Research Summaries I

Evaluation of environmental culture to determine herd status for Johne's disease on Atlantic Canadian dairy farms

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Introduction

Environmental culture (EC) is a non-invasive and relatively inexpensive method to detect the presence of *Mycobacterium avium* subsp *paratuberculosis* (MAP) in a herd. Research into the test characteristics of EC has generally focused on large, MAP-positive herds. This study was designed to evaluate the test characteristics of EC within typical Atlantic Canadian dairy herds. These herds tend to be smaller in size than herds that have been used in previous EC studies. Furthermore, many of these herds were suspected to be test-negative or have low MAP prevalence.

Materials and Methods

A total of 32 purposively selected herds from the Maritime provinces participated in this two-year project. Median herd size was 66 milking cows (range, 30 to 220). Individual fecal samples were collected from all milking cows biannually. Environmental samples were collected quarterly following a protocol adapted from the USDA-APHIS Voluntary Bovine Johne's Disease Control Program. Two samples were collected from the manure storage areas and four samples were collected from the mature cow housing areas. All manure samples were processed by the Maritime Quality Milk Laboratory, Atlantic Veterinary College, Prince Edward Island, Canada. For comparison purposes, individual cow fecal cultures (grouped by cow age) were combined into pools of five. All samples were cultured via the ESP® Culture System II (TREK Diagnostic Systems, Inc., Cleveland, Ohio, USA). Fecal samples were prepared and cultured in accordance with the manufacturer's instructions. After culture, all broth samples were examined microscopically for the presence of MAP with an acid-fast stain. For all samples that tested positive for MAP via culture or microscopic visualization, confirmatory PCR for detection of IS900 was performed by use of the VetAlert™ Johne's Real-Time PCR kit (Tetracore, Inc., Rockville, MD, USA). Statistical analysis was performed via GEE logistic models with exchangeable correlation structures to account for the repeated measures.

Results

Results of null logistic GEE models indicated that the sensitivity (Se) of EC was 71.4% (95% confidence interval (CI), 49.2% to 86.5%) and specificity (Sp) was 98.6% (95% CI, 94.8% to 99.6%). Season of EC collection, herd management system and within-herd fecal culture prevalence (WHP) were entered into the null model for Se. Only WHP was found to be significantly associated with Se; as WHP increased, Se increased. For example, when WHP was 1%, the Se of EC was 32% (95% CI, 19.7% to 48.0%) and when WHP was 74%, the Se of EC increased to 98.1% (95% CI, 87.2% to 99.7%). Sensitivity and Sp of EC using stepwise combinations of 2, 3, 4, 5, and 6 EC results were derived from separate null logistic regression models. Although there was not a statistical difference between Se and Sp estimates for the various combinations of results (most likely because of large 95% CIs), the greatest numerical improvement in Se was seen when two sets of EC were evaluated in parallel, compared with the Se to one EC set. Estimated WHP increased as the proportion of positive individual EC samples increased. For example, when one of six individual EC was positive, the estimated WHP was 0.7% (95% CI, 0.44% to 1.2%) and when all six individual ECs were positive, the estimated WHP was increased to 12.7% (95% CI, 10.8% to 14.9%).

Significance

The Se and Sp estimates from this study indicated that environmental culture was an acceptable tool to screen Atlantic Canadian dairy herds for MAP. Sensitivity of EC improved as WHP increased. Additionally, the proportion of positive individual EC allowed for an estimation of the MAP WHP, which is useful information when designing a MAP control program for a herd.