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Isolation of *Salmonella* Enteritidis Phage Type 21b from a Eurasian Eagle-Owl (*Bubo bubo*)

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ABSTRACT: A case of fatal salmonellosis in a Eurasian eagle-owl (*Bubo bubo*) from Bursa Province (northwestern Turkey) is described. The organs of the bird were examined histopathologically and microbiologically. Macroscopic and microscopic findings were consistent with a *Salmonella* infection. *Salmonella enterica* subspecies *enterica* serovar Enteritidis (*S.* Enteritidis) phage type (PT) 21b was isolated from the liver and spleen in pure culture and from the intestine. The isolate was susceptible to amoxycillin/clavulanic acid, ampicillin, chloramphenicol, gentamicin, kanamycin, tetracycline, and trimethoprim/sulphamethoxazole. This is the first report of an isolation of salmonellae from a wild bird species from Turkey and the first time *S.* Enteritidis PT21b has been reported from Turkey.

Key words: Antimicrobial susceptibility, Eurasian eagle owl, *Salmonella* Enteritidis phage type 21b, wild bird.

A male Eurasian eagle-owl (*Bubo bubo*) found dead in a private garden in Bursa Province in June 2005 was submitted to the Faculty of Veterinary Medicine, Uludag University, Bursa, Turkey, for diagnostic evaluation. At postmortem examination, small whitish nodules of 1–3 mm in diameter were observed on the liver surface. The proventriculus mucosa and the passage into the ventriculus were covered with hemorrhagic exudate.

Tissue samples from various organs were routinely processed, embedded in paraffin, sectioned at 5 μm, and stained with hematoxylin and eosin. Smears of tissues were stained using Gram and modified Ziehl-Neelsen methods. Tissue samples also were inoculated onto xylose-lysine-desoxycholate agar (Oxoid Ltd., Basingstoke, UK) as well as other culture media routinely used for pathogenic bacteria and fungi. Presumptive *Salmonella* colonies were chosen and subjected to biochemical tests. Species and subspecies differentiation of the isolate were performed according to the method described by Popoff (2001). Serotyping and phage typing were performed according to the Kauffman-White scheme (Rowe and Hall, 1989) and Ward et al. (1987), respectively. The susceptibility of the *Salmonella* isolate to various antibiotics was examined by the Kirby-Bauer disc diffusion method (Bauer et al., 1966) on Mueller-Hinton agar (Oxoid).

Microscopic examination of tissues revealed areas of coagulation necrosis in the hepatic parenchyma, together with mononuclear cell infiltrations in the subcapsular and periportal areas. Walls of branches of hepatic artery and portal vein showed signs of degeneration, and the lumina contained thrombi. Coccidian merozoites and oocysts were observed within the epithelial lining of the small intestine together with moderate inflammation. Focal mononuclear cell infiltrations, to a lesser extent, also were observed in the lamina propria and submucosa of the proventriculus, small intestine, and in the heart (together with focal myocyte degeneration). Thrombi were present in the middle-sized vessels in the lung.

Microscopically, gram-negative bacilli were observed in large numbers on liver smears and in lesser numbers on spleen smears. *Salmonella* was isolated from the liver and spleen in pure culture, and also from the intestine. The isolate was identified as *Salmonella enterica* subspecies *enterica* according to biochemical reaction

results. In serotyping, the strain had the following antigenic formula: 9, 12: g, m: −, corresponding to serovar Enteritidis. The phage type of the serovar was identified to be 21b (PT21b). The isolated agent was susceptible to amoxycillin/clavulanic acid, ampicillin, chloramphenicol, gentamicin, kanamycin, tetracycline, and trimethoprim/sulphamethoxazole, but resistant to cephalothin and nalidixic acid.

Serovar Typhimurium is a well-known cause of salmonellosis in many wild bird species (Pennycott et al., 1998; Refsum et al., 2002, 2003). In this report we describe a case of fatal salmonellosis in an owl caused by *S.* Enteritidis PT21b. Isolation of the bacteria from the liver, spleen, and intestine indicates the presence of a septicemic infection in the bird. Coccidia observed in the intestine could have enhanced the severity of *Salmonella* infection in this bird. Death associated with salmonellosis caused by nontyphoidal serovars of *S. enterica* usually occurs in birds coinfected with other agents, especially with coccidia, presumably due to the increased ability of *Salmonella* to colonize the intestinal tract (Qin et al., 1995).

In the last few decades, *S.* Enteritidis has emerged as a major cause of foodborne illness worldwide (Agron et al., 2001). The serovar has been most commonly isolated from humans (Erdem et al., 2005) and chickens (Carli et al., 2001) in Turkey as well. However, there are no data regarding the existence of the agent in wild birds of the Turkish fauna, which may also act as an important reservoir for transmission of salmonellae to humans (Kapperud et al., 1998; Hudson et al., 2000). Kapperud et al. (1998) reported that sporadic cases of domestically acquired human salmonellosis and cases of fatal salmonellosis among small passerines in Norway were most often observed at the same time of year, thus indicating an epidemiologic link. In addition, a molecular epidemiologic study has shown that 32% of the isolates recovered from human patients belonged to clones detected in the small passerine fauna in Norway (Heir et al., 2002).

This is the first report on the isolation of salmonellae from a wild bird species of Turkish fauna, and the first time this phage type was documented in Turkey. The study indicates that there is strong need to clarify the distribution of salmonellae in wild bird populations and the potential epidemiological relationship between salmonellae in wild birds and humans in Turkey.

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