Massive *Cryptosporidium parvum* Infection Associated with an Outbreak of Diarrhoea in Neonatal Goat Kids

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Abstract: There was a massive outbreak of watery diarrhoea in kids aged 5 to 15 days in a goat herd in Gönen district of Isparta province in Turkey in May 2003. The presence of *Cryptosporidium parvum* was investigated in the faeces of 130 diarrhoeic kids and 3 dead kids in this herd. All of the faecal samples were also examined for rotavirus and coronavirus. *C. parvum* were detected in all of the faecal specimens. Coronavirus and rotavirus were not detected in any of the animals.

Key Words: Goat, diarrhoea, Cyptosporidium parvum, rotavirus, coronavirus

Neonatal Dönemde Şiddetli İshal Semptomu Gösteren Oğlaklarda Yoğun Cryptosporidium parvum Enfeksiyonu

Özet: Mayıs 2003'de İsparta'nın Gönen ilçesinde bir keçi sürüsünde 5-15 günlük oğlaklarda şiddetli ishale bağlı olarak ölümlerin görüldüğü bildirilmiştir. Üç ölü oğlak ile 130 ishalli oğlağın dışkıları parazitolojik yönden muayene edilmiş ve incelenen bütün örneklerde yoğun *Cryptosporidium parvum* ookisti tespit edilmiştir. Dışkı örnekleri ayrıca rotavirus ve coronavirus varlığı yönünden de incelenmiş ancak hiç birinde bu viral etkenlere rastlanmamıştır.

Anahtar Sözcükler: Oğlak, ishal, Cyptosporidium parvum, rotavirus, coronavirus

Introduction

Infectious diarrhoea in neonatal animals is one of the most common and economically devastating conditions encountered in the animal agriculture industry. The agents causing neonatal diarrhoea in domestic ruminants include bacteria such as Escherichia coli, viruses such as rotavirus and coronavirus and. importantly. Cryptosporidium parvum as a parasitic protozoon in many animal species and in humans, which causes the disease known as cryptosporidiosis (1,2). The first documented case of goat cryptosporidiosis was reported in 1981 (3). Since then, C. parvum has been studied in both diarrhoea outbreaks and in randomly selected farms. C. parvum is usually most prevalent in kids between 5 and 10 days of age (2,4-8). The most evident symptoms of cryptosporidiosis in neonate small ruminants are apathy, depression, anorexia, abdominal pain, and mainly diarrhoea accompanied by the shedding of a large number of oocysts (4,6,9,10). In outbreaks of diarrhoea, morbidity and mortality can be very high in goat kids less than 2 weeks old. Mortality increases when the disease is associated with concurrent infections. In addition to its economic importance, infection due to *C. parvum* also constitutes a major public health issue (1,11).

Some reports have described Cryptosporidium infection in field cases of neonatal goat diarrhoea in Turkey (12,13). Cryptosporidiosis in diarrhoeic goat kids

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was first detected by microscopic and transmission electron microscopic examination in the vicinity of Ankara and Denizli in Turkey (12). Since then, in Turkey only one study has been carried out to determine the prevalence of Cryptosporidium infection in diarrhoeic and nondiarrhoeic goats using carbol-fuchsin staining (13). However, no studies involving the outbreak cases of cryptosporidiosis in ruminants have been reported. This study reports a massive outbreak of cryptosporidiosis among kids aged 5 to 15 days. Although rotavirus and coronavirus have been recovered from many animal species, little is known about the epidemiology, characteristics and natural history of rotavirus and coronaviruses in goats (14). In addition, the present study is the first researching rotavirus and coronavirus in goats in Turkey.

Materials and Methods

In May 2003, there was a massive outbreak of watery diarrhoea in 5-15-day-old kids in a goat herd in Gönen district of Isparta province in Turkey. Seventy kids in the herd had died due to diarrhoea. All the affected animals were dehydrated, anorectic and prostrated. This study was conducted to investigate the causes of death in the kids due to diarrhoea. The faeces of 3 dead and 130 diarrhoeic kids were examined for parasitic and viral infections.

Faecal samples were directly collected from the rectum in sterile plastic bottles. For the detection of Cryptosporidium oocysts, fresh smears were made from non-concentrated faecal samples on glass slides and oocysts were detected microscopically using a modified Ziehl-Neelsen's acid-fast method (15). Moreover, coproantigens of *C. parvum* were also detected by a commercial ELISA kit (Bio-X Diagnostics, Belgium).

The intensity of infection was assessed semiquantitatively by counting the number of oocysts in 10 randomly selected microscope fields at 100X magnification. The categories established were negative (absence of oocysts), slight (1–5 oocysts), moderate (5–50 oocysts), and severe (>50 oocysts) (16).

All the faecal samples were tested for the presence of rotavirus and coronavirus by a commercial ELISA kit (Bio-X Diagnostics, Belgium). The ELISA test was performed according to the manufacturer's instructions. Briefly faeces samples were distributed in the wells of microtitration plates sensitised by specific antibodies for the rotavirus or coronavirus. The captured virus was revealed with a peroxidase labelled anti-virus specific monoclonal antibody conjugate. The plates were read by ELISA reader using a 450 nm filter (Anthos II, Austria).

Results

An acute natural outbreak of cryptosporidiosis on a goat farm in Gönen district of Isparta province in Turkey resulted in the death of 70 kids ranging in age from 5 to 15 days. They had been treated with antibiotics (Lincomicin + Spectinomicin), together with intravenous fluid therapy. They had not responded to this treatment and had died. All of the 130 diarrhoeic kids had acute diarrhoea with pasty and yellowish faeces. The kids were apathetic and dehydrated. The faecal samples of 3 dead and 130 diarrhoeic kids were positive for the oocysts and coproantigens of *C. parvum* by MZN staining and ELISA. The high number of oocysts in microscopic examinations indicated that the intensity of infection was severe. Five of the 20 randomly selected adult goats had low oocyst counts.

No coronavirus or rotavirus was detected using bovine coronavirus and rotavirus ELISA.

Discussion

Cryptosporidiosis is a common disease among neonatal ruminants and is most commonly observed in calves. The disease has not been reported as frequently in small ruminants. *C. parvum* has been recognised as a cause of outbreaks of diarrhoea in goat kids in several countries (4,5,7,8). This is the first outbreak of goat cryptosporidiosis detected in Turkey. In the present study, all of the diarrhoeic goat kids were 5 to 15 days old, and the affected animals were dehydrated, anorectic and prostrated, and were shedding the oocysts in great numbers in their faeces. Cryptosporidium infection in these goat kids resembles the clinical outbreaks of diarrhoea with cryptosporidiosis in goats that have been reported in Brazil (7) and the Sultanate of Oman (8) and observations in other countries (4,5,17).

The aetiologic diagnosis of acute infectious diarrhoea is not determined for a large percentage of cases of neonatal diarrhoeas. *C. parvum* is commonly associated with diarrhoea in neonatal ruminants. Enteric cryptosporidiosis is characterised by diarrhoea and high morbidity; when the disease is characterised by high mortality, concurrent bacterial and viral infections are common. Among the bacterial causes of diarrhoea in neonatal animals, E. coli and Salmonella spp. are the most common and economically important. Rotaviruses and coronaviruses are by far the most commonly identified viral causes of diarrhoea in neonatal food animals (18). A recent study showed that the most frequent aetiologic agent involved in outbreaks of diarrhoea in goat kids was C. parvum, followed by E. coli, rotavirus, Clostridium perfringens and Salmonella spp. (6,19,20), whereas few authors (21,22) have reported the detection of coronavirus in faeces from diarrhoeic kids. In the present study, no coronavirus and rotavirus was detected using bovine coronavirus and rotavirus ELISA. On the bacterial culturing, E. coli was isolated in the faeces of 3 dead kids and of 5 of the randomly selected 20 diarrhoeic kids, but the strain detection of *E. coli* was not possible. However, the high mortality rate of the kids in this goat herd indicates that the *E. coli* strain may be pathogenic. This outbreak demonstrates that C. parvum is probably involved as the primary pathogen associated with

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naturally occurring outbreak of diarrhoea in 5-15-day-old kids and that cryptosporidiosis causes higher morbidity and mortality together with bacterial enteritis.

The maternal stool is the major origin of Cryptosporidium infection in newborns. Infection risk is greater in newborns fed insufficient colostrum and born premature, because their immune systems are not mature enough to avoid infections. Adult animals can act as asymptomatic carriers shedding small numbers of oocysts into the environment, which was shown to increase in number in the perinatal period and contribute to maintaining the infection between kidding periods. Most animals born in this environment become infected in the first few weeks of life (23,24). In the present study, the faeces of 20 randomly selected adult goats were examined for *C. parvum* oocysts by MZN staining, and in 5 of them low counts of C. parvum oocysts were detected. The results of the current study demonstrate that subclinically infected adult goats, insufficient colostrum intake in kids, and contamination of the kidding area may be associated with Cryptosporidium infection status.

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