

# A New Aspect in Neonatal Calf Diarrhea: Presence of *Escherichia coli* CS31A at Unexpected Ratio

## (Yeni Doğan Buzağı İshallerine Yeni Bir Bakış Açısı: Beklenmeyen Oranda *Escherichia coli* CS31A Varlığı)

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### Dear Editor,

Neonatal calf diarrhea (NCD), defined as diarrhea occurring during the first month of the life, is the most common disease and the cause of death of calves worldwide in this period of life. *Escherichia coli*, *Bovine rotavirus*, *Bovine coronavirus*, *Salmonella* spp., *Cryptosporidium parvum* are common etiological agents of NCD. These pathogens can cause disease as mono or co-infections and also may be found in clinically healthy animals<sup>[1]</sup>.

*Escherichia coli* is primarily known as a commensal colonizing bacterium in the gastrointestinal tract of calves early in life, but some strains being responsible for diarrhea, which can be severe in neonatal calves. Different *E. coli* pathotypes can settle in different parts of the gastrointestinal tract in animals, and the most predominant pathotype was reported as K99<sup>[1,2]</sup>. Binding to host cells is necessary for the occurrence of infection, and provided through factors such as fimbria and fimbrial adhesins<sup>[2]</sup>.

The fimbria, or adhesins of *E. coli* serotypes, do not give an idea about the virulence of the agent, but they are the tools used in the identification of the agent. One of the most commonly studied fimbrial antigens in vaccines is the plasmid-encoded F5 (K99) fimbria, which mediates adhesion to the ileum and constitutes the first steps of colonization in the bovine intestine. Therefore, knowledge on fimbriae and adhesins are essential for the development of efficient vaccination<sup>[3]</sup>.

CS31A is a plasmid encoded fimbrial adhesin of *E. coli*, and its antigen was first reported as a capsule-like surface protein<sup>[4]</sup>, CS31A was isolated from septicemic and diarrhetic neonatal calves in many countries and is currently considered as a major component of neonatal colibacillosis<sup>[5,6]</sup>. CS31A protein is coded by conjugative R plasmid associated with multiple-antibiotic resistance and aerobactin system. In one study performed on 391 calves from 14 farms, 378 were *E. coli* positive in which 211 samples were positive for CS31A (55.8%). More striking is that, 88.1% of CS31A isolates were multi-resistant for three or more antimicrobial groups and 15.6% of these samples were resistant to all six antimicrobial groups<sup>[7]</sup>. Prevalence of CS31A in neonatal calves in Turkey is not known well. Etiological diagnoses were performed by rapid kit on 51 samples and *E. coli* K99 + *E. coli* CSA31A was detected in 1.96% of the calves in Van<sup>[8]</sup>.

We have collected 33 neonatal calf fecal samples (average age of 8.06 days), which were presented to our hospital with the complaint of diarrhea, by taking informed consent forms from the owner. DNA extractions from feces was performed by DNA isolation kit (Pure Link® Genomic DNA mini kit, Invitrogen, Thermo Fisher) according to the manufacturer's instructions. Isolated DNAs were served as template for determining CS31A gene and K99 gene of isolates. The CS31A and F5 (K99) genes were analyzed by PCR as described before<sup>[9]</sup>. The bands of 403 and 314 bp were evaluated as positive for CS31A and F5 genes.

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As a result of the etiological data obtained in our observation, in which we evaluated the effectiveness of the cristobalite molecule in neonatal calf diarrhea, prevalence of CS31A was 51.51% and K99 was 9.09% as compared to those of the previous reports <sup>[1,4]</sup>. Lesser prevalence of K99 antigen in this group may be explained by extensive vaccination practices of pregnant cows and hyperimmune serums administered to the newborn calves in Turkey, but the significantly higher prevalence of CS31A must be considered carefully in the etiology of colibacillosis in the neonates.

Understanding the etiology of NCD and determining the prevalence of infectious agents in calf diarrhea is necessary for optimizing prophylactics, passive immunity, and vaccination protocols <sup>[10]</sup>. Currently, there is no vaccine providing protection for CS31A.

We think that, further investigation for the prevalence of *E. coli* pathotypes in field is essential and evaluation of the role of CS31A adhesin in pathogenesis in neonatal calf diarrhea, and investigation of CS31A antigen as well as F5 fimbrial antigen in vaccination studies are of vital importance in terms of prophylaxis.

#### AUTHOR CONTRIBUTIONS

Ü. Özcan, B. U. Sayılkan, and E. Küllük collected the faecal samples. The microbiological analyzes were performed by M. G. Sezener, V. E. Ergüden, and Ş. Yaman.

#### CONFLICT OF INTEREST

The authors report no conflicts of interest. The authors alone are responsible for the content and writing of paper.

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