

Case Report: Two Cases of Ocular form of Listeriosis in Cattle Herds

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ABSTRACT

Nowadays, the widespread use of silage has led to an increased frequency of bacterial contamination of *Listeria* spp in ruminants. The infection of ruminants is essentially linked to the consumption of silage of poor quality or of milk products. The disease is frequent and severe in ruminants and can be present in various forms: nervous, septicaemic, genital, mastitis and ocular. The ocular form of Listeriosis can serve as a sentinel for the disease at the herd level. In this case report, 2 herds were investigated for calves and adult mortalities. In the first case, a large majority of the calves presented with hypopion but neurological examination of the calves revealed no abnormalities. The majority of the dairy cows also presented with ocular disorders but neurological examination did not reveal any abnormalities. Calves received whole milk from the dairy herd. The dairy herd diet comprised maize and grass silage, and cereals. *Listeria monocytogenes* was found in milk and grass silage in significant amounts. In the second case, weight loss, abortions and mortalities in adult cattle were reported. Fifty percent of animals examined presented with hypopion, and/or keratoconjunctivitis, and/or uveitis. No neurological abnormality was found. The animals' ration comprised grass and maize silages. The grass silage was not well made. Analysis revealed the presence of *Schizophyllum commune* (fungus) and *L. monocytogenes* in significant amounts. Furthermore, analysis of aborted fetuses also demonstrated *L. monocytogenes*. Listeria infection in cattle can occur through ingestion of contaminated silage but calves can also be infected throughout the dams' milk. Ocular lesions are not pathognomonic for listeriosis but, accompanied with abortions and poor quality silage, this can be a major indication for ancillary exams for *Listeria monocytogenes* at a herd level.

KEYWORDS: Silage, milk, Listeria, ocular disease, abortion, ruminants

INTRODUCTION

Listeriosis is a bacterial infectious disease, common to human beings and other mammalian species. *Listeria* is a gram-positive intra-cellular and facultative aerobic organism. Only 2 species of *Listeria* have clinical relevance: *Listeria monocytogenes* and *Listeria ivanovii*. The latter, isolated in small ruminants and cattle, is of minor importance. The bacterium is an opportunist pathogen present in soil, silages, and rivers, but also in the digestive system, genital and nasal secretions of numerous animal species leading to the presence of healthy carriers. Furthermore, *Listeria* is resistant to numerous antiseptics, heavy metals, bile, freezing and thawing, and classical pasteurisation (58°C for 10 minutes). The bacteria can survive in the field for months (straw, soils, fecal material) and multiply at temperatures as low as 4°C. Animal contamination is essentially linked to the consumption of silage of poor quality (ruminants) or of milk or beef products (humans and carnivores).

The disease is frequent and severe in ruminants (Cooper & Walker 1998, Erdogan *et al.* 2001), particularly since the generalisation of the use of silages (Farber *et al.* 1988, Husu 1990). Different forms of Listeriosis are described in cattle: a nervous form (encephalitis usually leading to death), a septicaemic form (in the newborn or premature), a genital form (abortion), a mammary form (mastitis) (Low & Renton 1985, Fedio *et al.* 1990, Low &

Donachie 1997, Radostits *et al.* 2007), and an ocular form (uveitis, kerato-conjunctivitis, hypopion) (Welchman *et al.* 1997, Ravary 2000, Laven & Lawrence 2006, Radostits *et al.* 2007). *Listeria* has the particularity of forming micro-abscesses in organs. The nervous form is the easiest to diagnose due to its typical clinical pattern. However, the other clinical forms must not be under-estimated. For example, diagnosing the ocular form of listeriosis at herd level, could allow a prophylactic approach before confrontation with other clinical forms of the disease (Ravary 2000). The present case report shows how the ocular form of listeriosis can serve as a sentinel for the disease at a herd level.

CASE REPORT

Two herds were investigated for calf and adult mortalities. Both herds comprised beef (Belgian Blue (BB)) and dairy (Holstein) breeds.

Herd #1

This herd was a mixed population of beef and dairy cattle (50-50%) with 65 calvings per year concentrated between September and December. The milk quota (209,000l) was supplied by 30-35 Holstein cows. The farmer called the Clinic for diarrhoea, weight loss, and septicaemia in young calves. Generally, treatment of ill calves with antibiotics (enrofloxacin, 2.5 mg/kg body weight IV) proved successful. However, two calves died before

the veterinary farm visit. Only one of the latter calves underwent necropsy in the field. The lesions observed suggested a *Salmonella* infection but the bacteriological examination for *Salmonella* was negative. The calf had been treated with antibiotics before its death.

The main problems observed during the farm visit was bilateral white opacities in the eyes of young calves, both dairy and beef breeds. Indeed, out of 32 calves examined, 23 (72%) presented this clinical sign. The eye lesions were characterised by a decolouration of the iris (rare) (Figure 1) accompanied by turbidity in the anterior chamber (Figure 2). The latter lesion is indicative of hypopyon (pus in the anterior chamber of the eye) and could be a post-septicaemic localisation of infection. Neurological examination (exploration of cranial nerves) of the calves revealed no abnormality. Ten of the 32 calves examined (31%) presented diarrhoea (age range: 6-25 days) as well as ocular disorders.

Figure 1.



Figure 2.



Clinical examination of adult cattle was performed. Out of 30 dairy cows examined, 25 (83%) presented keratitis, or keratoconjunctivitis, and/or epiphora, and/or turbidity in the anterior chamber of the eye and/or cataract. Neurological examination (exploration of cranial nerves) of the cows revealed no abnormality. Abortions were not reported on the

farm. Occurrences of mastitis were recorded by the farmer but no bacteriological analyses were performed. Treatment against mastitis, by the veterinarian and sometimes by the farmer himself, gave good results. On the day of the visit, no clinical mastitis was observed. Beef cows (20 animals) did not present with any clinical disorder. Beef and dairy calves were mainly fed fresh milk produced by the dairy cows only. The milk was not pasteurised and was given directly to the calves after milking. The calves also received calf-starter, fresh water and hay of good quality. The hygiene in the barn housing the calves (on straw) was perfect. All calves received fresh and frozen colostrum from dairy cows (containing 150 grams of total protein per litre) immediately after birth (3-4 litres/calf). Dairy cows were vaccinated with Bovilis Lactovac®, Pfizer Limited (*Rotavirus, Coronavirus, E.Coli F5/F41*) or Trivacton®, Merial Animal Health Ltd (*E.Coli F5/F17/F41/31A, Rotavirus, Coronavirus*).

Dairy cows were fed grass and corn silage, hay, cereals, concentrate with 35% crude proteins and minerals. Beef cows were fed corn silage (small quantities), hay, cereals and another concentrate with proteins. The grass silage was made directly on the ground. Hygiene of the silage was poor (Figure 3). A large amount of mud contaminated the grass when giving the silage to the dairy cows. Moreover, the temperature of the silage was high (mean of 37°C with an external temperature of 5°C). The corn silage had a better macroscopic aspect and its temperature was normal (10°C).

Figure 3.



Bacterial cultures of the grass silage demonstrated the presence of *Listeria monocytogenes* (Test Rapid L.mono®). A sample of bulk milk was taken and also revealed the presence of *L. monocytogenes*.

Herd #2

This herd included beef and dairy cattle (50-50%) with 100 calvings per year. The milk quota was 170,000l. Vaccination against BVD and BHV-1 (IBR) but no anti-parasitic treatment was performed on the farm. The farmer called the Clinic for

problems such as weight loss, abortions (4 cases) and mortalities (3 cases) in adult cattle.

The adult cattle were in poor body condition, both dairy and beef breeds. At least 50% of the examined animals (30 BB and 25 Holstein) presented with hypopyon (Figure 4), and/or keratoconjunctivitis, and/or uveitis. On these animals, a rapid neurological examination was performed (test of cranial nerves) but no abnormality was observed. No mastitis was recorded by the farmer at the time of the visit. Calves presented with mild growth retardation but the farmer did not mention any specific problem (diarrhoea, respiratory trouble, septicaemia). The calves received milk-replacer, calf-starter, good hay and water. Colostrum from the dams was given to the calves during their first day of life. No precise information about quantity or quality of colostrum was available.

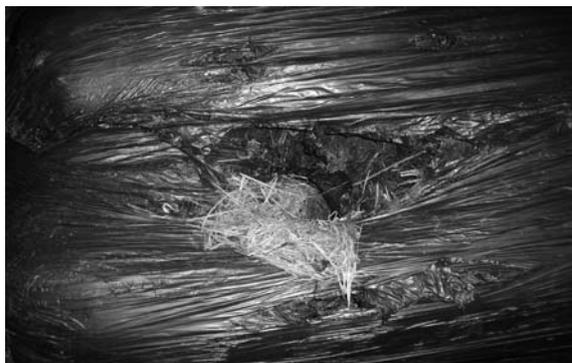
Figure 4.



The ration of animals was composed of grass and corn silages, and minerals for dry cows; grass and corn silages, cereals, hay and minerals for lactating animals. The corn silage was of good quality and well conserved (temperature and pH in normal range). Big bales of grass silage were pierced by the claws of the tractor and transported through the barn (Figure 5). The grass silage stayed for 3 to 7 days in the barn before being distributed. Moreover, several of the bales left outside were already pierced by birds. When opened, the bales of silage were wet, warm (40°C at depth) and smelt of alcohol. Large areas of white mould were present on the surface and at depth.

Necropsies (4) revealed the presence of multi-focal abscesses on the liver, lungs, and brain (3 cows). Lesions of liver fluke were also observed (4 cows). Analyses for BVD and BHV-1 (IBR) were negative. Analyses performed on aborted foetuses (2) revealed the presence of *L. monocytogenes*. Cultures were performed on the grass silage leading to the identification of a fungus, *Schizophyllum commune*. This fungus is a saprophytic species with no clinical significance to humans; however no data is available regarding its potential toxicity in cattle nutrition. Furthermore, *L. monocytogenes* was identified in two samples of grass silage.

Figure 4.



DISCUSSION AND CONCLUSIONS

In the first herd, infection of the calves by *Listeria monocytogenes* was due to consumption of milk produced by the farm's dairy cows. The latter cows were infected with *L. monocytogenes* by eating contaminated grass silage. Furthermore, liver fluke contributed actively to the poor body condition of the herd.

Listeriosis cannot be diagnosed exclusively on the basis of clinical signs. Differential diagnosis includes contagious ophthalmia, bovine keratoconjunctivitis, septicaemia of other aetiologies (ocular form), nervous ketosis, rabies, polioencephalomalacia, middle ear disease, BSE, *Salmonella* (enteric form), most causal agents of abortion such as *Salmonella*, bluetongue, *Brucella*, *Neospora*, IBR, *Leptospira* (abortive form), and most agents of subclinical mastitis (Radostits 2007).

Bacteriological testing (Walker 1999) and histological examination are the classical methods used for laboratory diagnosis of listeriosis in animal specimens. Bacteria can be isolated in bulk milk. The origin of the latter bacteria can either be environmental or due to the presence of a single, or several, infected animals with mastitis (Fedio *et al.* 1990, Schoder *et al.* 2003). Bacteriology can also be performed on conjunctivae (with sterile swab AMIES) in live animals, or sterile puncture of aqueous humour in dead animals. Serological diagnosis is not very useful because numerous healthy animals have antibodies to *L. monocytogenes*. Furthermore, development of antibodies is irregular and slow, even during clinical disease. Polymerase Chain Reaction (PCR) can detect bacteria in milk samples, even after administration of antibiotics in animals. Nevertheless, the combination of serology and PCR can lead to a diagnosis of listeriosis in ruminants, even after the administration of antibiotics (Amagliani *et al.* 2006).

When diagnosis on animals is not possible, *e.g.* because of lack of diagnostic means (PCR or serology), the presumption can be made on the basis of feedstuff. Culture of silage samples (often grass

silage) can show evidence of *Listeria*.

Poor nutritional status, stress of late pregnancy and parturition are host risks that increase the susceptibility of animals to *Listeria*. Poorly fermented silages, with pH above 5.5, aerobic deterioration, contamination by soil, or big bale silages (low density, poor fermentation, greater risk for mechanical damage to the plastic covering) are at risk for *Listeria* growth.

Finally, farmers who consume raw milk need to be aware of the risk of infection, especially if their cattle are fed silages. Moreover, conjunctivitis has been recorded in agricultural workers handling infected livestock.

Hypopyon in cattle fed silages may be suggestive of listeriosis but requires confirmation by complementary examinations, both in animals and in the feedstuffs they consume.

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