From the State Veterinary Research Station for Small Ruminants, Høyland, Sandnes, Norway.

CLINICAL INVESTIGATIONS IN A GOAT HERD WITH OUTBREAKS OF LISTERIOSIS*

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— The majority of 50 nonpregnant dairy goats in a herd showed acute symptoms of disease for 1—2 days after a sudden change in weather conditions, feeding regime and management. In 17 of the does the condition progressed with symptoms of septicaemia with depression, fever, inappetence and hypogalactia for a period of up to 5 weeks. Only 1 of these 17 animals died. Three weeks after the first symptoms were seen, 7 other animals showed typical symptoms of listeric encephalitis. All but 1 recovered within 2 weeks. Treatment with penicillin seemed to have good effect against both these disease forms.

The does with symptoms of septicaemia had significantly higher antibody titres against Listeria monocytogenes than the animals with encephalitis and the remainder.

Listeria monocytogenes; goats; septicaemia; encephalitis.

Listeric encephalitis is rather common in goats (King 1940, Gall 1962, Wood 1972, Dijkstra 1979). Although outbreaks of listeric abortion may occur sporadically (Amanshulov 1953, Sandbu 1956), only 1 case of listeric septicaemia in adult goats seems to have been reported (Jansen & van den Hurk 1943). The present work describes the investigation of outbreaks of assumed listeric septicaemia followed by encephalitis in a herd of non-pregnant goats.

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HISTORY OF THE OUTBREAKS

The herd consisted of 50 non-pregnant dairy goats and 17 kids about 7 months old, and 4 male goats not included in this investigation. Most of the animals belonged to the Norwegian breed, the remainder was cross-bred with Saanen. There had been no disease problems in the herd and the incidence of listeriosis had been low, with one case of encephalitis the previous winter.

Until Sept. 17th the goats had been kept on uncultivated pastures and fed a commercial supplementary mixture of concentrates which contained 12.5 % digestible crude protein. On that day the weather was unusually cold and rainy. The herd was therefore given grass silage from a newly opened silo, and the does also received 0.1 kg supplement consisting of dried fish and seaweed. The herd was still kept on pastures during that day. Since most of the does were ill on Sept. 18th, the herd was kept indoors from then on and fed hay and freshly harvested grass during the following week. Thereafter grass silage was again gradually introduced. Concentrates were given as usual all the time, but feeding of the fish and seaweed supplement was terminated after 2 days.

The herd was housed on slatted floors with the kids in a separate pen. The stables were overcrowded and had inadequate ventilation. The mating season started on Oct. 1st.

On Sept. 18th the majority of the does were depressed, had poor appetite and reduced milk yield, and some were febrile. In most does the symptoms lasted for 1—2 days only, but 7 does showed more severe and protracted symptoms for up to 5 weeks. Some does had fever up to 41° C, and some had diarrhoea with faeces occasionally mixed with blood. Later, another 10 does were similarly affected. As can be seen from Fig. 1 these 17 does became sick during a period of 3 weeks. One doe with severe symptoms from Sept. 18th died on Oct. 4th, whereas the remainder of the 17 does recovered within October. Ten does were treated intramuscularly with 2.5 g penicillin daily for 1—3 days, but only treatment for 3 days seemed to have good effect.

Two does severely affected were submitted to this laboratory on Sept. 25th. A clinical examination revealed similar symptoms as given for the other affected does. They recovered within few days when treated intramuscularly with penicillin.

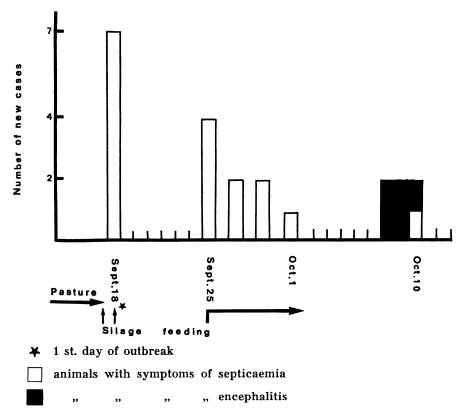


Figure 1. The daily number of new cases of assumed listeriosis during a period with outbreaks in a herd of 65 goats. Does with transient symptoms are not included.

During the period from Oct. 7th to Oct. 10th (Fig. 1), 5 other does and 2 kids showed typical symptoms of listeric encephalitis with depression, reduced appetite, incoordination and torticollis, and one had paralysis of facial muscles and salivation. They were all treated intramuscularly with 2.5 g penicillin/2.5 g dihydrostreptomycin daily for 3 days. One doe died but was not necropsied, whereas the 6 others recovered within 2 weeks.

LABORATORY METHODS

Bacteriological examination

Faeces from the 2 does submitted was examined for Yersinia enterocolitica as described by Krogstad (1974), for Listeria

monocytogenes (Lm) as described by $Gr\phi nst\phi l$ (1979a) and for Salmonella spp. by the method given by $Kj\phi lleberg$ (1972).

Bacteriological examination of a necropsied doe included inoculations from 6 organs on blood agar plates which were incubated at 37° C for 24 h. Intestinal contents were examined for Salmonella, and smears from the superficial layer of the mucosa were stained by the Ziehl-Neelsen and May-Grünewald-Giemsa methods. Toxicity test for Pulpy Kidney disease was performed on mice with ileal contents as described by *Stevens* (1959).

Samples of silage and fish and seaweed supplement taken on Oct. 12th were examined for Lm as described by *Grønstøl* (1979a).

Serological examination

Twenty of the 22 affected animals and 6 others were bled on Oct. 12th and the remaining 39 on Oct. 23rd. The sera were stored at -20° C until examined for haemagglutinating antibodies to Lm as described by $Gr \phi nst \phi l$ (1979a). Sera sampled on Oct. 12th were also examined for agglutinating antibodies to Yersinia enterocolitica type 2^* .

Haematological examination

Blood samples collected on Oct. 12th were examined for packed cell volume, number of erythrocytes and leucocytes, amount of haemoglobin and total serum protein. Blood sampled from the 2 does on the day when submitted was examined in the same way. In addition glucose, sodium and potassium in plasma and calcium, magnesium, inorganic phosphorus, iron, copper and zinc in serum were estimated. The methods routinely used in this laboratory have been described by Øverås (1969, 1974), Waldeland (1977) and Grønstøl (1980 b).

Egg counts

Faecal nematode egg counts per g (EPG) were estimated in the 2 does submitted by a slightly modified McMaster method described by Gordon & Whitlock (1939).

^{*} This examination was performed at the Department of Microbiology and Immunology, Veterinary College of Norway, Oslo.

Transmission experiment

Faeces from 7 does that became ill during the first outbreak was collected on Oct. 12th, mixed with water and given orally to 2 lambs. One lamb was dosed on the day of collection, the other after storage of the faeces at room temperature for 5 weeks. The lambs were examined clinically and the temperatures were recorded daily during an observation period of 2 months for the first dosed lamb and 1 month for the other.

Post mortem examination

The doe which died on Oct. 4th was necropsied in a routine manner. Sections of brain, liver, myocardium, lungs and kidneys were fixed in buffered 10 % formalin and stained with haematoxylin & eosin*.

RESULTS

Bacteriological examination

Neither Yersinia nor Salmonella were demonstrated in faeces from the 2 does. Lm serotype 4 was isolated from 1 of them. No pathogenic organisms were isolated from the necropsied carcass, and the clostridial toxin test was negative. Lm was not isolated from silage or supplement.

Serological examination

The reciprocal geometrical mean titre (GMT) of antibodies to Lm was 294 in sera from 16 of the 17 severely affected does, 25 in the 6 animals with encephalitis and 20 in the healthy animals. The difference between GMT for the 16 does and GMT for the healthy animals was statistically significant (P < 0.001), and also between the 16 does and the encephalitis group (P < 0.02). Titres of antibodies to Yersinia enterocolitica were < 10 (lowest dilution was 1/10) in 22 animals and ranged from 20 to 80 in the 4 remaining animals.

Haematological examination

The values were all within normal ranges.

^{*} The sections were prepared at the Regional Veterinary Laboratory, Sandnes, Norway.

Egg counts

Nematode EPG in the 2 does were 6000 and 400 respectively. Eggs of cestodes and oocysts of coccidia were not seen.

Transmission experiment

The 2 dosed lambs showed no symptoms or elevated temperatures.

Post mortem examination

No gross lesions were present at necropsy of the doe. Histopathological lesions of the brain consisted of perivascular cuffings of mononuclear and polymorphonuclear cells and multiple microabscesses in the medulla, with similar but not so pronounced lesions in pons and cerebellum. Some focal infiltrations of mononuclear and polymorphonuclear cells were present in the liver and kidneys. The liver was clearly affected with swollen and some degenerated hepatocytes.

DISCUSSION

The symptoms in the affected does indicated that different diseases had to be considered, such as septicaemia induced by infection of the alimentary tract. Therefore, the laboratory examinations included methods to diagnose listeriosis, salmonellosis and yersiniosis. In addition, subacute Pulpy Kidney disease was taken into consideration as it is very common in adult goats under our feeding conditions.

The diagnostic work revealed no evidence of salmonellosis, yersiniosis, or Pulpy Kidney disease. On the other hand, both the clinical and the laboratory examinations clearly indicated that the outbreak was caused by Lm-infection. Firstly, the symptoms and the course of disease in the 17 severely affected does followed the pattern of listeriosis in sheep described by *Palsson* (1963). However, the lethality was lower in the present outbreak in goats. The suggested minimum incubation period of about 1 day is short compared to the 3—4 days described in sheep by *Palsson* in septicaemia and *Grønstøl* (1979b) in an outbreak of listeric abortion.

No symptoms were observed in the kids during the first outbreak. However, weak and transient symptoms might have been overlooked by the owner since the kids were not milked and fed individually.

The GMT in the 16 of the 17 does with severe symptoms was significantly higher than that of the animals without pronounced symptoms as well as that of the animals with encephalitis. These results strongly indicate that the 16 sick does had listeric septicaemia. This is in accordance with findings in sheep ($Gr \phi nst \phi l$ 1979 b), and was further supported by high excretion rate of Lm in the herd during a few weeks after the outbreaks ($L\phi ken$ et al. 1982).

Only 1 of the 2 does presented for clinical examination had a high faecal egg count. They both recovered before treatment with an anthelmintic, and there is no reason to believe that parasites caused the outbreak.

The histopathological brain lesions in the one which died of the 17 severely affected does were characteristic for listeric encephalitis, and the findings in the liver and kidneys indicated a recent bacteraemia. Lm was not isolated from any organs, a fact which may be due to the antibiotic treatment 1 day after the symptoms were first observed. The unusually short incubation period of about 1 day was probably due to haematogenous spread of Lm to the brain in this case, although infection before Sept. 17th can not be ruled out.

The most common form of listeriosis in ruminants is encephalitis, whereas septicaemia is the most prevalent form in monogastric animals and newborn ruminants (Gray & Killinger 1966). Listeric encephalitis is a common disease in goats in Norway, and listeric abortion have been diagnosed in several outbreaks (Sandbu 1956, Grønstøl 1975 unpublished). In adult sheep individual cases of septicaemia have been found occasionally in this country (Grønstøl & Ulvund 1977), and there are reports of flock outbreaks of septicaemia in other countries (Gray 1963, Palsson). As a high proportion of healthy sheep seems to carry Lm (Grønstøl 1979a, 1980a), the animals are probably fairly resistant against the bacterium. In general, transient or inapparent listeric infections may be more prevalent among animals than reports indicate (Hyslop 1975).

Feeding of new silage of poor quality, together with sudden changes in feeding, management or weather conditions may be important predisposing factors of listeriosis in ruminants (*Gray*, Hyslap, Grønstøl 1979c). Palsson reported that outbreaks of listeriosis in sheep frequently started 3—4 days after introduction of new grass silage. Lehnert (1964) found that silage feeding might lead to a latent or transient infection with Lm in a large proportion of sheep and cattle, usually with bacteraemia and fever for 1—2 days.

In the present herd the feeding of grass silage from a newly opened silo, the introduction of fish and seaweed supplement and the sudden change in weather conditions might have disposed for the outbreak of disease about 1 day later.

Lm was not isolated from the fish and seaweed supplement, which was commercially prepared with preservatives and distributed in 50 kg's paper bags. No disease-inducing effect had been reported. Lm was neither isolated from the silage, although the samples had pH 4.4—4.6, a level somewhat above that indicating best quality. However, silage from the top layer given during the day before the outbreak was not available for examination at the time of investigation. It is therefore not possible to establish whether Lm had been present. The top layer of a silo is usually of inferior quality with a high pH, and may contain large numbers of Lm even if the bacteria can not be demonstrated in the rest of the silage ($Gr \phi nst \phi l$ 1979c). Gray found a wide variation in distribution and density of Lm within and among samples from each silo.

The pathogenesis seems to be similar in septicaemia and abortion, and differs from that of encephalitis (Grønstøl 1980a). In both septicaemia and encephalitis the bacteria seem to be introduced with the feed and cause a transient bacteraemia. The initial bacteraemia may occasionally progress to septicaemia (Hyslop), and may often induce listeric placentitis and abortion in pregnant animals (Njoku & Dennis 1973, Clegg 1975). In most cases of encephalitis, the bacteria probably migrate along the cranial nerves to the brain stem (Asahi 1963, Charlton & Garcia 1977), and cause an inflammation in the brain tissue. The incubation period may vary, but is probably usually as long as 2-3 weeks. However, Lehnert demonstrated that cerebral infections with Lm in sheep can occur after haematogenous spread of Lm. Incubation period of 1 day in listeric encephalitis in goats after change in management has been recorded by Wood (1972). An immunopathological reaction may also contribute to the

clinical symptoms as discussed by Kuchembuck (1978) and $Gr \phi nst \phi l$ (1980c), especially in animals previously sensitized to Lm.

It is noteworthy that none of the 16 sick does which recovered developed listeric encephalitis later, and that the 7 animals with encephalitis apparently recovered temporarily after the initial febrile period of 1—2 days. This supports the theory of different pathogenesis in septicaemia and encephalitis.

According to Gray, outbreaks of listeriosis in sheep flocks occur either as encephalitis or abortion. This is in agreement with observations in sheep and goats in this country; however, in 2 goat herds Sandbu and Grønstøl (1975 unpublished) diagnosed cases of both abortion and encephalitis in the same outbreak. In Iceland Palsson frequently observed more than one form in the same outbreak. He found that transient disease without characteristic symptoms often occurred initially. Some of these progressed further to septicaemia. When encephalitis was observed in outbreaks, the milder cases were often overlooked since these occurred earlier but were usually more numerous. The course of the present outbreaks was in accordance with the description given by Palsson.

The lethality of encephalitis in the present outbreak was surprisingly low compared with that observed in sheep (Lippmann 1969, Kummeneje 1979). In our experience the lethality of listeric encephalitis is higher in goats than in sheep. The question arises whether the does in the present herd had a low initial cell-mediated immunity against Lm and thus a weak immunopathological response causing less damage in the brain than usual, according to the hypothesis by Kuchembuck and Grønstøl (1980c). It might also have been of importance that the animals were not pregnant.

The present investigation indicates that changes in management, feeding and weather conditions may predispose for listeric septicaemia and encephalitis in goats, and that the pathogenesis probably is different for septicaemia and encephalitis in most cases. Compared to goats with encephalitis, the antibody production was higher in severely affected does with assumed septicaemia. Treatment with penicillin had an apparently good effect against both these conditions.

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SAMANDRAG

Kliniske undersøkingar i ein geiteflokk med utbrot av listeriose.

Størstedelen av 50 ikkje drektige geiter i ein flokk viste akutt sjukdom i 1—2 dagar, etter brått vèromslag og endring i fôring og oppstalling. Hjå 17 av geitene forverra sjukdomen seg, med symptom på septikemi i opptil 5 veker. Desse geitene var sløve og hadde feber og nedsatt fôropptak og mjølkeproduksjon, men berre 1 dauda. 3 veker etter første sjukdomsteikn viste 7 andre dyr typiske symptom på listeriaencephalitt. Også av desse dauda 1 geit, medan dei andre kom seg innan 2 veker. Penicillinbehandling syntest å ha god effekt ved båe utbrota.

Geitene med symptom på septikemi hadde signifikant høgre antistofftiter mot Listeria monocytogenes enn både dyra med encephalitt og dei andre. Serologisk undersøking synest difor å kunne brukast til å stille diagnosen listeria-septikemi hjå geit.

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