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P286 - Bovine coronavirus N gene detection and characterization in cattle: a preliminary study

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Bovine coronavirus (BCoV) belongs to the genus *Betacoronavirus* and subgenus *Embecovirus* in the family *Coronaviridae* and causes respiratory and enteric disease in cattle. The genome is a single-stranded, positive-sense RNA of approximately 31 kb. Although most molecular phylogenetic analyses are based on the S gene, genetic variability has been found also in HE and in N genes. This preliminary work is part of a larger study aimed at analysing the genetic variability of BCoV detected in cattle recently acquired from different European countries.

Nasal swab samples were collected from 66 cattle recently acquired from Northern Italy, France, Hungary, Romania, and Poland by 15 different farms. PCR methods were used to amplify and sequence the N gene [1]. Sequences were aligned with N gene sequences available in databases and the phylogenetic tree was inferred using the Maximum Likelihood method with Kimura 2-parameter as nucleotide substitution model. The rate variation among sites was modelled with a gamma distribution. The main sites of mutation were at positions T30041C, C30059T, C30071A, T30251C and G30257T in comparison with the sequence of the Mebus isolate (GenBank ID U00735.2). In addition, one sample showed synonymous mutations at positions C30056T and A30122T. The only non-synonymous mutation A30192C, corresponding to the substitution of an isoleucine with a leucine, was found in one strain from Northern Italy. Most samples showed the highest percentage of identity with the strain BCoV/FRA/EPI/Caen/2012/07 (GenBank ID KT318089.1) isolated in France in 2012 from cattle. A few samples showed the highest percentage of identity with the strain bcovizsm (GenBank ID MW074864.1) isolated in Southern Italy in 2020 from cattle [2]. Interestingly, one sample collected from cattle reared in Northern Italy showed the highest identity with bovine-like coronavirus detected in water buffalo in Southern Italy in 2007 (GenBank ID EU019216.1) [3] and then in dromedary in Morocco in 2016 (GenBank IDs MN514972.1 and MN514975.1) [4]. N protein plays an important role during virion assembly and in enhancing the efficiency of subgenomic viral RNA transcription as well as viral replication. The sequences obtained in this study showed some synonymous mutations rarely or not detected previously. The N gene is often the target of PCR diagnostic protocols thus monitoring of sequences of this gene is important to evaluate the conservation among sequences of viruses from different geographic regions. Further investigations will be aimed at sequencing other relevant and more variable genes. Particular attention will be paid to the sample showing high identity with bovine-like coronavirus previously detected in host different than cattle [3,4].

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[4] So et al. Diversity of Dromedary Camel Coronavirus HKU23 in African Camels Revealed Multiple Recombination Events among Closely Related Betacoronaviruses of the Subgenus Embecovirus, J Virol, 93(23):e01236-19, 2019.