Diagnosing, Differentiating, and Managing Infections with BVD Virus

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May 1, 2017

Bovine viral diarrhea virus (BVDV) causes significant gastrointestinal, respiratory and reproductive disease in cattle. The economic impact of BVDV on beef and dairy industries is well known by those who deal with the disease in the field. Tangible estimates of losses are found in several reports. For example, when acute infections reach 34% within a population, total annual losses were projected to be $20 per calving due to low-virulence strains and $57 per calving due to highly virulent strains. Furthermore, it should be noted that the true economic impact of BVDV on the beef and dairy industries is underestimated because of subtle losses due to subfertility and immune system suppression.

To a degree, the importance of BVDV as a disease causing organism and the costs of living with it are further demonstrated by the large number of licensed vaccines that are available (Table 1). However, it should be understood that vaccination provides important but *incomplete* protection. Modified-live vaccines are considered to provide greater fetal protection that killed vaccines (Table 2). **Based on recent studies, heifers should receive at least two doses of a modified-live vaccine containing type 1 and 2 strains of BVDV 30 days prior to breeding to maximize the protection which the dam's immune system can provide to the gestating fetus.** If fetuses are infected with BVDV during the first 150 days of gestation, they may be aborted or alter their immune system to allow the viral infection to persist for life (Figure 1). These persistently infected animals shed huge concentrations of infectious virus in feces, urine, saliva, nasal discharge, hair and secretions from the reproductive tract. These persistently infected animals, which are more common than any of us would like (Table 3), are the critical link for continued viral transmission and economic losses due to this virus.

Control of BVDV requires identification and removal of all persistently infected animals. Identification of persistently infected animals requires that animals in the herd be tested for virus. Producers are strongly encouraged to work with their local veterinarian in developing a strategy for testing animals for BVDV. Multiple assays are widely available for detection of persistently infected cattle (Table 4). Many diagnostic labs can perform the antigen capture enzyme linked immunosorbent assay (ELISA) test on ear notches submitted after freezing or brief refrigeration. Other labs commonly perform an immunohistochemistry test on ear notch tissue submitted in formalin. These tests are not affected by ingestion of maternal antibodies in colostrum, so they can be recommended on animals less than 3 months of age. The antigen capture ELISA test on serum samples or virus isolation from serum can be affected by ingestion of maternal antibodies in colostrum, so these tests are recommended only on animals over 6 months of age. While these tests are very reliable, no test is perfect. Thus, valuable animals that test positive should be retested 21 days later to verify if the animal is persistently infected with BVDV. If a non-pregnant animal is only transiently infected, the clinical impact may be mild and there is no reason to cull the animal. If a pregnant animal is transiently infected, the developing fetus may become persistently infected during the first 150 days of gestation. Persistently infected animals represent an infectious threat for all contacted animals, especially pregnant heifers or cows. If an animal is persistently infected, the animal should be sold for slaughter only or humanely euthanized. Detection and removal of persistently infected animals is the first step in controlling BVDV within beef herds. Maintaining appropriate biosecurity is the second step in controlling BVDV within beef operations. This is especially important for cow/calf

operations which can generate persistently infected animals to spread the pathogen throughout other segments of the beef industry.

Understanding the ecology of BVDV in cattle leads to definition of a few key status descriptors for individual animals. If an animal is (a) **persistently infected**, as classically defined by positive results for detection of virus on two tests performed three to four weeks apart, then this animal will consistently and persistently shed virus in all excretions and cause infection of naïve animals in direct contact. If an animal is (b) **tested free of virus**, then this animal is not persistently infected and cannot become persistently infected. The cow or heifer with this status can give birth to a persistently infected calf if exposed to virus during the first 5 months of gestation. If an animal is (c) **tested free of anti-BVDV antibodies and free of virus** then this animal is not persistently infected and has not been exposed to BVDV. If a cow or heifer meets this criteria within 3 months prior to calving, then the resulting calf will not be persistently infected. This third status can only be achieved when vaccination is not employed because immunization will result in the presence of anti-BVDV antibodies.

Important but less permanent status descriptors for individual animals include:

1. **Acute infections**- These animals shed low concentrations of virus that may (or may not) result in infection of contacted cattle. Acutely infected animals shed virus for 7 to 21 days after initial infection. In one study, 93% of persistently infected calves were born from acutely infected dams. The use of PCR or virus isolation assays are commonly required to diagnose acutely infected animals within the narrow window of opportunity to detect virus.
2. **Testicular infections**
    a. **Prolonged testicular infections**- After acute infection, some bulls will seroconvert and continue to produce semen that is contaminated with BVDV as determined by PCR for many months (up to two years). While this virus can be detected by PCR, anti-BVDV antibodies from the secondary sex glands prevent routine isolation of the virus from semen. Ultracentrifugation of this semen may result in the fractionation of infectious BVDV. Despite substantial research, the epidemiologic significance of prolonged testicular infections are questioned as viral transmission has not been detected by natural breeding or artificial insemination.
    b. **Persistent testicular infections**- Seropositive bulls with persistent testicular infections produce semen from which BVDV can be isolated. The concentration of virus in semen from these bulls is 10^4 to 10^7 CCID50/mL. This infection has been demonstrated to be epidemiologically significant as virus can be transmitted in this semen to seronegative heifers by natural breeding or artificial insemination. Euthanasia of a bull with a persistent testicular infection at 22 months of age resulted in isolation of virus only from testicular tissue. Notably, multiple bulls demonstrating a persistent testicular infection have cleared the infection and produced semen that is free of virus as determined using both virus isolation and PCR after months of monitoring the progression of this localized infection.

Understanding the ecology of BVDV, the prevalence of persistently infected animals, the advantages and disadvantages of various diagnostic tests and the strengths and weaknesses of available vaccines allows development of a control program that will minimize the impact of this pathogen on the cattle industry.

References available upon request.

Table 1. Vaccines available for bovine viral diarrhea virus.

Manufacturer	Vaccine	Modified Live (MLV) or Killed	Contains Type 1 BVDV			Contains Type II BVDV		Label Claims for BVDV*			Approved for Pregnant Cows	Dosage	2 Doses Initially	Route		Meat Withdrawal	Duration of Immunity
			Subtype	Biotype	Strain	Biotype	Strain	type 1	type 1b	type 2				IM	SQ		
Boehringer Ingelheim	Elite 9	Killed	1a	cp	Singer	cp	296	7			√	5 mL	√	√		21 days	
Boehringer Ingelheim	Triangle 10	Killed	1a	cp	Singer	cp	5912	8		8	√	5 mL	√	√	√	21 days	
Zoetis	CattleMaster Gold FP 5 L5	Killed	1a	cp	5960	cp	53637	8, 12		8, 12	√	5 mL	√		√	21 days	
			1	ncp	6309												
Elanco	Master Guard 10 HB	Killed	1a	cp	C24V	cp	125C	8			√	3 mL	√	√	√	21 days	
Elanco	ViraShield 6 + VL5	Killed	1a	cp	KY22	ncp	TN 131	8		8	√	5 mL	√		√	60 days	
			1	ncp													
Boehringer Ingelheim	Express FP 10	MLV	1a	cp	Singer	cp	296	8, 12		8, 12	Provisional	2 mL		√	√	21 days	12 mo
Boehringer Ingelheim	Pyramid 5 Presponse SQ	MLV	1a	cp	Singer	cp	5912	8, 12	7.5	8, 12	Provisional	2 mL		√	√	21 days	Rsp. 217 D
Zoetis	Bovi-Shield Gold FP VL5	MLV	1a	cp	NADL	cp	6	8, 12		8, 12	Provisional	2 mL	For preg revac	√	√	21 days	12 mo
Zoetis	PregGuard Gold FP 10	MLV	1a	cp	NADL	cp	53637	8, 12		8, 12	Provisional	2 mL	For preg revac	√		21 days	12 mo
Elanco	Titanium 5 L5 HB	MLV	1a	cp	C24V	cp	296	8		8	Provisional	2 mL	For BRSV	√	√	21 days	
Elanco	BRD Shield	MLV	1a	**ncp**		**ncp**		7.5		8	∅	2 mL	For BRSV		√	21 days	
Elanco (Allied Bio)	Viralign 6	MLV	1a	cp		cp		1,3	2,4,5,8	10	∅	2 mL			√	21 days	
			1b	cp													
Intervet/Merck	Vista 5 L5 SQ	MLV	1a	cp	Singer	cp	125A	7.5, 12.5		8, 12.5	Provisional	2 mL			√	21 days	206 D

Provisional= Use is approved in pregnant cows if cows have been vaccinated with this same product prebreeding. These vaccines are also provisionally approved for use in calves nursing pregnant cows.

BVDV label claims supported by the USDA Center for Veterinary Biologics

Claim (with identifying code number)

- An aid in the prevention of _____ caused by Bovine Virus Diarrhea Virus __ (BVDV __) infection.
  - **viremia**
    1. reduction of
    2. prevention of
  - **nasal shedding**
    3. reduction of
    4. prevention of
  - **leukopenia**
    5. reduction of
    6. prevention of
  - **clinical signs of disease**
    7. reduction of
    7.5 control of
    8. prevention of
  - **clinical disease and mortality**
    9. reduction of
    10. prevention of

- An aid in the prevention of _____ caused by Bovine Virus Diarrhea Virus __ (BVDV __) infection.
  - **abortion**
    11. prevention of
  - **persistently infected calves**
    12. prevention of
  - **fetal infection, including persistently infected calves**
    12.5 prevention of

Table 2. Results of studies to assess fetal protection resulting from vaccination.

Report	Publication year	Vaccination characteristics							Interval to challenge	Challenge characteristics						Viremia endpoint					Fetal infection endpoint	
		Killed or MLV	Name	Biotype	Genotype	Route	# of doses	Dose type		Source	Route	Duration	Genotype	Biotype	Day of gestation	Days PE	Serum Control	Serum Subject	WBC Control	WBC Subject	Control	Subject
Zimmerman et al.	2013	MLV	Express 5 VL5	CP	1a & 2	SQ	1	MID*	~ 1 y	Cell culture	IN	1d	1b (BJ)	NCP	80 to 90	2, 4, 6, 8, 10, 12, 14			19/23 83%	0/22 0%	15/18 83%	0/21 0%
										Cell culture	IN	1d	2 (PA131)	NCP	80 to 90				20/22 91%	1/18 6%	21/22 96%	0/18 0%
Meyer et al.	2012	MLV	Mucosiffa	CP	1a	IM	1	10^3.5	4 months	Cell culture	IN	1d	1f (02146Ha	NCP	49, 74, or 96						9/9 100%	0/15 0%
Xue et al.	2011	MLV	Vista 5 SQ	CP	1a & 2	SQ	1	Release	121 d	Cell culture	IN	1d	1b (SD02 BV	NCP	73 to 87	6 to 10			12/12 100%	1/23 4%	12/12 100%	1/23 4%
Givens et al.	2011	MLV	Express 5	CP	1a & 2	SQ	2	Release	102 d	3 PI cattle	exposure	56 d	1a, 1b & 2	NCP	68 to 124	6, 7, 8, 9, 10, 28	10/10 100%	0/20 0%	10/10 100%	4/20 20%	10/10 100%	0/19 0%
Leyh et al.	2011	Control nonvaccinates							124 d	8 PI cattle	exposure	14 d	1b	NCP	64 to 103	6, 9, 14			8/10 80%		10/10 100%	
		MLV	Bovishield Gold FP5 VI	CP	1a & 2	IM	1	MID*												1/20 5%		3/20 15%
		MLV	Bovishield Gold FP5 VI	CP	1a & 2	SQ	1	MID*												3/20 15%		3/20 15%
Rodning et al.	2010	Control nonvaccinates							91 d	3 PI cattle	exposure	58 d	1a, 1b & 2	NCP	68 to 126	6, 7, 8, 9, 10, 28	8/10 80%		10/10 100%		10/10 100%	
		MLV	Bovishield Gold FP5	CP	1a & 2	IM	4	Release										0/20 0%		0/20 0%	5-type 1a, 3-type 1b, 3-type 2	0/19 0%
		MLV	Pyramid 5	CP	1a & 2	SQ	4	Release										0/20 0%		1/20 5%		0/18 0%
		Inactive	Virashield 6	NCP & CP	1 & 2	SQ	4	Release										0/20 0%		10/20 50%		2/18 11%
																						1-type 1b, 1-type 2
Xue et al.	2009	MLV	Vista 5 SQ	CP	1a & 2	SQ	1	Release	191-205	Cell culture	IN	1d	1b (SD02 BV	NCP	163 to 177	5,6,7,8,9,10			12/12 100%	1/25 4%	11/11[a] 100%	4/22 18%
										Cell culture	IN	1d	2	NCP	163 to 177	5,6,7,8,9,10			18/18 100%	2/28 7%	18/18[b] 100%	7/28[c] 25%
Grooms et al.	2007	Inactive	Cattlemaster Gold FP 5-L5	CP	1a & 2	SQ	2	Release	69 d	3.5 PI cattle	exposure	98 d	2-1b, 2-2a	NCP	52 to 150	7, 9, 11			8/15 53%	3/15 20%	14/14 100%	4/15 27%
																					8-type 1, 11-type 2	4-type 1, 2-type 2
Schnackel et al.	2007	MLV	Pyramid 9	CP	1a	SQ	1	MID*	110 to 128 d	Cell culture	IN	1 d	2 (96B2222)	NCP	75 to 83						10/10 100%	0/25 0%
									114 to 150 d	Cell culture	IN	1 d	1b (97B1415)	NCP	75 to 97						6/6 100%	1/24 4%
Ellsworth et al.	2006	MLV	Bovishield Gold FP5 VL5	CP	1a & 2	IM	1	MID*	490 d	4 PI cattle	exposure	45 d	2a	NCP	149 to 262	7, 9, 12			8/10 80%	0/20 0%	10/10 100% 1 PI, 9 CI	1/20 5% 1 CI
Brock et al.	2006	MLV	Arsenal 4.1®	NCP	1a	IM or SQ	1		125 to 135 d	Cell culture	IN	1 d	1b (BJ)	NCP	=75						9/9 100%	3/23 13%
										Cell culture	IN	1 d	2 (PA131)	NCP	=75						8/8 100%	1/14 7%
Ficken et al.	2006	MLV	Bovishield 4	CP	1a	IM	1		125 d	Cell culture	IN	1 d	2 (94B-5359a)	NCP	75 to 100	2, 4, 6, 8, 10, 14			9/10 90%	1/20 5%	9/10 90%	6/18 33%
		MLV	Bovishield 4	CP	1a	IM	2		125 d	Cell culture	IN	1 d	2 (94B-5359a)	NCP	75 to 100	2, 4, 6, 8, 10, 14			9/10 90%	1/20 5%	9/10 90%	7/19 37%
		MLV	Bovishield FP5	CP	1a & 2	IM	1		146 d	Cell culture	IN	1 d	2 (94B-5359a)	NCP	62 to 104	6, 7, 8, 9, 10, 14			10/10 100%	0/20 0%	9/9 100%	0/18 0%
Fairbanks et al.	2004	MLV	Breedback FP	CP	1a & 2	SQ	1		120 to 132 d	Cell culture	IN	1 d	1b (BJ)	NCP	76 to 79	1, 3, 4, 6, 8, 10, 12			9/10 90%	1/18 6%	10/10 100%	0/18 0%
										Cell culture	IN	1 d	2 (PA131)	NCP	76 to 79				8/8 100%	0/19 0%	8/8 100%	1/19 5%
Kovacs et al.	2003	MLV	Breedback FP	CP	1a & 2	SQ	1		88 to 146 d	Cell culture	IN	1 d	1 (#9 KE9)	NCP	60 to 90	PCR every 2 to 40			8/8 100%	0/11 0%	8/8 100%	1/11 9%
										Cell culture	IN	1 d	2 (NY 93)	NCP	60 to 90				7/7 100%	0/11 0%	1/1 100%	0/9 0%
Dean et al.	2003	MLV	Jencine®	NCP	1a	IM or SQ	1		121 d	Cell culture	IV	1 d	1a (7443)	NCP	55 to 100	every 2 to 14					13/13 100%	2/27 7%
Patel et al.	2002	Inactive	Bovilis BVDV	CP	1a	IM	2		187 d	3 PI cattle	exposure	14 d	1a	NCP	57 to 101	1 to 12 except 6	7/7 100%	0/11 0%	7/7 100%	5/11 45%	7/7 100%	0/11 0%
Frey et al.	2002	Inactive Then MLV	Mucobovin® Vacoviron®	NCP CP	1a 1a	SQ IM	1 1		122 d	Cell culture	IN	1 d	1f & 2	NCP	30 to 120	3, 5, 7, 9			6/6 100%	1/9 11%	6/6 100% All type 2	0/9 0%
Van Campen et al.	2002	MLV	Pyramid®	CP	1a	SQ	1		102 to 138 d	Cell culture	IN	1 d	2	NCP	74 to 85	1 to 21			5/7 71%	6/21 29%	5/7 71%	0/21 0%
Cortese et al.	1998	MLV	Resvac 4	CP	1a (NADL)	IM	1	Release	107 d	Cell culture	IN	1 d	1b (BJ)	NCP	70 to 75	1 to 10			4/6 67%	0/12 0%	6/6 100% 1 was Type 2	2/12 17%
Brownlie et al.	1995	Inactive	Bovidec	NCP	1a	SQ	2		25 to 90 d	Cell culture	IN	1 d	1a (Pe515)	NCP	25 to 80	1 to 14			3/6 50%	0/6 0%	4/5 80%	0/4 0%
		Inactive	Bovidec	NCP	1a	SQ	3													0/9 0%		0/9 0%
Meyling et al.	1987																					
Harkness et al.	1987																					
McClurkin et al.	1975																					

*Minimum Immunizing Dose

[a]5/12 (42%) of controls VI positive from tissues when challenged at d177 of gestation and tested after euthanasia at birth

[b]17/18 (94%) of controls VI positive from tissues or buffy coat when challenged at d177 of gestation and tested after euthanasia at d237 of gestation

[c]2/28 (7%) of vaccinates VI positive from tissues or buffy coat when challenged at d177 of gestation and tested after euthanasia at d237 of gestation

Figure 1. The transmission cycle of bovine viral diarrhea virus in cattle.

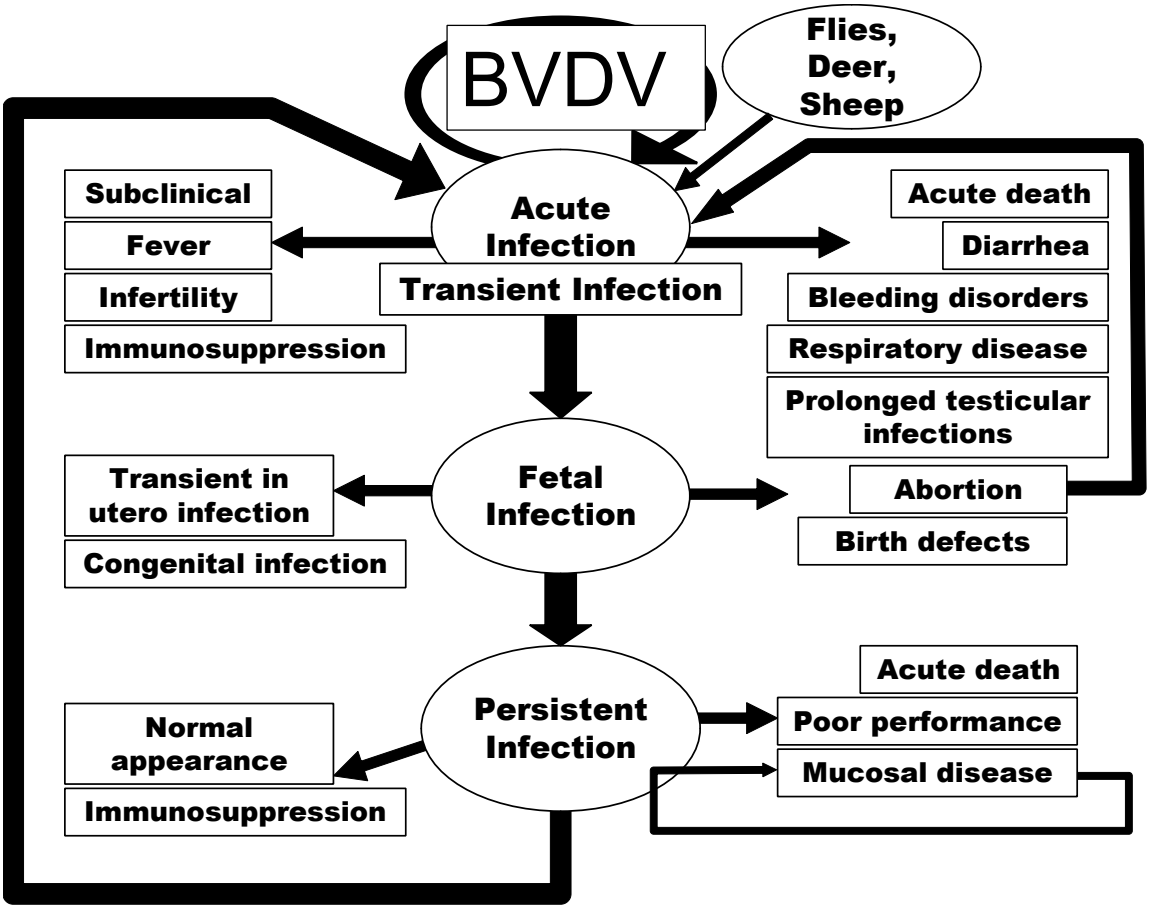

Table 3. The prevalence of cattle persistently infected (PI) with bovine viral diarrhea virus in various studies.

Report	Reference	Data collection year	Geographic Area	Population	PI Prevalence	Herd Prevalence
Hoar et al.	{1500}	2004	CA	Dairy Steers	3/900 (0.3%)	
Grooms & Keilen	{1145}	2000	MI	Dairy Neonates	6/332 (1.8%)	
Munoz-Zanzi et al.	{1190}	1999	CA	Dairy Neonates	2/434 (0.5%)	
Houe et al.	{439}	1995	MI	Dairy Herds	7/5,481 (0.13%)	3/20 (15%)
Mawhinney et al.	{1505}	2007	United Kingdom	Dairy Heifers & Cows	3/1,769 (0.2%)	
Grooms et al.	{1088}	2001	MI	Dairy & Beef cows	5/1,952 (0.26%)[d]	3/13 (23%)[d]
Stephenson et al.	{1593}	2005	Southeast U.S.	Stocker Beef calves	24/7,544 (0.3%)	
Cornish et al.	{1367}	2005	WY	Beef calves[c]	59/559 (10.5%)[c]	
O'Connor et al.	{1504}	2006	IO	Beef calves	12/12,030 (0.09%)[c]	4/102 (4%)
Wittum et al.	{1494}	1996	Multistate U.S.	Beef cow-calf	56/18,931 (0.3%)	3/76 (4%)
Fulton et al.	{1496}	2006	TX & OK	Beef cow-calf	26/4,407 (0.6%)	5/29 (17.2%)
Paisley et al.	{309}	1996	Multistate U.S.	Beef cow-calf	8/1,201 (0.67%)[a]	
Bolin et al.	{1495}	1985	Multistate U.S.	Beef cow-calf	54/3,157 (1.7%)[b]	6/66 (9%)[b]
NAHMS	{1646}	2007-2008	Multistate U.S.	Beef cow-calf	53/44,150 (0.12%)	18/53 (8.8%)
Cleveland et al.	{1253}	2003	CO	Endemically infected cattle herd	5/2,921 (0.17%)	
Taylor et al.	{352}	1992	Western Canada	Infected beef herd	51/560 (9.1%)	
Taylor et al.	{412}	1991	Western Canada	Feedlot	1/1,029 (<0.1%)	
Loneragan et al.	{1497}	1998	TX	Feedlot	8/4,000 (0.2%)	
Fulton et al.	{1476}	2004	KS	Feedlot	86/21,743 (0.4%)	
Larson et al.	{1478}	2004	MO	Feedlot	3/938 (0.32%)	
Loneragan et al.	{1347}	2002-03	KS, TX	Feedlot-arrival	6/2,000 (0.3%)	
Loneragan et al.	{1347}	2002-03	KS, TX	Feedlot-chronically ill	36/1,383 (2.6%)	
Loneragan et al.	{1347}	2002-03	KS, TX	Feedlot-dead	39/1,585 (2.5%)	
Howard, et al.	{104}	1988	Northeastern U.S.	AI Centers	12/1,538 (0.78%)	
Lawrence & McClure	{1535}	2005-07	34 U.S. States, Mexico, Canada	Cattle	3,489/866,602 (0.4%)	
Yan, et al.	{1686}	2006-2008	Mississippi	Cattle	111/27,932 (0.4%)	
Pogranichniy, et al.	{1536}	2006	Indiana	White-tailed deer	2/745 (0.3%)	
Duncan, VanCampen et al.	{1534}	2005-06	CO	Wild cervids	1/5,951 (0.02%)	

[a]1,201 animals tested of 1,223 previously screened to be seronegative of 3,894 samples submitted.

[b]Herds not randomly selected; contained herds with and without histories of BVDV infection.

[c]Herds not randomly selected; calves 1 to 5 months of age.

[d]Samples from farms suspecting problems due to BVDV.

Table 4. Diagnostic Assays to detect bovine viral diarrhea virus (BVDV).

		Virus isolation	Antigen capture ELISA	Antigen capture ELISA	Individual PCR	Pooled PCR	IHC
	Characteristics	Serum	Serum	Ear Notch	Buffy Coat	Ear Notch	Ear Notch
1	Sensitivity	☆☆☆☆	☆☆☆	☆☆☆☆	☆☆☆☆	☆☆☆	☆☆☆☆
2	Specificity	☆☆☆☆	☆☆☆☆	☆☆☆☆	☆☆☆☆	☆☆☆☆	☆☆☆☆
3	Repeatability	☆☆☆	☆☆☆☆	☆☆☆☆	☆☆☆☆	☆☆	☆☆☆☆
4	Turn around time	☆	☆☆☆☆	☆☆☆☆	☆☆☆☆	☆☆☆☆	☆☆☆
5	Cost per sample	Moderate	Low	Low	High	Low	Low
6	Availability	☆☆	☆☆☆☆	☆☆☆☆	☆☆	☆☆	☆☆☆
7	Distinguish PIs from transient infections	∅	∅	Usually	∅	∅	Usually
8	Ease of sample collection & handling	☆☆	☆☆	☆☆☆☆	☆☆	☆☆☆☆	☆☆☆
9	Facilitates sample pooling	Decrease sensitivity	∅	∅	√		∅
	Appropriate for:						
10	< 3 months of age	∅	∅	√	√	√	√
11	>3 months of age	√	√	√	√	√	√

Key
☆☆☆☆= best
☆= least desirable
∅ = No
√ = Yes