[Type here]

INTERPRETATIVE SUMMARY

Association of days in close up, gestation length and rumination around time of calving with disease and pregnancy outcomes in multiparous dairy cows. *By Cook et al.* In this study we showed the importance of days spent in close up and gestation length on early lactation health outcomes and demonstrated that early lactation health issues and rumination around calving can be used as predictors of pregnancy outcomes in later lactation. Understanding the importance of days spent in close up, gestation length and rumination around calving offers the potential for dairy producers to plan and adapt management strategies to avoid health issues that lead to poor pregnancy outcomes.

RUNNING HEAD: DAYS IN CLOSE UP, GESTATION LENGTH, RUMINATION TIME AND PREGNANCY OUTCOME

Title: Association of days in close up, gestation length and rumination around time of calving with disease and pregnancy outcomes in multiparous dairy cows

J. G. Cook *[1], P. T. Pepler †, L. Viora ‡

*** World Wide Sires, Yew Tree House, Carleton, Carlisle, Cumbria, CA1 3DP, United Kingdom**

† Institute for Biodiversity Animal Health and Comparative Medicine, College of Medical, Veterinary and Life Sciences, University of Glasgow, Glasgow, G12 8QQ, United Kingdom

‡ **Scottish Centre for Production Animal Health and Food Safety, School of Veterinary Medicine, College of Medical Veterinary and Life Sciences, University of Glasgow, Bearsden Road, Glasgow G61 1QH, United Kingdom**

[1] Corresponding author: John G. Cook: john.cook14@btinternet.com

ABSTRACT

The purpose of this study was to evaluate the effect of rumination times and the days spent in a close-up group prior to calving (DINCU) on early lactation health and reproductive outcomes in dairy cows. Data was gathered for 719 cows located in a single herd. Herd management and reproductive records were analysed for cows receiving treatment in the first 30 d of lactation (days in milk, DIM) for clinical mastitis, reproductive tract disease, ketosis, milk fever and displaced abomasum. Daily rumination times for each cow were downloaded daily from the herds automated collar system used to generate heat and health alerts for each cow beginning at 21 d pre-calving until 14 d post calving. During the first 30 DIM 121 cows (18%) developed at least one disease—any combination of ketosis (40 cows, 5.9% of total), mastitis (17 cows, 2.5%), metritis (75 cows, 11%), milk fever (17 cows, 2.5%), or displaced abomasum (28 cows, 4.1%), whilst 305 cows (45%) were pregnant again at 100 DIM, and an additional 139 cows (20%) were pregnant at 150 DIM. Principal component analysis was used to determine the relationship between gestation length (GL) and DINCU and their association with the odds of developing disease in early lactation. We did not find any significant association between pre-calving rumination time and disease within the first 30 DIM. Higher rumination time in the week prior to calving was shown to be strongly linked to a shorter time to subsequent pregnancy whilst rumination times post calving were not associated with changes in the time to pregnancy. Principal component analysis showed that a curvilinear combination of GL and DINCU (first principal component, PC1) was significantly associated with changes in disease incidence in the first 30 DIM. Gestation length and time spent in close up are important management factors in reducing the incidence of disease in early lactation, and rumination times around calving may help predict future reproductive outcomes.

Key words: days in close up, rumination time, pregnancy outcome, transition management, post-partum diseases

INTRODUCTION

The transition period has been identified as a time when cows are at increased risk of infectious (Cai et al., 1994; Pinedo et al., 2020) and metabolic diseases (Goldhawk et al., 2009; Venjakob et al., 2019; McArt and Neves, 2020). Greater than 90% of production diseases occur during the transition period (Ingvartsen et al., 2003) and over 50% of dairy cows become ill in the first 60 d of lactation (DIM), often suffering from multiple disorders (Santos et al., 2010). In addition, many post-partum diseases are often subclinical or may be so mild as to go unnoticed for extended periods of time despite having significant effects on the well-being and future productivity of the cow (Goff and Horst, 1997; Goldhawk et al., 2009, Santos et al., 2013).

Feed intake tends to be reduced around the time of calving with a 30% reduction in the 5-7 d prepartum (Bertics et al., 1992; Grummer et al., 2004), preceded by a more gradual decline beginning 3 wk before calving (Drackley, 1999). The factors influencing eating and rumination behaviour in dairy cows have recently been reviewed by Beauchemin (2018). Alongside feed intake, rumination diminishes prepartum and reaches a nadir on the day of calving (Soriani et al., 2012) or on the following day (Schirmann et al., 2012). However, rumination time and the prepartum decrease in rumination activity are subject to large individual variation (Soriani et al., 2012; Büchel and Sundrum, 2014). Daily rumination time recovers within 7 to 15 days postpartum, stabilising at levels seen before the prepartum decline (Soriani et al., 2012; Calamari et al., 2014).

Close monitoring of cows during the transition period, which includes monitoring of eating and rumination, may aid in early detection of subclinical problems and thus timely initiation of treatment or management change. Automated technologies for monitoring rumination behaviour are becoming more widely adopted and several studies have investigated their use as tools for early diagnosis of impending parturition (Schirmann et al., 2013) and peripartum health disorders (Liboreiro et al., 2015; Kaufmann et al., 2016; Kaufmann et al 2018). Management factors also influence transition cow behaviour (Grant and Albright, 1995; Huzzey et al., 2006; Proudfoot et al., 2009) and were identified as risk factors for disease post-partum (Huzzey et al., 2005, Huzzey et al., 2007, Neave et al., 2018). More recently Huzzey et al. (2014) showed that automated technologies can be used to quantify management risk factors likely to lead to post-partum disease, whilst the effect of periparturient subclinical and clinical hypocalcaemia on periparturient rumination time in cows entering their third or greater lactation are strongly correlated (Goff et al., 2020). Moreover, consistent negative changes in rumination activity were observed for each health condition during the 7 d prior to diagnosis (Paudyal et al., 2018).

Animal grouping strategies, and in particular group moves, influence aspects of cow behaviour and it is known that feeding behaviour can be altered for several days following a group change (Grant and Albright 2001). Newly introduced cows must establish a ranking in the group, and it appears reasonable that a longer stay in a pre-calving or close up group may be more beneficial than a shorter stay (Nordlund et al., 2006; Robinson et al., 2001). More recently the effect of gestation length (Viera-Neto et al., 2017) and deviations in dry period length (Olagaray et al., 2020) have been shown to influence health and performance in dairy cows. Shortened dry periods occurring for biological reasons had a greater negative effect than those occurring for management reasons whilst lengthened dry period occurring for management reasons had a greater negative effect. The negative effect of a shortened dry period occurring due to a shortened gestation length is likely to be a response to physiological stressors such as cows carrying twins or subject to heat stress in late pregnancy that also lead to impaired health and milk production. A lengthened dry period will carry an increased risk of excessive body condition and a metabolic scenario associated with poor DMI that would lead to an increased risk of metabolic disease in early lactation (Olagaray et al., 2020). Clearly the effects of DINCU may be confounded by gestation length as cows with shorter gestations are likely to experience less DINCU and vice versa.

Reproductive performance greatly influences dairy herd profitability (Louca and Legates, 1968; Oltenacu et al., 1981; Giordano et al., 2011; 2012) by its effect on revenue from milk sales and replacement costs. The timing at which pregnancy occurs is important in sustaining profitability, ideally occurring between 90 and 130 DIM (Giordano et al., 2011).

The biological environment in the relatively long period of follicular growth prior to ovulation was suggested to be a major factor contributing to sub-fertility in the dairy cow (Britt, 1992); however, to date only the long-term effect of heat stress had been demonstrated (Chebel et al., 2004). The mechanisms by which metabolic changes may influence fertility were reviewed (Leroy et al., 2008 a, b) and it has been shown that uterine infection creates perturbations in ovarian function that have residual effects (Williams et al., 2007). These have only been investigated in the short term as studies examining long-term effects are difficult to conduct (Leroy et al., 2008a). More recently Carvalho et al. (2014) demonstrated a relationship between changes in body condition score as a measure of energy balance and reproductive efficiency. Ribeiro et al. (2016) showed that the occurrence of disease at the preantral or antral stages of ovulatory follicle development was detrimental to reproduction. Moreover, it seems likely that metabolic changes and infection and inflammation around the time of calving may bear some responsibility for the biological environment that leads to sub-fertility in the cow, and that these changes in turn may be influenced by behavioural and management changes (Ribeiro et al., 2016).

While the association between eating time around parturition and reproductive performance has been explored (Hut et al., 2019), no studies have looked at the relationship between rumination time in the transition period and subsequent reproductive performance.
The objectives of this study were to evaluate the ability to predict the likelihood of disease occurring in early lactation and the timing when pregnancy occurs using differences in rumination times around calving and the timing of a pre-calving group change. Of particular interest was the ability to predict the occurrence of disease in the first 30 DIM, and the timing of the occurrence of pregnancy during the subsequent lactation. Our hypothesis was that rumination times around calving, and the timing of a pre-calving group change could be associated with reproductive performance and the occurrence of early lactation disease in the subsequent lactation.

MATERIALS AND METHODS

Herd Management

Data was gathered for 719 Holstein cows calving between December 1, 2017 and May 31, 2018, located in a single dairy herd in the United Kingdom. The herd was housed all year round, milked 3 ×⁄d and fed a TMR 1 ×⁄d whilst the far-off dry cow group and close to calving group were each fed a separate TMR 1 ×⁄d. All rations were formulated to meet animal requirements (NRC, 2001), details are shown in Table 1. Cows were scheduled to be dried off into a far-off dry cow group 47 to 53 d prior to the predicted calving date (227 to 233 d of gestation) and then moved to a close to calving group 21 to 27 d (253 to 259 d of gestation) prior to the predicted calving date. Between 14 and 21 d prior to calving selected cows were given a controlled release monensin bolus (Kexxtone, Elanco UK, Basingstoke, UK). Selection for treatment was based on a body condition score in excess of 3.5. Predicted calving date was based on a calculation in the herd management software with an assumed gestation length of 280 d. All dry cows were housed in deep bedded straw yards. Dry off and cow movements were carried out weekly on a Tuesday. The herd practiced ‘just in time’ calving. When either the amniotic sac or calf’s feet were visible at the vulva, animals were moved to individual straw pens; 24-hour calving supervision was provided. Post calving cows entered a fresh pen for a minimum of 7 d to facilitate monitoring of health conditions post calving before being moved into the main herd accommodation. Fresh cows and the main milking groups were housed in deep sand bedded free stalls.

The herd practiced a structured approach to health management with all cows being observed daily for assessment of appetite, made either by observing the cow to feed at least once daily or by daily assessment of the degree of rumen fill and ruminating behaviour. Rumen fill was assessed by observing the cows left flank from the rear according to the methods described by Zaajer and Noordhuizen (2003). The foremilk and udder of every cow in each herd was examined at least once daily and any quarter identified with abnormal milk received a course of antibiotics (150 mg penethamate hydriodide, 185 mg dihydrostreptomycin sulphate, 50 mg framycetin sulphate, 5 mg prednisolone, Tetra Delta, Zoetis, Leatherhead, UK, daily for 5 d) administered via the intramammary route and had a mastitis event recorded. Additional adjunct therapies (250 mg meloxicam, Metacam 20mg/mL, Boehringer Ingelheim Animal Health UK Ltd, Bracknell, UK) were given based on an assessment of the degree of udder inflammation as assessed by palpation of heat, swelling and degree of pain of affected quarters.

Cows generating health alerts using the proprietary index provided via the herd management software (DataFlow II, SCR Engineers Ltd, Netanya, Israel) were given a more detailed examination including recording of rectal temperature and assessment of any post-natal vulval discharges, which were further categorised based on the presence or absence of a foul-smelling odour. Detailed examination included measurement of whole blood βhydroxybutyrate (**BHB**) by taking a blood sample from the coccygeal vessels using a syringe and needle and immediately applying it to a handheld meter (Precision Xtra, Abbott, Alameda, USA). Any cow generating a health alert and demonstrating a combination of any two symptoms of pyrexia, poor appetite and malodorous discharge was scheduled to receive a course of systemic antibiotics (6000 mg procaine penicillin and 7500 mg dihydrostreptomycin, sulphate Pen & Strep, Norbrook Laboratories Ltd, Corby, UK, daily for 3 d) and reported as a metritis event. Milk fever was recorded when a cow displayed clinical signs that include muscle weakness, nervousness, muscle shaking, cold ears, eventually leading to an inability to rise. Displaced abomasum was recorded when a ping was detected by thumping or tapping the cow's body wall while simultaneously listening with a stethoscope in the area between the 9th and 12th ribs above and below an imaginary line extending from the hip to the elbow on each side of the animal on the abdominal wall. Ketosis was reported when a cow was identified with elevated BHB in the blood (> 1.2 mmol/L), and in the absence of other signs of concurrent disease. Data were extracted from the herd management software for any cow treated for any of ketosis, mastitis, metritis, milk fever and displaced abomasum in the first 30 DIM.

All health assessments were carried out by farm staff. The medicines and treatment protocols were in accordance with the written instructions and training provided by the herd veterinarian.

Health and treatment protocols were verified by the first author at a single herd visit prior to study commencement.

Reproductive management consisted of an automated heat detection program using neck mounted collars (Hi-Tag, SCR Engineers Ltd., Netanya, Israel) to identify and inseminate cows in heat based on changes in cow activity. Cows were considered eligible for insemination from 50 d post calving and all cows were enrolled in a Presynch-11 program

(Galvão et al., 2007) with treatments scheduled to ensure all cows eligible to be inseminated receive a first AI by 78 d of lactation (DIM). Briefly, cohorts of cows were enrolled weekly at 40 ± 3 DIM and 54 ± 3 DIM to receive an injection of $PGF_{2\alpha}$ (500 µg cloprostenol sodium, Estrumate 250 µg/mL, MSD Animal Health, Walton Manor, UK) and then enrolled into a timed AI (**TAI**) program 11 d later which consisted of an injection of GnRH (10 µg buserelin acetate, Receptal 4 µg/mL, MSD Animal Health, Walton Manor, UK) followed 7 d later by an injection of $PGF_{2\alpha}$ and then approximately 56 h later by a further injection of GnRH with TAI being performed approximately 16-20 h after the final GnRH injection. Those cows identified in oestrus between the second $PGF_{2\alpha}$ and the commencement of TAI were inseminated and withdrawn from the Presynch-11 program whilst cows identified as open at pregnancy were immediately re-enrolled to the TAI program. Pregnancy diagnosis was routinely carried out by transrectal ultrasonography on all cows at 34 to 40 d since last insemination and confirmed at 69 to 75 d.

Rumination monitoring and data collection

Daily rumination times were monitored 24 h/d using the same neck mounted collar used for oestrus detection as validated by Schirmann et al., (2009). The collars contained a small microphone that recorded each time a bolus was regurgitated, re-masticated, and swallowed to determine total time spent ruminating during each 2-h interval throughout the day. This information was transferred to the control unit via radio frequency. Data were backed up from the control unit and downloaded to the database daily. The twelve 2-h intervals each day were summed to determine total time spent ruminating per day per cow. If any 2-h interval was not recorded by the system, the entire day was reported as a missing value. Data were downloaded from 21 d prior to the date of the first cow calving and continued until 14 d after the final calving date to create a record of each cow’s daily rumination activity from 21 d prior to 14 d after calving.

In addition to daily rumination times, individual cow health and reproductive management data were downloaded. For each cow the unique identity, date of calving, lactation number, date of dry off, date of movement to the close up pen, date of administration of monensin bolus, and date of all health events including details of all medical treatments administered in the first 30 DIM were extracted and entered into a spreadsheet (Excel, Microsoft, Redmond, WA). Copies of hand-written treatment records were also used so that the accuracy of the electronic records could be cross checked. In the UK, all medicines administered to dairy cows are legally required to be recorded in accordance with the Veterinary Medicines Regulations 2013, which includes a record of the unique identity of the cow receiving the medicine and the date of administration of the product.

All data extraction and cross checking was carried out in November 2018, which allowed sufficient time to elapse for every cow to progress beyond 150 DIM.

Cows were categorized to allow adjustment for the effects of parity (lactation group, **LCTGP**), season of calving (**CALVSEAS**), and monensin treatment (**KEXX** = yes/no). LCTGP indicates whether the cow is in her first lactation (LCTGP1), second lactation (LCTGP2), or third or more (LCTGP3). CALVSEAS distinguishes cows calving in winter (December, January and February) from those calving in spring (March, April and May).

In addition to these data, the number of days spent in close up (DINCU) was calculated for each cow by subtracting the date of calving from the date of movement to the close up group along with gestation length (GL) by subtracting the date of calving from the date of last insemination.

Statistical analysis

We used multivariable logistic regression to model the odds of disease—any combination of ketosis, mastitis, metritis, milk fever, and displaced abomasum—within 30 DIM. The disease model was based on pre-calving rumination times and a linear combination of DINCU and GL. Strong correlation between DINCU and gestation length (GL; Figure 1) caused multicollinearity issues when both variables were included in the regression model. We therefore calculated the first principal component (PC1) of the covariance matrix of DINCU and GL and used the scores on this component as a proxy variable to simultaneously adjust for DINCU and GL. Principal components are orthogonal linear combinations of a set of correlated variables. The first principal component is the linear combination maximizing the variation in the observed data. The second principal component, orthogonal to the first component (and therefore uncorrelated with it), is the linear combination maximizing the remainder of the variation not accounted for by the first component. In the case of strong positive correlation between two variables, such as DINCU and GL, the first principal component of their covariance matrix will account for most of the variation observed in these two variables simultaneously. To model time to pregnancy, we fitted a Cox proportional hazards model on the PC1 scores, pre- and post-calving rumination time, and a variable indicating whether the cow had any disease within 30 DIM. LCTGP, CALVSEAS and KEXX were included as covariates in both models.

We considered three ways to include rumination time in these regression models: daily rumination times, three-day averages, and weekly averages. Weekly averages, for the three weeks prior to calving and two weeks after calving, were chosen as these smoothed out the variability of daily rumination time and avoided overfitting.

Stepwise variable selection based on Akaike's Information Criterion (AIC) was used to select the weekly rumination intervals to include in each model. LCTGP, CALVSEAS and PC1 were forced into the final models, regardless of their effect sizes or statistical significance.

The final regression models were fitted on all cows with complete data for the included predictor variables, specifically the weekly rumination averages. Consequently, there was a small difference in the subsets of cows used for the disease and pregnancy models.

With the DHARMa R package (Hartig 2020) we performed diagnostic checks and goodness-of-fit tests on the final model for disease. We constructed a receiver operating characteristic (ROC) curve for this model and calculated the area under the curve.

All statistical analysis was performed in R (R Core Team 2020).

RESULTS

There was a total of 719 cows in the study. Twenty-six were first lactation animals, of which only one with pre-calving rumination time available; therefore, they were excluded from the analysis. One animal with much shorter gestation length than the rest (220 days), six animals with more than 50 DINCU, and another six cows with weekly pre-calving rumination means of fewer than 300 minutes, we considered to be outliers and removed them from the sample. In the case of the last group, such low rumination levels indicate that the cow probably had disease, or some other problem, even before calving.

Of the remaining 680 animals, 258 (38%) were second lactation, and 422 (62%) had three or more (up to nine) lactations.

Month of calving ranged from December to May, with 276 cows (41%) calving in the three months of winter (Dec-Jan), and 404 cows (59%) calving in spring (Mar-May). Kexxtone bolus was applied to 261 (38%) of the cows.

Table 2 summarizes DINCU, GL and the weekly pre- and post-calving rumination means.

The three weekly rumination intervals before calving were strongly correlated, with rank correlations ranging from 0.64 to 0.76. Pre- and post-calving rumination had only weak positive correlations, ranging from 0.26 to 0.37. First and second week post-calving rumination had a moderate positive correlation of 0.45. Correlations between variables are given in Table 3.

DINCU ranged from 0 to 47 days (median: 19) and was weakly correlated with the mean weekly rumination times. Rumination three weeks pre-calving had the strongest correlation with DINCU (r = 0.27).

By 30 DIM, 121 cows (18%) developed at least one disease—any combination of ketosis (40 cows, 5.9% of total), mastitis (17 cows, 2.5%), metritis (75 cows, 11%), milk fever (17 cows, 2.5%), or left displaced abomasum (28 cows, 4.1%). No cows were diagnosed with right displaced abomasum. Figure 2 compares daily rumination time of cows without disease with those that developed any disease by 30 DIM.

305 cows (45%) were pregnant again at 100 DIM, and an additional 139 cows (20%) were pregnant at 150 DIM. Figure 3 shows daily rumination time around calving for the pregnant and not pregnant cows.

The first principal component of the covariance matrix of DINCU and GL was estimated as: PC1 = 0.87DINCU + 0.49GL. Larger values of DINCU and GL therefore give larger PC1 scores. PC1 accounts for 86% of the variation observed in these two variables.

Results for the final disease model is given in Table 4. For the disease model, 549 cows had data on all the covariates. Compared to second lactation animals, those with three or more lactations were more likely to develop disease (P = 0.001). Changes in DINCU and GL (PC1) is associated with changing odds of developing disease (P = 0.001). The mean odds of disease approached a minimum for PC1 scores of about 155–168 (Figure 4). The right-hand side of Figure 4 shows the combinations of DINCU and GL which minimizes disease risk. For example, animals with GL = 280 and 30 DINCU had the lowest disease risk, whereas animals with GL = 270 require 35 DINCU to have the same low risk of disease, assuming all other factors are the same We did not find any significant association between pre-calving rumination time and disease within 30 DIM.

For the final pregnancy model, data on all covariates were available for 563 of the 680 cows (Table 5). The association between PC1 and fertility was not statistically significant (*P* = 0.53).

Higher rumination time in the week prior to calving is strongly linked to pregnancy (*P* = 0.007). For each additional 15 minutes (weekly average) rumination time, risk of pregnancy increased by 4% (95% confidence interval: 1% - 7%). Variable selection with AIC included also rumination time in the first week after calving, but in the final model it was not significantly linked to pregnancy (*P* = 0.15).

A higher number of lactations (*P* = 0.02) and calving in spring rather than winter (*P* = 0.04), were associated with increased time to pregnancy. Disease within 30 DIM increased time to pregnancy, but this effect was not statistically significant (*P* = 0.14).

DISCUSSION

In this study we identified an association between rumination times around calving and the risk of pregnancy during the subsequent lactation. This finding is unsurprising as rumination times around calving have been shown to be reliable indicators of the presence of both ketosis (Kaufmann et al., 2016) and hypocalcaemia (Goff et al., 2020), both of which have negative consequences for reproductive performance (Walsh et al., 2007, Santos et al., 2010). However, what is surprising is that we were unable to show an association between PC1 or disease on risk of pregnancy. An explanation for this may be found by considering the reproductive management program used by the herd, Presync-11 (Galvão et al., 2007). This ensured that every cow received a first insemination by 78 DIM and most likely produced a bias in our findings in favour of cows with metabolic abnormalities. These cows are likely to have poorer rumination times around calving (Kaufmann et al., 2016, Goff et al., 2020) and are less likely to resume normal cyclicity (Santos et al., 2010) and be inseminated to a naturally expressed estrus. A further explanation for this finding may also lie in the approach taken by this herd to finding and treating disease in early lactation, aggressive diagnosis and rigorous application of written protocols may be mitigating against the negative effects of disease on reproduction. It also remains possible that the effect of rumination around calving on the risk of pregnancy is an effect independent of either PC1 or disease.

A number of studies have reported significant associations between rumination times and the occurrence of disease (Soriani et al., 2012, Stangaferro et al., 2016, Paudyal et al., 2018) or metabolic abnormalities (Goff et al., 2020) around calving or during early lactation and several studies reported that changes in rumination times have been associated with health conditions occurred either in early lactation (Calamari et al., 2014) or 1 to 2 d prior to the onset of clinical signs (Paudyal et al., 2018). The studies of Kaufmann et al. (2016) and Liboreiro et al. (2015) did however associate changes in pre partum rumination times with specific health conditions post-partum. In this study the same trend has been shown in Figure 2, but we were unable to firmly associate rumination times pre partum with the occurrence of disease in early lactation. The explanation for this finding may be due to the categorisation 'ill health' by aggregating together a number of conditions as a single binary outcome and it is conceivable that certain individual conditions such as ketosis and hypocalcaemia (Kaufmann et al., 2016, Goff et al., 2020) may have an impact on pre partum rumination times whereas other conditions such as metritis and mastitis may not (Liboreiro et al., 2015). We chose to carry out the analysis using the occurrence of disease as a single outcome as the incidence of disease was low even in a herd of this size and so any analysis conducted using individual disease category as outcome would be likely to be lacking in sufficient power to identify effects and reflects the challenges of conducting studies in well managed herds that on one hand keep excellent records but also strive to have a low incidence of illness.

An arbitrary cut off point of 30 DIM for the recording of ill health was used in the current study and it is possible that for some cows categorised as diseased that both the reported health event and the changes in rumination time preceding it both took place post-partum. A shorter cut off might have identified a relationship between pre partum rumination times and disease outcomes. Both ketosis (McArt et al., 2012, Walsh et al., 2007 and Suthar et al., 2013) and hypocalcaemia (Martinez et al., 2012) are conditions that are detrimental to cow health, welfare and productivity more likely to occur in the immediate post-partum period and have both been described as gateway disorders leading to the development of more serious disorders and poorer reproductive performance. An objective of this study however was to attempt to predict disease outcomes sufficiently in advance of a health event to potentially allow the opportunity for management changes aimed at preventing those events.

A potential limitation of this study that could have resulted in bias is that cows generating health alerts from the automated monitoring system may have received closer attention than other cows and cases of disease may have been missed in cows that did not show alerts. However, the likely financial losses and costs associated with the failure to diagnose and treat a case of disease (Liang et al., 2016) and the use of written protocols for assessing cows make it less likely that any bias could occur. The findings of this study highlight the need to focus future studies on rumination times around calving towards the identification of specific conditions so that the future opportunity to predict and prevent those conditions is not missed.

The effect of DINCU on milk yield and components has been demonstrated in only a limited number of studies (Robinson et al., 2001) but in this study PC1, the linear combination of DINCU and GL was found to influence cow health and was associated with changes in the likelihood of a cow being treated for disease during the first 30 DIM. The combination of

DINCU and GL required to minimise the likelihood of treatment during the first 30 DIM is shown in Figure 4 and it is clear that when establishing protocols governing movement of animals to the close up group, the expected gestation length should also be taken into consideration as variations in gestation length will clearly impact on the available time a cow can spend in a close up group and potentially limit any benefits obtained from such a strategy. Regrouping of cows in the vulnerable weeks prior to calving has long been suspected of having adverse effects on cow health (Nordlund et al., 2006) and we believe this is the first occasion where this effect has been formally demonstrated. The effect of regrouping on cow behaviour including feeding patterns and DMI is well documented (Grant and Albright, 1995, von Keyserlingk et al., 2008), as is the link between feed intake prior to calving and disease (Huzzey et al., 2005, Huzzey et al., 2007), and it seems likely that this is the mechanism behind the effects observed herein. As also demonstrated in this study, the effect of PC1 on cow health would be expected to vary depending on how close to calving a group change occurs as DMI is known to decrease most sharply in the 5-7 d immediately prior to calving and so the effect of group changes would be most severe at that time and less severe if the cow is given more time to acclimate and re-establish her social ranking prior to calving. It might also be expected as shown in this study that there would initially be a positive relationship between PC1 and reducing the likelihood of disease in early lactation but that this relationship would weaken and fall away as an optimum is reached allowing the majority of cows to adequately acclimatise to a new social ranking following a group change. As DINCU is a component of the total dry period length we speculate that the increase in the odds of disease observed in this study as PC1 increased beyond an optimum can be explained by an increase in DINCU contributing to an increase in total dry period length which has been shown to be detrimental to health (Dann et al., 2006, Pinedo et al., 2011) and production (Oligaray et al., 2020). Possible mechanisms for this effect could include simple under or over feeding (Dann et al., 2006) as well as changes in insulin sensitivity induced by prolonged feeding of acidogenic diets which favour a more lipolytic state potentially detrimental to health (Vieira-Neto et al., 2021).

We were unable to demonstrate a significant effect of PC1 on the time to pregnancy, strongly suggesting that any effect of DINCU on a cow's fertility potential is exerted via its effect on early lactation disease. The effect of disease on reproductive performance is well documented (Santos et al., 2010). In this study we were unable to demonstrate a relationship between reproductive performance and clinically diagnosed disease and it remains possible that there could be an effect of PC1 on reproductive performance via an influence on the incidence of sub clinical disease such as ketosis or hypocalcaemia (McArt et al., 2012, Caxieta et al., 2017) which was not measured in this study. However, whilst there may be reproductive consequences of both these conditions, an impact on the overall numbers of cows becoming pregnant may not be affected, as its effect diminishes with time (McArt et al., 2012). The timed AI protocol used in this herd also may have biased the numbers of cows pregnant in favour of cows with sub-clinical conditions.

A significant effect of parity was also demonstrated with cows in third parity and above being more likely to experience disease in the first 30 DIM and also less likely to become pregnant which is not surprising as the incidence of disease is known to rise as lactational age increases (Renata et al., 1999; Berge and Vertenten, 2014) and fertility has been shown to decrease as lactation age increases (Ray et al., 1992, Garcia-Ispierto et al., 2007). Our analysis was carried out comparing 2nd lactation versus older cows as few primiparous cows were available to be included in the final models as nulliparous animals were not fitted with collars until after they had calved for the first time.

Season of calving was shown to be a significant predictor of pregnancy outcomes but not for disease in the first 30 DIM with cows being more likely to become pregnant again after calving in WIN rather than SPR. At first this may seem unsurprising as heat stress has been shown to adversely affect reproductive performance (De Rensis and Scaramuzzi, 2003). However, heat stress also influences immune function particularly in transition cows (do Amaral et al., 2011) and so an effect on disease in early lactation would also be expected which was not observed. The prevailing climate conditions in the UK limit the number of days when cows are likely to be heat stressed and it is possible that in the relatively temperate climate of the UK other factors and social stressors outweigh climatic effects. In particular ketosis can be influenced by season and parity and is associated with common post parturient disorders (Berge and Vertenten, 2014) and may be more prevalent in WIN versus SPR. The findings of this study should be treated with caution as it was carried out on a single herd with higher than average standards of management and so may not truly be representative of all herds. In many herds post calving health monitoring may not be as structured and treatment decisions may be made on a more ad hoc basis and so the potential effects observed in this study may be lost due to poor standards of health monitoring and a failure to consistently apply definitions of disease.

CONCLUSIONS

The combined effect of GL and DINCU was shown to be significantly associated with the risk of disease treatment during early lactation but was not associated with the time to subsequent pregnancy. Parity was also associated with greater odds of disease, with older cows in their third or greater lactation being more likely to develop disease. No association between pre-calving rumination times and the occurrence of early lactation disease was identified. Higher rumination time in the week prior to calving was however linked to a shorter time to pregnancy. The occurrence of disease showed no effect on the time to pregnancy. Management groupings immediately prior to calving should consider both the distribution of gestation length and the desired length of stay in the group to minimise risk of disease treatment in early lactation.

REFERENCES

Beauchemin, K. A. 2018. Invited review: Current perspectives on eating and rumination activity in dairy cows. J. Dairy Sci. 101:4762-4784. https://doi.org/10.3168/jds.2017-13706

Berge, A. C., and G. Vertenten. 2014. A field study to determine the prevalence, dairy herd management systems, and fresh cow clinical conditions associated with ketosis in western European dairy herds. J. Dairy Sci. 97:2145-2154. https://doi.org/10.3168/jds.2013-7163.

Bertics, S. J., R. R. Grummer, C. Cadorniga-Valino, and E. E. Stoddard. 1992. Effect of prepartum dry matter intake on liver triglyceride concentration and early lactation. J. Dairy Sci. 75:1914-1922. https://doi.org/10.3168/jds.S0022-0302(92)77951-X.

Britt, J. H. 1992. Impacts of early postpartum metabolism on follicular development and fertility. Proc. Ann. Conv. Am. Assoc. Bovine Pract. 24:39-43.

Büchel, S., and A. Sundrum. 2014. Short communication: Decrease in rumination time as an indicator of the onset of calving. J. Dairy Sci. 97: 3120–3127. https://doi.org/10.3168/jds.2013-7613.

Cai, T. Q., P. G. Weston, L.A. Lund, B. Brodie, D. J. McKenna, and W. C. Wagner. 1994. Association between neutrophil functions and periparturient disorders in cows. Am. J. Vet. Res. 55: 934–943.

Calamari, L., N. Soriani, G. Panella, F. Petrera, A. Minuti, and E. Trevisi. 2014. Rumination time around calving: An early signal to detect cows at greater risk of disease. J. Dairy Sci. 97: 3635–3647. https://doi.org/10.3168/jds.2013-7709.

Caixeta, L. S., P. A. Ospina, M. B. Capel, and D. V. Nydam. 2017. Association between subclinical hypocalcemia in the first 3 days of lactation and reproductive performance of dairy cows. Theriogenology. 94:1-7. https://doi.org/10.1016/j.theriogenology.2017.01.039.

Carvalho, P. D., A. H. Souza, M. C. Amundson, K. S. Hackbart, M. J. Fuenzalida, M. M. Herlihy, H. Ayres, A. R. Dresch, L. M. Vieira, J. N. Guenther, R. R. Grummer, P. M. Fricke, R. D. Shaver, and M. C. Wiltbank. 2014. Relationships between fertility and postpartum changes in body condition and body weight in lactating dairy cows. J. Dairy Sci. 97:3666–3683. http://dx.doi.org/ 10.3168/jds.2013-7809.

Chebel, R. C., J. E. P. Santos, J. P. Reynolds, R. L. A. Cerri, S. O. Juchem, and M. Overton. 2004. Factors affecting conception rate after artificial insemination and pregnancy loss in lactating dairy cows. Anim. Reprod. Sci. 84:239–255. https://doi.org/10.1016/j.anireprosci.2003.12.012.

Dann, H. M., N. B. Litherland, J. P. Underwood, M. Bionaz, A. D'Angelo, J. W. McFadden, and J. K. Drackley. 2006. Diets during far-off and close-up dry periods affect periparturient metabolism and lactation in multiparous cows. J. Dairy Sci. 89:3563- 3577. https://doi.org/10.3168/jds.S0022-0302(06)72396-7.

De Rensis F., and R. J. Scaramuzzi. 2003. Heat stress and seasonal effects on reproduction in the dairy cow-A review. Theriogenology. 60: 1139-1151. https://doi.org/10.1016/S0093-691X(03)00126-2.

do Amaral, B.C., E. E. Connor, S. Tao, M. J. Hayen, J. W. Bubolz, and G. E. Dahl. 2011. Heat stress abatement during the dry period influences metabolic gene expression and improves immune status in the transition period of dairy cows. J. Dairy Sci. 94:86–96. https://doi.org/10.3168/jds.2009-3004.

Drackley, J. K. 1999. Biology of dairy cows during the transition period: the final frontier? J. Dairy Sci. 82:2259-2273. https://doi.org/10.3168/jds.S0022-0302(99)75474-3.

Florian Hartig (2020). DHARMa: Residual Diagnostics for Hierarchical (Multi-Level / Mixed) Regression Models. R package version 0.2.7. https://CRAN.R-project.org/package=DHARMa.

Galvão, K. N., M. F. Sá Filho, and J. E. P. Santos. 2007. Reducing the interval from presynchronization to initiation of timed artificial insemination improves fertility in dairy cows. J. Dairy Sci. 90:4212–4218. https://doi.org/10.3168/jds.2007-0182.

García-Ispierto, I., F. López-Gatius, P. Santolaria, J. L. Yániz, C. Nogareda, and M. López-Béjar. 2007. Factors affecting the fertility of high producing dairy herds in north-eastern Spain. Theriogenology. 67:632-638, https://doi.org/10.1016/j.theriogenology.2006.09.038.

Giordano, J. O., P. M. Fricke, M. C. Wiltbank, and V. E. Cabrera. 2011. An economic decision-making support system for selection of reproductive management programs on dairy farms. J. Dairy Sci. 94:6216–6232. https://doi.org/10.3168/jds.2011-4376.

Giordano, J. O., A. S. Kalantari, P. M. Fricke, M. C. Wiltbank, and V. E. Cabrera. 2012. A daily Markov chain model to study the economic and reproductive impact of reproductive programs combining timed artificial insemination and estrus detection. J. Dairy Sci. 95:5442–5460. https://doi.org/10.3168/jds.2011-4972.

Goff, J. P., A. Hohman, and L. L. Timms. 2020. Effect of subclinical and clinical hypocalcemia and dietary cation-anion difference on rumination activity in periparturient dairy cows. J. Dairy Sci. 103:2591–2601 https://doi.org/10.3168/jds.2019-17581.

Goff, J. P., R. L. Horst. 1997. Physiological changes at parturition and their relationship to metabolic disorders. J. Dairy Sci. 80: 1260–1268. https://doi:10.3168/jds.S0022-0302(97)76055-7.

Goldhawk, C., N. Chapinal, D. M. Veira, D. M. Weary, and M. A. G. Von Keyserlingk. 2009. Prepartum feeding behavior is an early indicator of subclinical ketosis. J. Dairy Sci. 92: 4971–4977. https://doi.org/10.3168/jds.2009-2242.

Grant, R. J. and J. L. Albright. 1995. Feeding behavior and management factors during the transition period in dairy cattle. J. Anim. Sci. 73:2791-2803. https://doi: 10.2527/1995.7392791x.

Grant, R. J. and J. L. Albright. 2001. Effect of animal grouping on feeding behavior and intake of dairy cattle. J. Dairy Sci. 84(E. Suppl.): E156-E163. https:/doi:10.3168/JDS.S0022-0302(01)70210-X.

Grummer, R. R., D. G. Mashek, and A. Hayirli. 2004. Dry matter intake and energy balance in the transition period. Vet. Clin. Food. Anim. 20:447–470. https://doi.org/10.1016/j.cvfa.2004.06.013.

Hut P. R., A. Mulder, J. van den Broek, J. H. J. L. Hulsen, G. A. Hooijer, E. N. Stassen, F. J. C. M. van Eerdenburg, and M. Nielen. 2019. Sensor based eating time variables of dairy cows in the transition period related to the time to first service. Prev. Vet. Med. 169:104694 https://doi.org/10.1016/j.prevetmed.2019.104694.

Huzzey, J. M., M. A. G. von Keyserlingk, and D. M. Weary. 2005. Changes in feeding, drinking, and standing behavior of dairy cows during the transition period. J. Dairy Sci. 88:2454–2461. https://doi.org/10.3168/jds.S0022-0302(05)72923-4.

Huzzey, J. M., T. J. DeVries, P. Valois, and M. A. G. von Keyserlingk. 2006. Stocking density and feed barrier design affect the feeding and social behavior of dairy cattle. J. Dairy Sci. 89:126–133. https://doi.org/10.3168/jds.S0022-0302(06)72075-6.

Huzzey, J. M., D. M. Veira, D. M. Weary, and M. A. G. von Keyserlingk. 2007. Prepartum behavior and dry matter intake identify dairy cows at risk for metritis. J. Dairy Sci. 90:3220–3233. https://doi.org/10.3168/jds.2006-807.

Huzzey, J. M., D. M. Weary, B. Y. F. Tiau, and M. A. G. von Keyserlingk. 2014. Short communication: Automatic detection of social competition using an electronic feeding system. J. Dairy Sci. 97 :2953–2958. https://dx.doi.org/ 10.3168/jds.2013-7434.

Ingvartsen, K. L. and J. B. Andersen. 2000. Integration of metabolism and intake regulation: A review focusing on periparturient animals. J. Dairy. Sci. 83:1573-1597. https://doi: 10.3168/jds.S0022-0302(00)75029-6.

Kaufman, E. I., S. J. LeBlanc, B. W. McBride, T. F. Duffield, and T. J. DeVries. 2016. Association of rumination time with subclinical ketosis in transition dairy cows. J. Dairy Sci. 99:5604–5618. https://dx.doi.org/10.3168/jds.2015-10509.

Kaufman, E. I., V. H. Asselstine, S. J. LeBlanc, T. F. Duffield, and T. J. DeVries. 2018. Association of rumination time and health status with milk yield and composition in early-lactation dairy cows. J. Dairy Sci. 101:462–471. https://doi.org/10.3168/jds.2017-12909.

Leroy, J. L. M. R., G. Opsomer, A. Van Soom, I. G. F. Goovaerts, and P. E. J. Bols. 2008a. Reduced fertility in high-yielding dairy cows: Are the oocyte and embryo in danger? Part I. The importance of negative energy balance and altered corpus luteum function to the reduction of oocyte and embryo quality in high-yielding dairy cows. Reprod. Domest. Anim. 43:612–622. https://doi.org/10.1111/j.1439-0531.2007.00960.x.

Leroy, J. L. M. R., G. Opsomer, A. Van Soom, I. G. F. Goovaerts, and P. E. J. Bols. 2008b. Reduced fertility in high-yielding dairy cows: Are the oocyte and embryo in danger? Part II. Mechanisms linking nutrition and reduced oocyte and embryo quality in high-yielding dairy cows. Reprod. Domest. Anim. 43:623–632. https://doi.org/10.1111/j.1439-0531.2007.00961.x.

Liang, D., L. M. Arnold, C. J. Stowe, R. J. Harmon, and J. M. Bewley. 2016. Estimating US dairy clinical disease costs with a stochastic simulation model. J. Dairy Sci. 100:1472-1486https://doi.org/10.3168/jds.2016-11565.

Liboreiro, D. N., K. S. Machado, P. R. B. Silva, M. M. Maturana, T. K. Nishimura, P. Brandão, M. I. Endres, and R. C. Chebel. 2015. Characterization of peripartum rumination and activity of cows diagnosed with metabolic and uterine diseases. J. Dairy Sci. 98:6812–6827. https://dx.doi.org/10.3168/jds.2014-8947.

Louca, A., and J. E. Legates. 1968. Production losses in dairy cattle due to days open. J. Dairy Sci. 1:573–583. https://doi.org/10.3168/jds.S0022-0302(68)87031-6.

Martinez, N., C. A. Risco, F. S. Lima, R. S. Bisinotto, L. F. Greco, E. S. Ribeiro, F. Maunsell, K. Galvão, and J. E. P. Santos. 2012. Evaluation of peripartal calcium status, energetic profile and neutrophil function in dairy cows at low or high risk of developing uterine disease. J. Dairy Sci. 95:7158–7172. http://dx.doi.org/ 10.3168/jds.2012-5812.

McArt, J. A. A. and R. C. Neves. 2020. Association of transient, persistent, or delayed subclinical hypocalcemia with early lactation disease, removal, and milk yield in Holstein cows. J. Dairy Sci. 103:690–701. https://doi.org/10.3168/jds.2019-17191.

McArt, J. A. A., D. V. Nydam, and G. R. Oetzel. 2012. Epidemiology of subclinical ketosis in early lactation dairy cattle. J. Dairy Sci. 95:5056–5066. https://dx.doi.org/10.3168/jds.2012-5443.

Neave, H. W., J. Lomb, D. M. Weary, S. J. LeBlanc, J. M. Huzzey, and M. A. G. von Keyserlingk. 2018. Behavioral changes before metritis diagnosis in dairy cows. J. Dairy Sci. 101:1–12. https://doi.org/10.3168/jds.2017-13078.

Nordlund, K. V., N. B. Cook, and G. R. Oetzel. 2006. Commingling dairy cows: pen moves, stocking density and health. Proc. Ann. Conf. Am. Assoc. Bovine Prac. 39:36-42.

NRC. 2001. Nutrient Requirements of Dairy Cattle. 7th rev. ed. Natl. Acad. Press, Washington, DC. https://doi.org/10.17226/9825.

Olagaray, K. E., M. W. Overton, and B. J. Bradford. 2020. Do biological and management reasons for a short or long dry period induce the same effects on dairy cattle productivity? J. Dairy Sci. 103:11857–11875. https://doi.org/10.3168/jds.2020-18462.

Oltenacu, P. A., T. R. Rounsaville, R. A. Milligan, and R. H. Foote. 1981. Systems analysis for designing reproductive management programs to increase production and profit in dairy herds. J. Dairy Sci. 64:2096–2104. https://doi.org/10.3168/jds.S0022-0302(81)82813-5.

Paudyal, S., F. P. Maunsell, J. T. Richeson, C. A. Risco, D. A. Donovan, and P. J. Pinedo. 2018. Rumination time and monitoring of health disorders during early lactation. Animal. 12:1484-1492. https://doi.org/10.1017/S1751731117002932.

Pinedo, P., C. Risco, and P. Melendez. 2011. A retrospective study on the association between different lengths of the dry period and subclinical mastitis, milk yield, reproductive performance, and culling in Chilean dairy cows. J. Dairy Sci. 94:106–115. https://doi.org/10.3168/jds.2010-3141.

Pinedo. P., J. E. P. Santos, R. C. Chebel, K. N. Galvão, G. M. Schuenemann, R. C. Bicalho, R. O. Gilbert, S. Rodriguez Zas, C. M. Seabury, G. Rosa, and W. W. Thatcher. 2020. Early-lactation diseases and fertility in 2 seasons of calving across US dairy herds. J. Dairy Sci. 103:10560–10576. https://doi.org/10.3168/jds.2019-17951.

Proudfoot, K. L., D. M. Veira, D. M. Weary, and M. A. G. von Keyserlingk. 2009. Competition at the feed bunk changes the feeding, standing, and social behavior of transition dairy cows. J. Dairy Sci. 92:3116–3123. https://doi.org/10.3168/jds.2008-1718.

R Core Team (2020). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Ray, D. E., T. J. Halbach, and D. V. Armstrong. 1992. Season and lactation number effects on milk production and reproduction of dairy cattle in Arizona. J. Dairy Sci. 75:2976-2983. https://doi.org/10.3168/jds.S0022-0302(92)78061-8.

Renata, T.E., S. van Dorp, W. Martin, M. M. Shoukri, J. P. T. M. Noordhuizen, and J. C. M. Dekkers. 1999. An epidemiologic study of disease in 32 registered Holstein dairy herds in British Columbia. Can. J. Vet. Res. 63,185–192.

Ribeiro, E. S., G. Gomes, L. F. Greco, R. L. A. Cerri, A. Vieira-Neto, P. L. J. Monteiro Jr., F. S. Lima, R. S. Bisinotto, W. W. Thatcher, and J. E. P. Santos. 2016. Carryover effect of postpartum inflammatory diseases on developmental biology and fertility in lactating dairy cows. J. Dairy Sci. 99:2201–2220. http://dx.doi.org/10.3168/jds.2015-10337.

Robinson, P. H., J. M. Moorby, M. Arana, R. Hinders, T. Graham, L. Castelanelli, and N. Barney. 2001. Influence of close up dry period protein supplementation on productive and reproductive performance of Holstein cows in their subsequent lactation. J. Dairy Sci. 84:2273-2283. https://doi: 10.3168/jds.S0022-0302(01)74674-7.

Santos, J. E. P., R. S. Bisinotto, E. S. Ribeiro, F. S. Lima, L. F. Greco, C. R. Staples, and W. W. Thatcher. 2010. Applying nutrition and physiology to improve reproduction in dairy cattle. Soc. Reprod. Fertil. Suppl. 67:387–403. https://doi: 10.7313/upo9781907284991.030.

Santos, J., R. Bisinotto, E. Ribeiro, N. Martinez, and F. Lima. 2013. Role of animal health on reproduction of dairy cows. Pages 32–48 in Proc. 2013 Dairy Cattle Reproduction Council Conference, Indianapolis, IN. Dairy Cattle Reproduction Council, Dublin, OH.

Schirmann, K., N. Chapinal, D. M. Weary, L. Vickers, and M. A. G. von Keyserlingk. 2013. Short communication: Rumination and feeding behavior before and after calving in dairy cows. J. Dairy Sci. 96:7088–7092. https://doi.org/10.3168/jds.2013-7023.

Schirmann, K., M. A. G. von Keyserlingk, D. M. Weary, D. M. Veira, and W. Heuwieser. 2009. Technical note: Validation of a system for monitoring rumination in dairy cows. J. Dairy Sci. 92:6052–6055. https://dx.doi.org/10.3168/jds.2009-2361.

Schirmann, K., N. Chapinal, D. M. Weary, W. Heuwieser, and M. A. G. Von Keyserlingk. 2012. Rumination and its relationship to feeding and lying behavior in Holstein dairy cows. J. Dairy Sci. 95: 3212–3217. https://doi.org/10.3168/jds.2011-4741.

Soriani, N., E. Trevisi, and L. Calamari. 2012. Relationships between rumination time, metabolic conditions, and health status in dairy cows during the transition period. J. Anim. Sci. 90:4544–4554. https://doi:10.2527/jas.2012-5064.

Stangaferro, M. L., R. Wijma, L. S. Caixeta, M. A. Al-Abri, and J. O. Giordano. 2016. Use of rumination and activity monitoring for the identification of dairy cows with health disorders: Part I. Metabolic and digestive disorders. J. Dairy Sci. 99:7395–7410. https://doi .org/10 .3168/jds.2016 -10907.

Suthar, V. S., J. Canelas-Raposo, A. Deniz, and W. Heuwieser. 2013. Prevalence of subclinical ketosis and relationships with postpartum diseases in European dairy cows. J. Dairy Sci. 96:2925–2938. http://dx.doi.org/10.3168/jds.2012-6035.

Venjakob, P. L., R. Staufenbiel, W. Heuwieser, and S. Borchardt. 2019. Serum calcium dynamics within the first 3 days in milk and the associated risk of acute puerperal metritis. J. Dairy Sci. 102:11428–11438. https://doi.org/10.3168/jds.2019-16721.

Vieira-Neto, A., K. N. Galvão, W. W. Thatcher, and J. E. P. Santos. 2017. Association among gestation length and health, production, and reproduction in Holstein cows and implications for their offspring. J. Dairy Sci. 100:3166–3181. https://doi.org/10.3168/jds.2016-11867.

Vieira-Neto, A., R. Zimpel, F. R. Lopes Jr., T. L. Scheffler, E. Block, W. W. Thatcher, and J. E. P. Santos. 2021. Duration and degree of diet-induced metabolic acidosis prepartum alter tissue responses to insulin in dairy cows. J Dairy Sci. 104:1660-1679. https://doi.org/10.3168/jds.2020-18787.

von Keyserlingk, M. A. G., D. Olenick, and D.M. Weary. 2008. Acute behavioral effects of regrouping dairy cows. J. Dairy Sci., 91:1011-1016. https://doi.org/10.3168/jds.2007-0532.

Walsh, R. B., J. S. Walton, D. F. Kelton, S. J. LeBlanc, K. E. Leslie, and T. F. Duffield. 2007. The effect of subclinical ketosis in early lactation on reproductive performance of postpartum dairy cows. J. Dairy Sci. 90:2788–2796. https://doi.org/10.3168/jds.2006-560.

Williams, E. J., D. P. Fischer, D. E. Noakes, G. E. England, A. Rycroft, H. Dobson, and I. M. Sheldon. 2007. Uterine infection perturbs ovarian function in the postpartum dairy cow. Theriogenology 68: 549-559. https://doi.org/10.1016/j.theriogenology.2007.04.056

Zaaijer, D., and J. P. Noordhuizen. 2003. A novel scoring system for monitoring the relationship between nutritional efficiency and fertility in dairy cows. Ir. Vet. J. 56:145–151.

Table 1. Ingredient and nutrient composition of diets fed

Item	Far off dry cow	Close up dry cow	Lactating cow
Ingredient, kg DM			
Rye grass silage	9.6		
Wheat Straw		6.9	0.6
Far off protein blend[1]	2.7		
Close up protein blend[2]		2.7	
Lactating blend[3]			16.3
Far off mineral mix[4]	0.1	0.1	
Close up mineral mix[5]		0.01	
Maize silage		3.8	9.1
Magnesium chloride		0.15	
Megalac[6]			0.3
Water, kg	15.0		
Total, kg DM	12.5	13.8	26.3
Chemical composition %			
DM, %	32.7	35.0	46.9
CP, % of DM	14.3	13.8	16.5
ADF, % of DM,	32.1	35.2	20.4
NDF % of DM	50.7	54.9	33.1
Starch, % of DM	12.0	14.0	30.5
ME, MJ/kg of DM	8.9	8.6	10.8
NEL MJ/kg of DM	1.36	1.32	1.64
Ca, % of DM	0.36	0.34	0.89
Mg, % of DM	0.34	0.44	0.34
P, % of DM	0.32	0.30	0.41
K, % of DM	1.46	1.17	1.02

Na, % of DM	0.12	0.13	0.55
Cl, % of DM	0.31	0.80	0.43
S, % of DM	0.23	0.20	0.25
DCAD mEq/kg of DM[7]	198	10	110

[1] containing 50% SoyPass (KW Feeds, UK), 49% rolled wheat.

[2] containing 19.8% rapeseed meal, 40.4% SoyPass (KW Feeds, UK), and 39.8% soybean meal.

[3] containing 43.73% rolled wheat, 24.7% rapeseed meal, 8.42% ground soybean hulls, 8.1% NIS, (nutritionally improved straw) (Sundown Products, UK), 4.67% SoyPass (KW Feeds, UK), 1.97% soybean meal, 1.74% sodium bicarbonate, 1.33% limestone flour, 1.1% Megafat 88 (Volac Wilmar Feeds, UK), 0.42% salt, 0.37% calcined magnesite, 0.74% mineral/vitamin premix (28.0% Ca, 2% P, 0.1% Mg, 8.0% Na, 8000 mg of Zn/kg, 2500 mg of Cu/kg, 1500 mg of Mn/kg, 45 mg of Se/kg, 50 mg of Co/kg, 130 mg of I/kg, 750 kIU/kg vitamin A, 350 kIU/kg vitamin D and 10000 IU/kg vitamin E.

[4] containing 2% Ca, 5% P, 20% Mg, 9000 mg/kg of Zn, 1500 mg/kg of Cu, 2000 mg/kg of Mn, 50 mg/kg of Se, 80 mg/kg of Co, 500 mg/kg of I, 750 kIU/kg vitamin A, 350 kIU/kg of vitamin D, 20000 IU/kg vitamin E.

[5] containing 99% far off mineral mix and 1% Meta Smart dry (Pestell Nutrition, New Hamburg, ON).

[6] Volac Wilmar Feed, UK.

[7] Dietary cation-anion difference, calculated as follows: DCAD = [(mEq of K) + (mEq Na)] − [(mEq of Cl) + (mEq of S)].

Table 2. Summary statistics of DINCU, GL and weekly pre- and post-calving rumination means (in minutes) for *n* = 680 cows analysed

	DINCU[1]	GL[2]	AVWK3[3]	AVWK2[4]	AVWK1[5]	FRWK1[6]	FRWK2[7]
Cows with data (% of total)	680 (100%)	680 (100%)	418 (61%)	549 (81%)	563 (83%)	680 (100%)	667 (98%)
Minimum	0	260	342	318	305	20	11
1st Quartile	14	277	503	504	481	411	522
Median	19	279	541	538	523	491	564
Mean	18.9	279	536	536	517	462	544
3rd Quartile	23	282	574	578	563	535	598
Maximum	47	290	663	675	669	654	694

[1] Days in close up
[2] Gestation length (days)
[3, 4, 5] Average rumination time (minutes/day) in the third, second and one weeks prior to calving, respectively
[6, 7] Average rumination time (minutes/day) in the first and second weeks after calving, respectively

Table 3. Correlation matrix (Spearman rank correlations) for DINCU, GL and the weekly pre- and post-calving rumination means

	DINCU[1]	GL[2]	AVWK3[3]	AVWK2[4]	AVWK1[5]	FRWK1[6]
GL[2]	0.68	–				
AVWK3[3]	0.27	0.17	–			
AVWK2[4]	0.19	0.16	0.73	–		
AVWK1[5]	-0.03	0.01	0.64	0.76	–	
FRWK1[6]	0.07	0.06	0.33	0.35	0.37	–
FRWK2[7]	0.06	0.04	0.31	0.31	0.26	0.45

[1] Days in close up
[2] Gestation length (days)
[3, 4, 5] Average rumination time (minutes/day) in the third, second and one weeks prior to calving, respectively
[6, 7] Average rumination time (minutes/day) in the first and second weeks after calving, respectively

Table 4. Regression coefficients, odds ratios (OR), 95% confidence intervals for the odds ratios (in square brackets), and *p*-values for the logistic regression model (n = 549) to predict any disease with 30 days from calving. For this model, the area under the ROC curve is 0.70

	Any disease within 30 DIM (Logistic regression)
Lactation group (2nd vs. 3 or more)	3 or more: + 0.998 OR: 2.71 [1.51; 5.13] $P = 0.001$
Calving season (winter vs. spring)	Winter: - 0.211 OR: 0.81 [0.48; 1.35] $P = 0.42$
Gestation length and Days in close up (PC1)	$- 1.074x + 0.003x^2$ OR: N/A $P = 0.001$
Average rumination time (minutes/day) in the second week before calving (AVWK2)	Coefficient: - 0.003 OR: 0.997 [0.993; 1.002] $P = 0.23$

Table 5. Regression coefficients, hazard ratios (HR), 95% confidence intervals for the hazard ratios (in square brackets), and *p*-values for the Cox proportional hazards regression model (n = 563) for pregnancy

	Pregnancy (Cox proportional hazards model)
Lactation group (2nd vs. 3 or more)	3 or more: - 0.26 HR: 0.77 [0.61; 0.97] *P* = 0.02
Calving season (winter vs. spring)	Winter: + 0.22 HR: 1.24 [1.01; 1.52] *P* = 0.04
Gestation length and Days in close up (PC1)	Coefficient: - 0.004 HR: 0.996 [0.98; 1.01] *P* = 0.53
Any disease (within 30 days from calving)	Yes: - 0.24 HR: 0.78 [0.57; 1.08] *P* = 0.14
Average rumination time (minutes/day) in the week before calving (AVWK1)	Coefficient: 0.003 HR: 1.003 [1.001; 1.005] *P* = 0.007
Average rumination time (minutes/day) in the week after calving (FRWK1)	Coefficient: 0.001 HR: 1.001 [1.000; 1.002] *P* = 0.15

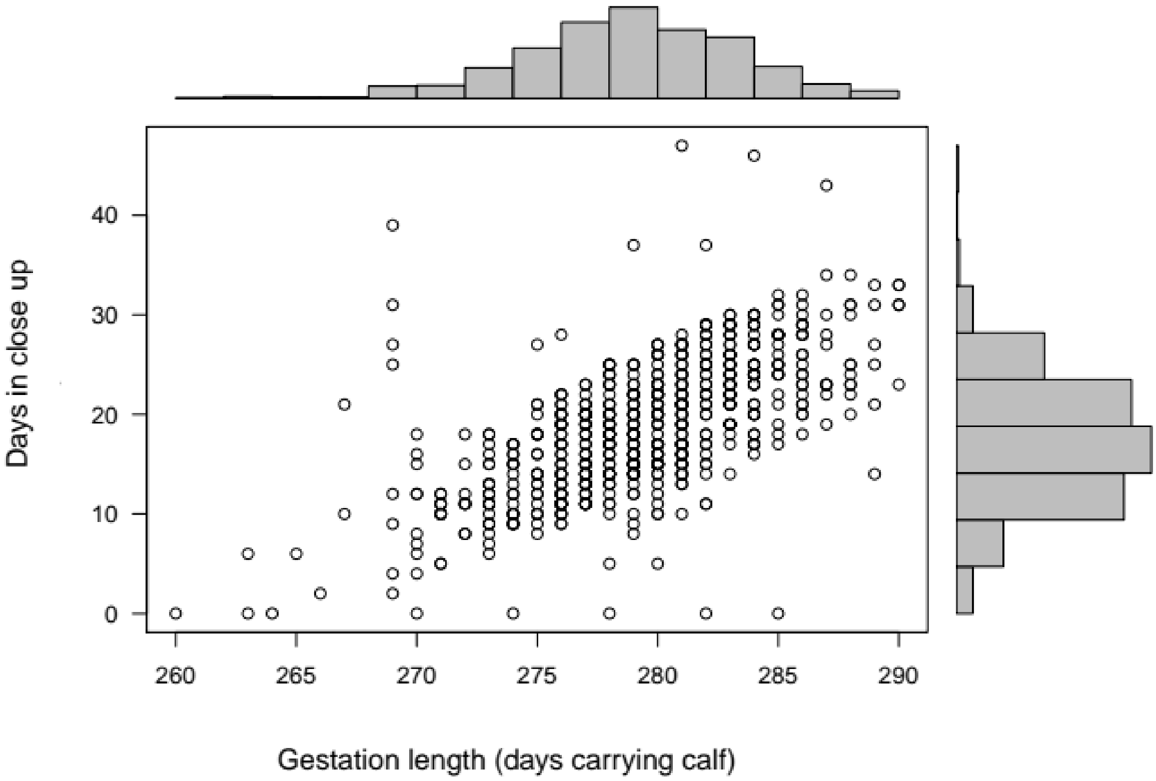

Figure 1: Correlation between gestation length and days in close up (*r* = 0.68) for the 680 dairy cows in our data. The histograms show the marginal distributions of the two variables.

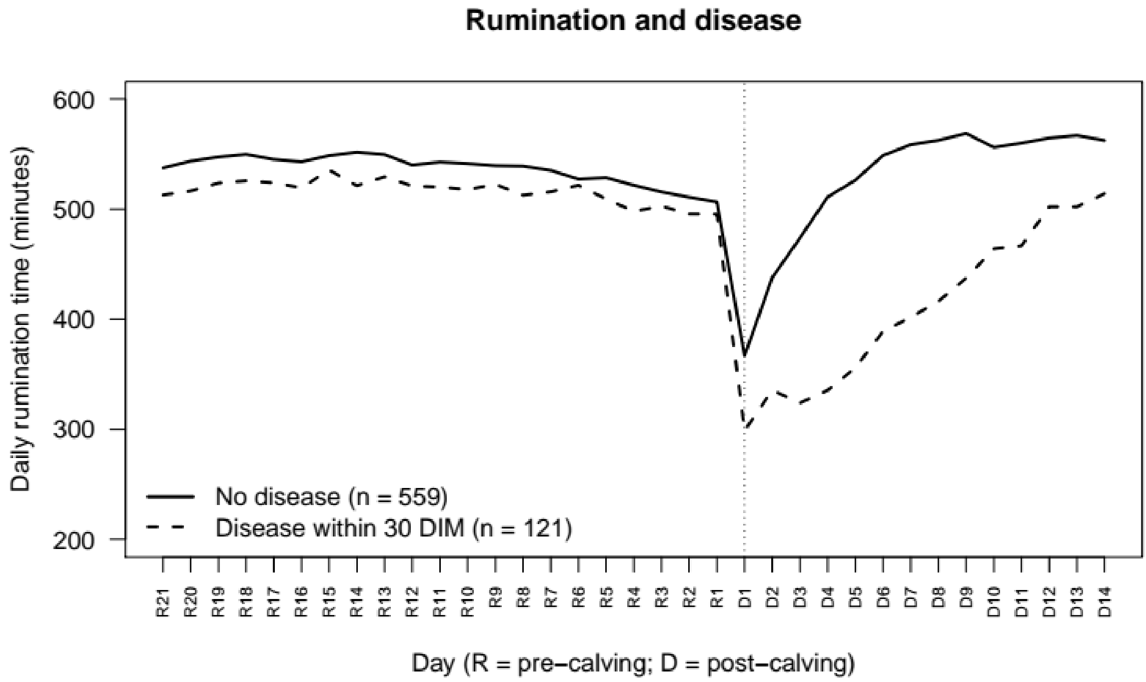

Figure 2: Daily rumination times for cows with no disease, and those that developed disease—ketosis (40 cows), mastitis (17), metritis (75), milk fever (17), or displaced abomasum (28)—within 30 days of calving. The dotted vertical line indicates the day of calving.

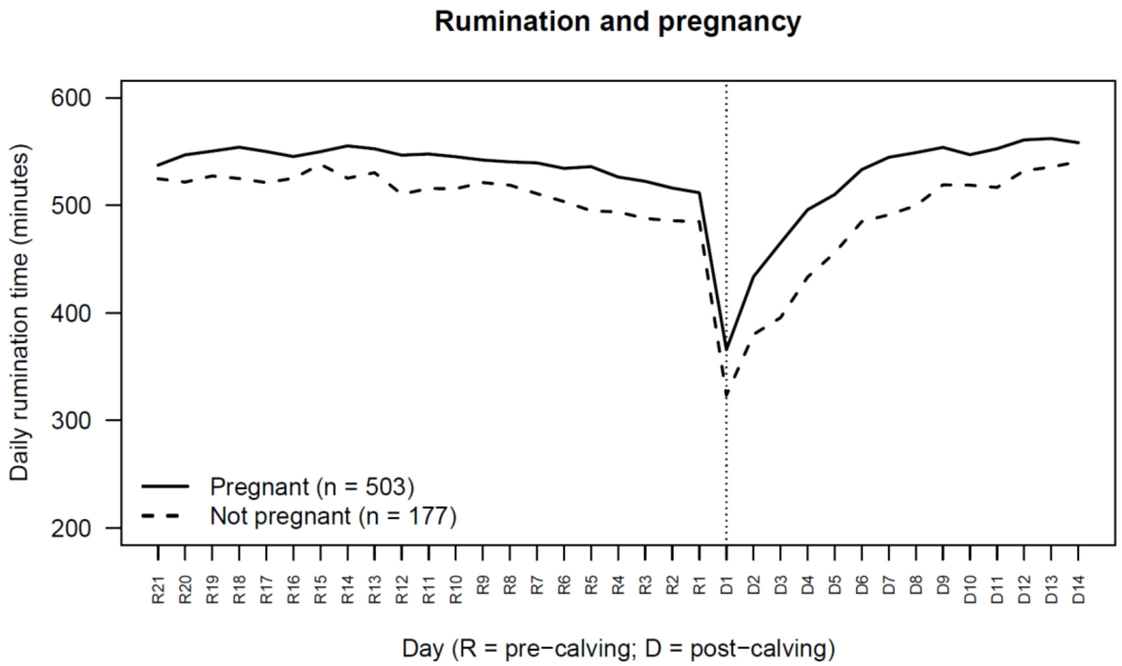

Figure 3: Mean daily rumination time around calving, for cows that became pregnant and cows not pregnant by the end of the follow-up period. The dotted vertical line indicates the day of calving.

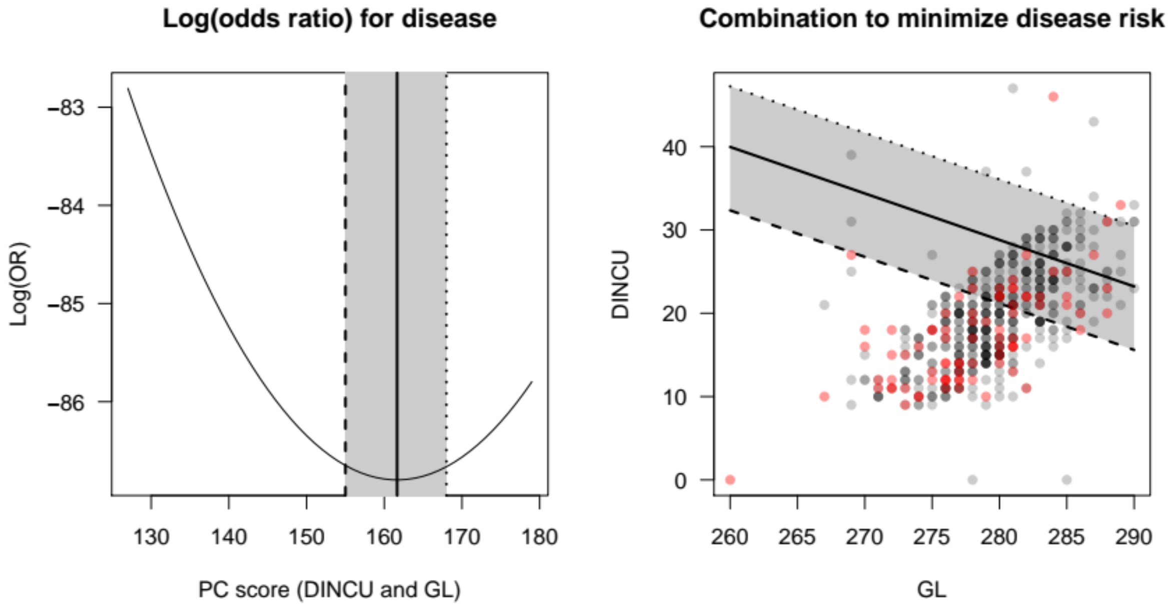

Figure 4: Left: Fitted effect for the principal component scores based on days in close up (DINCU) and gestation length (GL) in the disease model. The shaded areas and solid vertical line indicate the range of values corresponding to the lowest odds for disease within 30 DIM. Right: Combinations of DINCU and GL values which will minimize risk of disease within 30 DIM. The solid, dashed and dotted lines correspond to those in the graph on the left. Points indicate diseased (red) and disease-free (grey) animals by 30 DIM. The shaded grey area shows the combinations of DINCU and GL that were associated with minimum disease risk in our data; for example, animals with GL = 280 and 30 DINCU had the lowest disease risk, whereas animals with GL = 270 needed about 35 DINCU to have the same low risk of disease, assuming all other factors are the same.