The Diagnostic Reliability of Pooled Ear-Notch Antigan Capture Immunosorbent Assay Testing for Bovine Viral Diarrhea Persistently Infected Individuals and Economic Determination of Optimum Pool Size

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Introduction

In order to control BVD in cattle, the key component is finding and eliminating persistently infected (PI) individuals from the herd. Many of the testing methods available are cost-prohibitive to most producers for whole-herd screening. In order to address this, the diagnostic reliability of pooled antigen-capture enzyme-linked immunosorbent assay (ACE) was explored as a cost effective alternative.

Materials and Methods

Ear notches were collected from 37 BVD persistently infected calves as well as three BVD-PI negative calves to be used as test individuals. All samples were chilled and stored in 2 ml of PBS. Approximately 5500 PI negative ear notches were collected from feedlot populations to be used in configuring the sample pools. Pools were configured with 2, 3, 4, 5, 10 and 15 individuals for each of the forty experimental samples and tested by ACE for BVD. The economic effect of pooling was explored utilizing the formula: $E(Cherd) = rc[(k+1) - k(1-\pi)k]$. Where E(Cherd) is the total cost of testing a herd; r is the number of pools; c is the cost of the test; k is the number of individuals in the pool; and π is the expected prevalence of BVD for that herd. This formula was adapted from work done by Munoz-Zanzi et al. in 2000 on the economics of pooled PCR BVD testing. The formula includes factors for adjustment in cost based on prevalence, pool size, initial testing cost, and cost of re-tested pools for confirmation of positive individuals. It does not account for differences in sensitivity of a test, and assumes 100% sensitivity. A univariate analysis of variance including LS means analysis was conducted to determine significant differences between treatment groups. The 95% confidence intervals for the mean of each treatment were plotted by the program, as well.

Results

There were significant differences as pool size increased (P = <0.001), although it should be noted that at no time did a positive pool result in a suspect or negative test. Individual samples varied in their S/P ratios at different dilutions. The two lowest S/P ratio were observed when positive samples were tested in pool sizes of 10 and 15 individuals. Pools of 10 and 15 samples had significantly lower S/P ratios than the other pools in this study. However; the 95% confidence interval around the mean remains strongly in the positive zone across all treatments. When the economic analysis is run a clear point of reversal in economic benefit is present for each prevalence level. This data shows that populations with the lowest prevalence benefits most from large-pool testing, whereas sectors or even individual operations with a suspected high prevalence may need to consider smaller pool sizes.

Significance

These data show that pooling at these pool sizes did not have a negative effect on detecting positive samples. However, positive samples included in pools of 10 or 15 samples significantly decreased the S/P ratios relative to smaller pool sizes. The expected prevalence of BVD comes heavily into play when selecting a pool size for a testing protocol; the benefit of large pools increases as the prevalence of BVD decreases. In low prevalence situations it then becomes important to balance the benefit of pooling with the increased chance of obtaining false negative results. In many instances the benefit of large pools may not outweigh the risk. Some of the elements that must be considered when selecting pool size with BVD PI testing include the expected prevalence of BVD in the individual operation, the size of the pool which can be used, and the overall cost of false positive or negative individuals. Accurate estimates of the herd prevalence are important in designing pooled testing protocols. An increase in prevalence from 0.3% to only 0.5% changes the pooling size with the most economic benefit from 20 head to 15 head. A further increase to 1% prevalence moves the point of greatest return back even more to a pool size of 10 individuals