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Incidence of Salmonellae in Captive and Wild Free-Living Raptorial Birds in Central Spain

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With 3 tables

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Summary

A total of 595 faecal samples from raptorial birds, either captive or free-living, residing in GREFA Wildlife Hospital were bacteriologically examined using various selective media and an Automated Diagnostic Assay System for *Salmonella* detection. Serotype and phage type of the strains identified as *Salmonella* was determined. In the captive group, of the 285 samples examined, 21 (7.36%) were positive for *Salmonella*. Serotyping revealed that most of the individuals were infected by *Salmonella* serotype Havana. This result suggested that there could be a source of contamination in the Hospital although it could not be established. In the wild free-living group, over 310 samples examined (4.19%) were positive for *Salmonella*. The *Salmonella* isolates showed a major variety of serotypes: Enteritidis, Adelaide, Brandenburg, Newport, Typhimurium, Hadar, Saintpaul and Virchow. Most of them are similar to those commonly described in isolates from human and domestic animals. These results indicate that wild birds could be involved in the dissemination of *Salmonella* in humans or domestic animals or vice versa.

Introduction

Many studies about the incidence and the resistance profiles of *Salmonella* in domestic animals have been performed, but very little attention has been paid to the study of wild birds from various *Salmonella*-contaminated environments. In fact, the spread of *Salmonella* and its persistence in the environment may be enhanced by wild birds which, in view of their considerable motility, have been identified as one possible source of *Salmonella* infections in humans and farm animals (Coulson et al., 1983). Although, there is lack of documentary evidence on the incidence and distribution of salmonellae in raptorial birds, either captive or free-ranging.

This article describes the isolation and characterization of salmonellae from wild free-living and captive raptors in Spain by serotyping and phage typing during a 3-year period.

Materials and Methods

Sample collection

A total of 595 raptors of five different families, 285 captive and 310 free-ranging raptors, were sampled for detection of *Salmonella* in faecal microbiota (Table 1). The birds had been kept in captivity at least during the 3-year period of this study. Cloacal swabs were taken using small sterile wads of cotton with Amies transport medium.

Isolation procedures and identification

The faecal swabs were incubated in Selenyte Cystine broth (Oxoid, Madrid, Spain) and incubated at 42°C for 24 h. Vitek Inmuno Diagnostic Assay System (VIDAS) (BioMérieux, Marcy l'Etoile, France) assay was used for the detection of *Salmonella*. Positive cultures from this enrichment broth were then plated onto selective agar plates: *Salmonella Shigella* Agar (BioMérieux, Marcy l'Etoile, France) and Harlequim *Salmonella* Agar (International Diagnostic Group, Lancashire, UK) and cultivated at 37°C for 24 h. Presumed *Salmonella* colonies were identified by biochemical characterization using ID 32 GN-strips (BioMérieux, Marcy l' Etoile, France).

Serotyping

All the *Salmonella* strains were sent to the Centro Nacional de Referencia de Salmonella, Centro Nacional de Microbiología, Madrid (Spain) for serotyping and phage typing.

Results

The results of the detection of *Salmonella* in raptorial birds are shown in Table 1. From a total of 285 captive raptors, 21 (7.36%) presented *Salmonella* in the faecal microbiota. This percentage of *Salmonella* isolation correlates with the duration of time in captivity, 3 years. On the other hand, from a total of 310 free-ranging raptors, 13 (4.19%) presented *Salmonella*.

Serotyping and phage typing yielded the results given in Tables 2 and 3. From the group of captive raptors, the most frequent serotype isolated was Havana. Although, sixteen *Salmonella* serotype Havana were isolated from different species in the captive raptorial birds group. Two strains of *Salmonella* serotype Branderburg (S10 and S26) were isolated from a kestrel and a griffon vulture, respectively. One single strain of *Salmonella* serotype Newport (S28) was isolated from a little owl. *Salmonella* serotype Adelaide (S36), *Salmonella* serotype Enteritidis PT6a (S52) and *Salmonella* serotype Hadar PT1 (S57) were also isolated from a lesser kestrel, a scops owl and a kestrel, respectively.

Table 1. Incidence of *Salmonella* in the faecal samples of raptorial birds

Family	*Salmonella*	Captive		Free-ranging	
		+	–	+	–
Strigidae	Short-eared owl (*Asio flammeus*)	–	2	–	8
	Long-eared owl (*Asio otus*)	–	1	1	6
	Eagle owl (*Bubo bubo*)	–	6	–	16
	Tawny owl (*Strix aluco*)	–	2	–	10
	Scops owl (*Otus scops*)	1		–	8
	Little owl (*Athene noctua*)	5	10	3	26
Falconidae	Peregrine (*Falco peregrinus*)	–	4	–	8
	Hobby (*Falco subbteo*)	–	–	–	3
	Merlin (*Falco columbarius*)	–	–	–	2
	Kestrel (*Falco tinnunculus*)	3	38	–	44
	Lesser kestrel (*Falco naumanni*)	7	132	3	56
Accipitridae	Sparrow hawk (*Accipiter nisus*)	–	2	1	14
	Goshawk (*Accipiter gentilis*)	2	5	–	8
	Hen harrier (*Circus cyaneus*)	–	–	–	2
	Montagu's harrier (*Circus pygargus*)	1	2	–	6
	Marsh harrier (*Circus aeruginosus*)	–	–	–	1
	Black kite (*Milvus migrans*)	–	5	–	3
	Red kite (*Milvus milvus*)	–	–	–	3
	Honey buzzard (*Pernis apivorus*)	–	–	–	1
	Buzzard (*Buteo buteo*)	1	14	1	16
	Booted eagle (*Hieraaetus pennatus*)	–	2	–	6
	Short-toed eagle (*Circaetus gallicus*)	–	–	–	3
	Bonelli's eagle (*Hieraaetus fasciatus*)	–	6	–	7
	Imperial eagle (*Aquila heliaca*)	–	–	1	–
	Golden eagle (*Aquila chrysaetos*)	–	6	–	2
	Black vulture (*Aegytius monachus*)	1	2	–	8
	Griffon vulture (*Gyps fulvus*)	1	23	1	12
	Black-winged kite (*Elanus caeruleus*)	–	–	–	2
Pandionidae	Osprey (*Pandion haliaetus*)	–	–	–	5
Tytonidae	Barn owl (*Tyto alba*)	–	2	2	11
Total		21	264	13	297
%		7.36%	92.63%	4.19%	95.48%

Table 2. *Salmonella* strains solated from captive raptors

Isolate	Host	Serotype/phage type
S2	Black vulture	Havana
S7	Kestrel	Havana
S8	Goshawk	Havana
S10	Kestrel	Brandenburg
S15	Lesser kestrel	Havana
S18	Goshawk	Havana
S19	Little owl	Havana
S21	Little owl	Havana
S22	Little owl	Havana
S24	Little owl	Havana
S26	Griffon vulture	Brandenburg
S27	Little owl	Newport
S28	Montagu's harrier	Havana
S29	Buzzard	Havana
S36	Lesser kestrel	Adelaide
S39	Lesser kestrel	Havana
S52	Scops owl	Enteritidis PT6a
S57	Kestrel	Hadar PT1
S79	Lesser kestrel	Havana
S80	Lesser kestrel	Havana
S81	Lesser kestrel	Havana
S82	Lesser kestrel	Havana

From the group of free-ranging raptorial birds, more variety of serotypes were obtained. Two *Salmonella* serotype Enteritidis PT6a strains (S6 and S51) were isolated from two lesser kestrels; one *Salmonella* serotype Enteritidis PNR (S66) was isolated from a barn owl. Three strains of *Salmonella* serotype Adelaide (S9, S14 and S20) were isolated from a sparrow hawk, an imperial eagle and a little owl. One isolate of *Salmonella* serotype Brandenburg (S17) from a lesser kestrel. One isolate of *Salmonella* serotype Newport (S27) from a barn owl. Two isolates of *Salmonella* serotype Typhimurium (S76 and S86) DT 104b and DT 104, respectively. Single isolates of *Salmonella* serotype Hadar PT5 (S83), Saintpaul (S84) and Virchow PT8 (S87) were also isolated.

Discussion

The role of wild birds in the epidemiology of salmonellosis has been studied as the possible risk that wild birds represent in the possible spread of salmonellae (Kapperud and Rosef, 1983; Levre et al., 1989; Gopee et al., 2000).

The incidence of salmonellae in poultry and domestic animals has been pointed as a major source of contamination for humans (Baker and Goff, 1982; Ekperegin and Nagaraja, 1988), but little is known about the real role of wild birds in this zoonoses.

In our studies, the percentage of *Salmonella* carriers was 7.36% for the captive raptorial birds and 4.51% for the free-ranging birds and none of them presented any symptomatology of salmonellosis. Similar results were obtained by Bangert et al. (1988), Battisti et al. (1998) and Gopee et al. (2000), indicating that the number of *Salmonella* carriers was very little.

Table 3. *Salmonella* strains isolated from free-ranging raptors

Isolate	Host	Serotype/phage type
S6	Lesser kestrel	Enteritidis PT6a
S9	Sparrow hawk	Adelaide
S14	Imperial eagle	Adelaide
S17	Lesser kestrel	Brandenburg
S20	Little owl	Adelaide
S27	Barn owl	Newport
S51	Lesser kestrel	Enteritidis PT6a
S66	Barn owl	Enteritidis PNR
S76	Buzzard	Typhimurium DT104b
S83	Little owl	Hadar PT5
S84	Little owl	Saintpaul
S86	Long-eared owl	Typhimurium DT104
S87	Griffon vulture	Virchow PT8

From the captive raptorial birds, the most frequent serotype isolated was Havana. *Salmonella* serotype Havana is rarely isolated in Spain and has never been ranked in the list of the commonest serotypes isolated from human and non-human sources (Usera, 1999). On the other hand, this serotype has also been associated with raptors (Battisti et al., 1998). These facts suggest the possible clonal origin of the strains. To asses the epidemiological relationship of the strains different typing methods were carried out which showed that all these strains had a clonal origin, although the origin of contamination could not be established (Reche et al. 2002). It was thought that there was a source of contamination in the Wildlife Hospital, so a microbiological screening of the hospital installations and the stocked frozen chickens was performed but *Salmonella* serotype Havana was never detected.

From the group of free raptorial free-ranging birds, the serotypes isolated were more heterogeneous, most of them were similar to those frequently reported either in humans or in domestic animals.

Further studies are necessary to determine the origin of the salmonellae reported here and the exact role, if any, of the birds of prey in transmitting the infection to domestic animals and humans, or vice versa.

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