

Detection of *Mycobacterium avium paratuberculosis* in Naturally Exposed Dairy Calves

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Introduction

Johne's disease (JD) is a chronic disease of ruminants caused by *Mycobacterium avium paratuberculosis* (MAP). It has generally been accepted that cattle become infected as calves by ingesting MAP shed by adult cows. Due to MAP's slow-growing nature, JD has a long incubation period with clinical signs not becoming evident in infected cattle for years. However, infected cattle often shed MAP before the onset of clinical signs, albeit intermittently and at low levels. With a recent study documenting horizontal transmission of MAP from calf to calf, identifying infected cattle in the early stages of JD has become even more important. Fecal culture (FC) on Herrold's egg yolk media (HEYM) is only 50% sensitive in identifying subclinically infected adult animals. Studies using an automated liquid culture system report improved test sensitivity, capable of identifying MAP at lower levels than culture on HEYM. The purpose of this study was to determine if fecal shedding of MAP could be detected in naturally exposed dairy calves using a liquid culture system (ESP II).

Materials and Methods

A prospective, multi-cross-sectional study was conducted over 28 months on heifers from eight Michigan dairy herds. Ten heifers from four age groups: 0-3 months, 4-6 months, 7-14 months, and 15-24 months, were selected from each herd and tested for MAP using ESP II FC. The age groups were defined based on distinct, naturally occurring management groups found on most dairies representing four sets of environmental risk factors. Heifers from dams testing positive for JD by FC or serum ELISA were preferentially sampled, with the remainder of each age cohort filled with randomly selected calves. Other data collected included the within-herd JD prevalence and herd size at the time of sample collection. To assess the effect of age, herd prevalence, and herd size on the FC status of the heifer, logistic regression was used, accounting for clustering of data within herds and repeated measures on individual calves using generalized estimating equations in a subject-specific model.

Results

A total of 1,842 fecal samples were collected from 1,203 heifers, and 393 heifers were tested at least twice. Of samples originating from 27 heifers, 36 (2%) were culture-positive for MAP. Seven heifers were FC positive on two different dates, and one heifer was FC positive three times. The numbers of FC positive samples in each age group were as follows: 0-3 months, 4; 4-7 months, 2; 7-14 months, 18; and 15-24 months, 12. All pair-wise comparisons between the four age groups revealed no statistically significant difference between age groups 0-3 months and 4-6 months, or between age groups 7-14 months and 15-24 months. Thus, age group was collapsed into a dichotomous variable (<6 and >6 months) for regression analysis. The final multivariable regression model assessing the factors associated with heifers being FC positive included: age >6 months (OR=6.0, 95% CI: 2.1-17.4), within-herd JD prevalence >10% (OR=4.7, 95% CI: 2.0-11.1), and herd size >300 cows (OR=5.7, 95% CI: 2.4-13.4).

Significance

MAP was identified in the feces of 2% of the dairy heifers tested prior to 24 months of age using ESP II culture. It is unknown whether the low proportion of MAP positive FC's is due to a low prevalence of MAP-infected heifers in the study, intermittent MAP shedding in young infected heifers, or suboptimal culture sensitivity. The majority of positive FC's originated from heifers >6 months of age. Also associated with heifers culturing positive was within-herd JD prevalence >10% and herd size >300 cows. Based on this study, if a producer with a relatively high within-herd JD prevalence desires to aggressively identify and remove MAP-infected cattle by FC, targeted testing of heifers >6 months old may be of some benefit. However, further study is warranted to determine if heifers shedding MAP in their feces at a young age perform differently or develop clinical JD earlier than non-shedding herdmates.