

Comparison of Milk BVD ELISA Results with Ear Notch Testing, between Milk Sample Handling Methods and with Cow Characteristics

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Introduction

The primary objective was to study a milk ELISA test for detection of antibody against p80 (p80/125 non-structural protein) of bovine viral diarrhea virus (BVDV) in comparison with standard ear notch testing for cows persistently infected (PI) with BVDV. BVDV is an important cause of respiratory disease, diarrhea, abortion and infertility, and poor calf performance in dairy herds.

Secondary objectives included evaluation of four methods of handling Dairy Herd Improvement Association (DHIA) milk meter-collected milk samples for possible differences in BVD test results, and to study cow characteristics, such as parity and stage of lactation, and their association with anti-BVD antibody detection.

Materials and Methods

Milk samples collected using a DHIA milk meter were obtained from a dairy herd with past diagnoses of PI cows and abortions caused by BVD. BVD MLV vaccine was administered to calves three months and four months old, and to all cows at dry-off and 15 to 21 days-in-milk (DIM). A total of 247 and 258 cows were tested one month apart using a competitive ELISA for milk antibody (Ab) binding to p80 protein of BVD virus.

Results are reported as percentage of binding by a second test kit anti-BVD Ab after the milk sample was placed into p80 BVD protein-coated test wells and then washed. Higher second Ab binding means the milk sample had less anti-BVD p80 Ab. Interpretation of binding percentages of the second Ab was: 90-100%, little anti-BVD Ab - PI or vaccine failure if consistent; 60-89%, moderately low anti-BVD Ab; 30-59%, moderate anti-BVD Ab; 10-29%, high anti-BVD Ab; 0-9%, very high anti-BVD Ab.

Four samples from each cow were handled differently: fresh milk, fresh with preservative pill, frozen for seven days without preservative, and room temperature for seven days with preservative pill. Ear notches were sampled concurrently from all cows for BVD antigen (Ag) testing.

Results

For all cows, means were: 305ME (age, season, fat corrected Mature Equivalent) milk production 29,625 lb (13,450 kg); daily milk 88 lb (40 kg); 173 DIM.

No PI-BVD cows were found from ear notch Ag tests of 345 cows, including all cows tested at least once using the milk test. Milk handling method was significantly associated with differences in milk BVD results. Second antibody binding means were as follows: fresh milk 49%^a; fresh with pill 45%^b; frozen seven days 45%^b; pill preserved seven days 42%^c (different letters $P \leq 0.01$, ANOVA, Tukey's).

All subsequent results shown here are from fresh milk. Binding ranged from 3%-98%, quartiles 29%, 47%, and 62% in the first month, and 35%, 56%, and 71% in the second month. Fifteen cows had 90-98% binding, but all had this result on only one of the consecutive monthly tests. Fourteen of these cows were milking each month and all were below 90% on the other test; their mean second Ab binding the other month was 60%. For the 15 cows > 90%, on that monthly test their DIM range was 41-188 (mean 106 DIM), 305ME mean was 28,404 lb (12,896 kg), and daily milk mean was 97 lb (44 kg). Therefore, no PI or vaccine failures (consistently > 90% binding) were found by milk ELISA.

DIM significantly affected second Ab binding: 1-9 DIM, 16%*; 10-30 DIM, 34%*; 31-60 DIM, 46%; 61-150 DIM, 60%*; 151-300 DIM, 47%; 301-360 DIM, 40%; >360 DIM, 46% (* = $P \leq 0.025$, ANOVA, Tukey's). Lactation number did not affect binding. Second Ab binding by lactation number was: first 47%; second 50%; third 52%; fourth 48%; and fifth through sixth was 51%.

Significance

Cows varied in quantitative results of anti-BVD Ab in milk (inversely related to binding of a second Ab) from very low to very high. Milk handling methods such as freezing and preservation pills affected BVD test results. The milk ELISA agreed with ear notch testing by finding no PI cows. Anti-BVD Ab was high in early lactation, and then decreased as DIM and the time since last vaccination against BVD increased. Some cows tested once for very low anti-BVD antibody, defined as PI or vaccine failure, but this was not repeatable. The ELISA should be evaluated further as a BVD milk test, including comparison to serum anti-BVD antibody measurement, and in cows diagnosed as PI animals by ear notch testing.