

BVDV PI Testing Sensitivity

Which Test is Best

Many testing methodologies are available to diagnose BVDV PI infections. The sensitivity and specificity (accuracy) vary between these methodologies as does the cost of these tests. In a perfect world we would get 100% accuracy with the least expensive test. Unfortunately, we do not live in a perfect world. The most expensive test is the test with the lowest sensitivity.

Antigen Capture ELISA (ACE), IHC, Virus Isolation and PCR tests are the most common tests used today. There are many advantages and disadvantages for each of these testing methods. Most important is the accuracy of the test as even a few false negatives can prove to be very expensive for either the herd or in the feedlot.

Central States Testing utilizes the ACE as we feel it not only has the most advantages but also the best accuracy.

IHC testing is also very accurate but requires 7-9 days to receive results. Cost for IHC testing is slightly more expensive than CST's ACE test.

Virus Isolation, once considered the gold standard, is now recognized to be the least preferred method. Not only is the sensitivity poor but sample collection is difficult as serum or whole blood is the preferred sample. Also in order to be conclusive two samples must be tested at least two weeks apart.

PCR tests have been proclaimed to be so sensitive that you can pool many samples to reduce testing costs. Many recent studies have put PCR testing in doubt and have shown PCR sensitivity to be 85% or less. Although pooling does reduce the testing cost, it is at the expense of having as much as 22% false negative results. These outcomes completely remove the benefit of BVDV PI testing.

Following are studies illustrating the superior sensitivity of ACE testing when compared to PCR and other testing methods.

Edmondson MA, Givens MD, Walz PH, et al. Comparison of tests for detection of bovine viral diarrhea virus in diagnostic samples. *J Vet Diagn Invest* 19:376-381 (2007).

Samples were provided to 23 diagnostic laboratories using respective laboratories submission protocols. Results for IHC, ACE, VI and PCR are as follows on non-pooled samples.

<u>IHC (skin)</u>		<u>ACE (skin)</u>		<u>VI (serum)</u>		<u>PCR (serum)</u>	
<u>%CP</u>	<u>%CN</u>	<u>%CP</u>	<u>%CN</u>	<u>%CP</u>	<u>%CN</u>	<u>%CP</u>	<u>%CN</u>
90	98	100	100	69	98	85	89

CP- correct positive
CN- correct negative

Ridpath JF, Hessman BE, Neill JD, et al. Parameters of Ear Notch Samples for BVDV Testing: Stability, Size Requirements and Viral Load. Proceedings of AABP, September, 2006. St Paul Minnesota.

“The concentration range of virus in ear notch extractions and the detection limits of real-time PCR suggest that pooling of samples in surveillance programs must be approached cautiously. Pooling of 10 samples, where a sample pool includes one positive and nine negative samples, could result in the failure to detect 10% of the samples used in this study. Pooling of 100 samples, where sample pool includes one positive and 99 negative samples, could result in failure to detect over 50% of the samples used in this study.”

In some unpublished work samples sent to 3 diagnostic laboratories were tested as single samples and pools with ACE and PCR on skin samples. PCR correctly identified 78% of the samples positive as single tests and 39% correct in pools of 10. ACE correctly identified 100% of the samples positive as single tests and 85% correct in pools of 10.

Fulton RW, Hessman BE, Ridpath JF, et al. Utilization of Multiple Diagnostic Tests to Identify Cattle with Bovine Viral Diarrhea Virus and Duration of Positive Tests in Persistently Infected Cattle. Submitted for publication in *Vet Microb* 2007.

Twelve confirmed PI animals were tested monthly with multiple tests for 11 months. All PI cattle showed 100% sensitivity with ACE (serum and skin) and IHC on all samplings for the entire length of the study. PCR on serum showed 98.6% sensitivity and PCR on skin samples showed 83% sensitivity on initial monthly tests. All samples were tested individually and not pooled.

Although CST provides ACE, IHC and PCR testing methodologies we currently only recommend the ACE test for BVDV PI screening. Sampling is simple, turn around time for results are quick and the costs are comparable to other testing methods. The value of the ACE is greater than other tests as studies have shown that the accuracy of the ACE test is superior to other methods. False negative results can undermine any screening program and negate any and all of the benefits of BVDV PI testing. False negative results make whatever screening test you are using very expensive.

CST has been able to improve the sensitivity of the ACE test with multiple patent pending technologies. On occasions, **without** these technologies, false negative results would be reported. Although no testing methodology is 100%, our clients can be assured that CST has the best sensitivity of any lab in the US. An example of CST expertise is the famous Auburn 501 calf. This is a confirmed PI calf whose PI strain mutated, resulting in multiple negative test results by Auburn University and IDEXX Labs using their ACE test. In CST's hands, using our patent pending technology we were able to identify this calf as PI.

We know there are substances that can inhibit the BVDV making it difficult to identify PI animals and resulting in false negative outcomes. Not only can high levels of antibodies to BVDV inhibit the virus but substances yet to be identified have been found by CST that can also result in false negative outcomes. This is especially true if samples are pooled. Because of this increased risk of false negative outcomes CST **does not pool** samples. CST has identified confirmed PI animals that would result in false negative outcomes due to antibody production. CST has developed and uses an antibody screening test to evaluate samples for this potential inhibition. Mutations, though rare, are always a possible reason for false negative results as with the Auburn 501 calf. CST continues to conduct research and use novel technologies to limit these rare false negative results.

Unfortunately, due to the level of sensitivity CST can achieve, we do see a higher level of false positive results at approximately 5%. False positive results are much easier to deal with than a false negative result. Re-testing all positive results is recommended by CST prior to euthanasia of any animal. Re-sample the animal at least 10 days post initial testing and re-submit the sample with original identification as a "Re-test" and CST will confirm the PI status at **no charge**.