Stochastic simulations of a multi-group compartmental model for Johne's disease on US dairy herds with test-based culling intervention

Zhao Lu *, Ynte H. Schukken, Rebecca L. Smith, Yrjo T. Grohn

Department of Population Medicine and Diagnostic Sciences, College of Veterinary Medicine, Cornell University, Ithaca, NY 14853, USA

**A R T I C L E   I N F O**

Article history:
Received 10 October 2009
Received in revised form
4 February 2010
Accepted 22 March 2010
Available online 27 March 2010

Keywords:
Stochastic modeling
Johne's disease
Test-based culling
Uncertainty and sensitivity analyses

**A B S T R A C T**

Infection elimination may be an important goal of control programs. Only in stochastic infection models can true infection elimination be observed as a fadeout. The phenomena of fadeout and variable prevalence are important in understanding the transmission dynamics of infectious diseases and these phenomena are essential to evaluate the effectiveness of control measures. To investigate the stochastic dynamics of Mycobacterium avium subsp. paratuberculosis (MAP) infection on US dairy herds with test-based culling intervention, we developed a multi-group stochastic compartmental model (a continuous time Markov chain model) with both horizontal and vertical transmission. The stochastic model predicted fadeout and within-herd prevalence to have a large variance. Although test-based culling intervention generally decreased prevalence over time, it took longer than desired by producers to eliminate the endemic MAP infection from a herd. Uncertainty analysis showed that, using annual culture test and culling of only high shedders or culling of both low and high shedders with a 12-month delay in culling of low shedders, MAP infection persisted in many herds beyond 20 years. While using semi-annual culture test and culling of low and high shedders with a 6-month delay in culling of low shedders, MAP infection in many herds would be extinct within 20 years. Sensitivity analysis of the cumulative density function of fadeout suggested that combining test-based culling intervention and reduction of transmission rates through improved management between susceptible calves and shedding animals may be more effective than either alone in eliminating endemic MAP infection. We also discussed the effects of other factors such as herd size, heifer replacement, and adult cow infection on the probability of fadeout.

© 2010 Elsevier Ltd. All rights reserved.

**1. Introduction**

Johne's disease, caused by Mycobacterium avium subsp. paratuberculosis (MAP) infection, results in economic losses in dairy industry. In the United States, the cost to dairy producers due to the impact of Johne's disease on milk production was estimated to be more than $200 million per year (Ott et al., 1999). Johne's disease prevalence has been increasing and is currently one of the more important infectious diseases in cattle (NAHMS, 1997). Prevalence of MAP infection (the proportion of detected animals shedding low and high quantities of MAP within a herd) is generally low, but control of Johne's disease is difficult due to the long incubation period and low test sensitivities for animals shedding low quantities of MAP (Whitlock et al., 2000; NRC, 2003; Collins, 2005; Collins et al., 2006).

To lower the risk of MAP infection and reduce potential economic loss, producers have frequently applied test-based culling of infectious animals in herds as a control measure. Culling of animals shedding high levels of MAP is typically immediate, as high shedders are considered to be a high risk for spreading MAP and these animals also have a lower milk production (Dorshorst et al., 2006; Smith et al., 2009). Culling of animals shedding low levels of MAP is not usually implemented because low shedders are lower perceived risk for MAP transmission and their milk production is unaffected by MAP infection (Smith et al., 2009). Evaluation of the effectiveness of test-based culling intervention for herds infected with MAP is complicated, because it requires understanding the dynamics of MAP infection within a herd and the impacts of test-based culling on those dynamics. Collection of longitudinal field data is important for studying the dynamics of MAP infection and evaluating the culling effectiveness, but such studies are costly and time-consuming. An overall understanding of the dynamics of MAP infection can be acquired by building a mechanism-based mathematical model, in which current biological and epidemiological knowledge of MAP infection, transmission, and culling intervention on dairy herds are integrated.

Mathematical modeling of Johne's disease has been developed to study the dynamics of MAP infection and useful insights have been obtained for the evaluation of control measures (Collins and...
Morgan, 1992; Beyerbach et al., 2001; Groenendaal et al., 2002, 2003; Pouillot et al., 2004; Kudahl et al., 2007; Mitchell et al., 2008; Lu et al., 2008). In a previous deterministic model, we used the threshold property of the reproduction ratio (R) as a criterion to evaluate the effectiveness of test-based culling intervention (Lu et al., 2008). This criterion provides an important basis for understanding the impacts of test-based culling intervention on the transmission of MAP infection, but it only represents two possible outcomes in the long term: either MAP is predicted to be eliminated (R < 1) or not (R > 1). A more commonly used and more intuitive approach to assessing the effectiveness of test-based culling intervention is to compare prevalence over time in the presence/absence of test-based culling.

Although deterministic models are helpful in assessing the potential impact of test-based culling on MAP infection in dairy herds, they only provide average results, so the variance of prevalence cannot be addressed. When the number of total infected animals within a herd is small, fadeout becomes more important. However, previous stochastic models of Johne's disease on dairy herds did not consider the effect of test-based culling intervention on fadeout (Groenendaal et al., 2002, 2003; Pouillot et al., 2004; Kudahl et al., 2007). The objective of this study was to evaluate the effectiveness of test-based culling intervention and its impact on fadeout in Johne's disease control on dairy herds using a stochastic modeling approach.

2. Model and methods

To investigate fadeout and its effect on the dynamics of MAP infection under culling intervention, we developed a stochastic multi-group compartmental model with both horizontal and vertical transmission. The fadeout data from simulations were analyzed using a cumulative density function (CDF). To understand the effect of uncertainty of model parameters on MAP infection dynamics, we performed global uncertainty analyses for prevalence, the reproduction ratio, and the CDF of fadeout. To find the most influential model parameters, sensitivity analysis for the CDF of fadeout was carried out.

2.1. Model description

A multi-group compartmental model representing a typical commercial US dairy farm infected with MAP is illustrated in Fig. 1 (Mitchell et al., 2008). In this model, three groups were modeled: calves (< 1 year), heifers (1–2 years), and adult cows. According to current understanding of the pathobiology of MAP infection, calves were divided into susceptible (X₁) and transiently shedding (Tr₁) compartments; heifers were divided into resistant (X₃), transiently shedding (Tr₂), and latent (H₂) compartments; adult cows were divided into resistant (X₃), latent (H₃), low shedding (Y₁), and high shedding (Y₂) compartments.

Calves (X₁) were susceptible to MAP infection until one year of age when they became age resistant. Resistant heifers (X₃) and adult cows (X₃) have a very low risk of being infected by MAP due to their increasing age, and were considered resistant to MAP infection in the present model. Transiently shedding calves (Tr₁) and heifers (Tr₂) can shed low levels of MAP intermittently; latent heifers (H₂) and adult cows (H₃) either shed no MAP or the level of MAP shedding is not detectable by the current diagnostic methods; infectious adult cows can shed low levels of MAP (Y₁), ≤ 300 cfu/g, or high levels of MAP (Y₂), > 300 cfu/g (van Schaik et al., 2005; Whitleck, 2005a, b; van Roermund et al., 2007).

Susceptible calves (X₁) were infected through one of three direct transmission routes: calf–calf transmission (β₁y₁) and susceptible calves infected by either low (β₁yₙ) or high (β₂yₙ) shedding adult animals. The force of infection $λ₁$ to susceptible calves was parametrized as $λ₁ = β₁(Tr₁ + Tr₂) + β₁X₁Y₁ + β₁Y₂$, where transmission parameters are $β₁$, $β₁y₁$, and $β₁y₂$. While the fecal–oral transmission between susceptible calves and shedding adult animals was common, a recent study reported that transiently shedding calves were also able to infect susceptible calves in an experimental setting (van Roermund et al., 2007). Once susceptible calves were infected, they entered the transiently shedding compartment (Tr₁). Calves in the susceptible compartment (X₁) not infected by MAP moved to the resistant compartment (X₂) at rate $ρ₁$ and then entered the resistant adult compartment (X₃) at rate $ρ₂$. Although resistant heifers and cows may also be infected by MAP, the risk of their infection is very low; therefore, the forces of infection $λ₂$ and $λ₃$ to resistant heifers and cows, respectively, were assumed zero in this study. After transiently shedding, infected young animals became latent (H₂) at rate $φ₁$ and then moved to the adult latent group (H₃) at rate $φ₂$. The latent cows (H₃) progressively became low shedding animals (Y₁) at rate $σ$ and then entered the high shedding compartment (Y₂) at rate $ν$. High shedding animals with clinical signs were removed (via extra culling) from the herd at rate $x$. All animals were subject to a general removal rate (or base culling rate); the general removal rates of calves, heifers, and cows are $μ₁$, $μ₂$, and $μ₃$, respectively.

As seen in Fig. 1, vertical transmission, reflecting the in utero infection of a proportion of calves born to their infected (H₂) or infectious dams (Y₁ and Y₂), was also modeled. The proportion of live female calves through vertical transmission $γ$ was expressed as $μ₂γ₂H₂ + γ₁Y₁ + γ₂Y₂$, where $μ₂$ is the birth rate of female calves, and $γ₁$, $γ₂$, and $γ₃$ are the proportional ratios of calves being vertically infected, respectively.

Parameters used in the model and their baseline values are summarized in Table 1. Most parameters were directly estimated...
from US dairy herds. Transmission rates $\beta_1$ (transiently shedding animals) and $\beta_2$ (low shedding animals) were assumed constant and relatively small compared to the transmission rate $\beta_2$ (high shedding animals), because the majority of infections occurring in dairy herds were believed to be caused by high shedding animals ($Y_2$). The value of the transmission rate $\beta_2$ was varied within a range, which corresponds to an observed spectrum of management in dairy farms, from good to poor management (Lu et al., 2008). As most dairy farms are well managed with respect to hygiene, a relatively low transmission rate $\beta_2$ was assumed to match the relatively low proportion of shedding animals in a herd. However, for a farm of poor management (high transmission rate $\beta_2$), the proportion of shedding animals is high in a herd.

Numerical evaluation of test-based culling rates $\delta_1$ and $\delta_2$ was based on the properties of diagnostic methods such as fecal culture test, and the producers decision to cull low and high shedders. Expressions of these test-based culling rates were derived in our previous study (Lu et al., 2008):

$$\delta_1 = \left(1 - \frac{1}{S_{c1}}\right)^{-1},$$

$$\delta_2 = \left(1 - \frac{1}{S_{c2}}\right)^{-1},$$

where test sensitivities of low and high shedding animals are represented by $S_{c1}$ and $S_{c2}$, respectively; $T_1$ and $T_2$ are testing interval and test turnaround time, respectively. The $T_{ta}$ denotes the time in the interval between two subsequent tests at which an infected animal starts to shed low or high level of MAP. Here, $\eta$ was assumed to be $\frac{1}{2}$. The delay time between diagnosis and culling of low shedding animals, due to economic considerations, is represented by $T_{ac}$. When no test-based culling is implemented, culling rates $\delta_1$ and $\delta_2$ are zero. In this study, we assumed two types of culture-based culling decisions: culling of high shedding animals was immediately after diagnosis with either delayed culling or no culling of low shedders.

2.2. Stochastic simulations

All events and their rates used in the stochastic model were defined in Table 2. All nine compartmental variables labeled in Fig. 1 were represented by non-negative integer numbers. We used Gillespies direct algorithm for simulating the stochastic dynamics of MAP transmission with test-based culling intervention (Gillespie, 1976, 1977; Renshaw, 1993; Gillespie, 2007; Keeling and Rohani, 2008). The herd size selected was 100, including all animals in three groups. To keep herd size constant, an event of removal, test-based culling, or extra culling induced by clinical Johne's disease, was followed by an immediate replacement.

In this study we are most interested in evaluating the effectiveness of test-based culling intervention on endemically infected herds. The endemic infection was defined as a state in which the number of animals infected with MAP is finite over a relatively long term. In a deterministic compartmental model, the number of animals infected with MAP is a constant over time; while in a stochastic model the number of animals infected with MAP fluctuates around a constant level over time. We ran 100,000 simulations, starting from identical MAP-free equilibria (see Appendix A) after introduction of a primary infected calf for a long time ($\geq 50$ years) to obtain a set of approximately stochastic endemic infection. Next, using the obtained stochastic endemic MAP infection as the starting point, we ran the stochastic model with and without test-based culling intervention.

Prevalence in this study was defined as the proportion of the herd that is truly infected, as predicted by the stochastic model, i.e., $\text{Prevalence} = \frac{[T_{r1} + T_{r2} + H_2 + H_3 + Y_1 + Y_2]}{N}$. To study fadeout phenomena, we defined a fadeout as a simulation in which the total number of truly infected animals ($T_{r1} + T_{r2} + H_2 + H_3 + Y_1 + Y_2$) is zero at a time point, i.e., fadeout occurred when prevalence became zero. The empirical CDF of fadeout under different test-and-cull strategies was evaluated to quantitatively analyze simulated fadeout events. The maximum time scale was set as 25 years.

### Table 1

Parameters used in the model.

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Description</th>
<th>Value</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\mu_1$</td>
<td>Removal rate of calves</td>
<td>0.111 (per year)</td>
<td>NAHMS (2005)</td>
</tr>
<tr>
<td>$\mu_2$</td>
<td>Removal rate of heifers</td>
<td>0.007 (per year)</td>
<td>–</td>
</tr>
<tr>
<td>$\mu_3$</td>
<td>Removal rate of cows</td>
<td>0.332 (per year)</td>
<td>–</td>
</tr>
<tr>
<td>$\mu_4$</td>
<td>Female calves birth rate</td>
<td>0.45 (per year)</td>
<td>Estimated</td>
</tr>
<tr>
<td>$\gamma_k$</td>
<td>Portion infected at birth given infected adult latent dams</td>
<td>0.15</td>
<td>Sweeney et al. (1992)</td>
</tr>
<tr>
<td>$\gamma_1$</td>
<td>Portion infected at birth given infected low shedding dams</td>
<td>0.15</td>
<td>Whitlock et al. (2005a)</td>
</tr>
<tr>
<td>$\gamma_2$</td>
<td>Portion infected at birth given infected high shedding dams</td>
<td>0.17</td>
<td>–</td>
</tr>
<tr>
<td>$\rho_1$</td>
<td>Exit rate from calves to heifers</td>
<td>1.0 (per year)</td>
<td>Estimated</td>
</tr>
<tr>
<td>$\rho_2$</td>
<td>Exit rate from heifers to adult cows</td>
<td>1.0 (per year)</td>
<td>Estimated</td>
</tr>
<tr>
<td>$\phi$</td>
<td>Exit rate from transiently shedding heifers to latent heifers</td>
<td>2.0 (per year)</td>
<td>Rankin (1961)</td>
</tr>
<tr>
<td>$\sigma$</td>
<td>Exit rate from latent adult cows to low shedders</td>
<td>0.667 (per year)</td>
<td>van Schaik et al. (2003)</td>
</tr>
<tr>
<td>$\psi$</td>
<td>Exit rate from the low shedders to high shedders</td>
<td>0.33 (per year)</td>
<td>–</td>
</tr>
<tr>
<td>$\alpha$</td>
<td>Extra culling rate due to the clinical signs</td>
<td>0.70 (per year)</td>
<td>Mitchell et al. (2008)</td>
</tr>
<tr>
<td>$\theta$</td>
<td>Fraction of time between subsequent tests that at which an infected animal starts to shed low or high levels of MAP</td>
<td>0–1 (0.5 assumed)</td>
<td>Lu et al. (2008)</td>
</tr>
<tr>
<td>$S_{c1}$</td>
<td>Test sensitivity of culture tests for low shedders</td>
<td>0.50</td>
<td>Whitlock et al. (2000)</td>
</tr>
<tr>
<td>$S_{c2}$</td>
<td>Test sensitivity of culture tests for high shedders</td>
<td>0.90</td>
<td>Collins et al. (2006)</td>
</tr>
<tr>
<td>$T_{t1}$</td>
<td>Test turnaround time of culture tests</td>
<td>3/12 (year)</td>
<td>–</td>
</tr>
<tr>
<td>$T_{t2}$</td>
<td>Test turnaround time of culture tests</td>
<td>3/12 (year)</td>
<td>–</td>
</tr>
<tr>
<td>$T_{r1}$</td>
<td>Testing interval</td>
<td>12, 6 months</td>
<td>–</td>
</tr>
<tr>
<td>$T_{r2}$</td>
<td>Testing interval</td>
<td>12, 6 months</td>
<td>–</td>
</tr>
<tr>
<td>$T_{ta}$</td>
<td>The time delay in culling of low shedder shedders</td>
<td>12, 6 months</td>
<td>–</td>
</tr>
<tr>
<td>$\delta_1$</td>
<td>Culling rate for low shedders</td>
<td>Expression (1)</td>
<td>Lu et al. (2008)</td>
</tr>
<tr>
<td>$\delta_2$</td>
<td>Culling rate for high shedders</td>
<td>Expression (2)</td>
<td>–</td>
</tr>
<tr>
<td>$\beta_{r1}$</td>
<td>Transmission rate between transiently shedding animals and susceptibles</td>
<td>0.01 (per year)</td>
<td>Assumed</td>
</tr>
<tr>
<td>$\beta_{r2}$</td>
<td>Transmission rate between low shedders and susceptibles</td>
<td>0.02 (per year)</td>
<td>Assumed</td>
</tr>
<tr>
<td>$\beta_{r3}$</td>
<td>Transmission rate between high shedders and susceptibles</td>
<td>0.10 (per year)</td>
<td>Assumed</td>
</tr>
</tbody>
</table>
after test-and-cull intervention was initiated, which is long enough to observe fadeout of MAP infection in herds.

2.3. Global uncertainty and sensitivity analyses

The present stochastic model used 20 parameters as shown in Table 1. As parameters inevitably contain uncertainty to some extent, global parameter uncertainty analyses for prevalence over time and the CDF of fadeout are important (Saltelli et al., 2000; Marino et al., 2008). To provide an overview of prevalence over time and the CDF of fadeout with parameter uncertainty, we created 100 sample points in model parameter space using the Latin-Hypercube sampling technique (Saltelli et al., 2000; Helton and Davis, 2002). Parameters were described by a uniform distribution with 20% from the baseline values in Table 1. For each specified parameter set, we ran 10,000 simulations, starting with the approximately endemic MAP infection, to incorporate stochastic effects (Vose, 2008). In addition to uncertainty analyses of prevalence and the CDF of fadeout, the uncertainty of the reproduction ratio, $R$, was calculated. To find the most influential model parameters for fadeout, sensitivity analysis for the CDF of fadeout was performed, using a partial rank correlation coefficient method (Marino et al., 2008).

3. Results

3.1. Distribution of animals at endemic infection

Fig. 2 illustrates the distribution of animals within a 100-animal dairy herd in nine compartments at the state of endemic MAP infection without test-based culling intervention. For a well-managed farm (low transmission rate), numbers of infected animals in $T_r$, $T_h$, $H_h$, $I$, $Y$, compartments were generally lower than that of non-infected animals ($X_1$, $X_2$, and $X_3$).

### Table 2
Events and their rates of the stochastic model with test-based culling intervention.

<table>
<thead>
<tr>
<th>Event</th>
<th>Representation of transition</th>
<th>Rate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Removal of susceptible calves</td>
<td>$X_1 \rightarrow X_1 - 1$</td>
<td>$\mu_1 X_1$</td>
</tr>
<tr>
<td>Removal of resistant heifers</td>
<td>$X_2 \rightarrow X_2 - 1$</td>
<td>$\mu_2 X_2$</td>
</tr>
<tr>
<td>Removal of resistant cows</td>
<td>$X_3 \rightarrow X_3 - 1$</td>
<td>$\mu_3 X_3$</td>
</tr>
<tr>
<td>Removal of transiently shedding calves</td>
<td>$T_r \rightarrow T_r - 1$</td>
<td>$\mu_1 T_r$</td>
</tr>
<tr>
<td>Removal of transiently shedding heifers</td>
<td>$T_h \rightarrow T_h - 1$</td>
<td>$\mu_2 T_h$</td>
</tr>
<tr>
<td>Removal of latent heifers</td>
<td>$H_1 \rightarrow H_1 - 1$</td>
<td>$\mu_3 H_1$</td>
</tr>
<tr>
<td>Removal of latent cows</td>
<td>$H_2 \rightarrow H_2 - 1$</td>
<td>$\mu_2 H_2$</td>
</tr>
<tr>
<td>Removal of low shedders</td>
<td>$Y_1 \rightarrow Y_1 - 1$</td>
<td>$\mu_1 Y_1$</td>
</tr>
<tr>
<td>Removal of high shedders</td>
<td>$Y_2 \rightarrow Y_2 - 1$</td>
<td>$\mu_2 Y_2$</td>
</tr>
<tr>
<td>Test-based culling of low shedders</td>
<td>$Y_1 \rightarrow Y_1 - 1$</td>
<td>$\delta_1 Y_1$</td>
</tr>
<tr>
<td>Test-based culling of high shedders</td>
<td>$Y_2 \rightarrow Y_2 - 1$</td>
<td>$\delta_2 Y_2$</td>
</tr>
<tr>
<td>Extra culling due to clinical signs</td>
<td>$X_2 \rightarrow X_2 - 1$</td>
<td>$\alpha X_2$</td>
</tr>
<tr>
<td>Movement from $X_1$ to $X_2$</td>
<td>$X_1, X_2 \rightarrow X_1 - 1, X_2 + 1$</td>
<td>$\mu_1 X_1$</td>
</tr>
<tr>
<td>Movement from $X_2$ to $X_1$</td>
<td>$X_2X_1 \rightarrow X_2 + 1, X_1 + 1$</td>
<td>$\mu_2 X_2$</td>
</tr>
<tr>
<td>Movement from $T_r$ to $T_r$</td>
<td>$T_r, T_r \rightarrow T_r - 1, T_r + 1$</td>
<td>$\mu_1 T_r$</td>
</tr>
<tr>
<td>Movement from $T_h$ to $T_h$</td>
<td>$T_h, T_h \rightarrow T_h - 1, T_h + 1$</td>
<td>$\delta T_h$</td>
</tr>
<tr>
<td>Movement from $H_1$ to $H_1$</td>
<td>$H_1, H_1 \rightarrow H_1 - 1, H_1 + 1$</td>
<td>$\mu_3 H_1$</td>
</tr>
<tr>
<td>Movement from $H_2$ to $H_2$</td>
<td>$H_2, H_2 \rightarrow H_2 - 1, H_2 + 1$</td>
<td>$\mu_2 H_2$</td>
</tr>
<tr>
<td>Vertical transmission due to dams $H_1$</td>
<td>$T_r \rightarrow T_r + 1$</td>
<td>$\mu H_1 T_r$</td>
</tr>
<tr>
<td>Vertical transmission due to dams $Y_1$</td>
<td>$T_r \rightarrow T_r + 1$</td>
<td>$\mu H_1 Y_1$</td>
</tr>
<tr>
<td>Vertical transmission due to dams $Y_2$</td>
<td>$T_r \rightarrow T_r + 1$</td>
<td>$\mu H_2 Y_2$</td>
</tr>
<tr>
<td>Direct transmission between transiently shedding calves ($T_r$) and susceptibles ($X_1$)</td>
<td>$X_1, T_r \rightarrow X_1 - 1, T_r + 1$</td>
<td>$\beta T_r X_1$</td>
</tr>
<tr>
<td>Direct transmission between transiently shedding heifers ($T_h$) and susceptibles ($X_1$)</td>
<td>$X_1, T_r \rightarrow X_1 - 1, T_r + 1$</td>
<td>$\beta T_r X_1$</td>
</tr>
<tr>
<td>Direct transmission between low shedders ($Y_1$) and susceptibles ($X_1$)</td>
<td>$X_1, Y_1 \rightarrow X_1 - 1, Y_1 + 1$</td>
<td>$\alpha Y_1$</td>
</tr>
<tr>
<td>Direct transmission between high shedders ($Y_2$) and susceptibles ($X_1$)</td>
<td>$X_1, Y_2 \rightarrow X_1 - 1, Y_2 + 1$</td>
<td>$\alpha Y_2$</td>
</tr>
</tbody>
</table>

Fig. 2. The stochastic distribution of animals in a dairy herd of 100 animals at endemic infection. Numbers of the first year calves, the second year heifers, and adult animals ($\geq 2$ years) are $X_1 + T_r$, $X_2 + T_h + H_h$, and $X_3 + H_1 + Y_1 + Y_2$, respectively (see Fig. 1 for symbol codes).

Fig. 3. Demonstration of fadeout and persistence over time without test-based culling intervention. (a) Fadeout occurred at 16.5 years when all infected animals disappeared; (b) persistence of MAP infection was more than 25 years.
3.2. Impact of test-based culling on fadeout

To demonstrate fadeout and persistence, two specific realizations of stochastic simulations without test-based culling are shown in Fig. 3. Here, initial states of these two specific realizations were identical, \([X_t, X_0, X_Y, T_{r1}, T_{r2}, H_2, H_3, Y_t, Y_1, Y_2] = [14, 19, 42, 3, 1, 7, 9, 4, 1]\). We found four possible patterns of low and high shedders at any given time point: (1) both low and high shedders > 0; (2) low shedders > 0 and high shedders = 0; (3) low shedders = 0 and high shedders > 0; (4) both low and high shedders = 0. In Fig. 3(a), fadeout (elimination of MAP infection) occurred at approximately at the 17th year, when all infected animals disappeared. In Fig. 3(b), MAP infection persisted, i.e., the number of total infected animals was not zero prior to the 25th year, although numbers of low and high shedders were zero at some time points.

Fig. 4 showed the impacts of test-based culling on the cumulative probability function of fadeout. In Fig. 4(a), the probability of removing MAP infection from an endemically infected herd increased when: (1) testing frequency increased from annual to semi-annual tests; (2) in addition to immediate culling of high shedders after diagnosis, culling of low shedders was implemented (with a 6- or 12-month delay after diagnosis). The probability of fadeout increased very slowly in the first 10 years after test-based culling was initiated. For annual or semi-annual test-based culling of only high shedders, the probability of fadeout continued to slightly increase within 25 years. For annual or semi-annual test-based culling of both low and high shedders, the probability of fadeout quickly increased after 10 years. Twenty years after test-based culling was initiated, the probability of fadeout for annual or semi-annual test-based culling of only high shedders was below 0.14, while the probability of fadeout for culling of both low and high shedders with annual and semi-annual tests was 0.38 and 0.66, respectively. In Fig. 4(b), we found that the maximal rate of fadeout for culling of both low and high shedders, with a 6-month testing interval and a 6-month delay time to cull low shedders, was around 14 years after test-based culling was started.

3.3. Impact of test-based culling on prevalence and the proportion of shedders

Statistics of prevalence \((Y_1 + Y_2)/N\) over time with and without test-based culling was summarized in Fig. 5. In Fig. 5(a), the 10th–90th percentile of endemic prevalence was 8–36%. When no test-based culling was applied, the summary statistics of prevalence showed that MAP infection was in an approximately endemic state, although there was a slightly decreasing tendency due to fadeout. In Fig. 5(b), when using annual culture test and culling of only high shedders, prevalence slowly decreased within 25 years. At the 20th year, ~10% of simulation runs had zero prevalence. In Fig. 5(c), when using annual culture test and culling of low and high shedders with a 12-month delay in culling of low shedders, we found that at the 24th year, ~50% of simulation runs had zero prevalence. In Fig. 5(d), when using semi-annual culture test and culling of low and high shedders with a 6-month delay in culling of low shedders, we observed zero prevalence from ~75% of simulation runs at the 23rd year (the cumulative probability of fadeout for this test-and-cull strategy was 0.66 as shown in Fig. 4).

Fig. 6 showed the proportion of shedders \((Y_1 + Y_2)/N\) over 25 years in a herd with and without test-based culling intervention. In Fig. 6(a), the 10th–90th percentile of the proportion of shedders was 2–11%. As in Fig. 5, the dynamics of shedders in Fig. 6(b)–(d) had similar patterns with the same test-based culling strategies. However, the proportion of shedders is much lower than that of total infected animals (prevalence).

3.4. Impact of test-based culling on uncertainty of the mean prevalence, the reproduction ratio \(R\), and fadeout

In each panel of Fig. 7 100 lines of the mean prevalence were plotted with and without test-based culling. The mean prevalence for each parameter set was obtained from 10,000 simulation runs (the total simulation runs is 1,000,000 = 100 parameter sets \(\times 10,000\) simulation runs per parameter set; Vose, 2008). When no test-based culling was applied (shown in Fig. 7(a)), the mean prevalence was distributed between 10% and 42%. In Fig. 7(b)–(d), the mean prevalence dropped after test-based culling was initiated at the beginning, and then gradually decreased over time.

In Fig. 8, the frequency distribution of the reproduction ratio \(R\) was shown. For well-managed herds endemically infected with MAP with no test-based culling intervention, the distribution of the reproduction ratio \(R\) was almost entirely greater than 1 (MAP...
infection persisted). When test-based culling was applied, the reproduction ratio decreased and its frequency distribution shifted to the left. With culling of both low and high shedders, using either annual or semi-annual culture test and 12-month delay to cull low shedders; (d) culling of both low and high shedders with semi-annual test, and 6-month delay to cull low shedders. In each panel, statistics of prevalence were summarized by the 90th, 75th, 50th, 25th, 10th percentiles (curves from top to bottom), and the mean (solid black line).

Fig. 6. Statistics of the proportion of shedders \((Y_1 + Y_2)/N\) over time from stochastic simulations with and without test-based culling intervention using the baseline values of model parameters in Table 1: (a) no test-based culling; (b) culling of only high shedders with annual culture test; (c) culling of both low and high shedders with annual culture test, and 12-month delay to cull low shedders; (d) culling of both low and high shedders with semi-annual test, and 6-month delay to cull low shedders. In each panel, statistics of prevalence were summarized by the 90th, 75th, 50th, 25th, 10th percentiles (curves from top to bottom), and the mean (solid black line).

Fig. 9 provided an overview of uncertainty in the CDF of fadeout. Each CDF line was obtained using fadeout data produced by 10,000 simulations of at most 25 years (Vose, 2008). Because there were 100 independent samples of model parameters, 100 CDF lines were plotted. Without test-based culling, the probability of fadeout slowly increased over time (Fig. 9(a)). When test-based culling was applied, Fig. 9(b)–(d) showed that test-based culling generally increased the probability of fadeout. For endemically infected dairy herds, however, time to a 50% chance of fadeout was highly variable: in Fig. 9(a), it was well
beyond 25 years (none of CDF lines reached 0.5 at the 25th year); in Fig. 9(b), greater than 25 years (\(\frac{C}{24}\)\% of CDF lines reached 0.5 at the 25th year); and in Fig. 9(d), 10–25 years (\(\frac{C}{24}\)\% of CDF lines reached 0.5 at the 25th year).

3.5. Sensitivity analysis of fadeout

The partial rank correlation coefficients (PRCC) of model parameters based on the uncertainty of the CDF of fadeout at the 10th year in Fig. 9(a)–(d) are illustrated in Fig. 10(a)–(d), respectively. Only significant model parameters (\(p\) value < 0.01, using student t test) were shown. In Fig. 10(a), when no test-based culling was applied, we found that the exit rate \(r_1\) between calf and heifer groups, the extra culling rate \(a\), the general removal rate \(m\) and transmission rates \(\beta_{y2}\) and \(\beta_{y1}\) were significantly correlated to the cumulative probability of fadeout. When test-based culling was applied (Fig. 10(b)–(d)), we found that the most influential parameter was still the exit rate \(r_1\), but more parameters become significantly correlated with the cumulative probability of fadeout. For example, the calf–calf transmission rate \(\beta_{tr}\) was significantly different from zero in all these test-and-cull strategies.

We also performed sensitivity analysis on the CDF of fadeout at the 20th year (not shown). The values of PRCC for model parameters significantly correlated with the cumulative probability of fadeout were nearly identical to those of the 10th year. In addition, sensitivity analysis for the mean prevalence (Fig. 7) was also carried out at the 10th year and the 20th year. The results (not shown) indicated that the list of parameters significantly correlated with the mean prevalence was almost identical to that for the cumulative probability of fadeout. However, the signs of PRCC values for model parameters were opposite to those for fadeout because a higher probability of fadeout implied lower mean prevalence.

3.6. Herd size effect on fadeout

Fig. 11 illustrates the effect of herd size on CDF of fadeout. When herd size increased from 100 to 200, and up to 500, we found that probabilities of fadeout of large herd sizes decreased. For culling of both low and high shedders with a 6-month testing interval and a 6-month delay culling of low shedders, the probability of fadeout had a relatively large drop, but for culling...
of only high shedders with a 12-month testing interval, the probability of fadeout had a relatively small drop.

4. Discussion

4.1. Stochastic modeling

Our objective was to evaluate the effectiveness of test-based culling intervention for MAP control and investigate its impact on fadeout of MAP infection on dairy herds using a stochastic modeling approach. Stochastic modeling is appropriate when our interests focus on understanding fadeout and the variable proportion of shedding animals in field studies. The multi-group stochastic model developed in this study for Johne’s disease on dairy herds was able to predict a large variance in numbers of animals in each compartment as shown in Fig. 2 and fadeout events in Fig. 4(a) and (b). Due to random events, in Fig. 3(a) and (b), we have illustrated that an endemically MAP infected herd could have completely different outcomes over time, either early fadeout or persistence over a long time. From Fig. 4(a), we found that, using annual culture test and culling of...
The cumulative probability of fadeout over time with test-based culling shown in Fig. 4(a) was affected by a number of the following factors. First, large herd size decreased the probability of fadeout. In Fig. 11, when herd size increased from 100 to 200, and up to 500, the probability of fadeout decreased. Second, we assumed that resistant heifers ($X_2$) and adult cows ($X_2$) were not infected, i.e., the forces of infection, $\lambda_2$, and $\lambda_3$, were zero, because there was no published data supporting these possible infection routes. However, if the adult cow infection route was considered to have a low infection rate in the model, $\lambda_2$, and $\lambda_3$, were not zero but small, it is assumed that more animals would be infected by MAP, thus the probability of fadeout would be decreased. Third, we assumed that the farm under study was closed, i.e., no external heifers were purchased and the farm provided its own replacement animals. If external purchasing was allowed, then the probability of fadeout would be decreased because replacement heifers might be infected with MAP. However, the risk of purchasing infected heifers could be reduced if these replacement animals were from certified Johne’s disease-free herds. Therefore, we may increase the probability of fadeout in our model through decreasing herd size, and purchasing no external replacement heifers, or purchasing only from certified Johne’s disease-free herds. Of course, improved farm management is also important. Although we only studied fadeout events for a small closed herd using the continuous time stochastic model, large and open herds can be simulated in the similar way. However, to simulate the extinction of MAP infection for those large and open herds, as shown in Fig. 8, where the mean prevalence decreased very slowly within 20 years, and in Fig. 9(b), where 20 years after test-based culling was started almost all herds had a cumulative probability of fadeout of less than 0.50. When using annual culture tests and culling of both low and high shedders with a 12-month delay in culling of low shedders, the cumulative probability of fadeout at the 20th year was possible (but not guaranteed). The above findings from the cumulative probability of fadeout, as shown in Fig. 4 were also supported by the dynamics of prevalence and the proportion of shedders as shown in Figs. 5 and 6.

4.2. Effectiveness of test-based culling intervention

In our previous study, we evaluated the effectiveness of test-based culling strategies based on the threshold property of the reproduction ratio ($R$) (Lu et al., 2008). When $R$ was reduced below 1 (indicating that the disease will eventually die out or be eliminated), the test-based culling intervention was considered to be effective. However, this criterion is only valid in the long term, which may be beyond the time scale of interest for JD control. For discussion purposes, we considered a 20-year time window as the time frame of interest to see the results of the implemented control programs. Our results (Figs. 7 and 8) showed that the reproduction ratio $R$ only predicted the trend of the mean prevalence under test-based culling intervention. In Fig. 8, when using annual culture test and culling of only high shedders, approximately 50% of $R$ realizations were greater than 1, which explains the long term maintenance of MAP infection and indicates that this test-and-cull strategy was not likely to be effective for elimination. This was indeed reflected in Fig. 7(b), where the mean prevalence decreased very slowly within 20 years, and in Fig. 9(b), where 20 years after test-based culling was started almost all herds had a cumulative probability of fadeout of less than 0.50. When using annual culture tests and culling of both low and high shedders with a 12-month in culling of low shedders, as shown in Fig. 8, realizations of the reproduction ratio were almost entirely below 1, which indicates MAP infection would finally die out in the long term, and this test-and-cull strategy was likely to be effective in eliminating of the endemic MAP infection. However, although the mean prevalence decreased more in Fig. 7(c) than that in Fig. 7(b), Fig. 9(c) showed that many herds had a cumulative probability of fadeout of less than 0.5.
years after test-based culling was initiated, indicating MAP infection in many herds persisted more than 20 years. When using semi-annual culture test and culling of low and high shedders with a 6-month delay in culling of low shedders, the reproduction ratio was further reduced below 1, which means this test-and-cull strategy is very likely to be effective in eliminating the endemic MAP infection. The effectiveness of this test-based culling strategy was also seen in Fig. 7(d), in which the mean prevalence decreased even further, and in Fig. 9(d), 20 years after test-based culling was started, many herds had a cumulative probability of fadeout of more than 0.5, indicating MAP infection in many herds would be extinct within 20 years (but elimination of MAP infection was not guaranteed). We, therefore, argue that the criterion of evaluating the effectiveness of test-based culling in controlling JD would require both \( R \) (for the general trend) and the stochastic dynamics of MAP infection such as fadeout and prevalence within a limited time to obtain a valid indication of time to elimination.

Culling of low and high shedding cows in dairy farms is mainly determined by economic considerations. Culling of only high shedders is generally desirable because of lower milk yield in these animals (Smith et al., 2009). Although the fecal culture test is more time-consuming and expensive than the ELISA test, in this study, we used the fecal culture test rather than the ELISA test, because the ELISA test is not able to distinguish low and high shedders (the detected antibody level in the ELISA test is not necessarily linearly correlated with the shedding level of MAP infected cows). To reduce the risk of MAP transmission on dairy herds, it is important to find low and high shedders as soon as possible. From Expressions (1) and (2) (see Section 2.1), the culling rates for low and high shedders (or the average detection time of low and high shedders) are determined by a few factors, such as testing interval, test sensitivity, test turnaround time, and delay time for culling of low shedders. Therefore, improving diagnostic methods by increasing test sensitivities for low and high shedders and/or decreasing test turnaround time (such as PCR based diagnostic method, Wells et al., 2006; Whitlock et al., 2007), along with increasing testing frequency from annual to semi-annual or even to quarterly tests, would result in the shorter average detection time for low and high shedders, and consequently higher test-based culling rates and better control of MAP infection. The three test-and-cull strategies in Figs. 5(b)–(d) and 6(b)–(d) showed how the shorter average detection times (higher culling rates) were achieved through decreasing testing interval from 1 year to 6 months.

### 4.3. Effective control measures

In Figs. 7(c)–(d) and 9(c)–(d), we can see that test-based culling intervention was able to reduce prevalence and increase the probability of fadeout over time, but it took longer than desired by producers to eliminate the endemic MAP infection from dairy herds. Sensitivity analysis in Fig. 10 listed the most influential model parameters on the CDF of fadeout. Aiming at those parameters, we may design more effective measures for JD control. The most influential parameter with and without test-based culling is the exit rate \( \rho_1 \) from susceptible calves \((X_1)\) to resistant heifers \((X_2)\), which was positively correlated with the cumulative probability of fadeout, indicating that increasing \( \rho_1 \) increased the cumulative probability of fadeout. This may be understood as more susceptible calves becoming resistant heifers, leaving fewer susceptible calves available to be infected and thus resulting in a higher chance of fadeout. Exit of susceptible calves \((X_1)\) to resistant heifers \((X_2)\) represents the development of age-related immunity. Although aging is not subject to random variation, the parameter \( \rho_1 \) reflects the complement of becoming infected, i.e., a higher \( \rho_1 \) indicates a lower rate of infection from susceptible calves \((X_1)\) to transiently shedding calves \((Tr_1)\). However, management driven control of this mostly age related parameter is difficult. The extra culling rate \( x \) and the general removal rate \( \mu_g \) were also positively correlated with the cumulative probability of fadeout. However, increasing these two parameters may result in decreasing the size of lactating group, because more adult cows would be removed from the herd. Transmission rates \( \beta_{y_2}, \beta_{y_1}, \) and \( \beta_{tr} \) were negatively correlated with the cumulative probability of fadeout, which indicated that increasing transmission rates between susceptible calves and either transiently shedding calves/heifers or low/high shedders caused more infections, and leading consequently to more shedding animals and decreasing the cumulative probability of fadeout.

The transmission rate \( \beta_{tr} \) (in Fig. 10(b)–(d)) became an important parameter correlated with the cumulative probability of fadeout. This indicates that, when test-based culling was implemented, calf-calf transmission became important. Therefore, a more effective measure for JD control may combine test-based culling and reducing transmission rates \( \beta_{y_2}, \beta_{y_1}, \) and \( \beta_{tr} \). Control measures aiming at the reduction of transmission rates can be achieved through vaccination or improvement of management of dairy herds. Because current Johne's disease vaccines may interfere with the bovine tuberculosis (TB) test, vaccination to control MAP infection is very limited. Improved management procedures used to reduce transmission rates include: separating healthy calves from low \( (\beta_{y_1}) \) and high \( (\beta_{y_2}) \) shedding cows and from transiently shedding calves/heifers \( (\beta_{tr}) \) to avoid fecal–oral transmission. The combined test-based culling intervention and improved management strategy has been recommended and was put into practice for up to seven years in a group of dairy herds in Wisconsin; it was reported that the incidence rate of MAP infection in calves significantly decreased (Collins and Eggleston, 2009).

### 5. Summary

We have studied the impacts of test-based culling intervention on fadeout and the dynamics of MAP infection in dairy herds using a stochastic compartmental model. The stochastic model predicted fadeout and a large variance of within-herd prevalence. Although test-based culling intervention generally decreased prevalence over time, it took longer than desired by producers to eliminate the endemic MAP infection from a herd. Uncertainty analysis showed that, using annual culture test and culling of only high shedders or culling of both low and high shedders with a 12-month delay in culling of low shedders, MAP infection persisted in many herds beyond 20 years. While using semi-annual culture test and culling of low and high shedders with a 6-month delay in culling of low shedders, MAP infection in many herds would be extinct within 20 years. Sensitivity analysis of the cumulative density function of fadeout suggested that combining test-based culling intervention and reduction of transmission rates through improved management between susceptible calves and shedding animals may be more effective than either alone in eliminating endemic MAP infection. The effects of herd size, purchasing replacement heifers, and adult cow infection on the probability of fadeout were also discussed.

### Acknowledgments

The USDA (CSREES) Awards Number 2008-35204-04627 and 2009-7610-05104 provided funding for this study, as did the...
Appendix A. Deterministic model: a system of nonlinear ordinary differential equations

The nonlinear ordinary differential equations used to describe Fig. 1 are

$$\frac{dX_1}{dt} = \mu N - \mu_0 y - (\lambda_1 + \rho_1 + \mu_1) X_1,$$

$$\frac{dX_2}{dt} = \rho_1 X_1 - (\mu_2 + \rho_2) X_2,$$

$$\frac{dY_1}{dt} = \lambda_1 X_1 + \mu_y y - (\rho_1 + \mu_1) Y_1,$$

$$\frac{dY_2}{dt} = \rho_1 Y_1 - (\mu_2 + \rho_2) Y_2,$$

where the force of infection $\lambda_1$, the general replacement rate $\mu$ and the rate of vertical transmission $\gamma$ are

$$\lambda_1 = \beta y (1 - T_{TV} + T_{H2}) + \beta h y_1 + \beta^2 y_2,$$

$$\mu = \mu_1 (X_1 + T_{TV}) + \mu_2 (X_2 + T_{H2} + H_2) + \mu_3 (X_3 + H_3 + Y_1 + Y_2) + \delta_1 Y_1 + (\delta_2 + 2 \delta_3) Y_2 / N,$$

$$\gamma = (\gamma_1 h + \gamma_2 y_1 + \gamma_3 y_2) / N.$$

At MAP infection free equilibrium, the analytical solutions are

$$X_1 = \frac{(\mu_1 + \rho_2) \mu_3}{\rho_1 \rho_2 + \mu_3 (\rho_1 + \mu_2 + \mu_3)} N,$$

$$X_2 = \frac{\rho_1 \mu_3}{\rho_1 + \mu_2 + \mu_3 (\rho_1 + \mu_2 + \mu_3)} N,$$

$$X_3 = N - X_1 - X_2,$$

$$T_{R1} = T_{R2} = T_2 = H_2 = Y_1 = Y_2 = 0.$$

Appendix B. The basic reproduction ratio with test-based culling

The reproduction ratio $R_0$ can be evaluated using the next generation matrix method (van den Driessche and Watmough, 2002). For the multi-group compartmental model shown in Fig. 1, the $R_0$ is

$$R_0 = R_{D1} + R_{D2} + R_{V1} + R_{V2},$$

where

$$R_{D1} = \frac{1}{\mu_1 + \rho_1} + \frac{1}{\mu_1 + \rho_1 + \mu_2 + \rho_2} \mu_1 B_{D1},$$

$$R_{D2} = \frac{1}{\mu_1 + \rho_1 + \mu_2 + \rho_2} \mu_2 B_{D2},$$

$$R_{V1} = \frac{\rho_1}{\mu_1 + \rho_1} + \frac{\rho_2}{\mu_2 + \rho_2} \mu_2 B_{V1},$$

$$R_{V2} = \frac{\rho_1}{\mu_1 + \rho_1 + \mu_2 + \rho_2} \mu_2 B_{V2}.$$

References


Johne’s disease status on milk production in individual cows. J. Dairy Sci. 92, 2653–2661.