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HAEMOPROTEUS IWA N. SP. IN GREAT FRIGATEBIRDS (*FREGATA MINOR* [GMELIN]) FROM HAWAII: PARASITE MORPHOLOGY AND PREVALENCE

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ABSTRACT: We describe a new species of *Haemoproteus* Kruse, 1890 from great frigatebirds (*Fregata minor* [Gmelin]) captured on Tern Island–French Frigate Shoals and Laysan Island in Hawaii. Parasite prevalence on Laysan Island (35%) was not significantly different than that of Tern Island (36%). On Laysan, prevalence was highest in juveniles (52%), followed by adult males (29%) and adult females (19%). Prevalence on Tern was 36% both for adult females and juveniles, and 28% for adult males. Parasitemia was low (mean < 2 parasites/10,000 red blood cell). Parasitized red cells had significantly greater areas than unparasitized cells. We named this parasite *Haemoproteus iwa* after the Hawaiian name for frigatebirds (iwa). This is the first documentation of a hemoparasite from tropical pelagic seabirds in Hawaii and the first description of an endemic hemoparasite in the archipelago.

To date, the only documented host harboring *Haemoproteus* Kruse, 1890 in Hawaii is the pigeon (*Columbia livia* [Gmelin]) (Yager and Gleiser, 1946; Kartman, 1949). Among pelagic seabirds in Hawaii and worldwide, there appears to be a general absence of hemoparasites (Greiner et al., 1975; Bishop and Bennett, 1992; Bennett et al., 1994). There is an anecdotal description of a *Hepatozoon* Miller, 1908 in an undocumented species of albatross (Peirce and Prince, 1980). Lowery (1971) mentions infection by a species of *Haemoproteus* in great (*Fregata minor* [Gmelin]) and lesser (*Fregata ariel* [Gray]) frigatebirds from Aldabra Atoll, Indian Ocean, but gives little detail on prevalence by species or on parasite morphology.

We describe a new species of *Haemoproteus* from great frigatebirds (*F. minor*) captured on Laysan Island and Tern Island–French Frigate Shoals in Hawaii. Parasite morphology, prevalence, and effects on the host cell are also described.

MATERIALS AND METHODS

We captured 60 and 30 frigatebirds from Laysan and Tern Island, respectively, in March 1994, using hand nets. Birds were classified as adult females or males, or juveniles, based on plumage (Pratt et al., 1987) and bled from the cutaneous ulnar vein. We fixed thin blood smears in 100% methanol for 1 min, stained them with Giemsa (buffered to pH 7.2), and rinsed them in tap water.

Parasitemia was measured by counting 10,000 red cells under oil immersion (1,000×) for each smear. Using a calibrated ocular micrometer, we measured 31 parasitized, 30 unparasitized red cells, and 31 parasites according to the methods of Bennett and Campbell (1972). Additionally, we counted numbers of intracytoplasmic pigment granules in the parasite and calculated the nuclear displacement ratio (NDR), an index of lateral displacement of the host cell nucleus by the parasite (Bennett and Campbell, 1972). The area of parasitized and unparasitized host cell and nucleus was calculated using the formula for an ellipse πab: where a = major axis (length) and b = minor axis (breadth).

Prevalence of parasitized birds was calculated for island, age, and gender. Prevalence between islands was compared with the chi square test (Daniel, 1987). We compared the areas of parasitized vs. unparasitized red cell and red cell nucleus using student's *t*-test (Daniel, 1987). The level of significance for all tests was 0.05.

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RESULTS

Prevalence of *Haemoproteus iwa* in great frigatebirds on Laysan (21/60 [35%]) was not significantly different ($\chi^2 = 0.00606$, df = 1, $P = 0.9379$) than that on Tern (11/30 [37%]). On Laysan, prevalence in adult females, adult males, and juveniles was 4/21 (19%), 6/21 (29%), and 11/21 (52%), respectively. On Tern Island, prevalence in adult females and juveniles was 4/11 (36%) and 3/11 (28%) in adult males. Parasitized host cells had a significantly greater area than unparasitized cells ($t = 2.12$, df = 59, $P = 0.0382$). There was no difference between area of parasitized and unparasitized host cell nuclei ($t = -1.71$, df = 59, $P = 0.0921$) (Table I). Mean parasitemia was 1.8 ± 1.4 parasites/10,000 red cells and ranged from 1 to 7 parasites/10,000 red cells.

DESCRIPTION

Haemoproteus iwa n. sp.

Macrogametocyte (Fig. 1): Parasite is sausage-shaped to halteridial with a light blue finely granular cytoplasm; the ends of the parasite extend only partially around the host nucleus, and the parasite border is well defined. The parasite contains moderate numbers of small clear intracytoplasmic vacuoles scattered throughout that rarely coalesce into a larger single vacuole. The cytoplasm contains diffusely scattered brown to black pigment granules with a mean of 22 per parasite (Table I). The parasite is closely appressed to the host cell nucleus, usually occupies the entire cytoplasm on 1 side, and displaces the host cell nucleus to 1 side (mean NDR = 0.72). Mean parasite lengths and widths are 13.7 μm and 3.3 μm, respectively (Table I). The nucleus is round to oblong, pink, and is centrally to subterminally located. Mean parasite nucleus length and width are 2.3 μm and 1.8 μm, respectively (Table I).

Microgametocyte: None seen.

Immature parasites: Extremely rare (n = 2). These are round to irregular, pink to clear, measure ~2 μm diameter with 1–4 pigment granules, and are located near the pole or alongside the nucleus.

Avian host: Fregatidae: great frigatebird (*F. minor* [Gmelin]).

Locality: Type locality Laysan Island, Hawaii, U.S.A. (25°46′N 171°44′W). Parasites recorded from Tern Island (23°45′N 166°10′W) on French Frigate Shoals, Hawaii, U.S.A.

Type specimens: *Haemoproteus iwa* is described from parasites in an adult male great frigatebird (*F. minor*) from Laysan Island collected on 11 November 1993. Paratype material includes 32 slides from the same host; 21 collected from Laysan Island and 11 collected from Tern Island on French Frigate Shoals, Hawaii in March 1994. Parahapantotypes (accession nos. G212808, G212809, and G212810) were de-

TABLE I. Morphometrics of *Haemoproteus iwa*, number of granules in parasites, parasitized red blood cells (RBCs), RBC nuclei (n = 31), and unparasitized RBCs and RBC nuclei (n = 30).

	Mean ± SD	Range
Parasite		
Breadth (μm)	3.3 ± 0.5	2.5–4.0
Length (μm)	13.7 ± 1.9	8.5–16.0
Nucleus length (μm)	2.3 ± 0.7	1.5–4.5
Nucleus breadth (μm)	1.9 ± 0.4	1.0–3.0
No. granules	22 ± 6	7.0–32.0
Parasitized red cell		
Breadth (μm)	7.8 ± 0.6	7.0–9.5
Length (μm)	15.8 ± 0.8	14.0–17.5
Area (μm²)	386.5 ± 39.4	340.9–522.3
Parasitized red cell nucleus		
Breadth (μm)	2.2 ± 0.4	1.5–3.0
Length (μm)	6.7 ± 0.6	5.5–8.0
Area (μm²)	46.2 ± 9.0	30.6–70.7
NDR*	0.72 ± 0.17	0.22–1.00
Unparasitized red cell		
Breadth (μm)	7.7 ± 0.7	6.0–9.0
Length (μm)	15.3 ± 1.0	13.0–18.5
Area (μm²)	366.2 ± 35.4	301.6–452.4
Unparasitized red cell nucleus		
Breadth (μm)	2.4 ± 0.3	2.0–3.0
Length (μm)	6.7 ± 0.6	5.5–7.5
Area (μm²)	49.5 ± 5.5	37.7–58.9

* NDR = nuclear displacement ratio.

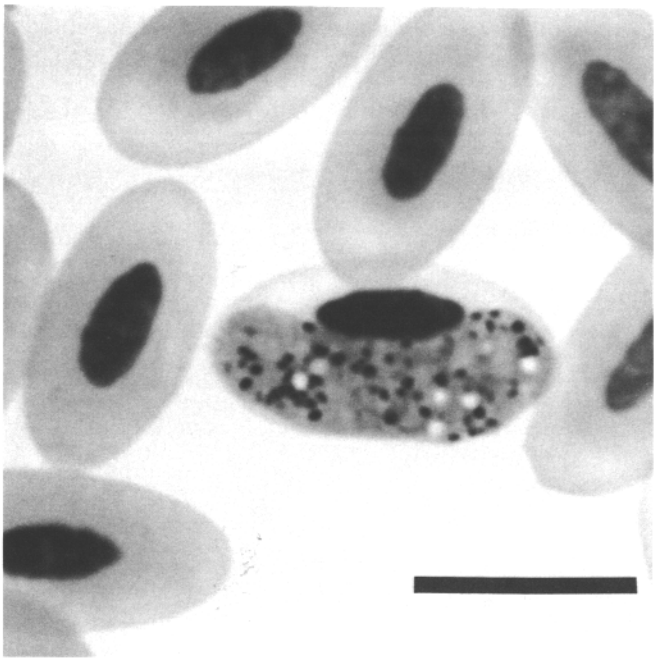

FIGURE 1. Great frigatebird red blood cell infected with a macrogametocyte of *Haemoproteus iwa*. Bar equals 10 μm.

posited at the International Reference Center for avian Haematozoa (Queensland Museum, Grey Street, South Brisbane, Queensland, 4101, Australia).

Etymology: The specific name refers to the Hawaiian name for the type host (iwa).

Remarks

Given the consistent parasite morphology in birds from different northwestern Hawaiian islands and the host, we believe we are dealing with 1 *Haemoproteus* species in great frigatebirds and not multiple species or genera. Morphology and presence of hematozoin pigment would rule out other sausage-shaped parasites such as *Hepatozoon* (Hawkey and Dennett, 1989). Species of *Plasmodium* Marchiafava and Celli, 1885 with elongate gametocytes such as *P. elongatum* Huff, 1930, *P. polare* Manwell, 1935 or *P. hexamerium* Huff, 1935 (Hewitt, 1940) rarely have thick-bodied forms. This, coupled with our inability to locate schizonts, would make *Plasmodium* unlikely; however, the most rigorous test for this hypothesis would necessitate either genetic or subinoculation studies. Also militating against *Plasmodium* is the lack of a suitable culicid vector in the islands where these birds were sampled (Nishida, 1992). Based on number of granules, shape, outline, and degree of displacement of host red cell nucleus, *H. iwa* would most closely resemble other avian haemoproteids such as *Haemoproteus nettionis* Johnston and Clelland, 1910, *Haemoproteus caprimulgi* Williams, Bennett & Mahrt, 1975, *Haemoproteus aegnithidae* de Mello, 1936 or *Haemoproteus witti* White, Bennett and Williams, 1979 (Bennett and Peirce, 1988). Our inability to find *Haemoproteus* microgametocytes is not too surprising considering the low parasitemia in the birds we examined; this is not a unique phenomenon (Greiner and Mundy, 1979).

Haemoproteus iwa is the first documentation of a hemoparasite in Fregatidae from Hawaii and the first native haematozoan from Hawaiian birds. Lowery (1971) documented 14% (9/62) prevalence of *Haemoproteus* in frigatebirds from Aldabra Atoll; however, his failure to distinguish between greater and lesser frigatebirds precludes comparisons with our data. Furthermore, he offered little detail on the parasite.

Frigatebirds in Hawaii that harbored the parasite appeared clinically healthy. It is unlikely that the parasite causes detectable pathology given the low parasitemia. To date, we have not detected schizogonic development of the parasite in 6 frigatebirds we necropsied (data not shown). We have been unable to demonstrate significant differences between parasitized and unparasitized birds when comparing hematology and blood chemistry (data not shown). The parasite causes slight, but significant, host cell hypertrophy; however, this is not a consistent characteristic of haemoproteid infections (Bennett and Campbell, 1972). Prevalence of the parasite on Tern and Laysan suggests *H. iwa* may be cosmopolitan as a clinically inapparent infection in great frigatebirds throughout the northwestern Hawaiian Islands.

Our data did not reveal the vector for this parasite. Haemoproteids are generally transmitted by ceratopogonid or hippoboscid biting flies (Atkinson, 1991). Hematophagous ceratopogonids have not been documented in Hawaii (Nishida, 1992), making them an unlikely vector. Like Lowery (1971), we noted several frigatebirds with hippoboscid infestations, presumably *Olfersia spinifera* (Leach) or *Olfersia anescens* Thomson (Maa, 1968); we judge these to be a more promising candidate. Other less likely vectors include ticks like *Ornithodoros capensis* Neumann (Garrett and Haramoto, 1967) or *Ixodes laysanensis* Wilson (Wilson, 1964). Interestingly, we have noted hippoboscids on species of seabirds of other families including Procellariidae, Phaethontidae, Sulidae, and Diomedeidae; however, we have never observed hemoparasites in any of these avian taxa (data not shown).

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LITERATURE CITED

ATKINSON, C. T. 1991. Vectors, epizootiology and pathogenesis of avian species of *Haemoproteus* (Haemosporina: Haemoproteidae). Bulletin of the Society of Vector Ecologists **16:** 109–126.

BENNETT, G. F., AND A. G. CAMPBELL. 1972. Avian haemoproteidae. I. Description of *Haemoproteus fallisi* n. sp. and a review of the haemoproteids of the family Turdidae. Canadian Journal of Zoology **50:** 1269–1275.

———, AND M. A. PEIRCE. 1988. Morphological form in the avian Haemoproteidae and an annotated checklist of the genus *Haemoproteus* Kruse, 1890. Journal of Natural History **22:** 1683–1686.

———, ———, AND R. A. EARLE. 1994. An annotated checklist of the valid avian species of *Haemoproteus, Leucocotyzoon* (Apicomplexa: Haemosporida) and *Hepatozoon* (Apicomplexa: Haemogregarinidae). Systematic Parasitology **29:** 61–73.

BISHOP, M. A., AND G. F. BENNETT. 1992. Host–parasite catalogue of the avian haematozoa supplement 1. Memorial University of Newfoundland Occasional Papers in Biology **15:** 1–244.

DANIEL, W. W. 1987. Biostatistics: A foundation for analysis in the health sciences. John Wiley & Sons, New York, New York, 734 p.

GARRETT, L. E., AND F. H. HARAMOTO. 1967. A catalog of Hawaiian Acarina. Proceedings of the Hawaiian Entomological Society **19:** 381–414.

GREINER, E. C., G. F. BENNETT, E. M. WHITE, AND R. F. COOMBS. 1975. Distribution of the avian hematozoa of North America. Canadian Journal of Zoology **53:** 1762–1787.

———, AND P. J. MUNDY. 1979. Hematozoa from southern African vultures, with a description of *Haemoproteus janovyi* sp. n. Journal of Parasitology **65:** 147–153.

HAWKEY, C. M., AND T. B. DENNETT. 1989. Color atlas of comparativ veterinary hematology. Iowa State University Press, Ames, Iowa 192 p.

HEWITT, R. 1940. Bird malaria. John Hopkins Press, Baltimore, Mary land, 228 p.

KARTMAN, L. 1949. Observations on the *Haemoproteus* of pigeons i Honolulu, Hawaii. Pacific Science **3:** 127–132.

LOWERY, R. S. 1971. Blood parasites of vertebrates on Aldabra. Philo sophical Transactions of the Royal Society of London B **260:** 577 580.

MAA, T. C. 1968. Records of Hippoboscidae (Diptera) from the Cen tral Pacific. Journal of Medical Entomology **5:** 325–328.

NISHIDA, G. M. 1992. Hawaiian terrestrial arthropod checklist. Bisho Museum, Honolulu, Hawaii, 262 p.

PEIRCE, M. A., AND P. A. PRINCE. 1980. *Hepatozoon albatrossi* sp nov. (Eucoccida: Hepatozoidae) from *Diomedea* spp. in the an tarctic. Journal of Natural History **14:** 447–452.

PRATT, H. D., P. L. BRUNER, AND D. G. BERRETT. 1987. A field guid to the birds of Hawaii and the Pacific. Princeton University Press Princeton, New Jersey, 454 p.

WILSON, N. 1964. *Ixodes laysanensis,* a new species of tick from bird on Laysan Island (Metastigmata: Ixodidae). Journal of Medica Entomology **1:** 165–168.

YAGER, R. H., AND C. A. GLEISER. 1946. *Trichomonas* and *Hemo proteus* infections and the experimental use of DDT in the contro of ectoparasites in a flock of signal corps pigeons in the Territor of Hawaii. Journal of the American Veterinary Medical Association **109:** 204–207.