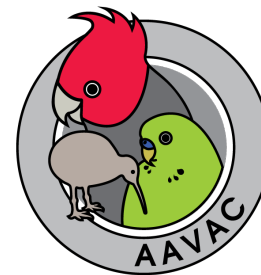


Costs of Avian Malaria in Australo-Papuan Avifauna



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Introduction

Avian malarial parasites are represented by a large diversity of haemosporida within Plasmodiidae and Haemoproteidae families. Other closely related haemosporida from Leukocytozoidae and Garniidae also infect birds and are often discussed under the umbrella of avian malaria. Each family differs in life-cycle, host and vector specificity, distribution, and pathogenicity.

Avian malarial infections are described as acute, chronic and abortive (Valkiunas 2004b; Valkiunas 2011). The acute phase is distinguished by a rising parasitaemia, occurring after a latent or prepatent period following inoculation. This acute phase is often associated with the most overt clinical signs including lethargy, dyspnoea, seizures, and death (Valkiunas 2004c). Survivors of the acute phase go on to carry the infections chronically at relatively low intensities and near life-long infections can occur (Bishop et al., 1938). Chronic avian malaria is associated with a variety of costs. In addition to mortality these include effects on investment in reproduction (Marzal et al., 2005) and immunity (Loiseau et al., 2011; Dunn et al., 2013), sexual ornaments and signaling (Hörak et al., 2001; Bosholn et al., 2016), and lifespan (Asghar et al., 2015). Abortive infections are those detected with PCR without evidence of completing reproduction within the vertebrate host or vector and the outcomes of such infections can range from minimal clinical signs to death (Olias et al., 2011; Cannell et al., 2013; Levin et al., 2013; Dimitrov et al., 2015).

The distribution of avian malaria spans both transmission and non-transmission zones. Transmission is the result of complex interactions between avian hosts, vectors, malarial organisms, and local environmental

factors. These parasites are thus not detected evenly throughout their distribution; instead a mosaic pattern nested within a general trend of higher diversity and prevalence at lower altitudes and latitudes is observed (Mendes et al., 2005; Wood et al., 2007; Clark et al., 2016). Large-scale migratory movements of avian hosts and near life-long infections mean these parasites can move within their avian hosts well beyond current transmission zones, effectively expanding their distribution.

Avian malaria is associated with island bird extinctions and can impose significant restrictions to avian distributions (Warner 1968; van Riper III et al., 1982). This is in stark contrast to evidence revealing avian hosts successfully completing large-scale movements while infected (Bensch et al., 2007). Some avian families, including penguins (Spheniscidae) have shown particular susceptibility to avian malaria, however this is not consistent for all species and populations. The introduction of a known vector of *Plasmodium relictum*, *Culex quinquefasciatus*, to the Galapagos Islands exposed the naïve island avifauna to avian malaria (Whiteman et al., 2005). Despite exposure evidenced by serological and molecular testing, clinical signs and parasitaemia were rarely observed in Galapagos Penguins (Palmer et al., 2013). Predicting the effects of avian malaria in a host is thus not a simple task. It is clear that the outcomes of infection with avian malaria are multifactorial and dependent on a range of broad concepts such as host immunity and tolerance, exposure dynamics, and parasite virulence. Costs are detected disproportionately less frequently in wild populations, possibly due to a sample bias towards less affected individuals or through removal of affected individuals by predation (Bennett et al., 1993; Valkiunas 2004a). Much of what is known of avian malaria comes from the northern hemisphere, and of all

the continents, Australia stands out for its paucity of avian malaria investigations (Clark et al., 2014).

The Australian continent is expansive and provides an unbroken land bridge between the tropics and temperate zones. Frequent continental avian movements connect avian populations on the mainland and nearby islands (Griffioen and Clarke 2002), and inter-continental migration further connects Australia to Papua New Guinea and beyond into the Northern Hemisphere (Dingle 2008). Unsurprisingly, Australia is host to a diverse population of avian malarial organisms (Adlard et al., 2004; Beadall et al., 2004). However studies of the effects and costs of these parasites in Australian avifauna are few. A single Australian study has determined how the distribution and diversity of avian malaria are associated with altitude, and simultaneously inferred costs by an association of avian species distributions with absence of avian malaria and vectors (Zamora-Vilchis et al., 2012). Direct costs of avian malaria in Australian avifauna are, however, limited to a handful of reports describing histological findings from sporadic mortality events of small groups or individuals (Peirce et al., 2004; Olias et al., 2011; Cannell et al., 2013).

Distributions of vector-borne diseases are not static and climate change is predicted to affect distributions of avian malaria (Garamszegi 2011; Pérez-Rodríguez et al., 2014). Rising temperatures are expected to allow transmission at higher altitudes and latitudes, and evidence suggests changes are already occurring (Loiseau et al., 2012; Atkinson et al., 2014). In Australia, avian malaria has been detected as far south as Perth (Cannell et al., 2013), where it caused an outbreak in the local penguin population. It is unclear what role climate change had in aiding this outbreak, but it is suggested that warmer than usual ocean surface created ideal environments for vector breeding.

There is no current understanding of the distributions of avian malaria lineages in Australian avifauna, nor is there an understanding of host-parasite dynamics for almost all the known malaria infecting Australian avifauna. This project focuses on Australo-Papuan avifauna in tropical Far North Queensland (FNQ) and Torres Strait (TS) and includes data spanning 10 years. This project is the first to investigate the effects of avian malaria in an endemic transmission zone in wild Australian avifaunal populations. It incorporates ecological survey methods and veterinary tests and interpretation. By targeting tropical avian communities, the potential diversity of avian

malarial organisms in north eastern Australia is described, and baseline expectations of effects in naturally infected populations are determined.

Sample and Data Collection

Birds were captured, sampled and released from communities in the north-western islands of TS and mainland FNQ in the years spanning 2006 and 2015. Each bird was banded, provided with a unique ID, and blood was sampled. Blood and data from approximately 1000 birds were collected for this project.

A broad approach was taken and numerous potential indices were measured. These included:

- Parasitaemia, prevalence, and infection intensity: manual cell counts (parasites per 10,000 erythrocytes) and qPCR;
- Immunity investment: manual differential white cell counts (cells per 10,000 erythrocytes);
- Plasma biochemistry: Carotenoid concentration, VetScan VS2® Avian Reptile Profile Plus biochemistry profile (VetScan VS2® Chemistry Analyzer, Abaxis, Inc.);
- Visual signaling: plumage colour measured by photospectroscopy; and
- Body condition: body mass (scaled to skeletal size), body condition score (pectoral muscle mass).

Results and Discussion

Approximately 67% (n=900) of all birds sampled in TS were positive by PCR for avian malaria and a lower prevalence, 37%, was detected in FNQ (n=104). Prevalence of malaria varied greatly between avian species, ranging from 0% to 98%, and infection intensity also differed significantly between species. Quick scans (scanning each slide for less than five minutes each) confirmed 69% of PCR positive birds, however the agreement increased as more time was applied to each slide. The relationship between PCR/qPCR and microscopy agreement varied for different avian hosts and malaria species. There was greatest agreement for *Haemoproteus* species, with agreement reaching 100% for some avian species. The majority of malaria were *haemoproteus* lineages and numerous novel lineages were detected. This is consistent with previous work in northern and south eastern Queensland (Adlard et al., 2004; Beadall et al., 2004). Prevalence was similar between dry and

wet seasons on Boigu Island, however infection intensity was greatest in the wet season. Presumably this is associated with vector abundance, timing of breeding, and other factors including seasonal mortality patterns.

The communities from which these samples were taken included active flying birds and so chronically infected birds were expected to comprise the majority of captured individuals. Chronic malaria infections show fewer clinical signs and costs are more subtle and difficult to detect (Asghar et al., 2015). Thus it was expected that few significant differences were detected between infected and non-infected birds for most of the indices measured in sampled birds. Differences were detected between positive and negative birds for plasma carotenoid concentration and indicators of erythrocytic turnover. There are also detectable and significant correlations with both infection intensity and body condition for these indices.

Conclusions

This project provides important baseline information for the current state of avian malaria in northern Australia. Outcomes demonstrate surveillance and health investigation of avian populations is possible even in remote areas of Australia. Most importantly it reveals costs to individuals associated with chronic avian malaria infections in Australian birds and subtle signs can be detected using samples and data collected under field conditions. The benefits of collaboration between disciplines (in this case ecology and veterinary science) are highlighted by this project and shows the value in samples collected by ecologists for veterinary use. Human facilitated movements of birds commonly occurs in Australian aviculture, and is becoming more important for conservation. The expansive size of Australia means that many of these movements can potentially transport naïve or infected avian hosts into or beyond current avian malaria distribution zones. Disease surveillance of wild avian populations and communities is thus an important aid for both wildlife and pet/aviary bird vets, especially for diseases with the potential to spread between wild and captive individuals. Translocations and release programs are increasingly an important component of conservation management of avian species. It is easy to see how understanding infectious diseases present at sites of collection, captive care, and release can be invaluable and equally as important as understanding baseline effects in stable populations.

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