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Hematozoa in Two Populations of the Threatened Red-billed Chough in Spain

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ABSTRACT: The prevalence of hematozoa in two populations of red-billed choughs (*Pyrrhocorax pyrrhocorax*) was sampled in 1992 and 1994 in Spain. Two blood parasites infected red-billed choughs. A species of *Plasmodium*, possibly *Plasmodium relictum*, and the piroplasm *Babesia frugilegica*, are described for the first time from this host. Low prevalence (1/178, <1%) of hematozoa in these populations, was evidence for a lack of effects of blood parasites on the life history and conservation of this threatened species in at least the two populations studied.

Key words: Red-billed chough, *Pyrrhocorax pyrrhocorax*, hematozoa, *Plasmodium* sp., piroplasm, *Babesia frugilegica*, new host record.

As part of a study on the biology of red-billed choughs (*Pyrrhocorax pyrrhocorax*) in northwestern Spain, we conducted a survey of blood parasites. Red-billed choughs are rare birds with a fragmented distribution in the Palearctic region and a history of declining populations and extinction over much of Europe (Guillou, 1981). Several factors may affect numbers and distribution of red-billed choughs in Europe, including human disturbance, especially changes in traditional agro-pastoral systems, persecution, (Bignal and Curtis, 1989) and disease (Bullock et al., 1983; Bignal et al., 1987a; Meyer and Simpson, 1988). Small isolated or fragmented populations of red-billed choughs may be especially vulnerable to the effects of chronic infections by parasites or to epizooties of disease because of their relatively slow recruitment rates (Bignal et al., 1987b; G. Blanco, J.L. Tella and J. Fargallo, unpubl. data). Despite the possible conservation implications of parasitic infections (May, 1988; Loye and Carroll, 1995) the impact of parasites and other disease agents on red-billed chough populations are not documented except for the effects of gapeworms, *Syngamus trachea* (Bignal et al., 1987a; Meyer and Simpson, 1988). There are only two previous records of hematozoa in red-billed choughs: *Leucocytozoon sakharoffi* from Poland (Ramisz, 1961) and microfilariae from Russia (Shakhmatov et al., 1972). However, there are records of *Plasmodium* sp. in the alpine chough (*Pyrrhocorax graculus*) in Spain (Hamerton, 1936, 1937, 1944, cited in Bennett et al., 1982). Here we present data on prevalence of hematozoa in two populations of red-billed choughs at the southern part of their range in Spain.

During 1992, 1994, and 1995, data on red-billed choughs and their hematozoan parasites were collected in Los Monegros, Aragón, northwestern Spain (41°20′N, 0°11′W) and southeastern Madrid province, central Spain (40°19′N 3°32′W). The study areas are described by Blanco et al. (1991) and Tella et al. (1993). Red-billed choughs were captured at night with large butterfly nets while roosting inside abandoned buildings located in Los Monegros and in large caves and crevices in southeastern (SE) Madrid. Breeding pairs were captured while roosting at the nest site and non-breeding choughs were captured in communal roosts. We sampled 178 individuals. Samples were grouped into three seasons: breeding season (April through June; Los Monegros $n = 35$), autumn (August and September; Los Monegros $n = 54$, SE Madrid $n = 28$), and winter (November through January; Los Monegros $n = 7$, SE Madrid $n = 54$).

Upon capture, each red-billed chough was banded, measured, weighed, and released. Blood samples were taken from the brachial vein and thin blood smears were prepared, air-dried, fixed with absolute ethanol and stained with Giemsa stain. On mounted slides, half a smear chosen at random was entirely scanned at 200× along its longitudinal axis, looking for extraerythrocytic protozoa (Merino and Potti, 1995). Numbers of intraerythrocytic parasites were estimated under oil at 1000× by counting the number of parasites per 2000 erythrocytes (Godfrey et al., 1987).

We distinguished first year from second-year birds and older birds (≥3 yr old) on the basis of plumage characteristics (Blanco et al., 1996) and because most of them were banded in previous years. Sex was determined according to discriminant functions extracted from morphological variables (Tella and Torre, 1993; Blanco et al., 1996).

We sampled all age and sex classes in both Los Monegros (1-yr-old: six males, eight females; 2-yr-old: 23 males, 21 females; ≥3-yr old: 19 males, 19 females) and SE Madrid (1-yr-old: 12 females; 2-yr-old: six males, five females; ≥3-yr-old: 28 males, 31 females) except for 1-yr-old males in SE Madrid. Within the older age class (≥3-yr-old) there were at least 14 breeders (six males, eight females).

We did not find blood parasites in smears from the 96 red-billed choughs captured in Los Monegros. In SE Madrid we found only one (1%) of 82 birds infected with two blood parasites (*Plasmodium* sp., and *Babesia frugilegica*). The infected bird, a male >3-yr-old, was captured and banded in November 1992 at a large communal roost comprising about 250 choughs; it had an intensity of infection of 52 parasites/2,000 erythrocytes. The round gametocyte displaced the nucleus of the host cell towards a pole (Fig. 1), characteristic of *Plasmodium* spp. from the subgenus *Haemamoeba*. This species of *Plasmodium* may be part of the group

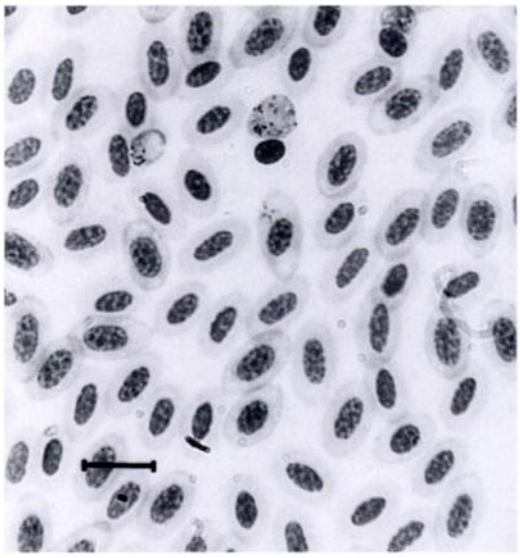

FIGURE 1. Chough erythrocytes infected by round gametocytes of *Plasmodium* sp. and forms of *Babesia frugilegica*. Giemsa. Scale bar = 14 μm.

cludes *P. relictum, P. giovannolai*, and *P. subpraecox,* which cannot be identified by morphological characters and host family (Greiner et al., 1975). Because *P. relictum* is cosmopolitan and reported from the family Corvidae (Greiner et al., 1975; Bennett et al., 1993) it may be the species involved. This is the first record of *Plasmodium* spp. infection in red-billed choughs. The piroplasm species was identified as *Babesia frugilegica,* a parasite founded in other two Corvid species before (M. A. Peirce, pers. comm.). The positive slide was deposited in the collection of the Museo Nacional de Ciencias Naturales, Madrid, Spain (access number: MNCN35.01/4).

The parasitized red-billed chough was never resighted or recaptured during a 3-yr period during which we eventually observed 15 (83%) of the 18 choughs of the same age and sex banded the same day as the infected individual. The weight of the infected red-billed chough (344 g) was typical (320 to 390 g) for ≥3-year-old male choughs weighed on the same date at the same site, although slightly below the mean ± SD (353.3 ± 15.2 g, n = 24). The

infected bird also had a slightly higher than average intensity of infestation by the mite (n = 10) *Neotrombicula autumnalis* (Acari, Trombiculidae; Suborder Actinedida) than choughs of the same age and sex captured at the same site and date (5.6 ± 6.6, n = 20; prevalence = 88%, n = 25; intensity of infection was not recorded on five individuals). The mite was deposited in the Museo Nacional de Ciencias Natural de Madrid; accession number 20.02/9023.

The low prevalence of hematozoa in the birds sampled, along with the infection appearing in a season with low probability of transmission due to the effects of climate (cold, scanty rainfall and thermic reversal characterizing inland Mediterranean latitudes) on suitable vectors (ornitophilic mosquitoes) are evidence that we detected an accidental infection. However, the survival of high numbers of mosquitoes (G. Blanco and J. Fargallo, pers. observation) and possibly the transmission of *Plasmodium* spp. by these mosquitoes (Seed and Manwell, 1977) may be allowed by the warm temperatures maintained during winter in large crevices where choughs congregate in large numbers to roost communally (Blanco et al., 1993). In fact, colonial birds attract ectoparasites and have frequent inter-individual contacts (Hubálek, 1994). The absence of suitable vectors in the abandoned farm-houses used by red-billed choughs in Los Monegros (Tella et al., 1995) may be a possible cause of the low prevalence of hematozoa in another threatened bird species, the lesser kestrel (*Falco naummani*), which also breeds in these houses. Ticks are vectors of *Babesia* spp. in birds (Peirce, 1973), and possibly that is the case in choughs. Although ticks may be present on the chough populations we studied, they are difficult to find in the intricate plumage of birds. However, unidentified ticks has been observed on crevices used by choughs (G. Blanco and J. Fargallo, pers. obs.).

The bird infected was never seen after capture and had a slightly higher infection by ectoparasites and a slightly lower mass than the other birds of the same sex and similar age. The symptoms of birds infected with *Plasmodium* sp. may include anemia, splenomegaly, and myocarditis (Seed and Manwell, 1977; Hubálek, 1994), and may cause death, at least in those species or birds not commonly infected with this parasite (Warner, 1968; Bennett et al., 1993). We do not know the extent of pathogenicity caused by *Babesia* spp. in birds, but the mixed infection may enhance pathogenicity in these hosts.

In conclusion, blood parasites do not appear to be a serious threat to the adults of two populations of red-billed choughs studied in Spain, although the presence of *Plasmodium* spp. may indicate a potential mortality factor in nestlings and fledgings. More work is required, especially studies of pathogens that might affect recruitment by causing nestling and juvenile mortality.

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