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Short communication

Haemorrhagic enterotoxemia by *Clostridium perfringens* type C and type A in silver foxes

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Abstract

Type C and type A of *C. perfringens* were detected in the seat of natural infections in silver foxes characterized by symptoms of haemorrhagic enterotoxemia. In all of the dead foxes characteristic changes were noted in the small intestine and parenchymatous organs. The production of *alpha* and *beta* toxins by isolated bacteria was confirmed by the bioassay using white mice and by PCR. The results of the drug sensitivity testing showed that isolated strains were highly susceptible to amoxicillin with clavulanic acid, metronidazole, doxycycline and penicillin with streptomycin.

Key words: *Clostridium perfringens* type C and type A, silver foxes, haemorrhagic enterotoxemia

Introduction

Anaerobic infections caused by bacteria of the genus *Clostridium* have recently played an increasingly important role as a cause of fatal diseases, predominantly in newborn animals. (Sasaki et al. 1999, Uzal et al. 2008). Most published reports on anaerobic infections in foxes and other fur-bearing animals deal with intoxications or toxoinfections caused by *Clostridium botulinum* (Uzal et al. 2008). There is little information on the occurrence and course of haemorrhagic enteropathy caused by *C. perfringens* type C and type A in silver foxes. High numbers of *C. perfringens*, mainly of the type A, may be present in the intestinal tract of animals with no visible signs of disease.

However, this phenotype as well as type C are consistently associated with enteric diseases, especially under conditions allowing the organisms to grow and produce toxins. Our observations show that generalized infections caused by *C. perfringens* can break out in silver foxes under farm conditions. The aim of the study was to isolate, identify and characterize anaerobic bacilli from the seat of natural infections in silver foxes characterized by symptoms of haemorrhagic enterotoxemia.

Materials and Methods

The study was conducted on a silver fox farm with a foundation stock of 160 females and 45 males aged

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1-4 years. All foxes that died in the peracute course of the disease were subjected to detailed laboratory examination including gross pathology, bacteriology and histopathology. Additionally, bacteriological examination was performed on feed samples. Colonies of the morphology suggestive of anaerobic bacteria were subjected to biochemical assay with the use of API 20A kit (Bio Merieux). Toxins were detected by biological assay using white mice according to Knight et al. 1990. Additionally, the presence of toxin genes encoding the production of *alpha* (CPA) and *beta* (CPB) toxin in isolated *C. perfringens* strains were confirmed by PCR according to Griffiths et al. 1997. Isolated bacterial colonies were subjected to drug sensitivity test according to the CLSI/NCCLS M11-A7 recommendations.

Results and Discussion

Characteristic clinical symptoms in the peracute course of the disease including copious foamy diarrhoea mixed with blood, dyspnea and respiratory and cardiac disturbances resulted in sudden death of 45% of the stock within 8-12 hours. Just before death the animals had subnormal body temperature and symptoms of considerable weakness, depression, and sometimes nervous disorders in the form of muscle tremors or paresis of the limbs.

Gross pathological examination revealed characteristic changes typical to haemorrhagic enterotoxaemia in all of the dead foxes. Histopathological examination revealed deep necrosis of the intestinal villi, in which numerous inflammatory cellular infiltrates were observed in the HE-stained slides. Bacteriological examination combined with the results of the biochemical assays using the API 20 A kit confirmed that the bacteria isolated from dead foxes and feed samples belonged to the genus of *C. perfringens*. The bioassay using white mice showed that isolated strains were capable of producing *alpha* and *beta* toxins. The presence of toxin genes, α/cpa and β/cpb , in isolated bacteria was confirmed by PCR. Drug sensitivity testing performed on the isolated *C. perfringens* strains showed that they were highly susceptible to amoxicillin with clavulanic acid, metronidazole, doxycycline and penicillin with streptomycin. The strains showed intermediate sensitivity to gentamicin

and streptomycin and were resistant to tetracycline, enrofloxacin, colistin, sulphonamides with TMP and cephalosporines.

The results of this study show that anaerobic infections by *C. perfringens* type A and C in breeding foxes can result in mass outbreaks of disease with a severe course, which have a high death rate and cause substantial economic losses.

The source of infection in such outbreaks is not easy to detect, especially as these bacteria can be isolated from the alimentary tract of healthy animals. In fur-bearing animals there is usually a secondary source of anaerobic bacilli limited to feed contaminated by spores or vegetative cells (Sasaki et al. 1999). Our study showed that the main source of infection in the foxes was feed containing *C. perfringens*. Germination of *Clostridium* spores and toxin production by vegetative forms of anaerobes were most likely caused by the use of uncooked feed and by feed preparation using intermediate products contaminated by spores (Wojdat et al. 2006). Prevention of anaerobic infections in fur-bearing animal farms should thus include, apart from general hygiene procedures, sanitary control over feed production and storage, as well as microbiological cultures of feed samples. Alternatively, the use of thermal processing to inactivate vegetative forms and spores is recommended.

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