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An Investigation of Blood Smears of Northern Australian Finches

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EMU Vol. 92, 114-117, 1992. Received 21-10-1990, accepted 8-4-1991

Haematology can be used to diagnose avian (Hawkey *et al.* 1985; Campbell & Dein 1984; Campbell 1988) and mammalian diseases (Bubenik 1987). It can be used to indicate the presence of intra- or extra-cellular parasites, a high white blood cell count or a low count of mature red blood cells (Campbell & Dein 1984).

The aim of this study was to investigate the possibility that Gouldian Finches *Erythrura gouldiae* were diseased, as far as it was possible to determine from blood smears, by comparing blood smears from them with those of co-occurring finches. The reason for focusing on the Gouldian Finch was that their numbers have declined in the wild (Blakers *et al.* 1984) to the point of their being endangered (Brouwer & Garnett 1990). If the measures of red and white cells of Gouldian Finches lay outside the ranges of the other finches or intra- or extra-cellular parasites were present in Gouldian Finches but not the others, it could suggest that Gouldian Finches might be affected by disease. More refined methods could then be used to re-examine blood composition and investigate the nature of the disease.

Methods

Blood was taken from eight species of finch, Long-tailed *Poephila acuticauda* (n = 81), Masked *P. person-*

ata (n = 30), Pictorella Mannikin *Lonchura pectoralis* (n = 19), Chestnut-breasted Mannikin *L. castaneothorax* (n = 19), Zebra Finch *Taeniopygia guttata* (n = 141), Star Finch *Neochmia ruficauda* (n = 77), Double-barred Finch *P. bichenovii* (n = 41) and Gouldian Finch (n = 77) from 11 locations across the Northern Territory: Newry Station (15°55′S, 129°14′E; 15°59′S, 129°09′E; 15°57′S, 129°05′E), Keep River National Park (15°58′S, 129°02′E), Auvergne Station (15°35′S, 129°20′E; 15°45′S, 129°52′E), Rosewood Station (16°28′S 129°07′E), Willeroo Station (15°17′S, 131°35′E), Ferguson River (14°05′S, 131°58′E) and Yinberrie Hills (14°08′S, 132°05′E; 14°08′S, 132°06′E).

Birds were caught in mist nets and blood samples taken between 0800 and 1000 h (to minimise diurnal variation) from mid-August to mid-October, 1986. A drop of blood was collected from the superficial ulnar vein in a heparinised microcapillary tube. The blood was transferred to a clean glass slide, smeared immediately, air-dried and fixed in absolute ethanol for three minutes. Blood smears were stained using Wright's-Giemsa.

Thirty monolayer fields (where cells were adjacent but not overlapping or deformed) per slide were examined (on 1000X magnification) for intraerythrocytic parasites such as *Haemoproteus* or *Plasmodium*. The smears were also searched for extracellular parasites, such as microfilariae and trypanosomes, particularly at the thicker end of the blood smear.

The number of white blood cells (lymphocytes, heterophils, monocytes, basophils and eosinophils) and thrombocytes were counted in 30 monolayer fields where cells were touching but not overlapping. Eosinophils were distinguished from heterophils by the presence of rounded eosinophilic cytoplasmic granules present in the former but not the latter. Thrombocytes were distinguished from small lymphocytes on the basis of nucleus shape, density of chromatin clumping and amount of cytoplasm relative to the size of the nucleus: thrombocytes have oval rather than rounded nuclei, more homogeneously stained nuclei, and a higher ratio of cytoplasm:nucleus than small lymphocytes. The total of each type of white cell, the sum of the white cells and the number of thrombocytes, over the 30 fields, were divided by five to provide the average number of each type of leucocyte per 1000 erythrocytes. (This method [described by Campbell & Dein 1984; Campbell 1988] was checked by counting the erythrocytes in ten samples of 30 fields [of touching but not overlapping cells] to ensure that their basis of number of red cells in 30 fields/5 = 1000 was valid.)

Erythrocytes were divided into three categories: (1) mature — an oval-shaped cell with an oval, centrally placed nucleus that is more condensed than (2) (below); (2) polychromatic erythrocyte — a round or oval-shaped cell with a larger nucleus than the mature cell and with irregularly clumped chromatin; and (3) erythroblast — a large round cell that has a large amount of cytoplasm in relation to its nucleus. An estimate was made of the percentage of each of the categories of red blood cells in each field and averaged for the 30 fields. (Counts of each of the categories were made over 10 fields to check that the estimates were within 6% of the counts.)

Results

No blood parasites, either intra- or extra-cellular, were found in any of the blood smears.

Lymphocytes were the most common of the white cells in all finches, then heterophils, monocytes and eosinophils (Table 1). No basophils were distinguished. Thrombocytes, the functional equivalent of the mammalian platelet, were in low numbers in all finches. Thrombocytes were sometimes clumped in fields where cells were more than one monolayer thick and so would not have been included in the counts. Cell counts of Gouldian Finches lay within the ranges of the other species except in the case of heterophils which were highest in the Gouldian but did not differ significantly from the others (Analysis of Variance: $F = 1.68$, $d.f. = 7, 475$; $0.2 < P < 0.5$) (Table 1).

Mature erythrocytes were more numerous than both the other stages and ranged from 62% in Zebra Finches to 76% in Longtailed. Mid-stage erythrocytes varied between 20 and 33% while immature eryhrocytes did not exceed 6%.

Discussion

Blood parasites were absent and so may not have affected the Gouldian Finch (see also Steadman *et al.* 1990). Blood smears are appropriate to determine the presence of parasites although they underestimate the prevalence of malaria (Herman *et al.* 1966) and microfilariae (Seegar 1979). Parasites exhibiting nocturnal periodicity, however, would not have been detected by the method used in this study.

Gouldian Finches did not appear to be diseased, as far as could be indicated by a blood smear, because counts of white blood cells did not differ from the range

Table 1 Counts from blood smears of white blood cells, thrombocytes and estimates (%) of red blood cells (with standard deviations in parentheses) in eight species of finch found in the northwest of the Northern Territory.

	Double-barred	Pictorella	Long-tailed	Masked	Star	Zebra	Chestnut-breasted	Gouldian
Lymphocyte	6.42 (3.07)	3.9 (2.8)	6.13 (3.7)	5.9 (3.15)	5.72 (2.61)	4.13 (2.4)	4.96 (2.27)	5.97 (2.59)
Heterophi	0.8 (0.74)	0.38 (0.49)	0.72 (0.63)	0.62 (0.53)	0.55 (0.79)	0.78 (0.95)	0.35 (0.39)	0.89 (0.81)
Monocyte	0.2 (0.23)	0.07 (0.11)	0.22 (0.25)	0.19 (0.29)	0.18 (0.17)	0.29 (0.28)	0.1 (0.14)	0.25 (0.22)
Eosinophil	0.07 (0.01)	0.01 (0.05)	0.05 (0.11)	0.03 (0.08)	0.03 (0.07)	0.12 (0.3)	0.01 (0.05)	0.06 (0.1)
Basophil	—	—	—	—	—	—	—	—
Sum white cells	7.42 (4.21)	4.35 (2.87)	7.12 (4.14)	6.69 (3.29)	6.45 (2.81)	5.29 (2.58)	5.43 (2.31)	7.04 (3.02)
Thrombocyte	0.32 (0.32)	0.04 (0.12)	0.18 (0.29)	0.27 (0.44)	0.18 (0.24)	0.15 (0.22)	0.17 (0.22)	0.23 (0.28)
Erythrocyte 1	70 (16)	63 (12)	76 (9)	69 (11)	67 (11)	62 (12)	73 (12)	72 (11)
Erythrocyte 2	26 (15)	33 (12)	20 (8)	26 (10)	29 (11)	33 (11)	22 (10)	23 (9)
Erythrocyte 3	6 (4)	4 (2)	4 (3)	4 (3)	4 (3)	5 (3)	5 (3)	5 (4)
n	41	19	81	30	77	141	19	77

encompassed by the other species. If the white cell counts indicated any form of infection, then all species were infected equally. The range of the sum of the white blood cells (per 1000 red cells) of the eight species in this study was smaller than that of 'normal' white blood cell counts of five species in a study by Stoskopf *et al.* (1983). The numbers of white cells in this study was possibly underestimated because no basophils were observed. Numbers of thrombocytes were probably also under-estimated because of their clumping in the thicker fields of view not included in the counts.

The numbers of white cells present can vary diurnally but the method we used minimised this source of variation. All birds were held in the same way for the same period before blood was taken; blood samples were taken early in the morning and from a large number of birds. A haemocytometer would have provided a more accurate measure of the numbers of white cells than a blood smear but the latter method was appropriate for use in the field and provided a relative measure of the blood cells present.

The proportions of erythrocytes of different ages were similar for all finches. Although up to a third of the erythrocytes were at intermediate stage, this is not considered pathologic in birds. Neither were there any spherical erythrocytes with oval nuclei, as reported from anaemic birds (Campbell & Dein 1984).

We conclude that the Gouldian Finch did not differ from sympatric species of finch: no blood parasites were observed and haematological counts were similar in all species. Disease as evidenced from blood smears does not appear to be responsible for the currently low numbers of Gouldian Finches in the wild. Even if diseases which could be detected from blood smears were factors in the decline of Gouldian Finches in the past, there may be no indication of their presence now.

Acknowledgements

Thanks to J. Purdie and L. Melville for histological assistance, N. Burley and S. Kerin for field assistance, R. Reece for references in the subject area and W. Freeland, K.

Richardson and D. Spratt for constructive comments on the manuscript.

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Habitat Use by Eastern Bristlebirds in Barren Grounds Nature Reserve

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EMU Vol. 92, 117-121, 1992. Received 1-10-1990, accepted 30-4-1991

Considerable cause for concern exists with regard to the long-term survival of the Eastern Bristlebird *Dasyornis brachypterus*. Though apparently widespread during the Tertiary Period (Smith 1977), this species is now restricted to a few small, isolated populations (Smith 1977; Blakers *et al.* 1984). The species is listed as endangered by Burbidge & Jenkins (1987) and CONCOM (1988), as threatened on Schedule 12 of the N.S.W. National Parks & Wildlife Act, 1974, and as vulnerable by Kennedy & Burton (1986). It appears to have declined in abundance in some areas during recent times (Blakers *et al.* 1984; Holmes 1989).

Little is known about the biology of the Eastern Bristlebird. It is apparently a species which is usually found near an ecotone, either between woodland and heath in the southern part of its range (e.g. Jordan 1987) or between open forest/woodland and rainforest in the northern part of its range (Holmes 1989). It is believed to feed predominantly on insects obtained from the leaf litter (Holmes 1989) but it may also eat seeds as its congeners *D. longirostris* and *D. broadbenti* do (Smith 1987, pers. comm.). Most nests have been found in grass tussocks, 10-45 cm above ground (McNamara 1946; Holmes 1989) and the most common clutch size is two eggs (Holmes 1989). It is extremely shy and difficult to observe (McNamara 1946; Robertson 1946) so that most of the information available for this species is anecdotal; no comprehensive behavioural or ecological study has been carried out.

Habitat use by Eastern Bristlebirds remains largely unknown. The ecotones associated with the species have been defined (see Jordan 1987; Holmes 1989) but the extent to which the species utilises areas at different distances from an ecotone has not been quantified and it