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HAEMATOZOA IN BIRDS FROM LA MACARENA NATIONAL NATURAL PARK (COLOMBIA)

Hematozoarios en aves del Parque Nacional Natural La Macarena (Colombia)

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ABSTRACT
Birds from 69 species in 25 families were collected from La Macarena National Natural Park in Colombia between June and November 2000 and examined for haematozoa. Eighty-two of the 342 birds (24%) were positive for one or more taxon. Microfilariae were the most commonly seen parasites (10.5%) and *Leucocytozoon* the least common (0.3%). Other parasites were species of the genera *Plasmodium* (4.4%), *Trypanosoma* (3.5%), *Hepatozoon* (3.5%) and *Haemoproteus* (3.2%). The low intensity of haemosporidian parasites agreed with other records from the Neotropics. Parasite prevalence in this Neotropical region was higher than levels found in other surveys in the Neotropics, but lower than levels found for the Nearctic area. A new host-parasite association is reported here, as well as avian species examined for haematozoa for the first time.

Key words. Birds, Colombia, haematozoa, haemoparasite, infection, Neotropics, prevalence.

RESUMEN
Se recolectaron aves pertenecientes a 69 especies y 25 familias en el parque nacional natural Sierra de La Macarena (Colombia), de junio a noviembre del año 2000, y se examinaron para hematozoarios. Ochenta y dos de las 342 aves (24%) fueron positivos para uno o más hematozoarios. En los preparados sanguíneos de aves positivas, las microfilarias fueron los parásitos más comunes (10,5%), y *Leucocytozoon* el mas escaso (0,3%). Otros parásitos observados pertenecieron a los géneros de

Plasmodium (4,4%)*, Trypanosoma* (3,5%), *Hepatozoon* (3,5%) y *Haemoproteus* (3,2%). Las bajas intensidades de parásitos hemosporidianos están de acuerdo con otros registros en el neotrópico. La prevalencia de parásitos en esta región es mayor que los niveles encontrados en otros estudios en la región neotropical, pero menores que los niveles encontrados para el área neártica. Nosotros reportamos aquí nuevas asociaciones parásito-hospedero, también como especies aviares examinadas por primera vez para hematozoarios.

Palabras clave. Aves, Colombia, hematozoarios, hemoparásitos, infección, neotrópico, prevalencia.

INTRODUCTION

Protozoan knowledge is currently linked to their clinical and economic importance; there is detailed information regarding some coccidian or haemoflagellate species which infect mammals whilst research on avian and reptilian Haemosporidia is more limited. Avian haematozoa infection has been related to behavioural changes, sexual selection and the reduction and extinction of native bird populations (VanRiper III *et al.* 1986, Garvin & Remsen 1997, Buchanan *et al.* 1999). However, the paucity of research concerning the distribution, prevalence and pathogenicity of these parasites has limited recognition of their importance in tropical bird demography and veterinary wildlife management. Despite great avian diversity in Colombia and the strong influence of migrant birds, this kind of study is scarce (Renjifo *et al.* 1952, Bennett & Borrero, 1976, Valkiünas *et al.* 2003, Matta *et al.* 2004).

It has been demonstrated that the distribution patterns and frequency of avian haemoparasite infections from the Neartic region differ from those found in Central and South America (Greiner *et al.* 1975, White *et al.* 1978). The main aim of this research was to determine the prevalence of haemotropic parasites of birds from La Macarena national natural park (NNP) (located in Meta, Colombia) and compare the results with previous studies carried out in this region and the neotropics.

MATERIALS AND METHODS

This work was carried out at El Cerrillo station in the La Macarena NNP, Colombia, between June and November 2000. The study area lies at 400-500m and has vegetation which includes gallery forest and savannah. The area has a mean annual temperature of 24°C with a 2,973 mm annual rainfall occurring mainly between June and August. Birds were mist netted and bled by clipping their claws to produce three thin smears on glass slides. The smears were fixed with 100% methanol and stained with Giemsa (7.2) for 40 min. The stained blood smears were examined by light microscope. Each smear was scanned double-blind at low magnification (100x) (10 min) for establishing the presence of *Leucocytozoon*, *Trypanosoma* and microfilariae and using oil immersion (1,000x) (20 min) for *Plasmodium*, *Haemoproteus* and *Hepatozoon*. Intensity of infection for microfilariae, *Plasmodium*, *Haemoproteus* and *Leucocytozoon* were established by counting 100 fields at 1,000 magnification by moving the slide in areas where the blood cells formed a single monolayer field at 100x oil objective containing around 100 erythrocytes. The intensity of infection was thus determined as a percentage of infected cells or the number of parasites/10,000 erythrocytes (Muñoz *et al.*1999).

RESULTS

Results of microscopic examination of blood smears are shown in table 1.These birds represented 69 species classified into in 25 families and nine orders. Eighty-two birds from 31 species were infected with either single or mixed infections (93 infections altogether) of parasites from six genera. Overall prevalence of infection was 24%. Microfilariae were the most common parasites encountered, occurring in 36 of the samples (10.5%), followed by *Plasmodium* in 15 birds (4.4%), *Trypanosoma* and *Hepatozoon* (3.5%), *Haemoproteus* (3.2%), *Leucocytozoon* (0.3%) and others (1.7%). The 69 avian host species examined included 10 species which were examined for blood parasites for the first time. 10 new host-parasite associations were also recorded.

Most parasitaemias were low (0.02%-0.04%), suggesting chronic rather than acute disease. *Haemoproteus columbae* were only found in columbids; *Haemoproteus caprimulgi* occurred in the nightjar *Caprimulgus nigrescens*. Other haemoproteids found resembled *Haemoproteus thraupi* in the tanager *Ramphocelus carbo*, *Haemoproteus nisi* in the hawk *Buteo magnirostris* and *Haemoproteus vireonis* in *Vireo olivaceus*. The trypanosomatids found were similar to descriptions of *Trypanosoma everetti* (Molineux 1973) in Tyrannidae and *T. paddae* in *Manacus manacus.* Unidentified species of *Trypanosoma* were observed in *Ramphocelus carbo*, *Thraupis episcopus*, *Arremonops conirostris*, *Pipra erithrocephala* (Fig. 1a), *Pachyramphus polychopterus* and *Vireo olivaceus*. *Hepatozoon atticorae* were found in the swallow *Neochelidon tibialis* (Fig. 1b); unidentified species of this genus were seen in *Dendrocincla fuliginosa* and *Glyphorhynchus spirurus* and in *Manacus manacus. Plasmodium* infection included the subgenus *Novyella* in the species *Psarocolius decumanus* (possibly *P. columbae*)*, Saltator maximus, Arremonops conirostris* and *Arremon taciturnus* (possibly *P. nucleophilum*). The subgenus *Haemamoeba* occurred in *Mionectes oleaginea* and *Myiarchus ferox* (possibly *P. relictum*) and *Arremonops conirostris*. *Plasmodium* subgenus *Giovannolaia* shizonts were found in *Ramphocelus carbo*. *Leucocytozoon* (probably *fringillinarum*) was detected in *Dendroica discolor* (Fig. 1c). Rounded gametocytes were observed in a male *Pipra erythrocephala*, as well as an unidentified member of the genus *Haemoproteus* or *Plasmodium* was seen in *Arremon taciturnus*; however, in the absence of mature and schizogonic forms, even a generic diagnosis was not possible.

Most of the birds captured (309) were Passeriformes, followed by Apodiformes (15) and Columbiformes (11); all other orders were represented by only a few individuals. Frequency of infection varied markedly amongst the 15 parasitized families. *Hepatozoon* was found in 5 out of 8 (62 %) woodcreepers (Dendrocolaptidae) and all of 5 swallows (Hirundinidae). The Fringillidae (56%, primarily infected with *Plasmodium*), Turdidae (53% infected with microfilariae) and Columbidae (36% primarily infected with *Haemoproteus*) showed the highest prevalence of infection with haematozoa. Other families, such as Pipridae (12%) and Tyrannidae (11%), had the lowest prevalence. All other families were represented by small samples (Table I).

DISCUSSION

Our results concerning parasitaemia, when compared to other surveys carried out in Neotropical countries were similar to those obtained by Renjifo *et al* (1952) in eastern Colombia, Sousa & Herman (1982) in Panama, and Rodríguez & Matta (2001) also in eastern Colombia (27%, 18% and 16%, respectively); these were, however, higher than those found by White *et al.* (1978) in Neotropical area (10.5%), Bennett *et al.* (1991a) in Bolivia (5%) and Valkiünas et al (2003) in Colombia (8%). These values were lower than those recorded for the Nearctic area (Greiner *et al.* 1975).

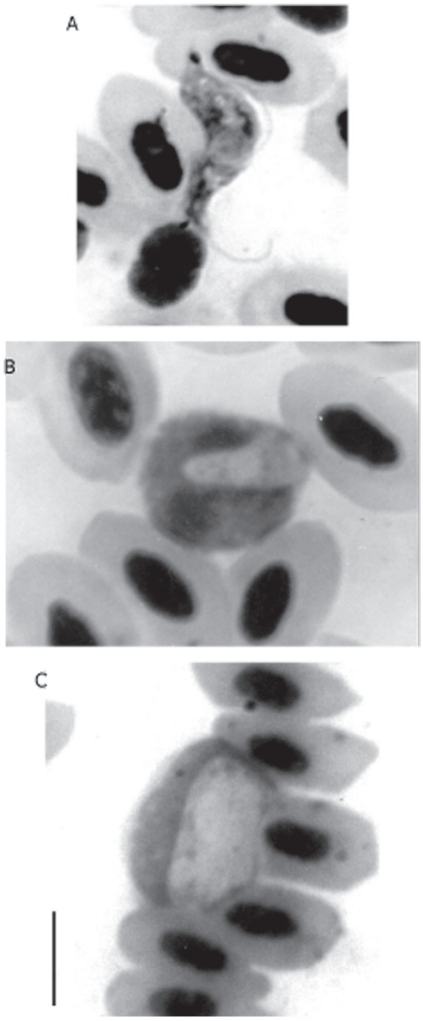

Figure 1. Microphotographs of blood parasites from Colombian avian hosts. A: *Trypanosoma* in *Pipra erithrocephala*; B: *Hepatozoon* in *Neochelidon tibialis*; C: *Leucocytozoon* in *Dendroica discolor*. Bar = 10 µm

The parasite index so found could also indicate higher vector potential in this area (1.13) when compared to other surveys in Neotropical regions such as Rodríguez & Matta (2001) in Colombia (1.08) and Bennett *et al.* (1991a) in Bolivia (1.03). This phenomenon was probably due to differences in the study areas' ecological conditions since it is difficult to compare the prevalence of parasites in a broad range of bird species sampled in different ecological systems at different times of the year. However, when specifically compared to studies carried out in Colombia (Renjifo *et al.*1952, Rodríguez & Matta 2001), in similar geographical areas, avian host species sampled and the same climatic season, our results suggest that the prevalence of blood parasites could be related to current conservation status and land use during the past fifty years. The differences in the prevalence of blood parasites compared to those obtained from birds from La Macarena NNP possibly reflects slower reduction of natural biotypes through intensive agriculture and cattle ranching which are more intense practised in areas closer to Villavicencio, situated on the edge of Colombia's eastern plains (Rodríguez & Matta 2001). A similar hypothesis has been proposed for European birds (Bennett *et al.*1982, Tella *et al.* 1999).

An interesting aspect of this survey was the high prevalence observed for certain blood parasites like *Hepatozoon*, *Trypanosoma* and microfilariae. The genus *Hepatozoon* was mainly found to be associated with families such as Hirundinidae and Dendrocolaptidae which usually occupy cavity or colony nests (Hilty & Brown 1986). Such nests frequently support large populations of mites, ticks, fleas and other relatively sedentary nest-occupying blood-feeding arthropods which could serve as potential vectors for this haematozoan genus (Kirkpatrick & Smith 1988, Bennett & Peirce 1989). The presence of microfilarial worms and trypanosomes was higher than that reported by Bennett *et al.* (1991a) in Bolivia. The microfilaries were not identified to the species level because only peripheral blood were collected, were the nematode adult is absent. Conversely, *Leucocytozoon* was restricted to a North-American migrant bird (*Dendroica discolor*) and was not found in resident birds. The apparent absence of leucocytozoids suggests a lack of suitable ornithophilic simuliid vectors, since North-

Table I. Blood parasites found in birds from La Macarena NNP (Colombia).

Host species	Total birds	Infected birds	Total birds infected						
			M	*P*	*T*	*Hp*	*H*	*L*	O
Accipritidae									
Buteo magnirostris	1	1					1		
Columbidae									
Claravis pretiosa	4	2					2		
*Leptotila rufaxilla**	4	1	1				1		
Zenaida auriculata	1	1					1		
Caprimulgidae									
Caprimulgus nigrescens	1	1					1		
Dendrocolaptidae									
*Dendrocincla fuliginosa**	7	4	2			3			
Glyphorhynchus spirurus	1	1				1			
Pipridae									
*Manacus manacus**	67	7	4		1	3			
Pipra erythrocephala	6	2		1	1				
Cotingidae									
*Pachyramphus polychopterus**	6	2			1		1		1
Tyrannidae									
Attila spadiceus	1	1	1						
Atalotriccus pilaris	5	1	1						
*Mionectes oleaginea**	42	4	1	1	3				
Myiarchus ferox	17	1		1					
Pitangus sulphuratus	2	1	1						
Tyrannus melancholicus	7	2	1		1				
Hirundinidae									
Neochelidon tibialis	5	5				5			
Corvidae									
Cyanocorax violaceus	1	1	1						
Turdidae									
Turdus ignobilis	25	13	13						
Turdus leucomelas	2	1	1						
Turdus nudigenis	3	2	2						
Vireonidae									
*Vireo olivaceus**	2	2			2		2		
Icteridae									
Psarocolius decumanus	4	2		2					
Parulidae									
Dendroica discolor	1	1						1	
Setophaga ruticilla	2	1	1						
Thraupidae									
*Ramphocelus carbo**	9	5	1	1	1		2		1
Tangara cayana	4	1	1						
*Thraupis episcopus**	2	2	1		1				1
Thraupis palmarum	9	1							1
Fringillidae									
*Arremon taciturnus**	3	3	1	1					2
*Arremonops conirostris**	9	7		7	1				
Saltator maximus	10	3	2	1					
Negative birds (see below)	263								
Total	342	82	36	15	12	12	11	1	6
% infected		24	10.5	4.4	3.5	3.5	3.2	0.3	1.7

M: microfilariae; *T*: *Trypanosoma*; *Hp*: *Hepatozoon*; *P*: *Plasmodium*; *H*: *Haemoproteus*; *L*: *Leucocytozoon*; O: others; *: species with double/triple infection.

Negative birds (number examined in parenthesis): Columbidae: *Columbina minuta* (1); *Columbina talpacoti* (1). Psittacidae: *Touit huetii* (4). Cuculidae: *Crotophaga ani* (2). Strigidae: *Athene cunicularia* (2). Trochilidae: *Agyrtria versicolor* (1); *Chlorestes notatus* (1); *Chlorostilbon mellisugus* (2); *Heliomaster longirostris* (4); *Phaethornis augusti* (4); *Phaethornis griseogularis* (1); unidentified hummingbirds (2). Momotidae: *Momotus momota* (2). Ramphastidae: *Pteroglossus pluricinctus* (1). Formicaridae: *Formicivora grisea* (2); *Thamnophilus amazonicus* (1). Pipridae: *Machaeropterus regulus* (3). Tyrannidae: *Elaenia parvirostris* (8); *Elaenia chiriquensis* (2); *Empidomonus varius* (2); *Leptopogon amaurocephalus* (5); *Megarhynchus pitangua* (2); *Myiozetetes cayanensis* (2); *Pitangus lictor* (1); *Tyrannus tyrannus* (1). Troglodytidae: *Troglodytes aedon* (1). Mimidae: *Mimus gilvus* (1). Coerebidae: *Cyanerpes caeruleus* (1); *Cyanerpes cyaneus* (1); *Dacnis cayana* (4). Thraupidae: *Euphonia laniirostris* (1); *Euphonia xanthogaster* (1); *Schistoclamys melanopis* (1); *Tachyphonus phoenicius* (8); *Tachyphonus surinamus* (2). Fringillidae: *Sporophila* sp. (1).

American migrants harbouring *Leucocytozoon* over-winter in the neotropics and could serve as a leucocytozoid reservoir for infecting resident birds (Herman 1968, Bennett *et al.* 1991b).

The presence of haemoproteids in some avian hosts indicates that suitable ceratopogonid and hippoboscid vectors are present in the area; however, the low number of infections recorded suggests that vector densities are low. The frequency of *Plasmodium* was lower than that found by Renjifo *et al.* (1952), but higher than that encountered in other Colombian surveys (Bennett & Borrero 1976, Rodriguez & Matta 2001). A possible explanation for the paucity of haematozoa in Colombian birds has been recently proposed by Valkiünas *et al.* (2003), suggesting that neotropical strains of avian hematozoa may be more virulent and kill more birds than other strains elsewhere, leading to low prevalence in mist-netted birds. While this is an interesting hypothesis, we have no data to support it.

The low intensity of parasitaemia obtained in the sampled birds has also been recorded previously (Bennett & Borrero 1976, Rodriguez & Matta 2001, Valkiünas *et al.* 2003), factors associated with the state of infection (acute or chronic) (Valkiünas *et al.* 2003), type of diagnostic test used (Kirkpatrick & Smith 1988), vector density and evolutionary relationship with their host (Bennett *et al.* 1980, Young *et al.*1993), could be affecting this result.

It is interesting to note that the origin of bird families could be related to the prevalence of haemoparasites. Bennett *et al.* (1980) suggested that Neartic avian families (i.e. Turdidae, Columbidae) have a higher prevalence of blood parasites than those from presumed Neotropical origins (i.e. Tyrannidae, Trochilidae) and our data are in accordance with this suggestion. Certain families have thus evolved with neoarctic and neotropical parasites, being thereby inherently more susceptible to infection by different blood parasites (Bennett *et al.* 1980). Behavioural aspects of various avian host families may also bring them into more frequent contact with suitable ornithophilic vectors (Kirkpatrick & Smith 1988). Trochillidae family members are rarely examined due to their difficult to catch, also the low prevalence detected in this birds, might be associated with their flight which could minimise host-vector contact (White *et al.* 1979)

The prevalence and intensity of haemoparasites in birds (as in other parasitic infections) is a result of a dynamic process involving the vector-host-environment. We have tried to demonstrate that the ecological conservation of an area is directly associated with the number of infections detected; however, the intensity of infection has continued to be low. Specific studies aimed at determining the pathogenicity of neotropical strains of avian hematozoa are therefore desirable, as would be studies on vectors of these parasites.

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