Veterinary Microbiology 173 (2014) 318–322

Contents lists available at ScienceDirect

Veterinary Microbiology

journal homepage: www.elsevier.com/locate/vetmic

Tularaemia in Norwegian dogs

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ARTICLE INFO

Article history:
Received 31 January 2014
Received in revised form 19 June 2014
Accepted 28 June 2014

Keywords:
Tularaemia
Francisella tularensis subspecies holarctica
Dogs
Lemmings
Hares
Humans

ABSTRACT

We describe tularaemia in a Norwegian dog caused by *Francisella tularensis* subspecies *holarctica*. A Hamilton Hound and his owner developed tulaeremia after hunting an infected mountain hare (*Lepus timidus*). The dog showed signs of lethargy, anorexia and fever during a period two to four days after hunting and thereafter fully recovered. Its antibody titers increased 32-fold from one to three weeks post exposure. Thereafter, the titer declined and leveled off at moderate positive values up to one year after exposure (end of study). This is believed to be the first case report of clinical *F. tularensis* subspecies *holarctica* infection in a European dog.

In 2011, enormous numbers of Norway lemmings (*Lemmus lemmus*) occurred in Finnmark, the northernmost county of Norway and many dogs caught and swallowed lemmings. Some of these dogs developed non-specific signs of disease and the owners consulted a veterinary surgeon, who suspected tularaemia. In order to investigate this hypothesis, serum samples from 33 dogs were examined for antibodies to *F. tularensis*. The dogs were allocated into three groups: Dogs from Finnmark that became sick (Group 1) or remained healthy following contact with lemmings (Group 2), and healthy control dogs from Oslo without known contact with lemmings (Group 3). All the serum samples were analyzed with a tube agglutination assay. Among dogs exposed to lemmings, 10/11 and 3/12 were antibody positive in Group 1 and Group 2, respectively, whereas none of the control dogs ($n = 10$) were positive for antibodies against *F. tularensis*. These results strongly indicate that the non-specific disease seen in the dogs in Finnmark was linked to *F. tularensis* infection acquired through contact with lemmings.

1. Introduction

Tularaemia is a zoonotic bacterial disease caused by *Francisella tularensis*. The two most important subspecies are *F. tularensis tularensis* (type A) occurring in North

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http://dx.doi.org/10.1016/j.vetmic.2014.06.031
0378-1135/

America and *F. tularensis holarctica* (type B) found in Europe, North America and Asia (World Health Organization, 2007). *F. tularensis* has a very broad host range. However, the susceptibility and sensitivity to infection varies greatly between animal species, and tularaemia is primarily a disease of lagomorphs and rodents (Hopla and Hopla, 1994). In Norway, the mountain hare (*Lepus timidus*) is the only wild lagomorph present. This hare species is very sensitive to *F. tularensis* infection and normally dies within a few days post exposure (Borg et al., 1969; Mörner and Sandstedt, 1983). Thus, the mountain hare should be regarded a good sentinel but not reservoir for *F. tularensis* in the Norwegian nature. The main animal reservoirs are considered to be rodents, particularly the Norway lemming (*Lemmus lemmus*) that occurs in enormous numbers in certain years (so-called lemming years). A human disease called lemming fever, almost certainly tularaemia, was described more than 100 years ago in Norway (Horne, 1911). This disease was associated with lemming years and contamination of local drinking water sources. Also later reported human outbreaks of tularaemia in Norway have been associated with high densities of lemmings and contamination of drinking water (Berdal et al., 2000; Fossum et al., 2002; Larssen et al., 2011).

Dogs are considered relatively resistant to *F. tularensis* (Foley and Nieto, 2010) and to the authors' knowledge no reports of canine tularaemia have been published from Europe. However, in the USA, canine infection has been associated with subclinical or mild disease (Calhoun et al., 1955; Schmid et al., 1983; Gustafson and DeBowes, 1996; Leighton et al., 2001; Meinkoth et al., 2004).

During the summer and autumn 2011, enormous numbers of Norway lemmings occurred in Finnmark, the northernmost county in Norway (Rolf Ims, University of Tromsø, personal communication). At the same time, several cases of tularaemia in the mountain hare were for the first time diagnosed in this county by the Norwegian Veterinary Institute (NVI) and 46 cases of human tularaemia were recorded (Norwegian Surveillance System for Communicable Diseases). In the same period, many dog owners consulted veterinarians in Finnmark with dogs showing non-specific signs of illness. The dogs had a history of exposure to lemmings and tularaemia was suspected. Serum samples from four dogs were examined for antibodies to *F. tularensis* and all tested positive. These observations indicated a relationship between clinical signs and infection with *F. tularensis*.

This paper presents a case report of natural infection in a dog and its owner after exposure to a tularaemia-sick mountain hare during hunting. In addition we report the results of a serological pilot study on *F. tularensis* conducted in groups of dogs after exposure to lemmings in 2011 and, for comparison, in dogs with no history of such contact.

2. Materials and methods

2.1. Case report

During hunting (September 2012) in Central Norway an adult mountain hare was caught and killed by the hunting dog, a 1.5-year-old Hamilton Hound. The hare was dressed by the hunter the same day and hung. A few days later the dog and the hunter became sick and the hunter suspected tularaemia. The hare carcass was delivered for post mortem examination at the NVI six days after hunting and the hunter was advised to take immediate contact with his physician for medical examination. He was also encouraged to visit a veterinary clinic to have his dog examined and blood sampled for serological testing on tularaemia.

The hare was submitted to the NVI as an eviscerated carcass with no internal organs and examined following standard procedures. The femurs were split and the bone marrow was inspected. Bone marrow samples were obtained for histopathological and immunohistochemical examinations as well as for bacterial culturing. Samples for histopathology and immunohistochemistry were fixed in 10% buffered formalin, embedded in paraffin and sectioned at 5 μm. Slides for histopathological examination were stained with haematoxylin and eosin, whereas those for immunohistochemical examination were processed using a commercial anti-*F. tularensis* primary monoclonal antibody (C86316M, BIO, Maine, USA). Bacterial culturing was performed on chocolate agar plates incubated in 10% CO_2 at 37 °C. Typical colonies were verified by realtime PCR on bacterial DNA from inactivated colony material using primers and probes specific for *F. tularensis* subspecies *holarctica* (realtime PCR developed at the Swedish Institute for Communicable Disease Control, unpublished).

The hunter was examined by his physician at day 6 after hunting. He was given antibiotic treatment on clinical suspicion of tularaemia. Paired blood samples taken on the day of consultation and 3 weeks later were analyzed for *F. tularensis* antibodies by in-house microagglutination (Bevanger et al., 1988) as well as IgM and IgG ELISA (Serion ELISA Classic *Francisella tularensis* IgM/IgG, Serion immunodiagnostica, Würzburg, Germany). Data from the hunter is included after informed written consent.

The hunting dog was examined at the veterinary clinic at day 7 after hunting. Blood samples were drawn from the dog for tularaemia serology, clinical biochemistry and hematology. Swab samples were obtained from the tonsils. Thereafter, convalescent-phase sera were sampled two weeks after the first sampling and subsequently at 3, 6, 9 and 12 months post exposure.

2.2. Pilot study

Three groups of dogs were included in the serological pilot study. Group 1 comprised dogs from Finnmark that became sick shortly after exposure to lemmings. They were examined in the acute stage at a veterinary clinic and treated with antibiotics. Some of the dogs were blood sampled during the consultation, whereas others were recalled and blood sampled in a period varying from 1 week to 8 months after the disease consultation. Group 2 included dogs from Finnmark with no history of clinical disease after exposure to lemmings. All dogs in groups 1 and 2 had, according to their owners, been observed while catching and swallowing lemmings. Group 3 comprised healthy dogs from the city of Oslo that reportedly never had been in areas with lemmings.