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Comparing ELISA test-positive prevalence, risk factors and management recommendations for Johne's disease prevention between organic and conventional dairy farms in Ontario, Canada

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ABSTRACT

Johne's disease (JD) is a chronic, infectious disease in cattle. Between 2010 and 2013, a voluntary JD control program was successfully launched in Ontario, Canada, including a Risk Assessment and Management Plan (RAMP) and JD ELISA testing of the entire milking herd. Over the last decade, the organic dairy sector has been growing. However, organic farming regulations and philosophies may influence the risk for JD transmission on Ontario organic dairy farms. The aim of this cross-sectional study was to investigate differences in JD ELISA test positive prevalence, risk factors for JD and recommendations for JD prevention between organic and conventional dairy herds in Ontario. RAMP results (i.e. RAMP scores and recommendations) and ELISA results were available for 2103 dairy herds, including 42 organic herds. If available, additional data on milk production, milk quality, and herd characteristics were gathered. Organic and conventional herds had a similar herd-level JD ELISA test-positive prevalence (26.2% and 27.2%, respectively). Organic herds (4.2%) had a higher within-herd JD ELISA test-positive prevalence compared to conventional herds (2.3%) if they had at least one JD test-positive animal on the farm. Organic farms had lower risk scores for biosecurity (9 points lower), and higher scores in the calving (7 points higher) and the calf-rearing management areas (4 points higher). After accounting for RAMP score, organic farms received fewer recommendations for the calving management area (Odds Ratio = 0.41) and more recommendations in the adult cow management area (Odds Ratio = 2.70). A zero-inflated negative binomial model was built with purchase of animals and the herd size included in the logistic portion of the model. Herd type (organic or conventional), colostrum and milk feeding practices, average bulk tank somatic cell count, and presence of non-Holstein breeds were included in the negative binomial portion of the model. Organic farms had a higher number of test positive animals (Count Ratio = 2.02). Further research is necessary to investigate the apparent disconnect between risk factors and recommendations on organic dairy farms.

1. Introduction

The organic industry is growing worldwide (Willer et al., 2008). In Canada, the production of organic milk increased from 100,000 hl in 2000 to 870,000 hl in 2009. Likewise, the number of farmers producing organic milk increased from 65 to 206 between 2000 and 2009 (Agriculture and Agri-Food Canada, 2009). Canadian Organic Standards (COS) were implemented in 2006 to regulate the organic dairy industry. Farmers can become certified as organic if they comply with the described management practices in the COS. Desirable management practices included in the standards focus on field

Abbreviations: BtSCC, average bulk tank somatic cell count; CI, confidence interval; COS, Canadian Organic Standards; CR, count ratio; HES, herd ELISA status; HP, high positive; ICC, intraclass correlation coefficient; IQR, interquartile range; JD, Johne's disease; Lntotnotest, logarithmic transformed number of cows tested; MAP, *Mycobacterium avium* ssp. *paratuberculosis*; OJEMAP, Ontario Johne's Education and Management Assistance Program; RAMP, Risk Assessment and Management Plan; ZINB, zero-inflated negative binomial.

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crop, manure, pasture, and livestock management (Government of Canada, 2006). While some organic management practices seem to be favorable for maintaining metabolic health and longevity of dairy cattle (Hardeng and Edge, 2001), the risks arising for infectious disease transmission are rarely investigated.

Johne's disease (JD) is a chronic, infectious disease of ruminants caused by *Mycobacterium avium* ssp. *paratuberculosis* (MAP). Subclinically infected cattle may show a drop in milk yield, while clinically affected animals develop profound diarrhea and emaciation. Estimates of the herd-level seroprevalence of JD in Canadian provinces range from 33 to 74% of herds (Tiwari et al., 2006). Routes of MAP transmission from adult cows to young calves include in utero transmission from dam to calf, and the ingestion of infectious feces, colostrum, or milk (McKenna et al., 2006). Therefore, it is commonly recommended to remove the calf from the adult cow environment quickly after birth and to provide it with low risk colostrum and milk replacer (Pieper et al., 2015a). This practice specifically is not consistent with organic farming practices, which encourages bonding between the calf and the dam through prolonged contact periods and suckling of colostrum and milk (Wagenaar and Langhout, 2007). While suckling is not a requirement within the COS, natural colostrum and milk has to be provided to the calves (Government of Canada, 2006). In emergency situations, milk replacer from organic sources may be used, but none is currently available in Canada from commercial sources.

Infection with MAP may remain undetected for years. Adult animals between 2.5 and 5.5 years of age have the highest probability of being identified as infected by in vivo diagnostic tests, either indirectly using antibodies against JD or directly based on fecal culture and/or PCR (Nielsen and Ersbøll, 2006). Furthermore, the probability of detecting JD infection increases with cow age (Jubb et al., 2004). Therefore, since the longevity of cows on organic farms is greater (Hardeng and Edge, 2001; Stiglbauer et al., 2013), there may be an increased opportunity for infected cows to contaminate the environment and transmit JD to the offspring or other calves.

The COS state that breeds and lines of cattle resistant to diseases should be used on the operation (Government of Canada, 2006). Crossbreeds have been shown to have better reproductive performance compared to purebred Holstein cows (Schaeffer et al., 2011). Therefore, organic farms may be more likely to keep minor breeds or crossbreeds. However, Sorge et al. (2011) showed that Channel Island breeds had greater odds of being JD positive compared to Holsteins. Consequently, organic farms might have a greater risk of having JD positive animals on their farm if they keep cows of Channel Island breeds.

To date, four studies have investigated JD prevalence on organic farms. Studies from the USA (Zwald et al., 2004; Sorge, 2014) suggested that the JD prevalence may be lower on organic than conventional farms (25.0% vs. 48.5% and 42.9% vs. 92.3% [large conventional herds], respectively). In Canada, Ramanantoanina et al. (2012) found a JD herd-level prevalence of 20.3% which they considered less than estimates from the literature. A study from The Netherlands found a similar herd-level (36% vs. 39%, respectively) and within-herd (1.2% vs. 1.7%, respectively) JD ELISA prevalence on organic and conventional farms (Kijlstra, 2005). Using a risk-based management assessment, this latter study also reported elevated risk scores in the calving and calf raising areas as well as for biosecurity, compared to conventional farms (Kijlstra, 2005). When considering the higher risk scores found by the aforementioned author, as well as the previously mentioned expectations of organic farming practices, it is surprising that these studies did not find a higher JD prevalence in the investigated populations of organic herds. However, three out of four studies did not consider herd size when comparing organic and conventional farms. Commonly, conventional farms are larger than organic farms (Stonehouse et al., 2001; Zwald et al., 2004) and larger farms are reported to have higher odds of having JD positive cows (Wells and Wagner, 2000; Muskens et al., 2003).

The objectives of this study were to compare (1) management practices associated with high risk for JD transmission, (2) farm-specific recommendations that were given for JD prevention, and (3) JD ELISA positive herd-level and within-herd prevalence between organic and conventional dairy herds participating in the Ontario Johne's Education and Management Assistance Program (OJEMAP).

2. Materials and methods

2.1. The Ontario Johne's Education and Management Assistance Program

In 2010, the Ontario dairy industry launched the voluntary OJEMAP to limit the effects of JD. Detailed information is provided on the program website (www.johnes.ca) or in Pieper et al. (2015a). Briefly, all Ontario dairy producers were encouraged to conduct an on-farm JD Risk Assessment and Management Plan (RAMP, Supplementary material) with their veterinarian. Between January 2010 and August 2013, dairy farmers were also eligible for a one-time milk or serum JD ELISA testing of the entire milking herd. Milk samples were analysed by CanWest Dairy Herd Improvement and serum samples by Animal Health Laboratory, University of Guelph, Canada. The PARACHEK milk ELISA (Prionics AG, Schlieren, Switzerland) had a sensitivity of 61.1% and a specificity of 94.7% compared to fecal culture and the IDEXX HerdChek *Mycobacterium paratuberculosis* serum ELISA (IDEXX Laboratories, Westbrook, Maine, USA) had a sensitivity of 73.6% and a specificity of 87.5% compared to fecal culture (Hendrick et al., 2005). The costs for these tests were covered by the program provided two additional requirements were met: (1) within 90 days of the test, the producers had to complete a JD RAMP with their herd veterinarian and (2) within 90 days after the test (or before the next calving, whichever occurred first) producers had to dispose of animals with a high positive JD ELISA test result (HP; sample-to-positive control ratio $\geq$1.0), while ensuring that those animals did not enter the human food chain or another dairy herd. The RAMP was a standardized one-page questionnaire administered by a veterinarian (supplementary material). Veterinarians received an on-farm training and were registered to do the RAMP. A handbook was provided to try to reach consistency among assessors. The RAMP was comprised of 5 sections: (1) general JD management (60 points), (2) calving area risk management (80 points), (3) pre-weaned calf area risk management (70 points), (4) post-weaned calf area risk management (40 points), and (5) adult cows risk management (50 points). The maximum score was 300 with increasing scores indicating increasing risk for JD transmission. The RAMP was found to be useful for developing a farm specific JD prevention strategy and in predicting the herd-level and within-herd JD ELISA test positive prevalence Pieper et al. (2015b).

2.2. Sample size justification

Since no Canadian data were available, data from research from the USA were used to estimate sample size. Based on a study by Zwald et al. (2004), a self-reported JD herd-level prevalence of 25% for organic and 49% for conventional dairy farms in the USA were considered for sample size calculation. The required sample sizes to detect this difference in a cross-sectional study was $n = 33$ for organic and $n = 1603$ for conventional dairy farms (confidence: 95%, power: 80%, 2-tailed test, ratio of sample sizes: 0.02).

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