Original article

LRH: N. J. Clark and S. M. Clegg

RRH: Vagrant hosts and insular parasite dynamics

The influence of vagrant hosts and weather patterns on the colonization and persistence of blood parasites in an island bird

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ABSTRACT

Aim Colonization and extinction are important drivers of island biogeography, but they are difficult to study. We used a long-term dataset to determine the mechanisms that contribute to colonization and persistence for vector-borne blood parasites in an island population of birds that regularly receives infected vagrant conspecifics and wind-assisted potential vectors from the mainland.

Location Heron Island (Australia) and the Australian mainland.

Methods We determined the prevalence, temporal stability and host-specificity of *Haemoproteus* and *Plasmodium* parasites in resident and mainland-vagrant silvereyes (*Zosterops lateralis*) on Heron Island over seven years (1999–2003 and 2012–2013). We carried out simulations using mainland infection data and vagrant arrival scenarios to test whether transmission from vagrants influences infection patterns in island residents. We tested whether variation in island parasite prevalence was predicted by abiotic factors associated with vector breeding and dispersal.

Results Parasite prevalence and composition on the island varied considerably across years. Host-specialist *Haemoproteus* species exhibited lower prevalence than expected and frequent absence despite a high probability of arriving via vagrants. In contrast, host-generalist *Plasmodium* species exhibited a low probability of arriving via vagrants but were temporally persistent in island silvereyes. Increases in prevalence and diversity of *Plasmodium* species were associated with episodes of offshore winds.

Main conclusions This study shows that parasites that are abundant in source populations do not necessarily exhibit increased colonization success via vagrant host movement. Vagrant silvereyes are not likely to shape infection patterns in island-resident silvereyes. Instead, indirect evidence of associations between weather patterns and parasite dynamics suggests that the insular parasite community may be limited by

vector establishment. Our results support the hypothesis that host-specificity is important in determining a parasite's ability to persist on islands, with host-specialists at greater risk of failing to establish after initial arrival.

Keywords

Australia, avian malaria, island biogeography, *Haemoproteus*, host-specificity, parasite colonization, *Plasmodium*, vagrant, wind dispersal, *Zosterops lateralis*.

INTRODUCTION

Islands offer exceptional models to study the processes that drive community dynamics (MacArthur & Wilson, 1967; Diamond, 1975). For islands, the interplay between colonization and extinction plays an important role in determining community composition by influencing species turnover and interspecific interactions (Lomolino, 2000; Losos & Ricklefs, 2010). Whether a species can effectively colonize an island depends on a number of factors, including the availability of suitable habitat, the number of colonists and the vagility of the species (Ricklefs & Lovette, 1999; Ahlroth *et al.*, 2003). Colonization–extinction dynamics are, however, difficult to investigate directly (Lees & Gilroy, 2014), and studies are often limited to inferring colonization histories from current patterns of presence and absence. Consequently, the mechanisms that contribute to island colonization and ultimately to community composition, are not well understood for many taxa (Lomolino, 2000).

For vector-borne parasites, patterns of island colonization and extinction are complex because of the habitat requirements and dispersal capacities of both intermediate and primary hosts (Sousa & Grosholz, 1991; Pérez-Tris & Bensch, 2005). Parasites that colonize or persist at low prevalence are at risk of extinction if the

population of either the vector or the primary host is reduced below a minimum transmission threshold (Bailey, 1957). Migratory or vagrant hosts that arrive from areas of higher parasite prevalence may therefore be important for maintaining island parasite populations (Clark *et al.*, 2014a). Additionally, differing levels of host-specificity are widely thought to determine whether some parasites effectively colonize or fail to persist following their initial arrival during a host's range expansion (Clay, 2003; MacLeod *et al.*, 2010). For instance, parasites able to infect a range of host species (generalists) may persist despite their prevalence being too low in a particular host if they are able to utilize multiple host reservoirs (Sturrock & Tompkins, 2008).

Generalists are therefore expected to be more successful at colonizing islands than specialists that rely on closely related hosts (Ewen *et al.*, 2012), particularly on small islands, where host population sizes may vary over time. However, specialized parasites may reach higher prevalence in their preferred hosts than do generalists (Keesing *et al.*, 2006), which can increase the parasite's chances of transport with dispersing hosts.

Avian haemosporidians of the genera *Plasmodium* and *Haemoproteus* are an abundant and diverse group of vector-transmitted blood parasites found all over the globe except Antarctica (Valkiūnas, 2005). These parasites exhibit a range of vertebrate host-specificity strategies, from strict host-specialists to widespread generalists (Valkiūnas, 2005). Advances in molecular screening have led to a growing number of avian haemosporidian island biogeography studies, revealing distribution anomalies indicative of complex colonization dynamics (Ishtiaq *et al.*, 2010; Cornuault *et al.*, 2012; Pérez-Rodríguez *et al.*, 2013) that may be partly driven by host-specificity strategies (Clark *et al.*, 2014b). In addition, because dispersing birds are thought to be important drivers of disease dispersal (Klaassen *et al.*, 2012; Levin *et al.*, 2013; van Dijk *et al.*, 2014), it has been suggested that vagrant birds act as dispersal vehicles for avian

haemosporidians in island systems (Santiago-Alarcon *et al.*, 2010; Valkiūnas *et al.*, 2010; Clark *et al.*, 2014a). Nevertheless, the role of vagrants in shaping island parasite communities remains untested because of a lack of empirical data for vagrant infection status and arrival frequencies.

Silvereyes, *Zosterops lateralis* (Latham, 1802), and related species within the Zosteropidae have high potential for dispersing parasites as they are remarkably good colonizers and have established island populations throughout the South Pacific (Lack, 1971). In addition, *Zosterops* species exhibit substantial levels of intraspecific variation (Clegg *et al.*, 2002; Clegg & Phillimore, 2010), a feature that may contribute to the high diversity of haemosporidian lineages carried by *Zosterops* hosts (Ishtiaq *et al.*, 2008, 2010). The Capricorn silvereye (*Z. l. chlorocephalus* Campbell & White, 1910) is the only regularly breeding passerine on Heron Island (Kikkawa, 1970), a small (0.12 km²), wooded cay 70 km from the Australian continental shore (28°26' S, 151°57' E; Fig. 1). The Capricorn silvereye is a subspecies that has diverged rapidly from the Australian mainland form (*Z. l. cornwalli* Mathews, 1912) (Clegg *et al.*, 2008). Nevertheless, mainland silvereyes regularly arrive on Heron Island as vagrants (Kikkawa, 1970) and are known to carry haemosporidian infections (Clark *et al.*, 2014a). Mainland vagrants can persist on the island for a number of weeks (Kikkawa, 1970), increasing the chance that infected vagrants can act as sources for the island parasite community. Importantly, vagrant arrival frequencies can be accurately estimated from observations made over a long-term study (> 40 years) of the Heron Island silvereye population (Kikkawa, 1970; Clegg *et al.*, 2008). Together, these features make the Heron Island silvereye population a tractable system to study the role of vagrant hosts in driving island parasite dynamics.

On the Australian mainland, silvereyes exhibit a high prevalence of haemosporidian infections, often reaching > 50% (Beadell *et al.*, 2007; Zamora-Vilchis *et*

al., 2012). In contrast, a recent study reported much lower prevalence (< 10%) in Heron Island's resident silvereyes (Clark *et al.*, 2014a), perhaps because of a lack of freshwater breeding grounds for arthropod vectors (Marks, 1969; Pierce & Adlard, 2004). The community composition of parasite lineages may also vary between mainland and Heron Island silvereyes. The majority of infections in mainland silvereyes are attributed to ZOSLAT04-H (Beadell *et al.*, 2007; Zamora-Vilchis *et al.*, 2012), a specialist *Haemoproteus* lineage representing a morphospecies that has only been recorded in hosts from the family Zosteropidae (*H. zosteropis* Chakravarty & Kar, 1945) (Valkiūnas, 2005; Bensch *et al.*, 2009). In contrast, 50% of infections in Heron Island silvereyes sampled in a single month (June 2012) were generalist *Plasmodium* lineages (Clark *et al.*, 2014a), suggesting that generalist lineages are more successful on the island than on the mainland. Nevertheless, silvereyes on Heron Island are prone to large fluctuations in population size (ranging from 225 to nearly 500 individuals; McCallum *et al.*, 2000) and under such conditions – small and variable vertebrate host density, low infection rates and limited vector breeding opportunities – stochastic parasite extinctions are likely, and temporal variation in both prevalence and community composition is expected (Bailey, 1957). The arrival of infected vagrant silvereyes may therefore be particularly important in shaping the community of parasite lineages infecting Heron Island's resident silvereyes.

In dynamic island systems, the colonization and local persistence of haemosporidian parasites might also be influenced by the population dynamics of vectors. Biting arthropods, the primary vectors for avian haemosporidians, are known to carry diseases among discrete populations during wind events (Ritchie & Rochester, 2001; Ducheyne *et al.*, 2007), and influxes of mosquitoes have been observed on Heron Island following periods of winds from the mainland (Fletcher, 1973), which approach

the island in a NNW to SSW direction (henceforth referred to as 'offshore winds'; Fig. 1). However, a tendency for vagrancy does not necessarily result in successful colonization (Lees & Gilroy, 2014). Infection dynamics may instead respond to weather conditions that promote vector breeding and development on the island (Zamora-Vilchis *et al.*, 2012; Galen & Witt, 2014).

We sampled silvereyes on Heron Island and the Australian mainland to test the hypothesis that vagrant birds can shape patterns of haemosporidian colonization and persistence in an island parasite community. We screened island silvereyes for haemosporidian infection across seven non-contiguous years to determine temporal patterns of lineage prevalence and composition. We then used a database of mainland silvereye haemosporidian infections to test whether vagrants could be shaping the observed infection patterns in island silvereyes. We also determined the host-specificity of parasite lineages to test whether host-specialists are less likely to colonize and persist on the island than host-generalists. Finally, we tested whether patterns of haemosporidian prevalence are associated with weather factors that may influence vector dispersal from the mainland (offshore winds) as well as vector breeding and population density on the island (rainfall and temperature).

MATERIALS AND METHODS

Sample collection

Resident and vagrant silvereyes were captured on Heron Island using mist nets and baited traps across seven years (1999–2003 and 2012–2013). Sampling on the island occurred during the non-breeding season from April to June each year. Morphological measurements of wing length, tail length, bill length, bill depth, bill width and weight were taken from each bird to differentiate island residents and mainland vagrants (see

Clegg *et al.*, 2002; Clark *et al.*, 2014a). Silvereyes were also captured using mist nets at sites in mainland eastern Australia (Fig. 1). A small blood sample was taken from the brachial vein of each bird and transferred to a microfuge tube containing lysis buffer (1% Sodium dodecyl sulfate, 20 mM NaCl, 10 mM TRIS pH 8.0 and 10 mM EDTA pH 8.0). Individuals were banded with an Australian Bird and Bat Banding Scheme (ABBBS) metal band, and a unique set of colour bands in the case of *Z. l. chlorocephalus*, and released at the site of capture.

Molecular methods

DNA was extracted using ammonium acetate/ethanol precipitation (Richardson *et al.*, 2001). Each bird was molecularly sexed following Griffiths *et al.* (1998), and this procedure also served to confirm extraction quality. Samples were screened for haemosporidian DNA (*Haemoproteus* and *Plasmodium*) using a nested polymerase chain reaction (PCR) to amplify 479 bp of the parasite cytochrome *b* (*cytb*) gene (Waldenström *et al.*, 2004) following reaction conditions given by Clark *et al.* (2014a). Positive amplifications were sequenced on an Applied Biosystems 3130xl Genetic Analyser (Foster City, California, USA) at the Griffith University DNA Sequencing Facility (Brisbane, Australia). Parasite sequences were aligned in GENEIOUS 5.4 (Biomatters, Auckland, New Zealand) and identified by comparison to sequences on GenBank and the avian malaria database, MalAvi (Bensch *et al.*, 2009). Mainland infection results were added to published data to bolster comparisons between island and mainland populations.

Host-specificity

We used data from avian host–parasite records (from MalAvi) to classify lineages as

either avian host-specialists or host-generalists. Lineages were classified as specialists if they have only been recorded to date in *Z. lateralis*. Lineages that have been amplified from at least one host species in addition to *Z. lateralis* were classified as generalists. However, to confidently determine if a haemosporidian parasite is a generalist, infection viability (i.e. circulating gametocytes in host blood) must be confirmed from a range of host taxa (Valkiūnas *et al.*, 2009). Such a classification is problematic due to the presumed high incidence of PCR-positive, abortive infections in natural populations (see Valkiūnas *et al.*, 2009; Galen & Witt, 2014). We therefore examined blood smears and parasite lineages from additional host-parasite occurrences in south-east Australia, as well as data from published sources, to repeat the analysis by classifying lineages as generalists using the criterion that gametocytes for that exact lineage must have been observed in blood smears from more than one avian species (see Appendix S1 in Supporting Information).

Weather variables

We accessed climate data for the Heron Island research station using the Australian Bureau of Meteorology's Data Services (<http://www.bom.gov.au/climate/data-services>). To test if weather variables commonly associated with vector breeding and abundance predicted haemosporidian prevalence, we calculated mean temperature and total number of days with rainfall (> 0.2 mm). To test whether offshore winds predicted haemosporidian prevalence, we calculated the total number of days on which mainland offshore winds were recorded. As the majority of haemosporidian infections are thought to be transmitted during the breeding season (Valkiūnas, 2005), weather variables (mean temperature, total days with rainfall and total days with offshore winds) were extracted to span the breeding/fledgling season for each year prior to

sampling (September of the previous year to March of the sampling year; Kikkawa, 1970).

Statistical analyses

All statistical analyses were conducted in the R statistical framework (version 3.0.1, R Development Core Team, 2008) except where specified. To test whether variation in parasite prevalence was explained by weather variables, we constructed generalized linear models (GLMs), specifying prevalence (number of birds infected and number uninfected) as the binomial response variable, weather variables as explanatory variables, and a binomial error structure (or quasibinomial when overdispersed, indicated by a dispersion parameter considerably greater than 1; Crawley, 2012). We could not fit a model with all interaction terms, because we had only seven data points (one for each year) and we therefore limited the number of explanatory variables in any model to three (Crawley, 2012). We first tested the overall prevalence of parasite lineages (*Plasmodium* and *Haemoproteus* species combined) against additive effects of number of days with offshore winds, mean temperature and number of days with rain, and then tested in subsequent models for each combination of two effects and their interaction. Because avian *Plasmodium* and *Haemoproteus* species are transmitted by different arthropod vectors (primarily *Culex* mosquitoes and *Culicoides* biting midges, respectively; Valkiūnas, 2005; Santiago-Alarcon *et al.*, 2012), models were repeated for *Plasmodium* and *Haemoproteus* separately. To account for the possibility of single years having a disproportionate influence on models due to the low number of data points, we used a bootstrapping procedure whereby one year was randomly excluded from the dataset and models were performed on the remaining data subset. This process was then repeated following replacement of the previously excluded year, allowing for each

year to have an equal probability of being excluded. We generated 1000 bootstrap replicates to calculate 95% confidence intervals of model statistics. We tested for autocorrelation in models by calculating the Durbin–Watson statistic, where values approaching zero indicate negative autocorrelation, values approaching four indicate positive autocorrelation, and values around two indicate no autocorrelation (Crawley, 2012).

To test whether variation in parasite prevalence in island silvereyes could be explained by the arrival of vagrant silvereyes, we drew pools of vagrant individuals from a simulated population of 100,000 mainland silvereyes. The simulated population was proportional to the number of infected and uninfected mainland silvereyes, with the infection prevalence of each parasite lineage equal to the prevalence identified in the mainland dataset (see Results). Although the number of vagrants arriving each year can vary from zero to a maximum record of 50 individuals (Kikkawa, 1970), observations of more than eight vagrants simultaneously on the island are rare (S.M.C., pers. obs.). We elected to use a conservative approach when simulating vagrant arrivals by repeating simulations using vagrant pools of varying size. We carried out five simulations in total, using vagrant pool sizes of 5, 8, 15, 30 and 50 individuals. For each simulation, we drew 10,000 pools from the null (mainland) distribution and determined the probability of occurrence for each parasite lineage by calculating the proportion of draws with at least one individual carrying that lineage.

To determine whether the frequencies of individual parasite lineages differed between island and mainland silvereyes, we tested whether the observed island parasite composition deviated from an expected null distribution based on infection frequencies recorded on the mainland. We simulated a second population of 100,000 mainland silvereyes in which all individuals were infected, using the observed

proportion of total infections for each parasite lineage from the mainland dataset. From this, we randomly drew 10,000 groups of 33 infected individuals (equal to the number of infections found in island silvereyes; see Results) and calculated the prevalence of each parasite lineage in each draw. Simulations were carried out using Monte Carlo randomizations with replacement.

We compared the observed proportion of generalist and specialist parasite infections between island and mainland populations using a chi-square test for equality of proportions. We repeated the analysis by excluding lineages that have been recovered from multiple host species but have not been microscopically confirmed (two *Plasmodium* lineages; see Results).

We compared parasite diversity between the mainland and island population using the non-parametric Chao2 estimator in the program ESTIMATES 9.1.0 (Colwell, 2001). This estimate was chosen to control for differences in sample size and to adjust for unsampled diversity based on the number of rare lineages detected (Chao, 1984; Colwell, 2001). Estimates were produced from 10,000 permutations.

RESULTS

We sampled 1014 island-resident silvereyes and detected 33 haemosporidian infections (3.3% prevalence). We sequenced 32 infections (one infection failed to produce a sequence), 17 of which were *Plasmodium* and 15 *Haemoproteus*. Total parasite prevalence varied considerably among years, ranging from 0 to 8.6% (Fig. 2, Table 1). *Plasmodium* infections were recorded in all years apart from 2003 and prevalence did not vary significantly (χ^2 test: d.f. = 6, $\chi^2 = 9.55$, $P = 0.15$), whereas *Haemoproteus* was absent in three of the seven years (Fig. 2, Table 1) and its prevalence varied significantly (χ^2 test: d.f. = 6, $\chi^2 = 33.86$, $P < 0.001$). Among the 32 sequenced infections, four parasite

lineages were recovered, three *Plasmodium* (lineages BELL01-P, ORW1-P and FANTAIL01-P) and one *Haemoproteus* (lineage ZOSLAT04-H; Table 1). Five of the eight mainland vagrants sampled on Heron Island were infected (63.5% prevalence), three with *Haemoproteus* ZOSLAT04-H, one with *Plasmodium* BELL01-P and one with *Plasmodium* ORW1-P (Table 1).

The mainland silvereye dataset consisted of 118 individuals, including 64 that were screened by Beadell *et al.* (2007), six screened by Zamora-Vilchis *et al.* (2012) and 48 new samples (see sample sites in Fig. 1). Silvereyes are widespread and common in eastern Australia, and although mainland sampling was not carried out adjacent to Heron Island (Fig. 1), individuals from the southern sampling region migrate north in winter months (Higgins *et al.*, 2006) and have been suggested as a source of vagrants on Heron Island (Kikkawa, 1970). In addition, all lineages detected in the southern region were present in the north in similar proportions (percentage of infections attributed to ZOSLAT04-H: 78.1% in the north, $n = 32$ infections; 79.4% in the south, $n = 34$ infections). In the mainland dataset, 65 individuals were infected (55.1% prevalence), 54 with *Haemoproteus* ZOSLAT04-H, one with *Haemoproteus* ZOSLAT07-H and 10 with *Plasmodium* spp. (seven with ORW1-P, three with ZOSXAN01-P; Table 1). When adjusted for sample size, parasite diversity estimates were similar for *Z. l. cornwalli* (mainland birds and vagrants combined; Chao2 estimate of 5.44 ± 1.18 lineages) and the island *Z. l. chlorocephalus* population (4.01 ± 0.50 lineages). When considering the island as a proportional subset of the mainland, the observed prevalence of *Haemoproteus* lineages in island residents was lower than expected, and the three *Plasmodium* lineages recovered in island residents were more prevalent than expected (Fig. 3).

The number of offshore wind days ranged from 34 to 62 across years and was

the only significant explanatory variable for the prevalence of *Plasmodium* spp. in island residents, a finding that was robust to the bootstrapping GLM procedure [bootstrap GLM including only offshore wind days; d.f. = 5, mean Z (95%CI) = 2.31(2.101, 2.600), mean $P = 0.02$]. Analysis of model residuals revealed no temporal autocorrelation (Durbin–Watson statistic, 1.96). The diversity of *Plasmodium* lineages in island silvereyes also increased with increasing offshore wind days (Appendix S2). No weather variables or two-way interactions significantly predicted variation in the prevalence of *Haemoproteus* spp. or of both genera combined (Appendix S3).

Vagrant pool simulations drawn from a population of infected and uninfected mainland silvereyes returned a 95% probability of detecting *Haemoproteus* ZOSLAT04-H if only five vagrants arrive on the island each year, increasing to 98.9% if eight vagrants arrive (Fig. 4a). The observed proportion of years in which *Haemoproteus* ZOSLAT04-H was detected in island residents was 57.1% (detected in four out of seven years; Figs 2 & 4a). For the three *Plasmodium* lineages detected in island silvereyes, vagrant pool simulations indicated a 42.2% probability of detection for all lineages combined when five vagrants arrive each year, and 48.9% when eight vagrants arrive (Fig. 4b). The observed proportion of years in which *Plasmodium* spp. infections were recorded in island silvereyes was 85.7% (Figs 2 & 4b). Our simulations suggest that between 15 and 30 mainland vagrants would need to arrive on the island each year to reach an 85.7% probability of detecting *Plasmodium* infections in each vagrant pool (Fig. 4b).

All four *Plasmodium* lineages recovered in this study have been recorded in a range of avian species and geographical locations (Bensch *et al.*, 2009; Appendix S1) and are classified as host generalists (low parasitaemia prevented morphological identification of *Plasmodium* lineages in blood smears). In contrast, *Haemoproteus*

ZOSLAT04-H and ZOSLAT07-H have only been recorded to date in hosts from the family Zosteropidae (Beadell *et al.*, 2007; Ishtiaq *et al.*, 2010; Zamora-Vilchis *et al.*, 2012), and both were microscopically confirmed to represent the *Zosterops*-restricted morphospecies *H. zosteropsis*. These two *Haemoproteus* lineages were therefore classified as host specialists in proportion analysis. The proportion of infections attributed to specialist and generalist lineages differed significantly between island and mainland populations (χ^2 test: d.f. = 1, $\chi^2 = 18.31$, $P < 0.001$), with generalist lineages accounting for 53.1% of island infections and 14.6% of mainland infections. When excluding generalists that have not been confirmed to infect hosts other than silvereyes (*Plasmodium* lineages FANTAIL01-P and ZOSXAN01-P; see Appendix S2), the results were similar (χ^2 test: d.f. = 1, $\chi^2 = 20.38$, $P < 0.001$).

DISCUSSION

The prevalence and composition of haemosporidian parasites in Heron Island silvereyes are markedly different from those of populations sampled in mainland Australia, displaying temporal dynamics of infection that are not consistent with the contemporary arrival of infected vagrant mainland silvereyes. Differences in colonization and persistence patterns observed between parasite genera appear to be influenced by variation in host-specificity and the potential for wind-assisted colonization. Compared to the mainland, host-generalist *Plasmodium* lineages were more frequent in silvereyes residing on Heron Island than host-specialist *Haemoproteus* lineages, supporting the notion that host generalism is a feature that facilitates successful island colonization and persistence. Indirect evidence provided by the relationships between weather variables and parasite prevalence further suggests that the composition of *Plasmodium* lineages on Heron Island is influenced by wind-aided

dispersal from the mainland.

Insular haemosporidian community composition and the role of vagrant silvereyes

Here we describe an ephemeral, low-prevalence insular haemosporidian community where stochastic extinction and avian host-mediated or vector-mediated recolonization must be important in shaping composition. Two lines of evidence, however, suggest that the dispersal of vagrant silvereyes has little influence on these dynamics. First, the observed prevalence of parasite lineages in island silvereyes was substantially different from that expected if the island population were a proportional subset of the mainland. Second, the observed detection probabilities across years for *Haemoproteus* and *Plasmodium* lineages in island silvereyes were substantially different from those expected under models that sample from the mainland parasite distribution based on realistic scenarios of vagrant arrivals. The complex life cycle of haemosporidians leaves them exposed to multiple transmission-limiting steps, and the relative importance of vector-associated or host-associated limits is likely to vary across systems. Reduced transmission efficiency may be particularly prevalent when colonization processes are involved, resulting in a 'drowning on arrival' phenomenon (MacLeod *et al.*, 2010). In Heron Island silvereyes, transmission does not appear to be limited by the arrival of infected vagrants. For instance, both empirical data and simulations suggest that *Haemoproteus* species are regularly carried by vagrants, often without successful transmission to island residents. Although this low level of transmission could reflect differences in immunity or vector-avoidance behaviours between island and mainland silvereyes, this seems doubtful because the Zosteropidae family generally exhibits high infection rates whenever avian haemosporidians are present (Ishtiaq *et al.*, 2010; Cornuault *et al.*, 2012). A growing number of studies have demonstrated differences in

haemosporidian parasite frequencies between island and mainland populations despite regular arrivals of dispersing avian hosts (Beadell *et al.*, 2007; Spurgin *et al.*, 2012; Pérez-Rodríguez *et al.*, 2013), and our results suggest that parasite host-specificity could explain this phenomenon.

Wind-assisted dispersal and parasite lineage dynamics

The influence of offshore winds on the prevalence and diversity of *Plasmodium* species suggests that wind-assisted dispersal may shape the observed island infection patterns. Although the arrival of vagrant silvereyes is likely to be influenced by offshore winds, our simulations suggest that arrival numbers would have to be unrealistically large to account for the patterns observed. Instead, the significant offshore wind effect could be attributed to wind-assisted vectors arriving from the mainland. A paucity of avian haemosporidian vectors has been reported for oceanic islands that lack permanent fresh water, as is the case for Heron Island (Marks, 1969; Pierce & Adlard, 2004). Island vector communities are therefore likely to be unstable, but near-shore islands may occasionally receive replenishment through wind-assisted dispersal (Fletcher, 1973; Chapman *et al.*, 1999). Mosquito species observed on Heron Island include *Aedes vigilax* and several *Culex* species (Marks, 1969), both of which are known to feed readily on birds (Santiago-Alarcon *et al.*, 2012). Species from both genera have been identified as vectors for avian haemosporidians in other bioregions (Valkiūnas, 2005), and haemosporidian DNA has been recovered from both genera in the South Pacific (Ishtiaq *et al.*, 2008). In addition, *A. vigilax* often collects in large numbers on the Australian mainland and can be readily dispersed across water bodies (Marks, 1969; Chapman *et al.*, 1999), but *A. vigilax* is a salt-marsh breeder with limited breeding opportunities on Heron Island. We suggest that *A. vigilax* is probably a transient component of the

island's vector community, with its presence governed by the frequency of offshore winds. The lack of temperature or rainfall effects on prevalence also suggests that vector numbers may often be too low to capitalize on favourable weather conditions for breeding. Associations (or lack thereof) between weather variables and *Plasmodium* species prevalence are consistent with the island having a transient vector community characterized by local extinction and recolonization. Nevertheless, although vagrant silvereyes are unlikely to produce the patterns observed, numerous other vagrant species temporarily occur on Heron Island in small numbers and the island is a stopover site for some migratory waders (Kikkawa, 1970). The roles of other avian species as parasite carriers on Heron Island could therefore contribute to the offshore wind effect.

We did not observe any influence of offshore winds on the prevalence of *Haemoproteus* species, which could be explained by the vector's movement patterns. Whereas some species of biting midges exhibit wind-assisted dispersal (Ducheyne *et al.*, 2007), other species restrict their movements to periods of low wind velocity (Murray, 1987). Because vagrant silvereyes are likely to carry *Haemoproteus* to the island, their frequent absence in island silvereyes and the lack of weather influences suggest that other factors are influencing the local abundance of vectors for *Haemoproteus*. The vectors responsible for transmitting the parasite lineages observed here are not known, and further research is needed to determine the mechanisms that restrict the transmission of *Haemoproteus* on the island. Nevertheless, the absence of *Haemoproteus* lineages on islands appears to be common across the globe (Clark *et al.*, 2014b) and may be indicative of limited vector dispersal and/or a paucity of breeding habitats.

Host-specificity

Explanation of the observed parasite prevalence and composition patterns requires a

broader perspective encompassing host-specificity with regard to both vector and vertebrate hosts. Our findings support the widely held hypothesis that host-specificity influences a parasite's colonization success (Clay, 2003; MacLeod *et al.*, 2010). The restriction of *H. zosteropsis* to *Zosterops* hosts limits this parasite's ability to persist on the island when its prevalence in silvereyes is low, which may explain the pattern of apparent extinction and recolonization. In contrast, the *Plasmodium* lineages found in Heron Island silvereyes are avian host-generalists. Other vagrant bird species could occasionally carry generalist *Plasmodium* lineages to the island, increasing the chances of persistence for particular lineages in silvereyes. For instance, the most stable lineage in island silvereyes, lineage ORW1-P, infects a variety of species in south-eastern Australia (Appendix S1). Generalist lineages could also exploit a wider set of resident host species on islands, increasing their chances of persistence by having larger reservoirs of infection (Ewen *et al.*, 2012).

If the vector community is ephemeral as suggested, the extent to which parasite lineages have generalist or specialist vector requirements will also affect their likelihood of persistence. Some *Plasmodium* lineages display generalist tendencies with respect to vector species (Njabo *et al.*, 2011; Inci *et al.*, 2012), which can be advantageous to a lineage's colonization and persistence on islands where avian and vector host abundance can vary over time. The global colonization success of *Plasmodium* species on islands (Clark *et al.*, 2014b), combined with the temporal persistence of *Plasmodium* lineages observed here, may partly be a result of generalist vector preferences, although more data are needed on specific vector species (Lima & Bensch, 2014). In contrast to *Plasmodium*, some lineages of *Haemoproteus* display specialist relationships with vector species (Martínez-de la Puente *et al.*, 2011), a trait that may lead to high prevalence when the vector is common but would be highly

restrictive when the vector is absent or at low abundance. An inability to use multiple vector species could explain why *Haemoproteus* infections were frequently absent from the island silvereye population.

‘Drowning on arrival’ phenomenon in a host-specialist parasite

The complex nature of interacting variables that can shape haemosporidian community dynamics on islands may best be summarized by the patterns displayed by a single lineage, *Haemoproteus* ZOSLAT04-H. This is the most common haemosporidian lineage in mainland silvereyes, and is regularly carried to the island by vagrant silvereyes, but does not occur in the island population at the prevalence expected if it were readily transmissible. In addition, this lineage is frequently absent from island residents, suggesting that ZOSLAT04-H exhibits a pattern of drowning on arrival. The lower success of ZOSLAT04-H on Heron Island than on the mainland is unlikely to be due to a single factor, but rather several interacting processes. First, the vectors for *Haemoproteus* may not be common or persistent on the island, nor arrive frequently enough via wind-assisted dispersal to replenish the island vector population. Second, vector and avian host specialization is likely to limit opportunities for the lineage to persist on the island. Vector information for ZOLAT04-H is not available but, like other *Haemoproteus* species, it may not be able to use multiple vector species to perpetuate infection on the island. Furthermore, this lineage is limited to avian hosts belonging to the genus *Zosterops* and therefore its probability of persistence is not raised via the mechanism of infecting other avian species.

CONCLUSIONS

To our knowledge, this is the first study to test whether contemporary dispersal of

vagrant hosts contributes to colonization and persistence in an island parasite system. Despite a propensity for infections to be carried by vagrants, the temporally unstable prevalence and lineage composition patterns in island silvereyes are not consistent with models of vagrant arrival from the mainland. The level of vector and host specialization, along with propensity for wind-assisted dispersal may be more important variables for explaining parasite community composition in island systems. Our results, from a relatively simplified system, highlight the complex and potentially lineage-specific interactions of abiotic, vector and host-related limits on the colonization and persistence of vector-borne parasites in small island populations.

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REFERENCES

- Ahlroth, P., Alatalo, R.V., Holopainen, A., Kumpulainen, T. & Suhonen, J. (2003) Founder population size and number of source populations enhance colonization success in waterstriders. *Oecologia*, **137**, 617–620.
- Bailey, N.T.J. (1957) *The mathematical theory of epidemics*. Hafner, New York.
- Beadell, J.S., Atkins, C., Cashion, E., Jonker, M. & Fleischer, R.C. (2007) Immunological change in a parasite-impooverished environment: divergent signals from four island taxa. *PLoS ONE*, **2**, e896.
- 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**, 1353–1358.
- Chao, A. (1984) Nonparametric estimation of the number of classes in a population. *Scandinavian Journal of Statistics*, **11**, 265–270.
- Chapman, H.F., Hughes, J.M., Jennings, C., Kay, B.H. & Ritchie, S.A. (1999) Population structure and dispersal of the saltmarsh mosquito *Aedes vigilax* in Queensland, Australia. *Medical and Veterinary Entomology*, **13**, 423–430.
- Clark, N.J., Adlard, R.D. & Clegg, S.M. (2014a) First evidence of avian malaria in Capricorn Silvereyes (*Zosterops lateralis chlorocephalus*) on Heron Island. *The Sunbird*, **44**, 1–11.
- Clark, N.J., Clegg, S.M. & Lima, M.R. (2014b) A review of global diversity in avian haemosporidians (*Plasmodium* and *Haemoproteus*: Haemosporida): new insights from molecular data. *International Journal for Parasitology*, **44**, 329–338.
- Clay, K. (2003) Parasites lost. *Nature*, **421**, 585–586.

- Clegg, S.M. & Phillimore, A.B. (2010) The influence of gene flow and drift on genetic and phenotypic divergence in two species of *Zosterops* in Vanuatu. *Philosophical Transactions of the Royal Society B: Biological Sciences*, **365**, 1077–1092.
- Clegg, S.M., Degnan, S.M., Moritz, C., Estoup, A., Kikkawa, J. & Owens, I.P.F. (2002) Microevolution in island forms: the roles of drift and directional selection in morphological divergence of a passerine bird. *Evolution*, **56**, 2090–2099.
- Clegg, S.M., Frentiu, F.D., Kikkawa, J., Tavecchia, G. & Owens, I.P.F. (2008) 4000 years of phenotypic change in an island bird: heterogeneity of selection over three microevolutionary timescales. *Evolution*, **62**, 2393–2410.
- Colwell, R. (2001) *EstimateS: statistical estimation of species richness and shared species from samples, version 9.1.0*. University of Connecticut, Storrs, CT. Available at: <http://viceroy.eeb.uconn.edu/EstimateS/>.
- Cornuault, J., Bataillard, A., Warren, B.H., Lootvoet, A., Mirleau, P., Duval, T., Milá, B., Thébaud, C. & Heeb, P. (2012) The role of immigration and *in-situ* radiation in explaining blood parasite assemblages in an island bird clade. *Molecular Ecology*, **21**, 1438–1452.
- Crawley, M.J. (2012) *The R book*. John Wiley & Sons, Chichester, UK.
- Diamond, J.M. (1975) The island dilemma: lessons of modern biogeographic studies for the design of natural reserves. *Biological Conservation*, **7**, 129–146.
- van Dijk, J.G.B., Hoyer, B.J., Verhagen, J.H., Nolet, B.A., Fouchier, R.A.M. & Klaassen, M. (2014) Juveniles and migrants as drivers for seasonal epizootics of avian influenza virus. *Journal of Animal Ecology*, **83**, 266–275.
- Ducheyne, E., De Deken, R., Bécu, S., Codina, B., Nomikou, K., Mangana-Vougiaki, O., Georgiev, G., Purse, B.V. & Hendrickx, G. (2007) Quantifying the wind dispersal of *Culicoides* species in Greece and Bulgaria. *Geospatial Health*, **1**, 177–189.

- Ewen, J.G., Bensch, S., Blackburn, T.M., Bonneaud, C., Brown, R., Cassey, P., Clarke, R.H. & Pérez-Tris, J. (2012) Establishment of exotic parasites: the origins and characteristics of an avian malaria community in an isolated island avifauna. *Ecology Letters*, **15**, 1112–1119.
- Fletcher, B.S. (1973) Observations on a movement of insects at Heron Island, Queensland. *Australian Journal of Entomology*, **12**, 157–160.
- 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**, 374–386.
- Griffiths, R., Double, M.C., Orr, K. & Dawson, R.J.G. (1998) A DNA test to sex most birds. *Molecular Ecology*, **7**, 1071–1075.
- Higgins, P.J., Peter, J.M. & Cowling, S.J. (eds) (2006) *Handbook of Australian, New Zealand and Antarctic birds*, Vol. 7, *Boatbill to Starlings*. Oxford University Press, Melbourne.
- Inci, A., Yildirim, A., Njabo, K.Y., Duzlu, O., Biskin, Z. & Ciloglu, A. (2012) Detection and molecular characterization of avian *Plasmodium* from mosquitoes in central Turkey. *Veterinary Parasitology*, **188**, 179–184.
- Ishtiaq, F., Guillaumot, L., Clegg, S.M., Phillimore, A.B., Black, R.A., Owens, I.P.F., Mundy, N.I. & Sheldon, B.C. (2008) Avian haematozoan parasites and their associations with mosquitoes across Southwest Pacific Islands. *Molecular Ecology*, **17**, 4545–4555.
- Ishtiaq, F., Clegg, S.M., Phillimore, A.B., Black, R.A., Owens, I.P.F. & Sheldon, B.C. (2010) Biogeographical patterns of blood parasite lineage diversity in avian hosts from southern Melanesian islands. *Journal of Biogeography*, **37**, 120–132.
- Keesing, F., Holt, R.D. & Ostfeld, R.S. (2006) Effects of species diversity on disease risk. *Ecology Letters*, **9**, 485–498.
- Kikkawa, J. (1970) Birds recorded at Heron Island. *The Sunbird*, **1**, 34–48.

- Klaassen, M., Hoyer, B.J., Nolet, B.A. & Buttemer, W.A. (2012) Ecophysiology of avian migration in the face of current global hazards. *Philosophical Transactions of the Royal Society B: Biological Sciences*, **367**, 1719–1732.
- Lack, D.L. (1971) *Ecological isolation in birds*. Blackwell Scientific, Oxford.
- Lees, A.C. & Gilroy, J.J. (2014) Vagrancy fails to predict colonization of oceanic islands. *Global Ecology and Biogeography*, **23**, 405–413.
- Levin, I.I., Zwiars, P., Deem, S.L., Geest, E.A., Higashiguchi, J.M., Iezhova, T.A., Jiménez-Uzcátegui, G., Kim, D.H., Morton, J.P., Perlut, N.G., Renfrew, R.B., Sari, E.H.R., Valkiūnas, G. & Parker, P.G. (2013) Multiple lineages of avian malaria parasites (*Plasmodium*) in the Galapagos Islands and evidence for arrival via migratory birds. *Conservation Biology*, **27**, 1366–1377.
- Lima, M.R. & Bensch, S. (2014) Why some parasites are widespread and abundant while others are local and rare? *Molecular Ecology*, **23**, 3130–3132.
- Lomolino, M.V. (2000) A call for a new paradigm of island biogeography. *Global Ecology and Biogeography*, **9**, 1–6.
- Losos, J.B. & Ricklefs, R.E. (eds) (2010) *The theory of island biogeography revisited*. Princeton University Press, Princeton, NJ.
- MacArthur, R.H. & Wilson, E.O. (1967) *The theory of island biogeography*. Princeton University Press, Princeton, NJ.
- MacLeod, C.J., Paterson, A.M., Tompkins, D.M. & Duncan, R.P. (2010) Parasites lost – do invaders miss the boat or drown on arrival? *Ecology Letters*, **13**, 516–527.
- Marks, E.N. (1969) Mosquitoes (Culicidae) on Queensland's coral cays. *Queensland Naturalist*, **19**, 94–98.
- Martínez-de la Puente, J., Martínez, J., Rivero-de Aguilar, J., Herrero, J. & Merino, S. (2011) On the specificity of avian blood parasites: revealing specific and generalist

- relationships between haemosporidians and biting midges. *Molecular Ecology*, **20**, 3275–3287.
- McCallum, H., Kikkawa, J. & Catterall, C. (2000) Density dependence in an island population of silvereyes. *Ecology Letters*, **3**, 95–100.
- Murray, M.D. (1987) Local dispersal of the biting-midge *Culicoides brevitarsis* Kieffer (Diptera, Ceratopogonidae) in southeastern Australia. *Australian Journal of Zoology*, **35**, 559–573.
- Njabo, K.Y., Cornel, A.J., Bonneaud, C., Toffelmier, E., Sehgal, R.N.M., Valkiūnas, G., Russell, A.F. & Smith, T.B. (2011) Nonspecific patterns of vector, host and avian malaria parasite associations in a central African rainforest. *Molecular Ecology*, **20**, 1049–1061.
- Peirce, M.A. & Adlard, R.D. (2004) Absence of blood parasites in birds on Heron Island, Great Barrier Reef. *Corella*, **28**, 38–39.
- Pérez-Rodríguez, A., Ramírez, Á., Richardson, D.S. & Pérez-Tris, J. (2013) Evolution of parasite island syndromes without long-term host population isolation: parasite dynamics in Macaronesian blackcaps *Sylvia atricapilla*. *Global Ecology and Biogeography*, **22**, 1272–1281.
- Pérez-Tris, J. & Bensch, S. (2005) Dispersal increases local transmission of avian malarial parasites. *Ecology Letters*, **8**, 838–845.
- R Development Core Team (2008) *R: a language and environment for statistical computing*. R Foundation for Statistical Computing, Vienna, Austria.
- Richardson, D.S., Jury, F.L., Blaakmeer, K., Komdeur, J. & Burke, T. (2001) Parentage assignment and extra-group paternity in a cooperative breeder: the Seychelles warbler (*Acrocephalus sechellensis*). *Molecular Ecology*, **10**, 2263–2273.
- Ricklefs, R.E. & Lovette, I.J. (1999) The roles of island area *per se* and habitat diversity in

- the species–area relationships of four Lesser Antillean faunal groups. *Journal of Animal Ecology*, **68**, 1142–1160.
- Ritchie, S.A. & Rochester, W. (2001) Wind-blown mosquitoes and introduction of Japanese encephalitis into Australia. *Emerging Infectious Diseases*, **7**, 900–903.
- Santiago-Alarcon, D., Outlaw, D.C., Ricklefs, R.E. & Parker, P.G. (2010) Phylogenetic relationships of haemosporidian parasites in New World Columbiformes, with emphasis on the endemic Galapagos dove. *International Journal for Parasitology*, **40**, 463–470.
- 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**, 928–964.
- Sousa, W.P. & Grosholz, E.D. (1991) The influence of habitat structure on the transmission of parasites. *Habitat structure* (ed. by S.S. Bell, E.D. McCoy and H.R. Mushinsky), pp. 300–324. Springer, Dordrecht.
- Spurgin, L.G., Illera, J.C., Padilla, D.P. & Richardson, D.S. (2012) Biogeographical patterns and co-occurrence of pathogenic infection across island populations of Berthelot's pipit (*Anthus berthelotii*). *Oecologia*, **168**, 691–701.
- Sturrock, H.J.W. & Tompkins, D.M. (2008) Avian malaria parasites (*Plasmodium* spp.) in Dunedin and on the Otago Peninsula, southern New Zealand. *New Zealand Journal of Ecology*, **32**, 98–102.
- Valkiūnas, G. (2005) *Avian malaria parasites and other Haemosporidia*. CRC Press, Boca Raton, FL.
- Valkiūnas, G., Iezhova, T.A., Loiseau, C. & Sehgal, R.N.M. (2009) Nested cytochrome *b* polymerase chain reaction diagnostics detect sporozoites of hemosporidian parasites in peripheral blood of naturally infected birds. *Journal of Parasitology*, **95**, 1512–

1515.

- Valkiūnas, G., Santiago-Alarcon, D., Levin, I.I., Iezhova, T.A. & Parker, P.G. (2010) A new *Haemoproteus* species (Haemosporida: Haemoproteidae) from the endemic Galapagos dove *Zenaida galapagoensis*, with remarks on the parasite distribution, vectors, and molecular diagnostics. *Journal of Parasitology*, **96**, 783–792.
- Waldenström, J., Bensch, S., Hasselquist, D. & Östman, Ö. (2004) A new nested polymerase chain reaction method very efficient in detecting *Plasmodium* and *Haemoproteus* infections from avian blood. *Journal of Parasitology*, **90**, 191–194.
- Zamora-Vilchis, I., Williams, S.E. & Johnson, C.N. (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.

SUPPORTING INFORMATION

Additional Supporting Information may be found in the online version of this article:

Appendix S1 Alternative lineage names, GenBank accession numbers and host-specificity of avian haemosporidian lineages.

Appendix S2 Linear regression of *Plasmodium* spp. prevalence in silvereyes resident on Heron Island against number of offshore wind days.

Appendix S3 GLM results for tests of weather variables against parasite prevalence.

BIOSKETCH

Nicholas Clark is a PhD student (supervised by Sonya Clegg) interested in host–parasite relationships and their effects on host distribution and divergence patterns in Australia and the South Pacific.

Sonya Clegg is interested in island biogeography patterns and the microevolutionary mechanisms that underlie genetic and phenotypic divergence in wild populations.

Editor: Brent Emerson

Table 1 Sample sizes and prevalence of avian *Plasmodium* (with suffix P) and *Haemoproteus* (with suffix H) lineages recovered from silvereyes (*Zosterops lateralis*) on Heron Island and mainland Australia. Island residents were sampled across seven years. Plas.: *Plasmodium*; Haem.: *Haemoproteus*.

Sample population	Year	<i>n</i>	<i>Plasmodium</i>					<i>Haemoproteus</i>		
			ORW1-P	BELL01-P	FANTAIL01-P	ZOSXAN01-P	Prevalence	ZOSLAT04-H	ZOSLAT07-H	Prevalence
Island-residents	1999	158	4	0	0	0	2.5%	0	0	0.0%
	2000	108	1	0	0	0	0.9%	0	0	0.0%
	2001	93	0	1	0	0	1.1%	7	0	7.5%
	2002	126	2	1	1	0	3.2%	1	0	0.8%
	2003	92	0	0	0	0	0.0%	0	0	0.0%
	2012	195	3	3	0	0	3.1%	5	0	2.6%
	2013	242	1	0	0	0	0.4%	2	0	0.8%
Island vagrants		8	1	1	0	0	25.0%	3	0	37.5%
Mainland		118	7	0	0	3	8.5%	54	1	46.6%

FIGURE LEGENDS

Figure 1 Map of the sampling area. Sampling locations for silvereyes (*Zosterops lateralis*) on the Australian mainland and on Heron Island are represented by black dots. Shaded areas show sampling regions of published lineages included in the mainland silvereye dataset.

Figure 2 Prevalence of avian *Plasmodium* lineages (grey bars) and *Haemoproteus* lineages (black bars) infecting silvereyes (*Zosterops lateralis*) resident on Heron Island, Australia, across sample years. Numbers above bars indicate absolute number of infected individuals.

Figure 3 Expected (grey bars $\pm$ SD) and observed (black bars) prevalence of each mainland parasite lineage of *Plasmodium* or *Haemoproteus* in silvereyes (*Zosterops lateralis*) resident on Heron Island, Australia. The expected prevalence for each lineage was generated from 10,000 Monte Carlo simulations from a null distribution of 100,000 infected mainland silvereyes.

Figure 4 Expected probability of detecting (a) avian *Haemoproteus* infections and (b) avian *Plasmodium* infections in pools of vagrant-mainland silvereyes (*Zosterops lateralis*) in Australia. Probabilities were generated from 10,000 Monte Carlo simulations from a null distribution of 100,000 mainland silvereyes using the observed prevalence data presented in this study. Dashed lines represent observed proportion of years in which avian *Haemoproteus* (a) and *Plasmodium* (b) lineages were recovered in Heron Island's resident silvereyes.