A deterministic mathematical model of Mycobacterium avium subsp. paratuberculosis transmission on commercial US dairy farms

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ABSTRACT

Prevalence of Johne’s disease on U.S. dairy farms is estimated at one-fifth of all herds, and larger herds are found to be infected more often. Despite a low prevalence of high-shedding animals, elimination of MAP from herds has proven exceptionally challenging, with few published reports of successful eradication from infected farms. Mathematical modeling may help us understand this apparent contradiction. A deterministic mathematical model of MAP transmission on commercial US dairies was developed. It builds upon and modifies the assumptions in previous work to best reflect the pathobiology of the disease. Of the animals which test positive for MAP, high levels of bacterial shedding are noted in only a small proportion. Transmission was modeled using ordinary differential equations. Calculation of transmission parameters in these models is necessarily non-linear. Previous models of infection utilized linear dynamics only and therefore lack sensitivity to changes in susceptible population size. Animal turnover rates were obtained from the literature and transition from disease states were calculated from retrospective fecal culture data from herds in New York and Pennsylvania. The model shows that aggressive test-and-cull strategies do not result in successful elimination of MAP in a short timeframe. Transmission is relatively insensitive to the presence of high shedding animals. According to the model, multiple levels of contagiousness among infected adult animals and MAP shedding by infected calves explained the maintenance of low prevalence infections in herds. Although previous experimental studies support the potential of infectious transmission among young calves, further research with natural infections is needed to verify the existence of a pool of infectious young animals. If this group of animals is diagnostically identifiable, elimination strategies may need to address this route of transmission.

Key words: Calf, model, transmission, transient shedding, vertical transmission

INTRODUCTION

Johne’s disease is caused by Mycobacterium avium subsp. paratuberculosis (MAP), a slow-growing, Mycobactin-J dependent bacterium. Prevalence of MAP on US dairy farms at a herd level is estimated at 22% of all dairy herds, and 40% of all herds larger than 300 animals (Wells and Wagner, 2000). The high prevalence of infected herds makes transmission of MAP in a farm environment of interest both from animal welfare and economic perspectives. Recent research that considers MAP presence as a risk factor for human Crohn’s disease raises transmission of MAP as a potential human health hazard (Shulaw and Larew-Naugle, 2003). Although 2.6% of animals typically test positive for MAP on each infected farm (Groenendaal and Galligan, 1999), high levels of bacterial shedding is noted in only a small proportion of these animals (Whitlock et al., 2000). However, elimination of MAP from herds has proven exceptionally challenging, with few published reports of successful eradication of MAP from infected farms. Several previous models address not only how MAP is spread on a farm, but also incorporate intervention strategies to determine successful methods of eliminating the infection (Collins and Morgan, 1991; Collins et al., 1991; Groenendaal and Galligan, 1999; Beyerbach et al., 2001a; Beyerbach et al., 2001b; Groenendaal et al., 2002). The transmission of MAP in these models focuses on infection of young calves via MAP in
colostrum or milk, *in utero* infection, or exposure to infected feces. All models assign a shedding status to infected adult animals and base incident infections on the number of infectious adult animals present.

Collins and Morgan developed the first dynamic deterministic model of MAP transmission, which tracked the prevalence of four categories of animals (Collins and Morgan, 1991; Collins et al., 1991; Collins and Morgan, 1992). In this model, young animals remained susceptible up to age one year after which uninfected animals entered a resistant category and infected animals were clinically normal and not infectious. At two years of age, infected animals entered an infectious compartment for the remaining time in the herd. This model assumed homogenous mixing with equal risk of contact between susceptible animals and all herdmates and with calves from infected dams having the same risk of infection as all other calves.

Groenendaal and Galligan’s (1999) stochastic model introduced two adult shedding categories (low and high) and an increased risk of infection in selected individual animals via assumptions of non-homogenous mixing. This more complex model allowed for multiple ages at initial infection to better reflect disease dynamics following targeted management changes. Infected animals entered the high shedding category with a probability distribution based on age at infection, and were then retroactively assigned to have entered the low shedding compartment two lactations previously. These two separate infectious categories contributed differently to infection transmission, with the probability of an infectious contact decreasing for low shedding animals. Exposure and infection could occur as a result of vertical transmission and point source exposures (dam → daughter, colostrum → calf, infected milk → calf). Thus high-risk cohorts of animals or individuals existed rather than a constant exposure status among animals of the same age. Farm level risk factors established a base level of exposure as a result of environmental contamination.

Although these models served as the first and most widely recognized transmission models developed for paratuberculosis in a dairy setting, they are not “transmission dynamic”. That is, the assumption that halving the transmission / exposure will halve the infection incidence is not necessarily the case, since transmission dynamics are necessarily non-linear and are determined by susceptibility as well as exposure (Edmunds et al., 1999). These models reflect our current knowledge of the biology of MAP transmission, with limited input as to infectious status of calves and age at which animals are no longer susceptible to infection.

The objective of this paper is to develop a series of mathematical models of MAP transmission building upon and modifying the assumptions in previous work to best reflect the pathobiology of infection transmission. In this paper a series of models is used to explore sensitivity of the transmission cycle to a variety of current assumptions. Ultimately the goal of developing this series of models is to aid in determining effective control strategies for eliminating clinical paratuberculosis and MAP infection from the herd.

**MATERIALS AND METHODS**

We have developed a series of models which seek to mimic herd dynamics and disease process at an increasingly detailed level. Three such models will be developed in this paper. All models calculate a density dependent rate of contact rather than a population size dependent one. This allows models to reflect current data showing that within-herd prevalence is not greater in large herds (Collins et al., 1994; van Schaik et al., 2003). All model parameters are defined in Table 1.
Table 1. Definition of all symbols used in compartmental models. Values used in outputs and sources of data are provided.

<table>
<thead>
<tr>
<th>Symbol</th>
<th>Definition</th>
<th>Value</th>
<th>Source</th>
</tr>
</thead>
<tbody>
<tr>
<td>( t )</td>
<td>Duration of timestep</td>
<td>1 month</td>
<td>1</td>
</tr>
<tr>
<td>( \beta )</td>
<td>force of infection</td>
<td>8</td>
<td>2</td>
</tr>
<tr>
<td>( \beta_1 )</td>
<td>force of infection from Transients (Model C)</td>
<td>8</td>
<td></td>
</tr>
<tr>
<td>( \beta_2 )</td>
<td>force of infection from Y1 &amp; Y2 (Model C)</td>
<td>8</td>
<td></td>
</tr>
<tr>
<td>( \epsilon )</td>
<td>Increased infectiousness of Y2 vs Y1</td>
<td>1-1000</td>
<td>1</td>
</tr>
<tr>
<td>( \mu_d )</td>
<td>Universal death rate</td>
<td>0.223 /year</td>
<td>3</td>
</tr>
<tr>
<td>( \mu_1 )</td>
<td>Death rate Category 1</td>
<td>0.111/year</td>
<td>3</td>
</tr>
<tr>
<td>( \mu_2 )</td>
<td>Death rate Category 2</td>
<td>0.007/year</td>
<td>3</td>
</tr>
<tr>
<td>( \mu_{3vol} )</td>
<td>Voluntary cull rate Category 3</td>
<td>0.285/year</td>
<td>3</td>
</tr>
<tr>
<td>( \mu_{3inv} )</td>
<td>Involuntary cull rate Category 3</td>
<td>0.048/year</td>
<td>3</td>
</tr>
<tr>
<td>( \mu_3 )</td>
<td>Total cull rate Category 3*</td>
<td>0.333/year</td>
<td>3</td>
</tr>
<tr>
<td>( \alpha )</td>
<td>Additional death rate from Y/Y2</td>
<td>0.5/year</td>
<td>4</td>
</tr>
<tr>
<td>( \rho_i )</td>
<td>Rate of aging Category i</td>
<td>1/year</td>
<td>5</td>
</tr>
<tr>
<td>( \delta_i )</td>
<td>Additional cull rate of infected animals</td>
<td>( \delta_1=0.25/\text{year}; \delta_2=1/\text{year} )</td>
<td>1</td>
</tr>
<tr>
<td>( \gamma )</td>
<td>Rate of infection at birth given infected dam</td>
<td>( \delta=\text{average } \delta_1+\delta_2 )</td>
<td>6</td>
</tr>
<tr>
<td>( \Phi )</td>
<td>Rate of Exit Tr/Tr</td>
<td>Mode B =1/year; Mode C=1/year</td>
<td>7</td>
</tr>
<tr>
<td>( \sigma )</td>
<td>Rate of exit H</td>
<td>Model B = 0.667/year; Model C=1/year</td>
<td>4</td>
</tr>
<tr>
<td>( \nu )</td>
<td>Rate of exit Y1</td>
<td>0.33/year</td>
<td>4</td>
</tr>
</tbody>
</table>

1: user defined; 2: estimated by model sensitivity analysis 3:(2003) 4: (Whitlock et al., 2000; van Schaal et al., 2003) 5: calculated 6: (Benedictus et al, (Sweeney et al., 1992); 7: (Rankin, 1961) 

*Culling in the adult herd (\( \mu_3 \)) is maintained by decreasing voluntary culls when there are MAP culls (\( \mu_3=\mu_{3inv}+\mu_{3vol}-\alpha-\delta \)). This calculation reflects turnover in a commercial herd, where increased culling for MAP may result in reduced culling for other voluntary causes. This stable herd turnover maintains constant input of susceptible youngstock.

Model A: Most basic state transition model. Three states: Susceptible (\( X_1 \)), Infected (\( Y \)) and Resistant (\( X_2 \)). Transmission between states is modeled using rates defined in Table 1.

The first, most basic, model utilizes 3 infection states with one infectious stage (\( Y \)). All animals enter into a susceptible (\( X \)) category and progress to either resistant (\( X_2 \)) status via aging at a rate \( \rho \) or to infected (\( Y \)) status at a rate \( \beta \). Death rate of animals is constant for all stages (\( \mu_d \)) with the exception of \( Y \), in which there is additional death due to clinical disease (\( \alpha \)) and optional additional death from culling (\( \delta \)). \( R_0 \) is defined by a single equation: \( R_0=\beta/(\mu+\alpha) \)
A more developed model maintains a homogenous population of cattle (no age dependent death, homogenous mixing) but introduces more disease states. Following susceptibility ($X_1$), infected animals enter a transient shedding compartment (Tr). Animals exit this transient shedding state at a rate $\phi$ and enter an undetectable shedding or latent stage (H). Latent animals progress to low shedding (Y1) and high shedding (Y2) stages. $R_0$ is defined by a separate $R_0$ for each infectious compartment. Overall $R_0$ is the sum of the three. Homogenous mixing across all disease states requires that the transient shedding group has the same contact rate with $X_1$ as do other infectious compartments (Y1 and Y2).

$$R_{0\text{Tr}} = \frac{b_1}{(\phi + \mu)}$$
$$R_{0\text{Y1}} = \frac{b_2}{(\phi + \mu)(\sigma + \mu)(\nu + \mu)}$$
$$R_{0\text{Y2}} = \frac{e b_2}{(\phi + \mu)(\sigma + \mu)(\nu + \mu)(\alpha + \mu)}$$

**Model B**: Intermediate state transition model. 5 states: Susceptible ($X_1$), Transient (Tr), Latent (H), Low Shedding (Y1), High Shedding (Y2) and Resistant ($X_2$). Transmission between states is modeled using rates defined in Table 1.

**Model C**: Complex state transition model. Nine states reflect Susceptible youngstock ($X_1$), Transiently shedding youngstock (Tr$_1$), Transiently shedding intermediates (Tr$_2$), Latent intermediates (H$_2$), Resistant intermediates ($X_2$), Resistant adults ($X_3$), Latent adults (H$_3$), Low shedding adults (Y1) and High shedding adults (Y2). Transition between states is modeled from values in Table 1.

The most developed model in this series incorporates age dependent death which splits several compartments with otherwise similar characteristics (Tr$_1$/Tr$_2$, H$_1$/H$_2$, $X_2$/X$_3$). Animals thus progress through the same disease state in two separate age categories. In addition, this model allows age-dependent contact rates (homogenous mixing within age-category, but not between age categories), with transient
shedding young animals (Tr₁ and Tr₂) having potentially much higher contact with their age-cohort animals (X₁) than low and high shedding adults (Y₁ and Y₂). The expression of R is tedious due to the age-categories. The basic definition is quite similar to that in the previous model, with a series of equations reflecting the larger number of compartments.

This model has an additional flow of animals entering the system (γ) accounting for calves infected at birth which is evaluated independently in model validation. Calves infected at or before birth enter into the population in the category 1 transient stage (Tr₁) rather than as susceptible young animals (X₁). Input of youngstock from higher or lower risk replacement herds can be modulated to account for different assumptions of open or closed herd status.

**Simulation studies**

State transition models were constructed in Modelmaker Version 4.0 software (Cherwell Scientific Ltd, Oxford, UK). Simulations were run to determine model output with a range of input parameters to test sensitivity of the model to our assumptions. All simulations follow an average herd during the introduction of MAP at t=0. MAP introduction occurs by the addition of two shedding animals into a naive herd. In Models B and C these are low shedders, while in Model A there is no distinction and all shedding animals are assumed to be equivalence. All herds are modeled as 100 animals appropriately divided into age categories where appropriate; however, as the model is insensitive to herd size, any size herd could be selected with the same output prevalence.
Output 1: Percent of the total herd infected with different ratios of infectiveness ($\varepsilon$) between low ($Y_1$) and high ($Y_2$) shedding adults. In this simulation there is no transient shedding, and all calves are assumed to be born susceptible and uninfected. Output from Models A, B and C when the ratio of infectiousness ($\varepsilon$) is 1 or 100 are shown in Panels A, B and C respectively. Lines are from simulations with no intervention strategies employed (grey: $\varepsilon=100$, black: $\varepsilon=1$). Symbols the same shade reflect disease transmission during a test-and-cull intervention strategy with the values listed in Table 1 (grey triangles: $\varepsilon=100$, black squares: $\varepsilon=1$).

Output 1 tracks prevalence of MAP infection in herds when the ratio of infectiousness ($\varepsilon$) between Low ($Y_1$) and High ($Y_2$) shedding is 1 or 100. Panel A is output from Model A, which allows disease persistence at both ratios of infectiousness when a test-and-cull intervention is modeled. Panels B and C, which are output
from models B and C respectively, do not allow disease persistence at a ratio of 1:100 during intervention. Results for a ratio of 1:10 produce the same decline in disease under a test-and-cull strategy with parameters outlined in Table 1 (data not shown). Disease prevalence when no intervention is introduced is similar for all ratios of infectiousness for the same model, but there is a large difference in prevalence between models B & C and model A.

**Output 2**: Model C transient shedding calves contribute to force of infection ($\beta_1$) at a value 0, 1 or 10 times that of adult cattle($\beta_2$). Herd level disease prevalence is tracked for all 4 values, with lines representing the simulations without an intervention strategy and symbols representing the same simulation with the addition of an active test-and-cull intervention.

Output 2 shows the impact of a potential infectious contribution ($\beta_1$) from transiently shedding ($\text{Tr}_1$ & $\text{Tr}_2$) juveniles in model C. The base model without transient shedding calves does not maintain disease under a test-and-cull elimination strategy in this model (which utilizes values listed in Table 1 for all parameters, with $\varepsilon=100$). Introduction of $\beta_1$ equivalent to $\beta_2$ does not prevent successful elimination. Not only does increasing the contact rate of calves with calves to 10 times the contact rate with infectious adults result in higher disease prevalence in a non-intervention setting ($\beta_1=80$ assuming equivalent infectiousness of an X-Y1 contact and an X-Tr contact) but it also results in stable disease prevalence at a low level despite an aggressive intervention.
Output 3: Model C and Model C1 are the most complex model with (Model C1) and without (Model C) vertical transmission. Addition of vertical transmission at different ratios of infectiousness ($\varepsilon$) of low and high shedding animals (1 and 100). Black lines (thin = without vertical transmission, bold = with vertical transmission) track herd level prevalence at $\varepsilon = 1$, and grey (symbols = without vertical transmission, bold line = with vertical transmission) at $\varepsilon = 100$.

The effect of vertical transmission (Output 3, Model C1) is a moderate increase in prevalence of infection. This increase in prevalence occurs at all ratios of infectiousness of low and high shedding animals. Without calf-to-calf transmission however, this increased prevalence does not mean the infection cannot be eliminated (data not shown).

DISCUSSION

The contribution of greater model complexity (B, C vs A) can be evaluated by examining its output. If a complex model produces output equivalent to a simpler version, we would select the simplest model to explore the system. Models B and C offered different results from Model A in the series of panels in Output 1. Based on this and other results (not shown), Model C’s output will be discussed.

Supershedders

Recent data examining colony counts of high shedding animals has indicated that there may be a Negative Binomial distribution of shedding levels of MAP among infected adults (Whitlock et al 8ICP). Even prior to exhibiting clinical signs, cows may be shedding in excess of $10^{10}$ bacteria/day. These animals contaminate the environment with massive amounts of bacteria and increase the chances for transmission. Intuitively, one would assume that removing these “super-shedding” animals should result in the virtual elimination of MAP from the farm. If high shedding animals were >100,000x more infectious than low shedders, the input of low shedders should be negligible in controlling infection spread. However, the series of models developed here (Output 1) conclude that these super-shedding animals, while they are responsible for the majority of new infections (potentially greater than 90% on a highly infected farm), are not responsible for all new cases on infection. If model inputs allow super-shedding animals to be even 100 times more contagious than low shedders, all models show that test-and-cull strategies would be an effective mechanism of infection control except the most basic (Output 1: Model A). Eliminating super-shedding animals would drop $R_{\text{effective}}$ below 1 and result in decline of MAP and its eradication over the course of several years (results not shown).
This model output indicates that although high shedders are much more infectious than low shedding animals their rate of contact with other animals limits transmission. High shedders are important in a high prevalence environment, where many calves are likely to come into contact with one, but removing high shedders is insufficient to address long term persistence of MAP. To understand sustained transmission in a test-and-cull environment we must look to other sources for potential contributions.

**Transient Shedding**

Dam-to-calf transmission alone is believed insufficient to maintain infection in a population. This was recently shown for *Neospora caninum* in dairy herds (French et al., 1999). Following contact with cows at birth, calves come into contact with other young animals more often than they do adults for the next twelve to twenty-four months of life. If young animals are excreting bacteria into the calf environment, there is a high risk of exposure and transmission to susceptible animals.

Peer-reviewed literature has described the biologic plausibility of young calves actively shedding MAP. (Taylor, 1953; Rankin, 1959; Collins and Zhao, 1994; Waters et al., 2003). Rankin (1961) used an experimental infection transmission design to determine whether and when shedding would occur following natural calf-to-calf infection. While these animals may have been exposed to a greater dose of MAP relative to farm levels (maybe even the equivalent of high shedding adult animals), the experiment showed that animals infected from other shedding calves produced culture-positive samples within a short timeframe. Although there are no peer-reviewed studies of young animals with positive culture results due to natural vs. experimental infection, risk factors for MAP at a farm level include an association with group-housing young calves (Wells and Wagner, 2000; USDA, 2005). While other factors associated with group housing may contribute to this finding, infected young animals shedding MAP via pass-through or active shedding would explain this association.

When calf-to-calf transmission is modeled by giving an infectious value to a transient shedding group of young animals (models B and C), model output reaches a stable low prevalence MAP infection at a herd level even with an increased rate of infectiousness of Y2 animals (e). Herds which previously maintained high levels of disease through the presence of super-shedding adults could sustain infection at a low level with the input of this transient compartment. These transiently shedding young calves may contribute to the persistence of infection in herds when high shedding animals are eliminated.

**Dam-daughter transmission**

Fetuses from high shedding MAP-infected dams can culture positive from *in utero* culture studies (Seitz et al., 1989; Sweeney et al., 1992). Recent data analysis (Benedictus et al, 8ICP) of a dataset collected by Dr. Robert Whitlock has shown that calves born to low shedding or latent dams also carry a significantly increased risk of being test-positive as adults vs. calves from consistently fecal culture negative dams. This information presents a challenge from an eradication standpoint, as not only does the most recent calf born from a high shedding dam become a high risk animal, but any calf produced throughout her lifespan. Given this model assumes herd size is constant, over the lifespan of a dam one daughter is expected to survive and become a productive member of a herd. Addressing dam-to-daughter transmission requires an elimination strategy which traces an animal’s pedigree to cull daughters born early in the adult lifespan of an infected dam, but should not be excessively costly in comparison to other aggressive test-and-cull regimens. If high risk daughters are added to our most developed model (C) using the values calculated based on fetal infection and increased risk of infection with MAP positive dams, vertical transmission is sufficient to increase the stable threshold of infection by a factor of 1.2-1.5 (Output 3) depending on assumptions concerning relative infectiousness of low and high shedding dams. This increased infection prevalence would not be an impediment to control in the long term although it may be detrimental in the short term due to increased immediate culling. If there were no calf-to-calf transmission of infection, these animals could be removed before they had any impact on herd transmission dynamics. However, when the effect of vertical transmission is combined with the presence of infective transiently shedding animals (Tr11, Tr12), then these animals may have a real impact on infection spread.
CONCLUSION

All models demonstrated that “super-shedding” animals can have a significant impact on the prevalence of MAP infected animals in a herd, yet more complex models illustrate that these animals are not predominantly responsible for infection persistence. The maintenance of infection in a low prevalence herd that aggressively tests for and culls shedding animals is caused by other factors. Elimination of infection is a greater challenge in a model which reflects calf-to-calf transmission and dam-to-daughter fetal transmission for dams of unknown status. Further work with this model and field studies will provide opportunity to validate/invalidate the contribution of these sources of MAP to persistence in a dairy environment.

REFERENCES


