

**Evaluation of Diagnostic Tests
Used for Detection of Bovine
Viral Diarrhea Virus and
Prevalence of BVDV Subtypes
1a, 1b, and 2a in Persistently
Infected Cattle Entering a
Feedlot**

**Dr. Robert W. Fulton
Oklahoma State University**



Project Support

- **Grant from The Noble Foundation, Ardmore, Oklahoma (NF)**
 - **Drs. A. Confer and R. Fulton, Project Directors**
- **Collaboration**
 - **Agricultural Division (NF)**
 - **Dr. Billy Cook (NF)**



Publication of Study Results

Journal of American Veterinary Medical Association , February 15, 2006

“ Evaluation of Diagnostic Tests used for the Detection of Bovine Viral Diarrhea Virus and Prevalence of BVDV subtypes 1a,1b,and 2a in Persistently Infected Cattle Entering a Feedlot”



Investigators

- **Bill Hessman, DVM , Sublette,KS**
- **Julia Ridpath, PhD, USDA NADC**
- **Bill Johnson, DVM, OSU OADDL**
- **Jerry Saliki, DVM, PhD, OSU OADDL**
- **Dave Sjeklocha, DVM, Sublette,KS**
- **A.W. Confer, DVM, PhD, OSU VPB**
- **Lurinda Burge, MS, OSU VPB**
- **Mark Payton, PhD, OSU Dept Statistics**
- **Rebecca Funk, OSU VPB**



Materials and Methods

- **Feedlot collections: July-December 2004**
- **21,743 cattle entering feedlot**
- **Initial collection: Fresh ear notch in PBS**
- **Initial test: Antigen capture ELISA (ACE) at veterinary clinic**
- **Subsequent collections for confirmation at OSU CVHS**



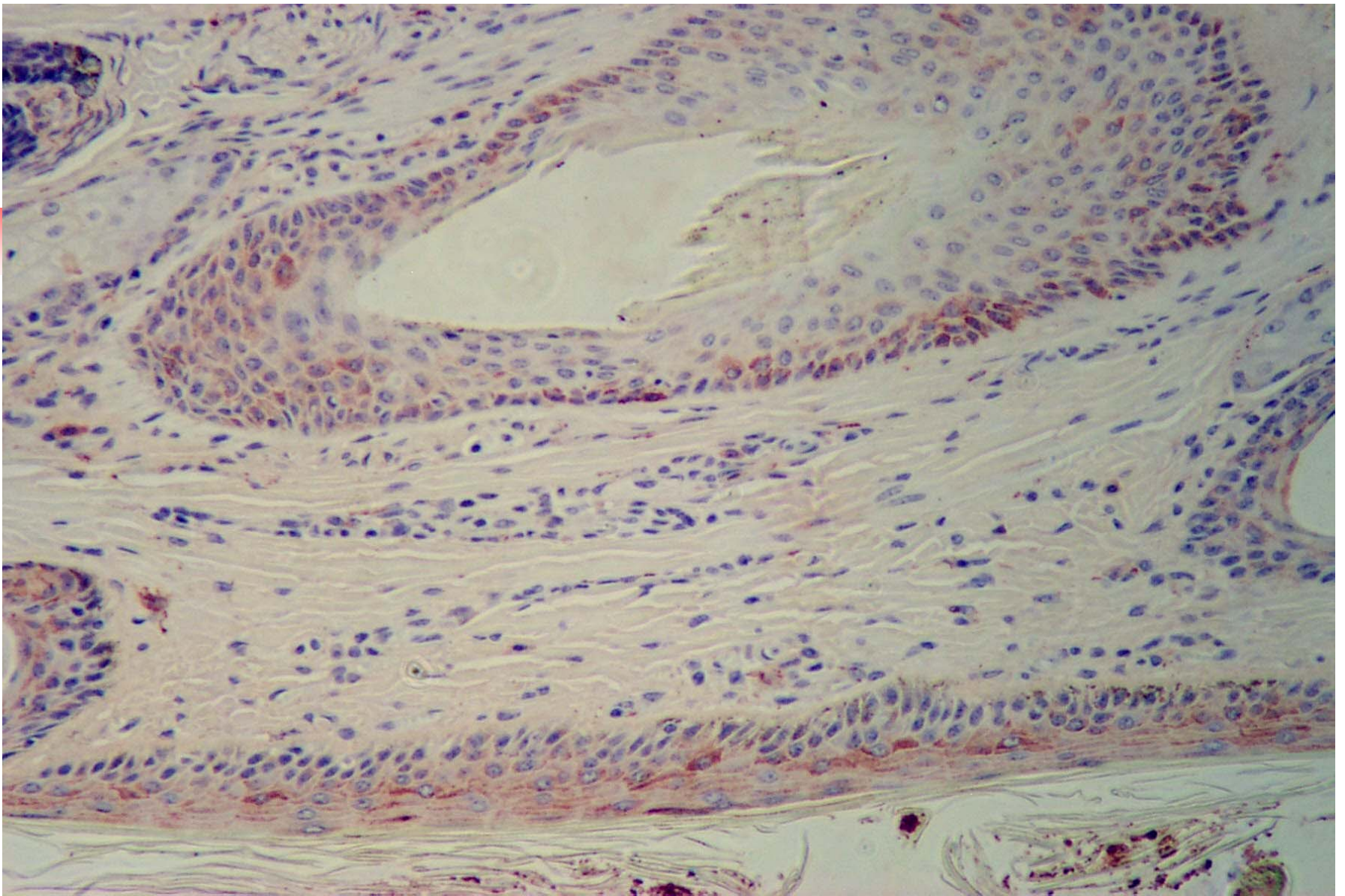
Subsequent collection on ACE positives on initial test

- All initial collections of ACE + were repeated: >90% within 48 hours after initial test
- Second fresh ear notch in PBS for ACE
- Formalin notch for IHC
- Serums and nasal swabs for PCR and virus isolation and titration
- Subtyping for BVDV 1a, 1b, 2a, and 2b



Results of Second (Confirmatory Testing)

- 86/88 were ACE positive and 2 were ACE negative , IHC neg, VI neg
 - 86/86 ACE positives also IHC positive
 - 86/86 ACE+ and IHC+ = Viral isolation+ and PCR + on serum
 - The two ACE+ on initial test and ACE/IHC/VI negative on second test were potential false positive for PI
- **86 of 21,743 = PI (0.4%)**



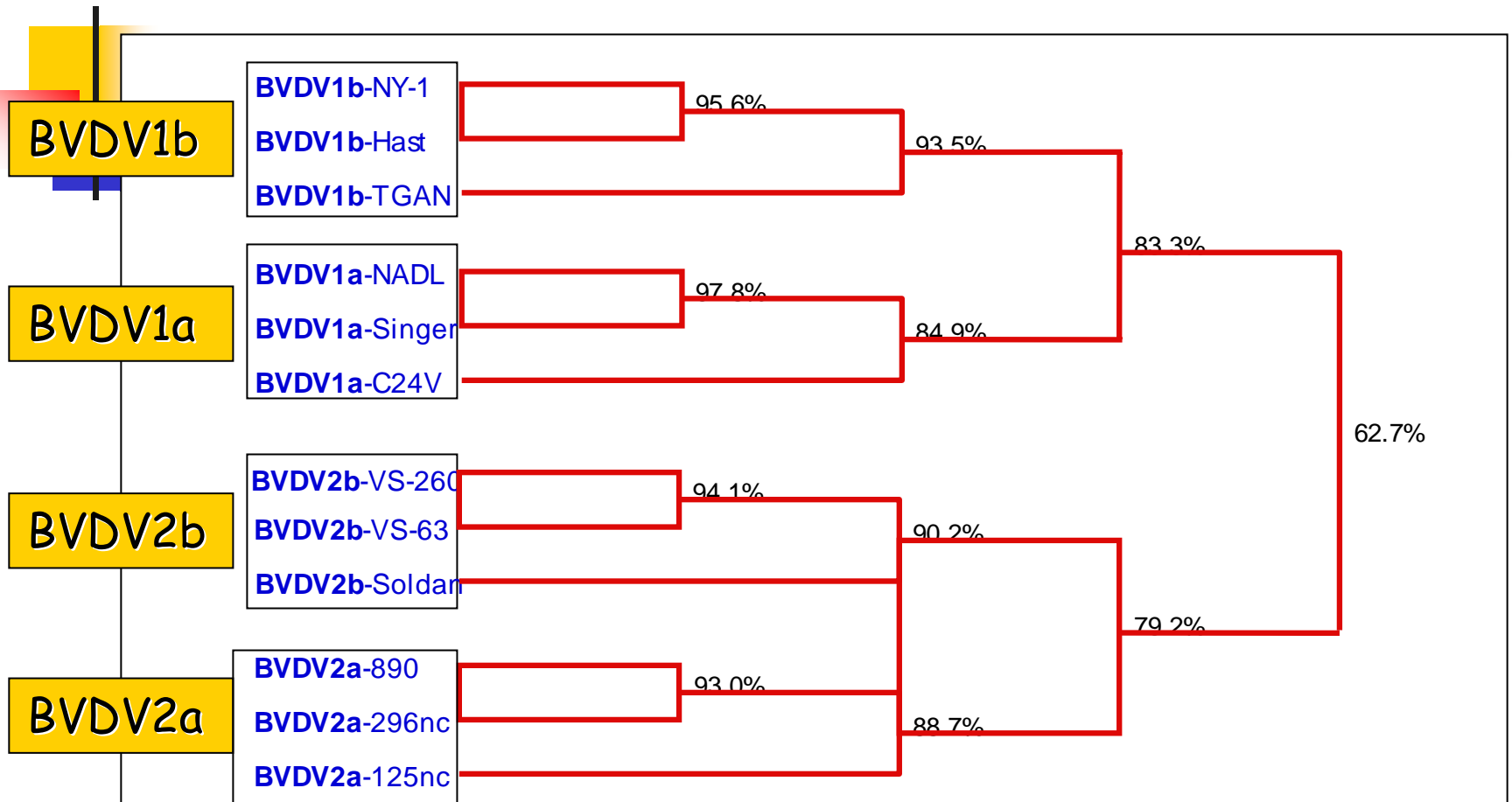
**Positive IHC BVDV ear notch from PI calf: OADDL
Dr. Bill Johnson: Feedyard Calf**



Predictive value of positive ACE test on ear notch at processing

- 86 of 88 initial ACE positive = PI
- 97.7 % considered as PI using additional tests

BVDV Subgenotypes





BVDV subtypes in US vaccines

- **BVDV1a CP**
- **BVDV1 NCP**
- **BVDV2a CP**
- **BVDV1a NCP**
- **BVDV 1 not specified**



Distribution of BVDV subtypes in diagnostic lab cases

- BVDV1a 37/131 (28.2%)
- BVDV2a 34/131 (26.0%)
- BVDV1b 60/131 (45.8%)

Veterinary Microbiology (2005),
111,pp 35-40



Distribution of BVDV subtypes in PI cattle entering the feedlot : 2004 current study

- **BVDV 1a: 10/86 (11.6%)**
- **BVDV 2a: 9/86 (10.5%)**
- **BVDV 1b: 67/86 (77.9%)**



Distribution of BVDV subtypes in PI cattle entering the feedlot : 2005-2006 follow up to current study

- **Kansas feedlot**

- BVDV 2a: 10/78(12.8%)
- BVDV 1a: 8/78 (10.3%)
- **BVDV 1b: 60/78 (76.9%)**

- **Clayton NMSU study**

- BVDV 1a: 3/17 (17.6%)
- **BVDV 1b: 14/17 (82.4%)**

Order buyer location and PI pens

State	No.	PI	%PI	PI pens	PI pen rate
Arkansas	403	8	2.0%	3/4	75.0%
N.Carolina	851	4	0.47%	4/11	36.4%
Florida	1930	3	0.16%	3/18	16.7%
Kentucky	415	1	0.24%	1/4	25.0%
Missouri	1323	2	0.15%	2/15	13.3%
Mississippi	756	1	0.13%	1/8	12.5%
Oklahoma	8184	42	0.51%	36/88	40.9%
Tennessee	1227	7	0.57%	7/14	50.0%
Texas	5691	15	0.26%	15/67	22.4%
Virginia	963	3	0.31%	2/11	18.2%
Total	21,743	86	0.40%	74/240	30.8%

Individual order buyer: PI

Buyer	State	No.	PI	%PI	PI pens	PI pen rate
1	FL	1604	2	0.12%	2/15	13.3%
2	FL	326	1	0.31%	1/3	33.3%
3	TN	1227	7	0.57%	7/14	50.0%
4	NC	850	4	0.47%	4/11	36.4%
5	VA	277	2	0.72%	1/3	33.3%
6	VA	686	1	0.15%	1/8	12.5%
7	KY	314	0	0.00%	0	0.00%
8	KY	102	1	0.98%	1/1	100.0%
9	MS	756	1	0.13%	1/8	12.5%

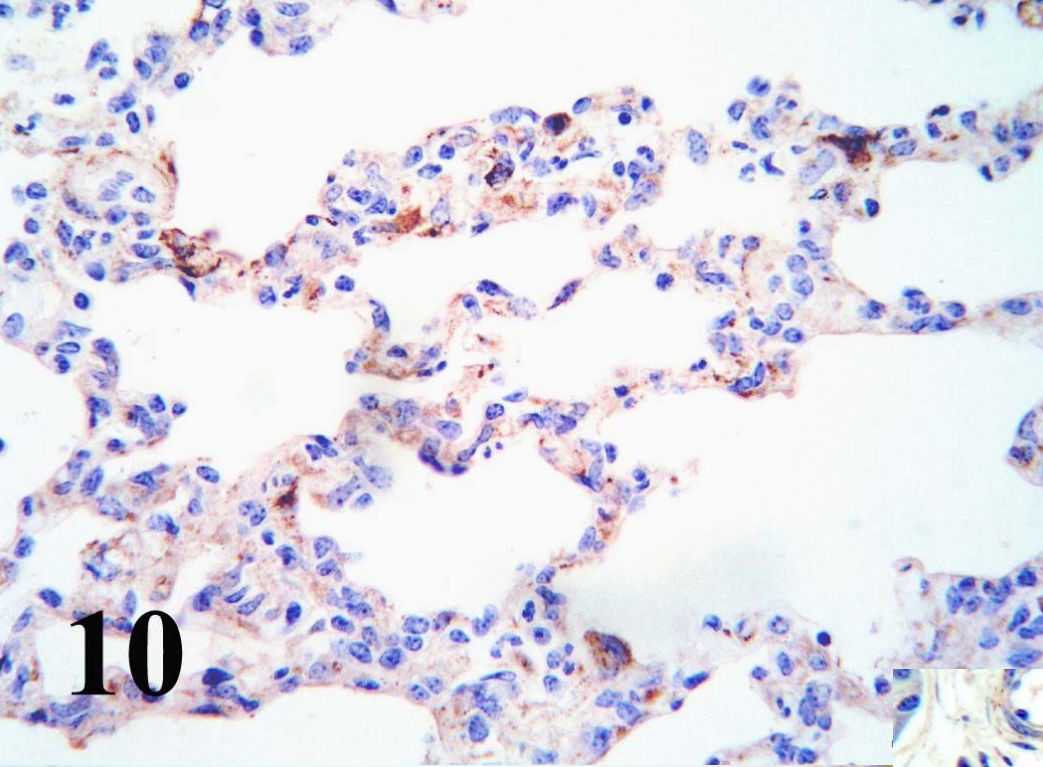
Individual order buyer: PI

Buyer	State	No.	PI	%PI	PI pens	PI pen rate
10	MO	1323	2	0.15%	2/15	13.3%
11	Ark	403	8	2.0%	3/4	75.0%
12	TEX	3698	12	0.33%	12/45	26.7%
13	TEX	1575	1	0.06%	1/17	5.9%
14	TEX	427	2	0.47%	2/5	40.0%
15	OK	7480	38	0.51%	32/80	40.0%
16	OK	221	1	0.45%	1/2	50.0%
17	OK	100	1	1.0%	1/1	100.0%
18	OK	383	2	0.52%	2/5	40.0%

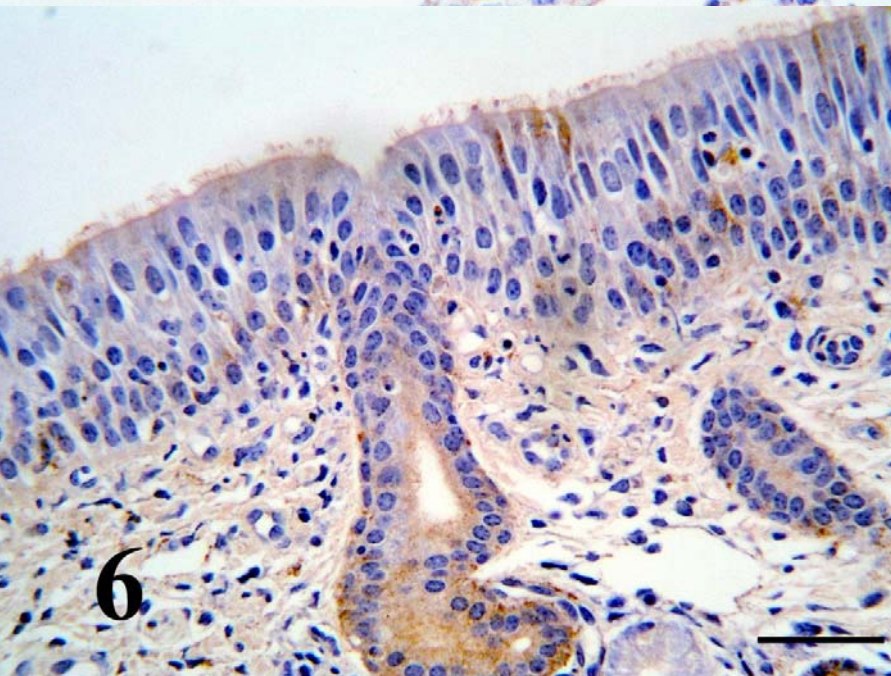
Viral Titers in Serums and Nasal Swabs from PI Calves

Calf no.	Sample	Viral titer/ml
4276	serum	4×10^4
	nasal swab	1.4×10^4
4061	serum	2.2×10^4
	nasal swab	4.5×10^3
13984	serum	2.2×10^4
	nasal swab	4.5×10^4

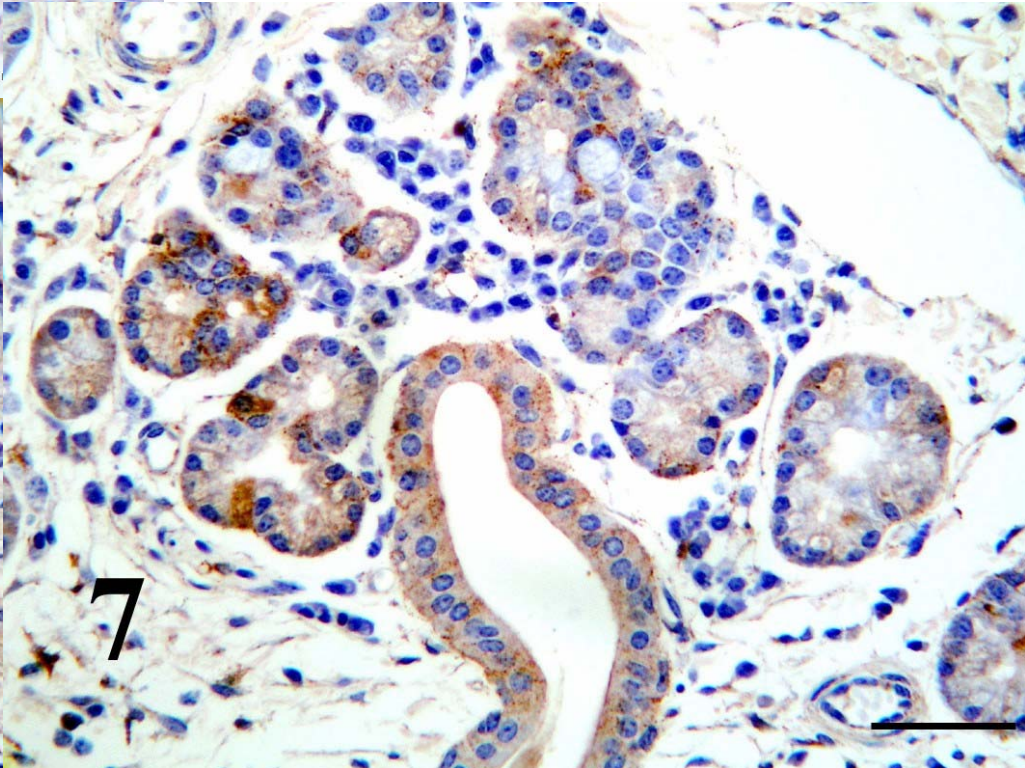
**BVDV IHC
In PI calf
Respiratory
Tract**



10



6



7



Diagnostic Tests for BVDV

- IHC on ear notches : formalin notch
- Antigen capture ELISA (ACE): PBS fresh notch
- ACE on serum
- PCR on serum
- PCR on fresh notch in PBS
- PCR on whole blood
- Viral isolation (VI) on serum/whole blood



Diagnostic Tests for BVDV

- IHC on ear notches : formalin
- Antigen capture ELISA (ACE): PBS notch
- These two tests less likely to get positive result with acute infection or MLV vaccination



Diagnostic Tests for BVDV detection for feedlot ,stocker, and breeding herd

- **Antigen capture ELISA (ACE): PBS notch**
 - Isolate and Retest calf
 - Potential false positive for PI status
 - OD readings positive but very close to positive should be retested



Diagnostic Tests for BVDV detection

- ACE on serum
- PCR on serum
- PCR on whole blood
- Viral isolation (VI) on serum/whole blood

- These tests could/will give positive result for some acute infections or MLV vaccinates



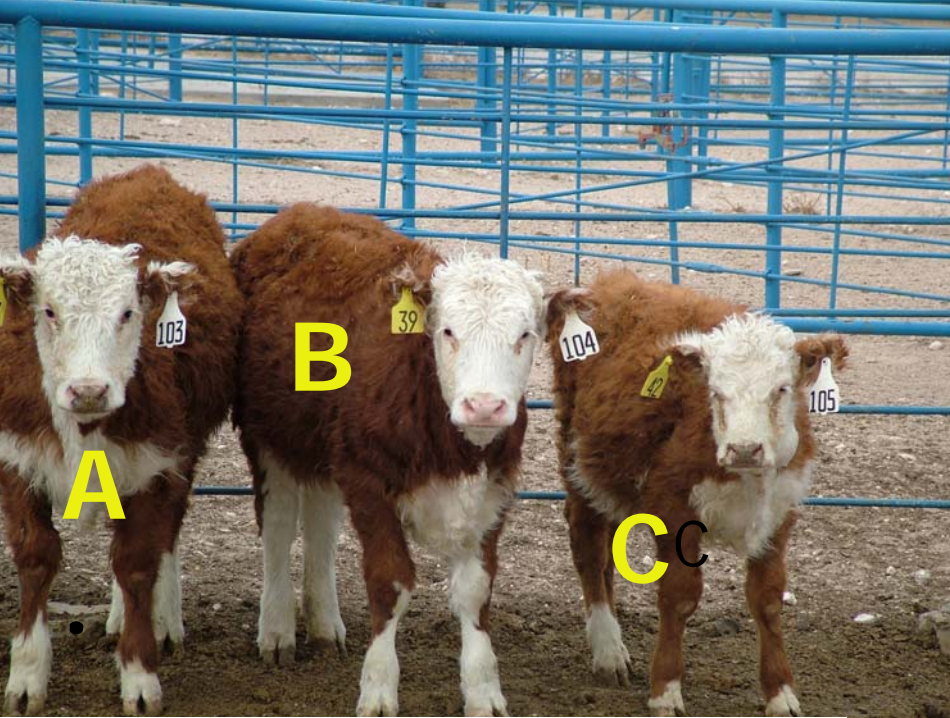
OSU Experiences with Tests

- **ACE on PBS notches**
 - PI positives (with confirming tests; IHC and VI) have stayed positive on repeated tests
 - Have not found ACE + notch in acute infections
- **Occasional ACE PBS notch + will be IHC negative: False positive, thus repeat**
- **PCR on Ear notches**
 - Some PI calves: PCR negative in fresh notches
 - Acutely infected BVDV calves can be PCR positive in serum
 - Dilutions of 1:2, 1:5, and 1:10 of one positive in PBS



Current and future projects for BVDV testing

- **Evaluation of management options for BVDV test positive cattle: stockers and feedlots**
- **Examination of additional tests**
- **Evaluation of BVDV vaccine responses to cattle exposed to PI cattle**
- **Focusing on breeding herd testing as goal**
- **Stressing animal ID to assist breeding herd decisions/biosecurity/vaccinations**



**Which animal(s) is/are
persistently
infected with Bovine Viral
Diarrhea
Virus ?**