

Brief report

Absence of haematozoa on colonial White Storks *Ciconia ciconia* throughout their distribution range in Spain

Roger Jovani, José L. Tella, Guillermo Blanco & Marcelo Bertellotti

*Jovani, R., Tella, J. L. & Bertellotti, M., Department of Applied Biology, Estación Biológica de Doñana, C.S.L.C., Avda. M Luisa s/n, E-41013 Sevilla, Spain.
Blanco, G., Department of Biology, University of Saskatchewan, Saskatoon, S7N0W0, Canada.*

The White Stork (*Ciconia ciconia*) is distributed primarily in Europe, with about 8000 pairs breeding in Spain out of the 120 000–150 000 European breeding pairs (Tucker & Heath 1994). A large population decrease occurred during the twentieth century, particularly in western Europe, and therefore the White Stork has an unfavourable conservation status in Europe (Tucker & Heath 1994). Therefore, the White Stork is a species for which information about parasites and diseases should be taken into account in managing programs.

Blood parasite surveys are being incorporated in the monitoring of vertebrate species (e.g., Michot *et al.* 1995, Shutler *et al.* 1996). Blood parasites could affect life history traits of their vertebrate hosts (reviewed in Møller 1997), and hence its study is important for a correct and complete understanding of the factors shaping their population trends. On a review paper of the Haemoproteidae of the Ciconiidae, Forrester *et al.* (1977) only reported negative records of a captive White Stork in France. Later, Bennett *et al.* (1982) reported three species of haematozoa for White Storks, including one species of *Haemoproteus*, one *Leucocytozoon* and one microfilaria. However, these authors did not offer data on

prevalence in their review. The only review offering prevalence data on this species showed that none of the six White Storks sampled in Western Europe were infected by haematozoa (Peirce 1981). The aim of our study was to determine the prevalence and intensity of blood parasites in Spanish White Storks using a large sample of individuals. To our knowledge, ours is the first study searching for blood parasites on nestling White Storks, and the first specifically focused on this species. Given that infection with blood parasites may show geographical variations in a single host species (Bennett *et al.* 1995, Merilä *et al.* 1995, Sol *et al.* 2000), we sampled several colonies throughout the distribution range of White Storks in Spain.

Field work was conducted during three breeding seasons (1998–2000) in seven different areas (Fig. 1) which hold the highest breeding densities of White Storks in Spain (SEO/BirdLife 1994). Nestlings were bled from the brachial vein, and a thin smear was made using a drop of blood. Blood smears were air dried, fixed with ethanol in the field, and stained in the laboratory with Giemsa stain. A total of 130 blood smears were sampled from nestlings, covering an adequate sampling of the different habitats and geographical areas oc



Fig. 1. Location, main habitats surrounding the colonies, and number of White Storks sampled in Spain. a — Cinca valley (41°80'N, 0°20'E, freshwater rivers and irrigated cultures, n = 9); b — Ebro valley (41°70'N, 1°50'W, freshwater rivers and irrigated cultures, n = 12); c — Soto del Real (40°55'N, 3°90'W, humid pastures in low mountains, n = 15); d — Huelva (37°60'N, 7°20'W, oak forests and crops, n = 3); e — Donana area (37°00'N, 6°30'W, marshes and Mediterranean scrubland, n = 59); f — Guadaiquivir valley (37°30'N, 5°40'W, cereal crops, n = 12); g — Jerez (36°60'N, 6°15'E, urban area, n = 5); h — Cáceres (39°30'N, 6°50'W, oak forests, n = 15). Location of the two wildlife rehabilitation centres coincides with the areas 3 (n = 8) and 5 (n = 14). The geographical distribution of White Storks in Spain is indicated by a point-marked area.

occupied by White Storks in Spain (Fig.1). Nestlings were sampled on average at 55 days old (range: 45–68 days), thus a few days prior to fledging. In addition, 22 adult White Storks were sampled during *the* breeding season when they arrived to two wildlife rehabilitation centres (Fig.1) in a poor state of health.

Each blood smear was inspected for blood parasites for 10 mm under oil immersion (1000x). The average of five smears indicated that about 28000 erythrocytes were inspected for each blood smear. No parasites were detected in the 152 birds sampled.

The inspection of blood smears is not the best method to detect *Plasmodium* (Forrester *et al.* 1974), *Trypanosoma* or microfilarias (Apanius 1991). However, Merino and Potti (1995) found a high prevalence of *Trypanosoma* in Spanish Pied Flycatchers (*Ficedula hypoleuca*) nestlings by

analysing blood smears. Therefore, our results could also be indicative of the absence or very low prevalence of *Trypanosoma* in the sampled birds.

As far as we know no information is available on the prepatent period for any of the blood parasites found neither in White Storks nor for any other species of the family Ciconiidae (Bennett *et al.* 1982). However, the age of nestling White Storks at sampling fairly exceeds the minimum prepatent period reported for some blood parasites in nestlings of other European avian species [13 days for Sparrowhawks (*Accipiter nisus*) Ashford *et al.* (1991); 9 days for Pied Flycatchers Merino & Potti (1995); 14 days for Goshawks (*Accipiter gentilis*) Toyne & Ashford (1997)]. Moreover, the number of erythrocytes inspected per blood smear was sufficient to detect infections of high intensity, which are typical in nestlings of several species because of their naive immune system to parasites (Memo & Potti 1995, Dawson & Bortolotti 1999). Therefore, there is no reason to think that nestlings had blood parasites not yet expressed in circulating blood, or undetected by us. The 22 adults sampled in the rehabilitation centres reinforce the results obtained from nestlings, since damaged or diseased birds are presumed to be immunodepressed and thus would favour the relapse of latent blood parasite infections (Blanco *et al.* 1998). Moreover, haematocrit infections usually peak during the breeding season (Allander & Sundberg 1997), when we sampled all the birds. Therefore, the lack of blood parasites reported here could not be explained because of an inappropriate sampling or methodology.

Studies failing to detect blood parasites are as important as those reporting high haematocrit prevalences, because there is the need of comparing infected and non infected bird species or populations to address which host, parasite or habitat characteristics are the important cues shaping avian blood parasite distributions. Studies conducted in Spain have shown a great variability among bird species in their occurrence of blood parasites. Several studies have failed to find blood parasites, or have found very low prevalences (Tella *et al.* 1995, Figuerola *et al.* 1996, Blanco *et al.* 1997, Forero *et al.* 1997, Gonzalez-Soills & Abella 1997, Blanco *et al.* 1998, Merino & Minguéz 1998, Tella *et al.* 1999, Merino *et al.* 2000). On the contrary,

other studies reported high prevalences (Merino & Potti 1995, Ruiz *et al.* 1995, Bosch *et al.* 1997, Figuerola *et al.* 1999, Sol *et al.* 2000). In the case of White Storks, we can not discern whether the absence of blood parasites is related to the lack of appropriate parasite species, the scarcity of vectors, or both. Spanish raptors inhabiting open habitats, as is the case for the White Stork, have been found to present lower prevalences than those in forested areas (Tella *et al.* 1999). In Feral Pigeons (*Columba livia*), differences in blood parasite prevalence among localities have been experimentally proved to be shaped by variability in vector abundance (Sot *et al.* 2000). Therefore, habitat could play a role in the geographical distribution of blood parasites mediated by vector abundance (e.g. Figuerola 1999). Duration of the embryonic development period could also play a role in some avian species by increasing their ability to fight against haematzoa infections (Ricklefs 1992, Tella *et al.* 1999). However, this possibility has not been investigated in many avian groups and would need further study on the White Stork. In addition, future comparative studies including other places and other Stork species could add insight on the reasons for the absence of blood parasites we have reported.

Acknowledgements: We are grateful to M. C. Quintero, M. Vázquez, J. Marchamalo, F. Martínez, J. A. Pinzolas, the staff of Jeréz Zoo, and the Equipo de Seguimiento de Procesos Naturales from the Estación Biológica de Doñana for their help during the field work, and to J. C. Senar (Museu de Zoologia, Barcelona) for allowing us the use of a laboratory. We thank J. Colas and M. Corroto from GREFA rehabilitation centre for kindly providing us with several blood smears. J.L.T. and G.B. were supported by postdoctoral grants of the M.E.C. Jukka Jokimäki, Juha Merilä, and an anonymous referee provided helpful comments on the manuscript.

Selostus: Espanjan kattohaikaroilla ci tavattu veriloisia pesimäaikana

Kattohaikarat ovat vähentyneet viime aikoina erityisesti Länsi-Euroopassa. Koska veriloiset voivat vaikuttaa lintujen kasvuun ja kehitykseen, on uhanalaisille lintulajeille suojeleohjelmia tehtäessä selvitettävä myös veriloisten esiintymistä lintulajeissa. Kirjoittajat analysoivat veriloisten

esiintymistä ja infektioiden intensiteettiä espanjalaisissa kattohaikaroissa. Kattohaikarat pesivät seitsemässä koloniassa, joista näytteitä kerättiin 130 poikasesta. Lisäksi näytteet otettiin 22 aikuisesta linnusta, jotka ohjautuivat heikkokuntoisina kuntoutuslaitokseen. Yhdestäkään näytteestä ei löytynyt veriloisia. Espanjassa useilla lintulajeilla ei ole havaittu lainkaan veriloisia tai niiden esiintyminen on ollut vähäistä. Toisaalta, joiltakin lajeilta on raportoitu korkeita esiintymisfrekvenssejä. Kirjoittajat arvioivat, että lintujen elinympäristö voi vaikuttaa veriloisten esiintymiseen esimerkiksi väli-isäntien esiintymisen kautta. Myös erot eri lintulajien alkionkehityksen pituudessa voivat vaikuttaa lajien välisiin eroihin veriloisten esiintymisessä. Kirjoittajat eivät olleet varmoja siitä, johtuiko veriloisten puuttuminen espanjalaisista kattohaikaroista sopivien loislajien puutteesta vai loisten väli-isäntien vahyydesta vaiko molemmista edellä mainituista tekijöistä. Kirjoittajien mukaan on yhtä tärkeää raportoida loisten puuttumisesta kuin niiden esiintymisestäkin, jotta veriloisten yleisyydestä ei saada vääristynyttä kuvaa.

References

- Allander, K. & Sundberg, J. 1997: Temporal variation and reliability of blood parasite levels in captive Yellowhammer males *Emberiza citrinella*. — *Journal of Avian Biology* 28: 325–330.
- Apanius, V. 1991: Avian trypanosomes as models of Hemoflagellate evolution. — *Parasitology Today* 7: 87–90.
- Ashford, R. W., Green, E. E., Holmes, P. R. & Lucas, A. J. 1991: Leucocytozoon toddi in British sparrowhawks *Accipiter nisus*: patterns of infection in nestlings. — *Journal of Natural History* 25: 269–277.
- Bennett, G. F., Whiteway, M. & Woodworth-Lynas, C. 1982: Host-parasite catalogue of avian Haematzoa. Memorial University of Newfoundland. — *Occasional Papers in Biology*. Number 5. Newfoundland. Canada.
- Bennett, G. F., Squires-Parsons, D., Siikamäki, P., Huhta, E., Allander, K. & Hillström, L. 1995: A comparison of the blood parasites of three Fennoscandian populations of the Pied Flycatcher *Ficedula hypoleuca*. — *Journal of Avian Biology* 26: 33–38.
- Blanco, G., Merino, S., Tella, J. L., Fargallo, J. A. & Gajón, A. 1997: Hematozoa in two populations of the threatened Red-billed Cough in Spain. — *Journal of Wildlife Diseases* 33: 642–645.
- Blanco, G., Gajón, A., Doval, G. & Martínez, F. 1998: Absence of blood parasites in Griffon vultures from

- Spain. — *Journal of Wildlife Diseases* 34: 640—643.
- Bosch, M., Figuerola, J., Cantos, F. J. & Velarde, R. 1997: Intracolony differences in the infestation by *Haemoproteus lan* on Yellow-legged Gulls *Larus cachinnans*. — *Omis Fennica* 74: 105—112.
- Dawson, R. & Bortolotti, G. R. 1999: Prevalence and intensity of hematozoan infections in a population of American kestrels. — *Canadian Journal of Zoology* 77: 162—170.
- Figuerola, J., Velarde, R., Bertolero, A. & Cerdà, F. 1996: Absence of haematozoa in a breeding population of Kentish Plover *Charadrius alexandrinus* in North East Spain. — *Journal für Ornithologie* 137: 523—524.
- Figuerola, J. 1999: Effects of salinity on rates of infestation of waterbirds by haematozoa. *Ecography* 22: 681—685.
- Figuerola, J., Muñoz, E., Gutiérrez, R. & Ferrer, D. 1999: Blood parasites, leucocytes and plumage brightness in the Cirl Bunting, *Emberiza cirius*. — *Functional Ecology* 13: 594—601.
- Forero, M. G., Tella, J. L. & Gajón, A. 1997: Absence of blood parasites in the Red-necked nightjar. — *Journal of Field Ornithology* 68: 575—579.
- Forrester, D. J., Hon, L. T., Williams Jr., L. E. & Austin, D. H. 1974: Blood protozoa of wild turkeys in Florida. *The Journal of Protozoology* 21: 494—497.
- Forrester, D. J., Greiner, E.C., Bennett, G.F. & Kigaye, M.K. 1977. Avian Haemoproteidae. 7. A review of the haemoproteids of the family Ciconiidae (storks) and descriptions of *Haemoproteus brodkorbi* sp.nov. and *H. peircei* sp.nov. *Canadian Journal of Zoology* 55: 1268—1274.
- Gonzalez-Soils, J., & Abella, J.C. 1997. Negative record of haematozoan parasites on Cory's Shearwater *Calonectric diomedea*. — *Omis Fennica* 74: 153—155.
- Merilä, J., Björklund, M. & Bennett, G. F. 1995: Geographic and individual variation in haematozoan infections in the greenfinch, *Carduelis chlois*. — *Canadian Journal of Zoology* 73: 1798—1804.
- Merino, S. & Minguéz, E. 1998: Absence of haematozoa in a breeding colony of the Storm Petrel *Hydrobates pelagicus*. — *Ibis* 140: 180—181.
- Merino, S. & Potti, J. 1995: High prevalence of haematozoa in nestlings of a passerine species, the Pied flycatcher (*Ficedula hypoleuca*). — *Auk* 112: 1041—1043.
- Menino, S., Seoane, J., De la Puente, J. & Bermejo, A. 2000: Low prevalence of infection by haemoparasites in Cetti's Warblers *Cettia cetti* from Central Spain. — *Ardeola* 47: 269—271.
- Michot, T. C., Garvin, M. C. & Weidner, P. H. 1995: Survey for blood parasites in Redheads (*Aythya americana*) wintering at the Chandeleur islands, Louisiana. — *Journal of Wildlife Diseases* 31: 90—92.
- Møller, A. P. 1997: Parasitism and the evolution of host life history. — **In:** Clayton, D. H. & Moore, J. (eds.), *Host-parasite evolution. General principles and avian models*: 105—127. Oxford University Press, Oxford, U.K.
- Peirce, M. A. 1981: Distribution and host-parasite checklist of the haematozoa of birds in Western Europe. — *Journal of Natural History* 15: 419—458.
- Ricklefs, R. E. 1992: Embryonic development period and the prevalence of avian blood parasites. — *Proceedings National Academy of Science USA* 89: 4722—4725.
- Ruiz, X., Oro, D. & Gonzalez-Soils, J. 1995: Incidence of a *Haemoproteus lan* parasitemia in a threatened Gull: *Larus audouinii*. — *Omis Fennica* 72: 159—164.
- SEO/BirdLife. 1994: V Censo nacional de Ciguena Blanca. — SEO/BirdLife, Madrid, Spain.
- Shutler, D., Ankney, C. D. & Dennis, D. G. 1996: Could the blood parasite *Leucocytozoon* deter mallard range expansion? — *Journal of Wildlife Management* 60: 569—580.
- Sol, D., Jovani, R. & Tomes, J. 2000: Geographical variation in blood parasites in feral pigeons: the role of vectors. — *Ecography* 23: 307—314.
- Tella, J. L., Gortazar, C., Gajón, A. & Osácar, J. J. 1995: Apparent lack of effects of a high louse-fly infestation (Diptera, Hippoboscidae) on adult colonial Alpine swifts. — *Ardea* 83: 435—439.
- Tella, J. L., Blanco, G., Forero, M. G., Gajón, A., Donazar, J. A. & Hiraldo, F. 1999: Habitat, world geographic range, and embryonic development of hosts explain the prevalence of avian haematozoa at small spatial and phylogenetic scales. — *Proceedings National Academy of Science USA* 96: 1785—1789.
- Toyne, E. P. & Ashford, R. W. 1997: Blood parasites of nestling Goshawks. — *Journal of Raptor Research* 31: 81—83.
- Tucker, G. M. & Heath, M. F. 1994: *Birds in Europe: their conservation status*. — Cambridge, U.K.: BirdLife International (BirdLife Conservation Series no. 3). The Burlington Press, Ltd, Cambridge.