

# Genetic and Antigenic Characterization of Bovine Viral Diarrhea Field Isolates

**B.M. Hause, MS; R.F. Bey, PhD; D.L. Stine, DVM, PhD; R.R. Simonson, PhD**  
*Newport Laboratories, Worthington, MN 56187*

## Introduction

Bovine viral diarrhea virus (BVDV) is commonly found in cattle and causes significant economic losses. Despite long term and widespread use of commercial modified live vaccines, BVD remains problematic. BVDV are classified into two genotypes, BVDV-1 and BVDV-2, and are further subdivided within the genotype by sequence analysis. Most genetic analysis is performed on the 5'-untranslated region of the genome as it is relatively conserved, however, this region does not code for an antigen. The objective of this study was to characterize recent BVDV field isolates using genetic analysis of the E2 gene in combination with antigenic methods using antisera generated against multiple isolates in a high throughput serum neutralization assay.

## Materials and Methods

BVDV was isolated from samples submitted to Newport Laboratories. Isolates were genetically characterized by partial E2 gene sequencing and DNA sequences were translated into amino acids and aligned by pairwise cluster analysis using commercial software. Hyperimmune antisera were generated in swine for both field and reference isolates of BVD1a (x2), BVD1b (x4), and BVDV2 (x4). Thirty-eight recent field isolates were analyzed using a high throughput serum neutralization assay incorporating normalized hyperimmune antisera run at a single dilution to serotype isolates.

## Results

Sequence analysis of the E2 gene classified 38 field isolates as BVD1a (6, 15.8%), BVD1b (27, 71.1%), and BVDV2 (5, 13.2%). Amino acid similarities between isolates within subtypes were 80-100%. Isolates were next antigenically categorized using reference antisera in a high throughput serum neutralization assay. Isolates of BVDV1a, BVDV1b, and BVDV2 were neutralized by homologous cluster antisera 100%, 92.5%, and 80% of the time, respectively. Cross reaction between BVDV1a, BVDV1b, and BVDV2 isolates and heterologous antisera was minimal with isolates neutralized 4.2%, 7.4%, and 10% of the time, respectively.

## Significance

Genetic analysis of the E2 gene indicated that BVDV1b was the most common subtype found in our sample set. This is in agreement with prevalence studies performed by others using 5'UTR analysis. Despite possessing up to 20% amino acid differences in the E2 gene within a subtype, BVDV field isolates were neutralized by homologous subtype antisera with high frequency. Conversely, isolates were poorly neutralized by heterologous subtype antisera. These results suggest that antibody cross reactivity between BVDV subtypes is minimal. Veterinarians should consider using vaccines incorporating BVDV1a, BVDV1b, and BVDV2 isolates to ensure coverage of circulating strains.