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RESEARCH ARTICLE

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An inverse latitudinal gradient in infection probability and phylogenetic diversity for *Leucocytozoon* blood parasites in New World birds

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Abstract

1. Geographic variation in environmental conditions as well as host traits that promote parasite transmission may impact infection rates and community assembly of vector-transmitted parasites.
2. Identifying the ecological, environmental and historical determinants of parasite distributions and diversity is therefore necessary to understand disease outbreaks under changing environments. Here, we identified the predictors and contributions of infection probability and phylogenetic diversity of *Leucocytozoon* (an avian blood parasite) at site and species levels across the New World.
3. To explore spatial patterns in infection probability and lineage diversity for *Leucocytozoon* parasites, we surveyed 69 bird communities from Alaska to Patagonia. Using phylogenetic Bayesian hierarchical models and high-resolution satellite remote-sensing data, we determined the relative influence of climate, landscape, geography and host phylogeny on regional parasite community assembly.

4. Infection rates and parasite diversity exhibited considerable variation across regions in the Americas. In opposition to the latitudinal gradient hypothesis, both the diversity and prevalence of *Leucocytozoon* parasites decreased towards the equator. Host relatedness and traits known to promote vector exposure neither predicted infection probability nor parasite diversity. Instead, the probability of a bird being infected with *Leucocytozoon* increased with increasing vegetation cover (NDVI) and moisture levels (NDWI), whereas the diversity of parasite lineages decreased with increasing NDVI. Infection rates and parasite diversity also tended to be higher in cooler regions and higher latitudes.
5. Whereas temperature partially constrains *Leucocytozoon* diversity and infection rates, landscape features, such as vegetation cover and water body availability, play a significant role in modulating the probability of a bird being infected. This suggests that, for *Leucocytozoon*, the barriers to host shifting and parasite host range expansion are jointly determined by environmental filtering and landscape, but not by host phylogeny. Our results show that integrating host traits, host ancestry, bioclimatic data and microhabitat characteristics that are important for vector reproduction are imperative to understand and predict infection prevalence and diversity of vector-transmitted parasites. Unlike other vector-transmitted diseases, our results show that *Leucocytozoon* diversity and prevalence will likely decrease with warming temperatures.

KEYWORDS
community assembly, latitudinal diversity gradient, macroecology, NDVI, parasite distribution, parasite diversity, phylogenetic diversity

1 | INTRODUCTION

Understanding the geographic distribution, host range expansion and infection prevalence of pathogens under climate change and across heterogeneous landscapes has become a central goal in disease ecology (Parratt, Numminen, & Laine, 2016; Stephens et al., 2016). Yet, for the infectious agents causing a multitude of human and zoonotic diseases, the climatic, ecological and historical determinants of diversity, geographical distribution and host usage are not resolved (Stephens et al., 2016). This lack of basic knowledge presents a clear impediment to understanding when and where a pathogen will emerge, thus limiting our ability to predict and prevent future disease outbreaks.

Climate change impacts parasite infection rates because spatiotemporal variation in host community structure, caused by shifts in temperature and precipitation, alters host–parasite contact rates (Canard et al., 2014). Host traits that limit infection can also fluctuate in response to environmental conditions (Joseph, Stutz, & Johnson, 2016). Warming temperatures, for instance, can alter the phenology of avian hosts by changing the onset of their breeding period (Walther et al., 2002). Therefore, environmental changes can have important consequences on host–pathogen interactions and may increase the spread of infectious diseases, especially for those pathogens and parasites transmitted by hematophagous dipteran insects as they are dependent on water and temperature for reproduction and host-seeking activity.

Geographical variation in infection prevalence for many vector-borne parasites is influenced not only by climate heterogeneity but also by local landscape factors (Haque et al., 2010; Woolhouse et al., 1997). Vectors' breeding sites are heterogeneously distributed across the landscape; thus, accurate characterization of aquatic habitat availability (potential breeding sites) across host communities is useful for predicting the spatial distribution of vector-transmitted parasites. Water bodies and microhabitat (e.g. vegetation cover) availability induce local geographic reproduction of vectors, both of which may trigger changes in pathogen transmission dynamics (Lafferty, 2009; Santiago-Alarcon, Palinauskas, & Schaefer, 2012). Detecting landscape correlates using high-resolution satellite remote-sensing services is becoming an increasingly important step towards understanding variation in parasite infection probabilities (Pullan et al., 2011; Soares Magalhães, Clements, Patil, Gething, & Brooker, 2011).

Though free-living organisms exhibit macroecological patterns such as the marked increase in species richness from high to low latitudes, known as the latitudinal diversity gradient (hereafter 'LDG'; Hillebrand, 2004; Pianka, 1966; Rohde, 1992), a recent meta-analysis showed that this phenomenon does not hold true for parasitic organisms (Kamiya, O'Dwyer, Nakagawa, & Poulin, 2014). In free-living

organisms, the LDG may be caused by multiple factors including climate stability in tropical zones, lower rates of diversification in temperate zones, ecological interactions, variation in size of continental landmasses and increased productivity near the equator (Mittelbach et al., 2007; Pianka, 1966; Rohde, 1992). Studies of spatial patterns of parasite diversity are less common and have found conflicting results (Bordes, Morand, Krasnov, & Poulin, 2010). When there is support for the LDG among parasite taxa, variation in host species richness is usually recognized as the explanatory factor for the pattern (Poulin, 2007, 2014). The absence of a latitudinal gradient for endoparasitic helminths of mammals and birds (Poulin, 1995) might be explained by the relative stability of their environment, since host internal body temperature is relatively constant (Rohde & Heap, 1998). In contrast, parasites such as those in the order Haemosporida that are vectored by blood-feeding insects are exposed to and dependent on external environmental conditions (temperature and precipitation) that change with latitude. However, the only two studies to test LDG for haemosporidian parasites in bird communities did not find a significant relationship between parasite diversity in temperate Chile (Merino et al., 2008) or across a global scale (Clark, 2018).

Haemosporidians (Phylum Apicomplexa, Order Haemosporida) are protozoan parasites that infect vertebrate blood cells and are transmitted by hematophagous dipteran vectors (Santiago-Alarcon et al., 2012; Valkiūnas, 2005). Birds host the highest species diversity of haemosporidian parasites, which are traditionally placed within three genera: *Haemoproteus* (containing two distinct subgenera *Haemoproteus* and *Parahaemoproteus*), *Leucocytozoon* and *Plasmodium* (Valkiūnas, 2005). *Plasmodium* is widely distributed in vertebrate hosts with either nucleated (birds and reptiles) or anucleated (mammals) red blood cells. In contrast, *Haemoproteus* and *Leucocytozoon* exclusively infect birds (Valkiūnas, 2005).

Historically, the genus *Leucocytozoon* has been vastly undersampled, with limited information on prevalence, distribution, diversity, taxonomy and its relationships with avian and vector hosts (Fecchio et al., 2018; Galen, Nunes, Sweet, & Perkins, 2018; Lotta et al., 2016; Lutz et al., 2015; Valkiūnas, 2005). It has been recently suggested that high temperature is an environmental filter for *Leucocytozoon* that might constrain parasite prevalences and distributions in lowland South America (Fecchio et al., 2018). This hypothesis was based on the scarcity of *Leucocytozoon* infections reported in Brazilian lowlands (Fecchio et al., 2018), whereas higher prevalence has been reported in the tropical Andes, especially above 2,000 m (Galen & Witt, 2014; Harrigan et al., 2014; Lotta et al., 2016), where the annual average temperature is lower compared to lowland tropical sites. Fecchio et al. (2018) linked the scarcity of *Leucocytozoon* infections with warmer temperatures in the tropical lowlands rather than a lack of transmission opportunity, as the vectors for this genus (black flies in the family Simuliidae) are abundant and diverse in lowland regions. Nevertheless, some studies have found strong support for the relationship between prevalence of *Leucocytozoon* and altitude (Barrow et al., 2019; Galen & Witt, 2014; González et al., 2014; Harrigan et al., 2014; Lotta et al., 2016) or latitude (Merino et al., 2008). However, the combined effect of latitude and altitude that ultimately reflects a gradient of temperature, precipitation and host diversity have never been tested for *Leucocytozoon* infections across the New World (but see Barrow et al., 2019 for a local analysis across the Tropical Andes of Peru).

Here, we used data on the prevalence and distribution of *Leucocytozoon* parasites to explore macroecological patterns in diversity and infection rates across Neotropical and Nearctic bird communities. Using phylogenetic Bayesian hierarchical models, we asked whether patterns of parasite diversity and probability of infection across the New World are driven by site-level (climate, latitude, altitude and landscape) or host species-level predictors (host phylogenetic relationships, diet, height of foraging and sex). We predicted that *Leucocytozoon* infection probability would be higher in regions with lower temperature (Fecchio et al., 2018; Valkiūnas, 2005). If parasite host shifting decreases continuously with increasing phylogenetic distance between its original host and potential new hosts (Clark & Clegg, 2017; Poulin, Krasnov, & Mouillot, 2011), we would expect similar infection rates among closely related host species. We also expect higher parasite infection probabilities at sites closer to water bodies and increased vegetation cover, as these landscape factors might be correlated with higher vector abundance (Oakgrove et al., 2014). Collectively, our analyses identify the predictors and contributions of climate, geography, landscape and host ancestry to regional community assembly and infection rates of *Leucocytozoon*, one of the most prevalent parasites of birds in the New World.

2 | MATERIALS AND METHODS

2.1 | Sample collection

We collected 6,293 blood and tissue samples from 909 avian species across the New World. Our sampling included 69 bird communities surveyed across 12 bioregions in South America (Argentina, Brazil, Chile, Peru), Central America (Honduras, Nicaragua) and North America (Mexico and the United States of America) (Figure 1). The majority of samples were collected during the summer from the period of 2010 to 2017. Netted birds were bled by brachial venipuncture, and blood was collected in heparinized capillary tubes. After blood collection, birds were either ringed and released or killed and prepared as museum specimens. Birds not likely to be captured with mist nets were collected with firearms in Brazil, Honduras and Peru. Liver samples were taken during specimen preparation and stored in liquid nitrogen or RNAlater until DNA extraction. Blood samples were stored in 95% ethanol or on FTA cards. All tissue samples and birds were collected in accordance with appropriate permits in each of the eight countries listed above.

2.2 | Molecular detection of parasites

DNA was extracted from bird tissues using the Qiagen DNeasy 96 Blood and Tissue kit (Qiagen), following the Qiagen tissue protocol

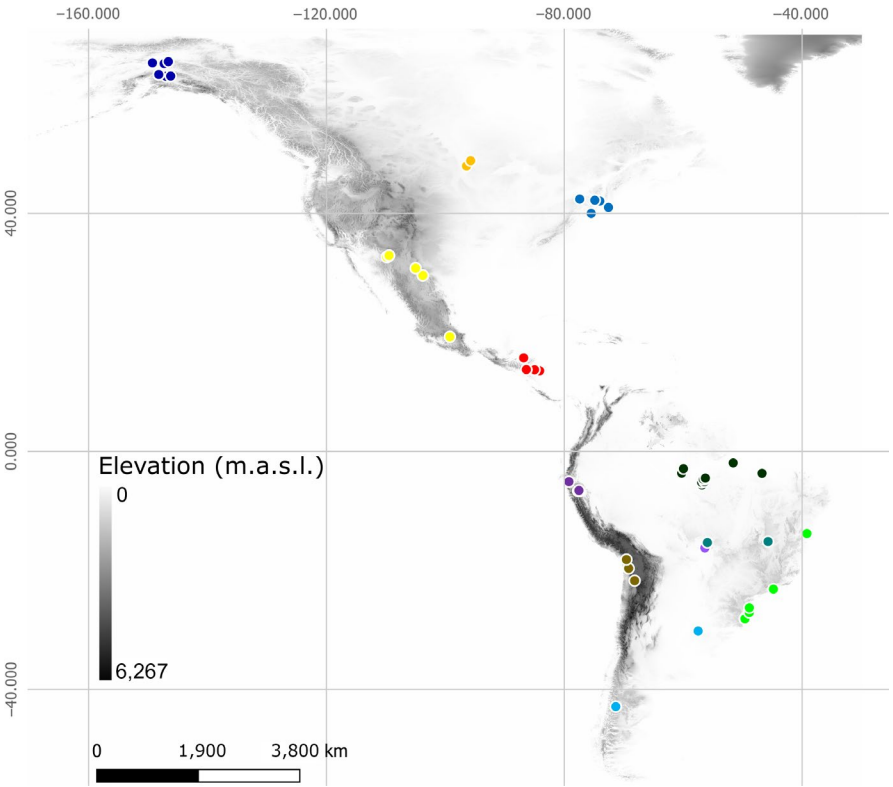

FIGURE 1 Sampling sites distributed across elevational and latitudinal gradients in the New World. Colours depict the site classification into one of 12 biogeographical regions denoted in Table 1

for both blood and tissue stored in 95% ethanol. For most samples, the protocols of Bell, Weckstein, Fecchio, and Tkach (2015) were followed to both initially screen samples for haemosporidian DNA with real-time PCR and then to amplify a 477-bp region of the *Leucocytozoon* cytochrome *b* gene (cyt-*b*) from positive samples using nested PCR. Samples from Alaska and Minnesota were screened and amplified following the protocols of Hellgren, Waldenström, and Bensch (2004), which amplifies the same 477-bp region of cyt-*b*. All PCR amplifications included both negative and positive controls. All PCR products were run on 1.25% agarose gels, stained with ethidium bromide, visualized under UV light and photographed. Positive PCR products were purified using ExoSAP-IT (Affymetrix) and sequenced using BigDye terminator v3.1 cycle sequencing kit (Applied Biosystems) on ABI 1300 and 3100 DNA sequencers (Applied Biosystems).

Sequence identities were verified with a local BLAST against the MalAvi database (Bensch, Hellgren, & Pérez-Tris, 2009) using BioEdit v7.2.0 (Hall, 1999). Given the evidence indicating that avian haemosporidian haplotypes differing by one cyt-*b* nucleotide may be reproductively isolated entities (Bensch, Pérez-Tris, Waldenström, & Hellgren, 2004), we followed the conventional practice of referring to each unique cyt-*b* haplotype as a unique parasite lineage. New lineages were named after the host of origin following a standard protocol (Bensch et al., 2009), and DNA sequences of all new lineages were deposited within GenBank (MG714922-MG714926, MG726098-MG726173, MK947467-MK947908, MK972879-MK972904) and the MalAvi database (http://mbio-serv2.mbioekol.lu.se/Malavi/).

2.3 | Site-level altitude, climate and landscape descriptors

A primary goal of this study was to identify important correlates of avian individuals' probabilities of carrying *Leucocytozoon* parasites. For each of the 69 sampling sites in our dataset, we extracted altitude as well as several long-term climate variables that could relate to *Leucocytozoon* infection prevalence by influencing the distributions and densities of vectors (see Appendix S1). Previous studies have found evidence that temperature and precipitation variables, particularly those that capture variation in summer and winter climates, may be linked to important aspects of haemosporidian community assembly and transmission (Clark et al., 2018; Fecchio, Wells, et al., 2019; Oakgrove et al., 2014; Sehgal et al., 2011). We included maximum temperature of the warmest month, maximum rainfall of the wettest month, minimum temperature of the coldest month and minimum rainfall of the driest month as predictors in our analysis. Each of these variables was downloaded from Worldclim (http://worldclim.org/version2), a database of interpolated climate layers that describe long-term average conditions (from the years 1970 to 2000).

We extracted remote-sensed measurements for two variables that reflect local variation in vegetation, moisture and the presence of water bodies, all of which can impact the densities and distributions of simuliid black flies that act as vectors for *Leucocytozoon* parasites (Oakgrove et al., 2014; Santiago-Alarcon et al., 2012; Sehgal, 2015). These variables were Normalized Difference Vegetation Index (NDVI) and Normalized Difference Water Index (NDWI),

both obtained at 10 km × 10 km resolution using functions in the MODISTools R package (Tuck et al., 2014). This package accesses remote-sensing images from NASA's MODerate-resolution Imaging Spectroradiometer (MODIS) satellite (Justice et al., 1998). To ensure that these variables accurately reflected conditions when vectors were likely to be active, we calculated mean NDVI and NDWI values for each site across the primary growing season. For the Northern Hemisphere, this included values from May to August (inclusive), and for the Southern Hemisphere, we used values from December to March (inclusive).

2.4 | Ecological and phylogenetic relationships of avian hosts

Avian host species-level covariates that may influence susceptibility to vectorborne blood parasites were extracted from the EltonTraits v1.0 database (Wilman et al., 2014). The selected traits included the relative proportions of time species spend foraging in lower and upper level canopies as well as species' average body masses. In addition to these ecological traits, we extracted data on species' evolutionary relationships from an open-source phylogenetic supertree that includes all extant avian species (Jetz, Thomas, Joy, Hartmann, & Mooers, 2012). To capture uncertainty in avian host phylogenetic relationships, we downloaded 100 possible tree topologies from the supertree's 'Ericsson All Species' Bayesian posterior distribution (available at Birdtree.org/subsets/).

2.5 | Phylogenetic logistic regressions to predict infection probability

We filtered the dataset to only include species for which at least ten individual birds had been sampled for parasites. This allowed us to focus on potential host species for which we had high confidence of detecting parasites, even when the true background infection prevalence was low (e.g. restricting the analysis to species with at least 10 individuals results in a ~70% probability of detecting infections when the true prevalence is just 10%). This filtering resulted in a dataset with 3,938 individual birds sampled from 155 different species.

To quantify factors associated with variation in *Leucocytozoon* infection probability among birds sampled in the Americas, we used a phylogenetic generalized linear model fitted in a Bayesian framework. We assumed the observed presence–absence of parasite infection Inf(*i*) from host individual *i* at sampling site *s* was a random Bernoulli draw contingent on surrounding environmental conditions, host phylogenetic ancestry and species-level ecological variables:

$$\text{Inf}(i) \sim \text{Bernoulli}[\Psi(i)] \quad (1)$$

Using a logit link function, we modelled the infection probability $\Psi(i)$ of each host individual using a hierarchical linear regression of the form:

$$\text{logit}(\Psi(i)) = \alpha(i) + \Phi(i) + \beta ET(i,s) \quad (2)$$

Here, α represents a non-phylogenetic species-level random intercept, which we implemented to capture aspects of avian species susceptibility that were not described by the selected ecological variables or by avian phylogenetic relationships. Φ represents a phylogenetic random term capturing evolutionary relationships among avian host species. This term was drawn from a zero-centred multivariate normal distribution that was parameterized with an inverse phylogenetic variance–covariance (VCV) matrix Ω:

$$\Phi(i) \sim \text{MVN}[0, \Omega] \quad (3)$$

This multivariate random effect models the assumption that closely related avian species may harbour similar infection rates. To assess the effect of phylogenetic uncertainty on our estimates, we followed de Villemereuil, Wells, Edwards, and Blomberg (2012) by placing a categorical prior distribution on the set of phylogenetic VCV matrices, allowing the sampler to randomly select one of these matrices (with equal probabilities) in each iteration. This allowed for more precise estimation of regression coefficients than using a single consensus tree and was considered more appropriate than running sequential models across different trees. Finally, the β coefficients in Equation (2) describe effects of 12 additional species-level and site-level covariates on each bird's infection probability. Chosen species-level variables included sex (binary), body mass (continuous), lower canopy affinity (continuous) and upper canopy affinity (continuous). We used these host traits because they are known to influence haemosporidian diversity and prevalence and are available in open-source data repositories. The predicted effects of these traits specifically on *Leucocytozoon* infection have been debated previously (Barrow et al., 2019; Lutz et al., 2015). As sex information was missing for 1,424 birds, we imputed these missing values using Bernoulli draws with a prior belief of $p = .5$. Site-level predictor variables were as follows: altitude, latitude (absolute value), maximum temperature of the warmest month, maximum rainfall of the wettest month, minimum temperature of the coldest month, minimum rainfall of the driest month, mean NDVI and mean NDWI (all site-level variables were continuous and scaled to unit variance prior to analysis to facilitate direct comparisons of coefficients).

We fitted the model and estimated coefficients using Markov chain Monte Carlo (MCMC) sampling with the Gibbs sampler provided in the open-source software JAGS (Plummer, 2003). Priors for species-level non-phylogenetic intercepts (α) were sampled from normal distributions with mean = 0 and $SD = 10$ (see Appendices S1 and S2 for model specifications in JAGS language). To reduce the model's parameter space and identify the most important predictors, we implemented a form of Gibbs variable selection (GVS) when estimating β coefficients that ensured effects were shrunk towards zero when evidence for their inclusion in the model was limited (Fecchio, Wells, et al., 2019; O'Hara & Sillanpää, 2009). Our GVS method implemented a spike and slab prior of the form $P(\beta|ind) = N(0, SD = 10)(1 - ind) + N(0, SD = 0.01)(ind)$, where variable selection indicator (ind) was a Bernoulli draw with a low prior belief of inclusion ($p \sim .2$). We used vague normal priors for estimates when variables were

selected for inclusion (e.g. we sampled from the 'slab' when *ind* = 1). The pseudo-priors (sampled from the 'spike' when *ind* = 0) forced estimates towards zero when they received little empirical support, while still allowing the sampler to efficiently move between the distributions (Wells, Lakim, & O'Hara, 2014). This method had the added benefit that posterior distributions of GVS indicators were reflective of each variable's relative importance. To allow for variation in latitudinal infection gradients between the two hemispheres, as has been detected previously for a number of ecological phenomena (Chown, Sinclair, Leinaas, & Gaston, 2004; Zhang, Zhang, & Ma, 2016), we allowed the latitude β coefficient to vary among the Northern and Southern Hemispheres.

We ran two MCMC chains for a burn-in period of 100,000 iterations and gathered 1,000 parameter estimates. Mixing of chains was assessed both visually and with the Gelman–Rubin diagnostic (all values were <1.2). A posterior predictive check was used to assess whether our model's assumptions yielded good approximations of the data generating process. Here, Bayesian *p*-values ~ .5 indicate a lack of discrepancy between predictions and observed values (indicating good fit), while values near 0 or 1 indicate poor fit (Gelman, Meng, & Stern, 1996).

2.6 | Phylogenetic signal and comparison with a non-phylogenetic model

We estimated phylogenetic signal in species' infection probabilities by calculating Pagel's lambda (λ; Pagel, 1999). Following Hadfield and Nakagawa (Hadfield & Nakagawa, 2010), we estimated λ as the proportion of variance that was attributed to variance in phylogenetic intercepts (Φ). To describe how aspects of species identity that were not captured by our chosen traits or by phylogenetic relationships may contribute to infection probability, we also estimated the proportion of variance that was attributed to non-phylogenetic species intercepts (α).

Initial models showed that estimates of λ were very low and that non-phylogenetic intercepts accounted for the majority of variance in species' average infection rats (see Results below). In addition, convergence for the phylogenetic model was difficult to reach (Appendix S2). As a result, we fit an equivalent model that did not include the phylogenetic error term. Run parameters and convergence assessments were the same for this model (Appendix S2).

2.7 | Latitudinal variation in parasite phylogenetic diversity

We tested whether phylogenetic diversity of *Leucocytozoon* parasite communities showed patterns consistent with the LDG hypothesis. All *Leucocytozoon* cytochrome *b* sequences that overlapped with a 477 bp in length fragment were aligned using Bioedit (Hall, 1999). This alignment of 216 unique parasite sequences was used to reconstruct their phylogenetic relationships. Parasite phylogenies were estimated using Bayesian inference in the program BEAST v. 1.8.4 (Drummond, Suchard, Xie, & Rambaut, 2012). Run parameters were as follows: the GTR + I + G model of evolution, a Yule process tree prior and a log-normal relaxed clock using the substitution rate of 0.006 per lineage per million years (Ricklefs & Outlaw, 2010). We ran 100 million generations of MCMC (Markov chain Monte Carlo) sampled every 5,000 generations, discarding 10% of generations as burn-in to generate a posterior distribution of 18,001 total trees.

Parasite phylogenetic diversity of each sampling region was calculated by generating a region by parasite count matrix and generating an abundance-weighted and sample-size-corrected estimate of diversity using functions available in the iNextPD R package (Hsieh & Chao, 2017). This method uses an integration of rarefaction and extrapolation techniques to account for variation in sample size and sample completeness among sampling sites, both of which are known to have large influences on resulting estimates of diversity. As above, we accounted for phylogenetic uncertainty in our diversity estimates by repeating calculations across a distribution of 100 parasite trees that were randomly drawn (without replacement) from the phylogenetic posterior distribution.

We used a hierarchical linear regression to test the LDG hypothesis. Our outcome of interest was standardized phylogenetic diversity for each region. However, because several regions harboured low parasite recoveries, interpolation estimates of diversity were not possible. We excluded these regions (*N* = 3; Amazonia, Atacama Desert and Cerrado), which resulted in a vector of 900 total diversity estimates for analysis (nine regional estimates across 100 different parasite trees). Rather than simply using mean values for predictors, we accounted for observed variation in latitudes, altitudes and landscape descriptors across sampling sites in each region by including these predictors as latent variables in our model. This was done by calculating the means and standard deviations for each quantity and using these values to parameterize normal distributions from which each of the 100 observations per region was drawn (see Appendix S3 for model specifications in JAGS language). For example, if a region's mean altitude was 50 m and the *SD* was 10 m, we drew our 100 altitude observations from the distribution *N*(50, 10). Predictors included in the model were latitude (absolute value), altitude, NDVI and NDWI. We did not include climate variables because Clark (2018) found no influence of climate variation on parasite phylogenetic diversity. We included a random intercept term for region to capture region-level variation that was not described by our chosen predictors.

Priors for regression coefficients in our linear model were specified as vague normal distributions (see Appendix S3 for model specifications in JAGS language). We ran two MCMC chains for a burn-in period of 75,000 iterations and gathered 1,000 parameter estimates. Convergence and posterior predictive checks were assessed as above. For all parameters from both models, we report posterior modes and 95% highest posterior density credible intervals. All analyses were conducted in R and primarily made use of the following packages: DPLYR (Wickham & Francois, 2017), RJAGS (Plummer, 2003) and READXL

(Wickham & Bryan, 2017). R code to replicate predictor variable extraction and regression analyses are supplied in Appendices S1-S3.

3 | RESULTS

3.1 | Predictors of infection risk

3.1.1 | Species-level

In total, 747 out of 6,293 individual birds were infected with *Leucocytozoon* (overall prevalence of 12%, 129 host species, see raw data in Appendix S4). There was wide variation in our estimates of average prevalence (ranging from zero to 100%) for the 155 well-sampled bird species (minimum 10 individuals screened per species; Appendix S5). We found little evidence that host phylogenetic relationships played a role in modulating *Leucocytozoon* infection risk (95% CI for λ estimates: 0.01, 0.05; Appendix S2). Instead, the non-phylogenetic intercepts captured the majority of species-level variance (95% CI of species vs. phylo terms: 0.66, 0.94). As a result, and because convergence for the phylogenetic model was difficult to reach (Appendix S2), we report results from an equivalent model that did not include the phylogenetic error term. From this model, we found that infection probability did not correlate with any of the species-level ecological variables (sex, body mass, canopy affinity; GVS indicators for each of these variables were below 0.1, suggesting they were readily excluded; Appendix S2), which suggests that most of the variation in prevalence across species could be determined by the species-level random intercept terms.

3.1.2 | Site level

The prevalence of *Leucocytozoon* was heterogeneous across the 69 bird communities and among the 12 biogeographical regions (ranging from zero to 84%; Table 1). Among the eight landscape variables that could explain such heterogeneity, only maximum temperature of the warmest month, NDVI, NDWI and Latitude was found to be important site-level predictors (GVS indicator variables for these three covariates were all above 0.6; Figure 2). Infection was more likely to occur in areas with cooler summers (95% CI for maximum temperature of the warmest month: −1.52, −0.26), with higher growing season vegetation cover (95% CI for NDVI: 0.23, 0.95), with higher growing season moisture content (95% CI for NDWI: 0.09, 0.89) and at higher absolute latitudes (95% CI for Latitude: 0.98, 3.11). There was no evidence that latitudinal patterns differed across hemispheres (95% CIs for these estimates strongly overlapped; see Appendix S2). By using the standard deviations of maximum temperature of the warmest month and Latitude, we calculated effect sizes (NDVI and NDWI are standardized, unitless indices and so calculating an effect sizes does not help with interpretation). We estimated that a five-degree increase in maximum temperature of the warmest month resulted in a 4-fold decrease in a bird's infection probability, whereas an increase in absolute Latitude of 10 degrees resulted in a 10-fold increase in infection probability.

3.2 | Predictors of parasite diversity

Among the nine regions with at least six infections recorded, NDVI and Latitude were the strongest predictors of parasite community phylogenetic diversity, collectively capturing ~99% of the explained variation (Figure 3). There was a trend of increasing diversity at higher absolute latitudes, though two regions (Mesoamerica and Prairie Parkland) did not follow this trend (Appendix S6). Communities were also found to be less diverse with increasing growing season vegetation cover (NDVI), though this relationship was comparatively weak and the regions of Southwest and Temperate Forest appeared to be outliers (Appendix S7). We used prediction heatmaps to further visualize these estimated trends (Figure 4). Here, it was clear

TABLE 1 Sample sizes (sites, individual birds, host species and parasite lineages found), number of infections and prevalence for the 12 biogeographical regions

Biogeographical region	# sites	Mean latitude	Mean longitude	# Bird species	# Bird individuals	# Infections	# Lineages	Prevalence (%)
Boreal Forest	9	64	−147.87	37	397	335	72	84.4
Prairie Parkland	2	48.26	−96.22	27	228	51	9	22.4
Temperate Forest	6	40.23	−75.32	81	1,287	225	96	17.5
Southwest	14	24.05	−102.31	57	136	10	5	7.4
Mesoamerica	6	14.17	−85.7	160	470	16	14	3.4
Amazonia	10	−4.21	−54.46	244	1,239	5	1	0.4
Huallaga	4	−6.45	−77.75	270	1,173	13	12	1.1
Cerrado	2	−15.3	−53.44	34	147	0	0	0.0
Pantanal	1	−16.25	−56.37	57	122	10	7	8.2
Atacama Desert	3	−19.7	−69.07	18	68	2	3	2.9
Atlantic Forest	10	−22.2	−45.13	87	341	16	7	4.7
Andean–Patagonian Forest and Espinal	2	−38.92	−67	80	685	64	28	9.3

Note: Colour coding corresponds to sampling localities in Figure 1.

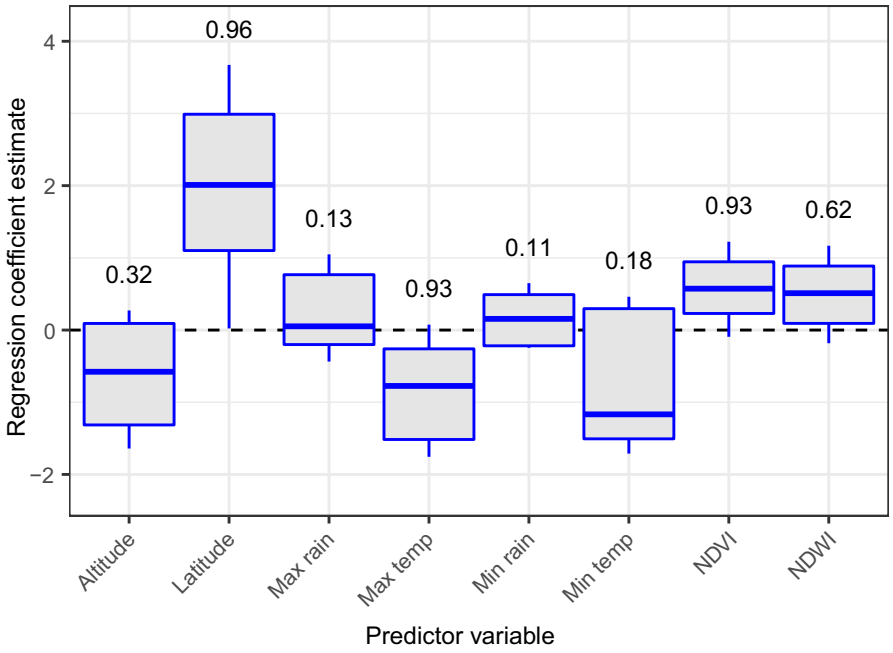

FIGURE 2 Regression coefficient estimates for site-level predictors of a bird's probability of being infected with *Leucocytozoon* parasites in the New World. Boxplots represent posterior modes (lines), 95% highest posterior density credible intervals (hinges) and minimums and maximums (whiskers). Numbers above boxes represent the mean Gibbs variable selection indicator for each variable, which indicates the proportion of iterations in which the variable was included in the model (higher values indicate stronger support that a particular variable improves the model likelihood)

that Latitude was found to be a stronger predictor of community diversity.

4 | DISCUSSION

Our models revealed that host ecological traits and phylogenetic relationships played no role in modulating the probability of a bird being infected with *Leucocytozoon* in the New World. Rather, the heterogeneity in infection rates was driven by predictors at the site level, with higher probabilities of infection in areas at higher absolute Latitudes, cooler summers, with higher cover of vegetation (NDVI) and with higher moisture levels (NDWI). Latitude and NDVI were also the strongest predictors of *Leucocytozoon* phylogenetic diversity across the New World, with a trend of increasing parasite diversity at higher absolute Latitudes and with decreasing average vegetation cover. These results are in opposition with the assumption that infection rates and parasite diversity are higher in tropical habitats or towards the equator.

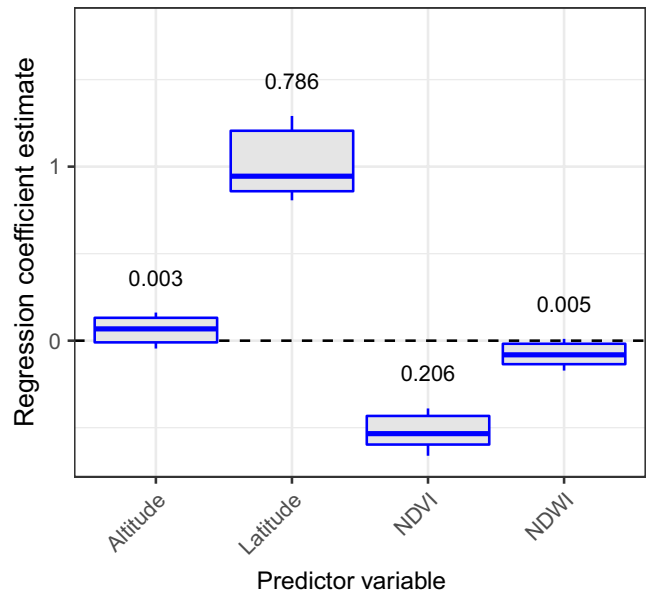

FIGURE 3 Regression coefficient estimates for site-level predictors of *Leucocytozoon* parasite community phylogenetic diversity in the New World. Boxplots represent posterior modes (lines), 95% highest posterior density credible intervals (hinges) and minimums and maximums (whiskers). Numbers above boxes represent the proportions of variance in estimated community diversity that were explained by each variable

The highest probability of a bird being infected with *Leucocytozoon* was consistently found in regions with cooler summers and towards the pole, demonstrating that high temperature appears to be an important environmental barrier for *Leucocytozoon* distribution. Previous studies have shown that climate has little or no influence on the diversity and distributions of two related genera (*Plasmodium* and *Haemoproteus*) in both Nearctic (Ellis et al., 2015) and Neotropical birds (Fecchio, Bell, et al., 2019), whereas temperature explained haemosporidian prevalence (including *Leucocytozoon*) in Arctic birds (Oakgrove et al., 2014). Although *Plasmodium* prevalence and distribution are constrained by low temperature due to thermal limits on parasite development within mosquito vectors (LaPointe, Goff, & Atkinson, 2010), *Leucocytozoon* might be constrained by high temperature (Fecchio et al., 2018). Experimental infections under laboratory conditions will be necessary to confirm whether high temperature is a limiting factor for *Leucocytozoon* parasite development within their vectors, which ultimately affects prevalence and distributions among their avian hosts, thus constraining lineage diversity across biogeographical regions. Nevertheless, higher prevalence of *Leucocytozoon* in boreal (Galen et al., 2018; Oakgrove et al., 2014; this study) and temperate regions (Merino et al., 2008, this study) supports the idea that *Leucocytozoon* parasites (and

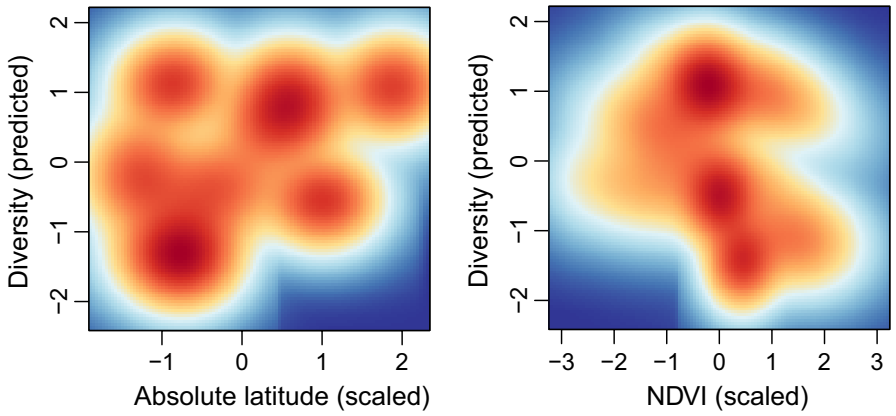

FIGURE 4 Bivariate prediction heatmaps representing posterior predictions of standardized parasite community phylogenetic diversity across regions ordered by absolute latitude (left-hand plot) and Normalized Difference Vegetation Index (NDVI) (right-hand plot). Cooler blue colours indicate fewer predictions occurred within a particular multivariate space, while warmer reds indicate higher densities of posterior predictions. Collectively, Latitude and NDVI accounted for >99% of explained variance in community phylogenetic diversity

perhaps their vectors) may be better adapted for development and transmission in colder environments (Valkiūnas, 2005).

Contrary to previous studies demonstrating an increase of prevalence with increasing elevation across the tropical Andes (Barrow et al., 2019; Galen & Witt, 2014; González et al., 2014; Harrigan et al., 2014; Lotta et al., 2016), we have shown no effect of altitude on infection probability and phylogenetic diversity of *Leucocytozoon* across bird communities at a continental scale. This pattern may be driven by the scarcity of *Leucocytozoon* infections in the highland Atlantic Forest and the moderate prevalence of *Leucocytozoo*n in some low-elevation sites such as the Pantanal and temperate forest. Therefore, differences in altitude seem to not capture variation in the required combinations of environmental determinants of *Leucocytozoon* prevalence when latitude is taken into consideration at a larger scale. Moreover, there is no strong evidence that the density nor diversity of black flies (the main vectors for *Leucocytozoon* parasites) follows an altitudinal gradient (Currie & Adler, 2008; McCreadie, Adler, & Hamada, 2005).

Host ecological attributes and phylogenetic relatedness would be expected to promote host range expansion in parasitic organisms (Clark & Clegg, 2017; Poulin et al., 2011) and thus determine the parasite prevalence and diversity across host communities. However, for multi-host parasites, such as *Leucocytozoon*, the parasite host range might be decoupled from vertebrate host phylogenetic signal (Galen, Speer, & Perkins, 2019), as parasite sharing does not occur necessarily between avian taxa that share a common ancestor, but instead through cross-species transmission between unrelated host taxa mediated via vectors. Blackflies have been shown to harbour high diversities of *Leucocytozoon* parasites within a single species (Murdock, Adler, Frank, & Perkins, 2015), supporting the idea that vectors may play important roles in facilitating transmission of *Leucocytozoon* infections across distantly related bird species. Furthermore, many parasites and pathogens can infect unrelated host species with similar ecological niches, such as habitat use, nesting behaviour and diet (Clark et al., 2018; Poulin, 2007). Whereas these host traits are often phylogenetically conserved, they may also be influenced by environmental conditions. Therefore, rather than host ecology and evolution, regional climate determined primarily by temperature seems to have an important impact on *Leucocytozoon* host range expansion as demonstrated recently for its two related genera (Fecchio, Wells, et al., 2019).

Our hierarchical linear regression clearly demonstrates a positive relationship between parasite diversity and Latitude across New World bird communities, which is in opposition with the LDG hypothesis and contrasts with previous studies showing that species richness increases towards the equator in some parasitic and pathogenic organisms (Fecchio, Bell, et al., 2019; Guernier, Hochberg, & Guégan, 2004; Nunn, Altizer, Sechrest, & Cunningham, 2005; Rohde & Heap, 1998). Whether or not pathogens consistently obey this macroecological phenomenon, as observed for a multitude of free-living organisms, has received conflicting support and a growing body of evidence supports a lack of an effect of latitude on species richness, abundance, diversity or prevalence for parasitic organisms (Clark, 2018; Kamiya et al., 2014; Poulin, 1995; Rohde & Heap, 1998). Because the distribution of parasites is largely dependent on the distribution of their hosts, one might expect that parasitic organisms would adhere to the LDG patterns by following the diversity of their hosts. The inverse patterns in diversity between *Leucocytozoon* (higher towards Poles) and avian hosts (higher towards Equator; Duchêne & Cardillo, 2015) in the New World suggest that parasite phylogenetic diversity is driven by the diversity of its black fly vectors (higher richness in temperate streams than in tropical streams; McCreadie et al., 2005).

Infection prevalence and diversity of *Leucocytozoon* were not solely predicted by Latitude and its effects on bioclimatic conditions. Local landscape features, such as NDVI and NDWI, are known to capture microhabitat characteristics that are important for vector reproduction and therefore should explain prevalence, and possibly diversity, of blood parasites (Pullan et al., 2011; Sehgal et al., 2011; Soares Magalhães et al., 2011). Whereas NDVI was consistently found to be an important predictor of *Leucocytozoon* community assembly and infection rates across the New World (this study) and Alaska (Oakgrove et al., 2014), NDWI only influenced prevalence.

This reinforces that NDWI may not accurately reflect breeding site availability for vector-transmitted parasites and pathogens. Black flies (the vectors for *Leucocytozoon*) are dependent on the flowing streams created by sustained snow melt in temperate and boreal regions. In these regions, sufficient snow melt would produce the flowing small streams that these vectors need for reproduction in areas with limited elevation gradient. This lack of cold water and associated high dissolved oxygen needed for larval development and vector reproduction in lowland tropical sites may explain the low prevalence and lineage diversity in low latitudes.

Quality data on vector biology are missing for most avian haemosporidian parasites, but this is especially true for *Leucocytozoon*. The simuliid vectors of *Leucocytozoon* (Valkiūnas, 2005) show unique life-history characteristics as compared to the vectors of *Plasmodium* or *Parahaemoproteus*, which should affect their ecological distribution and evolutionary history in ways that differ from *Plasmodium* or *Parahaemoproteus*. Unlike the vectors of *Plasmodium* (mosquitoes) and *Parahaemoproteus* (biting midges), the vectors of *Leucocytozoon* (black flies) are diurnal feeders; therefore, *Leucocytozoon* transmission occurs during the day when most birds are active. Since most birds rest at night, mosquitoes and midges have good opportunities to take blood without encountering a host defensive reaction. In contrast, black flies must contend with feeding on fully awake birds. However, the window of opportunity for diurnal blood feeding during the summer months is much wider in temperate zones (i.e. long days and short nights) than in tropical zones where the seasonal variation in photoperiod is more uniform. Because of the extended feeding time, the per capita blood-feeding success of black flies in temperate biomes would be proportionally higher than in tropical biomes. More blood meals would yield greater likelihood of transmission and, in turn, yield higher infection risk in the local bird populations in the temperate and boreal regions.

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AUTHORS' CONTRIBUTIONS

A.F. and N.J.C. conceived the ideas and designed methodology; A.F., J.A.B., M.B., V.V.T., H.L.L., V.R.C., C.A.G., D.G.A., J.A.V., C.S., D.K., K.J.M.C., K.K., J.B.P., J.B., C.S.F., K.Z.,. S.S., J.H.D., S.C.G. and J.D.W. conducted field/laboratory research; N.J.C. analysed the data; A.F. and N.J.C. led the writing of the manuscript with significant contributions from J.A.B.. All authors contributed critically to the drafts and gave final approval for publication.

DATA AVAILABILITY STATEMENT

The raw dataset and R code to replicate analyses are available in Figshare Digital Repository at: https://doi.org/10.6084/m9.figshare.9916328.v2 (Clark, 2019). All DNA sequences are available in GenBank (Accession numbers MG714922-MG714926, MG726098-MG726173, MK947467-MK947908).

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REFERENCES

Barrow, L. N., McNew, S. M., Mitchell, N., Galen, S. C., Lutz, H. L., Skeen, H., … Witt, C. C. (2019). Deeply conserved susceptibility in a multihost, multi-parasite system. *Ecology Letters*, *22*(2), 987–998. https://doi.org/10.1111/ele.13263

Bell, J. A., Weckstein, J. D., Fecchio, A., & Tkach, V. V. (2015). A new real-time PCR protocol for detection of avian haemosporidians. *Parasites & Vectors*, *8*, 383. https://doi.org/10.1186/s13071-015-0993-0

Bensch, S., Hellgren, O., & Pérez-Tris, J. (2009). MalAvi: A public database of malaria parasites and related haemosporidians in avian hosts based on mitochondrial cytochrome b lineages. *Molecular Ecology Resources*, *9*(5), 1353–1358. https://doi.org/10.1111/j.1755-0998.2009.02692.x

Bensch, S., Pérez-Tris, J., Waldenström, J., & Hellgren, O. (2004). Linkage between nuclear and mitochondrial DNA sequences in avian malaria parasites: Multiple cases of cryptic speciation? *Evolution*, *58*(7), 1617–1621. https://doi.org/10.1554/04-026

Bordes, F., Morand, S., Krasnov, B. R., & Poulin, R. (2010). Parasite diversity and latitudinal gradients in terrestrial mammals. In S. Morand, & B. R. Krasnov (Eds.), *The biogeography of host-parasite interactions* (pp. 89–98). New York, NY: Oxford University Press.

Canard, E. F., Mouquet, N., Mouillot, D., Stanko, M., Miklisova, D., & Gravel, D. (2014). Empirical evaluation of neutral interactions in host-parasite networks. *American Naturalist*, *183*(4), 468–479. https://doi.org/10.1086/675363

Chown, S. L., Sinclair, B. J., Leinaas, H. P., & Gaston, K. J. (2004). Hemispheric asymmetries in biodiversity-a serious matter for ecology. *PLoS Biology*, *2*(11), e406. https://doi.org/10.1371/journal.pbio.0020406

Clark, N. J. (2018). Phylogenetic uniqueness, not latitude, explains the diversity of avian blood parasite communities worldwide. *Global Ecology and Biogeography*, *27*(6), 744–755. https://doi.org/10.1111/geb.12741

Clark, N. (2019). Data from: An inverse latitudinal gradient in infection probability and phylogenetic diversity for *Leucocytozoon* blood parasites in New World birds. *Figshare Digital Repository*, https://doi.org/10.6084/m9.figshare.9916328.v2

Clark, N. J., & Clegg, S. M. (2017). Integrating phylogenetic and ecological distances reveals new insights into parasite host-specificity. *Molecular Ecology*, *26*(11), 3074–3086. https://doi.org/10.1111/mec.14101

Clark, N. J., Clegg, S. M., Sam, K., Goulding, W., Koane, B., & Wells, K. (2018). Climate, host phylogeny and the connectivity of host communities govern regional parasite assembly. *Diversity and Distributions*, *24*(1), 13–23. https://doi.org/10.1111/ddi.12661

Currie, D. C., & Adler, P. H. (2008). Global diversity of black flies (Diptera: Simuliidae) in freshwater. *Hydrobiologia*, *595*, 469–475. https://doi.org/10.1007/s10750-007-9114-1

de Villemereuil, P., Wells, J. A., Edwards, R. D., & Blomberg, S. P. (2012). Bayesian models for comparative analysis integrating phylogenetic uncertainty. *BMC Evolutionary Biology*, *12*, 102. https://doi.org/10.1186/1471-2148-12-102

Drummond, A. J., Suchard, M. A., Xie, D., & Rambaut, A. (2012). Bayesian phylogenetics with BEAUti and the BEAST 1.7. *Molecular Biology and Evolution*, *29*(8), 1969–1973. https://doi.org/10.1093/molbev/mss075

Duchêne, D. A., & Cardillo, M. (2015). Phylogenetic patterns in the geographic distributions of birds support the tropical conservatism hypothesis. *Global Ecology and Biogeography*, *24*(11), 1261–1268. https://doi.org/10.1111/geb.12370

Ellis, V. A., Collins, M. D., Medeiros, M. C. I., Sari, E. H. R., Coffey, E. D., Dickerson, R. C., … Ricklefs, R. E. (2015). Local host specialization, host-switching, and dispersal shape the regional distributions of avian haemosporidian parasites. *Proceedings of the National Academy of Sciences of the United States of America*, *112*(36), 11294–11299. https://doi.org/10.1073/pnas.1515309112

Fecchio, A., Bell, J. A., Pinheiro, R. B. P., Cueto, V. R., Gorosito, C. A., Lutz, H. L., … Collins, M. D. (2019). Avian host composition, local speciation, and dispersal drive the regional assembly of avian malaria parasites in South American birds. *Molecular Ecology*, *28*(10), 2681–2693. https://doi.org/10.1111/mec.15094

Fecchio, A., Silveira, P., Weckstein, J. D., Dispoto, J. H., Anciães, M., Bosholn, M., … Bell, J. A. (2018). First record of *Leucocytozoon* (Haemosporida: Leucocytozoidae) in Amazonia: Evidence for rarity in Neotropical lowlands or lack of sampling for this parasite genus? *The Journal of Parasitology*, *104*(2), 168–172. https://doi.org/10.1645/17-182

Fecchio, A., Wells, K., Bell, J. A., Tkach, V. V., Lutz, H. L., Weckstein, J. D., … Clark, N. J. (2019). Climate variation influences host specificity in avian malaria parasites. *Ecology Letters*, *22*(3), 547–557. https://doi.org/10.1111/ele.13215

Galen, S. C., Nunes, R., Sweet, P. R., & Perkins, S. L. (2018). Integrating coalescent species delimitation with analysis of host specificity reveals extensive cryptic diversity despite minimal mitochondrial divergence in the malaria parasite genus *Leucocytozoon*. *BMC Evolutionary Biology*, *18*, 128. https://doi.org/10.1186/s12862-018-1242-x

Galen, S. C., Speer, K. A., & Perkins, S. L. (2019). Evolutionary lability of host associations promotes phylogenetic overdispersion of co-infecting blood parasites. *Journal of Animal Ecology*, 1–14. https://doi.org/10.1111/1365-2656.13089

Galen, S. C., & Witt, C. C. (2014). Diverse avian malaria and other haemosporidian parasites in Andean house wrens: Evidence for regional co-diversification by host-switching. *Journal of Avian Biology*, *45*(4), 374–386. https://doi.org/10.1111/jav.00375

Gelman, A., Meng, X.-L., & Stern, H. (1996). Posterior predictive assessment of model fitness via realized discrepancies. *Statistica Sinica*, *6*(4), 733–760.

González, A. D., Matta, N. E., Ellis, V. A., Miller, E. T., Ricklefs, R. E., & Gutiérrez, H. R. (2014). Mixed species flock, nest height, and elevation partially explain avian haemoparasite prevalence in Colombia. *PLoS ONE*, *9*(6), e100695. https://doi.org/10.1371/journal.pone.0100695

Guernier, V., Hochberg, M. E., & Guégan, J.-F. (2004). Ecology drives the worldwide distribution of human diseases. *PLoS Biology*, *2*(6), e141. https://doi.org/10.1371/journal.pbio.0020141

Hadfield, J. D., & Nakagawa, S. (2010). General quantitative genetic methods for comparative biology: Phylogenies, taxonomies and multi-trait models for continuous and categorical characters. *Journal of Evolutionary Biology*, *23*(3), 494–508. https://doi.org/10.1111/j.1420-9101.2009.01915.x

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.

Haque, U., Magalhães, R. J. S., Reid, H. L., Clements, A. C. A., Ahmed, S. M., Islam, A., … Glass, G. E. (2010). Spatial prediction of malaria prevalence in an endemic area of Bangladesh. *Malaria Journal*, *9*, 120. https://doi.org/10.1186/1475-2875-9-120

Harrigan, R. J., Sedano, R. E., Chasar, A. C., Chaves, J. A., Nguyen, J. T., Whitaker, A., & Smith, T. C. (2014). New host and lineage diversity of avian haemosporidia in the northern Andes. *Evolutionary Applications*, *7*(7), 799–811. https://doi.org/10.1111/eva.12176

Hellgren, O., Waldenström, J., & Bensch, S. (2004). A new PCR assay for simultaneous studies of *Leucocytozoon*, *Plasmodium*, and *Haemoproteus* from avian blood. *Journal of Parasitology*, *90*(4), 797–802. https://doi.org/10.1645/GE-184R1

Hillebrand, H. (2004). On the generality of the latitudinal diversity gradient. *The American Naturalist*, *163*(2), 192–211. https://doi.org/10.1086/381004

Hsieh, T. C., & Chao, A. (2017). Rarefaction and extrapolation: Making fair comparison of abundance-sensitive phylogenetic diversity among multiple assemblages. *Systematic Biology*, *66*(1), 100–111. https://doi.org/10.1093/sysbio/syw073

Jetz, W., Thomas, G. H., Joy, J. B., Hartmann, K., & Mooers, A. O. (2012). The global diversity of birds in space and time. *Nature*, *491*, 444–448. https://doi.org/10.1038/nature11631

Joseph, M. B., Stutz, W. E., & Johnson, P. T. J. (2016). Multilevel models for the distribution of hosts and symbionts. *PLoS ONE*, *11*(11), e0165768. https://doi.org/10.1371/journal.pone.0165768

Justice, C. O., Vermote, E., Townshend, J., Defries, R., Roy, D. P., Hall, D. K., … Barnsley, M. J. (1998). The Moderate Resolution Imaging Spectroradiometer (MODIS): Land remote sensing for global change research. *IEEE Transactions on Geoscience and Remote Sensing*, *36*, 1228–1249. https://doi.org/10.1109/36.701075

Kamiya, T., O'Dwyer, K., Nakagawa, S., & Poulin, P. (2014). What determines species richness of parasitic organisms? A meta-analysis across animal, plant and fungal hosts. *Biological Reviews*, *89*(1), 123–134. https://doi.org/10.1111/brv.12046

Lafferty, K. D. (2009). The ecology of climate change and infectious diseases. *Ecology*, *90*(4), 888–900. https://doi.org/10.1890/08-0079.1

LaPointe, D. A., Goff, M. L., & Atkinson, C. T. (2010). Thermal constraints to the sporogonic development and altitudinal distribution of avian

malaria *Plasmodium relictum* in Hawai'i. *The Journal of Parasitology*, *96*(2), 318–324. https://doi.org/10.1645/GE-2290.1

Lotta, I. A., Pacheco, M. A., Escalante, A. A., González, A. D., Mantilla, J. S., Moncada, L. I., … Matta, N. E. (2016). *Leucocytozoon* diversity and possible vectors in the Neotropical highlands of Colombia. *Protist*, *167*(2), 185–204. https://doi.org/10.1016/j.protis.2016.02.002

Lutz, H. L., Hochachka, W. M., Engel, J. I., Bell, J. A., Tkach, V. V., Bates, J. M., … Weckstein, J. D. (2015). Parasite prevalence corresponds to host life history in a diverse assemblage of Afrotropical birds and haemosporidian parasites. *PLoS ONE*, *10*(4), e0121254. https://doi.org/10.1371/journal.pone.0121254

McCreadie, J. W., Adler, P. H., & Hamada, N. (2005). Patterns of species richness for blackflies (Diptera: Simuliidae) in the Nearctic and Neotropical regions. *Ecological Entomology*, *30*(2), 201–209. https://doi.org/10.1111/j.0307-6946.2005.00681.x

Merino, S., Moreno, J., Vásquez, R. A., Martínez, J., Sánchez-monsálvez, I., Estades, C. F., … Mcgehee, S. (2008). Haematozoa in forest birds from southern Chile: Latitudinal gradients in prevalence and parasite lineage richness. *Austral Ecology*, *33*(3), 329–340. https://doi.org/10.1111/j.1442-9993.2008.01820.x

Mittelbach, G. G., Schemske, D. W., Cornell, H. V., Allen, A. P., Brown, J. M., Bush, M. B., … Turelli, M. (2007). Evolution and the latitudinal diversity gradient: Speciation, extinction and biogeography. *Ecology Letters*, *10*(4), 315–331. https://doi.org/10.1111/j.1461-0248.2007.01020.x

Murdock, C. C., Adler, P. H., Frank, J., & Perkins, S. L. (2015). Molecular analyses of host-seeking black flies (Diptera: Simuliidae) reveal a diverse assemblage of *Leucocytozoon* (Apicomplexa: Haemospororida) parasites in an alpine ecosystem. *Parasites & Vectors*, *8*, 343. https://doi.org/10.1186/s13071-015-0952-9

Nunn, C. L., Altizer, S. M., Sechrest, W., & Cunningham, A. A.. (2005). Latitudinal gradients of parasite species richness in primates. *Diversity and Distributions*, *11*(3), 249–256. https://doi.org/10.1111/j.1366-9516.2005.00160.x

Oakgrove, K. S., Harrigan, R. J., Loiseau, C., Guers, S., Seppi, B., & Sehgal, R. N. M. (2014). Distribution, diversity and drivers of blood-borne parasite co-infections in Alaskan bird populations. *International Journal for Parasitology*, *44*(10), 717–727. https://doi.org/10.1016/j.ijpara.2014.04.011

O'Hara, R. B., & Sillanpää, M. J. (2009). A review of Bayesian variable selection methods: What, how and which. *Bayesian Analysis*, *4*(1), 85–117. https://doi.org/10.1214/09-BA403

Pagel, M. (1999). Inferring the historical patterns of biological evolution. *Nature*, *401*, 877–884. https://doi.org/10.1038/44766

Parratt, S. R., Numminen, E., & Laine, A.-L. (2016). Infectious disease dynamics in heterogeneous landscapes. *Annual Review of Ecology, Evolution, and Systematics*, *47*(1), 283–306. https://doi.org/10.1146/annurev-ecolsys-121415-032

Pianka, E. R. (1966). Latitudinal gradients in species diversity: A review of concepts. *The American Naturalist*, *100*(910), 33–46. https://doi.org/10.1086/282398

Plummer, M. (2003). JAGS: A program for analysis of Bayesian graphical models using Gibbs sampling. In: *Proceedings of the 3rd international workshop on distributed statistical computing* (p. 125).

Poulin, R. (1995). Phylogeny, ecology, and the richness of parasite communities in vertebrates. *Ecological Monographs*, *65*(3), 283–302. https://doi.org/10.2307/2937061

Poulin, R. (2007). *Evolutionary ecology of parasites* (2nd ed.). Princeton, NJ: Princeton University Press.

Poulin, R. (2014). Parasite biodiversity revisited: Frontiers and constraints. *International Journal for Parasitology*, *44*(9), 581–589. https://doi.org/10.1016/j.ijpara.2014.02.003

Poulin, R., Krasnov, B. R., & Mouillot, D. (2011). Host specificity in phylogenetic and geographic space. *Trends in Parasitology*, *27*(8), 355–361. https://doi.org/10.1016/j.pt.2011.05.003

Pullan, R. L., Gething, P. W., Smith, J. L., Mwandawiro, C. S., Sturrock, H. J. W., Gitonga, C. W., … Brooker, S. (2011). Spatial modelling of soil-transmitted Helminth infections in Kenya: A disease control planning tool. *PLoS Neglected Tropical Diseases*, *5*(2), e958. https://doi.org/10.1371/journal.pntd.0000958

Ricklefs, R. E., & Outlaw, D. C. (2010). A molecular clock for malaria parasites. *Science*, *329*(5988), 226–229. https://doi.org/10.1126/science.1188954

Rohde, K. (1992). Latitudinal gradients in species diversity: The search for the primary cause. *Oikos*, *65*(3), 514–527. https://doi.org/10.2307/3545569

Rohde, K., & Heap, M. (1998). Latitudinal differences in species and community richness and in community structure of metazoan endo- and ectoparasites of marine teleost fish. *International Journal for Parasitology*, *28*(3), 461–474. https://doi.org/10.1016/S0020-7519(97)00209-9

Santiago-Alarcon, D., Palinauskas, V., & Schaefer, H. M. (2012). Diptera vectors of avian Haemosporidian parasites: Untangling parasite life cycles and their taxonomy. *Biological Reviews*, *87*(4), 928–964. https://doi.org/10.1111/j.1469-185X.2012.00234.x

Sehgal, R. N. M. (2015). Manifold habitat effects on the prevalence and diversity of avian blood parasites. *International Journal for Parasitology: Parasites and Wildlife*, *4*(3), 421–430. https://doi.org/10.1016/j.ijppaw.2015.09.001

Sehgal, R. N. M., Buermann, W., Harrigan, R. J., Bonneaud, C., Loiseau, C., Chasar, A., … Smith, T. B. (2011). Spatially explicit predictions of blood parasites in a widely distributed African rainforest bird. *Proceedings of the Royal Society B: Biological Sciences*, *278*(1708), 1025–1033. https://doi.org/10.1098/rspb.2010.1720

Soares Magalhães, R. J., Clements, A. C. A., Patil, A. P., Gething, P. W., & Brooker, S. (2011). The applications of model-based geostatistics in helminth epidemiology and control. *Advances in Parasitology*, *74*, 267–296. https://doi.org/10.1016/B978-0-12-385897-9.00005-7

Stephens, P. R., Altizer, S., Smith, K. F., Alonso Aguirre, A., Brown, J. H., Budischak, S. A., … Poulin, R. (2016). The macroecology of infectious diseases: A new perspective on global-scale drivers of pathogen distributions and impacts. *Ecology Letters*, *19*(9), 1159–1171. https://doi.org/10.1111/ele.12644

Tuck, S. L., Phillips, H. R. P., Hintzen, R. E., Scharlemann, J. P. W., Purvis, A., & Hudson, L. N. (2014). MODISTools - downloading and processing MODIS remotely sensed data in R. *Ecology and Evolution*, *4*(24), 4658–4668. https://doi.org/10.1002/ece3.1273

Valkiūnas, G. (2005). *Avian malaria parasites and other haemosporidian* (2nd ed.). Boca Raton, FL: CRC Press.

Walther, G.-R., Post, E., Convey, P., Menzel, A., Parmesan, C., Beebee, T. J. C., … Bairlein, F. (2002). Ecological responses to recent climate change. *Nature*, *416*, 389–395. https://doi.org/10.1038/416389a

Wells, K., Lakim, M. B., & O'Hara, R. B. (2014). Shifts from native to invasive small mammals across gradients from tropical forest to urban habitat in Borneo. *Biodiversity and Conservation*, *23*(9), 2289–2303. https://doi.org/10.1007/s10531-014-0723-5

Wickham, H., & Bryan, J. (2017). *readxl: Read excel files. R package version 1.0*. Retrieved from https://CRAN.R-project.org/package=readxl

Wickham, H., & Francois, R. (2017). *dplyr: A grammar of data manipulation. R package version 0.7.2*.

Wilman, H., Belmaker, J., Simpson, J., de la Rosa, C., Rivadeneira, M. M., & Jetz, W. (2014). EltonTraits 1.0: Species-level foraging attributes of the world's birds and mammals. *Ecology*, *95*(7), 2027–2027. https://doi.org/10.1890/13-1917.1

Woolhouse, M. E. J., Dye, C., Etard, J.-F., Smith, T., Charlwood, J. D., Garnett, G. P., … Anderson, R. M. (1997). Heterogeneities in the transmission of infectious agents: Implications for the design of control programs. *Proceedings of the National Academy of Sciences of the United States of America*, *94*(1), 338–342. https://doi.org/10.1073/pnas.94.1.338

Zhang, S., Zhang, Y., & Ma, K. (2016). Latitudinal variation in herbivory: Hemispheric asymmetries and the role of climatic drivers. *Journal*

of Ecology, *104*(4), 1089–1095. https://doi.org/10.1111/1365-2745.12588

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section.

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