*RAFFLES BULLETIN OF ZOOLOGY* **63**: 279–286
Date of publication: 26 June 2015
http://zoobank.org/urn:lsid:zoobank.org:pub:511E96EF-4EB2-4D39-89FE-DC23266053F8

# Prevalence of avian haemosporidians among understorey birds of Mt. Banahaw de Lucban, Philippines.

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**Abstract.** Understanding vector-borne diseases such as avian hemosporidiosis provides clues regarding environmental changes that can be crucial for conservation efforts. Haemosporidian prevalence in understorey birds from Mt. Banahaw de Lucban was studied to survey parasites occurring in the area, and test the difference between prevalence in high and low altitudes. Ninety-five birds were captured from May–June 2013 at 745–800 m, and 1500–1600 m, and were screened for haemosporidians via peripheral blood smear microscopy. *Leucocytozoon*, *Haemoproteus*, and *Plasmodium* were found in 28.4% of the birds sampled, with *Haemoproteus* most frequently occurring. Half of the 24 bird species screened showed positive infections, and six species were identified as new Philippine host records of avian haemosporidians. Prevalence of infection did not significantly differ between altitudes and could be attributed to factors that warrant further investigation. Relatively high prevalence of predominantly chronic infections suggests birds are currently able to keep infection intensities in check but this cannot be guaranteed once they are immunocompromised, leading to potential disease outbreaks. Long term monitoring of haemosporidian prevalence and strict security against anthropogenic disturbance in Mt. Banahaw de Lucban may help reduce the risk of epizootic outbreaks that threaten Philippine endemic avifauna.

**Key words.** *Haemoproteus*, *Leucocytozoon*, *Plasmodium*, birds, Mt. Banahaw, Philippines

## INTRODUCTION

Avian haemosporidians are protozoan blood parasites belonging to the genera *Plasmodium*, *Haemoproteus*, and *Leucocytozoon.* These parasites are transmitted to susceptible birds by insect vectors, which include mosquitoes, black flies, sand flies, biting midges, and louse flies (Valkiūnas, 2005). Documented avian haemosporidian vectors in the Philippines include *Simulium* sp. for *Leucocytozoon* (Vidal, 1968), *Culex fatigans* for *Plasmodium juxtanucleare* (Directo, 1969), and *Culex quinquefasciatus fatigans* for *Plasmodium capistrani* (Russell, 1932). Avian malaria has been a recent focus in climate change studies due to some aspects of both vector and parasite biology that are temperature/climate dependent, such as the temperature threshold for malaria oocyst development (Benning et al., 2002, in Ahumada et al., 2004), as well as the requirement of an aqueous substrate for mosquito development. Studies have also shown that the increase in global temperatures favour both avian malaria vectors and parasites, thereby extending their distributions to more polar regions and higher elevations (Freed et al., 2005; Sehgal, 2010; Garamszegi, 2011; Samuel et al., 2011; Zamora-Vilchis et al., 2012). The recent extinction of some Hawaiian honeycreepers due to avian malaria (Freed et al., 2005), as well as the emergence of avian malaria in arctic regions such as Alaska (Loiseau et al., 2012), are testaments to how climate change can potentially threaten biodiversity and wildlife populations via changing the distributions of parasites and their respective vectors.

In the Philippines, the earliest known studies on avian malaria reported the presence of *Plasmodium* and *Haemoproteus* in several wild bird species (Russell, 1932; Dy & Soriano, 1940). Eduardo (2007) provided an extensive list of avian malaria species known to parasitise Philippine avifauna, which mostly cited the works done by Bennett and his colleagues. The most recent study on the prevalence of avian haemosporidians was by Silva-Iturriza et al. (2012) in the central Philippine islands. These studies mostly contribute to baseline data on the taxonomy and prevalence of avian haemosporidia in the Philippines, however, to our knowledge no known local studies have explored the relationship between avian malaria prevalence and elevation differences. Recent studies have reported a positive association between avian malaria prevalence and increasing temperatures, thereby potentially widening the distribution of avian malaria (via their vectors) to higher elevations (Freed et al., 2005; Sehgal, 2010; Garamszegi, 2011; Samuel et al., 2011; Imura et al., 2012). A better understanding of disease transmission dynamics in a spatial aspect can help improve biodiversity conservation plans across the multitude of Philippine protected areas,

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ISSN 2345-7600 (electronic) | ISSN 0217-2445 (print)

such as Mt. Banahaw–San Cristobal Protected Landscape (MBSCPL) in Southern Luzon, Philippines.

Our objective is to identify avian haemosporidians associated with understorey birds from Mt. Banahaw de Lucban (one of four peaks within MBSCPL) at high and low elevations, and to determine the relationship between avian haemosporidian prevalence among their bird hosts and elevation.

## MATERIAL AND METHODS

Birds were sampled with mist nets in two sites along the northeastern slope of Mt. Banahaw de Lucban, the northeastern peak of Mt. Banahaw volcano complex (highest peak at 2,170 m) within MBSCPL from 26 May–12 June 2013. This period was approaching the end of the dry season, and coincided with the breeding/nesting period of many of the birds surveyed (Kennedy et al., 2000). The high altitude sampling site was at an elevation of 1500–1600 m (14°4’N 121°30’E) and the low altitude sampling site ranged from 745–800 m (14°5’N 121°31’E). The boundary of the protected area is at 745 m, making it the lowest possible sampling area. The 1500–1600 m sampling area was selected based on the recommendation of field guides as the highest possible elevation range that can maximise trapping success. Sampling at high and low elevation consisted of 9 and 7 net days, respectively. Nets were opened from 0500–1700 h. Birds were identified following the taxonomy and nomenclature in Kennedy et al. (2000). Each captured bird was marked by clipping its rightmost tail feather, and morphometric data were gathered. The brachial vein of each bird was pricked using a sterile 30-gauge needle. A few drops of blood were collected in heparinised capillary tubes and then smeared onto a clean glass slide. Blood smears (2–3 slides) were prepared for each bird. The bird was then released. Blood smears were air dried and fixed with absolute methanol for 3–5 minutes. Procedures on storage, and staining of blood smears followed those by Valkiūnas et al. (2008a). Each blood film was examined for parasites using a standard light microscope. Digital photographs of parasites were taken using a Lenovo Q350 USB micronCAM fitted into the microscope eyepiece. Approximately 100 fields each were examined for low (×400) and high (×1000) magnifications for a total of at least 200 fields (Valkiūnas et al., 2008a). Unconfirmed infections were characterised by parasite morphology that can resemble either very young *Haemoproteus* gametocytes or *Plasmodium* trophozoites. The relative intensity of infection was determined as described by Valkiūnas (2005); briefly, +, and ++, for 1–10, and 11–100 parasites in 100 thin film fields, respectively, and +++, and ++++, for 1–10, and >10 parasites in one thin film field, respectively. For the purposes of discussion, +, ++, +++, and ++++, corresponds to low, moderate, high, and very high, respectively. The open source software Quantitative Parasitology 3.0 (Reiczigel & Rózsa, 2005) was used to test if there was a significant difference in parasite prevalence between the sampling sites. Specifically, Chi-square was used to compare prevalence values of the two elevations.

## RESULTS

**Prevalence of parasites.** A total of 95 birds representing 24 species, 15 families, and five orders, were captured in Mt. Banahaw de Lucban (Figs. 2, 3). There were no recaptures. Philippine endemics comprised 62.5% (n=15) of captured species, and none are endangered. All captured individuals were screened for blood parasites. Hematozoa from the genera *Plasmodium* (*P*), *Haemoproteus* (*H*), and *Leucocytozoon* (*L*) were detected. Bird species that have been screened for the first time include *Batrachostomus septimus*, *Loriculus philippensis*, *Dicaeum hypoleucum*, *Eumyias panayensis*, *Pachycephala albiventris*, *Stachyris whiteheadi*, *Ficedula hyperythra*, *Turdus poliocephalus*, *Zoothera andromedae*, and *Brachypteryx montana.* Among those screened for the first time, *F. hyperythra*, *Z. andromedae*, *S. whiteheadi*, and *T. poliocephalus* were infected with avian haemosporidians. Other infected birds include *Parus elegans* and *Phylloscopus cebuensis*, both of which were diagnosed as uninfected in previous studies. We report these six species as new host records for avian haemosporidia. Infected birds comprised 27 of 95 (28.4%) individuals, representing 12 of 24 (50%) species, 10 of 15 (66.7%) families, and three of five (60%) orders. *Haemoproteus* was the most commonly encountered parasite genus with a prevalence of 9.5%, followed by *Leucocytozoon* with 7.4%, *Plasmodium* with 4.2%, unconfirmed *Plasmodium* and/or *Haemoproteus* (*P/H*) with 3.2%, and multiple infections (*Haemoproteus* and *Leucocytozoon,* or *Plasmodium* and *Leucocytozoon*) with 4.2%. *Haemoproteus* infections were observed in three species belonging to three families, and were most prevalent in Dicruridae and Timaliidae (57% and 46.2% respectively). Six species belonging to four families were positive for *Leucocytozoon*, which was most prevalent in families Alcedinidae and Timaliidae (75% and 30.8%, respectively). Four species belonging to four families were found positive for *Plasmodium*. Unconfirmed *Plasmodium* and/or *Haemoproteus* infections (n=3) were found in two families represented by two species of birds. The absence of diagnostic stages had made identification to genus level impossible with only microscopy. Mixed infections were detected in four birds (Table 1). Dicruridae, Alcedinidae, and Timaliidae, have the highest prevalence of haemosporidians at 85.7%, 75%, and 61.5%, respectively. Muscicapidae had the lowest prevalence at 9.5% (Table 2). Birds trapped at 1500–1600 m had a prevalence of 35.5%; whereas birds trapped at 745–800 m had a prevalence of of 25% (Table 3).

**Relative intensity of parasites.** The majority of infections were of low intensity, regardless of whether they were single or mixed infections. *Haemoproteus* was the only genus observed at high intensity (two individuals).

## DISCUSSION

**Prevalence of haemosporidians.** The relatively high overall prevalence suggests that the corresponding arthropod vectors of the three haemosporidian genera are present in the sampling areas since many of the bird species examined are non-migratory. It should be noted, however, that both the

Table 1. Summary of 24 species from 15 families screened for haemosporidians

| **Host Species** | **Common Name** | **N** | **(+)** | **Parasite**[a] | | | | |
|---|---|---|---|---|---|---|---|---|
| | | | | **_P_** | **_H_** | **_L_** | **_P/H_** | **MI** |
| **ALCEDINIDAE** | | **4** | **3** | | | | | |
| _Actenoides lindsayi_* | Spotted wood kingfisher | 3 | 2 | | | 1 | | 1_PL_ |
| _Halcyon smyrnensis_ | White-throated kingfisher | 1 | 1 | | | 1 | | |
| **COLUMBIDAE** | | **10** | **1** | | | | | |
| _Chalcophaps indica_ | Emerald dove | 1 | 0 | | | | | |
| _Phapitreron leucotis_* | White-eared brown dove | 9 | 1 | | | 1 | | |
| **PODARGIDAE** | | **1** | **0** | | | | | |
| _Batrachostomus septimus_* | Philippine frogmouth | 1 | 0 | | | | | |
| **PSITTACIDAE** | | **1** | **0** | | | | | |
| _Loriculus philippensis_* | Philippine hanging parrot | 1 | 0 | | | | | |
| **DICAEIDAE** | | **3** | **0** | | | | | |
| _Dicaeum hypoleucum_* | Buzzing flowerpecker | 3 | 0 | | | | | |
| **DICRURIDAE** | | **7** | **6** | | | | | |
| _Dicrurus balicassius_* | Balicassiao | 7 | 6 | | 4 | | 2 | |
| **MUSCICAPIDAE** | | **21** | **2** | | | | | |
| _Eumyias panayensis_ | Mountain verditer flycatcher | 3 | 0 | | | | | |
| _Ficedula hyperythra_† | Snowy-browed flycatcher | 4 | 2 | 2 | | | | |
| _Rhipidura cyaniceps_* | Blue-headed fantail | 14 | 0 | | | | | |
| **PACHYCEPHALIDAE** | | **9** | **0** | | | | | |
| _Pachycephala albiventris_* | Green-backed whistler | 8 | 0 | | | | | |
| _Pachycephala philippinensis_* | Yellow-bellied whistler | 1 | 0 | | | | | |
| **PARIDAE** | | **2** | **1** | | | | | |
| _Parus elegans_*† | Elegant tit | 2 | 1 | 1 | | | | |
| **SYLVIIDAE** | | **3** | **1** | | | | | |
| _Phylloscopus cebuensis_*† | Lemon-throated leaf warbler | 3 | 1 | | | | 1 | |
| **PYCNONOTIDAE** | | **5** | **1** | | | | | |
| _Hypsipetes philippinus_* | Philippine bulbul | 4 | 1 | | 1 | | | |
| _Pycnonotus goiavier_ | Yellow-vented bulbul | 1 | 0 | | | | | |
| **SITTIDAE** | | **2** | **1** | | | | | |
| _Sitta frontalis_* | Velvet-fronted nuthatch | 2 | 1 | 1 | | | | |
| **TIMALIIDAE** | | **13** | **8** | | | | | |
| _Stachyris whiteheadi_*† | Chestnut-faced babbler | 13 | 8 | | 4 | 2 | | 2_HL_ |
| **TURDIDAE** | | **10** | **3** | | | | | |
| _Brachypteryx montana_ | White-browed shortwing | 7 | 0 | | | | | |
| _Turdus poliocephalus_† | Island thrush | 1 | 1 | | | | | 1_HL_ |
| _Zoothera andromedae_† | Sunda ground thrush | 2 | 2 | | | 2 | | |
| **ZOSTEROPIDAE** | | **4** | **0** | | | | | |
| _Zosterops nigrorum_* | Yellowish white-eye | 3 | 0 | | | | | |
| _Zosterops montanus_ | Mountain white-eye | 1 | 0 | | | | | |
| **TOTAL** | | **95** | **27** | **4** | **9** | **7** | **3** | **4** |

*Philippine endemic; †New host record; N = No. of individuals; (+) = No. of infected individuals; [a] _P_ = _Plasmodium_ spp.; _H_ = _Haemoproteus_ spp.; _L_ = _Leucocytozoon_ spp.; _P/H_ = unconfirmed _Plasmodium_ and/or _Haemoproteus_; MI = Mixed infection

Table 2. Prevalence of all haemosporidians within infected bird families

| Host Family | Common Name | # Screened | # Positive | Prevalence |
|---|---|---|---|---|
| Alcedinidae | Kingfishers | 4 | 3 | 75% |
| Columbidae | Doves and pigeons | 10 | 1 | 10% |
| Dicruridae | Drongos | 7 | 6 | 85.7% |
| Muscicapidae | Flycatchers and Fantails | 21 | 2 | 9.5% |
| Paridae | Tits | 2 | 1 | 50% |
| Pycnonotidae | Bulbuls | 5 | 1 | 20% |
| Sittidae | Nuthatches | 2 | 1 | 50% |
| Sylviidae | Leaf warblers | 3 | 1 | 33.3% |
| Timaliidae | Babblers | 13 | 8 | 61.5% |
| Turdidae | Thrushes | 10 | 3 | 30% |

Table 3. Prevalence of Haemosporidians by Altitude

| Altitude | # Screened | # Positive | Overall Prevalence (%) | Parasite Prevalence (%)[a] | | | | |
|---|---|---|---|---|---|---|---|---|
| | | | | P | H | L | P/H | MI |
| High | 31 | 11 | 35.5 | 3.2 | 12.9 | 9.7 | 0.0 | 9.7 |
| Low | 64 | 16 | 25 | 4.7 | 7.8 | 6.3 | 4.7 | 1.6 |
| TOTAL | 95 | 27 | 28.4 | 4.2 | 9.5 | 7.4 | 3.2 | 4.2 |

[a] *P* = *Plasmodium* spp.; *H* = *Haemoproteus* spp.; *L* = *Leucocytozoon* spp.; *P/H* = unconfirmed *Plasmodium* and/or *Haemoproteus*; MI = Mixed infection

small sampling size and relatively short sampling period in this study might have only provided estimates for prevalence and relative intensity. The prevalence of vector-borne diseases is also affected by seasonal variations and abiotic factors such as temperature, precipitation, and humidity, all of which can vary within a single forest (LaPointe et al., 2012). In general, warmer temperatures and abundant rainfall contribute to increased prevalence of vector-borne diseases such as hemosporidiosis and malaria because these conditions provide optimal breeding conditions for vectors, and higher rates of parasite development (Garamszegi, 2011; Imura et al., 2012; Zamora-Vilchis et al., 2012). Ahumada et al. (2004) presented a population model showing temperature and rainfall as main determinants of *C. quinquefasciatus* population abundance. *Plasmodium* development can occur between 16–30°C and is inhibited in temperatures lower than 16°C (Zamora-Vilchis et al., 2012). In this study, average morning and afternoon temperatures at 1500–1600 m netting stations during the time of sampling were 19.7°C and 19.4°C respectively. For netting stations at 745–800 m average morning and afternoon temperatures were 22.9°C and 23.3°C respectively. Sampling of vertebrate hosts occurred approaching the end of the dry season, but with incidences of sporadic rainfall.

*Haemoproteus* infections were observed to be the most common, followed by *Leucocytozoon*, and lastly, *Plasmodium.* Similar results were reported in different studies with several plausible explanations (Rodriguez & Matta, 2001; Beadell et al., 2004; del Cerro et al., 2010; Marzal & Albayrak, 2012; Silva-Iturriza et al., 2012; Zamora-Vilchis et al., 2012). Infections with *Plasmodium* produce the highest mortality among the three parasite genera in this study (Greiner & Ritchie, 1994; Savage et al., 2009; Atkinson & Samuel, 2010). In relation to this is the inherent bias of the mist-netting technique; susceptible individuals and those with acute infections become less active and are less likely to be captured in mist nets compared to their more resistant, hence, more mobile counterparts (Valkiūnas, 2005; Atkinson, 2008). *Haemoproteus* gametocytes persist longer in peripheral blood compared to *Leucocytozoon* and *Plasmodium*; some species of the latter two may be abundant in visceral blood or fixed tissues but rare or absent in peripheral blood, or may have highly periodic cycles or diurnal fluctuations in peripheral blood that render diagnostic stages undetectable by microscopy (Fallon et al., 2003; Valkiūnas, 2005, 2008a; Bunbury et al., 2006; Kilpatrick et al. 2006; Paperna et al., 2008). *Haemoproteus* and *Leucocytozoon* are transmitted by vector species belonging to families Hippoboscidae (louse flies) and Simuliidae (black flies), which are more ornithophilic and are capable of crawling beneath feathers to blood feed (Valkiūnas, 2005; Savage et al., 2009; Atkinson, n.d). Some vector species were also reported to have spatial preferences within a forest, with some species having preference for the canopy, others at ground level, and some with no apparent preference (Atkinson, n.d.; Černý et al*.,* 2011). The shortcomings of microscopy may have also contributed to observed prevalence values. Some infections are of very low intensity and may remain undetected despite extensive screening efforts (Valkiūnas et al., 2008a).

Prevalence of all haemosporidians among infected bird families ranged from 9.5% in Muscicapidae to 85.7% in Dicruridae, while some families had zero prevalence. Dicruridae and Alcedinidae have the highest prevalence values, similar to the findings of Silva-Iturriza et al. (2012). It was also observed that the same species that were diagnosed as negative came out as positive in this study, and vice versa (Silva-Iturriza et al., 2012). These findings suggest that these species are capable of being infected by the different haemosporidians despite the observed absence of parasites in certain species in either study. Similarly, they found no incidence of infection in family Columbidae, while in this study only one individual was infected. This is especially surprising since members of this family are known to have high infection rates (Silva-Iturriza et al*.,* 2012; Atkinson, n.d.). Absence of infection observed in certain families may be explained by little to no vectors that specialise on these bird groups within the study site, or that some species are simply less susceptible to infection, although these have to be confirmed (Atkinson, n.d.; Silva-Iturriza et al., 2012). Sample sizes however were highly varied and caution must be taken when comparing these prevalence values. Some aspects of vertebrate host morphology, ecology, and behavior may also influence their chances of being infected (Atkinson, n.d.; Valkiūnas, 2005). These include the size of the bird, the nest architecture, flock behavior, defensive behavior, mobility, and niche preference in the different forest layers (Valkiūnas 2005; Darbro & Harrington, 2007; Marzal et al*.,* 2008; Černý et al*.,* 2011). Smaller sized birds have reduced probability of infection, perhaps due to a smaller effective surface area for blood feeding, and that larger sized birds may be more attractive to vectors (Valkiūnas, 2005). Birds that are more stationary or inactive are also said to have an increased chance of infection (Valkiūnas, 2005). Flock or colonial behavior allows for closer proximity among bird hosts, thereby increasing chances of infection (Valkiūnas 2005; Marzal et al*.,* 2008). Closed types of nests, and those that are found close to the ground, are somehow protected from certain vector species (Atkinson, n.d.; Valkiūnas, 2005; Černý et al*.,* 2011). Each of these factors cannot exclusively explain the observed results because in natural settings they may work simultaneously (synergistically, antagonistically) to influence the spread of infection. An example is *S. whiteheadi,* which is a small and highly active passerine species that occurs in flocks. Although its size and level of activity may effectively reduce transmission of the disease, perhaps flock behavior, and/or another unknown factor, was a more predominant determinant in the spread of infection in this species.

**Comparison of prevalence between elevations.** Despite the seemingly large difference in prevalence between altitudes, the result was not statistically significant ($\chi^2 = 1.128$, $p = 0.288$). Further analyses on vector, bird, and parasite biology, and the environmental factors that influence their life histories are needed to better understand haemosporidian prevalence and distribution. Results of several studies were contrary to what was observed perhaps due to latitudinal differences of the chosen study sites (Ishtiaq et al., 2007; Marzal & Albayrak, 2012; Zamora-Vilchis et al., 2012). These were situated in either higher or lower latitudes with respect to the Philippines, thus having relatively cooler climates. It is likely that sampling should have taken place at higher elevations for reported trends to be evident, or trends in prevalence may altogether be different in tropical areas. Specific differences in each sampling area may also influence the transmission of hemosporidiosis. Sampling areas in 745–800 m were near the edge of the protected area. The immediate perimeter surrounding Mt. Banahaw de Lucban, outside the protected area of MBSCPL was composed of various agricultural lands and built-up areas including telecommunications infrastructures and a local university. Similar studies reported overall higher prevalence values of haemosporidians in undisturbed habitats (Matta et al., 2004; Bonneaud et al., 2009; Chasar et al., 2009). Higher pollution in disturbed areas may have negative impacts leading to less suitable vector breeding grounds, or undersampling of susceptible avian hosts due to reduced host fitness. It is of note that the prevalence for *Haemoproteus* and *Leucocytozoon* is higher in high elevation, while the prevalence of *Plasmodium* is higher in low elevation (Fig. 1). Similar findings were observed in *Parus major* populations in Switzerland (van Rooyen et al*.,* 2013b). This was said to possibly be due to the fact that *Leucocytozoon* parasites are able to complete sporogony at lower temperatures. In this study, very small samples sizes could no longer allow further statistical analysis, but the plausibility of an altitudinal variation in parasite prevalence merits further examination.

**Mixed infections.** A total of four birds were diagnosed with multiple infections of *Haemoproteus* and *Leucocytozoon* (n=3), and *Plasmodium* and *Leucocytozoon* (n=1). Only mixed infections of different parasite genera have been considered in this study. Multiple infections, particularly those of congeneric haemosporidia or morphologically similar taxa are also probable but may have been overlooked due to the limitations of morphological analysis. It was observed that the common parasite involving mixed infections was with *Leucocytozoon*; van Rooyen et al. (2013a) attributed this similar observation to the likelihood of *Leucocytozoon* being an opportunistic parasite that can best establish itself in a

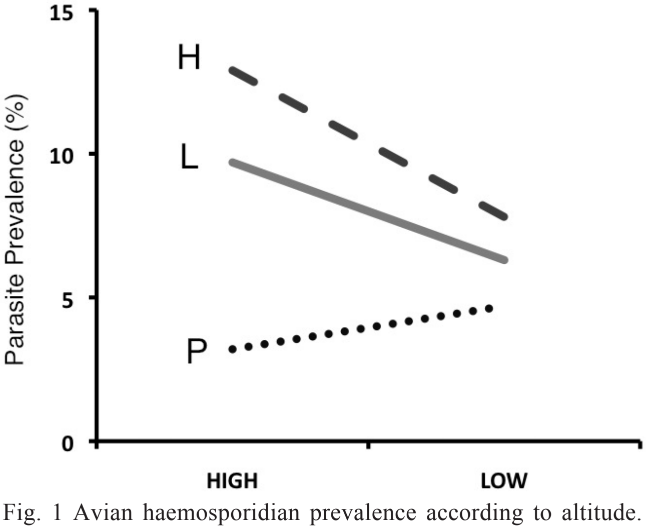

Fig. 1 Avian haemosporidian prevalence according to altitude. P=*Plasmodium*, L=*Leucocytozoon*, H=*Haemoproteus*.

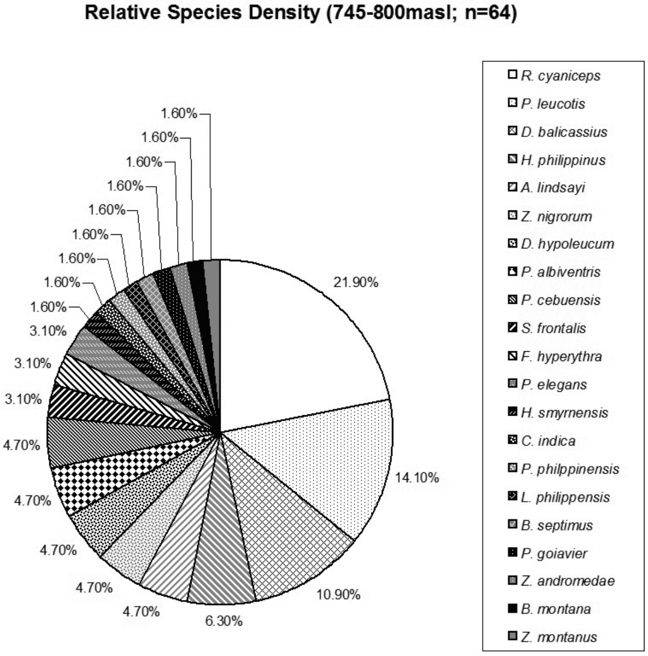

Fig. 2. Species composition of birds captured in 745–800 m sampling area

host system already weakened by a prior infection. Multiple infections may induce resource competition within the host, and these may have cumulative effects on host physiology, and/or trigger immune host immune response, thus potentially selecting for more virulent parasites (Marzal et al., 2008; del Cerro et al., 2010; Silva-Iturriza et al., 2012; Lachish et al., 2011; van Rooyen et al., 2013a). Infected birds often maintain the parasites for years or even throughout life; species with mixed infections such as *Stachyris whiteheadi*, *Actenoides lindsayi*, and *Turdus poliocephalus* may serve as reservoirs for more virulent lineages of haemosporidians, although this has yet to be confirmed.

**Relative intensity of infection.** Low parasitemias in peripheral blood smears suggest chronic infections, whereas high parasitemias likely indicate recent infections (Jarvi et al., 2003; Beadell et al., 2004; Freed et al., 2005; Ishtiaq et al., 2007; Valkiūnas et al., 2008a, b; Krams et al., 2012). The presence of moderate (*Leucocytozoon*, *Haemoproteus*) and high (*Haemoproteus*) intensity infections can perhaps be explained by a possible relapse of parasitemia that coincides with the vertebrate host breeding season, or frequent reinfection (Valkiūnas, 2005; Savage et al*.,* 2009).

**Conservation implications.** Although the prevalence of haemosporidians is relatively high, the majority of parasitemias are of low intensity. This is quite typical in wildlife assemblages, and as such the perpetuation of the disease is assured since these birds are reservoir hosts that serve as potential sources of new infections (Valkiūnas, 2005). In the Philippines, where hemosporidiosis naturally occurs, it is possible that its associated avian hosts have developed immunotolerance to maintain parasite intensity to chronic levels. But when hosts are immunocompromised, seemingly benign parasitic diseases may become problematic (Wobeser, 2008). Our findings suggest that climate change provides just a fraction of the whole picture on avian haemosporidian transmission. Pristine areas are often thought to be sanctuaries for threatened species, but this may soon change depending on how birds, vectors, and parasites respond to environmental changes. Human-induced disturbances in natural ecosystems may increase the risk of epizootic outbreaks in wild populations by compounding on the effects of other selection pressures on wildlife (Bonneaud, 2009). The current design in many Philippine protected areas, in which they have been reduced to island refugia surrounded by anthropogenic pressures, may also intensify the risk of outbreak by virtue of genetic drift (Whiteman et al., 2006; Jamieson et al., 2008). Hypothetically, wildlife populations become isolated and are unable to diversify their existing gene pools that allow for the selection of traits related to parasite resistance. Reducing avoidable human-induced pressures on pristine environments such as Mt. Banahaw de Lucban (within MBSCPL) may help ensure that infection intensities in avifauna are kept at bay.

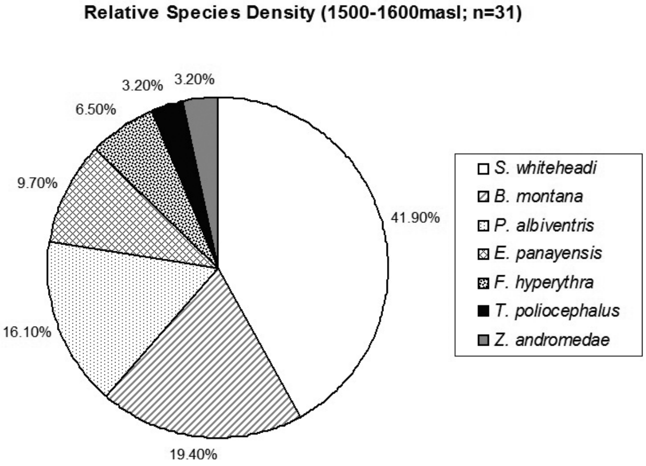

Fig. 3. Species composition of birds captured in 1500–1600 m sampling area

## ACKNOWLEDGEMENTS

We would like to thank Mario Nañola, George Rosalda, Glen Reantaso, and Severino Salvosa for their assistance in the field. We also thank the Department of Environment and Natural Resources, Southern Luzon State University, and the IACUC for the approval of the protocols and issuance of permits to catch birds. Special thanks to Commissioner Louie Palmera of the National Police Commission and Major Calma of the Philippine National Police for ensuring the team's security on field, and Prof. Noel Sabino of Institute if Biological Sciences (UPLB) for the free use of his USB digital microscope camera. We also thank Dr. Robert Adlard of the Queensland Museum, and Dr. Billy Divina and Dr. Salcedo Eduardo of the UPLB College of Veterinary Medicine, for their advice on parasite identification.

## LITERATURE CITED

Ahumada JA, Lapointe D & Samuel MD (2004) Modeling the population dynamics of *Culex quinquefasciatus* (Diptera: Culicidae), along an elevational gradient in Hawaii. Journal of Medical Entomology, 41(6): 1157–1170.

Atkinson CT (n.d.) Hemosporidosis. In: Field Manual of Wildlife Diseases:Birds. http://www.nwhc.usgs.gov/publications/field_manual/chapter_24.pdf

Atkinson CT (2008). Avian malaria. In: Atkinson CT, Thomas NJ & Hunter DB (eds) Parasitic Diseases of Wild Birds. Wiley-Blackwell: USA. Pp. 35–53.

Atkinson CT & Samuel MD (2010) Avian malaria *Plasmodium relictum* in native Hawaiian forest birds: epizootiology and demographic impacts on `apapane *Himatione sanguinea*. Journal of Avian Biology, 41: 357–266.

Beadell JS, Gering E, Austin J, Dumbacher JP, Peirce MA, Pratt TK, Atkinson CT & Fleischer RC (2004) Prevalence and differential host-specificity of two avian blood parasite genera in the Australo-Papuan region. Molecular Ecology, 13(12): 3829–3844.

Bonneaud C, Sepil I, Milá B, Buermann W, Pollinger J, Sehgal RNM, Valkiūnas G, Iezhova TA, Saatchi S & Smith TB (2009) The prevalence of avian *Plasmodium* is higher in undisturbed tropical forests of Cameroon. Journal of Tropical Ecology, 25(4): 439–447.

Bunbury N, Barton E, Jones CG, Greenwood AG, Tyler KM & Bell DJ (2006) Avian blood parasites in an endangered columbid: *Leucocytozoon marchouxi* in the Mauritian Pink Pigeon *Columba mayeri*. Parasitology, 134(6): 794–808.

Černý O, Votýpka J & Svobodová M (2011) Spatial feeding preferences of ornithophilic mosquitoes, blackflies and biting midges. Medical and Veterinary Entomology, 25(1): 1–5.

Chasar A, Loiseau C, Valkiūnas G, Iezhova T, Smith TB & Sehgal RNM (2009) Prevalence and diversity patterns of avian blood parasites in degraded African rainforest habitats. Molecular Ecology, 18: 4121–4133.

Darbro J & Harrington L (2007) Avian defensive behavior and blood-feeding success of the West Nile vector mosquito, *Culex pipiens*. Behavioral Ecology, 18(4): 750–757.

del Cerro S, Merino S, de la Puente JM, Lobato E, Ruiz-de-Castañeda R, Rivero-de Aguilar J, Martínez J, Morales J, Tomás G & Moreno J (2010) Carotenoid-based plumage coloration is associated with blood parasite richness and stress protein levels in blue tits (*Cyanistes caeruleus*). Oecologia, 162(4): 825–835.

Directo CC (1969) A Preliminary Study of the Mosquito Intermediate Host of *Plasmodium juxtanucleare* Versianni and Gomes, 1941 in the Philippines. Unpublished Thesis. University of the Philippines, Los Baños, Laguna.

Dy FJ & Soriano LJ (1940) The incidence of *Plasmodium* and *Haemoproteus* infections in some Philippine birds. Philippine Journal of Science, 72(3): 251–253.

Eduardo SL (2007) Synopsis of the Parasites of Birds in the Philippines. National Academy of Science and Technology Phils., Department of Science and Technology, University of the Philippines Los Baños, Philippines, 390 pp.

Fallon SM, Ricklefs RE, Swanson BL & Bermingham E (2003) Detecting avian malaria: an improved polymerase chain reaction diagnostic. The Journal of Parasitology, 89(5): 1044–1047.

Freed LA, Cann RL, Goff ML, Kuntz WA & Bodner GR (2005) Increase in avian malaria at upper elevation in Hawai'i. The Condor, 107: 753–764.

Garamszegi LZ (2011) Climate change increases the risk of malaria in birds. Global Change Biology, 17: 1751–1759.

Greiner EC & Ritchie BW (1994) Parasites. In Ritchie BW, Harrison GJ & Harrison LR (eds.) Avian Medicine: Principles and Application*,* Wingers Publishing, Inc., Lake Worth, FL. Pp. 1007–1029.

Imura T, Suzuki Y, Ejiri H, Sato Y, Ishida K, Sumiyama D, Murata K & Yukawa M (2012) Prevalence of avian haematozoa in wild birds in a high-altitude forest in Japan. Veterinary Parasitology, 183(3–4): 244–248.

Ishtiaq F, Gering E, Rappole JH, Rahmani AR, Jhala YV, Dove CJ, Milensky C, Olson SL, Peirce MA & Fleischer RC (2007) Prevalence and diversity of avian hematozoan oarasites in Asia: a regional survey. Journal of Wildlife Diseases, 43(3): 382–398.

Jamieson IG, Grueber CE, Waters JM & Gleeson DM (2008) Managing genetic diversity in threatened populations: a New Zealand perspective. New Zealand Journal of Ecology, 32(1): 130–137

Jarvi SI, Farias MEM, Baker H, Freifeld HB, Baker PE, Gelder EV, Massey JG & Atkinson CT (2003) Detection of avian malaria (*Plasmodium* spp.) in native land birds of American Samoa. Conservation Genetics, 4: 629–637.

Kennedy RS, Gonzales PC, Dickinson EC, Miranda HC & Fisher TH (2000) A Guide to the Birds of the Philippines. Oxford University Press Inc., New York, 369 pp.

Kilpatrick AM, LaPointe DA, Atkinson CT, Woodworth BL, Lease JK, Reiter ME & Gross K (2006) Effects of chronic avian malaria (*Plasmodium relictum*) infection on reproductive success of Hawaii amakihi (*Hemignathus virens*). The Auk, 123(3): 764–774.

Krams I, Suraka V, Cīrule D, Hukkanen M, Tummeleht L, Mieraukas P, Rytkönen S, Rantala MJ, Vrublevska J, Orell M & Krama T (2012) A comparison of microscopy and PCR diagnostics for low intensity infections of haemosporidian parasites in the Siberian tit *Poecile cinctus*. Annales Zoologici Fennici, 49(5/6): 331–340.

Lachish S, Knowles SCL, Alves R, Wood MJ & Sheldon BC (2011) Fitness effects of endemic malaria infections in a wild bird population: the importance of ecological structure. Journal of Animal Ecology, 80(6): 1196–1206.

LaPointe DA, Atkinson CT & Samuel MD (2012) Ecology and conservation biology of avian malaria. Annals of the New York Academy of Sciences, 249(2012): 211–226.

Loiseau C, Harrigan RJ, Cornel AJ, Guers SL, Dodge M, Marzec T, Carlson JS, Seppi B & Sehgal RNM (2012) First evidence and predictions of plasmodium transmission in Alaskan bird populations. PLoS ONE, 7(9): e44729.

Marzal A, Bensch S, Reviriego M, Balbontin J & De Lope F (2008) Effects of malaria double infection in birds: one plus one is not two. Journal of Evolutionary Biology, 21(4): 979–987.

Marzal A & Albayrak T (2012) Geographical variation of haemosporidian parasites in Turkish populations of Krüper's Nuthatch *Sitta krueperi*. Journal of Ornithology, 153(4): 1225–1231.

Matta NE, Basto N, Gutierrez R, Rodriguez OA & Greiner EC (2004) Prevalence of blood parasites In: Tyrannidae (Flycatchers) in the Eastern Plains of Colombia. Memórias do Instituto Oswaldo Cruz, 99 (3): 271–274.

Paperna I, Keong MSC & May CYA (2008) Haemosporozoan parasites found in birds in Peninsular Malaysia, Singapore, Sarawak and Java. The Raffles Bulletin of Zoology, 56(2): 211–243.

Reiczigel J & Rózsa L (2005) Quantitative Parasitology 3.0. Reiczigel J, & Rózsa L, Budapest. http://www.zoologia.hu/qp/qp.html (Accessed 28 January 2014).

Rodriguez OA & Matta NE (2001) Blood parasites in some birds from eastern plains of Colombia. Memórias do Instituto Oswaldo Cruz, 96(8): 1173–1176.

Russell PF (1932a) Avian Malaria Studies, IV: *Haemoproteus* and *Plasmodium* In: Birds of Luzon Philippine Islands. Philippine Journal of Science, 48(2): 263–268.

Russell PF (1932b) Avian malaria studies, V: "*Plasmodium capistrani*" sp. nov., an avian malaria parasite in the Philippines. Philippine Journal of Science, 48(2): 269–287

Samuel MD, Hobbelen PHF, DeCastro F, Ahumada JA, LaPointe DA, Atkinson CT, Woodworth BL, Hart PJ & Duffy DC (2011) The dynamics, transmission, and population impacts of avian malaria in native Hawaiian birds: a modeling approach. Ecological Applications, 21(8): 2960–2973.

Savage AF, Robert V, Goodman SM, Raharimanga V, Raherilalao MJ, Andrianarimisa A, Ariey F & Greiner EC (2009) Blood Parasites in Birds from Madagascar. Journal of Wildlife Diseases, 45(4): 907–920.

Sehgal RNM (2010) Deforestation and avian infectious diseases. The Journal of Experimental Biology, 213: 955–960.

Silva-Iturriza A, Ketmaier V & Tiedemann R (2012) Prevalence of avian haemosporidian parasites and their host fidelity in the central Philippine islands. Parasitology International, 61: 650–657.

Valkiūnas G (2005) Avian malaria parasites and other haemosporidia. CRC Press, Boca Raton, Florida, 946 pp.

Valkiūnas G, Iezhova TA., Križanauskienė A, Palinauskas V, Sehgal RNM & Bensch S (2008a) A Comparative Analysis of Microscopy and PCR-Based Detection Methods for Blood Parasites. Journal of Parasitology, 94(6): 1395–1401.

Valkiūnas G, Iezhova TA, Loiseau C, Chasar A, Smith TB & Sehgal RNM (2008b) New species of haemosporidian parasites (Haemosporida) from African rainforest birds, with remarks on their classification. Parasitology Research, 103(5): 1213–1228.

van Rooyen J, Lalubin F, Glaizot O & Christe P (2013a) Avian haemosporidian persistence and co-infection in great tits at the individual level. Malaria Journal, 12(40): 1–8.

van Rooyen J, Lalubin F, Glaizot O & Christe P (2013b) Altitudinal variation in haemosporidian parasite distribution in great tit populations. Parasites & Vectors, 6(139): 1–10.

Vidal OM (1968) A Study on the Occurrence of Blood Parasites in Philippine Ducks. Unpublished Thesis. University of the Philippines Los Baños, Laguna.

Whiteman NK, Matson KD, Bollmer JL & Parker PG (2006) Disease ecology in the Galapagos Hawk (*Buteo galapagoensis*): host genetic diversity, parasite load and natural antibodies. Proceedings of The Royal Society B, 273: 797–804.

Wobeser GA (2008) Parasitism: cost and effects. In: Atkinson CT, Thomas NJ & Hunter DB (eds) Parasitic Diseases of Wild Birds. Wiley-Blackwell, USA. Pp. 3–9.

Zamora-Vilchis I, Williams SE & Johnson CN (2012) Environmental temperature affects prevalence of blood parasites of birds on an elevation gradient: implications for disease in a warming climate. PLoS ONE, 7(6): 1–8.