilds. It is apparent that a range of fine scale measurements of environmental variability and accurate area estimates are required to correctly record species richness patterns with elevation before possible mechanisms and explanatory variables underlying observed relationships can be conclusively determined. Limitations of this study were primarily associated with not being able to account for the sampling effort put into each of the elevational zones and having to use interpolation rather than actual records to confirm species distribution ranges. It is important to note that the close association between some of the variables with elevation, and problems associated with interpolation, may result in confounded results when trying to tease apart their separate effects. However, the limitations of interpolation for estimating environmental vary depending on the variable being estimated. For instance, interpolation of temperature variables with elevation is justified as temperature declines consistently with elevation along mountains relative to the adiabatic lapse rate, and as such missing values can be interpolated with a high level of confidence (Figure 7.1a). For variables such as precipitation and NPP, relationships with elevation are much more variable, and as such, estimates from each elevation band are required to rigorously test the overall importance of these variables in shaping observed relationships. In summary, both environmental variables and area are needed to predict species richness patterns of birds with elevation. Different foraging guilds of birds showed varying patterns of richness with elevation and as such some knowledge of ecology is required to accurately predict patterns of species richness with elevation for neotropical birds. This study indicates that although mid-

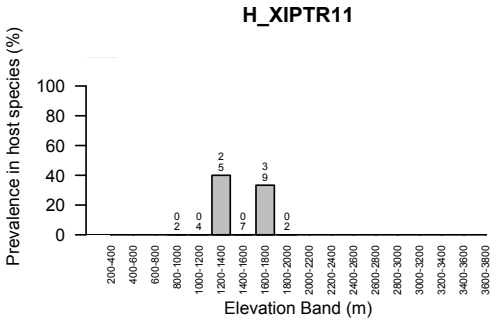
domain and Rapoport effects do not have general applicability for birds along the elevation gradient, their potential influence in shaping observed species richness needs to be investigated on an individual bases. Specifically, nectarivorous bird were the only guild that showed a significant fit to mid-domain null predictions. Rapport's rule of increasing range amplitude with elevation was supported for all resident bird species and each of the insectivore, frugivore and nectarivore bird guilds, but not for carnivores and granivores. Environmental variables, NPP and area explained the greatest proportion of the observed variation between species richness and elevation, but as the range of richness patterns shows, diversity is responding to biological factors and not simply the abiotic variables recorded here. More detailed analyses are needed to discern the underlying mechanisms shaping observed relationships and to evaluate the extent to which patterns are driven by ecological, environmental, evolutionary, historical or other processes.

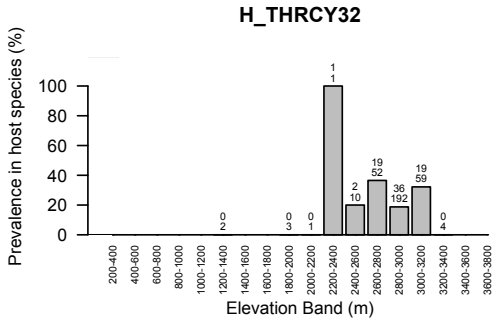
Appendix 2

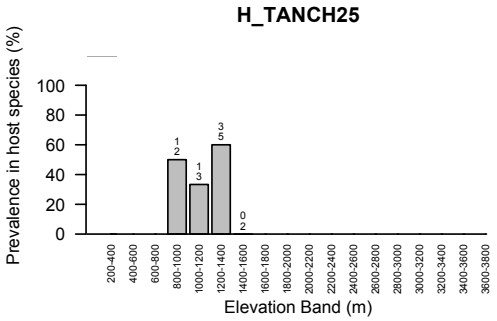
Supplementary figures and tables

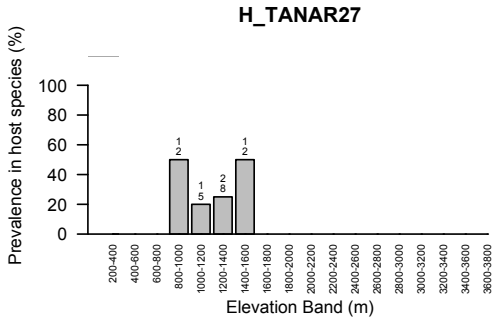
Figure 4.1 Prevalence of parasite lineages recorded a minimum of 5 times in all records of their resident hosts. These were used in the analyses testing if lineages were more prevalent in the outer vs. innner proportion of their elevation ranges. Parasite range limits are defined as the maximum and minimum records at which the lineage was recovered from their host species. Figures also show the number of parasite records (top number) and host species captures (bottom number) above each prevalence bar to help visualise the extent to which a lineage's distribution mirrored that of its hosts, relative to sample size.

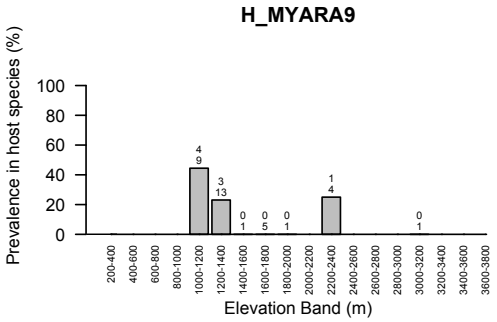


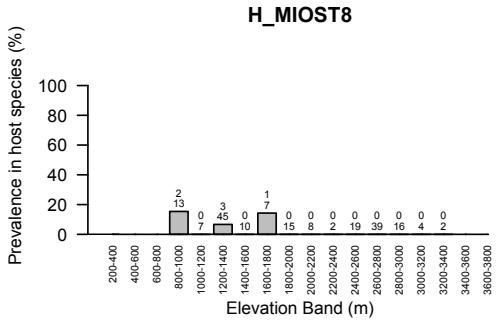

H_XIPTR11
Prevalence in host species (%)
Elevation Band (m)

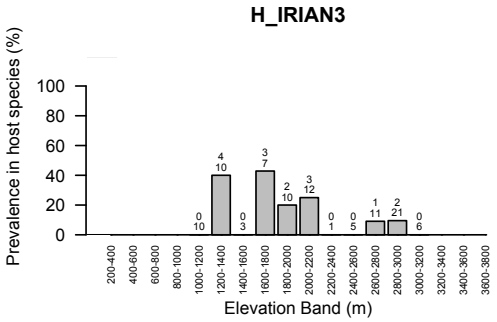

H_THRCY32
Prevalence in host species (%)
Elevation Band (m)

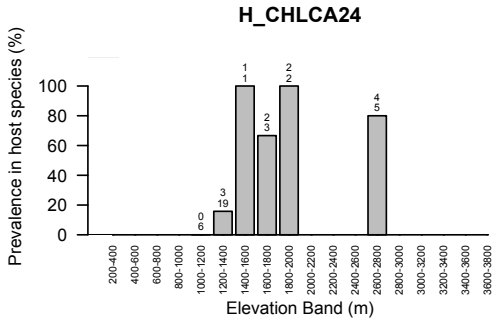

H_TANCH25
Prevalence in host species (%)
Elevation Band (m)

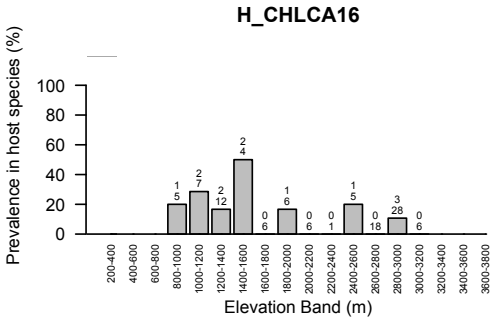

H_TANAR27
Prevalence in host species (%)
Elevation Band (m)

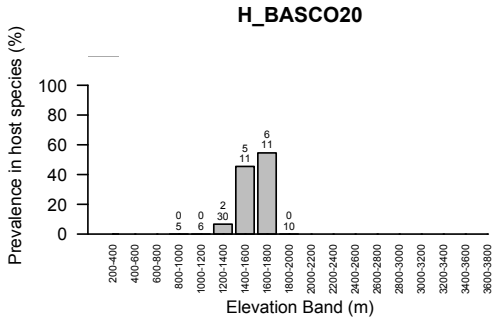

H_MYARA9
Prevalence in host species (%)
Elevation Band (m)

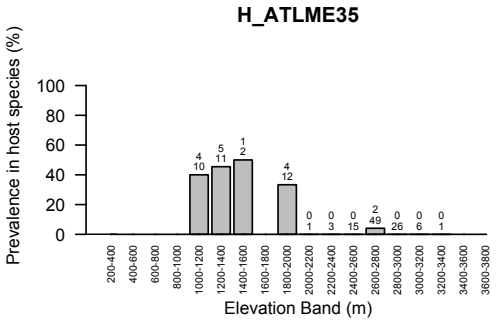

H_MIOST8
Prevalence in host species (%)
Elevation Band (m)

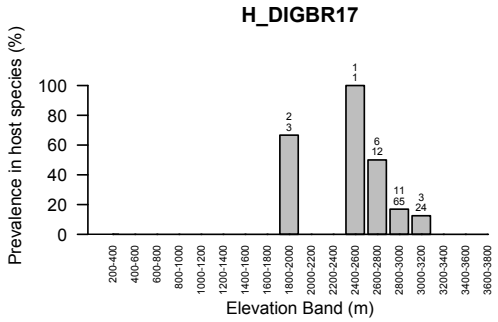

H_IRIAN3
Prevalence in host species (%)
Elevation Band (m)

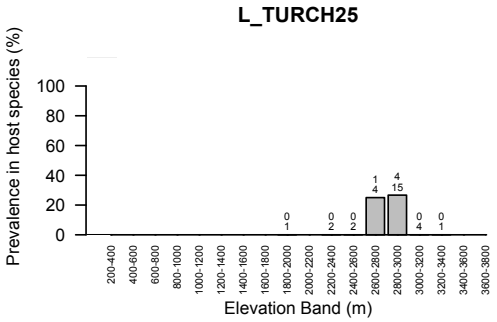

H_CHLCA24
Prevalence in host species (%)
Elevation Band (m)

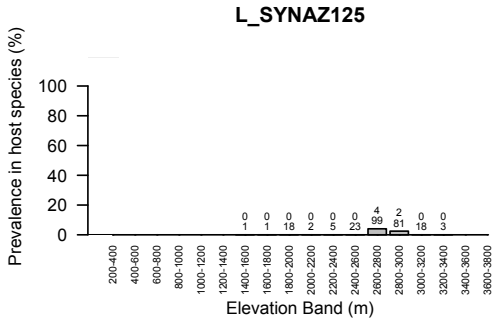

H_CHLCA16
Prevalence in host species (%)
Elevation Band (m)

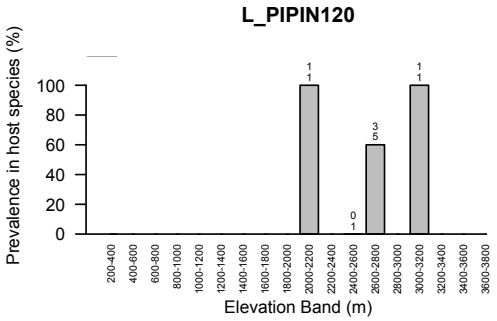

H_BASCO20
Prevalence in host species (%)
Elevation Band (m)

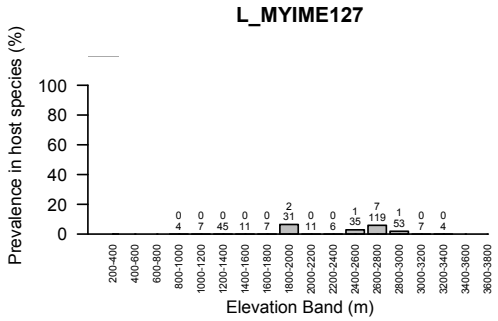

H_ATLME35
Prevalence in host species (%)
Elevation Band (m)

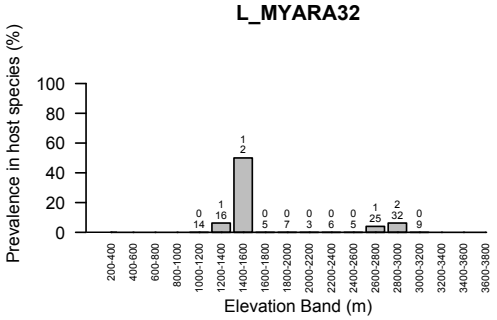

H_DIGBR17
Prevalence in host species (%)
Elevation Band (m)

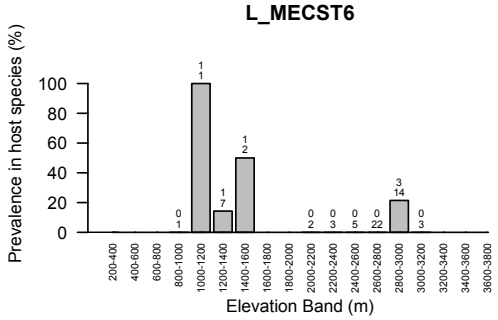

L_TURCH25
Prevalence in host species (%)
Elevation Band (m)

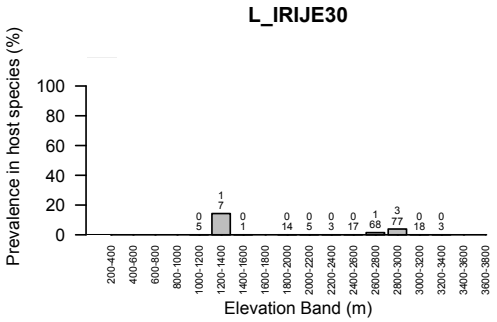

L_SYNAZ125
Prevalence in host species (%)
Elevation Band (m)

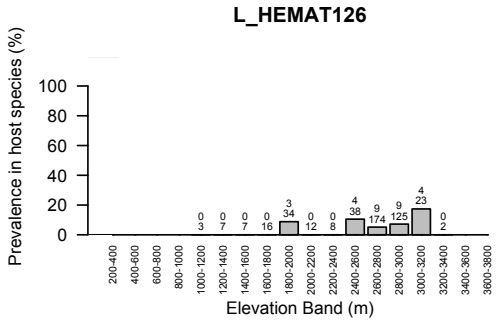

L_PIPIN120
Prevalence in host species (%)
Elevation Band (m)

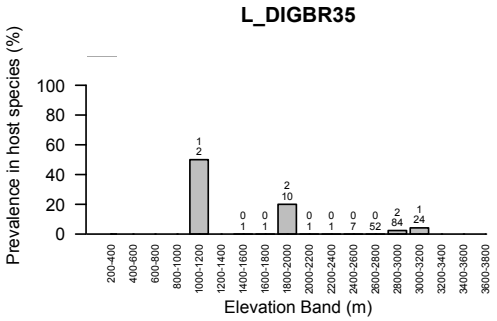

L_MYIME127
Prevalence in host species (%)
Elevation Band (m)

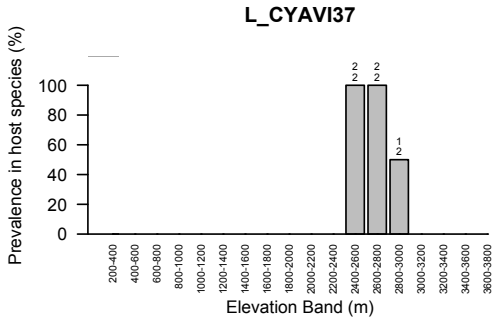

L_MYARA32
Prevalence in host species (%)
Elevation Band (m)

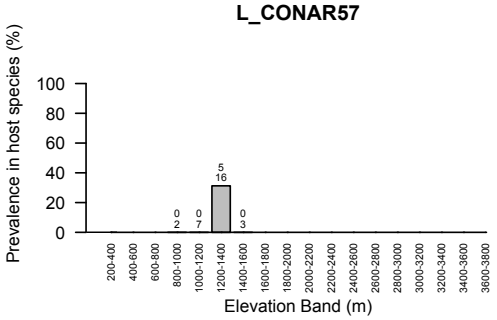

L_MECST6
Prevalence in host species (%)
Elevation Band (m)

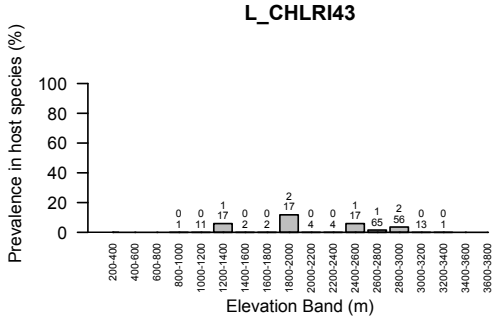

L_IRIJE30
Prevalence in host species (%)
Elevation Band (m)

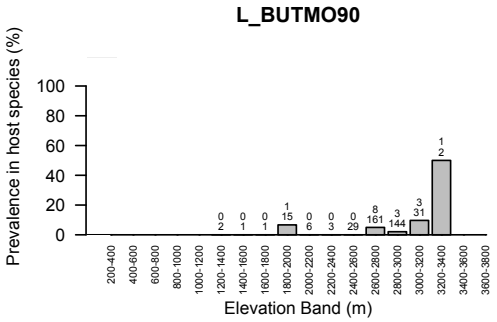

L_HEMAT126
Prevalence in host species (%)
Elevation Band (m)

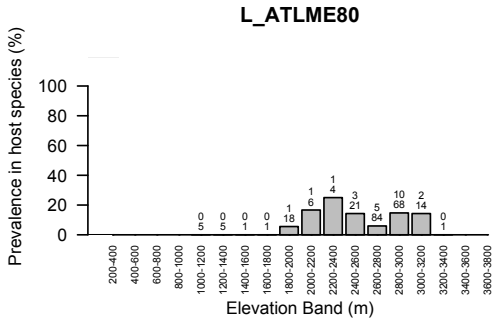

L_DIGBR35
Prevalence in host species (%)
Elevation Band (m)

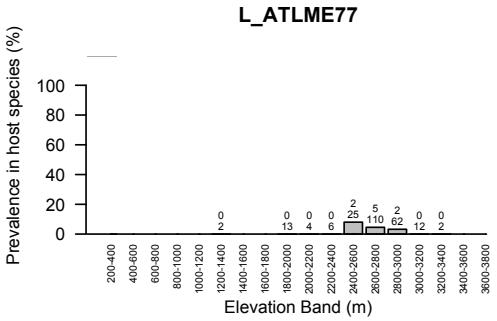

L_CYAVI37
Prevalence in host species (%)
Elevation Band (m)

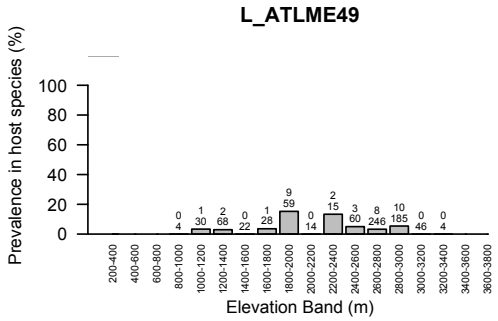

L_CONAR57
Prevalence in host species (%)
Elevation Band (m)

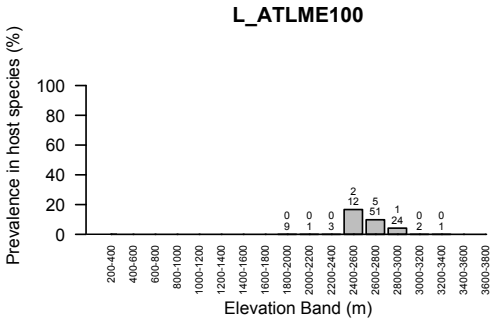

L_CHLRI43
Prevalence in host species (%)
Elevation Band (m)

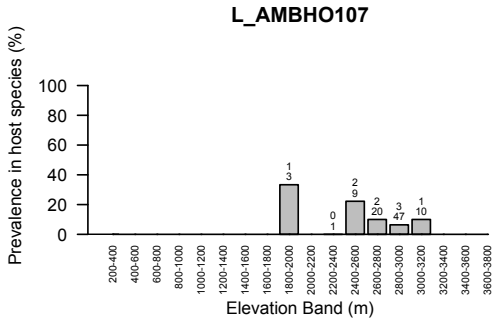

L_BUTMO90
Prevalence in host species (%)
Elevation Band (m)

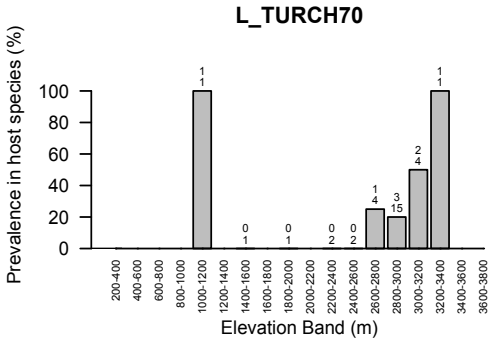

L_ATLME80
Prevalence in host species (%)
Elevation Band (m)


L_ATLME77
Prevalence in host species (%)
Elevation Band (m)


L_ATLME49
Prevalence in host species (%)
Elevation Band (m)


L_ATLME100
Prevalence in host species (%)
Elevation Band (m)


L_AMBHO107
Prevalence in host species (%)
Elevation Band (m)


L_TURCH70
Prevalence in host species (%)
Elevation Band (m)

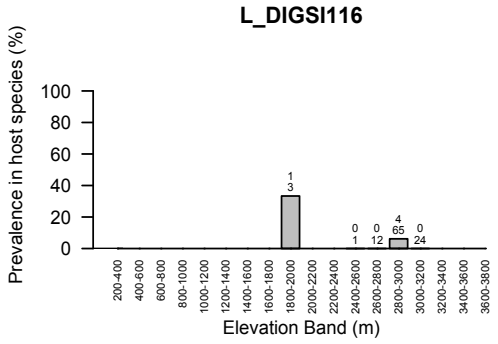

L_DIGSI116
Prevalence in host species (%)
Elevation Band (m)

Table 5.3 Parsite lineages recorded 5 or more times and their prevalence in resident bird speices. These host-parasite associations were used to test if paraiste lineages were abundace-centred within their own elevation ranges.

Lineage	Total lineage records	Family	Resident bird species	No. infections in host sp.	Total sp. captures	% Infected
P_CATUS1	6	Turdidae	*Entomodestes leucotis*	1	16	6
			Myadestes ralloides	3	32	9
P_MIOOL14	6	Tyrannidae	*Mionectes oleagineus*	1	26	4
			Mionectes striaticollis	1	178	1
			Mionectes olivaceus	1	50	2
			Ramphocelus carbo	1	8	13
		Thamnophilidae	*Epinecrophylla ornata*	1	7	14
			Thamnophilus palliatus	1	1	100
P_MYARA13	40	Turdidae	*Myadestes ralloides*	1	32	3
			Turdus fuscater	5	10	50
		Thraupidae	*Anisognathus igniventris*	1	31	3
			Diglossa cyanea	8	60	13
			Diglossa brunneiventris	6	100	6
			Buthraupis montana	4	15	27
			Diglossa mystacalis	1	23	4
			Iridosornis jelskii	3	28	11
			Thraupis cyanocephala	1	45	2
		Parulidae	*Basileuterus luteoviridis*	1	43	2
		Formicariidae	*Grallaricula ferrugineipectus*	2	7	29
		Emberizidae	*Atlapetes melanolaemus*	4	93	4
			Zonotrichia capensis	1	14	7
			Arremon torquatus	1	10	10
		Cotingidae	*Pipreola intermedia*	1	8	13
P_MYCHI5	24	Tyrannidae	*Phylloscartes ophthalmicus*	2	3	67
			Leptopogon superciliaris	5	18	28
			Myiodynastes chrysocephalus	1	1	100
			Mionectes olivaceus	4	50	8
			Mionectes striaticollis	8	178	4
			Mionectes oleagineus	2	26	8
			Lophotriccus pileatus	2	6	33
P_PHYOP7	8	Tyrannidae	*Mionectes olivaceus*	4	50	8
			Leptopogon superciliaris	3	18	17
			Phylloscartes ophthalmicus	1	3	33
P_TURNI2	12	Turdidae	*Turdus chiguanco*	2	9	22
			Turdus fuscater	2	10	20
			Turdus leucops	1	2	50
			Turdus serranus	3	10	30
		Thraupidae	*Thraupis bonariensis*	1	6	17
H_ATLME35	16	Thraupidae	*Chlorospingus flavigularis*	9	19	47
			Chlorospingus parvirostris	1	5	20
		Emberizidae	*Atlapetes melanolaemus*	4	93	4
			Zonotrichia capensis	1	14	7
		Thraupidae	*Chlorospingus ophthalmicus*	1	5	20
H_BASCO20	13	Parulidae	*Basileuterus coronatus*	8	33	24
			Basileuterus bivittatus	2	24	8
			Basileuterus tristriatus	2	7	29
		Thraupidae	*Trichothraupis melanops*	1	9	11
H_CHLCA16	12	Thraupidae	*Tangara nigroviridis*	1	7	14
			Tangara arthus	1	9	11
			Trichothraupis melanops	3	9	33
			Hemispingus melanotis	1	15	7
			Chlorothraupis carmioli	1	5	20

			Thraupis cyanocephala	4	45	9
			Chlorornis riefferii	1	14	7
H_CHLCA24	12	Thraupidae	*Tangara nigroviridis*	5	7	71
			Tangara xanthocephala	1	1	100
			Chlorochrysa calliparaea	1	16	6
			Tangara chilensis	1	6	17
			Tangara vassorii	4	6	67
H_DIGBR17	23	Thraupidae	*Diglossa brunneiventris*	20	100	20
			Diglossa sittoides	3	5	60
H_IRIAN3	15	Thraupidae	*Hemispingus melanotis*	6	15	40
			Anisognathus somptuosus	1	3	33
			Iridosornis analis	2	19	11
			Trichothraupis melanops	2	9	22
			Thraupis cyanocephala	3	45	7
		Emberizidae	*Arremon brunneinucha*	1	5	20
H_MIOST8	6	Tyrannidae	*Mionectes striaticollis*	4	178	2
			Leptopogon amaurocephalus	1	3	33
			Myiotriccus ornatus	1	6	17
H_MYARA9	8	Turdidae	*Myadestes ralloides*	7	32	22
			Catharus dryas	1	2	50
H_TANAR27	5	Thraupidae	*Tangara schrankii*	1	2	50
			Tangara chilensis	3	6	50
			Tangara arthus	1	9	11
H_TANCH25	5	Thraupidae	*Tangara schrankii*	1	2	50
			Tangara arthus	3	9	33
			Tangara chrysotis	1	1	100
H_THRCY32	77	Thraupidae	*Diglossa cyanea*	6	60	10
			Chlorornis riefferii	9	14	64
			Thraupis cyanocephala	17	45	38
			Buthraupis montana	8	15	53
			Anisognathus igniventris	24	31	77
			Diglossa brunneiventris	6	100	6
			Diglossa mystacalis	4	23	17
			Thraupis bonariensis	1	6	17
			Iridosornis jelskii	1	28	4
			Delothraupis castaneoventris	1	2	50
H_XIPTR11	5	Furnariidae	*Simoxenops ucayalae*	1	3	33
			Anabacerthia striaticollis	1	11	9
			Syndactyla rufosuperciliata	2	5	40
			Xiphorhynchus triangularis	1	10	10
L_AMBHO10 7	9	Thraupidae	*Thraupis cyanocephala*	4	45	9
			Chlorospingus ophthalmicus	1	5	20
			Anisognathus igniventris	2	31	6
		Icteridae	*Amblycercus holosericeus*	2	9	22
L_ATLME10 0	8	Emberizidae	*Atlapetes melanolaemus*	6	93	6
			Arremon torquatus	2	10	20
L_ATLME49	36	Tyrannidae	*Mionectes striaticollis*	1	178	1
			Pyrrhomyias cinnamomeus	1	3	33
			Ochthoeca pulchella	1	54	2
		Turdidae	*Entomodestes leucotis*	1	16	6
			Myadestes ralloides	1	32	3
		Thraupidae	*Tangara vassorii*	2	6	33
			Diglossa sittoides	2	5	40
			Chlorospingus ophthalmicus	2	5	40
			Diglossa cyanea	4	60	7
			Diglossa brunneiventris	2	100	2
			Buthraupis montana	1	15	7
			Delothraupis castaneoventris	1	2	50
		Parulidae	*Myioborus miniatus*	1	11	9
			Basileuterus coronatus	1	33	3
			Basileuterus luteoviridis	1	43	2

			Myioborus melanocephalus	1	15	7
			Basileuterus signatus	1	31	3
		Furnariidae	*Synallaxis azarae*	7	75	9
		Fringillidae	*Carduelis olivacea*	1	2	50
			Carduelis magellanica	1	2	50
		Emberizidae	*Atlapetes melanolaemus*	3	93	3
L_ATLME77	9	Thraupidae	*Diglossa cyanea*	3	60	5
		Parulidae	*Basileuterus luteoviridis*	3	43	7
			Basileuterus signatus	1	31	3
		Icteridae	*Amblycercus holosericeus*	1	9	11
		Emberizidae	*Atlapetes melanolaemus*	1	93	1
L_ATLME80	23	Thraupidae	*Cnemoscopus rubrirostris*	1	2	50
			Anisognathus somptuosus	1	3	33
			Iridosornis analis	1	19	5
			Thraupis cyanocephala	12	45	27
			Hemispingus atropileus	2	18	11
			Buthraupis montana	1	15	7
			Tangara vassorii	1	6	17
		Icteridae	*Amblycercus holosericeus*	1	9	11
		Furnariidae	*Margarornis squamiger*	1	18	6
		Emberizidae	*Atlapetes melanolaemus*	2	93	2
L_BUTMO90	16	Tyrannidae	*Ochthoeca pulchella*	1	54	2
		Troglodytidae	*Cinnycerthia fulva*	1	32	3
		Thraupidae	*Chlorospingus ophthalmicus*	1	5	20
			Iridosornis jelskii	2	28	7
			Thraupis cyanocephala	1	45	2
			Buthraupis montana	3	15	20
			Diglossa cyanea	1	60	2
			Anisognathus igniventris	1	31	3
		Parulidae	*Basileuterus signatus*	1	31	3
		Icteridae	*Amblycercus holosericeus*	1	9	11
		Furnariidae	*Synallaxis azarae*	1	75	1
		Emberizidae	*Arremon torquatus*	2	10	20
L_CHLRI43	7	Thraupidae	*Iridosornis analis*	1	19	5
			Tangara nigroviridis	1	7	14
			Chlorornis riefferii	1	14	7
			Buthraupis montana	1	15	7
			Thraupis cyanocephala	1	45	2
		Furnariidae	*Thripadectes melanorhynchus*	1	17	6
		Emberizidae	*Atlapetes melanolaemus*	1	93	1
L_CONAR57	6	Furnariidae	*Thripadectes melanorhynchus*	1	17	6
		Conopophagidae	*Conopophaga ardesiaca*	4	11	36
L_CYAVI37	5	Corvidae	*Cyanolyca viridicyanus*	5	6	83
L_DIGBR35	6	Thraupidae	*Thraupis bonariensis*	1	6	17
			Diglossa brunneiventris	3	100	3
		Furnariidae	*Synallaxis azarae*	1	75	1
		Fringillidae	*Carduelis olivacea*	1	2	50
L_DIGSI116	5	Thraupidae	*Diglossa sittoides*	1	5	20
			Diglossa brunneiventris	4	100	4
L_HEMAT126	29	Tyrannidae	*Myiotheretes fuscorufus*	1	1	100
		Troglodytidae	*Cinnycerthia fulva*	1	32	3
		Thraupidae	*Hemispingus melanotis*	1	15	7
			Hemispingus atropileus	2	18	11
			Thraupis cyanocephala	4	45	9
			Hemispingus trifasciatus	1	2	50
			Anisognathus igniventris	1	31	3
			Iridosornis jelskii	1	28	4
		Parulidae	*Basileuterus signatus*	5	31	16
			Basileuterus coronatus	1	33	3
			Basileuterus luteoviridis	4	43	9

		Furnariidae	*Synallaxis azarae*	1	75	1
		Emberizidae	*Haplospiza rustica*	1	2	50
			Atlapetes melanolaemus	5	93	5
L_IRIJE30	5	Thraupidae	*Iridosornis analis*	1	19	5
			Hemispingus atropileus	1	18	6
			Iridosornis jelskii	1	28	4
			Diglossa cyanea	1	60	2
		Emberizidae	*Atlapetes melanolaemus*	1	93	1
L_MECST6	6	Tyrannidae	*Mecocerculus stictopterus*	1	9	11
			Ochthoeca cinnamomeiventris	1	8	13
		Troglodytidae	*Cinnycerthia fulva*	1	32	3
		Conopophagidae	*Conopophaga ardesiaca*	3	11	27
L_MYARA32	5	Turdidae	*Myadestes ralloides*	1	32	3
		Troglodytidae	*Troglodytes solstitialis*	1	18	6
		Thraupidae	*Iridosornis analis*	1	19	5
			Catamblyrhynchus diadema	1	10	10
			Thraupis cyanocephala	1	45	2
L_MYIME127	11	Tyrannidae	*Mionectes striaticollis*	1	178	1
			Hemispingus frontalis	1	4	25
			Hemispingus atropileus	1	18	6
			Dubusia taeniata	1	1	100
		Parulidae	*Myioborus melanocephalus*	2	15	13
			Basileuterus signatus	1	31	3
		Emberizidae	*Atlapetes melanolaemus*	4	93	4
L_PIPIN120	5	Cotingidae	*Pipreola intermedia*	5	8	63
L_SYNAZ125	6	Thraupidae	*Buthraupis montana*	1	15	7
			Thraupis cyanocephala	1	45	2
			Diglossa mystacalis	1	23	4
		Furnariidae	*Synallaxis azarae*	1	75	1
		Emberizidae	*Atlapetes melanolaemus*	2	93	2
L_TURCH25	5	Turdidae	*Turdus serranus*	1	10	10
			Turdus fuscater	1	10	10
			Turdus chiguanco	3	9	33
L_TURCH70	8	Turdidae	*Turdus leucops*	1	2	50
			Turdus serranus	2	10	20
			Turdus fuscater	3	10	30
			Turdus chiguanco	2	9	22

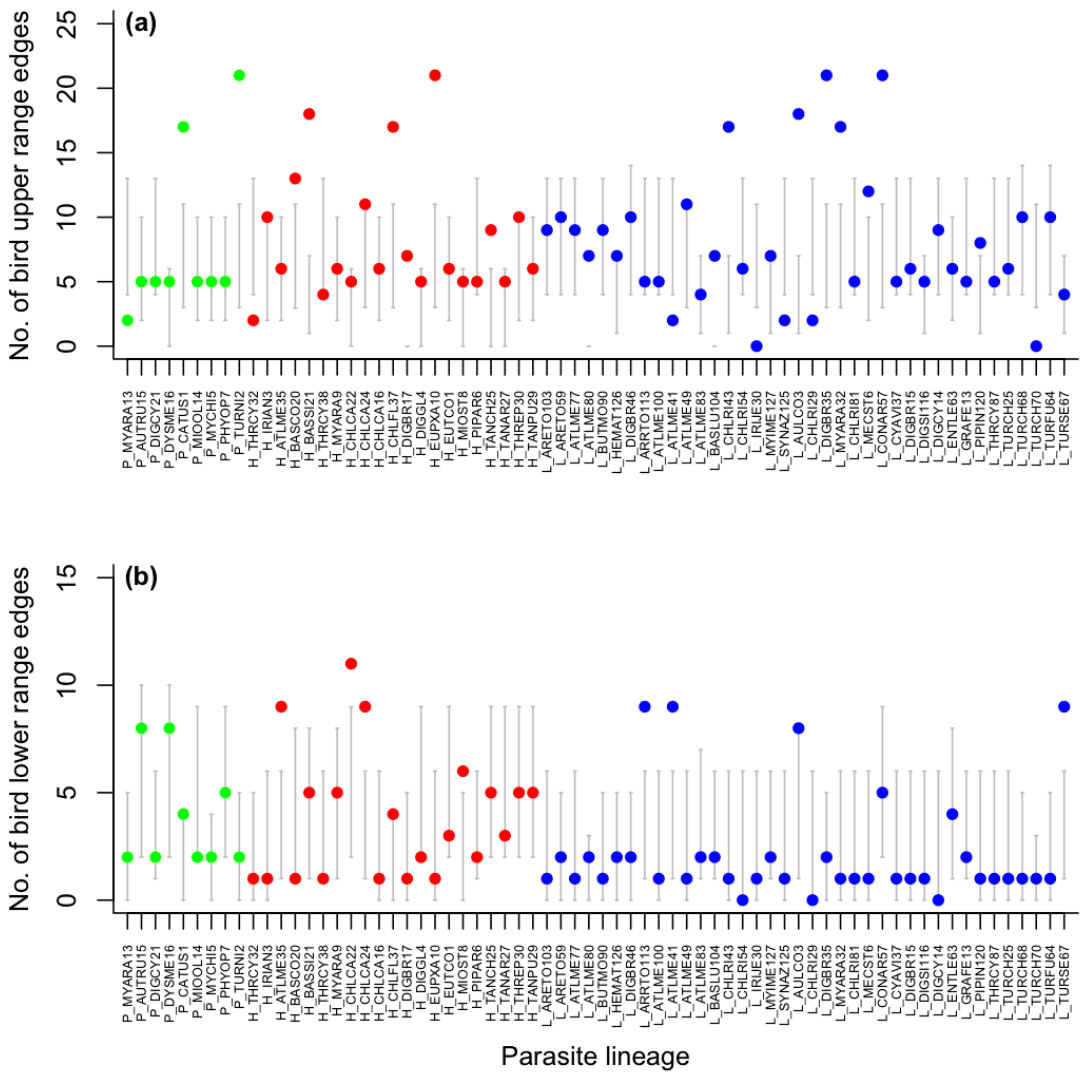


Figure 5.1 The number of empirical (filled circles) and expected (95% CI vertical bars) bird species' ranges abutting parasite lineage ranges. Circles falling outside the estimated range of expected values show parasite lineages that abutted significantly more (above) or fewer (below) bird species' than expected by change. Results are shown for (a) upper range limiting parasite lineages and (b) lower range limiting parasite lineages and for each of the three major genera (Plasmodium [green], Heamoproteus [red], and Leucocytozoan [blue]). Each parasite range boundary was defined as 100 m into the 95% confidence intervals of all recorded infections.

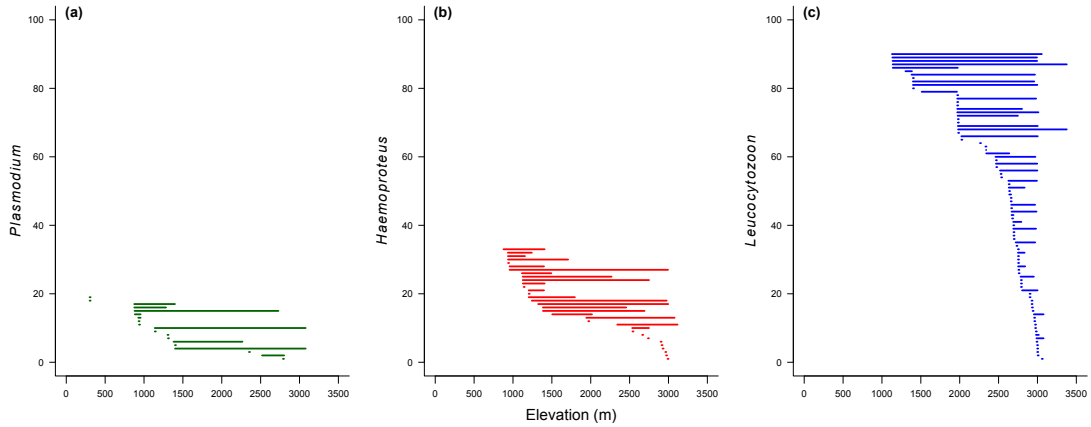


Figure 5.2 The elevation range of all parasite lineages recorded form resident birds along the elevation transect. Green lines represent *Plasmodium* elevation ranges (a), red *Haemoproteus* ranges (b), and blue *Leucocytozoon* ranges (c).

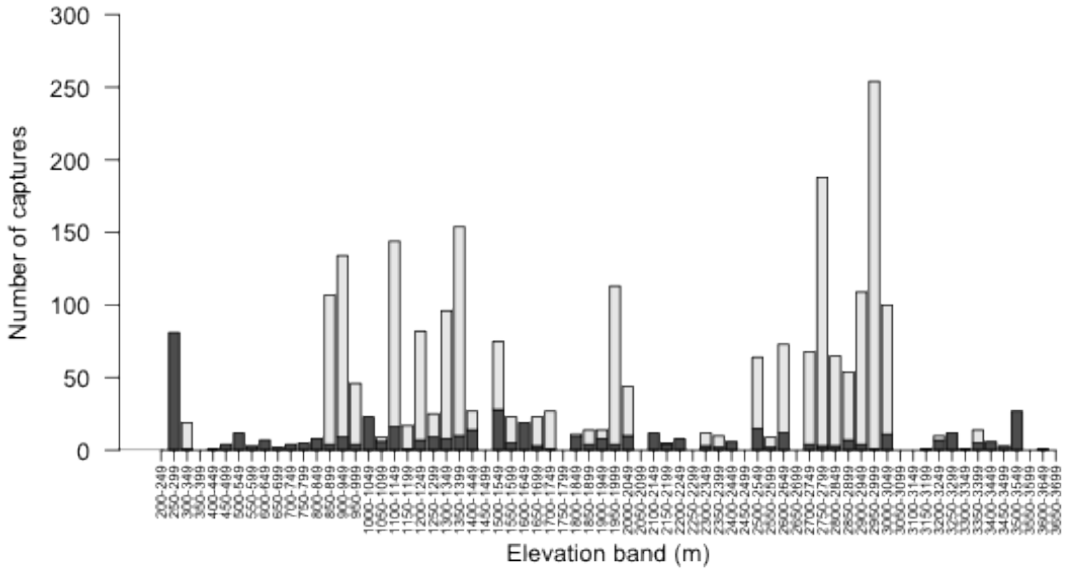


Figure 5.3 The number of bird captures (light bars) and species turnover (dark bars) in 50 m bands along the elevation transect.

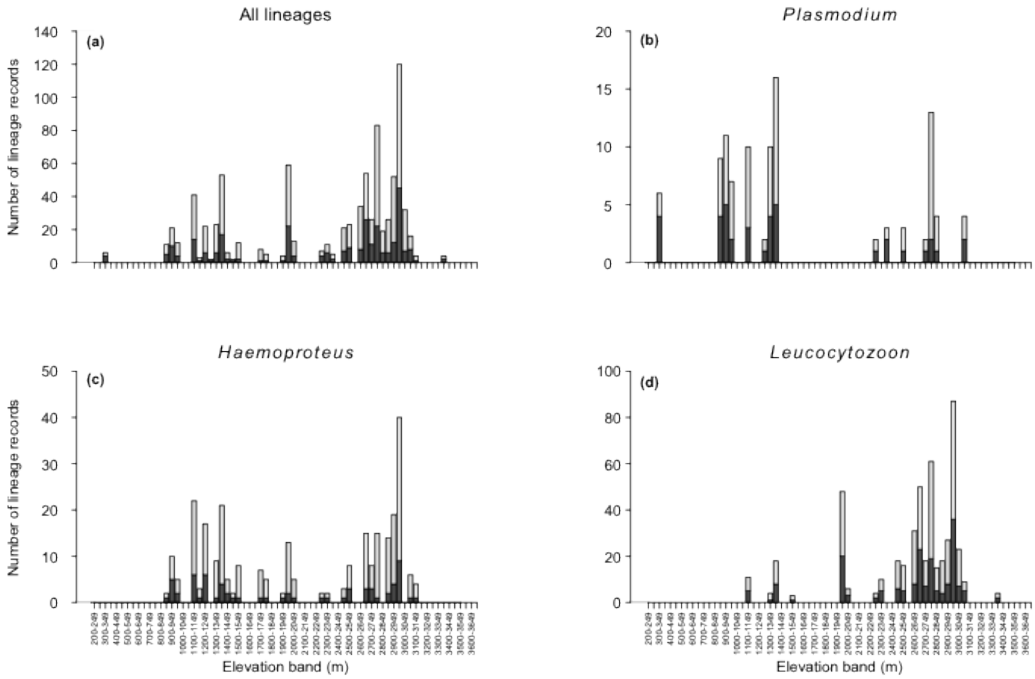


Figure 5.4 The number of infected birds in 50 m wide elevation bands and the turnover in parasite lineages for (a) Overall infections (any of the three parasite genera), (b) *Plasmodium* infections, (c) *Haemoproteus* infections, and (d) *Leucocytozoon* infections.

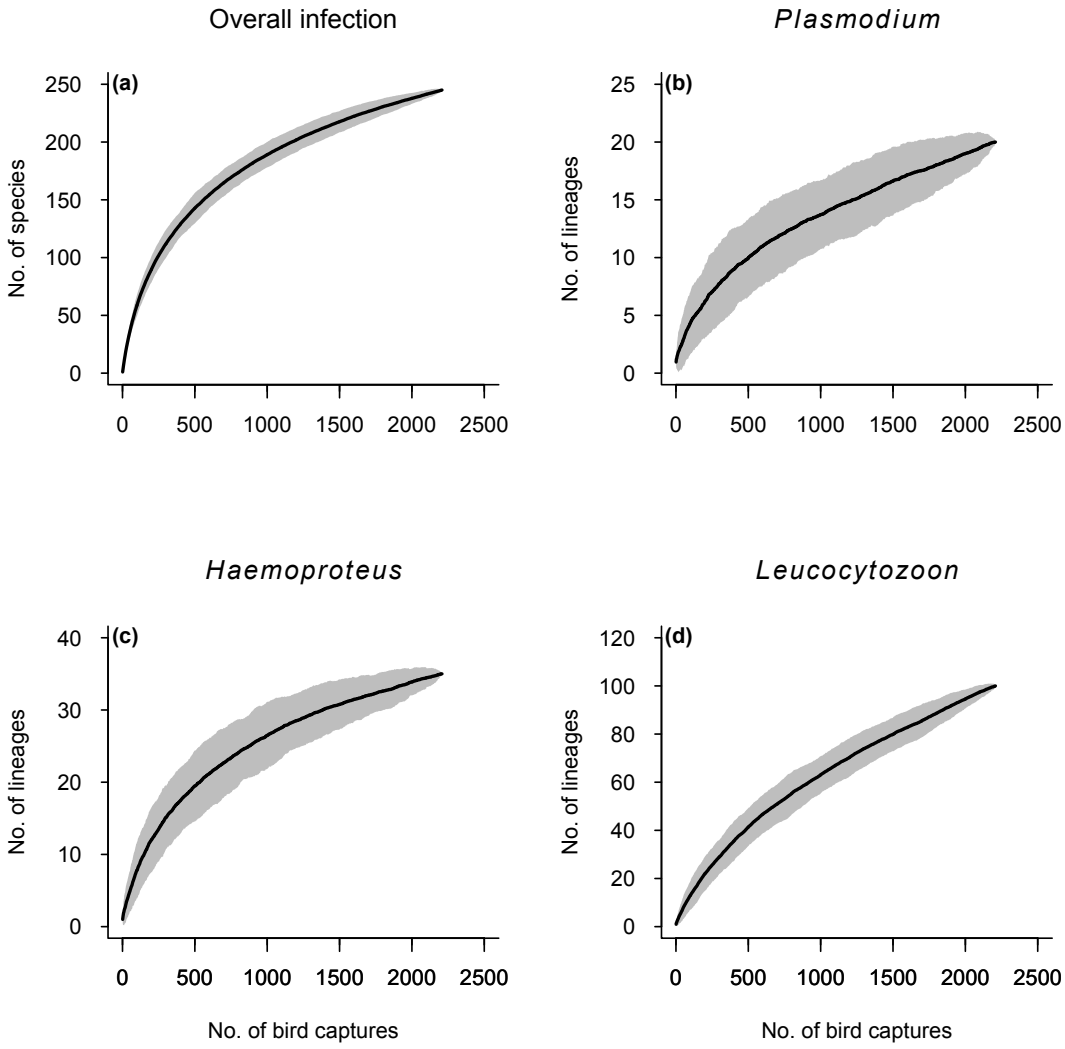


Figure 5.5 Rarefaction estimates of species richness for (a) birds, and for lineages of each of the three haemosporidian parasite genera recorded from these: (b) *Plasmodium* lineages, (c) *Haemoproteus* lineages and (d) *Leucocytozoon* lineages. Grey shading represents the measure of variance around the estimated richness.

Table 5.1 Species richness and species diversity indices (± se) (Chao, 1st order Jackknife, 2nd order Jackknife and Bootstrap) of 2188 captured birds and the parasite lineages recorded from these along the elevation transect.

	Species	Chao ± se	Jackknife1 ± se	Jackknife2	Bootstrap ± se
Bird species	245	298.44 ± 17.09	311.97 ± 8.18	336.97	276.93 ± 4.61
Plasmodium sp.	20	45.00 ± 24.24	30.00 ± 3.16	37.99	23.99 ± 1.61
Haemoproteus sp.	35	53.00 ± 14.39	46.99 ± 3.46	54.99	40.24 ± 1.88
Leucocytozoon sp.	100	268.20 ± 70.35	157.97 ± 7.61	205.93	123.28 ± 3.86

Table 5.2 Number of range limiting lineages when infection zones are classified as 100 m into parasite lineage maximum and minimum recorded elevations.

	100m into parasite lineage maximum and minimum elevation CI's											
Genus	Lineage	Low CI	Low limit	Mean elevation	Up CI	Up limit	Infected birds	Infected bird spp.	Infected low limited bird spp.	Low limited bird spp.	Infected up limited bird spp.	Up limit bird spp.
Plasmodium	P_MYARA13	2613	1399	2843	3004	3079	40	15	0	1	0	11
	P_ARRTA9	300	300	300	300	300	1	1	1	82	0	0
	P_ARRTO12	2787	2787	2787	2787	2787	1	1	0	2	0	9
	P_AULPR10	936	936	936	936	936	1	1	0	9	0	5
	P_AUTRU15	886	882	919	952	956	2	2	0	8	0	5
	P_DIGCY21	2532	2518	2660	2788	2802	2	2	0	0	0	5
	P_DYSME16	928	926	944	956	956	4	1	0	8	0	5
	P_ENTLE4	1139	1139	1139	1139	1139	1	1	0	11	0	6
	P_CATUS1	1408	1380	1798	2224	2266	4	2	0	3	0	11
	P_MIOOL14	895	875	1082	1244	1283	6	6	0	4	0	5
	P_HYLNA18	930	930	930	930	930	1	1	0	9	0	5
	P_HYPSU3	1302	1302	1302	1302	1302	1	1	0	7	0	21
	P_MYCHI5	889	878	1422	2451	2729	24	7	0	2	0	5
	P_PHYOP7	896	874	1229	1397	1398	8	3	0	5	0	5
	P_PYRLE11	1308	1308	1308	1308	1308	1	1	0	7	0	21
	P_RAMFU8	300	300	300	300	300	1	1	1	82	0	0
	P_TURNI2	1469	1138	2426	3049	3079	9	5	0	1	0	6
	P_TROPE17	2350	2350	2350	2350	2350	1	1	0	4	0	4
	P_TURIG20	1398	1398	1398	1398	1398	1	1	0	5	0	11
Haemoproteus	H_THRCY32	2633	2341	2882	3075	3117	77	10	0	0	0	4
	H_IRIAN3	1243	1242	1925	2976	2977	15	6	0	1	0	10
	H_ATLME35	1128	1125	1608	2684	2750	16	5	0	2	0	6
	H_BASCO20	1279	1202	1578	1773	1796	13	4	0	1	0	10
	H_BASSI21	1531	1505	1760	1990	2016	2	2	0	3	0	18
	H_THRCY38	2899	2899	2899	2899	2899	2	2	0	1	0	4
	H_MYARA9	1131	1125	1367	1963	2268	8	2	0	3	0	6
	H_CHLCA22	938	935	1046	1152	1155	4	4	0	11	0	5
	H_CHLCA23	1202	1202	1202	1202	1202	1	1	0	2	0	10
	H_CHLCA24	1390	1385	1965	2692	2692	12	5	0	9	0	11
	H_CHLCA16	1052	956	1867	2975	2994	12	7	0	1	0	9
	H_CHLFL37	1444	1385	1940	2408	2456	3	2	0	2	0	11
	H_CYAVI13	2537	2537	2537	2537	2537	1	1	0	8	0	5
	H_DIGBR17	2023	1943	2802	3051	3079	23	2	0	1	0	11
	H_DIGBR19	2911	2911	2911	2911	2911	1	1	0	1	0	10
	H_DIGBR18	2959	2959	2959	2959	2959	1	1	0	1	0	10
	H_DIGBR31	2969	2969	2969	2969	2969	1	1	0	1	0	10
	H_DIGGL4	949	934	1088	1227	1242	2	2	0	2	0	5
	H_ELAPA12	2924	2924	2924	2924	2924	1	1	0	1	0	10
	H_EUPXA10	1485	1324	2420	2991	2997	3	2	0	1	0	21
	H_EUTCO1	1116	1115	1246	1457	1493	3	2	0	3	0	6
	H_HYLOC14	937	937	937	937	937	1	1	0	9	0	5

	H_MIOST8	939	936	1269	1632	1709	6	3	0	1	0	5
	H_MICLA2	1138	1138	1138	1138	1138	1	1	0	11	0	6
	H_MIOST7	2736	2736	2736	2736	2736	1	1	0	2	1	6
	H_PIPAR6	2549	2533	2659	2744	2749	3	1	0	2	0	5
	H_TANCH25	990	956	1245	1399	1399	5	3	0	5	0	9
	H_TANAR27	925	878	1238	1405	1407	5	3	0	3	0	5
	H_THREP30	1212	1202	1300	1389	1399	2	2	0	5	0	10
	H_TANPU29	1134	1127	1245	1386	1407	3	2	0	3	0	6
	H_THLRU26	1968	1968	1968	1968	1968	1	1	0	5	0	7
	H_THRCY28	2662	2662	2662	2662	2662	1	1	0	9	0	5
	H_ZONCA34	2988	2988	2988	2988	2988	1	1	0	1	0	10
Leucocytozoon	L_AMBHO132	2674	2674	2674	2674	2674	1	1	1	9	0	5
	L_ARETO103	2784	2782	2830	2930	2954	4	3	0	1	0	9
	L_ARETO59	2830	2802	2947	3001	3002	4	4	0	2	0	10
	L_ATLME77	2488	2460	2731	2960	2974	9	5	0	1	0	9
	L_ATLME80	2048	1976	2698	3000	3004	23	10	0	2	0	7
	L_BUTMO90	2458	1979	2803	3097	3377	16	12	0	0	0	7
	L_HEMAT126	1976	1968	2725	3005	3013	29	14	1	2	0	7
	L_DIGBR46	2982	2981	2998	3016	3018	3	3	0	2	0	10
	L_ARETO96	2787	2787	2787	2787	2787	1	1	0	2	0	9
	L_ARRTO113	2668	2667	2680	2691	2692	2	2	0	9	0	5
	L_ATLME100	2509	2466	2699	2925	2997	8	2	0	1	0	9
	L_ATLME133	2656	2656	2656	2656	2656	1	1	0	9	0	5
	L_ATLME138	2466	2466	2466	2466	2466	1	1	0	2	0	9
	L_ATLME141	2788	2788	2788	2788	2788	1	1	0	2	0	9
	L_ATLME17	2662	2662	2662	2662	2662	1	1	0	9	0	5
	L_ATLME40	2730	2730	2730	2730	2730	1	1	0	2	0	6
	L_ATLME41	2638	2637	2649	2660	2661	2	2	0	9	0	2
	L_ATLME49	1399	1137	2401	2991	2996	36	21	0	1	0	6
	L_ATLME53	2630	2630	2630	2630	2630	1	1	0	9	0	2
	L_ATLME83	2339	2339	2340	2341	2341	2	2	0	2	0	4
	L_ATLME99	2929	2929	2929	2929	2929	1	1	0	1	0	10
	L_BASLU104	2011	1973	2358	2709	2749	3	2	0	2	0	7
	L_CHLRI43	1571	1398	2362	2992	2999	7	7	0	1	0	11
	L_CHLRI54	2749	2749	2778	2827	2835	3	3	0	0	0	6
	L_IRIJE30	1678	1398	2595	2955	2959	5	5	0	1	0	11
	L_MYIME127	1972	1968	2552	2794	2802	11	7	0	0	0	7
	L_SYNAZ125	2634	2626	2783	2976	2994	6	5	0	1	0	2
	L_AULCO3	1528	1511	1719	1937	1965	3	3	0	5	1	18
	L_AULCO4	1968	1968	1968	1968	1968	1	1	0	5	0	7
	L_CHLRI29	2646	2631	2750	2830	2835	3	2	0	0	0	2
	L_BUTMO19	3003	3003	3003	3003	3003	1	1	0	2	0	0
	L_BUTMO20	2899	2899	2899	2899	2899	1	1	0	1	0	4
	L_BUTMO58	2692	2692	2692	2692	2692	1	1	0	9	0	5
	L_CACCH48	2261	2261	2261	2261	2261	1	1	0	3	0	1
	L_DIGBR35	1341	1129	2337	3029	3056	6	4	0	1	0	6
	L_CATDI139	2959	2959	2959	2959	2959	1	1	0	1	0	10
	L_MYARA32	1416	1380	2307	2941	2968	5	5	0	1	0	11
	L_CATFU24	1968	1968	1968	1968	1968	1	1	0	5	0	7
	L_CHLRI76	2924	2924	2924	2924	2924	1	1	0	1	0	10

L_CHLRI81	2689	2689	2787	2953	2982	3	1	0	1	0	5
L_MECST6	1197	1134	2130	2982	2994	6	4	0	1	0	6
L_CONAR57	1303	1302	1336	1383	1386	5	2	0	5	0	21
L_CYAVI37	2522	2518	2727	2957	2999	5	1	0	1	0	5
L_CYAVI9	2997	2997	2997	2997	2997	1	1	0	1	1	10
L_DIGBR101	2899	2899	2899	2899	2899	1	1	0	1	0	4
L_DIGBR102	2969	2969	2969	2969	2969	1	1	0	1	0	10
L_DIGBR136	3056	3056	3056	3056	3056	1	1	0	1	0	1
L_DIGBR15	2733	2720	2846	2958	2971	2	1	0	1	0	6
L_DIGBR18	2957	2957	2957	2957	2957	1	1	0	1	0	10
L_DIGBR45	2750	2750	2750	2750	2750	1	1	0	2	0	12
L_DIGBR47	2970	2970	2970	2970	2970	1	1	0	1	0	10
L_DIGSI116	2159	1968	2764	2981	2983	5	2	0	1	0	7
L_DIGCY14	2756	2751	2796	2837	2842	2	2	0	0	0	9
L_DIGCY34	1399	1399	1399	1399	1399	1	1	0	5	0	11
L_DIGMY135	2938	2938	2938	2938	2938	1	1	0	1	0	10
L_DIGMY98	2995	2995	2995	2995	2995	1	1	0	1	0	10
L_ELAPA2	2755	2755	2755	2755	2755	1	1	0	2	0	9
L_ELAPA44	2750	2750	2750	2750	2750	1	1	0	2	0	12
L_ENTLE22	2021	2021	2021	2021	2021	1	1	0	3	0	7
L_ENTLE27	1976	1976	1976	1976	1976	1	1	0	5	0	7
L_ENTLE63	1139	1139	1418	1892	1976	3	1	0	5	0	6
L_EUBVE10	1968	1968	1968	1968	1968	1	1	0	5	0	7
L_GRAER1	1980	1980	1980	1980	1980	1	1	0	5	0	7
L_GRAFE13	2693	2688	2740	2788	2793	2	1	0	2	0	5
L_GRAFE5	2788	2788	2788	2788	2788	1	1	0	2	0	9
L_IRIAN86	1976	1976	1976	1976	1976	1	1	0	5	0	7
L_IRIJE95	2924	2924	2924	2924	2924	1	1	0	1	0	10
L_MIOST21	2689	2689	2689	2689	2689	1	1	0	9	1	5
L_PIPAR11	2533	2533	2533	2533	2533	1	1	0	8	0	5
L_PIPAR118	2749	2749	2749	2749	2749	1	1	0	2	0	6
L_PIPAR119	2695	2695	2695	2695	2695	1	1	1	9	0	5
L_PIPIN12	2751	2751	2751	2751	2751	1	1	0	2	0	9
L_PIPIN120	2141	2021	2634	2963	3004	5	1	0	2	0	7
L_PIPIN121	2758	2758	2758	2758	2758	1	1	0	2	0	9
L_PIPIN137	2526	2526	2526	2526	2526	1	1	0	8	0	5
L_SYNAZ38	2470	2470	2470	2470	2470	1	1	0	2	0	9
L_TANVA97	2692	2692	2692	2692	2692	1	1	0	9	0	5
L_THRBO16	2988	2988	2988	2988	2988	1	1	0	1	0	10
L_THRBO39	2979	2979	2979	2979	2979	1	1	0	1	0	10
L_THRCY140	2656	2656	2656	2656	2656	1	1	0	9	0	5
L_THRCY52	2635	2635	2635	2635	2635	1	1	0	9	0	2
L_THRCY87	2677	2662	2816	2955	2970	2	1	0	1	0	5
L_TURCH25	2708	2667	2875	2975	2988	5	3	0	1	0	5
L_TURCH26	2997	2997	2997	2997	2997	1	1	0	1	0	10
L_TURCH68	2960	2954	3016	3073	3079	2	2	0	1	0	10
L_TURCH70	1661	1138	2740	3246	3377	8	4	0	0	0	6
L_TURFU64	2992	2987	3033	3074	3079	2	1	0	1	0	10
L_TURIG65	1398	1398	1398	1398	1398	1	1	0	5	0	11
L_TURSE28	2331	2331	2331	2331	2331	1	1	0	2	0	4

	L_TURSE67	2362	2341	2510	2629	2638	3	1	0	9	0	4
Total limiting lineages									5		4	

Table 5.3 Number of range limiting lineages when infection zones are classified as 200 m into parasite lineage maximum and minimum recorded elevations.

200 m into parasite lineage maximum and minimum elevation												
Genus	Lineage	Low CI	Low limit	Mean elevation	Up CI	Up limit	Infected birds	Infected bird spp.	Infected low limited bird spp.	Low limited bird spp.	Infected up limited bird spp.	Up limit bird spp.
Plasmodium	P_MYARA13	2613	1399	2843	3004	3079	40	15	0	2	0	32
	P_ARRTA9	300	300	300	300	300	1	1	1	82	0	4
	P_ARRTO12	2787	2787	2787	2787	2787	1	1	0	11	0	13
	P_AULPR10	936	936	936	936	936	1	1	0	21	0	16
	P_AUTRU15	886	882	919	952	956	2	2	0	18	0	14
	P_DIGCY21	2532	2518	2660	2788	2802	2	2	0	2	0	7
	P_DYSME16	928	926	944	956	956	4	1	0	18	0	16
	P_ENTLE4	1139	1139	1139	1139	1139	1	1	0	33	0	16
	P_CATUS1	1408	1380	1798	2224	2266	4	2	0	9	0	32
	P_MIOOL14	895	875	1082	1244	1283	6	6	0	15	0	14
	P_HYLNA18	930	930	930	930	930	1	1	0	21	0	16
	P_HYPSU3	1302	1302	1302	1302	1302	1	1	0	9	0	38
	P_MYCHI5	889	878	1422	2451	2729	24	7	0	11	0	14
	P_PHYOP7	896	874	1229	1397	1398	8	3	0	9	0	14
	P_PYRLE11	1308	1308	1308	1308	1308	1	1	0	9	0	38
	P_RAMFU8	300	300	300	300	300	1	1	1	82	0	4
	P_TURNI2	1469	1138	2426	3049	3079	9	5	0	2	0	16
	P_TROPE17	2350	2350	2350	2350	2350	1	1	0	8	0	13
	P_TURIG20	1398	1398	1398	1398	1398	1	1	0	9	1	32
Haemoproteus	H_THRCY32	2633	2341	2882	3075	3117	77	10	0	2	0	11
	H_IRIAN3	1243	1242	1925	2976	2977	15	6	0	2	0	31
	H_ATLME35	1128	1125	1608	2684	2750	16	5	0	11	0	16
	H_BASCO20	1279	1202	1578	1773	1796	13	4	0	6	0	31
	H_BASSI21	1531	1505	1760	1990	2016	2	2	0	11	0	21
	H_THRCY38	2899	2899	2899	2899	2899	2	2	0	3	1	14
	H_MYARA9	1131	1125	1367	1963	2268	8	2	0	9	0	16
	H_CHLCA22	938	935	1046	1152	1155	4	4	2	31	0	16
	H_CHLCA23	1202	1202	1202	1202	1202	1	1	0	13	0	31
	H_CHLCA24	1390	1385	1965	2692	2692	12	5	0	17	0	32
	H_CHLCA16	1052	956	1867	2975	2994	12	7	0	2	0	15
	H_CHLFL37	1444	1385	1940	2408	2456	3	2	0	6	0	32
	H_CYAVI13	2537	2537	2537	2537	2537	1	1	0	12	0	7
	H_DIGBR17	2023	1943	2802	3051	3079	23	2	0	2	0	18
	H_DIGBR19	2911	2911	2911	2911	2911	1	1	0	1	0	10
	H_DIGBR18	2959	2959	2959	2959	2959	1	1	0	2	0	11
	H_DIGBR31	2969	2969	2969	2969	2969	1	1	0	2	0	11

	H_DIGGL4	949	934	1088	1227	1242	2	2	0	13	0	16
	H_ELAPA12	2924	2924	2924	2924	2924	1	1	0	1	0	10
	H_EUPXA10	1485	1324	2420	2991	2997	3	2	0	2	0	38
	H_EUTCO1	1116	1115	1246	1457	1493	3	2	0	8	0	16
	H_HYLOC14	937	937	937	937	937	1	1	0	21	1	16
	H_MIOST8	939	936	1269	1632	1709	6	3	0	7	1	16
	H_MICLA2	1138	1138	1138	1138	1138	1	1	0	33	0	16
	H_MIOST7	2736	2736	2736	2736	2736	1	1	0	11	1	16
	H_PIPAR6	2549	2533	2659	2744	2749	3	1	1	11	0	7
	H_TANCH25	990	956	1245	1399	1399	5	3	0	9	1	15
	H_TANAR27	925	878	1238	1405	1407	5	3	0	10	0	14
	H_THREP30	1212	1202	1300	1389	1399	2	2	0	9	0	31
	H_TANPU29	1134	1127	1245	1386	1407	3	2	0	10	0	16
	H_THLRU26	1968	1968	1968	1968	1968	1	1	0	9	0	17
	H_THRCY28	2662	2662	2662	2662	2662	1	1	0	17	0	14
	H_ZONCA34	2988	2988	2988	2988	2988	1	1	0	2	0	11
Leucocytozoon	L_AMBHO132	2674	2674	2674	2674	2674	1	1	1	17	0	14
	L_ARETO103	2784	2782	2830	2930	2954	4	3	0	2	0	13
	L_ARETO59	2830	2802	2947	3001	3002	4	4	0	3	1	20
	L_ATLME77	2488	2460	2731	2960	2974	9	5	0	2	0	12
	L_ATLME80	2048	1976	2698	3000	3004	23	10	0	3	0	17
	L_BUTMO90	2458	1979	2803	3097	3377	16	12	0	2	0	17
	L_HEMAT126	1976	1968	2725	3005	3013	29	14	1	3	0	17
	L_DIGBR46	2982	2981	2998	3016	3018	3	3	0	3	0	11
	L_ARETO96	2787	2787	2787	2787	2787	1	1	0	11	0	13
	L_ARRTO113	2668	2667	2680	2691	2692	2	2	1	17	0	14
	L_ATLME100	2509	2466	2699	2925	2997	8	2	0	2	0	12
	L_ATLME133	2656	2656	2656	2656	2656	1	1	0	17	0	14
	L_ATLME138	2466	2466	2466	2466	2466	1	1	0	6	0	12
	L_ATLME141	2788	2788	2788	2788	2788	1	1	0	11	0	13
	L_ATLME17	2662	2662	2662	2662	2662	1	1	0	17	0	14
	L_ATLME40	2730	2730	2730	2730	2730	1	1	0	11	0	16
	L_ATLME41	2638	2637	2649	2660	2661	2	2	0	17	0	8
	L_ATLME49	1399	1137	2401	2991	2996	36	21	0	2	0	16
	L_ATLME53	2630	2630	2630	2630	2630	1	1	0	17	0	8
	L_ATLME83	2339	2339	2340	2341	2341	2	2	1	6	0	11
	L_ATLME99	2929	2929	2929	2929	2929	1	1	0	1	0	10
	L_BASLU104	2011	1973	2358	2709	2749	3	2	0	11	0	17
	L_CHLRI43	1571	1398	2362	2992	2999	7	7	0	2	0	32
	L_CHLRI54	2749	2749	2778	2827	2835	3	3	0	2	0	16
	L_IRIJE30	1678	1398	2595	2955	2959	5	5	0	2	0	32
	L_MYIME127	1972	1968	2552	2794	2802	11	7	0	2	0	17
	L_SYNAZ125	2634	2626	2783	2976	2994	6	5	0	2	0	8
	L_AULCO3	1528	1511	1719	1937	1965	3	3	0	9	1	21
	L_AULCO4	1968	1968	1968	1968	1968	1	1	0	9	0	17
	L_CHLRI29	2646	2631	2750	2830	2835	3	2	0	2	0	8
	L_BUTMO19	3003	3003	3003	3003	3003	1	1	0	3	0	7
	L_BUTMO20	2899	2899	2899	2899	2899	1	1	0	3	0	14
	L_BUTMO58	2692	2692	2692	2692	2692	1	1	1	17	0	14
	L_CACCH48	2261	2261	2261	2261	2261	1	1	0	9	0	5

L_DIGBR35	1341	1129	2337	3029	3056	6	4	0	2	0	16
L_CATDI139	2959	2959	2959	2959	2959	1	1	0	2	0	11
L_MYARA32	1416	1380	2307	2941	2968	5	5	0	2	0	32
L_CATFU24	1968	1968	1968	1968	1968	1	1	0	9	0	17
L_CHLRI76	2924	2924	2924	2924	2924	1	1	0	1	0	10
L_CHLRI81	2689	2689	2787	2953	2982	3	1	0	2	0	14
L_MECST6	1197	1134	2130	2982	2994	6	4	0	2	0	16
L_CONAR57	1303	1302	1336	1383	1386	5	2	0	9	0	38
L_CYAVI37	2522	2518	2727	2957	2999	5	1	0	2	0	7
L_CYAVI9	2997	2997	2997	2997	2997	1	1	0	2	1	11
L_DIGBR101	2899	2899	2899	2899	2899	1	1	0	3	0	14
L_DIGBR102	2969	2969	2969	2969	2969	1	1	0	2	0	11
L_DIGBR136	3056	3056	3056	3056	3056	1	1	0	2	0	18
L_DIGBR15	2733	2720	2846	2958	2971	2	1	0	2	0	16
L_DIGBR18	2957	2957	2957	2957	2957	1	1	0	2	0	11
L_DIGBR45	2750	2750	2750	2750	2750	1	1	1	11	0	16
L_DIGBR47	2970	2970	2970	2970	2970	1	1	0	2	0	11
L_DIGSI116	2159	1968	2764	2981	2983	5	2	0	2	0	17
L_DIGCY14	2756	2751	2796	2837	2842	2	2	0	2	0	13
L_DIGCY34	1399	1399	1399	1399	1399	1	1	0	9	0	32
L_DIGMY135	2938	2938	2938	2938	2938	1	1	0	1	0	10
L_DIGMY98	2995	2995	2995	2995	2995	1	1	0	2	0	11
L_ELAPA2	2755	2755	2755	2755	2755	1	1	0	11	0	13
L_ELAPA44	2750	2750	2750	2750	2750	1	1	0	11	0	16
L_ENTLE22	2021	2021	2021	2021	2021	1	1	0	11	0	15
L_ENTLE27	1976	1976	1976	1976	1976	1	1	0	9	0	17
L_ENTLE63	1139	1139	1418	1892	1976	3	1	0	9	0	16
L_EUBVE10	1968	1968	1968	1968	1968	1	1	0	9	1	17
L_GRAER1	1980	1980	1980	1980	1980	1	1	0	9	0	17
L_GRAFE13	2693	2688	2740	2788	2793	2	1	1	11	0	14
L_GRAFE5	2788	2788	2788	2788	2788	1	1	1	11	0	13
L_IRIAN86	1976	1976	1976	1976	1976	1	1	0	9	0	17
L_IRIJE95	2924	2924	2924	2924	2924	1	1	0	1	0	10
L_MIOST21	2689	2689	2689	2689	2689	1	1	0	17	1	14
L_PIPAR11	2533	2533	2533	2533	2533	1	1	0	12	0	7
L_PIPAR118	2749	2749	2749	2749	2749	1	1	1	11	0	16
L_PIPAR119	2695	2695	2695	2695	2695	1	1	1	17	0	14
L_PIPIN12	2751	2751	2751	2751	2751	1	1	0	11	1	13
L_PIPIN120	2141	2021	2634	2963	3004	5	1	0	3	0	15
L_PIPIN121	2758	2758	2758	2758	2758	1	1	0	11	1	13
L_PIPIN137	2526	2526	2526	2526	2526	1	1	0	12	0	7
L_SYNAZ38	2470	2470	2470	2470	2470	1	1	0	6	0	12
L_TANVA97	2692	2692	2692	2692	2692	1	1	0	17	0	14
L_THRBO16	2988	2988	2988	2988	2988	1	1	0	2	0	11
L_THRBO39	2979	2979	2979	2979	2979	1	1	0	2	0	11
L_THRCY140	2656	2656	2656	2656	2656	1	1	0	17	0	14
L_THRCY52	2635	2635	2635	2635	2635	1	1	0	17	0	8
L_THRCY87	2677	2662	2816	2955	2970	2	1	0	2	0	14
L_TURCH25	2708	2667	2875	2975	2988	5	3	0	2	0	14
L_TURCH26	2997	2997	2997	2997	2997	1	1	0	2	0	11

	L_TURCH68	2960	2954	3016	3073	3079	2	2	0	2	0	11
	L_TURCH70	1661	1138	2740	3246	3377	8	4	0	2	0	16
	L_TURFU64	2992	2987	3033	3074	3079	2	1	0	2	0	11
	L_TURIG65	1398	1398	1398	1398	1398	1	1	0	9	1	32
	L_TURSE28	2331	2331	2331	2331	2331	1	1	0	6	0	11
	L_TURSE67	2362	2341	2510	2629	2638	3	1	0	17	0	11
Total limiting lineages									14		14	

Table 5.4 Number of range limiting lineages when infection zones are classified as 100 m into parasite lineage 95 % CI's.

100 m into parasite lineage 95% CI's												
Genus	Lineage	Low CI	Low limit	Mean elevation	Up CI	Up limit	Infected birds	Infected bird spp.	Infected low limited bird spp.	Low limited bird spp.	Infected up limited bird spp.	Up limit bird spp.
Plasmodium	P_MYARA13	2613	1399	2843	3004	3079	40	15	0	2	0	2
	P_ARRTA9	300	300	300	300	300	1	1	1	82	0	0
	P_ARRTO12	2787	2787	2787	2787	2787	1	1	0	2	0	9
	P_AULPR10	936	936	936	936	936	1	1	0	9	0	5
	P_AUTRU15	886	882	919	952	956	2	2	0	8	0	5
	P_DIGCY21	2532	2518	2660	2788	2802	2	2	0	2	0	5
	P_DYSME16	928	926	944	956	956	4	1	0	8	0	5
	P_ENTLE4	1139	1139	1139	1139	1139	1	1	0	11	0	6
	P_CATUS1	1408	1380	1798	2224	2266	4	2	0	4	0	17
	P_MIOOL14	895	875	1082	1244	1283	6	6	0	2	0	5
	P_HYLNA18	930	930	930	930	930	1	1	0	9	0	5
	P_HYPSU3	1302	1302	1302	1302	1302	1	1	0	7	0	21
	P_MYCHI5	889	878	1422	2451	2729	24	7	0	2	0	5
	P_PHYOP7	896	874	1229	1397	1398	8	3	0	5	0	5
	P_PYRLE11	1308	1308	1308	1308	1308	1	1	0	7	0	21
	P_RAMFU8	300	300	300	300	300	1	1	1	82	0	0
	P_TURNI2	1469	1138	2426	3049	3079	9	5	0	2	0	21
	P_TROPE17	2350	2350	2350	2350	2350	1	1	0	4	0	4
	P_TURIG20	1398	1398	1398	1398	1398	1	1	0	5	0	11
Haemoproteus	H_THRCY32	2633	2341	2882	3075	3117	77	10	0	1	0	2
	H_IRIAN3	1243	1242	1925	2976	2977	15	6	0	1	0	10
	H_ATLME35	1128	1125	1608	2684	2750	16	5	0	9	0	6
	H_BASCO20	1279	1202	1578	1773	1796	13	4	0	1	0	13
	H_BASSI21	1531	1505	1760	1990	2016	2	2	0	5	0	18
	H_THRCY38	2899	2899	2899	2899	2899	2	2	0	1	0	4
	H_MYARA9	1131	1125	1367	1963	2268	8	2	0	5	0	6
	H_CHLCA22	938	935	1046	1152	1155	4	4	0	11	0	5
	H_CHLCA23	1202	1202	1202	1202	1202	1	1	0	2	0	10
	H_CHLCA24	1390	1385	1965	2692	2692	12	5	0	9	0	11
	H_CHLCA16	1052	956	1867	2975	2994	12	7	0	1	0	6
	H_CHLFL37	1444	1385	1940	2408	2456	3	2	0	4	0	17

	H_CYAVI13	2537	2537	2537	2537	2537	1	1	0	8	0	5
	H_DIGBR17	2023	1943	2802	3051	3079	23	2	0	1	0	7
	H_DIGBR19	2911	2911	2911	2911	2911	1	1	0	1	0	10
	H_DIGBR18	2959	2959	2959	2959	2959	1	1	0	1	0	10
	H_DIGBR31	2969	2969	2969	2969	2969	1	1	0	1	0	10
	H_DIGGL4	949	934	1088	1227	1242	2	2	0	2	0	5
	H_ELAPA12	2924	2924	2924	2924	2924	1	1	0	1	0	10
	H_EUPXA10	1485	1324	2420	2991	2997	3	2	0	1	0	21
	H_EUTCO1	1116	1115	1246	1457	1493	3	2	0	3	0	6
	H_HYLOC14	937	937	937	937	937	1	1	0	9	0	5
	H_MIOST8	939	936	1269	1632	1709	6	3	0	6	0	5
	H_MICLA2	1138	1138	1138	1138	1138	1	1	0	11	0	6
	H_MIOST7	2736	2736	2736	2736	2736	1	1	0	2	1	6
	H_PIPAR6	2549	2533	2659	2744	2749	3	1	0	2	0	5
	H_TANCH25	990	956	1245	1399	1399	5	3	0	5	0	9
	H_TANAR27	925	878	1238	1405	1407	5	3	0	3	0	5
	H_THREP30	1212	1202	1300	1389	1399	2	2	0	5	0	10
	H_TANPU29	1134	1127	1245	1386	1407	3	2	0	5	0	6
	H_THLRU26	1968	1968	1968	1968	1968	1	1	0	5	0	7
	H_THRCY28	2662	2662	2662	2662	2662	1	1	0	9	0	5
	H_ZONCA34	2988	2988	2988	2988	2988	1	1	0	1	0	10
Leucocytozoon	L_AMBHO132	2674	2674	2674	2674	2674	1	1	1	9	0	5
	L_ARETO103	2784	2782	2830	2930	2954	4	3	0	1	0	9
	L_ARETO59	2830	2802	2947	3001	3002	4	4	0	2	0	10
	L_ATLME77	2488	2460	2731	2960	2974	9	5	0	1	0	9
	L_ATLME80	2048	1976	2698	3000	3004	23	10	0	2	0	7
	L_BUTMO90	2458	1979	2803	3097	3377	16	12	0	1	0	9
	L_HEMAT126	1976	1968	2725	3005	3013	29	14	1	2	0	7
	L_DIGBR46	2982	2981	2998	3016	3018	3	3	0	2	0	10
	L_ARETO96	2787	2787	2787	2787	2787	1	1	0	2	0	9
	L_ARRTO113	2668	2667	2680	2691	2692	2	2	0	9	0	5
	L_ATLME100	2509	2466	2699	2925	2997	8	2	0	1	0	5
	L_ATLME133	2656	2656	2656	2656	2656	1	1	0	9	0	5
	L_ATLME138	2466	2466	2466	2466	2466	1	1	0	2	0	9
	L_ATLME141	2788	2788	2788	2788	2788	1	1	0	2	0	9
	L_ATLME17	2662	2662	2662	2662	2662	1	1	0	9	0	5
	L_ATLME40	2730	2730	2730	2730	2730	1	1	0	2	0	6
	L_ATLME41	2638	2637	2649	2660	2661	2	2	0	9	0	2
	L_ATLME49	1399	1137	2401	2991	2996	36	21	0	1	0	11
	L_ATLME53	2630	2630	2630	2630	2630	1	1	0	9	0	2
	L_ATLME83	2339	2339	2340	2341	2341	2	2	0	2	0	4
	L_ATLME99	2929	2929	2929	2929	2929	1	1	0	1	0	10
	L_BASLU104	2011	1973	2358	2709	2749	3	2	0	2	0	7
	L_CHLRI43	1571	1398	2362	2992	2999	7	7	0	1	0	17
	L_CHLRI54	2749	2749	2778	2827	2835	3	3	0	0	0	6
	L_IRIJE30	1678	1398	2595	2955	2959	5	5	0	1	0	0
	L_MYIME127	1972	1968	2552	2794	2802	11	7	0	2	0	7
	L_SYNAZ125	2634	2626	2783	2976	2994	6	5	0	1	0	2
	L_AULCO3	1528	1511	1719	1937	1965	3	3	0	8	0	18
	L_AULCO4	1968	1968	1968	1968	1968	1	1	0	5	0	7

L_CHLRI29	2646	2631	2750	2830	2835	3	2	0	0	0	2
L_BUTMO19	3003	3003	3003	3003	3003	1	1	0	2	0	0
L_BUTMO20	2899	2899	2899	2899	2899	1	1	0	1	0	4
L_BUTMO58	2692	2692	2692	2692	2692	1	1	0	9	0	5
L_CACCH48	2261	2261	2261	2261	2261	1	1	0	3	0	1
L_DIGBR35	1341	1129	2337	3029	3056	6	4	0	2	0	21
L_CATDI139	2959	2959	2959	2959	2959	1	1	0	1	0	10
L_MYARA32	1416	1380	2307	2941	2968	5	5	0	1	0	17
L_CATFU24	1968	1968	1968	1968	1968	1	1	0	5	0	7
L_CHLRI76	2924	2924	2924	2924	2924	1	1	0	1	0	10
L_CHLRI81	2689	2689	2787	2953	2982	3	1	0	1	0	5
L_MECST6	1197	1134	2130	2982	2994	6	4	0	1	0	12
L_CONAR57	1303	1302	1336	1383	1386	5	2	0	5	0	21
L_CYAVI37	2522	2518	2727	2957	2999	5	1	0	1	0	5
L_CYAVI9	2997	2997	2997	2997	2997	1	1	0	1	1	10
L_DIGBR101	2899	2899	2899	2899	2899	1	1	0	1	0	4
L_DIGBR102	2969	2969	2969	2969	2969	1	1	0	1	0	10
L_DIGBR136	3056	3056	3056	3056	3056	1	1	0	1	0	1
L_DIGBR15	2733	2720	2846	2958	2971	2	1	0	1	0	6
L_DIGBR18	2957	2957	2957	2957	2957	1	1	0	1	0	10
L_DIGBR45	2750	2750	2750	2750	2750	1	1	0	2	0	12
L_DIGBR47	2970	2970	2970	2970	2970	1	1	0	1	0	10
L_DIGSI116	2159	1968	2764	2981	2983	5	2	0	1	0	5
L_DIGCY14	2756	2751	2796	2837	2842	2	2	0	0	0	9
L_DIGCY34	1399	1399	1399	1399	1399	1	1	0	5	0	11
L_DIGMY135	2938	2938	2938	2938	2938	1	1	0	1	0	10
L_DIGMY98	2995	2995	2995	2995	2995	1	1	0	1	0	10
L_ELAPA2	2755	2755	2755	2755	2755	1	1	0	2	0	9
L_ELAPA44	2750	2750	2750	2750	2750	1	1	0	2	0	12
L_ENTLE22	2021	2021	2021	2021	2021	1	1	0	3	0	7
L_ENTLE27	1976	1976	1976	1976	1976	1	1	0	5	0	7
L_ENTLE63	1139	1139	1418	1892	1976	3	1	0	4	0	6
L_EUBVE10	1968	1968	1968	1968	1968	1	1	0	5	0	7
L_GRAER1	1980	1980	1980	1980	1980	1	1	0	5	0	7
L_GRAFE13	2693	2688	2740	2788	2793	2	1	0	2	0	5
L_GRAFE5	2788	2788	2788	2788	2788	1	1	0	2	0	9
L_IRIAN86	1976	1976	1976	1976	1976	1	1	0	5	0	7
L_IRIJE95	2924	2924	2924	2924	2924	1	1	0	1	0	10
L_MIOST21	2689	2689	2689	2689	2689	1	1	0	9	1	5
L_PIPAR11	2533	2533	2533	2533	2533	1	1	0	8	0	5
L_PIPAR118	2749	2749	2749	2749	2749	1	1	0	2	0	6
L_PIPAR119	2695	2695	2695	2695	2695	1	1	1	9	0	5
L_PIPIN12	2751	2751	2751	2751	2751	1	1	0	2	0	9
L_PIPIN120	2141	2021	2634	2963	3004	5	1	0	1	0	8
L_PIPIN121	2758	2758	2758	2758	2758	1	1	0	2	0	9
L_PIPIN137	2526	2526	2526	2526	2526	1	1	0	8	0	5
L_SYNAZ38	2470	2470	2470	2470	2470	1	1	0	2	0	9
L_TANVA97	2692	2692	2692	2692	2692	1	1	0	9	0	5
L_THRBO16	2988	2988	2988	2988	2988	1	1	0	1	0	10
L_THRBO39	2979	2979	2979	2979	2979	1	1	0	1	0	10

	L_THRCY140	2656	2656	2656	2656	2656	1	1	0	9	0	5
	L_THRCY52	2635	2635	2635	2635	2635	1	1	0	9	0	2
	L_THRCY87	2677	2662	2816	2955	2970	2	1	0	1	0	5
	L_TURCH25	2708	2667	2875	2975	2988	5	3	0	1	0	6
	L_TURCH26	2997	2997	2997	2997	2997	1	1	0	1	0	10
	L_TURCH68	2960	2954	3016	3073	3079	2	2	0	1	0	10
	L_TURCH70	1661	1138	2740	3246	3377	8	4	0	1	0	0
	L_TURFU64	2992	2987	3033	3074	3079	2	1	0	1	0	10
	L_TURIG65	1398	1398	1398	1398	1398	1	1	0	5	0	11
	L_TURSE28	2331	2331	2331	2331	2331	1	1	0	2	0	4
	L_TURSE67	2362	2341	2510	2629	2638	3	1	0	9	0	4
Total limiting lineages									5		3	

Table 5.5 Number of range limiting lineages when infection zones are classified as 200 m into parasite lineage 95 % CI's.

200 m into parasite lineage 95% CI's												
Genus	Lineage	Low CI	Low limit	Mean elevation	Up CI	Up limit	Infected birds	Infected bird spp.	Infected low limited bird spp.	Low limited bird spp.	Infected up limited bird spp.	Up limit bird spp.
Plasmodium	P_MYARA13	2613	1399	2843	3004	3079	40	15	0	3	0	8
	P_ARRTA9	300	300	300	300	300	1	1	1	82	0	4
	P_ARRTO12	2787	2787	2787	2787	2787	1	1	0	11	0	13
	P_AULPR10	936	936	936	936	936	1	1	0	21	0	16
	P_AUTRU15	886	882	919	952	956	2	2	0	18	0	14
	P_DIGCY21	2532	2518	2660	2788	2802	2	2	0	11	0	7
	P_DYSME16	928	926	944	956	956	4	1	0	18	0	16
	P_ENTLE4	1139	1139	1139	1139	1139	1	1	0	33	0	16
	P_CATUS1	1408	1380	1798	2224	2266	4	2	0	9	0	35
	P_MIOOL14	895	875	1082	1244	1283	6	6	0	13	0	14
	P_HYLNA18	930	930	930	930	930	1	1	0	21	0	16
	P_HYPSU3	1302	1302	1302	1302	1302	1	1	0	9	0	38
	P_MYCHI5	889	878	1422	2451	2729	24	7	0	6	0	14
	P_PHYOP7	896	874	1229	1397	1398	8	3	0	9	0	14
	P_PYRLE11	1308	1308	1308	1308	1308	1	1	0	9	0	38
	P_RAMFU8	300	300	300	300	300	1	1	1	82	0	4
	P_TURNI2	1469	1138	2426	3049	3079	9	5	0	3	0	38
	P_TROPE17	2350	2350	2350	2350	2350	1	1	0	8	0	13
	P_TURIG20	1398	1398	1398	1398	1398	1	1	0	9	1	32
Haemoproteus	H_THRCY32	2633	2341	2882	3075	3117	77	10	0	2	0	8
	H_IRIAN3	1243	1242	1925	2976	2977	15	6	0	2	0	31
	H_ATLME35	1128	1125	1608	2684	2750	16	5	0	17	0	16
	H_BASCO20	1279	1202	1578	1773	1796	13	4	0	6	0	24
	H_BASSI21	1531	1505	1760	1990	2016	2	2	0	9	0	21
	H_THRCY38	2899	2899	2899	2899	2899	2	2	0	3	1	14
	H_MYARA9	1131	1125	1367	1963	2268	8	2	0	9	0	16

	H_CHLCA22	938	935	1046	1152	1155	4	4	1	31	0	16
	H_CHLCA23	1202	1202	1202	1202	1202	1	1	0	13	0	31
	H_CHLCA24	1390	1385	1965	2692	2692	12	5	0	17	0	32
	H_CHLCA16	1052	956	1867	2975	2994	12	7	0	2	0	18
	H_CHLFL37	1444	1385	1940	2408	2456	3	2	0	6	0	35
	H_CYAVI13	2537	2537	2537	2537	2537	1	1	0	12	0	7
	H_DIGBR17	2023	1943	2802	3051	3079	23	2	0	2	0	15
	H_DIGBR19	2911	2911	2911	2911	2911	1	1	0	1	0	10
	H_DIGBR18	2959	2959	2959	2959	2959	1	1	0	2	0	11
	H_DIGBR31	2969	2969	2969	2969	2969	1	1	0	2	0	11
	H_DIGGL4	949	934	1088	1227	1242	2	2	0	13	0	16
	H_ELAPA12	2924	2924	2924	2924	2924	1	1	0	1	0	10
	H_EUPXA10	1485	1324	2420	2991	2997	3	2	0	2	0	38
	H_EUTCO1	1116	1115	1246	1457	1493	3	2	0	8	0	16
	H_HYLOC14	937	937	937	937	937	1	1	0	21	1	16
	H_MIOST8	939	936	1269	1632	1709	6	3	0	17	0	16
	H_MICLA2	1138	1138	1138	1138	1138	1	1	0	33	0	16
	H_MIOST7	2736	2736	2736	2736	2736	1	1	0	11	1	16
	H_PIPAR6	2549	2533	2659	2744	2749	3	1	1	11	0	7
	H_TANCH25	990	956	1245	1399	1399	5	3	0	9	0	15
	H_TANAR27	925	878	1238	1405	1407	5	3	0	10	0	16
	H_THREP30	1212	1202	1300	1389	1399	2	2	0	9	0	31
	H_TANPU29	1134	1127	1245	1386	1407	3	2	0	9	0	16
	H_THLRU26	1968	1968	1968	1968	1968	1	1	0	9	0	17
	H_THRCY28	2662	2662	2662	2662	2662	1	1	0	17	0	14
	H_ZONCA34	2988	2988	2988	2988	2988	1	1	0	2	0	11
Leucocytozoon	L_AMBHO132	2674	2674	2674	2674	2674	1	1	1	17	0	14
	L_ARETO103	2784	2782	2830	2930	2954	4	3	0	1	0	13
	L_ARETO59	2830	2802	2947	3001	3002	4	4	0	3	1	20
	L_ATLME77	2488	2460	2731	2960	2974	9	5	0	2	0	12
	L_ATLME80	2048	1976	2698	3000	3004	23	10	0	3	0	15
	L_BUTMO90	2458	1979	2803	3097	3377	16	12	0	2	0	12
	L_HEMAT126	1976	1968	2725	3005	3013	29	14	1	3	0	17
	L_DIGBR46	2982	2981	2998	3016	3018	3	3	0	3	0	11
	L_ARETO96	2787	2787	2787	2787	2787	1	1	0	11	0	13
	L_ARRTO113	2668	2667	2680	2691	2692	2	2	0	17	0	14
	L_ATLME100	2509	2466	2699	2925	2997	8	2	0	1	0	7
	L_ATLME133	2656	2656	2656	2656	2656	1	1	0	17	0	14
	L_ATLME138	2466	2466	2466	2466	2466	1	1	0	6	0	12
	L_ATLME141	2788	2788	2788	2788	2788	1	1	0	11	0	13
	L_ATLME17	2662	2662	2662	2662	2662	1	1	0	17	0	14
	L_ATLME40	2730	2730	2730	2730	2730	1	1	0	11	0	16
	L_ATLME41	2638	2637	2649	2660	2661	2	2	0	17	0	8
	L_ATLME49	1399	1137	2401	2991	2996	36	21	0	2	0	32
	L_ATLME53	2630	2630	2630	2630	2630	1	1	0	17	0	8
	L_ATLME83	2339	2339	2340	2341	2341	2	2	0	6	0	11
	L_ATLME99	2929	2929	2929	2929	2929	1	1	0	1	0	10
	L_BASLU104	2011	1973	2358	2709	2749	3	2	0	11	0	15
	L_CHLRI43	1571	1398	2362	2992	2999	7	7	0	2	0	17
	L_CHLRI54	2749	2749	2778	2827	2835	3	3	0	2	0	16

L_IRIJE30	1678	1398	2595	2955	2959	5	5	0	2	0	9
L_MYIME127	1972	1968	2552	2794	2802	11	7	1	11	0	17
L_SYNAZ125	2634	2626	2783	2976	2994	6	5	0	2	0	8
L_AULCO3	1528	1511	1719	1937	1965	3	3	0	9	0	21
L_AULCO4	1968	1968	1968	1968	1968	1	1	0	9	0	17
L_CHLRI29	2646	2631	2750	2830	2835	3	2	0	2	0	8
L_BUTMO19	3003	3003	3003	3003	3003	1	1	0	3	0	7
L_BUTMO20	2899	2899	2899	2899	2899	1	1	0	3	0	14
L_BUTMO58	2692	2692	2692	2692	2692	1	1	1	17	0	14
L_CACCH48	2261	2261	2261	2261	2261	1	1	0	9	0	5
L_DIGBR35	1341	1129	2337	3029	3056	6	4	0	3	0	38
L_CATDI139	2959	2959	2959	2959	2959	1	1	0	2	0	11
L_MYARA32	1416	1380	2307	2941	2968	5	5	0	1	0	35
L_CATFU24	1968	1968	1968	1968	1968	1	1	0	9	0	17
L_CHLRI76	2924	2924	2924	2924	2924	1	1	0	1	0	10
L_CHLRI81	2689	2689	2787	2953	2982	3	1	0	2	0	14
L_MECST6	1197	1134	2130	2982	2994	6	4	0	2	0	25
L_CONAR57	1303	1302	1336	1383	1386	5	2	0	9	0	38
L_CYAVI37	2522	2518	2727	2957	2999	5	1	0	2	0	7
L_CYAVI9	2997	2997	2997	2997	2997	1	1	0	2	1	11
L_DIGBR101	2899	2899	2899	2899	2899	1	1	0	3	0	14
L_DIGBR102	2969	2969	2969	2969	2969	1	1	0	2	0	11
L_DIGBR136	3056	3056	3056	3056	3056	1	1	0	2	0	18
L_DIGBR15	2733	2720	2846	2958	2971	2	1	0	2	0	16
L_DIGBR18	2957	2957	2957	2957	2957	1	1	0	2	0	11
L_DIGBR45	2750	2750	2750	2750	2750	1	1	1	11	0	16
L_DIGBR47	2970	2970	2970	2970	2970	1	1	0	2	0	11
L_DIGSI116	2159	1968	2764	2981	2983	5	2	0	2	0	6
L_DIGCY14	2756	2751	2796	2837	2842	2	2	0	2	0	13
L_DIGCY34	1399	1399	1399	1399	1399	1	1	0	9	0	32
L_DIGMY135	2938	2938	2938	2938	2938	1	1	0	1	0	10
L_DIGMY98	2995	2995	2995	2995	2995	1	1	0	2	0	11
L_ELAPA2	2755	2755	2755	2755	2755	1	1	0	11	0	13
L_ELAPA44	2750	2750	2750	2750	2750	1	1	0	11	0	16
L_ENTLE22	2021	2021	2021	2021	2021	1	1	0	11	0	15
L_ENTLE27	1976	1976	1976	1976	1976	1	1	0	9	0	17
L_ENTLE63	1139	1139	1418	1892	1976	3	1	0	5	0	16
L_EUBVE10	1968	1968	1968	1968	1968	1	1	0	9	1	17
L_GRAER1	1980	1980	1980	1980	1980	1	1	0	9	0	17
L_GRAFE13	2693	2688	2740	2788	2793	2	1	1	11	0	14
L_GRAFE5	2788	2788	2788	2788	2788	1	1	1	11	0	13
L_IRIAN86	1976	1976	1976	1976	1976	1	1	0	9	0	17
L_IRIJE95	2924	2924	2924	2924	2924	1	1	0	1	0	10
L_MIOST21	2689	2689	2689	2689	2689	1	1	0	17	1	14
L_PIPAR11	2533	2533	2533	2533	2533	1	1	0	12	0	7
L_PIPAR118	2749	2749	2749	2749	2749	1	1	1	11	0	16
L_PIPAR119	2695	2695	2695	2695	2695	1	1	1	17	0	14
L_PIPIN12	2751	2751	2751	2751	2751	1	1	0	11	1	13
L_PIPIN120	2141	2021	2634	2963	3004	5	1	0	2	0	9
L_PIPIN121	2758	2758	2758	2758	2758	1	1	0	11	1	13

	L_PIPIN137	2526	2526	2526	2526	2526	1	1	0	12	0	7
	L_SYNAZ38	2470	2470	2470	2470	2470	1	1	0	6	0	12
	L_TANVA97	2692	2692	2692	2692	2692	1	1	0	17	0	14
	L_THRBO16	2988	2988	2988	2988	2988	1	1	0	2	0	11
	L_THRBO39	2979	2979	2979	2979	2979	1	1	0	2	0	11
	L_THRCY140	2656	2656	2656	2656	2656	1	1	0	17	0	14
	L_THRCY52	2635	2635	2635	2635	2635	1	1	0	17	0	8
	L_THRCY87	2677	2662	2816	2955	2970	2	1	0	2	0	14
	L_TURCH25	2708	2667	2875	2975	2988	5	3	0	2	0	16
	L_TURCH26	2997	2997	2997	2997	2997	1	1	0	2	0	11
	L_TURCH68	2960	2954	3016	3073	3079	2	2	0	2	0	11
	L_TURCH70	1661	1138	2740	3246	3377	8	4	0	1	0	9
	L_TURFU64	2992	2987	3033	3074	3079	2	1	0	2	0	11
	L_TURIG65	1398	1398	1398	1398	1398	1	1	0	9	1	32
	L_TURSE28	2331	2331	2331	2331	2331	1	1	0	6	0	11
	L_TURSE67	2362	2341	2510	2629	2638	3	1	0	17	0	13
Total limiting lineages									13		11	

Table 5.6 Number of range limiting lineages when infection zones are classified as 100 m above and below parasite lineages maximum and minimum elevation records.

100 m above and below parasite lineage maximum and minimum elevation												
Genus	Lineage	Low CI	Low limit	Mean elevation	Up CI	Up limit	Infected birds	Infected bird spp.	Infected low limited bird spp.	Low limited bird spp.	Infected up limited bird spp.	Up limit bird spp.
Plasmodium	P_MYARA13	1399	1399	2843	3079	3079	40	15	0	1	0	24
	P_ARRTA9	300	300	300	300	300	1	1	1	83	0	0
	P_ARRTO12	2787	2787	2787	2787	2787	1	1	0	3	0	14
	P_AULPR10	936	936	936	936	936	1	1	0	31	0	9
	P_AUTRU15	882	882	919	956	956	2	2	0	28	0	7
	P_DIGCY21	2518	2518	2660	2802	2802	2	2	0	1	0	12
	P_DYSME16	926	926	944	956	956	4	1	0	28	0	9
	P_ENTLE4	1139	1139	1139	1139	1139	1	1	0	13	0	17
	P_CATUS1	1380	1380	1798	2266	2266	4	2	0	7	0	24
	P_MIOOL14	875	875	1082	1283	1283	6	6	0	9	0	7
	P_HYLNA18	930	930	930	930	930	1	1	0	31	0	9
	P_HYPSU3	1302	1302	1302	1302	1302	1	1	0	10	0	31
	P_MYCHI5	878	878	1422	2729	2729	24	7	0	2	0	7
	P_PHYOP7	874	874	1229	1398	1398	8	3	0	8	0	7
	P_PYRLE11	1308	1308	1308	1308	1308	1	1	0	10	0	31
	P_RAMFU8	300	300	300	300	300	1	1	1	83	0	0
	P_TURNI2	1138	1138	2426	3079	3079	9	5	0	1	0	17
	P_TROPE17	2350	2350	2350	2350	2350	1	1	0	6	0	5
	P_TURIG20	1398	1398	1398	1398	1398	1	1	0	8	0	24
Haemoproteus	H_THRCY32	2341	2341	2882	3117	3117	77	10	0	1	0	5
	H_IRIAN3	1242	1242	1925	2977	2977	15	6	0	2	0	16

	H_ATLME35	1125	1125	1608	2750	2750	16	5	0	3	0	17
	H_BASCO20	1202	1202	1578	1796	1796	13	4	0	5	0	16
	H_BASSI21	1505	1505	1760	2016	2016	2	2	0	8	0	35
	H_THRCY38	2899	2899	2899	2899	2899	2	2	0	2	0	13
	H_MYARA9	1125	1125	1367	2268	2268	8	2	0	7	0	17
	H_CHLCA22	935	935	1046	1155	1155	4	4	0	15	0	9
	H_CHLCA23	1202	1202	1202	1202	1202	1	1	0	9	0	16
	H_CHLCA24	1385	1385	1965	2692	2692	12	5	0	11	0	24
	H_CHLCA16	956	956	1867	2994	2994	12	7	0	2	0	14
	H_CHLFL37	1385	1385	1940	2456	2456	3	2	0	10	0	24
	H_CYAVI13	2537	2537	2537	2537	2537	1	1	0	17	0	12
	H_DIGBR17	1943	1943	2802	3079	3079	23	2	0	1	0	15
	H_DIGBR19	2911	2911	2911	2911	2911	1	1	0	3	0	20
	H_DIGBR18	2959	2959	2959	2959	2959	1	1	0	2	0	14
	H_DIGBR31	2969	2969	2969	2969	2969	1	1	0	2	0	14
	H_DIGGL4	934	934	1088	1242	1242	2	2	0	9	0	9
	H_ELAPA12	2924	2924	2924	2924	2924	1	1	0	3	0	20
	H_EUPXA10	1324	1324	2420	2997	2997	3	2	0	2	0	31
	H_EUTCO1	1115	1115	1246	1493	1493	3	2	0	15	0	17
	H_HYLOC14	937	937	937	937	937	1	1	0	31	0	9
	H_MIOST8	936	936	1269	1709	1709	6	3	0	2	0	9
	H_MICLA2	1138	1138	1138	1138	1138	1	1	0	13	0	17
	H_MIOST7	2736	2736	2736	2736	2736	1	1	0	2	1	8
	H_PIPAR6	2533	2533	2659	2749	2749	3	1	0	2	0	12
	H_TANCH25	956	956	1245	1399	1399	5	3	0	8	0	14
	H_TANAR27	878	878	1238	1407	1407	5	3	0	14	0	7
	H_THREP30	1202	1202	1300	1399	1399	2	2	0	8	0	16
	H_TANPU29	1127	1127	1245	1407	1407	3	2	0	14	0	17
	H_THLRU26	1968	1968	1968	1968	1968	1	1	0	8	0	14
	H_THRCY28	2662	2662	2662	2662	2662	1	1	0	11	0	8
	H_ZONCA34	2988	2988	2988	2988	2988	1	1	0	2	0	14
Leucocytozoon	L_AMBHO132	2674	2674	2674	2674	2674	1	1	1	11	0	8
	L_ARETO103	2782	2782	2830	2954	2954	4	3	0	2	0	14
	L_ARETO59	2802	2802	2947	3002	3002	4	4	0	2	0	16
	L_ATLME77	2460	2460	2731	2974	2974	9	5	0	2	0	13
	L_ATLME80	1976	1976	2698	3004	3004	23	10	0	2	0	14
	L_BUTMO90	1979	1979	2803	3377	3377	16	12	0	0	0	14
	L_HEMAT126	1968	1968	2725	3013	3013	29	14	1	2	0	14
	L_DIGBR46	2981	2981	2998	3018	3018	3	3	0	2	0	14
	L_ARETO96	2787	2787	2787	2787	2787	1	1	0	3	0	14
	L_ARRTO113	2667	2667	2680	2692	2692	2	2	0	11	0	8
	L_ATLME100	2466	2466	2699	2997	2997	8	2	0	2	0	13
	L_ATLME133	2656	2656	2656	2656	2656	1	1	0	11	0	8
	L_ATLME138	2466	2466	2466	2466	2466	1	1	0	10	0	13
	L_ATLME141	2788	2788	2788	2788	2788	1	1	0	3	0	14
	L_ATLME17	2662	2662	2662	2662	2662	1	1	0	11	0	8
	L_ATLME40	2730	2730	2730	2730	2730	1	1	0	2	0	8
	L_ATLME41	2637	2637	2649	2661	2661	2	2	0	11	0	7
	L_ATLME49	1137	1137	2401	2996	2996	36	21	0	2	0	17
	L_ATLME53	2630	2630	2630	2630	2630	1	1	0	11	0	7

L_ATLME83	2339	2339	2340	2341	2341	2	2	0	6	0	5
L_ATLME99	2929	2929	2929	2929	2929	1	1	0	3	0	20
L_BASLU104	1973	1973	2358	2749	2749	3	2	0	2	0	14
L_CHLRI43	1398	1398	2362	2999	2999	7	7	0	2	0	24
L_CHLRI54	2749	2749	2778	2835	2835	3	3	0	1	0	8
L_IRIJE30	1398	1398	2595	2959	2959	5	5	0	2	0	24
L_MYIME127	1968	1968	2552	2802	2802	11	7	0	1	0	14
L_SYNAZ125	2626	2626	2783	2994	2994	6	5	0	2	0	7
L_AULCO3	1511	1511	1719	1965	1965	3	3	0	8	1	35
L_AULCO4	1968	1968	1968	1968	1968	1	1	0	8	0	14
L_CHLRI29	2631	2631	2750	2835	2835	3	2	0	1	0	7
L_BUTMO19	3003	3003	3003	3003	3003	1	1	0	2	0	10
L_BUTMO20	2899	2899	2899	2899	2899	1	1	0	2	0	13
L_BUTMO58	2692	2692	2692	2692	2692	1	1	0	11	0	8
L_CACCH48	2261	2261	2261	2261	2261	1	1	1	7	0	6
L_DIGBR35	1129	1129	2337	3056	3056	6	4	0	1	0	17
L_CATDI139	2959	2959	2959	2959	2959	1	1	0	2	0	14
L_MYARA32	1380	1380	2307	2968	2968	5	5	0	2	0	24
L_CATFU24	1968	1968	1968	1968	1968	1	1	0	8	0	14
L_CHLRI76	2924	2924	2924	2924	2924	1	1	0	3	0	20
L_CHLRI81	2689	2689	2787	2982	2982	3	1	0	2	0	8
L_MECST6	1134	1134	2130	2994	2994	6	4	0	2	0	17
L_CONAR57	1302	1302	1336	1386	1386	5	2	0	8	0	31
L_CYAVI37	2518	2518	2727	2999	2999	5	1	0	2	0	12
L_CYAVI9	2997	2997	2997	2997	2997	1	1	0	2	1	14
L_DIGBR101	2899	2899	2899	2899	2899	1	1	0	2	0	13
L_DIGBR102	2969	2969	2969	2969	2969	1	1	0	2	0	14
L_DIGBR136	3056	3056	3056	3056	3056	1	1	0	1	0	11
L_DIGBR15	2720	2720	2846	2971	2971	2	1	0	2	0	8
L_DIGBR18	2957	2957	2957	2957	2957	1	1	0	2	0	14
L_DIGBR45	2750	2750	2750	2750	2750	1	1	0	3	0	14
L_DIGBR47	2970	2970	2970	2970	2970	1	1	0	2	0	14
L_DIGSI116	1968	1968	2764	2983	2983	5	2	0	2	0	14
L_DIGCY14	2751	2751	2796	2842	2842	2	2	0	1	0	14
L_DIGCY34	1399	1399	1399	1399	1399	1	1	0	8	0	24
L_DIGMY135	2938	2938	2938	2938	2938	1	1	0	3	0	20
L_DIGMY98	2995	2995	2995	2995	2995	1	1	0	2	0	14
L_ELAPA2	2755	2755	2755	2755	2755	1	1	0	3	0	14
L_ELAPA44	2750	2750	2750	2750	2750	1	1	0	3	0	14
L_ENTLE22	2021	2021	2021	2021	2021	1	1	0	8	0	18
L_ENTLE27	1976	1976	1976	1976	1976	1	1	0	8	0	14
L_ENTLE63	1139	1139	1418	1976	1976	3	1	0	8	0	17
L_EUBVE10	1968	1968	1968	1968	1968	1	1	0	8	0	14
L_GRAER1	1980	1980	1980	1980	1980	1	1	0	8	0	14
L_GRAFE13	2688	2688	2740	2793	2793	2	1	0	3	0	8
L_GRAFE5	2788	2788	2788	2788	2788	1	1	0	3	0	14
L_IRIAN86	1976	1976	1976	1976	1976	1	1	0	8	0	14
L_IRIJE95	2924	2924	2924	2924	2924	1	1	0	3	0	20
L_MIOST21	2689	2689	2689	2689	2689	1	1	0	11	1	8
L_PIPAR11	2533	2533	2533	2533	2533	1	1	1	17	0	12

L_PIPAR118	2749	2749	2749	2749	2749	1	1	0	2	0	8
L_PIPAR119	2695	2695	2695	2695	2695	1	1	1	11	0	8
L_PIPIN12	2751	2751	2751	2751	2751	1	1	0	3	0	14
L_PIPIN120	2021	2021	2634	3004	3004	5	1	0	2	0	18
L_PIPIN121	2758	2758	2758	2758	2758	1	1	0	3	0	14
L_PIPIN137	2526	2526	2526	2526	2526	1	1	0	17	0	12
L_SYNAZ38	2470	2470	2470	2470	2470	1	1	0	10	0	13
L_TANVA97	2692	2692	2692	2692	2692	1	1	0	11	0	8
L_THRBO16	2988	2988	2988	2988	2988	1	1	0	2	0	14
L_THRBO39	2979	2979	2979	2979	2979	1	1	0	2	0	14
L_THRCY140	2656	2656	2656	2656	2656	1	1	0	11	0	8
L_THRCY52	2635	2635	2635	2635	2635	1	1	0	11	0	7
L_THRCY87	2662	2662	2816	2970	2970	2	1	0	2	0	8
L_TURCH25	2667	2667	2875	2988	2988	5	3	0	2	0	8
L_TURCH26	2997	2997	2997	2997	2997	1	1	0	2	0	14
L_TURCH68	2954	2954	3016	3079	3079	2	2	0	1	0	14
L_TURCH70	1138	1138	2740	3377	3377	8	4	0	0	0	17
L_TURFU64	2987	2987	3033	3079	3079	2	1	0	1	0	14
L_TURIG65	1398	1398	1398	1398	1398	1	1	0	8	0	24
L_TURSE28	2331	2331	2331	2331	2331	1	1	0	6	0	5
L_TURSE67	2341	2341	2510	2638	2638	3	1	0	11	0	5
Total limiting lineages								7		4	

Table 5.7 Genbank accession numbers for avian malaria parasite lineages recorded from sampled bird species in the Kosnipata valley, Peru.

No.	Lineage	Accession No.	No.	Lineage	Accession No.	No.	Lineage	Accession No.
1	P_TURNI2	KF874666	**52**	H_DIGGL4	KF874717	**103**	L_GRAFE13	KF874768
2	P_ENTLE4	KF874667	**53**	L_THRBO39	KF874718	**104**	L_ELAPA2	KF874769
3	P_DIGCY21	KF874668	**54**	H_EUPXA10	KF874719	**105**	L_ATLME83	KF874770
4	P_TURIG20	KF874669	**55**	H_TANCH25	KF874720	**106**	L_EUBVE10	KF874771
5	P_HYLNA18	KF874670	**56**	H_IRIAN3	KF874721	**107**	L_DIGMY98	KF874772
6	H_BASSI21	KF874671	**57**	H_BASCO20	KF874722	**108**	L_GRAFE5	KF874773
7	H_CHLFL37	KF874672	**58**	H_DIGBR19	KF874723	**109**	L_DIGMY135	KF874774
8	H_CYAVI13	KF874673	**59**	H_THRCY28	KF874724	**110**	H_THRCY32	KF874775
9	H_DIGBR31	KF874674	**60**	H_DIGBR18	KF874725	**111**	L_ATLME77	KF874776
10	H_CHLCA23	KF874675	**61**	H_DIGBR17	KF874726	**112**	L_ATLME80	KF874777
11	H_MIOST8	KF874676	**62**	L_CYAVI37	KF874727	**113**	L_ATLME138	KF874778
12	P_AULPR10	KF874677	**63**	H_THLRU26	KF874728	**114**	L_HEMAT126	KF874779
13	P_TROPE17	KF874678	**64**	H_THREP30	KF874729	**115**	L_BUTMO90	KF874780
14	H_ZONCA34	KF874679	**65**	L_CATUS73	KF874730	**116**	L_AMBHO93	KF874781
15	H_CHLCA22	KF874680	**66**	L_ATLME141	KF874731	**117**	L_ARETO60	KF874782
16	P_ARRTO12	KF874681	**67**	L_ATLME53	KF874732	**118**	L_DIGBR18	KF874783
17	P_AUTRU15	KF874682	**68**	L_ATLME40	KF874733	**119**	L_BUTMO20	KF874784
18	P_HYPSU3	KF874683	**69**	P_MYCHI5	KF874734	**120**	L_BASLU105	KF874785
19	P_PYRLE11	KF874684	**70**	L_DIGBR46	KF874735	**121**	L_ATLME100	KF874786
20	P_MIOOL14	KF874685	**71**	L_DIGCY34	KF874736	**122**	L_DIGBR101	KF874787
21	P_PHYOP7	KF874686	**72**	L_SYNAZ38	KF874737	**123**	L_IRIAN86	KF874788
22	P_MYARA13	KF874687	**73**	L_DIGBR45	KF874738	**124**	L_ARRTO113	KF874789
23	P_CATUS1	KF874688	**74**	L_CHLRI76	KF874739	**125**	L_BASLU104	KF874790
24	P_DYSME16	KF874689	**75**	L_PIPIN120	KF874740	**126**	L_ENTLE27	KF874791
25	H_ELAPA12	KF874690	**76**	L_CATUS72	KF874741	**127**	L_TANVA97	KF874792
26	H_PIPAR6	KF874691	**77**	L_CHLRI43	KF874742	**128**	L_PIPAR11	KF874793
27	H_TANAR27	KF874692	**78**	L_BUTMO58	KF874743	**129**	L_PIPIN12	KF874794
28	L_THRCY52	KF874693	**79**	L_CHLRI54	KF874744	**130**	L_THRCY87	KF874795
29	L_ARETO59	KF874694	**80**	L_CONAR57	KF874745	**131**	L_PIPAR119	KF874796
30	L_MYARA32	KF874695	**81**	L_CHLRI29	KF874746	**132**	L_PIPIN121	KF874797
31	L_THRCY140	KF874696	**82**	L_CACCH48	KF874747	**133**	L_TURFU64	KF874798
32	L_IRIJE30	KF874697	**83**	L_ELAPA44	KF874748	**134**	L_AMBHO131	KF874799
33	L_DIGBR35	KF874698	**84**	L_THRBO16	KF874749	**135**	L_TURSE67	KF874800
34	H_MIOST7	KF874699	**85**	L_CYAVI9	KF874750	**136**	L_TURIG65	KF874801
35	H_EUTCO1	KF874700	**86**	L_DIGBR15	KF874751	**137**	L_TURCH70	KF874802
36	L_PIPAR118	KF874701	**87**	L_CATFU24	KF874752	**138**	L_ATLME133	KF874803
37	H_MICLA2	KF874702	**88**	L_MYIME127	KF874753	**139**	L_ATLME41	KF874804
38	H_XIPTR11	KF874703	**89**	L_AMBHO132	KF874754	**140**	L_ATLME99	KF874805
39	H_CHLCA16	KF874704	**90**	L_DIGBR136	KF874755	**141**	L_ARETO103	KF874806
40	P_ARRTA9	KF874705	**91**	L_CATDI139	KF874756	**142**	L_ARETO96	KF874807
41	P_RAMFU8	KF874706	**92**	L_DIGCY14	KF874757	**143**	L_TURCH26	KF874808
42	H_MYARA9	KF874707	**93**	L_AULCO3	KF874758	**144**	L_ATLME17	KF874809
43	H_HYLOC14	KF874708	**94**	L_MECST6	KF874759	**145**	L_MIOST21	KF874810
44	H_ATLME35	KF874709	**95**	L_AULCO4	KF874760	**146**	L_CHLRI81	KF874811
45	H_TANPU29	KF874710	**96**	L_IRIJE95	KF874761	**147**	L_DIGSI116	KF874812
46	H_THRCY38	KF874711	**97**	L_CATUS23	KF874762	**148**	L_BUTMO19	KF874813
47	H_CHLCA24	KF874712	**98**	L_CATUS75	KF874763	**149**	L_PIPIN137	KF874814
48	L_ATLME49	KF874713	**99**	L_GRAER1	KF874764	**150**	L_DIGBR102	KF874815
49	L_AMBHO107	KF874714	**100**	L_CATUS74	KF874765	**151**	L_TURCH25	KF874816
50	L_ENTLE63	KF874715	**101**	L_SYNAZ125	KF874766	**152**	L_TURCH68	KF874817
51	L_DIGBR47	KF874716	**102**	L_ENTLE22	KF874767	**153**	L_TURSE28	KF874818