RecombiVirus™ Bovine Viral Diarrhea Virus (BVDV1-3) Vaccines ELISA and DIVA Tests

RecombiVirus™ series of ELISA kits are 2nd generation ELISA and quantitative ELISA kits to detect BVDV1-3 virus specific antibodies using recombinant and highly purified viral antigens. BVDV3 ELISA detects antibodies to the latest strain (hobi). Advantages of **RecombiVirus™** ELISA kits:

- **Recombinant viral antigens**: Safe, no risk of contamination
- **Qualitative or Quantitative**: Use single Positive antibody calibrator at 100 U/ml for +ve or –ve samples (Qualitative) or use full standard curve to measure antibody concentration in animals (Quantitative).
- **Rapid tests:** assay time ~105 mins
- **Sensitive**: higher sensitivity allows sample dilution of 1::100 or more. Less background.
- **Define Sensitivity**: Antibody detection to ~1.0 ng/ml.
- **Convenient**: Room temp incubations, all reagents in stable solution format; strips of 8-wells for maximum usage
- **Stable**: 1 year shelf life
- **BVDVE2 or BVDV Erns antibody** ELISA kits can be used to assess the antibody status of vaccinated animals or normal animals. **BVDV Erns antibody** ELISA kit may serve to establish efficacy of novel designer vaccines that enable **DIVA test** (differentiation of infected from vaccinated animals) and raise antibodies to BVDV Erns protein**.** ELISA kits for rabbits and mice (not a natural host for BVDV disease) are being made available to test various vaccines and adjuvants. Note: The BVDV ELISA kits are neither intended nor approved for disease diagnosis, viral serotyping, prevention or cure of the disease in animals or humans.

Assay Procedure: Arrange required number of strips on the plate.

Step 1. Add **100 µl of pre-diluted anti-BVDV IgG standards** (0, 3, 10, 30, 100 U/ml) and **100 ul samples** (diluted 1:100 or higher) into respective wells. Mix gently and incubate at room temp for **60 mins** (25-28oC; no shaking necessary).

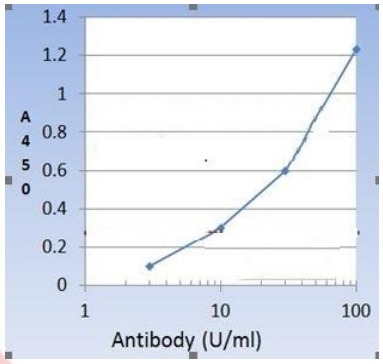
Step 2. Aspirate well contents and wash 3X with wash buffer. Add **100 ul of supplied antibody-HRP Conjugate** into all wells; mix gently and incubate at RT for **30 mins**.

Step 3. Aspirate or wash 5x with wash buffer. Tap plates over paper towels. Add **100 ul of TMB Substrate**. Mix gently and Incubate for 15 min at RT. Blue color develops in positive wells.

Step 4. Add **100 ul of stop solution** into each well and mix gently (blue color turns yellow). Measure yellow color at 450 nm.

Calculation of Results

Results can be expressed as simple –ve and +ve as compared to standards or BVDV antibody concn (U/ml) determined from standard curve.



List of Porcine circovirusELISA Kits available from ADI.

Product details, data sheets, and pricing available (http://4adi.com/commerce/catalog/spcategory.jsp?category_id=2802)

	Product Description	Rabbit	Mouse
*BVDV1-3 E2	Recombivirus™ **BVDV1-3 E**2 **IgG** ELISA kit, Quantitative, 96 tests or 5x96 tests	RV-500260-1 RV-500260-5	RV-500280-1 RV-500280-5
*BVDV1-3 Erns	Recombivirus™ **BVDV1-3 Erns IgG** ELISA kit, Quantitative, 96 tests or 5x96 tests (**DIVA**)	RV-500160-1 RV-500160-5	RV-500180-1 RV-500180-5

BVDV1-3 Antibody, Recombinant Proteins, and Controls

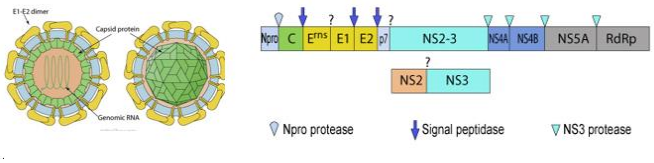
Catalog#	Product Description	Product Type
BVDE11-C	Recombinant BVDV1 Envelope glycoprotein 2 (BVDV1 E2) Protein control for western blot	Western Control
BVDE11-S	Anti-Bovine Viral Diarrhea Virus 1 Envelope glycoprotein 2 (BVDV1-E2) antiserum	Rabbit-Poly
BVDE15-R-10	Recombinant (E. coli) Bovine Viral Diarrhea Virus 1 Envelope glycoprotein 2 (BVDV1-E2, his tag)	Recom. protein
BVDE21-C	Recombinant Bovine Viral Diarrhea Virus1 Envelope Protein 2 (BVDV E2) control for western blot	Western Control
BVDE21-S	Anti-Bovine Viral Diarrhea Virus 2 Envelope glycoprotein 1 (BVDV2 E2) protein antiserum	Rabbit-poly
BVDE25-R-10	Recombinant (E. coli) Purified Bovine Viral Diarrhea Virus 2 E2 protein (BVDV2-E2, his tag)	Recom. protein
BVDE31-C	Recombinant (E. coli) Purified Bovine Viral Diarrhea Virus 3 E2 (BVDV3 E2) Protein control for	Western Control
BVDE31-S	Anti-Bovine Viral Diarrhea Virus 3 E2 (BVDV3-E2) protein antiserum	Rabbit-poly
BVDE31-S	Anti-Bovine Viral Diarrhea Virus 3 (BVDV3) E2 Protein antiserum	Rabbit-Poly
BVDE35-R-10	Recombinant (E. coli) Purified Bovine Viral Diarrhea Virus 3 E2 (BVDV3 E2) Protein (>95%)	Recom. protein
BVDE35-R-10	Recombinant (E. coli) Purified Bovine Viral Diarrhea Virus 3 E2 protein (BVDV3-E2, his tag) purified	Recom. protein
BVDR12-C	Recombinant Bovine Viral Diarrhea Virus 1 (BVDV1) Erns Protein control for western blot	Western Control
BVDR12-S	Anti-Bovine Viral Diarrhea Virus 1 Erns (BVDV1- Erns) Protein antiserum	Rabbit-Poly
BVDR16-R-10	Recombinant (E. coli) Purified Bovine Viral Diarrhea Virus 1 Erns Protein (BVDV1-Erns, his tag)	Recom. Protein
RV-500100-01N	Bovine Viral Diarrhea Virus1 E2 (BVDV1 E2) IgG negative control serum	Disease sera
RV-500100-02P	Bovine Viral Diarrhea Virus1 E2 (BVDV1 E2) IgG positive control serum	Disease sera
RV-500100-03N	Bovine Viral Diarrhea Virus 2 E2 (BVDV2 E2) IgG negative control serum	Disease sera
RV-500100-04P	Bovine Viral Diarrhea Virus 2 E2 (BVDV2 E2) IgG positive control serum	Disease sera
RV-500100-05N	Bovine Viral Diarrhea Virus 3 E2 (BVDV3 E2) IgG negative control serum	Disease sera
RV-500100-06P	Bovine Viral Diarrhea Virus 3 E2 (BVDV3 E2) IgG positive control serum	Disease sera

Bovine Viral Diarrhea Virus (BVDV1-3) Vaccine, Antibodies and ELISA Kits

Bovine viral diarrhea virus (BVDV) is the prototypic member of the genus pestivirus in the family Flaviviridae. The genus Pestivirus is composed of 4 important pathogens of livestock: **Bovine viral diarrhea virus 1 and 2 (BVDV-1 and BVDV-2**), **Classical swine fever virus (CSFV),** and **Border disease virus of sheep (BDV)**. Viruses in the genus Pestivirus infect mammals, including members of the family Bovidae (which includes, but is not limited to, **cattle, sheep, and goats**) and the family Suidae (which includes various species of **swine**). BVDV is widespread and one of the most economically important diseases of cattle. BVDV-associated diseases can range from clinically undetectable to severe. Infection with BVDV causes heavy economic losses for livestock industries worldwide due to abortions, production of weak calves, congenital deformities, respiratory infections, thrombocytopenia, persistent infections (PI), and mucosal disease. Fetal infections are an important manifestation of BVDV, particularly when susceptible pregnant heifers/cows develop a viremia after the initial acute infection. The economic impact of BVDV infections has lead a number of countries in Europe to start eradication or control programs, respectively.

BVDV has a world-wide distribution and is characterized by high prevalence and relatively low morbidity. In addition to **cows**, other artiodactyla (**sheep, goats, wild ruminants, pigs**) can be infected. Although those other animals do not get Mucosal Disease they may, among others, suffer from reduced fertility. Numerous investigations show differing Seroprevalence and incidence of Persistent infected (PI) animals. However, in areas where BVD is endemic they mainly range between 60 and 80% (Seroprevalence), or 0.5-2% (PI animals).

BVDV are classified as biotypes and genotypes. The biotypes are based on presence or absence of visual cytopathic effects in infected cell cultures: cytopathic (CP) or noncytopathic (NCP). The genotypes are based on nucleic acid differences in the viral genome based on PCR of specific region as well as genomic sequencing. There are also antigenic differences associated with the genotypes. The **predominant genotypes** in U.S. cattle are **BVDV1 and BVDV2**. However there are subtypes of BVDV1 represented by BVDV1a and BVDV1b. BVDV-1 and BVDV-2 contain 11 and 3 subtypes respectively, which have been demonstrated by analysis of the 5'UTR. **BVDV-1 is widely distributed world-wide** while **BVDV-2 is mainly found in North America**, although it has also been detected in various European countries. Both BVDV-1 and BVDV-2 can trigger serious disease, the majority (70-90 %) of the infections, however, are asymptomatic (i.e., differences in genetic structure do not account for virulence). Both genotypes occur as cytopathic as well as non-cytopathic biotypes. Non-cytopathic BVDVs of both genotypes can cause persistent infections.



A new putative pestivirus species, tentatively called "**HoBi-like**," "**BVDV-3**," or "atypical pestiviruses," was first identified in Europe in fetal bovine serum (FBS) imported from Brazil. It is estimated that 30% of FBS processed in South America, Europe, Australia, Canada, Mexico, and USA are contaminated with BVDV-3. Natural infection in cattle with HoBi-like viruses has being reported in Southeast Asia, Italy, and Brazil. BVDV-3 is genetically distant from BVDV-1 and BVDV-2, and diagnostics and vaccines that work well to detect and control the 2 species of BVDV are less efficacious for BVDV-3. The disease caused by these new viruses resembles clinical presentations historically associated with BVDV infection, including growth retardation, reduced milk production, respiratory disease, reduced reproductive performance, and increased mortality among young stock.

BVDV is a small, enveloped virus (40-60 nm) with a ssRNA(+V) **~12.5kb.** It encodes **N^{pro}, Capsid, E^{rns}, E1, E2 and p7,** which code for structural proteins, followed by **NS2-3, NS4A, NS4B, NS5A and NS5B** encoding non-structural proteins. Erns, E1 and E2 are all glycoproteins, with Erns possessing RNase activity involved in viral replication and pathogenesis. The **E2 structural protein** is the major target of virus-neutralizing antibodies.

Diagnosis of animals persistently infected with BVDV can be identified by virus isolation from whole blood or other tissues, ImmunoPerioxidase Monolayer Assay; IPMA, immunohistochemistry (IHC) staining of viral antigen in skin biopsies, enzyme-linked immunosorbent assay (ELISA) and PCR methods.

BVDV Vaccines: Two different types of BVD vaccines are available, modified live vaccines (MLV) and the killed vaccines. **MLVs contain attenuated strains of BVDV that replicates in the host to elicit an immune response**. Many vaccines contain BVDV Type 1 or 2 or both. Both vaccines will also help prevent transmission of BVD virus to the fetus and therefore prevent abortions and/or the birth of persistently infected calves. No vaccine, however, will give complete fetal protection.

Vaccine Type	Strains	Genotype/Biotype
MLV		
Express 5	Singer	1a CP
	296 2	CP
BoviShield 4	NADL	1a CP
Pyramid 4	Singer	1a CP
Reliant 4	NADL	1a CP
Frontier 4 plus	C24V	1a CP
	296	2 CP
Titanium 5	C24V	1a CP
	296	2 CP
Jencine 4	WRL	1 NCP
Herd Vac 3	Singer	1a CP
Killed		
Elite4	Singer	1a CP
Horizon 4 plus	C24V	1a CP
	125C	2 CP
Master Guard 5	C24V	1a CP
	125C	2 CP
Respishield 4	Singer	1a CP
Triangle 4+ type II	Singer	1a CP
	5912	2 CP
CattleMaster 4	5960	1a CP
	6309	1 NCP
ViraShield 5	KY22	1a CP
	TN 131	2 NCP
Surround 4	Singer	1a CP
	NY	1b NCP

Most **MLV vaccines** elicits antibodies against the BVDV E2, NS2/3 and also E1 and Erns proteins. **Killed vaccines** mainly induce antibodies against E2. Infection with BVDV also induces the same set of antibodies. Therefore, antibody tests against various structural proteins can be indicative of vaccine status or infection or both. Until now **DIVA** test for BVDV is not possible using classical MLV or Killed vaccines. New chimeric pestivirus vaccines consisting of a BVDV backbone with a swap of CSFV E2 (CP7_E2alf) or E1 and E2 (CP7_E1E2alf) are promising live marker vaccine candidate for DIVA applications.

Drawbacks of current BVDV Vaccine: The very large antigenic and genetic diversity of BVDV strains (1-3) is well documented and required vaccination against each strain. No vaccine available for the new strain of BVDV3. None of the commerical vaccines offer clear DIVA tests. However the antigenic variation between these strains was unknown.

BVDV-Bovine-Viral-Diarrhea-Virus-Vaccine-ELISA-Flr-u 160416A

India Contact:
Life Technologies (India) Pvt. Ltd.
306, Aggarwal City Mall, Opposite M2K Pitampura, Delhi – 110034 (INDIA). Ph: +91-11-42208000, 42208111, 42208222, Mobile: +91-9810521400, Fax: +91-11-42208444
Email: customerservice@lifetechindia.com Website: www.lifetechindia.com