Descriptive analysis of the changes in Johne’s disease management practices on Ontario dairy farms through repeat risk assessment

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Descriptive analysis of the changes in Johne’s disease management practices on-farm as a means to assess herd disease risk more accurately.

Key words: Johne’s disease, risk assessment, dairy

INTRODUCTION

Johne’s disease (JD) is a chronic progressive gastrointestinal disease of ruminants. An untreatable infection with a typical incubation period of 2 to 6 yr, JD is challenging in terms of both detection and control. In its clinical stage, it is characterized by weight loss with intermittent to persistent diarrhea. Sequelae to the clinical effects of JD are reduced milk production, early culling, and lower slaughter value. The estimated financial cost of JD is US$198 million for the American dairy industry and between US$17 and $28 million for their Canadian counterparts (Rasmussen et al., 2021). Control of JD is often based on the culling of positive animals and the adoption of management practices that minimize exposure of calves to the causative pathogen, Mycobacterium avium ssp. paratuberculosis (MAP; Collins et al., 2010).

From 2010 to 2013, the dairy industry in Ontario, Canada, instituted a voluntary JD control program called the Ontario Johne’s Education and Management Assistance Program (OJEMAP). The program consisted of whole-herd testing and a Risk Assessment and Management Plan (RAMP). The RAMP consisted of 30 questions that evaluated 5 different management areas (animal purchases, calving management, calf management, heifer and cow cleanliness, and heifer and cow management) to characterize herd risk for MAP introduction and transmission. The veterinarian-administered RAMP was then prompted to provide up to 3 recommendations for changes to management practices on-farm as a means to assess herd disease risk more accurately.

The RAMP focused on animal purchases, calving management, calf management, and heifer and cow cleanliness and management. In the summer of 2019, the RAMP was repeated on 180 farms that had participated in the JD program of 2010 to 2013 and had bulk tank milk ELISA results from 2013 and 2017. This cross-sectional study demonstrated that many producers changed management practices over the 4- to 7-year period. Producers changed their cattle buying practices, with a reduction in purchasing from multiple sources and more herds refraining from buying in animals. However, overall scores were higher in 2019 than in 2013. The 2019 RAMP indicated that fewer farms were utilizing individual calving pens in 2019 than in 2013 (13% vs. 26%), yet more farms had policies in place to deal with sick or suspect JD cows entering the maternity area (92% vs. 74%). Management changes occurred over time, some of which represent increased risk (crowded maternity pens) and others decreased risk (closed herd, protocols in place for JD-positive cows) for MAP introduction and transmission. These results highlight the importance of frequent risk assessments and the documentation of changes to management practices on-farm as a means to assess herd disease risk more accurately.
RAMP questions. A total of 4,168 herds were eligible for the OJEMAP program; 52% of these herds participated in the program and 26% of these had one or more test-positive cows. On an individual cow basis, through serum or milk ELISA, 139,893 animals were tested, with 1% (1,385) testing positive for JD (http://www.johnes.ca).

The RAMP produced a numerical score, with higher scores associated with a higher risk of JD transmission. A challenge to the utility of risk assessments as they pertain to management practices on a farm is that they are temporally dependent (Sorge et al., 2012). Temporal dependence means that the management characteristics recorded in the RAMP represent the state of the farm on that day; however, it is unclear how long these practices have been in place or how long they will remain unchanged. The slow progression of JD further complicates this matter in that changes in management practices will not result in changes to subclinical and clinical prevalence until months or years later (Stabel, 1998; Tiwari et al., 2006). It is not uncommon for certain practices to evolve over time due to shifts in farm personnel or changes in physical facilities, or to accommodate changing market conditions. For instance, producers may have purchased cattle to accommodate the increased quota allocation in the Canadian dairy market during this period, but the consequences of bringing those animals into the herd may not be recognized until months or years later.

In general, diagnostic testing for JD is performed with individual or pooled samples. Assessing changes to within-herd prevalence is best achieved with whole-herd testing of individual animals (van Schaik et al., 2007). Although this method of testing has better sensitivity in defining positive herds, its use can be limited by financial or labor constraints (Tiwari et al., 2006; Nielsen, 2014). To reduce cost and labor, pooled testing methods such as bulk tank milk ELISA and quantitative PCR and environmental cultures have been used to assess herd-level prevalence (Christensen and Gardner, 2000; Kalis et al., 2000; Bauman et al., 2019).

The objective of this study was to describe the management changes that pertain to JD control and the corresponding changes in bulk tank milk ELISA status that have occurred on a sample of Ontario dairy farms between the original program (2010–2013) and 2019 using a cross-sectional study approach. We hypothesized that changes have occurred in farm management practices since the original RAMP were completed and that some of these changes likely represent a change in risk for JD transmission. With the implementation of the OJEMAP program and the corresponding management changes intended to reduce JD transmission, herd-level JD prevalence, as estimated using bulk tank (BT) testing, would be expected to decrease.

**MATERIALS AND METHODS**

**Bulk Tank Milk Sampling and Testing**

Bulk tank milk (BTM) samples from all Ontario dairy farms (n = 3,909) were collected in 2013, at the end of the voluntary JD program, and were tested using a modified ELISA technique described by Wilson et al. (2010) using a commercial *Mycobacterium paratuberculosis* antibody test kit (IDEXX). In 2017, a follow-up set of BTM samples from all Ontario dairy farms (n = 3,581) were tested using the same ELISA test and protocol. The samples of BTM were classified as positive if they exceeded the cutoff value of 0.089 corrected optical density (OD), based on work by Innes (2011). The samples used in this study were commingled bulk tank samples collected by milk transporter/processors. The study did not involve any direct contact with animals; thus, no Animal Utilization Protocol was required.

**Classification of BTM Tests**

As the ELISA corrected OD is measured on a continuous scale, results near the proposed cut-point of 0.089 could change to the opposite (dichotomous) result just by slight variation on the test-day (Collins et al., 1993). With fluctuations in antibody production and in which cows are contributing to BT samples and how much milk they are contributing, defining samples as positive and negative may be inappropriate (Slana et al., 2008; Nielsen and Toft, 2014; Beaver et al., 2017). Low-risk and high-risk samples are more relevant to the discussion; a “negative” sample may just indicate a low risk of having infected cows but with the recognition that this risk was not zero. Thus, a positive BT ELISA result was considered a high-risk JD (HRJD) status herd (high risk of having prevalent MAP infection on the farm) and a negative BT ELISA was considered to represent a low-risk JD (LRJD) status. For each bulk tank test, each farm was classified LRJD or HRJD; thus, each farm had 2 risk classifications, one for their 2013 BT test and another for 2017.

**Initial Risk Assessment**

Between January 1, 2010, and October 31, 2013, trained herd veterinarians completed RAMP on client farms as part of the OJEMAP program. The results...
of the RAMP were transcribed and stored in an Excel spreadsheet (Microsoft Corp.).

**Follow-Up Risk Assessment: Study Farm Selection and Recruitment**

To be included in the sampling timeframe for follow-up RAMP, farms had to have both 2013 and 2017 BTM milk tests, have participated in the original OJEMAP program with a completed RAMP, and be located in southwestern or eastern Ontario. Exclusion based on geographical location was determined based on the logistics of having members of the research team able to visit the farm location. Of the Ontario dairy herds that were producing and shipping milk, 3,207 had BT tested for JD in both 2013 and 2017. With the paired samples, we were able to subdivide these herds into those that maintained a LRJD BT (604; 18.8%), those that changed from LRJD to a HRJD BT (1,100; 34.3%), those that tested as HRJD on both occasions (1,184; 36.9%), and those that changed from HRJD to LRJD (319; 10%) (Table 1). A subset of farms with paired BT results had participated in the original OJEMAP (n = 1,828). Of these 1,828 farms, 113 were excluded due to the absence of a risk assessment from 2013, and 112 farms were excluded due to geographical location. Another 916 farms were excluded due to one or both BT test results being near the 0.089 corrected OD cutoff (results between 0.07 and 0.14), leaving 687 farms to be included in the sampling frame (Figure 1). As stated above, it is possible to misclassify herd status when the ELISA corrected OD is close to the cutoff value. Therefore, the rationale for selecting herds that were on the more extreme ends of the spectrum was to minimize the misclassification of BTM results. In 2019, we planned to conduct a follow-up RAMP on a subset of farms (n = 180) from the 687 farms in the sampling frame. We used farm BTM ELISA results to guide sampling to capture a representative sample of different JD experiences. Equal numbers of farms (n = 50) from the 4 categories described above were randomly selected, using a random number generator, for a phone contact list. New farms were added to this list when the initial list of producers had been invited and either did not respond or declined involvement. Recruitment ended after 180 farms had consented to participate in the follow-up RAMP. Due to the lack of clear sample size calculation for multiple choice survey studies, purposive sampling was used with the goal of acquiring a meaningful representation of farms. The distribution of farms that had follow-up risk assessments conducted can be found in Table 1.

**Follow-Up Risk Assessment: Data Collection**

Follow-up RAMP were completed on-farm between June 18, 2019, and January 22, 2020, by a team of 3 researchers trained by the developer of the original RAMP. The interpretation of management practices and corresponding scores were outlined in the RAMP user guide (http://www.johnes.ca/pdf%20files/RAMP%20user%20guide.pdf), which had not been changed since the original RAMP were completed. Section 1 of the RAMP (5 questions on cattle purchasing behavior, number of farms from which animals were purchased, what type of animals were bought, and disease status before purchase) had a possible total score of 60. Section 2 of the RAMP (8 questions that evaluated the management of calving and the maternity pen) had a total possible score of 80. Section 3 (7 questions on cattle purchasing behavior, number of farms from which animals were purchased, what type of animals were bought, and disease status before purchase) had a possible total score of 60. Section 2 of the RAMP (8 questions that evaluated the management of calving and the maternity pen) had a total possible score of 80. Section 3 (7 questions on management and weaning of calves) had a total possible score of 70. Section 4 and section 5 evaluated the cleanliness and management of weaned and bred heifers, and cleanliness and environmental hygiene of dry and milking cows, respectively, with total possible scores of 40 and 50, respectively. Each of the individual questions within these 5 sections were assigned numerical scores, with the highest number corresponding to the management practice associated with the highest risk of JD transmission. The entire RAMP had a total possible score of 300 (http://www.johnes.ca/pdf%20files/Programs-CattleHealthDeclaration.pdf).

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**Table 1. Risk status of bulk tank (BT) milk samples from Ontario dairy farms with tests from both 2013 and 2017**

<table>
<thead>
<tr>
<th>Herd status1 (2013, 2017)</th>
<th>All paired BT (%)</th>
<th>Participants in OJEMAP2 (%)</th>
<th>Eligible for follow-up (%)</th>
<th>Included in follow-up RAMP2 (%)</th>
<th>Indigible for follow-up (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Low, low (LRJD, LRJD)</td>
<td>604 (18.8)</td>
<td>352 (19.3)</td>
<td>193 (28.1)</td>
<td>44 (24.4)</td>
<td>411 (16.3)</td>
</tr>
<tr>
<td>Low, high (LRJD, HRJD)</td>
<td>1,100 (34.3)</td>
<td>662 (36.2)</td>
<td>231 (33.6)</td>
<td>61 (33.9)</td>
<td>869 (34.5)</td>
</tr>
<tr>
<td>High, high (HRJD, HRJD)</td>
<td>1,184 (36.9)</td>
<td>632 (34.6)</td>
<td>212 (30.9)</td>
<td>57 (31.7)</td>
<td>972 (38.6)</td>
</tr>
<tr>
<td>High, low (HRJD, LRJD)</td>
<td>319 (10.0)</td>
<td>182 (9.9)</td>
<td>51 (7.4)</td>
<td>18 (10.0)</td>
<td>268 (10.6)</td>
</tr>
</tbody>
</table>

1BT samples that had >0.089 corrected optical density were defined as positive or high-risk Johne’s disease (HRJD); those with ≤0.089 corrected optical density were defined as negative or low-risk Johne’s disease (LRJD).

2OJEMAP = Ontario Johne’s Education and Management Assistance Program; RAMP = Risk Assessment and Management Plan.
The 2019 RAMP scores were entered into an electronic database, which was then exported and stored in Excel (Microsoft Corp.) for summary and analysis. For each herd in our cross-sectional study, the initial 2013 RAMP results were combined with the responses in the follow-up RAMP data set for comparison and descriptive analysis. Although scores were numeric, they were based on question scoring that was ordinal in nature (e.g., 1, 4, 7, and 10) and therefore nonparametric tests were used. Wilcoxon signed rank tests were performed to determine the significance (P < 0.05) of changes between test scores of the 2 RAMP. McNemar’s χ² tests were used to compare the questions between the 2 RAMP periods (P < 0.05) for section 1; a Yates correction was used for those contingency tables with cells that contained a value <5. For questions with more than 2 possible answers, a McNemar-Bowker global test of symmetry (P < 0.05) was used in place of McNemar’s χ² test. The nature of some of the questions within section 1 resulted in unequal denominators between the 2 RAMP periods, and thus a nonparametric repeated-measure ANOVA, Brunner improved Friedman test, was used to compare the questions between the 2 periods (P < 0.05). Because some questions consisted of 4 ordinal levels of scoring (1, 4, 7, and 10), the Brunner improved Friedman test, was also used to compare questions between 2 RAMP periods for sections 2, 3, 4, and 5 (P < 0.05). Herd demographics, production levels, farm layout, and cattle movement details were retrieved and reviewed through milk recording system for those herds enrolled in the milk recording program (Canwest DHI). Changes in average BT ELISA corrected OD and herd demographics were compared using a 2-tailed paired t-test (P < 0.05).

RESULTS AND DISCUSSION

BTM Sampling and Testing

Bulk tanks from 3,909 and 3,581 herds were tested in 2013 and 2017, respectively. In 2013, 46.8% (1,828/3,909) of bulk tanks were classified as HRJD compared with 71.4% (2,557/3,581) of HRJD bulk tanks in 2017, using the 0.089 corrected OD cut-point (Table 2). A total of 3,207 herds had BTM tested during both time periods. In 2013, 46.8% of these herds (1,503/3,207) had an HRJD BT result, whereas in 2017, 71.2% of the same 3,207 were HRJD. Of these herds with paired BT tests, a relatively small proportion (10.0%) were able to achieve successful JD control (i.e., HRJD to LRJD BT results), whereas 34.3% went from LRJD to HRJD BT results. The proportions of the OJEMAP farms as well as the farms in our final sampling frame and the 180 farms in our study can be found in Table 1.

From the Ontario dairy farms that had bulk tanks tested for JD, there was a dramatic increase from 2013 to 2017 of those with a BT result >0.089, an increase of 24.6% (Table 2) over the span of 4 years. A 24.6% increase from 2013 in BT classified as high risk could be due to increased introduction of MAP into Ontario dairy herds. Introducing JD into a herd would primarily be through cattle additions. New subclinically infected cattle may have been purchased by herd owners to accommodate the increases in milk that farms were able to ship. Between the 2 BTM tests, the available quota for Ontario dairy producers increased by 14.28 million kilograms (Agriculture Canada, 2021b). An alternative explanation could be due to the slowly progressive nature of JD. Some herds that were low risk in 2013 may still have had subclinically infected cattle present within these herds that began to progress and start shedding antibodies, resulting in their becoming HRJD in 2017.
The true cause of the increase likely incorporates both scenarios. Some of this apparent increase may also be attributed to poor test sensitivity of the BTM ELISA. Although whole-herd individual animal Johne’s disease testing is the most sensitive method of determining herd status, it can be quite costly. The cost per test can range from $8 to $15 per animal, in addition to the cost of sample collection and transport to the laboratory. Efforts to reduce the financial burden of testing have centered around pooled sampling techniques (e.g., pooled fecal samples, environmental testing, and BTM testing; van Schaik et al., 2007; Bauman et al., 2019). Apart from lower cost, pooled sampling techniques have the benefit of easier, more convenient sampling. Although it offers a more cost- and time-efficient mode of testing, pooled sampling does have challenges.

One challenge with this approach is the interpretation of the BTM ELISA test, which gives a continuous numerical result and is determined “positive” or “negative” based on a predetermined cut-point. Sensitivity analysis and receiver operator characteristic (ROC) curves have been used to estimate the ideal cut-point to determine positivity, but challenges rooted in the complexity of the disease and the causal pathogen remain (Nielsen and Toft, 2014). This complexity is amplified by the additional layer of pooled (BT) testing. Milk from many negative animals included with a few positive animals may dilute the sample below the detectable threshold of the test.

Historically, the use of BTM in ELISA tests has been met with limited success due to poor sensitivity. Recent studies have used a modified ELISA technique first described by Wilson et al. (2010) to increase the test sensitivity. In a 2011 study, the sensitivity was estimated to be 54.5% (specificity = 90.6%) when using a cutoff value of 0.1 OD, and herds with ≥1 test-positive cows were classified as positive. This sensitivity increased to 63.3% (specificity = 84.2%) when ≥2 positive cows classified a positive herd (Innes, 2011).

The testing laboratory defined positive results as a corrected OD ≥0.1, with negative results <0.1. Within our analysis of the modified BT ELISA results, we used 0.089 as the cutoff value to define a HRJD BT result. This would, in theory, maximize the likelihood of accurately detecting positive herds through BTM (by decreasing the false-negative fraction) at the expense of increasing false positives. Although a potential increase in false-positive results is not ideal, at this time the consequences of such a result do not pose any appreciable risk. This may change if restrictions or requirements are placed on farms that test positive for JD.

Follow-Up Risk Assessment

In our study of 180 farms that underwent repeat RAMP in 2019, 129 herds were enrolled in DHI in the first testing period (2013) and 125 of those were still in the DHI program in the second testing period. The average herd size for the 129 herds in the original RAMP period (2010–2013) was 87 and their average milk production was 30.7 kg/cow per day. The average herd size for the 125 herds still enrolled in DHI during the follow-up risk assessment period (2019–2020) was 106, with an average milk production of 34.4 kg/cow per day.

The average time between risk assessments for an individual farm was 7 yr (range: 5–9 yr). The overall average total score of the RAMP for our 180 study herds increased from 126.0 in 2013 to 144.4 in 2019 ($P < 0.001$). Among the study herds, 125 herds (69.4%) had a higher score on their follow-up RAMP with an average increase of 43, 53 herds (29.4%) decreased their score with an average decrease of 26, and 2 (1.1%) had the same score.

Based on comparisons of scores in section 1 between 2013 and 2019, the cattle purchasing behavior of the 180 study farms changed over the 7-yr period. The overall average section 1 score decreased from 33.8 in 2013 to 27.5 in 2019 ($P = 0.002$), representing a decrease in JD introduction and transmission risk. There was a 10-percentage-point reduction ($P = 0.027$) in the number of herds buying cattle over the period between the 2 risk assessments (71.6% in 2013 vs 61.6%...
in 2019; Table 3). Fewer herds were purchasing from multiple herds, either purchasing from single herds or not buying cattle ($P = 0.009$). More producers were electing to purchase younger animals, 18.9% (21/111) versus 12.4% (16/129), although this change was not significant ($P = 0.155$; Table 3). There were 12 occurrences where section 1, question 1 of the RAMP was scored as 0 (“have not purchased cattle in the past 5 years”) despite evidence of cattle movement onto the farm based on data obtained through the farms’ DHI records. Of the 129 herds enrolled in DHI, 58.9% (76/129) had made cattle additions since 2013; 46.0% of these farms (35/76) used only Ontario or Canadian sources, 25.0% (19/76) had only brought cattle in from the United States, and 29.0% (22/76) used both foreign and domestic sources of cattle.

Contrary to our original expectations, fewer herds reported purchasing animals within the last 5 yr. This finding is possible evidence that the education component of the Johne’s control program was effective to some degree, as reported by Roche et al. (2015), who investigated the use of participatory learning models and their effects on producer management changes. It is also possible that producers provided answers biased toward what they believed was socially desirable. For example, producers bought cattle despite knowing that the advice was to keep a closed herd and felt uncomfortable answering truthfully.

Those producers that continued to purchase replacement animals seemed to have incorporated actions to reduce their risk through purchase (buying from one instead of multiple herds, inquiring on disease status for incoming cattle, buying younger animals). Bringing animals in from outside sources has been found to be a predictor of herd positivity (Sorge et al., 2012; Wolf et al., 2016). A systematic review by Rangel et al. (2015) supports the idea that the purchase of animals from outside sources increases risk of JD introduction and transmission. Increasing the number of sources from which cattle are purchased would increase this risk. Although purchasing cattle still represents risk, buying from only one source may help reduce this risk. The current belief is that young animals are at a lower risk of spreading MAP. However, this belief must be balanced with the knowledge of the environment in which these young animals are raised. As Weber et al. (2010) demonstrated, young animals can also shed the bacterium if exposed to a high dose early in life. All the above noted changes represent positive changes toward the reduction of JD risk on-farm. It is not clear from the follow-up risk assessments whether these changes were inspired by previous veterinary recommendations or not. However, as Pieper et al. (2015) found in their analysis of the 2010–2013 OJEMAP program, the most common recommendation was “Do not purchase more cows, minimize purchases, or buy from low-risk herds you know.”

Overall, in 2019, more producers decreased their risk ($n = 55$) from their 2013 assessment of cattle purchasing behavior than increased their JD risk ($n = 30$); 95 producers remained at the same level of risk.

### Table 3. Section 1: cattle addition risk assessment results from original and follow-up risk assessments from 180 Ontario farms

<table>
<thead>
<tr>
<th>Section 1 question</th>
<th>2013 response, number (%)</th>
<th>2019 response, number (%)</th>
<th>$P$-value$^2$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Have you purchased cattle in the last 5 yr?</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes, from one herd</td>
<td>51 (28.3)</td>
<td>69 (38.3)</td>
<td>0.009</td>
</tr>
<tr>
<td>Yes, from multiple herds</td>
<td>21 (11.7)</td>
<td>24 (13.3)</td>
<td></td>
</tr>
<tr>
<td>No</td>
<td>108 (60.0)</td>
<td>87 (48.4)</td>
<td></td>
</tr>
<tr>
<td>Herds purchased:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cows + heifers + bulls</td>
<td>8/129 (6.2)</td>
<td>3/111 (2.7)</td>
<td>0.415</td>
</tr>
<tr>
<td>Cows + heifers</td>
<td>39/129 (30.2)</td>
<td>28/111 (25.2)</td>
<td>0.125</td>
</tr>
<tr>
<td>Cows + bulls</td>
<td>8/129 (6.2)</td>
<td>2/111 (1.8)</td>
<td>0.317</td>
</tr>
<tr>
<td>Heifers + bulls</td>
<td>3/129 (2.3)</td>
<td>0/111</td>
<td>0.155</td>
</tr>
<tr>
<td>Just cows</td>
<td>51/129 (39.5)</td>
<td>55/111 (49.5)</td>
<td>0.125</td>
</tr>
<tr>
<td>Just heifers</td>
<td>16/129 (12.4)</td>
<td>21/111 (18.9)</td>
<td>0.155</td>
</tr>
<tr>
<td>Just bulls</td>
<td>3/129 (2.3)</td>
<td>2/111 (1.8)</td>
<td>0.317</td>
</tr>
</tbody>
</table>

1Values in bold font represent responses that increased in frequency for their question from the recorded 2013 risk assessments.

2Brunner’s improved Friedman test.

3In 2013, one herd was recorded to have been purchasing cattle but did not indicate the demographics of the animals entering the herd; this farm was recorded as buying from multiple herds and not asking about disease status before purchase.
chasing behavior, 32 transitioned from buying cattle from multiple herds to closing their herds to incoming animals; 13 producers changed from purchasing their cattle from multiple herds to using only one source; and the remaining 10 producers changed from purchasing cattle from a single source to becoming a closed herd. All of the changes in purchasing behavior were assessed using a McNemar-Bowker global test of symmetry \((P = 0.058)\).

An important factor in these risk assessments is that they function as a snapshot in time, and these specific RAMP assess the animal purchases within the prior 5 yr. There is evidence that some of these producers who had responded as not purchasing cattle had records of cows from other Ontario sources and the United States entering their herd. It is possible that these herds purchased these cows before the 5-yr window assessed in the RAMP. This indicates that there are limitations in the application of these risk assessments and provides further support that, in order to gather accurate and representative data, more frequent assessments, possibly yearly, should be performed.

Within section 2 (calving and maternity pen management) of the RAMP, there was an increase in the number of farms calving multiple cows in the calving area; that is, farms scoring 10 (37.8% of farms in 2013 vs. 61.1% of farms in 2019; Table 4). More farms reported keeping sick and lame cows out of the maternity pen, scoring 0 (38.0% in 2013 vs. 46.7% in 2019), and more producers were keeping calves with their dam for longer than 30 min, scoring 10 (35.6% in 2013 vs. 60.0% in 2019; Table 4). Section 2 saw a modest increase in average section score, from 34.4 in 2013 to 39.4 in 2019 \((P < 0.001)\).

Some of the changes in response to the questions in section 2 of the RAMP represent an increase in JD risk (multiple cows in the same calving area, calves left with dam for 30 min or more). These changes would account for the increase in average overall section score (representing the overall risk) from 2010 to 2013 (34.4 in 2013 vs. 39.5 in 2019). Research by Künzler et al. (2014) and Pithua et al. (2013) found that the use of group calving pens increased the risk of JD compared with individual calving pens. Although there has yet to be sufficient research indicating the optimal time to remove calves from the dam, it stands to reason that limiting this highly susceptible age group to potential MAP exposure should be strongly considered for JD risk reduction. It is unclear what may have initiated this change, but we hypothesize that higher stocking density along with better reproduction could have functioned as a catalyst for these changes. Although it is also possible that increasing herd sizes resulting in a decreased amount of time for individual cows within

<table>
<thead>
<tr>
<th>Table 4. Section 2: calving area risk assessment scores from original and follow-up risk assessments from 180 farms; each point level (1, 4, 7, and 10) represents risk level pertaining to Johne’s disease (JD) for each question.</th>
<th>2013 response, number (%)</th>
<th>2019 response, number (%)</th>
<th>(P)-value</th>
<th>(\chi^2)</th>
</tr>
</thead>
</table>
| **Single or multiple cows in calving area?** | 68 (35.6) | 88 (48.9) | 37 (20.5) | 12 (6.7) | 46 (25.6) | 83 (46.1) | 35 (19.4) | 17 (9.4) | <0.001 | 110
| **Manure build up, risk for calf exposure?** | 43 (23.3) | 88 (48.9) | 37 (20.5) | 12 (6.7) | 42 (23.3) | 85 (47.2) | 36 (19.9) | 10 (5.5) | 0.584 | 18
| **Manure soiled udders and legs of cows?** | 59 (32.8) | 94 (52.2) | 25 (13.9) | 2 (1.1) | 46 (25.6) | 88 (47.2) | 32 (17.7) | 0 (0) | 0.005 | 28
| **Calving area used for sick of lame cows?** | 68 (35.6) | 72 (40.0) | 26 (14.5) | 13 (7.3) | 46 (25.6) | 84 (46.1) | 32 (17.7) | 10 (5.5) | 0.013 | 84
| **Calving area used by JD clinical or test-positive cows?** | 80 (44.4) | 72 (40.0) | 26 (14.5) | 13 (7.3) | 84 (46.1) | 32 (17.7) | 10 (5.5) | 0 (0) | <0.001 | 108
| **Birth of calves in areas other than designated calving area?** | 75 (41.1) | 72 (40.0) | 26 (14.5) | 13 (7.3) | 105 (57.8) | 47 (26.1) | 28 (15.0) | 10 (5.5) | <0.001 | 110
| **Likelihood of calves nursing cow(s)?** | 100 (55.6) | 72 (40.0) | 26 (14.5) | 13 (7.3) | 134 (73.4) | 47 (26.1) | 28 (15.0) | 10 (5.5) | <0.001 | 110
| **Length of time calves stay with cow(s)?** | 75 (41.1) | 72 (40.0) | 26 (14.5) | 13 (7.3) | 134 (73.4) | 47 (26.1) | 28 (15.0) | 10 (5.5) | <0.001 | 110

1 Values in bold font represent responses that have increased in frequency for their question from the recorded 2013 risk assessments.

2 Brunner improved Friedman test was used to assess for significant difference between the distribution of answers given in 2013 and those given in 2019.

3 Four missing values from 2019 risk assessments.

4 One missing value from 2013 risk assessments.

5 Trimmed mean of Friedman test used to assess for significant difference between the distribution of answers given in 2013 risk assessments.

6 Missing values from 2013 risk assessments.
the pen could explain this, herd size remains relatively constant under Canada’s supply management system.

Positive changes—those that reduce JD risk—were also reported, with fewer producers stating they would move sick and lame cows into the maternity area, and more herds had calving protocols in place if JD-positive animals were present (Table 4). Both results may indicate that producers have received the message of keeping sick animals away from the most susceptible (young calves). Actions targeting the protection of calf health are a significant action in the prevention of JD as well as other contagious diseases. Sorge et al. (2010) found that in a voluntary risk assessment-based JD control program that took place across 5 Canadian provinces, “Do not calve JD-positive cows in the same pen as JD-negative cows” was one of the common recommendations given by veterinarians, whereas the most common recommendations for JD control surrounded the prompt removal of calves from their dams and the feeding of low-risk colostrum; these were also common recommendations in the OJEMAP program.

Average section scores for section 3 (preweaning heifer management) of the RAMP increased from 20.1 in 2013 to 28.3 in 2019 ($P < 0.001$). More producers reported feeding more colostrum to calves, with 71.0% (vs. 58.3% in 2013) stating that they were feeding 3 to 4 L within the first 6 h, scoring 0. There was a large shift toward herds classified as feeding low-risk or artificial colostrum. More herds fed calves colostrum from only their own dam (31.1% in 2013 vs. 60.6% in 2019) versus feeding colostrum from other cows (pooled or frozen). However, fewer herds were scored at the lowest risk level: “All calves receive fresh colostrum only from their own test negative mother, or from a single low risk (selected because test negative or young) “donor” cow or are fed artificial colostrum” (57.8% in 2013 vs. 5.6% in 2019). The proportion of producers feeding nonsaleable milk (score of 10) to their calves had increased from 20% in 2013 to 30% in 2019. The proportion of producers feeding BTM (score of 10) also increased from the time of the original RAMP (18.3% in 2013 vs. 52% in 2019; Table 5). Group housing of calves also increased since the original RAMP, with 35.2% of producers reporting housing calves in groups of 9 or fewer (score of 7) in 2019, up from 15.0% in 2013.

Despite the increase in the average section 3 score, which represents an increase in JD transmission risk, many practices represented positive changes for reducing JD transmission risk, such as feeding more colostrum and feeding calves colostrum from their own mother. The movement away from feeding calves pooled colostrum is likely to reduce JD risk; Nielsen et al. (2008) identify pooling of colostrum as a potential risk factor in JD transmission, with the risk that the batch of colostrum is contaminated with MAP increasing with the number of cows that contribute. The large shift away from producers feeding the lowest risk colostrum (from test-negative cows or artificial colostrum) may be a consequence of fewer farms still participating in ongoing testing and thus not knowing which cows were actually test negative. With respect to milk feeding, more herds were feeding calves BTM, which potentially increases risk of JD. The feeding of pooled milk or BTM as well as waste or treated milk has tradition-

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### Table 5. Section 3: preweaning heifer risk assessment scores from original and follow-up risk assessments from 180 farms; each point level (1, 4, 7, and 10) represents risk level pertaining to Johnne’s for each question

<table>
<thead>
<tr>
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</tr>
</thead>
<tbody>
<tr>
<td>Are calves fed low-risk cow or artificial colostrum?</td>
<td>104 (57.8) 56 (31.1) 13 (7.2) 7 (3.9)</td>
<td>10 (5.6) 109 (60.6) 52 (28.9) 9 (5.0)</td>
<td>0</td>
<td>0</td>
<td></td>
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</tr>
<tr>
<td>Are 3 to 4 L of colostrum consumed in the first 6 h?</td>
<td>105 (58.3) 53 (29.4) 11 (6.1) 11 (6.1)</td>
<td>128 (71.1) 45 (25.0) 5 (2.8) 2 (1.1)</td>
<td>0.001</td>
<td>0.001</td>
<td></td>
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</tr>
<tr>
<td>Are calves fed low-risk whole milk or milk replacer?</td>
<td>95 (52.8) 37 (20.6) 15 (8.3) 33 (18.3)</td>
<td>59 (33) 14 (7.8) 13 (7.2) 93 (52.0)</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td></td>
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<tr>
<td>How often is nonsaleable milk (high risk) fed?</td>
<td>74 (41.1) 29 (16.1) 41 (22.8) 36 (20.0)</td>
<td>97 (53.9) 15 (8.3) 14 (7.8) 54 (30.0)</td>
<td>0.257</td>
<td>0.257</td>
<td></td>
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</tr>
<tr>
<td>Are calves housed in individual or group pens?</td>
<td>109 (60.6) 33 (18.3) 27 (15.0) 11 (6.1)</td>
<td>75 (41.9) 25 (14.0) 64 (35.2) 16 (8.9)</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Exposure to cow manure in calf housing area?</td>
<td>149 (82.8) 20 (11.1) 8 (4.4) 3 (1.7)</td>
<td>98 (54.4) 27 (15.0) 48 (26.7) 7 (3.9)</td>
<td>&lt;0.001</td>
<td>&lt;0.001</td>
<td></td>
<td></td>
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<tr>
<td>Exposure to cow manure by watering or feeding utensils?</td>
<td>161 (89.4) 16 (8.9) 2 (1.1) 1 (0.6)</td>
<td>95 (52.8) 65 (36.1) 17 (9.4) 3 (1.7)</td>
<td>0</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>

1Values in bold font represent responses that have increased in frequency for their question from the recorded 2013 risk assessments.
2Brunner improved Friedman test was used to assess for significant difference between the distribution of answers given in 2013 and those given in 2019.
3One missing value from 2019 risk assessments.
ally been considered a risk factor in the transmission of JD. However, more recent evidence demonstrates that it may not have as large an effect on JD prevalence as previously thought (Nielsen et al., 2008; Doré et al., 2012). The increased feeding of BTM could be due to better reproduction and animal retention, allowing for excess milk to be made, or to the high price sometimes associated with feeding milk replacer. In their study on the use of milk replacers on dairy farms, Heinrichs et al. (1995) found that larger farms were more likely to feed waste milk. Although our sample set did include some small farms, the average herd size was 87 for the original OJEMAP RAMP and 106 for the follow-up RAMP. Recommendations for the ad libitum feeding of whole milk may be a possible explanation for this change (Vasseur et al., 2010). Also, more calves were being housed in groups rather than individually (Table 5). Some producers have found group housing of calves to be easier to manage than individual hutches. There has also been increased interest from welfare and behavior researchers to consider the effects of isolation of calves during their growth and development (Barkema et al., 2015; Costa et al., 2016).

The largest change in average section scores was seen in section 4 (weaned and bred heifer management). The average section 4 score in 2013 was 15.3 (out of a possible 40) versus 24.4 in 2019 ($P < 0.001$). A higher proportion of producers were housing their bred heifers closer to their mature cows (score of 10; 67.2% vs. 20%; Table 6). In general, the cleanliness of weaned and bred heifers decreased, corresponding to higher scores in the follow-up RAMP. Whereas in 2013, relatively few farms scored 10 (questions scored out of 10) in their weaned heifers (6.1% of farms in 2013), in 2019 27% of farms scored 10 on their weaned heifer cleanliness: “If manure is present above the hocks/knees AND/OR is present on the flanks.” Similarly, bred heifer cleanliness decreased, with 8.9% of farms scoring 10 in 2013 versus 24% in 2019 (Table 6).

Decreased cleanliness and increased exposure to mature cows represent increased risks in JD transmission. The changes in score may indicate that many producers believe that these animals are at lower risk of JD infection, and the priority for cleanliness and limiting their exposure is lower than that of young calves. This has been the typical belief for JD and JD control practices; however, there is growing evidence that older animals are also susceptible to infection if exposed to a high enough dose of MAP (Windsor and Whittington, 2010).

The data from section 5 (dry and milking cow management) of the 2019 follow-up RAMP indicated that herds had more contaminated dry cow and milking cow environments compared with that at the time of the original RAMP. More farms scored 7 (“manure is clearly visible OR mangers and water troughs cleaned less than once a month”) in their dry cow area: 30% versus 10% in 2013. A similar increase was seen in the milking cow area scores, with 21% scoring 7 versus 7% in 2013 (Table 7). The average score for section 5 increased from 18.1 in 2013 to 24.8 in 2019 ($P < 0.001$), representing an increase in the risk of JD transmission.

As with heifers, decreased cow cleanliness (i.e., increased score) represents a higher risk of JD disease. This result may be explained by shortages of bedding material or could be due to the survey administrator. Pieper et al. (2015) found variability in the scoring of risk assessments between veterinarians. As previous surveys were done by herd veterinarians, there may have been stigmatization associated with scoring anything as “dirty.” The perceptions and biases that may exist within a specific veterinarian-client relationship can influence the final results of the risk assessment.

Table 6. Section 4: weaned and bred heifer risk assessment scores from original and follow-up risk assessments from 180 farms; each point level (1, 4, 7, and 10) represents risk level pertaining to Johne’s for each question

<table>
<thead>
<tr>
<th>Section 4 question</th>
<th>2013 response, number (%)</th>
<th>2019 response, number (%)</th>
<th>$P$-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Are weaned heifers exposed to cows/cow manure at any time?</td>
<td>1 point</td>
<td>4 points</td>
<td>7 points</td>
</tr>
<tr>
<td>Are bred heifers exposed to cows/cow manure at any time?</td>
<td>122 (67.8)</td>
<td>33 (18.3)</td>
<td>21 (11.7)</td>
</tr>
<tr>
<td>Overall weaned heifer hygiene and cleanliness score</td>
<td>70 (38.9)</td>
<td>32 (17.8)</td>
<td>42 (23.3)</td>
</tr>
<tr>
<td>Overall bred heifer hygiene and cleanliness score</td>
<td>67 (37.2)</td>
<td>79 (43.9)</td>
<td>23 (12.8)</td>
</tr>
</tbody>
</table>

$^1$Values in bold font represent responses that have increased in frequency for their question from the recorded 2013 risk assessments.

$^2$Brunner improved Friedman test was used to assess for significant difference between the distribution of answers given in 2013 and those given in 2019.

$^3$One missing value from 2019 risk assessments.

$^4$Two missing values from 2013 risk assessments.
Increased cleanliness scores could also result from the market for bedding. Increased cost and decreased availability of good quality, clean, dry bedding material may result in producers using less to bed their animals, resulting in a more contaminated environment and more soiled animals. Similarly, increasing herd sizes and overcrowding of pens could result in more contaminated environments.

The initial intention was to conduct follow-up RAMP on equal numbers of farms from each of the 4 categories. This distribution was not achieved; however, the proportions across the 4 groups do somewhat mirror the distribution across the groups when considering the 3,207 farms (Table 1). Another consideration in the study would be the differences in farm demographics between the 2 testing periods. In 2013, there were 318,600 cows, 166,600 heifers, and 3,997 farms shipping milk. In 2017, at the time of the second BTM test, there were 318,500 cows, 152,000 heifers, and 3,613 farms (Agriculture Canada, 2021a).

At the time of writing, we have observed a consistent trend of decreasing numbers of dairy farms in the province (and country), with 3,367 registered dairy farms. Average herd size for the tiestall farms was 61.5 cows (in milk and dry), whereas the average herd size for freestall herds was 141.8 cows (in milk and dry; Agriculture Canada, 2021a). In comparison, the average herd size of our 180 study herds with freestall facilities was 111 in 2013 and 140 in 2019. The average herd size for our tiestall study herds was 50 in 2013 and 56 in 2019.

As survey enrollment was voluntary, it is possible that producers who participated in the study used different management practices than those who did not participate. If this bias exists, it would be reasonable to assume that the participants were more proactive and progressive in their management. Another possible bias would be the administration of the survey; the original RAMP were conducted by a large number of trained veterinarians, whereas the follow-up RAMP were done by a smaller number of research assistants. Although all research assistants were trained by the developer of the RAMP and followed the same guide as the veterinarians, subjectivity still plays a role in cleanliness scoring. Variability in RAMP scores due to differences in survey administrators is a challenge; ideally, the same individual should be used for repeated assessments. Unfortunately, due to logistics, this was not possible for our study. Convenience sampling may also have biased the results; regional isolation may result in different management practices in the more northern Ontario herds. Our sample goal was 180 herds. Although an appropriate sample size is difficult to determine in a study investigating changes in survey answers, we believed that we captured a good representation of the sample population by including farms with different BT results (Table 1).

A limitation of BTM ELISA is that dry cows are not included in the test. Dilution of antibody is also an issue, in that if the prevalence of JD is very low or positive cows contribute much less to the bulk tank than their negative herd mates, these herds may be misclassified as low risk. Further to this, ELISA corrected OD values close to the 0.089 cutoff could have the opposite dichotomous result due to slight variations on test-day (Collins et al., 1993). Although within-batch variability can exist in commercial ELISA tests (Köhler et al., 2022), the magnitude of change seen in the BT test results in this study is beyond what would be expected due to batch variability over the study’s 5-yr testing in-

<table>
<thead>
<tr>
<th>Table 7. Section 5: milking and dry cow risk assessment scores from original and follow-up risk assessments from 180 farms; each point level (1, 4, 7, and 10) represents risk level pertaining to Johne’s for each question1</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Section 5 question</strong></td>
</tr>
<tr>
<td>-----------------------------------------------</td>
</tr>
<tr>
<td>Dry cow area environment hygiene score3</td>
</tr>
<tr>
<td>Milking cow area environment hygiene score</td>
</tr>
<tr>
<td>Dry cow cleanliness</td>
</tr>
<tr>
<td>Milking cow cleanliness</td>
</tr>
<tr>
<td>Spreading manure on hay</td>
</tr>
<tr>
<td>Spreading manure on alfalfa</td>
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</tbody>
</table>

1Values in bold font represent responses that have increased in frequency for their question from the recorded 2013 risk assessments.
2Brunner improved Friedman test was used to assess for significant difference between the distribution of answers given in 2013 and those given in 2019.
3One missing value from 2019 risk assessments.
terval. Further research is needed to investigate factors in herds that showed increased risk of JD as described by their BT status. There is also a need for more investigations into the use and interpretation of the modified BTM ELISA in JD surveillance.

To our knowledge, few other studies have attempted to quantify specific management changes over an extended period, as captured in our study, pertaining to the control of JD. The time frame in our study was approximately 6 yr (2013–2019). Wells et al. (2008) found that a reduction in risk assessment scores over a 6-yr period was associated with a decrease in the seroprevalence in both beef and dairy herds participating in a JD control program. Ferrouillet et al. (2009) reported in their longitudinal study that herds that decreased their risk assessment scores across 5 yr of annual risk assessment had a lower prevalence of ELISA-positive animals and fewer fecal shedders. Although these studies did capture a considerable time period, the specifics of the management changes that occurred were not reported.

Further research is required to investigate some of the motivational factors behind changes in management. Qualitative research methods may prove useful in this. Both focus groups and in-depth one-on-one interviews may serve as useful tools in discovering some of the factors underlying producer behavior in the context of Johne’s control on Ontario dairy farms.

CONCLUSIONS

Our primary hypothesis that a decrease in RAMP scores and a corresponding decrease in BT prevalence would be present after the implementation of the OJE-MAP control program was incorrect according to the results of the follow-up risk assessment study. Many changes in management practices occurred since the first RAMP were administered in 2010 to 2013. Some of those changes suggested that advice given during the Johne’s control program had been adopted by producers to decrease their risk of JD. Most notably, changes in cattle buying practices resulted in fewer herds purchasing from multiple sources or purchasing animals outright. The idea of keeping the maternity pen free from sick animals seems to be a priority for more herd managers; however, the use of space for multiple calving still presents some challenges for JD control. The advice given during the Johne’s control program was likely balanced against internal and external pressures on the producer. The result was that some practices were changed such that JD risk was lowered, whereas others resulted in the unintended consequence of increasing risk. The apparent increase of BT positivity suggests that researchers and veterinary professionals must carefully consider the hidden pervasive nature of JD.

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