

Case report

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ACUTE BOVINE MASTITIS CAUSED BY *KLEBSIELLA PNEUMONIAE* – CASE REPORT

Milan Ninković^{1*}, Jadranka Žutić¹, Jovan Bojkovski², Sveta Arsić²,
Dimitrije Glišić¹, Zorana Zurovac Sapundžić¹, Nemanja Zdravković¹

¹ Scientific Institute of Veterinary Medicine of Serbia, Department
for Healthcare, Belgrade, Republic of Serbia

² University of Belgrade, Faculty of Veterinary medicine, Department
of Ruminants and Swine disease, Belgrade, Republic of Serbia

Abstract

This case report describes the acute mastitis with signs of endotoxemia in the 32-month-old Simmental cow, antimicrobial susceptibility of *Klebsiella pneumoniae* isolated from a milk sample, and demonstrates the effectiveness of the applied therapy. Case presentation included the cow-lying syndrome together with the presence of clinical signs of endotoxemia accompanied by milk discoloration and “clots” formation. Bacteriology finding of the *K. pneumoniae* and antibiogram pointed that the isolate was sensitive to ceftriaxone, and trimethoprim/ sulfamethoxazole, while resistant to amoxicillin, gentamicin, ampicillin, ceftiofur, and cephalexin. The selection of treatment options came from the availability of an adequate route of administration, urgency for treatment, and lack of time for assessment of other body functions. The therapy by ceftriaxone at an intravenous dose rate of 8mg/kg with adequate supportive therapy NSAID, vitamin C, and correction dehydration showed a successful effect in life-saving procedures. The outcome of *K. pneumoniae* mastitis with endotoxemia did not affect the outcome of pregnancy.

Key words: ceftriaxone, endotoxemia, *Klebsiella pneumoniae*, mastitis

^{1*} Corresponding Author: milan.ninkovic1992@gmail.com

AKUTNI GOVEĐI MASTITIS IZAZVAN BAKTERIJOM *KLEBSIELLA PNEUMONIAE* – PRIKAZ SLUČAJA

Milan Ninković^{1*}, Jadranka Žutić¹, Jovan Bojkovski², Sveta Arsić²,
Dimitrije Glišić¹, Zorana Zurovac Sapundžić¹, Nemanja Zdravković¹

¹ Naučni institut za veterinarstvo Srbije, Odeljenje za
zdravstvenu zaštitu, Beograd, Republika Srbija

² Univerzitet u Beogradu, Fakultet veterinarske medicine,
Katedra za bolesti papkara, Beograd, Republika Srbija

Kratak sadržaj

Ovaj prikaz slučaja opisuje slučaj akutnog mastitisa sa znacima endotoksemije kod krave simentalske rase, stare 32 meseca, antimikrobnu osetljivost *K. pneumoniae* izolovanu iz uzorka mleka i prikaz primenjene terapije. Prikaz slučaja je uključivao prisustvo sindroma ležeće krave sa prisustvom kliničkih znakova endotoksemije praćene promenom boje mleka i prisutnim ugrušcima mleka. Bakteriološki nalaz *K. pneumoniae* i antibiogram pokazali su da je izolat osetljiv na ceftriakson i trimetoprim/sulfametoksazol, dok je otporan na amoksicilin, gentamicin, ampicilin, ceftiofur i cefaleksin. Izbor opcije za lečenje je proizašao iz dostupnosti, hitnosti lečenja i nedostatka vremena za procenu ostalih telesnih funkcija. Primena ceftriaksona u intravenskoj dozi od 8 mg/kg uz adekvatnu suportivnu terapiju NSAIL, vitamin C i korekciju dehidracije pokazala je uspešan efekat u lečenju mastitisa. Ishod mastitisa *K. pneumoniae* nije uticao na ishod graviditeta.

Ključne reči: ceftriakson, endotoksemija, *Klebsiella pneumoniae*, mastitis

CASE PRESENTATION

Forty percent of all clinical mastitis are caused by coliform bacteria *E. coli*, *Klebsiella spp.*, *Enterobacter spp.* (Schukken et al., 2012) with significant losses in milk production and a high economic cost of treatment (Munoz et al., 2007). The primary source of *K. pneumoniae* in dairy farms was faecal discharges, as

well as the presence of organic materials, such as wheat straw, barley straw, sand, and wood (Munoz et al., 2006), while the environmental factors, unhygienic conditions, and organic bedding are associated with the appearance of mastitis (Ngu Ngwa et al., 2018), where the opportunistic pathogens like *K. pneumoniae* additionally grow on organic and inorganic bedding materials (Zdanowicz et al., 2004). A 32-month-old pregnant Simmental cow with a daily milk yield of 25 kg (before the onset of mastitis) milking thrice per day, two months pregnant (pregnancy was confirmed by ultrasound a couple of days before mastitis onset) was presented with swollen, painful, and stiff udder. The California mastitis test (CMT) score was (+++), i.e., a severe reaction to the CMT. The milk was a watery grey colour with clots. The cow had a rectal temperature of 39.6°C and displayed the lying-cow syndrome, similar to milk fever with preserved ruminal contraction. The daily milk yield drop was drastic, to only 1kg per 24 h. Diagnosis of mastitis was based on general clinical examination, physical examination of the udder, physical changes in the milk, the California mastitis test (CMT) score, and later findings of microbiological analysis of the milk. Hematological and biochemical blood analyses were also performed. Each mammary gland quarter sample was collected aseptically and transported to the laboratory under refrigeration (4–8°C).

After collection, the samples were placed in ice-cooled hand freezers and transported to the laboratory within 4 hours. The microbiology examination was conducted on routine methods (Markey et al., 2013). In brief: the milk samples were put on Columbia blood agar, MacConkey agar, and Sabouraud dextrose agar (all Torlak, Serbia) and kept at 37°C under aerobic conditions for 24 h, while Sabouraud agar was reincubated under aerobic conditions at 25°C up to 72 h. The primary isolate was stained by Gram procedure (Biomereux, France), followed by catalase and oxidase reaction (HiMedia, India). Presumptive diagnosis based on growth characteristics and said reactions were subjected to further biochemical testing using commercial kits for enteric non-fermenter bacteria (BBL Crystal, E/N, ID kit, Becton, Dickinson and company, USA). There was no growth on Sabouraud dextrose agar.

The antibiograms were done following the ISO 20776-1:2006 standard by disk diffusion method using antimicrobial test disks (BBL, USA) to determine susceptibility toward and other antibacterials and antibiotics: amoxicillin with clavulanic acid (20/10 µg), ampicillin (5 µg), cephalexin (30 µg), ceftriaxone (30 µg), gentamicin (10 µg), trimethoprim+sulfamethoxazole (23.75/1.25 µg), enrofloxacin (5 µg). Interpretative zones were estimated following the standard in antimicrobial testing (CLSI).

DISCUSSION

A bacteriology test with antibiogram revealed *K. pneumoniae* mastitis in one quarter with two antimicrobial options: ceftriaxone and trimethoprim/sulfamethoxazole. The treatment option was influenced by the availability of an adequate route of administration, treatment urgency requirement, confirmed pregnancy, and lack of time for assessment of other parameters (such as renal function, hypoglycaemia, or folate deficiency) on the animal with presumptive endotoxemia. The therapy by ceftriaxone at an intravenous dose rate of 8mg/kg with adequate supportive therapy NSAID, vitamin C, and body fluid status correction was started immediately. The clinical appearance of the udder with systemic health deterioration with the effects of endotoxemia indicated the appearance of coliform mastitis. Previous records showed a couple of *Escherichia coli* mastitis isolates on the farm, all susceptible to ceftriaxone. In addition to the presence of cow-lying syndrome, the biochemical analysis of blood showed that calcium and magnesium were at the limit values of 2.2 mmol/L and 0.9 mmol/L, respectively. The cow was treated with ceftriaxone daily during five days, and supportively with a large volume of Hartman's solution (100ml/kg body weight per 24h), glucose 10% (1000 ml per 24h), flunixin meglumine (2.2 mg/kg b.wt intravenously once daily during five days) and vitamin C (4 mg/kg b.wt intravenously once daily during three days). Three days after treatment, the CMT reaction showed only mild reaction to CMT. Five days after therapy, the CMT reaction was negative, and no clinical signs were present.

The previous history of mastitis in the herd was unrelated to the incidence of *K. pneumoniae*. The set of new wheat straw as bedding material a few days before the onset of mastitis was a potential way of introducing *K. pneumoniae* to the farm, adequate to previous findings that the presence of organic matter as bedding may be the cause of mastitis (Munoz et al., 2006).

The antimicrobial susceptibility of *this* bacteria varies between different studies due to a large number of strains (Osman et al., 2014). The present case showed the antibiogram to be sensitive to ceftriaxone and trimethoprim/sulfamethoxazole; however, other studies (Enferad and Mahdavi, 2021) established that most strains of *K. pneumoniae* were 75% and 55% resistant to ceftriaxone and oxytetracycline, respectively. The duration of treatment (5 days) and persistence of clinical signs in our case followed previously reported study (Hoe and Ruegg, 2005). Supportive treatment of fluids and electrolytes using NSAID (flunixin meglumine) reduced the clinical signs of endotoxemia and accelerated recovery. We consider supportive therapy with vitamin C to reduce the duration of mastitis because ascorbic acid affects the recovery after clini-

cal mastitis (Naresh et al., 2002). After 15 days from the completed treatment, daily milk production had increased to 20 L; at the same point, pregnancy control was performed and reperformed two months after treatment, confirming pregnancy while assuring the success of the applied therapy.

In the present case, ceftriaxone administration was justified because of the narrowed choice of effective antibiotics available. After 15 days, the cow regained 80% of its daily milk production. We conclude that correction of dehydration, executive elimination of *K. pneumoniae* systemic endotoxins, and ceftriaxone as the antibiotic of choice based on the antibiogram, assisted by vitamin C supportive therapy, resulted in a quick recovery. The outcome of mastitis in this cow or treatment option did not affect the pregnancy outcome. Educating the farmers about the potential source of infection is essential to prevent new episodes of mastitis.

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Author's contribution:

MN and JŽ made contributions to conception, design of the study, and collected of samples, NZ and DG conducted the microbiological analysis, SA and ZZS involved in drafting the manuscript and analysed data, JB revised the manuscript critically and together with MN prepared the final draft of the manuscript.

Competing interest

The authors declare that they have no conflict of interest.

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