

GENETIC HETEROGENEITY OF BOVINE VIRAL DIARRHOEA VIRUS (BVDV) ISOLATES FROM ITALY: IDENTIFICATION OF NEW BVDV-1 GENOTYPES

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Introduction

Sequence analysis of 5'-UTR can distinguish BVDV-1 and BVDV-2 genotype, moreover the same region can subdivide BVDV-1 into at least eleven genetic groups. Cluster analysis of combined nucleotide sequences from 5'-UTR and Npro region, lead to a highest statistical support by bootstrap analysis. In this study we analysed a broad range of BVDV Italian isolates by genetic typing of 5'-UTR, supported by selected comparison within the Npro coding region.

Materials and Methods

Eighty-eight BVDV strains have been collected mainly during a period of 8 years (2000-2007) from 12 Italian regions from cattle (n=81), buffalo (n=3) and sheep (n=4). The conserved 5'-UTR (all viruses) and the Npro (18 selected viruses) regions were sequenced and analysed.

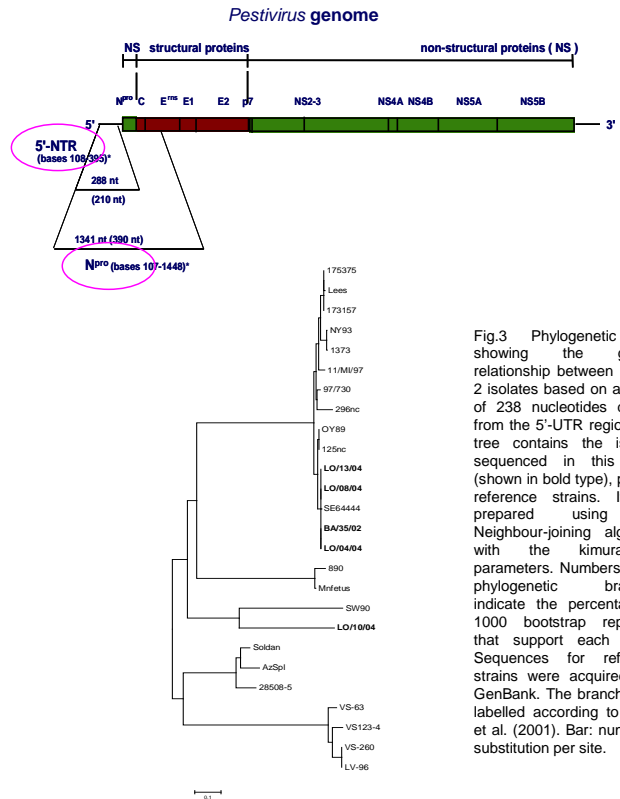


Fig.3 Phylogenetic tree showing the genetic relationship between BVDV-2 isolates based on analysis of 238 nucleotides derived from the 5'-UTR region. The tree contains the isolates sequenced in this study (shown in bold type), plus 18 reference strains. It was prepared using the Neighbour-joining algorithm with the kimura 2-parameters. Numbers at the phylogenetic branches indicate the percentage of 1000 bootstrap replicates that support each group. Sequences for reference strains were acquired from GenBank. The branches are labelled according to Vilcek et al. (2001). Bar: number of substitution per site.

Tab.1. Percentage of similarity of pair wise distances within and between genetic group.

a	b	d	E	f	g	H	
77.82-99.15	73.32-90.75	76.56-94.95	74.00-91.17	74.05-88.23	67.64-87.81	75.31-87.39	A
	75.46-100	71.80-97.00	70.50-89.00	73.52-88.23	67.38-88.65	73.52-87.39	B
		84.03-97.05	72.68-87.80	73.52-88.23	69.74-87.81	76.47-87.81	D
			76.10-100	75.63-90.33	68.48-90.33	74.36-90.33	E
				84.03-98.31	70.50-92.85	77.31-92.01	F
					77.73-100	70.58-98.31	G
						85.71-100	h

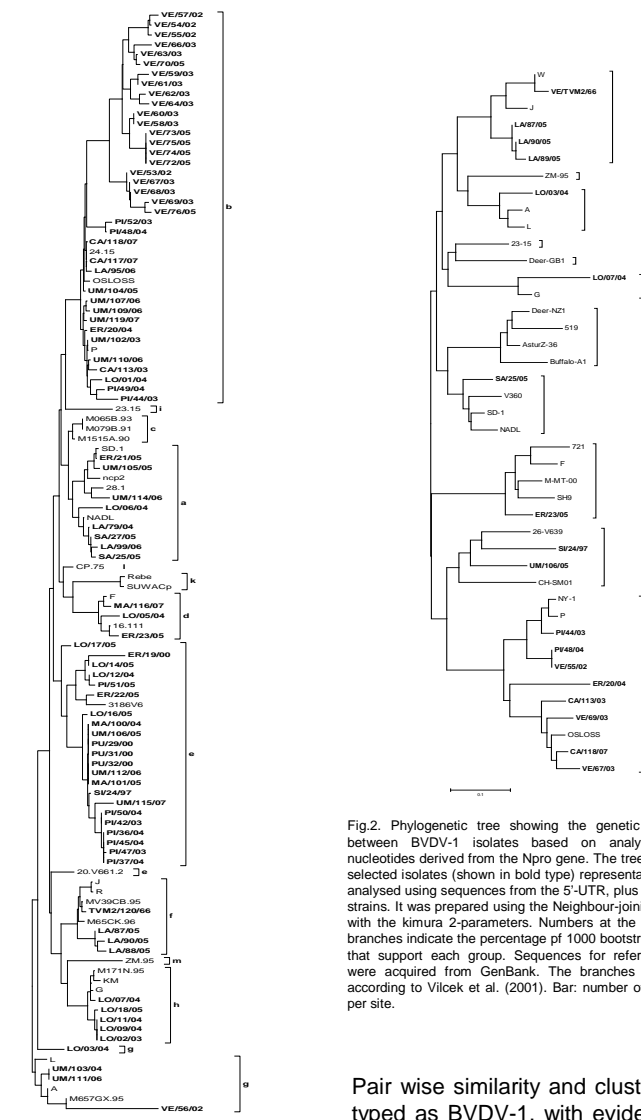


Fig.2. Phylogenetic tree showing the genetic relationship between BVDV-1 isolates based on analysis of 390 nucleotides derived from the Npro gene. The tree contains 18 selected isolates (shown in bold type) representative of those analysed using sequences from the 5'-UTR, plus 28 reference strains. It was prepared using the Neighbour-joining algorithm with the kimura 2-parameters. Numbers at the phylogenetic branches indicate the percentage of 1000 bootstrap replicates that support each group. Sequences for reference strains were acquired from GenBank. The branches are labelled according to Vilcek et al. (2001). Bar: number of substitution per site.

Fig.1. Phylogenetic tree showing the genetic relationship between BVDV-1 isolates based on analysis of 238 nucleotides derived from the 5'-UTR region. The tree contains all the isolates sequenced in this study (shown in bold type), plus 28 reference strains. It was prepared using the Neighbour-joining algorithm with the kimura 2-parameters. Numbers at the phylogenetic branches indicate the percentage of 1000 bootstrap replicates that support each group. Sequences for reference strains were acquired from GenBank. The branches are labelled according to Vilcek et al. (2001). Bar: number of substitution per site.

Discussion

Pair wise similarity and cluster analysis provided a clear-cut assignation to 7 distinct genotypes of 83 isolates typed as BVDV-1, with evidence for circulation of BVDV-1a and BVDV-1g additional genotypes, never shown before in Italy. Five field viruses were typed as BVDV-2 and were clustered into the genotypes BVDV-2a and BVDV-2b, this latter being reported for the first time in Italy. In summary, the results presented in this work revealed a high BVDV genetic heterogeneity in Italy. This evokes a question as to whether BVD vaccines which have been historically contained only BVDV-1a and BVDV-1b genotypes, can protect against infection with all of the highly diverse BVDV-1 isolates.