LETTER TO THE EDITOR

Exogenous source of PRRSV antibody in positive oral-fluid ELISA results

We wish to alert swine producers, practicing veterinarians, and diagnosticians to environmental sources of porcine reproductive and respiratory syndrome virus (PRRSV) antibody capable of producing positive reactions on a PRRSV oral-fluid antibody ELISA recently implemented in a number of veterinary diagnostic laboratories in North America. ¹

An exogenous source of PRRSV antibody was initially identified in the course of investigating a diagnostic case involving unexpected PRRSV antibody-positive results on oral fluids from a PRRSV-negative production system. The system was considered free of PRRSV on the basis of monthly testing of the sow farm (30 serum samples tested on the PRRS X3 Antibody ELISA; Idexx Laboratories Inc, Westbrook, Maine) and bimonthly testing of nursery and the finishing sites (oral-fluid samples tested by PRRSV reverse transcriptase-polymerase chain reaction [RT-PCR]).

Initially, four oral-fluid samples collected from a nursery site were tested by both PRRSV RT-PCR and the oral-fluid antibody ELISA. All four oral-fluid samples were PCR-negative, but three oral-fluid samples (Barns 2, 3, and 4) tested antibody-positive on the ELISA (sample-to-positive [S:P] ratios = 1.85, 1.95, and 1.80). These results prompted a series of PCR and ELISA re-tests in the laboratory, in which the original results were duplicated.

Simultaneously, new samples were collected, including 91 oral-fluid and 40 serum samples from the four barns on the nursery site and 120 sera from the sow farm. All serum samples were ELISA-negative and both serum and oral fluids were negative by PRRSV RT-PCR. In contrast, PRRSV oral-fluid antibody ELISA testing showed that all oral-fluid samples from Barn 1 (n = 23) were antibody-negative, whereas all oral-fluid samples from nursery Barns 2, 3, and 4 (n = 68) were antibody-positive. These results were confirmed by testing at a second diagnostic laboratory.

An explanation for oral-fluid antibody-positive results in a PRRSV-negative herd was suggested by Drs William Dubois and Scanlon Daniels. Specifically, they hypothesized that spray-dried porcine plasma incorporated into the nursery transition diet contained PRRSV antibody and that pigs consuming this ration contaminated the oral-fluid sample with PRRSV antibody from this exogenous source. If true, this hypothesis also explained the ELISA results from nursery Barn 1 (all negative) versus Barns 2, 3, and 4 (all positive).

To test the hypothesis, a feed sample representing the transition diet fed in nursery Barns 2, 3, and 4 was submitted for analysis. In the laboratory, 5 g of feed and 20 mL of PRRS X3 ELISA sample diluent were combined in a 50-mL centrifuge tube and thoroughly mixed. Using this mixture as a stock solution, two-fold serial dilutions (1:2 to 1:4096) were made using sample diluent and tested using the PRRSV oral-fluid antibody ELISA. As shown in the table, all sample dilutions through 1:64 (shaded) exceeded the PRRSV oral-fluid antibody ELISA S:P cutoff of = 0.40. Testing at a second diagnostic laboratory duplicated these results.

While this case was the first to document the presence of PRRSV antibody in feed products containing porcine spray-dried plasma, several similar cases have subsequently been identified. In each case, testing of the suspected source, eg, feed or milk replacer, revealed the presence of high levels of PRRSV antibody. Thus, producers and veterinarians using the PRRSV oral-fluid antibody ELISA to monitor herd status should be aware of exogenous sources of PRRSV antibody. Diagnosticians seeking to interpret seemingly contradictory diagnostic

Sample-to-positive (S:P) ratio response when feed samples containing porcine plasma were diluted and tested on a PRRSV oral-fluid antibody ELISA¹

Feed-sample dilution	S:P ratio
1:2	4.14
1:4	3.09
1:8	2.24
1:16	1.44
1:32	0.91
1:64	0.49
1:128	0.28
1:256	0.13
1:512	0.05
1:1024	0.02
1:2048	0.01
1:4096	0.00

results should be aware that PRRSV antibody in feedstuffs is easily detected by direct testing of the product in question.

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Reference

1. Kittawornrat A, Prickett J, Wang C, Panyasing Y, Ballagi A, Rice A, Main R, Johnson J, Rademacher C, Hoogland M, Rowland R, Zimmerman J. Detection of porcine reproductive and respiratory syndrome virus (PRRSV) antibodies in oral fluid specimens using a commercial PRRSV serum antibody enzyme-linked immunosorbent assay. *J Vet Diagn Invest*. 2012;24:262–269.