The association of lameness and mastitis with return-to-service oestrus detection in the dairy cow

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Abstract

Oestrus detection is an important part of maintaining efficient reproductive performance in dairy herds. Both lameness and mastitis are common diseases of dairy cows that may impact oestrus detection. A set of data from 28 herds identified as having good recording of clinical mastitis and lameness incidents was used for the study. Logistic regression was used to identify associations between disease episodes within 100 days of insemination and changes in the probability of re-insemination at either 18-24 or 19-26 days after an unsuccessful insemination. Population attributable risk was calculated to understand the impact these diseases may have at a herd level. Lameness 0-28 days after the first insemination of the interval decreased the odds of a re-insemination at an appropriate time by approximately 20%. Clinical mastitis 1-28 days prior to the first insemination of the interval increased the odds of re-insemination at the expected time by approximately 20%. The associations were similar for either inter-service interval outcome. Population attributable risk suggested that the effect of these diseases on the probability of re-insemination at the expected time at a population level would likely be extremely small.

Introduction

Efficient oestrus detection is essential to maintain good reproductive performance in dairy herds. The effectiveness of oestrus detection on a dairy farm can be measured in numerous ways. Some approaches focus on trends over time, for example the proportion of cows eligible for insemination that are inseminated in a 21 day period (21-day insemination risk). Other approaches look at the timing of inseminations, for example by days in milk or in relation to previous inseminations [1; 2]. Oestrus detection is also frequently divided into first insemination oestrus detection and return insemination oestrus detection (for subsequent inseminations). One measure of return oestrus detection is the proportion of cows that are re-inseminated at an appropriate interval (usually 18-24 days) from a previous, unsuccessful insemination. There is some evidence that longer intervals than

the traditionally accepted normal range of 18-24 are more common [3; 4], with Remnant and others [5] suggesting that an interval of 19-26 days may be more appropriate.

It is well accepted that disease in cattle will impact on their reproductive performance [6; 7]. Both mastitis and lameness are common problems in dairy cows. Clear negative associations with overall reproductive performance have been demonstrated for both clinical mastitis and elevated milk somatic cell counts (SCC) [8-11] and for lameness [12-15].

The associations of these diseases with overall reproductive performance could be related to effects on conception, oestrus detection or both. Clinical mastitis has been shown to reduce pregnancies per AI [16]. A similar reduction in conception rate has been demonstrated in cows with elevated somatic cell counts [17; 18]. Mastitis also has the potential to impact on the apparent (measured) oestrus detection efficiency by leading to embryonic death and irregular returns [19] or by direct effects on ovarian function [20; 21]. This includes potential impact on the apparent inter-ovulatory interval of the cow [22]. Similar findings have been demonstrated for cases of lameness, with evidence to support a decrease in conception rate in lame animals [14] and other studies showing a decrease in oestrus behaviour [23].

Whilst it is clear that both lameness and mastitis have a negative association with reproductive performance their impact on return oestrus detection specifically has not been evaluated on a large scale. The aim of this study was to explore and quantify the impact of lameness and mastitis on return oestrus detection at an individual cow level as well as exploring the impact of a different "expected" interval on any apparent associations.

Materials and methods

Data collection and organisation

Farm management data were collected as part of a wider project [10; 24]. Data were contributed by 20 farm animal veterinary surgeons from across England and Wales from a total of 468 dairy herds

considered to have good quality records. These data were converted into a common format and screened for fertility data quality before selectingherds that contained regular lameness treatment records, clinical mastitis records with a plausible incidence rate and consistent recording and milk recording data collected at a regular monthly interval. These data were structured so that each insemination was a single line of data along with the animal and herd identity, cow parity, days in milk, 305 day milk yield for that lactation, the number of inseminations so far that lactation, the year the cow calved and the month of the insemination. Lameness and clinical mastitis records kept according to normal farm detection and recording procedures were converted to an interval in days from each disease event to the insemination. These disease records were then converted to binary categories by timeframe relative to the insemination (whether there was a case of clinical mastitis or lameness 29-100 days before the insemination, 1-28 days before the insemination, 0-28 days after the insemination and 29-100 days after an insemination). Neither clinical mastitis aetiology or lameness lesion identification were collected. Where milk recording was carried out within the period 31 days prior to the insemination and 31 days after the insemination, the individual cow SCC both before and after the insemination were recorded and the natural logarithm of SCC treated as a continuous variable. SCC status was also categorised based on whether the SCC before and after insemination stayed below 200,000 cells/ml (uninfected), passed from below 200,000 cells/ml (new infection), decreased from above 200,000 cells/ml to below (cure) or stayed above 200,000 cells/ml (chronic). The interval to the next insemination in that cow in that lactation was calculated in days (Inter-service interval, ISI). Any inseminations not followed by a subsequent insemination, likely to be due to pregnancy or culling, were excluded from the data, as were ISIs of 1 or 2 days as these were considered likely be related to the same oestrus event. ISIs of over 200 days were also excluded as they were considered likely to represent recording errors or abortion events. As a result, only cows where there was an apparent intention to re-inseminate were included in the study. Two binary outcome variables were calculated from this ISI corresponding to whether the ISI was within the expected range of 18-24 days, and whether it was within the alternative range 19-26 days as a

measurement of return oestrus detection. The final data set contained 19,011 inseminations for 6,749 cows calving between 2000 and 2008 from 28 dairy herds.

Regression modelling

Logistic multivariable regression models were built with the outcome representing whether or not a cow received a re-insemination at the expected interval. Two similar models were fitted, one with an expected interval of 18-24 days as the outcome and one with an expected interval of 19-26 days. Herd was included as a random effect to account for variation in herd level oestrus detection efficiency. A cow-level random effect was also tested, but model fit was poor when assessed using a modified Hosmer-Lemeshow approach, and so a two level structure was used (inseminations within herds). Both models were built by stepwise forward selection, with each variable being offered to the model, and retained if the magnitude of its estimated coefficient was at least double the standard error of the estimate (equivalent to $p<0.05$). All rejected variables were re-offered to the final models, and retained if they met the criteria described above. Variables offered to the models are shown in Table 1. Biologically plausible interactions with the variables of interest (significant lameness and clinical mastitis variables with milk yield and month) were also tested. The model took the conventional form

$$re-insemination_{ij} \sim Bernoulli\left(mean = \pi_{ij}\right) \quad (1)$$

$$\ln\left(\frac{\pi_{ij}}{1-\pi_{ij}}\right) = \beta_0 + \boldsymbol{\beta}\boldsymbol{x}_{ij} + u_{0j} \quad (2)$$

$$u_{0j} \sim N(0, \sigma_{u0}^2) \quad (3)$$

where re-insemination$_{ij}$ is whether the ith insemination in the jth herd was followed by a re-insemination at either 18-24 days (model 1) or 19-26 days (model 2); π_{ij} is the fitted probability of re-insemination$_{ij}$; β_0 is the regression intercept, $\boldsymbol{\beta}$ is the vector of coefficients for the vector of predictor variables $\mathbf{x}$; u_{0j} is the random effect to represent herd level variation.

The model was fitted using MLwiN version 2.35 [25]. Initial parameter estimates were calculated using iterative generalised least squares (IGLS) and final parameter estimates generated using a Bayesian approach, Markov Chain Monte Carlo (MCMC) with Gibbs sampling [26; 27]. A burn-in length of 1,000 iterations was used followed by a monitoring chain of 10,000 iterations. MCMC chains for the parameter estimates were visually checked to ensure adequate convergence. Model fit was checked by comparing observed and predicted number of re-inseminations for each decile of risk using a modified Hosmer-Lemeshow approach [28].

To aid in interpretation, each model was used to predict the probability of a cow receiving a re-insemination at the expected interval with and without cases of lameness and clinical mastitis, with all other variables fixed at their population means. Predictions were illustrated using bar charts, showing the mean predicted effect as well as the 95% credible interval around the mean prediction.

Table 1 Variables tested for inclusion in a logistic regression model with the outcome of whether a re-insemination occurs at the expected interval

Variable	Type
Parity	Categorical (1,…,4+)
Days in milk at insemination	Continuous
305 day lactation milk yield	Continuous
Year of calving	Categorical (2000,…,2007)
Month of insemination	Categorical (Jan,…,Dec)
Insemination number	Categorical (1,…,5+)
Lameness 29-100 before insemination	Binary
Lameness 1-28 before insemination	Binary
Lameness 0-28 after insemination	Binary
Lameness 29-100 after insemination	Binary
Clinical mastitis 29-100 days before insemination	Binary
Clinical mastitis 28-1 days before insemination	Binary
Clinical mastitis 0-28 days after insemination	Binary
Clinical mastitis 29-100 days after insemination	Binary
Log_e SCC before	Continuous
Log_e SCC after	Continuous

SCC Status	Categorical (Uninfected, new infection, chronic infection, cure)
Inseminated again at 18-24 days?	Binary outcome (traditional)
Inseminated again at 19-26 days?	Binary outcome (modified)

The distribution of intervals across the traditional inter-service interval categories[1; 29] was calculated for inseminations with and without any clinical mastitis and lameness cases in the timeframes retained in the regression model.

Population attributable risk

To aid understanding of the potential effect of disease at a population level accounting for effect size and prevalence of lameness and mastitis, the population attributable risk was calculated [30]. A prediction was produced for each insemination in the dataset at each iteration of the MCMC chains. The process was then repeated to give a predicted outcome for each insemination in a hypothetical situation where there was no mastitis (i.e. a posterior prediction where every line of data was changed to have no case of clinical mastitis in the 28 days prior to the first insemination). The median values and 95% confidence intervals of these posterior predictions were calculated and compared. The same process was repeated for lameness variables, giving three sets of posterior predictions – the study population as it was, the study population in a hypothetical situation with no mastitis and the study population in a hypothetical situation with no lameness.

Results

Descriptive data

Of the 19,011 inseminations included in the analysis, 7,693 (40.5%) were followed by an insemination at 18-24 days and 8,741 (46.0%) were followed by an insemination at 19-26 days. The mean 305 day yield for lactations included in the final analysis was 8,854 litres. There were 1,188 inseminations with a case of lameness recorded 29-100 days before, 565 inseminations with a case 1-28 days before, 656 inseminations with a case 0-28 days after and 1,615 inseminations with a case

of lameness recorded 29-100 days after. There were 2,157 inseminations with a case of clinical mastitis recorded 29-100 days before, 902 inseminations with a case 28-1 days before, 982 inseminations with a case 0-28 days after and 1,843 inseminations with a case of clinical mastitis recorded 28-100 days after. These are summarised in Table 2.

Table 2 The number of inseminations with a disease incidence recorded for each condition and each time period used in the analysis, total number of inseminations in the analysis was 19,011

	Time relative to first insemination of interval			
Condition	**29-100 days before**	**1-28 days before**	**0-28 days after**	**29-100 days after**
Lameness treatment	1188	565	656	1615
Clinical mastitis	2157	902	982	1843
Both	156	20	30	160

Regression modelling

The parameter estimates for each of the regression models are shown in Table 3. There was no significant association of 305 day milk yield, days in milk or year of calving with either outcome. Parity was not significantly associated with the probability of re-insemination when using 19-26 day ISIs as an outcome but was when using 18-24 days. In both the 18-24 day model and the 19-26 day model there was a significant negative association of lameness treatments carried out 0-28 days after the first insemination. Odds of re-insemination at 18-24 days were reduced by 18% and those of re-insemination at 19-26 days by 17%. There was no significant association with lameness treatments occurring in other time periods. There was a significant positive association of clinical mastitis recorded 1-28 days before the first insemination, with the odds of re-insemination at 18-24 days increased by 21% and those of re-insemination at 19-26 days by 19%. There was no significant association with clinical mastitis at other time periods or with any of the representations of SCC.

Predicted probabilities from the models are illustrated in Figure 1. Effect sizes were similar in both models. Effect sizes for month of service and insemination number were similar between the two models, with increasing numbers of previous inseminations having a positive association and August having a negative association on the probability of re-insemination.

The distribution of inter-service intervals across the traditional categories is shown in Table 4.

Population attributable risk

The effect of both lameness and mastitis were considered similar in both models and so population attributable risk was calculated for model 2 (19-26 day interval outcome). The median predicted probability of being re-served at 19-26 days was 45.3% (95% credible interval 29.3-66.6%). In a hypothetical scenario, the same population with no cases of lameness in the 0-28 day window after an insemination would lead to a probability of 45.4% (95% credible interval 29.5-66.7%), the same population with no cases of clinical mastitis in the 1-28 days window before an insemination would lead to a probability of 45.1% (95% credible interval 29.2%-66.3%).

Table 3 Model parameters and odds ratios from two logistic regression models predicting whether an insemination is followed by another insemination at the expected interval

Variable	Model 1 (re-inseminated at 18-24 days)		Model 2 (re-inseminated at 19-26 days)	
	Coefficient (Standard error)	Odds ratio (95% credible interval)	Coefficient (Standard error)	Odds ratio (95% credible interval)
Intercept	-0.5 (0.09)		-0.41 (0.08)	
Parity 1	reference category		reference category	
Parity 2	-0.01 (0.04)	0.99 (0.91-1.08)	not significant	
Parity 3	-0.1 (0.05)	0.91 (0.83-0.99)	not significant	
Parity 4+	-0.18 (0.04)	0.84 (0.77-0.91)	not significant	
January	reference category		reference category	
February	-0.07 (0.07)	0.93 (0.81-1.07)	-0.05 (0.07)	0.95 (0.83-1.09)
March	-0.1 (0.07)	0.91 (0.79-1.04)	-0.14 (0.07)	0.87 (0.76-0.99)
April	-0.09 (0.07)	0.92 (0.79-1.06)	-0.06 (0.07)	0.94 (0.82-1.08)
May	-0.02 (0.08)	0.98 (0.84-1.13)	0.01 (0.07)	1.01 (0.87-1.16)
June	-0.11 (0.08)	0.89 (0.77-1.04)	-0.15 (0.07)	0.87 (0.75-0.99)
July	-0.07 (0.07)	0.94 (0.81-1.08)	-0.07 (0.07)	0.94 (0.81-1.08)
August	-0.21 (0.08)	0.81 (0.69-0.94)	-0.24 (0.08)	0.79 (0.68-0.91)
September	0.05 (0.08)	1.05 (0.9-1.21)	0.04 (0.07)	1.04 (0.9-1.19)
October	0.03 (0.07)	1.03 (0.89-1.18)	0.05 (0.07)	1.05 (0.91-1.2)
November	-0.02 (0.07)	0.98 (0.85-1.12)	-0.01 (0.07)	0.99 (0.87-1.13)
December	-0.03 (0.07)	0.97 (0.84-1.11)	0 (0.07)	1 (0.88-1.15)
First insemination	reference category		reference category	
Second insemination	0.2 (0.04)	1.22 (1.14-1.31)	0.26 (0.04)	1.29 (1.2-1.39)
Third insemination	0.35 (0.04)	1.42 (1.31-1.55)	0.43 (0.04)	1.53 (1.4-1.67)
Fourth insemination	0.56 (0.06)	1.74 (1.55-1.96)	0.66 (0.06)	1.93 (1.72-2.17)
Fifth+ insemination	0.88 (0.07)	2.4 (2.09-2.76)	0.91 (0.07)	2.48 (2.16-2.84)
Not lame 0-28 days post insemination	reference category		reference category	
lame 0-28 days post insemination	-0.2 (0.09)	0.82 (0.69-0.97)	-0.19 (0.08)	0.83 (0.71-0.97)
no CM 28-1 day previous	reference category		reference category	
CM 28-1day previous	0.19 (0.07)	1.21 (1.05-1.39)	0.17 (0.07)	1.19 (1.03-1.36)
Herd level random effect	0.08 (0.03)		0.08 0.03)	

Table 4 The distribution of inter-service intervals presented using the traditional categories relative to the expect return to oestrus for a cow with and without disease incidences

Inter-service interval category (days)	**<18**	**18-24**	**25-35**	**36-48**	**>48**
Lameness within 28 days after the first insemination	7.3%	35.5%	15.7%	23.8%	17.7%
Mastitis in the 28 days preceding the first insemination	8.4%	44.2%	17.3%	14.9%	15.2%
Neither	8.0%	40.5%	17.1%	17.5%	16.9%

Discussion

Cases of both lameness and clinical mastitis appear to be associated with the probability of a cow being re-inseminated at the expected time after an unsuccessful insemination. Cases of lameness after the first insemination of the interval are associated with a reduced risk of re-insemination at the expected interval. Cases of clinical mastitis before the first insemination of the interval are associated with an increased risk of subsequent insemination at the expected time. These associations were extremely similar whether the traditional (18-24 day) or modified (19-26 day) expected interval was used. The effect of disease on the probability of re-insemination at the expected interval was statistically significant at an individual cow level, with a decrease in odds of nearly 20% and an increase in odds of re-insemination of 20% for lameness and mastitis cases respectively, however the impact was much smaller at a population level. This is supportive of other work in this area suggesting that at a herd level reducing lameness or mastitis is unlikely to have a clinically relevant increase in herd level reproductive performance [11; 15]. It is worth noting that herds with this level of data recording may have higher health performance than average and the impact of disease is likely be higher in herds with very high lameness prevalence or mastitis incidence (in this study average lactation level incidence of mastitis was 26% and lameness was 19%). This is

because the population attributable risk calculated in this work uses the prevalence of disease in the study population to estimate the impact of eliminating that disease.

The apparent negative association of lameness at cow level with oestrus detection is supported by the existing literature. Lame cows have a longer calving to first service interval [31]; have been shown to spend more time lying and less time standing, walking and expressing oestrus behaviour [23] and appear to express oestrus less intensely [32]. Lameness has been shown to reduce the time oestrus cows are mounted by their herd mates and to reduce the intensity of oestrus behaviour, Walker and others [33] showed that lame cows were approximately a third less likely to be observed in oestrus. These studies suggest an explanation for the temporal relationship between lameness cases and oestrus detection identified in the current study. If the presence of lameness reduces the expression of oestrous behaviour then the greatest impact on return oestrus detection will occur when the lameness occurs before the second oestrus is due. The current study has shown a relatively small effect size at an individual cow level, with the probability of re-insemination at the expected time decreasing by approximately 10% (Figure 1).

It is likely that this represents a conservative estimate as the current study relied on farmer recorded lameness treatments. It has been shown that farmer recorded lameness treatments often represent an underestimate due to delays in detection and treatment [34-36]. This may have resulted in the misclassification of lame animals in this dataset, with some lame animals being recorded as non-lame because they were not treated or recorded as lame. Therefore some of the true effect of being lame may have been 'absorbed' in to the non-lame category, reducing the odds ratio. Conversely, it is also possible that when fewer cases are recorded that these only represent the most severe ones, potentially leading to the overestimation of the association of return insemination submission rate and lameness. This delayed treatment and recording may also mean that the temporal association of the onset of lameness may be different to the temporal association with lameness treatments.

Further studies examining the relationship between oestrus detection and lameness using mobility score data are warranted.

The positive association of clinical mastitis cases with the probability of a return insemination at the expected interval is harder to explain. Clinical mastitis has been shown to reduce reproductive performance in dairy cows [8; 37] although other studies have found no effect [6]. Moore and others [22] found that clinical mastitis resulted in a greater number of abnormal interoestrus intervals, defined as those falling outside of the expected 18-24 day range. However, this was not consistent and of the two herds studied, the effect was much stronger in the herd with predominantly gram negative mastitis cases. Another possible explanation for the difference in findings is the study methodology. In contrast to the current study, Moore and others [22] looked for the interval around a case of clinical mastitis (i.e. with the case occurring after the first insemination and before the second). In the current study the only significant association of return oestrus detection with clinical mastitis was when the case of mastitis occurred prior to the first insemination of the interval. Clinical mastitis has consistently been shown to reduce the chance of conception and maintenance of pregnancy after artificial insemination [8; 16]. A possible explanation for the positive association detected in the current study is that the case of clinical mastitis reduced the chance of conception at the first insemination. This could potentially reduce the likelihood of an abnormal return to oestrus due to late embryonic death because the mastitis case prior to insemination may have prevented fertilisation occurring at all, or may cause pregnancy failure before maternal recognition of pregnancy leading to a “normal” cycle length. Alternatively, Hockett and others [38] demonstrated that some cows with experimentally induced mastitis failed to express oestrus behaviour and that in these cows cyclicity was abnormal. It is possible that in the current study, using return oestrus detection as an outcome selected for cows that cycled normally in the presence of clinical mastitis (i.e. cows with clinical mastitis prior to the first insemination would not have had a first insemination if it affected their oestrous cycle). It is important to note by eliminating inseminations not followed by another insemination in this study,

the impact of disease on oestrus detection and not on establishment of pregnancy is being measured.

There was a very similar association of both clinical mastitis and lameness with the traditional (18-24 day) and modified (19-26 day) outcome interval. This suggests that these approaches are comparable and that neither is more or less sensitive to the effect of disease on return oestrus detection as well as suggesting that these associations are not a result of the interval that is selected. It is also suggestive that embryonic death is not the cause of the longer 'expected' interval identified in previous work, as it would seem likely that if it were, the impact of disease would vary more with the chosen expected interval. Interestingly, the confounding of parity when using the traditional interval did not occur when using the modified interval.

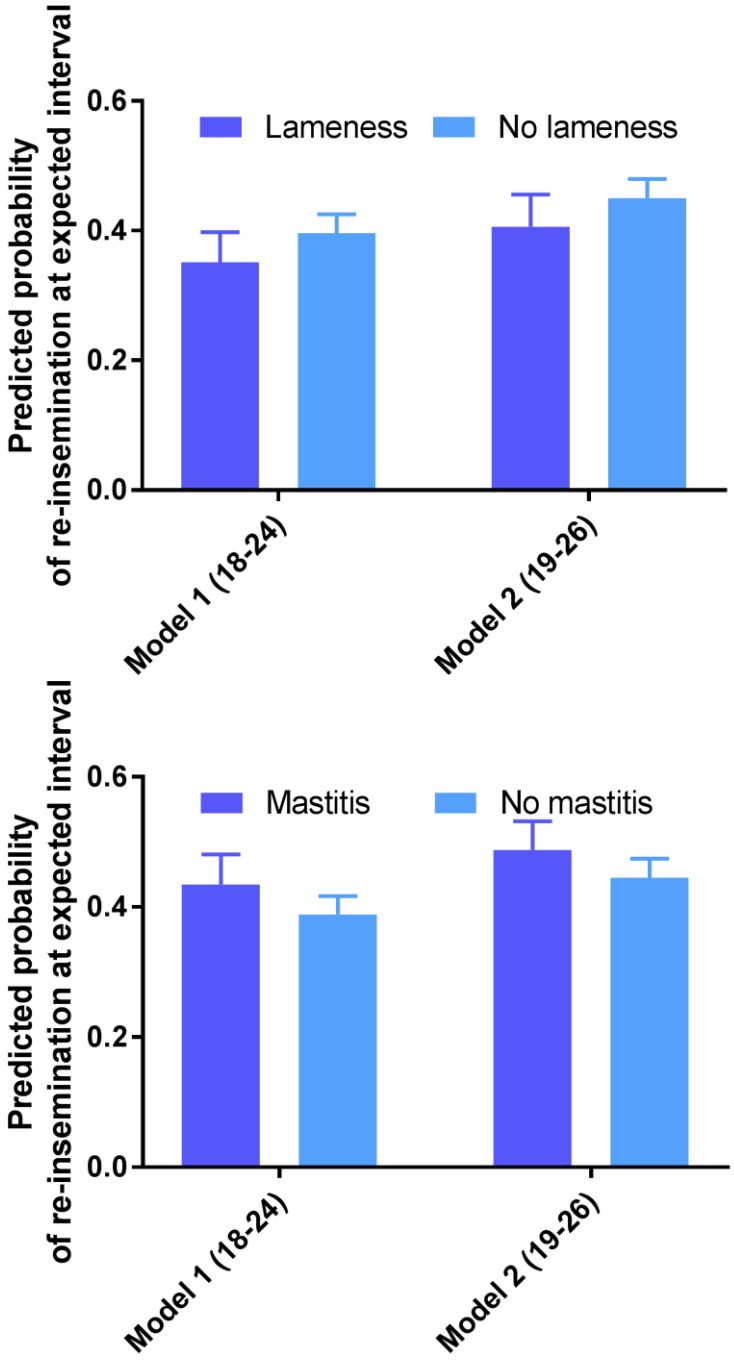
These findings highlight the value of presenting results as predicted relative risk in addition to odds ratios. Classically, findings from logistic regression models are presented as odds ratios (as in Table 3), as these are easier to calculate directly from the model coefficients. However, odds ratios can be harder to interpret as humans tend to find probability and risk (as shown in Figure 1) more intuitive than odds [39]. In this study the odds ratios show a change of about 20% whereas the predicted risk only changes by about 10%. This difference between odds ratio and relative risk is typical for studies such as this where baseline risk is high (approximately 50% in this study) [40]. The population attributable risk is then useful to put these findings in the context of the whole population [41].

Conclusion

Cases of lameness and clinical mastitis are respectively, negatively and positively associated with re-insemination at the expected time at an individual cow level. At a population level the impact of these conditions on return oestrus detection appears very small. These associations are very similar whether a traditional expected interval of 18-24 days or a modified expected interval of 19-26 days is used as the outcome.

Figures

Figure 1 Bar chart showing the predicted probabilities and 95% credible interval from two logistic regression models predicting the probability a cow is re-inseminated at 18-24 (model 1) or 19-26 (model 2) days following an insemination for cows that have a case of lameness within 28 days after the first insemination (top) or a case of clinical mastitis in the 28 days preceding the first insemination (bottom)



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