

Inferential and Predictive Modelling of Transition Success in Dairy Cows on Automatic Milking Systems

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Abstract

The health and welfare of the transition cow impacts the economic, environmental and social sustainability of the dairy industry. Despite significant improvements in our understanding of the physiological challenges this period poses, limited improvement in the morbidity and mortality rates associated with transition have been reported in the previous two decades. Transition cow monitoring programs (TMPs) are a commonly advocated means of reducing the impact of poor transition health. To date, such programs have been largely diagnostic in nature, focusing on the detection of specific disease states utilising labourintensive monitoring techniques. Automatic milking systems (AMS) offer an opportunity to develop fully automated monitoring systems which may be applied using a prognostic rather than diagnostic approach. Prognostic TMPs provide dairy producers with predictions relating to long-term performance outcomes which may be used to facilitate preemptive management practices aimed at preventing or mitigating the losses associated with poor transition health. The aim of this thesis was to investigate the relationship between production and behaviour data as collected by AMS in the early post-partum period, and subsequent performance assessed using milk production relative to expected, reproductive performance and the risk of removal from the herd in early lactation. An emphasis was placed on the predictive power of this data and its potential utility within a prognostic transition cow monitoring program.

A convenience sample of 46 herds was recruited on a voluntary basis from the UK and Republic of Ireland. Criteria for inclusion was the use of a Lely Astronaut milking robot under free-flow traffic conditions in conjunction with rumination and physical activity monitoring technology. Production variables analysed relating to milk quantity and quality included milk yield, milk yield acceleration, fat and protein content as well as conductivity. Behaviour parameters available via AMS included the number and nature of cow-robot interactions. In addition to this, auxiliary data sources including daily rumination and activity parameters, recorded using a neck mounted accelerometer and, historical cow-level production data as recorded by farm management software were also available. Within this thesis four outcomes were investigated. Chapters 3 and 4 explore the relationship between AMS data collected over days 1-3 post-partum and Yield Deviation, defined as the disparity between recorded milk production and expected milk production over the first 30 days post-partum. Chapters 5 and 6 examine the relationship between data collected from 1-21 days in milk (DIM) and two reproductive outcomes, Expression of Oestrus or Insemination (EOI); The recording of an oestrus or insemination event between DIM 22 and 65, and Conception to First Insemination (CFI), the conception rate to a first insemination between DIM 22 and 80. Finally, Chapter 7 examines data collected over days 1-3 post-partum and subsequent survival using removal from the herd by 100 DIM.

Mixed-effect multivariable models reported in Chapters 3, 5, and 7 serve to quantify the statistically significant associations between AMS production and behaviour data, and their respective outcomes while accounting for the random effect of herd and confounding variables. The development, and external validation of machine learning models for the prediction of production and fertility outcomes, described in Chapters 4 and 6 respectively, assess the degree to which AMS data, in combination with auxiliary data sources, may be leveraged into meaningful improvements in animal health through predictive TMPs. Chapter 6 also examines the marginal effects of auxiliary data sources on model performance by assessing the accuracy with which AMS data can predict reproductive performance with and without rumination, activity and historical production data. Chapter 7 provides a direct comparison of mixed-effect inferential modelling and machine learning predictive models for the odds of removal from the herd by DIM 100 using AMS production and behaviour data in isolation.

In Chapters 3 and 7 we demonstrate that AMS production and behaviour data collected overs days 1-3 post-partum has significant association with Yield Deviation at 30 DIM, and the risk of removal from the herd by 100 DIM. Likewise, in Chapter 5, data collected prior to day 22 post-partum demonstrated significant association with reproductive outcomes up to 80 DIM. Across all outcomes, variables relating to milk yield, rate of milk yield acceleration and fat-to-protein ratio were found to be statistically significant. These associations highlight the transition period, and in particular days 1-3 post-partum as a critical inflection point within the lactation cycle. Furthermore, it demonstrates the potential for AMS sensor data collected during this time to be incorporated into a prognostic TMP. However, the coefficient of determination attributed to the fixed effects within the final models for both reproductive and survival outcomes were found to be low, indicating that the explanatory power of these variables is limited.

Assessed in Chapters 4, 6, and 7, transition cow data demonstrated moderate group level-predictive power for Yield Deviation at 30 DIM, and reproductive outcomes EOI and CFI, but failed to demonstrate predictive power for the risk of removal by 100 DIM. The predictive power of AMS and auxiliary data sources examined in Chapters 4 and 6 represents a critically important finding in support of the premise of prognostic TMPs. While predictive performance is moderate, these findings highlight the potential utility of this data to identify animals likely to experience poor production or fertility performance in the early stages of lactation and should encourage further investigation of how this data may be applied within TMPs. However, the absence of predictive power for the risk of removal in early lactation, reported in Chapter 7, highlights a potential limitation of this approach to transition cow monitoring, particularly where the lag between observations and outcomes is prolonged. These results also serve to demonstrate the risks in the use of inferential models to imply predictive power and the need for externally validated predictive models to be incorporated into the assessment of the potential utility of novel data sources. The failure

to demonstrate a statistically significant increase in model performance following the incorporation of auxiliary data sources, as reported in Chapter 6, highlights the challenges of balancing model accuracy with generalisability and ease of deployment in an environment of rapidly increasing data complexity.

The work presented within this thesis examines a novel means of transition cow monitoring, one which seeks to assess transition health using subsequent production, fertility and survival outcomes. The inferential models reported demonstrate significant statistical association between AMS data and each outcome of interest. However, the predictive power of this data remains limited when applied at the level of the individual, particularly as it relates to the risk of removal within the first 100 days post-partum. Despite this, group level classification of milk production and fertility outcomes demonstrated potential for incorporation into prognostic TMPs. This represents a critical advancement in the field of transition cow monitoring and may offer an effective means to improve the health of transition cows and hence, the sustainability of the dairy industry.

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Contents

| Chapter 1 Introduction | 18 |
|---|----------|
| 1.1 Background | 18 |
| 1.2 Transition Monitoring Programs | 22 |
| 1.3 Monthly Test-Day Milk Recording | 22 |
| 1.4 Biomarkers for Health Status | 24 |
| 1.4.1 Hypocalcaemia | |
| 1.5 Precision Dairy Farming | 26 |
| 1.5.1 Automatic Milking Systems | 27 |
| 1.6 Production and Behaviour Data | 28 |
| 1.6.1 Production Data | 28 |
| Milk Yield | |
| Milk ConstituentsMilk Temperature | |
| 1.6.2 Behaviour Data | |
| Rumination | |
| Activity | |
| Visit Behaviour | |
| 1.7 The Future of Transition Monitoring Programs | |
| 1.7.1 Defining the Cost and Consequences of Poor Transition Health 1.7.2 Removals in Early lactation 1.7.3 Milk Production 1.7.4 Reproduction | 35 37 |
| 1.8 Conclusions and Objectives | |
| Chapter 2 General Introduction to Material and Methods | |
| 2.1 Introduction to the Study Data | 42 |
| 2.1.1 Farm Recruitment | 45 |
| 2.2 Data Quality | 46 |
| 2.2.1 Data Access Limitations | 46 |
| 2.3 Production and Behaviour Data | 51 |
| 2.3.1 Cow-Robot Data | |
| 2.4 Introduction to Methods | 58 |

| 2.4.1 Inferential Analysis 2.4.2 Predictive Modelling 2.4.3 Decision Trees and Random Forests | 59 |
|--|----|
| 2.5 Model Generalisability and Deployment | 61 |
| 2.5.1 Feature Selection | 62 |
| 2.6 Measuring Predictive Performance | 63 |
| 2.6.1 Classification Models | 63 |
| 2.7 Discussion | 64 |
| Chapter 3 The Association of Production and Behaviour Data with Yi Deviation in Transition Cows on Automatic Milking Systems | |
| 3.1 Introduction | 66 |
| 3.2 Materials and Methods | 67 |
| 3.2.1 Study Population | |
| 3.2.2 Step 1: Modelling of Expected Yield | 68 |
| 3.2.3 Step 2. Assessment of 30-Day Yield Deviation | |
| partum Variables | |
| 3.3 Results | |
| 3.4 Discussion | 77 |
| 3.5 Conclusions | 81 |
| Chapter 4 Predictive Modelling of Deviation from Expected Milk Yield Transition Cows on Automatic Milking Systems | |
| 4.1 Introduction | 82 |
| 4.2 Materials and Methods | 83 |
| 4.2.1 Study Population | 83 |
| 4.2.2 Data Analysis | 84 |
| Step 1: Modelling Expected 30-Day Cumulative Yield | |
| Step 2: Assessment of 30-Day Yield Deviation | |
| Data Preparation and Feature Engineering | |
| Model construction | 87 |
| Final Model Training | |
| Final Model Assessment | |
| 4.3 Results | |
| 4.3.1 Modelling Expected 30-Day Cumulative Yield | |
| Feature Selection and Model Tuning | |
| Final Model Assessment | |

| 4.4 Discussion | 95 |
|--|------------|
| 4.5 Conclusions | 96 |
| Chapter 5 The Association of Production and Behaviour Data with Fertility Performance | |
| 5.1 Introduction | 97 |
| 5.2 Materials and Methods | 98 |
| 5.2.1 Study Population | 99 |
| 5.3 Results | 101 |
| 5.3.1 EOI Model | |
| 5.4 Discussion | 109 |
| 5.5 Conclusion | 112 |
| Systems | |
| | |
| 6.2 Materials and Methods | |
| 6.2.1 Data Preparation | 116 117 |
| 6.3 Results | 119 |
| 6.3.1 Expression of Oestrus and Insemination Events | |
| 6.4 Discussion | 130 |
| 6.5 Conclusions | 133 |
| Chapter 7 Factors Associated with the Risk of Removal in Early Lactation for Dairy Cows in Automatic Milking Systems | 135 |
| 7.1 Introduction | 135 |
| 7.2 Materials and Methods | |
| 7.2.1 Study Population | |
| 7.2.2 Data Analysis | 136 |
| 7.2.3 Inferential Modelling7.2.4 Predictive Modelling | |
| 7.3 Results | 140 |

| 7.3.1 Inferential Modelling7.3.2 Predictive Modelling | |
|--|------|
| 7.4 Discussion | .147 |
| 7.5 Conclusions | .150 |
| Chapter 8 Discussion and Conclusion | .151 |
| 8.1 Introduction | .151 |
| 8.2 Inferential Modelling | .153 |
| 8.2.1 The transition period – A key inflection point | .153 |
| monitoring | .155 |
| Assessment of Cow-Robot Interactions | |
| 8.3 Predictive Modelling | .158 |
| 8.3.1 Prognostic TMPs – A viable tool for transition cow management 8.3.2 Optimising model development in an environment of increasing | |
| data complexity | .160 |
| 8.4 Future Studies | .162 |
| 8.5 Conclusions | .164 |
| Chapter 9 References | .165 |
| Chapter 10 Appendices | .191 |
| 10.1 | 101 |

Figures

| Figure 1-1 Example scatter plot analysis of early lactation milk production for the identification of outliers within a herd |
|--|
| Figure 1-2 Graphical demonstration of the relationship between Yield deviation from expected yield and the diagnosis of clinical disease 38 |
| Figure 2-1 Location of participating Lely Centres in the UK and Republic of Ireland |
| Figure 2-2 No. of successful milking events recorded by year across all herds |
| Figure 2-3 Mean Herd-Level 305-day milk yield across all 39 herds. Calculated as an average of cumulative 305-day yields recorded in 2020 and 2021 |
| Figure 2-4 Parity demographics for all available lactations across all herds from 2016-2023 |
| Figure 2-5 Cow-Lactations recording rumination events by year across all herds |
| Figure 2-6 Cow-lactations recording activity events by year across all herds |
| Figure 2-7 Distribution of successful milking visits recorded from days in milk 1-21 across all herds from 2016-2023 |
| Figure 2-8 Distribution of refusals recorded from days in milk 1-21 across all herds from 2016-2023 |
| Figure 2-9 Distribution of daily milk yields recorded from days in milk 1-21 across all herds from 2016-2023 |
| Figure 2-10 Distribution of milk temperature recorded from days in milk 1-21 across all herds from 2016-2023 |
| Figure 2-11 Distribution of milk fat indications recorded from days in milk 1-21 across all herds from 2016-2023 |
| Figure 2-12 Distribution of milk protein indications recorded from days in milk 1-21 across all herds from 2016-2023 |
| Figure 2-13 Distribution of concentrate feed dispensed recorded from days in milk 1-21 across all herds from 2016-2023 |
| Figure 2-14 Distribution of 2-hour activity records for days in milk 1-21 across all herds from 2020-2023 |
| Figure 2-15 Distribution of 2-hour rumination records for days in milk 1-21 across all herds from 2020-2023 |
| Figure 3-1 Scatter Plot of residuals plotted against fitted values for modelling of expected yield for Expected Yield Model in Step 1 72 |
| Figure 3-2 Histogram of Yield Deviation 73 |

| Figure 3-3 Effects plot for the interaction term Mean Milk Yield X Yield Acceleration |
|---|
| Figure 3-4 Effects plot for interaction term Mean Milk Yield X Mean Milkings |
| Figure 3-5 Effects plot for interaction term Mean Milk Yield X Mean Refusals |
| Figure 4-1 Scatter Plot of residuals plotted against fitted values for modelling of expected yield for Expected Yield Model in Step 1 88 |
| Figure 4-2 Distribution of 30-Day Yield Deviation (YD) 89 |
| Figure 4-3 Calibration plot for class membership of the test dataset comprised of 628 lactations from 6 herds |
| Figure 4-4 Calibration plots for the prediction of group membership for all 6 herds within the test dataset |
| Figure 5-1 Frequency distribution for the timing of oestrus or insemination events across the 9,638 lactations in the EOI dataset 103 |
| Figure 5-2 Frequency distribution for the timing of first insemination events across the 8,635 lactations in the CFI dataset |
| Figure 5-3 Frequency distribution for the estimated VWP adopted on each herd as calculated by the mean DIM at first service for the earliest 5% of services |
| Figure 5-4 Effects plot for the interaction term Mean Milk Yield X Mean Concentrate Dispensed |
| Figure 6-1 Workflow for data cleaning, feature engineering and preparation of the final datasets used for model construction and external validation |
| Figure 6-2 Frequency distribution for days in milk at EOI event (oestrus detection or insemination) within the final EOI dataset |
| Figure 6-3 Frequency distribution for days in milk at first insemination within the final CFI dataset |
| Figure 6-4 ROC for EOI-RBT and EOI-RBT+ random forest models for the predictions of expression of oestrus between days 22 and 65 post-partum as evaluated on the test dataset |
| Figure 6-5 Calibration plot for EOI-RBT and EOI-RBT+ random forest models for the predicted probability of expression of oestrus or insemination (EOI) between DIM 22-60 DIM as evaluated on the test dataset |
| Figure 6-6 ROC for CFI-RBT and CFI-RBT+ random forest models for the prediction of conception to first insemination (CFI), between days 22 and 80 post-partum as evaluated on the test dataset |
| Figure 6-7 Calibration plot for the predicted probability of conception to first insemination (CFI), between days 22 and 80 post-partum as evaluated on the test dataset. |

| Figure 7-1 Data preparation for inferential and predictive modelling . 13 | 38 |
|--|----|
| Figure 7-2 Days in Milk at removal for all 9,139 cow-lactations from 21 herds retained in the final dataset | 12 |
| Figure 7-3 Percentage of the milking herd removed in the first 100 day of lactation (RD100) | |
| Figure 7-4 The probability of removal by 100-day post-partum (RD100) plotted against AMS production and behaviour variables retained in the final mixed-effects multivariable logistic | é |
| Figure 7-5 Calibration plot for the predicted probability of the risk of removal from the herd by 100 days in milk as evaluated on the test dataset | 46 |

Tables

| Table 1-1 Commonly Diagnosed Transition Cow Diseases |
|--|
| Table 2-1 Number of recruited by each participating Lely centre 44 |
| Table 2-2 API requests used for data extraction |
| Table 2-3 Descriptive statistics for the 39 herds with accessible data. 48 |
| Table 3-1 Linear mixed model for Expected Cumulative 30-day Yield for 12,295 cow-lactations from 30 herds between 2017 and 2023 as described in Step 1 |
| Table 3-2 Descriptive statistics for herds included in the final multivariable model in Step 3 |
| Table 3-3 Descriptive statistics for independent variables examined in Step 3 |
| Table 3-4 Linear mixed model for 30-day Yield Deviation for 7,417 cowlactations from 30 herds between 2017 and 2023 as described in Step 3 |
| Table 4-1 Descriptive statistics for recruited herds |
| Table 4-2 Linear mixed model for Expected Cumulative 30-day Yield for 8659 cow-lactations from 31 herds between January 2017 and January 2022 |
| Table 4-3 Descriptive statistics for independent variables examined in Step 3 |
| Table 4-4 Regression Performance for Elastic Net, Random Forest Regression and ¹ Multivariable Adaptive Regression Splines90 |
| Table 4-5 Variable importance following recursive feature elimination of the final random forest model as selected for the minimisation of MAE in the model training process |
| Table 4-6 . Regression performance for the prediction of Yield Deviation within the test dataset as achieved by the final random forest model, selected for minimisation of MAE in the model training process |
| Table 4-7 Classification Performance for the final random forest model as selected for the minimisation of MAE in the model training process, of RED, RED + AMBER, and GREEN groups within the test dataset . 92 |
| Table 4-8 Classification performance of the final random forest model 93 |
| Table 5-1 Descriptive statistics for herds included in the final EOI multivariable model |
| Table 5-2 Descriptive statistics for herds included in the final CFI multivariable model |
| Table 5-3 Descriptive statistics for independent variables in final EOI dataset |

| Table 5-4 Descriptive statistics for independent variables in final CFI dataset |
|---|
| Table 5-5 Fixed effects retained in multivariable mixed logistic regression model assessing the association between early lactation AMS production and behaviour data and the risk of expression of oestrus or insemination (EOI). |
| Table 5-6 Performance metrics for EOI and CFI Mixed-effect Multivariable Logistic Models assessed through internal validation 107 |
| Table 5-7 Fixed effects retained in multivariable mixed logistic regression model assessing the association between early lactation AMS production and behaviour data and the risk of conception to first insemination (CFI). |
| Table 6-1 General descriptive statistics for herds which contributed cow-lactations to EOI and CFI dataset |
| Table 6-2 Descriptive statistics for cow-lactations retained in the final EOI and EOI+ random forest models for the prediction of expression of oestrus between DIM 22 and 65 |
| Table 6-3 Descriptive statistics for cow-lactations retained in the final CFI and CFI+ random forest models for the prediction of conception to first insemination between DIM 22 and 80 |
| Table 6-4 Variables analysed within the RBT datasets 123 |
| Table 6-5 Additional variables analysed within the RBT+ Datasets 123 |
| Table 6-6 Scaled Variable importance for the EOI-RBT Model, a random forest model for the prediction of Expression of Oestrus from DIM 22-65 (EOI) utilising AMS data exclusively |
| Table 6-7 Classification performance of all random forest models built utilising AMS data exclusively (RBT) and AMS data in conjunction with auxiliary data (RBT+) for the prediction of Expression of Oestrus or Insemination from DIM 22-65 (EOI) and Conception to First Insemination from DIM 22-80 (CFI) |
| Table 6-8 Scaled variable importance for the EOI-RBT+ Model, a random forest model for the prediction of Expression of Oestrus from DIM 22-65 (EOI) utilising AMS and Auxiliary data |
| Table 6-9 Classification Accuracy (%) per quartile of all random forest models for the prediction of Expression of Oestrus from DIM 22-65 (EOI) and Conception to First Insemination from DIM 22-80 (CFI) as assessed on the test dataset |
| Table 6-10 Scaled variable importance for the CFI-RBT Model, a random forest model predicting conception to first insemination from DIM 22-80 utilising AMS data exclusively (CFI-RBT) |
| Table 6-11 Scaled variable importance for the CFI-RBT+ Model, a random forest model predicting conception to first insemination from DIM 22-80 utilising AMS and auxiliary data |

| Table 7-1 Descriptive statistics for herds contributing cow-lactations to the final dataset |
|---|
| Table 7-2 Descriptive statistics for cow-lactations retained in the final mixed-effect logistic and XGBoost model |
| Table 7-3 Descriptive statistics for independent variables assessed in the mixed-effect logistic and XGBoost model |
| Table 7-4 Results of the multivariable mixed logistic regression model assessing the association between early lactation AMS production data and the risk of removal from the herd by 100 days in milk |
| Table 7-5 Performance metrics for mixed-effect multivariable logistic model assessing the association between early lactation AMS production data and the risk of removal from the herd by 100 days in milk, with and without the random effect of Herd |
| Table 7-6 Performance Metrics for XGBoost model for the prediction of removal by 100 days post-partum as assessed on the test dataset 146 |
| Table 7-7 Herd-level performance metrics for the XGBoost model for the prediction of removal by 100 days post-partum as assessed on the test dataset |
| Table 7-8 Scaled variable importance for independent variables in the XGBoost model for the prediction of removal from herd with the first 100 days post-partum |
| |

Abbreviations

| AFC | Age at first calving | MAPE | mean absolute percentage error; |
|------|----------------------------|--------|---------------------------------|
| AUC- | Area under the | NEB | Negative energy |
| ROC | receiver operator curve | INLU | balance |
| API | Application | NPV | negative predictive |
| / (1 | programming interface | 141 V | value |
| Al | Artificial insemination | NEFA | Non-esterified fatty |
| Ai | Artificial insertification | INCIA | acid |
| AYR | All year-round calving | OR | Odds-ratio |
| | pattern | | |
| AMS | automatic milking | PPV | positive predictive |
| | system | | value; |
| AU | arbitrary unit; | PDT | Precision dairy |
| | , | | technology |
| BCS | Body condition score | PFT | Potential fixed time |
| | | | insemination |
| BHB | Beta-hydroxy butyrate | PV-DD | Previously lactation |
| | | | day spent dry |
| CI | Confidence interval | PV- DC | Previously lactation |
| | | | conductivity at dry |
| | | | off |
| CFI | Conception to first | PV- DO | Previously lactation |
| | insemination | | DIM at dry off |
| DIM | days in milk | SCH | Sub-clinical |
| | | | hypocalcaemia |
| DL | Decilitre | SCC | Somatic cell count |
| ECY | expected 30-day | SCK | Sub-clinical ketosis |
| | cumulative yield; | | |
| EOI | Expression of oestrus | SE | Seasonal calving |
| | or insemination event | | pattern |
| FMS | Farm management | TMP | Transition cow |
| | support | | monitoring program |
| FPR | Fat-to-protein ratio | TCI | Transition cow index |
| IQR | Inter-quartile range | TRM | Targeted |
| | | | reproductive |
| | | | management |
| Kg | Kilogram | TSM | Transition success |
| | | | measure |
| LDA | Left displace | YD | 30-Day Yield |
| | abomasum | | Deviation |
| MAE | mean absolute error | | |

Chapter 1 Introduction

1.1 Background

In 1988 the World Commission on Environment and Development published a report describing the environmental challenges facing the global community and their long-term strategies for achieving sustainable global development. This was defined as, development that meets the needs of the present without compromising the ability of future generations to meet their own needs (Keeble, 1988). This idea continues to form the basis for agricultural development today as described by the United Nations Sustainable Development Goals (The United Nations, 2016). In line with these goals, the dairy industry is seeking to increase the efficiency of production without compromising the health of the animals which produce our food, the humans who consume it or the environment which sustains us.

The health and welfare of animals within the dairy industry impacts all three dimensions of its' sustainability, these being, economic, environmental, and social (Segerkvist et al., 2020). In this regard there is perhaps no time with greater influence on sustainability than the transition period. Commonly defined as the 3 weeks pre- and postpartum, the transition period is pivotal in the production cycle of the dairy cow. To meet the challenges associated with calving and initiation of lactation, a carefully coordinated response across metabolic, inflammatory, and immune pathways is required (Pascottini et al., 2020). Where an animal fails to adequately regulate this response, a range of interrelated clinical and sub-clinical disease states results (Mulligan et al., 2006a). The most commonly diagnosed transition diseases are briefly described in Table 1-1, including ketosis, hypocalcaemia, retained foetal membranes and metritis. The segregation of the transition cow's physiological status into discrete disease states as presented in Table 1-1 serves to ease diagnosis and disease recording. However, this fails to adequately capture the complexity of transition cow physiology, specifically the interdependency between metabolic pathways and the often-blurred line between physiology and pathophysiology. This topic has been reviewed in depth by Sundrum, (2015), however, a brief example is provided here.

The challenge posed by the onset of lactation often commences with an increase in demand for energy in the days prior to calving. This elevation of energy requirements, in support of both a full-term calf and colostrum synthesis, is accompanied by a reduction in feed intake and results in a near ubiquitous incidence of negative energy balance among dairy cows in the peri-parturient period (LeBlanc, 2010). Concurrently, the metabolic pathways associated with calcium homeostasis are challenged through the abrupt increase in milk synthesis (Caixeta & Omontese, 2021). In response to these challenges, fat stores are mobilised, and Non-Esterified Fatty Acids (NEFA) moved to the liver to generate energy via oxidation. Likewise,

bone resorption of calcium is upregulated while absorption via the gut and kidneys is increased. Where these homeorhetic responses are sufficient to meet demand for their respective metabolites, they can be down regulated appropriately, and transition health maintained. However, where these processes are insufficient to meet demand and continue uncontrolled, they begin to move from a physiological to a pathophysiological response (Ingvartsen et al., 2003). Where calcium demands cannot be met, clinical or sub-clinical milk fever occurs. This in turn leads to decreased food intake and exacerbation of negative energy balance. Furthermore, the efficiency of immune cells, in particular neutrophils, is markedly reduced (Sordillo et. al., 2013). In response to a continued state of negative energy balance, a greater number of fat cells are mobilised (Bradford et al., 2009). Excessive NEFA serum concentration alters the function of immune cells, enhancing pro-inflammatory pathways and initiating a positive feedback loop, further increasing fat mobilisation. The capacity of the liver to process these fat cells is overwhelmed resulting in partial oxidation and the generation of ketone bodies. These processes, combined with the inherent exposure to infectious pathogens via the reproductive tract during calving, results in an immune compromised, and energy deficient animal being required to launch a significant immune response (Sordillo et. al., 2009).

Any animal experiencing the physiological dysregulation described here may be diagnosed with none or all of the transition diseases presented in Table 1-1. This will depend on the resilience of the individual animal and the thoroughness of the transition cow monitoring program implemented on farm. However, to view these as unrelated, binary diagnoses, or the process which led to their development as wholly pathological, is not in keeping with our current understanding of transition cow physiology. A more appropriate framing may be that each transition cow's physiological status lies somewhere on a spectrum between that which leads to complete fulfilment of her genetically determined production potential, and that leading to complete loss of this potential through death or cull. Between these two extremes a broad range of physiological states may be experienced with a wide range of consequences for animal health, welfare and production. Utilising this framing of transition performance may prove to be of greater value to producers as it facilitates a more objective, long-term assessment of transition success.

Transition disease has been shown to exert a negative effect on animal performance including milk production (Carvalho et al., 2019), fertility (Pascottini et al., 2020), and survival (Probo et al., 2018), long beyond the course of the disease itself. This was demonstrated by Carvalho et al., 2019, in their analysis of the long-term outcomes for milk yield, reproduction and cull risk in animals diagnosed with clinical disease within the first 21 days in milk. Individual 305-day milk yield was reduced by 4%, pregnancy rate reduced by 19%, and culling risk by 305 DIM increased by 95% in cows diagnosed with at least one postpartum clinical disease. When these losses were apportioned to those

occurring within the first 21 days (broadly within the time during which clinical disease was detected) only 24% of milk losses, 36% of cull difference and 0% of reproductive difference were realised within this period. This, demonstrates how the success with which an animal navigates the transition period will in large part, determine the success of the entire lactation.

The impact of poor transition health on the dairy industry is difficult to overstate. It has been estimated that approximately 30-50 percent of dairy cow will experience some form of metabolic or infectious disease during this period (LeBlanc, 2010). Furthermore, removal of early lactation animals from the herd are reported as 7% annually (Hanks et. al., 2023). The detrimental effects on economic sustainability through the direct and indirect costs associated with disease (Galligan, 2006). involuntary culling (Orpin & Esslemont, 2010), as well as reproductive failure (Cabrera, 2014) are well documented. However, beyond economics, the morbidity and mortality associated with the transition period represents one of the most serious welfare issues affecting modern dairy farming (von Keyserlingk et. al., 2009). This impacts both the social and environmental sustainability of the industry. As such, the development of strategies aimed at improving the management of transition cows has been an industry priority for over two decades. Described as the "final frontier" in 1999 (Drackley, 1999), our understanding of the physiology of transition has grown exponentially in the intervening years. This has facilitated developments in transition cow nutrition (Cardoso et al., 2020), housing (Cook & Nordlund, 2004), and health management (Mulligan et al., 2006b). The impact of these developments on transition health, however, remains unclear. While a lack of large-scale studies hampers our ability to assess changes in transition cow health and welfare definitively, it is generally accepted that current disease incidence remains unacceptably high (Mulligan et al., 2006a; Daros et al., 2022). It would appear therefore, that we have failed to translate our understanding of transition cow physiology into meaningful improvements in transition cow health. The extent to which this can be addressed going forward will be a key determinant of the future sustainability of the dairy industry.

Table 1-1 Commonly Diagnosed Transition Cow Diseases

Ketosis

An increase in concentration of ketone bodies (Acetone, Acetoacetate, B-Hydroxybutyrate (BHB) in body fluids

Incidence: Subclinical: 40-60%, Clinical: 2-15% (McArt et al., 2012)

Pathophysiology: Non-Esterified Fatty Acids (NEFAs) mobilised in response to a state of negative energy balance undergo partial oxidisation in the liver resulting in the generation of ketone bodies.

Diagnosis: Evaluation of ketone concentration (most commonly Beta Hydroxy Butyrate) in serum, urine or milk

Impact: Animals diagnosed with sub-clinical ketosis demonstrate reduced milk yield, increased incidence of displaced abomasum, increased risk of culling (Duffield et al., 2009), reduced fertility (Walsh et al., 2007). Clinical ketosis can be fatal.

Hypocalcaemia

Reduced concentrations of serum calcium

Incidence: Clinical 5%, Subclinical 40% (Multiparous dairy cows) (Seifi & Kia, 2017)

Pathophysiology: Failure of homeorhetic processes designed to increase available calcium, including bone resorption, renal and gut absorption capacity to maintain calcium level in the face of a large increase in calcium demand concurrent with the initiation of lactation.

Diagnosis: Total calcium concentration assessed on serum

Impact: The occurrence of milk fever has been associated with reduced feed intake (Hansen et al., 2003), immunosuppression (Kimura et al., 2006), dystocia and decreased reproductive performance (Correa et al., 1993, Caixeta et al., 2017) Clinical milk fever can be fatal.

Retained Foetal Membranes

Failure to expel the foetal membranes within 24 hours post-partum

Incidence: 4-16% (Sheldon et al., 2008)

Pathophysiology: Normal explosion occurs within 3-8 hours. Failure of placental detachment has been linked with the premature delivery, twinning, traumatic calving and hypocalcaemia.

Diagnosis: Observed presence of foetal members 24 hours post-partum

Impact: Retained foetal membranes has been associated with increased risk of metritis, displaced abomasum, increased risk of culling (Tucho & Ahmed, 2017)

Metritis

Inflammation and infection of the uterine wall

Incidence: 25-40% (Sheldon et al., 2008)

Pathophysiology: Bacterial invasion of the deep lining of the uterus following calving may be facilitated by poor hygiene at the point of calving, trauma to the uterine wall or retention of foetal membranes.

Diagnosis: Enlarged uterus, fetid watery red-brown vaginal discharge with or without signs of systemic illness within 21 DIM.

Impact: Reduced milk production, reduce reproductive performance, increased risk of culling (Giuliodori et al., 2013)

1.2 Transition Monitoring Programs

A corner stone of modern transition cow management has been the development of transition cow monitoring programs (TMP). Applied at the level of the individual, these aim to provide early detection of poor transition health and thereby, an opportunity to limit the costs and consequences of disease, reduced animal performance, or welfare (LeBlanc, 2010).

At their inception TMP served to detect animals experiencing overt clinical disease and facilitate prompt initiation of treatment. They achieved this utilising manual assessment of variables related to clinical health, such as general demeanour, interest in food, rumen fill, udder tautness, and faecal consistency (Guterbock, 2004). This approach to transition monitoring has proven to be popular and the practice continues to play a large role in transition monitoring today. Espadamala et al., (2016) reported the transition cow monitoring protocols in place across 45 herds in the United States ranging in size from 450 to 9,500 lactating cows. Non-specific, subjective measures were the most commonly deployed techniques within TMPs on these herds. This included the assessment of variables such as demeanour, appetite and vaginal discharge. While qualitative scales for parameters such as demeanour, and vaginal discharge have been developed to reduce the subjectivity of their assessment, these were not found to be in use. Indeed, standard operating procedures of any kind for transition cow monitoring were reported as available in only 4 dairies surveyed. These results are mirrored by a survey conducted across 429 farms in Germany which reported that the majority of fresh cow assessments were subjective in nature with only 33% utilising objective measures such as rectal temperature (König et al., 2023).

Applied on commercial dairy farms, the manual observation of animals offers simplicity and flexibility as a broad transition cow monitoring program. It is, however, highly labour-intensive, a key limitation in an industry where lack of skilled labour is apparent. Furthermore, their subjective, non-specific nature leads to inherent issues relating to the reliability and repeatability of measurements between observers and across farms. An alternative means of assessment commonly available on modern dairy farms is monthly test-day milk recording. While this approach has inherent limitations relating to the timing and frequency of data collection, it offers value to producers as an objective means of monitoring performance in early lactation.

1.3 Monthly Test-Day Milk Recording

The advent of monthly milk testing regimes provided a means to standardise the monitoring of production metrics for transition cows. Their initial application was as an objective but non-specific assessment of transition health. This took the form of an assessment of milk production at a single time point, generally first-test day yield or peak yield (Caixeta & Omontese, 2021). The success or failure of transition management was judged at the herd level by the comparison of observed yields with producers' expectations, examination of variation among herd mates and the inspection of outliers. An example of such analysis is presented in Figure 1-1. The premise of such analysis is

simple, optimal transition management is expected to lead to early lactation milk yield in line with the producers' expectations and low variance in production between herd mates. Where this is not achieved, an investigation of transition management is advocated. The objective nature and low labour costs associated with this form of monitoring has led to it becoming a well-established assessment of transition health (Eicker et al., 2002) with several variations of this analysis being made available through commercial farm software (e.g., Week Four Milk via Dairy Comp®, Summit Milk via Dairy Herd Information).

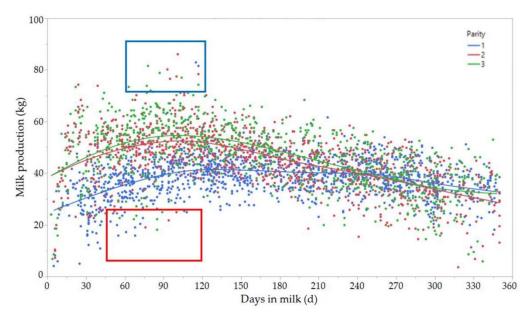


Figure 1-1 Example scatter plot analysis of early lactation milk production for the identification of outliers within a herd. Image description as per source, Caixeta & Omontese, (2021): Milk production (kg; vertical axis) by days in milk (DIM; horizontal axis) for different parity groups. The upper blue square highlights peak milk production between 60 and 120 DIM. The lower red rectangle highlights problem cows (outliers characterized by low milk production, compared with the rest of the herd between 50 and 120 DIM). Parity 1 (lactation = 1; blue dots), Parity 2 (lactation = 2; red dots), Parity 3 (lactation > 3; green dots). Continuous lines represent the average milk production for each parity group by days in milk.

The analysis of milk constituents attempted to refine this approach further by utilising monthly test-day data to provide more objective information relating to the risk of transition diseases, specifically ketosis. Through the assessment of milk fat and protein percentages assessed on test-day milk samples, herd-level risk for this disease was derived with moderate success. A study by Duffield et al., (2002) found that herds with elevated levels of ketosis within the first 2 weeks post calving (defined as >20% prevalence), could be identified with a sensitivity of 69% and specificity of 83% using fat-to-protein ratio. As with the analysis of early lactation milk yield, examining fat-to-protein ratio in this manner is designed to prompt a more detailed investigation of transition health within the herd. Their use is advocated as a means of assessing

compliance with management protocols or the effect of substantial herd level changes (e.g., a change in transition cow ration).

While the utility of monthly test-day data for the assessment of transition health at the herd-level is clear, its application within TMPs for the early detection of individuals expected to suffer poor transition health remains limited. The nature of monthly sampling regimes leads to large variance in days in milk at first sampling with individuals often sampled outside of the high-risk period of days 1-21 post-partum. In addition to this, the data generated is inherently retrospective in nature and offers little opportunity for the proactive management of individual animals (Eicker et al., 2002).

1.4 Biomarkers for Health Status

The analysis of biomarkers as part of TMPs represented a significant development in our ability to objectively monitor individual transition cows. While recognition that both clinical and sub-clinical disease was accompanied by measurable chemical changes in blood, milk and urine is not recent, the large-scale investigation of these changes has been limited to the past 25 years (Overton et al., 2017). The ability to quantify the changes in concentration of key metabolites such as calcium, ketone bodies and NEFAs during transition demonstrated potential for the early and accurate identification of transition disease (Ceciliani et al., 2018). Within a TMP, this offered the potential to move away from the subjective assessment of clinical disease and towards a more objective approach to monitoring.

1.4.1 Hypocalcaemia

The analysis of serum calcium level in the periparturient period for the identification of sub-clinical hypocalcaemia can be achieved through laboratory analysis of blood samples, ideally obtained between 12- and 24-hours post-partum (Goff, 2008). Hypocalcaemia is the quintessential "gateway" transition disease. Animals experiencing the disease in early lactation have increased risk of developing ketosis, abomasal displacement, retained foetal membranes, metritis and mastitis (Seifi & Kia, 2018). Despite the availability of laboratory-based diagnostics for its identification, assessment of serum calcium at the level of the individual within TMPs is effectively null. The limited uptake of this monitoring approach can likely be attributed to the labour-intensive nature of serum sampling and the lack of cow-side tests. Instead, calcium assessment has been largely limited to herd-level analysis. For example, intermittent sampling where a total of 12 animals within 48 hours of calving are selected. An alert level of over 5 animals recording calcium levels below 2mmol/l indicates the need for further investigation (Seifi & Kia, 2018). The limited extent to which individual sampling has been incorporated into TMPs demonstrates the reluctance of dairy producers to employ labour-intensive monitoring techniques, even when the broad range of negative health outcomes stemming from hypocalcaemia are extremely well documented.

1.4.2 Negative energy Balance

Both NEFA and Ketone bodies (acetone, acetoacetate, Beta Hydroxy Butyrate) have been established as indicators of negative energy balance in the transition cow. However, by far the most commonly utilised indicator as part of a TMP is the analysis of serum for Beta Hydroxy Butyrate (BHB). This ketone body is regarded as the gold standard for diagnosis of ketosis (Oetzel, 2015) and a widely accepted measure of the cow's adaption to NEB (LeBlanc, 2014). Its use on farm has been facilitated by the availability of a range of cow-side tests utilising serum, urine or milk as a substrate (Geishauser et al., 2001, Jansen et al., 2021). A range of handheld meters have been validated for the analysis of BHB levels in serum with sensitivity and specificity above 90% observed (Geishauser et al., 2001, Sailer et al., 2018). The accuracy of serum tests for the detection of sub-clinical ketosis generally exceeds that of cow-side tests utilising milk, which report sensitivity of 7 – 91% and specificity of 56-100% across a range of commercially available tests (Geishauser et al., 2001). Despite this, the use of milk remains an attractive option due to the ease with which the substrate can be obtained.

The Incorporation of cow-side metabolic testing into a TMP allows for the prompt diagnosis of sub-clinical NEB, and thus earlier initiation of treatment. Monitoring programs call for the testing of transition cows in both the first- and second-week post-partum, an approach reported to detect between 79% and 95% of all animals which will experience sub-clinical ketosis within the first two months post-partum. This percentage drops to between 69% to 86% when cows were tested in the second week only, and 30% to 56% when cows were tested in the first week only (Geishauser et al., 2001).

Given the ubiquitous nature of negative energy balance in the early post-partum period and the central role this process plays in the development of transition cow disease, assessment of BHBs represents a powerful tool in transition cow monitoring. Despite this however, its uptake on farm remains low. Across 45 herds surveyed in California, none utilised routine ketone monitoring within their TMP and only 7% evaluated ketone levels in fresh cows displaying signs of ill health (Espadamala et al., 2016). A survey in Germany returned similar results with only 3% of the 429 surveyed herds reporting the use of cow side ketones testing, though no distinction was made in this study as to the manner of its use (i.e., proactive or reactive) (König et al., 2023).

As our understanding of the relationship between key physiological variables and transition health has developed, the potential for the application of metabolic health indicators within TMPs has grown. The strengths and limitations of its use are somewhat exemplified by the development and adoption BHB analysis as an objective assessment of energy balance. Through large scale studies, the detrimental effect of subclinical ketosis on transition health has been widely demonstrated (Dohoo & Martin, 1984) and monitoring programs, using a range of cow-side tests have been described (Geishauser et al., 2001).

However, the degree to which this has been incorporated within TMPs remains disappointing.

In Europe, recent trends within the dairy industry have seen a reduction in the number of dairy farms, coupled with an increasing farm size and number of cows managed per unit of labour (Jongeneel. et al., 2023). The limited labour market which predominates within the UK, is likely a barrier in the uptake of labour-intensive sampling protocols such as those advocated for BHB monitoring. However, as labour shortages provide motivation for the adoption of new technology, this challenge also brings opportunity (White et al., 2005). The exponential growth in the adoption of sensor technology on dairy farms in recent years has increased the research attention directed at the automation of transition cow monitoring. The application of precision farming techniques in this regard represents another significant development for the future of TMPs, one which has the potential to allow the objective monitoring of transition cow health with minimal labour input.

1.5 Precision Dairy Farming

Precision dairy farming is the use of technologies to measure physiological, behavioural, and production indicators of individual animals in order to improve management strategies and farm performance (Bewley, 2010). Its utilisation on farm can be broken into four stages (Rutten et al., 2013).

- Measurement: techniques that record a variable (e.g., activity)
- Interpretation: changes in the sensor data (e.g., increase in activity) are used to classify the cow's status (e.g., oestrus)
- Integration: where sensor data is supplemented with other information (e.g., cull cow value) to produce advice (e.g., whether to inseminate a cow or not)
- Action: the farmer makes a decision, or the sensor system makes the decision autonomously (e.g., the AI technician is called).

Key amongst what precision dairy farming aims to deliver for the industry is to help producers make objective, informed management decisions based on automatically collected data. With this comes the potential for improved efficiency of production, health, and welfare of farmed animals and in particular transition cows (Bewley, 2010). The application of precision dairy farming has been facilitated by a large and expanding range of precision dairy technology (PDT). Over the past two decades we have seen an exponential increase in the availability of low cost, multifunctional sensor technology on dairy farms (Rutten et al., 2013). These range from wearable technology including accelerometers, thermometers, and microphones to parlour technology including milk meters and milk conductivity and constituent sensors.

This increase in the commercial availability of PDT has created greater opportunity for the application of precision farming techniques. However, concerns have been raised as to the driving forces behind

PDT development (Hogeveen & Ouweltjes, 2003). The opportunity to quickly develop and market animal sensor technology provides an attractive commercial proposition. Indeed, their sale in recent years has been subsidised in the UK through government grants (Rural Payment Agency, 2020). However, the availability of sensor technology represents only the first step in the realisation of the potential benefits of precision farming. When not accompanied by validated tools to facilitate data interpretation and integration, the adoption of PDT may fail to deliver a positive impact for the industry. This was highlighted by Neethirajanet al., (2017) who noted a lack of commercial impact despite widespread adoption of sensor technology, a phenomenon the authors attribute to a failure of the manufacturers' ability to deliver commercially relevant tools. Though large quantities of data can now be economically and reliably collected on farm, its interpretation, integration, and translation into action – the end goal of precision farming, remains difficult (Stone, 2020).

1.5.1 Automatic Milking Systems

Within the realm of precision dairy technology, automatic milking systems (AMS) are perhaps best positioned to deliver on the promise of precision dairy farming. Since their introduction to the marketplace in 1992 their use has grown rapidly with operating units estimated at 50,000 worldwide in 2020 (Filho et al., 2020). Adoption of AMS has been largely driven by their decreased labour requirement when compared with conventional milking systems (Koning & Rodenburg, 2004). Robotic milking, however, has the potential to provide solutions for the dairy sector far beyond a reduction in required labour.

Modern AMS incorporate a wide array of sensor technology, capable of collecting production and behaviour data in real time. Of perhaps greater value still, is the opportunity provided by AMS for the interpretation and integration of this data. Through purpose built onfarm software, AMS offer a platform to amalgamate these data sources, centralise integration and provide decision support. In an environment of increasingly fragmented data sources, the value provided by a single comprehensive system such as offered by AMS cannot be overstated.

The collection of production and behaviour data on modern AMS offers an opportunity for the automated, objective assessment of transition cow health. Applied within a TMP, such data may have utility in the early identification of animals likely to suffer the cost and consequence of poor transition health. The following section reviews the literature surrounding selected production and behaviour variables commonly recorded on commercial AMS in the UK, their ability to reflect physiological status, and potential for incorporation into an automated transition cow monitoring program.

1.6 Production and Behaviour Data

1.6.1 Production Data

Milk Yield

Milk production rate, evaluated subjectively through udder tautness, or objectively via monthly test-day milk records have formed a part of transition monitoring programs since their inception. The advent of AMS offers not only real time assessment of milk quantity through integrated milk meters, but also the assessment of quality, specifically relating to milk fat and protein percentage as well as milk conductivity.

Within AMS, daily milk volume provides an easily measured objective assessment of milk synthesis rate. This metric is well established as a marker of health in the dairy cow (Mansell, 2003). The response of milk production in the face of disease, however, is complex. This may be in part due to the biological prioritisation of milk synthesis, particularly during early lactation (Martens, 2020) leading to the continued production of milk in the face of disease and negative energy balance (Rajala & Gröhn, 1998). Despite this, the investigation of milk production in response to disease has by and large found a reduction in milk yield days and in some cases weeks, prior to diagnosis of clinical disease. Edwards & Tozer, (2004) reported an average of a 15kg/day reduction in milk yield for animals diagnosed with transition disease vs healthy herd mates. For those diagnosed with an LDA, this reduction was detected 6 days prior to clinical diagnosis. These results align with those of Lukas et al., (2009) who reported a reduction in yield for all digestive disorders and pneumonia 4-9 days prior to diagnosis as well as Stangaferro et al., (2016) who reported a significant reduction in yield in response to a range of metabolic and digestive conditions including LDA, clinical hypocalcaemia and metritis.

Conflicting reports exist as to the association between milk yield and metabolic indicators of negative energy balance such as BHB and NEFA (Gross & Bruckmaier, 2019; King, et al., 2018). Across a large prospective cohort study involving 91 herds and 2,290 lactations, Ospina et al., (2010) reported a decrease in 305-day milk production in multiparous animals with elevated NEFA and BHB post-partum. However, milk yields in primiparous animals recorded a positive association with both metabolites. Controlling for BCS and calving season, while dichotomising BHB values (> 10ml/DL), NEFA values were returned as the sole significant parameter affecting milk yield (NEFA dichotomised ≥0.57 mEg/L associated with +488kg for 305 D Yield, P = 0.02) within this group. Replication of this analysis in multiparous animals, again returned NEFA concentration as the single significant predictor (NEFA ≥0.72 mEg/L associated with a 647kg decrease in 305 D yield, P = 0.001). The difference in physiological status of primiparous animals, when compared with older animals, which are not required to support the same level of growth, may to some extent explain this finding. However, while the balance of evidence points towards a negative association between indicators of

negative energy balance and milk production, sufficient contrary evidence exists to substantiate the likelihood of true variance in this association between individual animals. This is an area reviewed in greater detail by Duffield, (2000) and Horst et al., (2021). However, in brief, a logical premise for this phenomenon is that high yielding animals may mobilise large fat reserves to meet their energy needs. Individual animals may be capable of mobilising these large quantities within the parameters of an appropriate physiological response, (though concentrations of NEFA or BHBs may exceed accepted reference ranges) and thus avoid compromising their milk production potential. The conflicting findings as regards the association of metabolic indication for NEB and milk yield likely reflect the inherent inaccuracies associated with the use of strict diagnosis of disease states. Despite this, milk volume remains a crucial means of assessing physiological status in early lactation, particularly if this can be conducted early enough in lactation to enable pro-active disease management.

Milk Constituents

Due to the variance in production of milk constituents such as lactose, fat and protein, volume alone does not provide a complete account of the physiological costs of milk production. While lactose is the primary carbohydrate of milk and the main driver of volume, fat significantly affects the energy and nutrient cost of milk synthesis. The application of milk constituent data at the herd level where it has been more widely applied, has been described in Section 1.3. However, the advent of inline fat and protein sensors has led to an increased interest in the utility of milk constituents to reflect physiological status at the individual level.

In response to negative energy balance, early lactation dairy cows mobilise fat reserves leading to increased circulation of fat cells and the elevation of milk fat percentage. Similarly, as demands for energy increase, protein is broken down at an increased rate, reducing the quantity available for incorporation into milk. The outcome of these physiological responses is the elevation of the milk fat-to-protein ratio.

Evaluating the association between milk production traits and energy status, Mäntysaari et al., (2019) utilised concurrent sample of serum analysis of NEFA concentration with milk sampling at weeks 2,3 and 20 post-partum. Following assessment of energy status using serum samples, a forward stepwise regression was built using milk traits to explain individual animal energy status. Fat-to-Protein ratio (FPR) and milk yield variables formed the final model ($R^2 = 0.47$), with FPR being the most informative trait. Given the establishment of fat and protein as a marker of energy balance (Friggens et al., 2007, Gross & Bruckmaier, 2019), these associations are unsurprising. However, the use of laboratory grade spectrometers to quantify milk constituents as used in these studies cannot be overlooked. In-line fat and protein sensors, as used in AMS have received far less research attention. Across 484 cows from 9 AMS herds, (King et al., 2019) assessed in-line milk fat and protein monitors for the detection of ketosis as diagnosed by serum samples taken over the first 3 weeks post-partum. Statistically

significant association between FPR and serum BHB was observed (p < 0.001). However, large variation was also observed with low R^2 returned across the various FPR metrics analysed (R^2 = 0.04 to 0.09), a result the authors attribute to sensor calibration (See section 2.3.1 for further discussion). In-line fat and protein sensors are a novel technology. While peer reviewed analysis documenting their utility is lacking, it stands to reason that where close correlation with gold standard spectrometer values can be achieved, they can provide a useful assessment of energy balance in transition cows.

Milk Temperature

Body temperature, generally assessed as rectal temperature, is reported as one of the most commonly applied objective assessment of transition cow health within TMPs (Espadamala et al., 2016, König et al., 2023). While it has demonstrated some utility as a means of monitoring physiological status, its use, particularly in the days immediately postpartum, does appear to be limited (Kristula et al., 2001). Grouping animals based on disease diagnosis within the first ten days post calving (Infectious Disease, Metabolic Disease or None), Wenz et al., (2011) reported a significant elevation in temperature within both diseased groups (P = < 0.001). The highest temperatures and largest variations in temperature were observed in animals suffering infectious disease followed by those diagnosed with metabolic disease. While a febrile response to infectious disease could be expected and has been demonstrated to precede diagnoses such as metritis (Benzaguen et al., 2007). The change in temperature in response to metabolic disease is of interest as it may indicate the utility of temperature to reflect subtle change in physiological status, however, no further substantive evidence to support this is apparent. A single herd study (n= 217), in which vaginal temperature loggers were implanted for days 2-10 postpartum reported a significant elevation of body temperature in response to hyperketonaemia in primiparous cows. However, the number of cows was extremely small (n= 12), and this effect was not observed in multiparous animals (Burfeind et al., 2014). The variance in core body temperature during the early post-partum period means that any assessment of temperature must be interpreted with caution. This was demonstrated by Kristula et al., (2001), who found that over the first 10 days post-partum, 48% of clinically normal animals recorded at least one elevated temperature. The interpretation of temperature is complicated further within modern AMS, as milk temperature is measured as a proxy for body temperature. Reports of its utility for the assessment of physiological status are limited, however, a reduction in milk temperature has been associated with the diagnosis of disease such as LDA, lameness and, in contrast to (Burfeind et al., 2014), subclinical ketosis (King, et al., 2018). It has also been incorporated into statistical models designed for the detection of mastitis (Nagvi et al., 2022).

1.6.2 Behaviour Data

Behavioural changes in response to a change in physiological status may be profound and easily recognisable on a single inspection of the animal (e.g. Recumbency due to hypocalcaemia). However, more subtle changes such as motivated sickness behaviours, which represent an adaptive response to physiological stress can also be evaluated over repeated observations (Weary et al., 2009). Continuous monitoring of behaviour, as facilitated by PDT offers the opportunity to utilise changes in the intensity or frequency of observed behaviour as a marker of physiological status. Four metrics of dairy cow behaviour are commonly available on commercial AMS in the UK. These are the frequency and nature of cow-robot interactions, and quantity of concentrate dispensed, recorded by the milking robot itself, as well as rumination and activity data, recorded via wearable sensors.

Rumination

Rumination is a vital process for the maintenance of rumen health and efficiency of digestion (Krause & Oetzel, 2006). It is also regarded as an expression of natural behaviour and thus, an indicator of dairy cow welfare (Wang et al., 2016). The trajectory of rumination activity over the transition period in a healthy animal reaches its nadir on the day of calving. Pahl et al., (2014) reported a progressive decline in the week before calving, followed by complete cessation of rumination on average 123 minutes before calving until 355 minutes after. This sharp decline appears highly reliable and has been utilised in the automatic prediction of calving (Borchers et al., 2017). From here, a sharp increase is observed over the first week of lactation (Soriani et al., 2012). Peak rumination has been reported from day 9 (Stevenson et al., 2020) to 50 post-partum (Paudyal et al., 2018) before a plateau is reached. Reported average rumination times for Holsteins generally agree with those reported by Stevenson et al., (2020), approximately 400-450 minutes spent ruminating per day during the two weeks precalving, reaching a plateau of approximately 500 – 550 minutes per day post-partum.

A reduction in absolute rumination time has been reported in response to a range of clinical transition diseases. Comparing 403 animals diagnosed with transition disease between days 5-12 post-partum, with 300 healthy herd mates, a significant difference in total rumination times was detected between groups (Steensels et al., 2017). This difference peaked at 3 days prior to diagnosis of ketosis or metritis, with sick animals ruminating on average 90 mins less per day than their healthy herd mates. Adding weight to this association is the observation within this study of a recovery in rumination time following diagnosis and treatment. These results have been replicated across a number of studies (Stangaferro et al., 2016, King, et al., 2017, Paudyal et al., 2018). While these have in general, been composed of a small number of lactations from single herd studies, the reproducibility of the response of rumination to clinical disease in the transition cow adds weight to this association.

The effect of sub-clinical transition disease such as sub-clinical ketosis (SCK) and sub-clinical hypocalcaemia (SCH) on rumination time and has also been reported. Similar to the studies investigating the effect of clinical transition disease, participant numbers within these analyses tend to be limited in scale with interpretation clouded further by variation in the timing of sampling and cut offs used for diagnosis of disease. However, some through lines can be identified. Goff et al., (2020) reported a strong correlation (r=0.75) between rumination rates on days 1 post-partum and calcium concentration 12 hours after calving across a twenty-six-cow dataset. Across a larger dataset (n= 286) Liboreiro et al., (2015) found only a weak correlation (r= 0.15) between calcium concentration and rumination time. Though this finding which may be attributed to calcium sampling in this study taking place at any point within 72 hours of calving. Nevertheless, a significant reduction in rumination was evident from day 1-3 for animals suffering sub-clinical hypocalcaemia.

In the same study a weak correlation (r=0.08) between BHB and rumination time was detected, with a significant reduction in rumination time associated with the diagnosis of sub-clinical ketosis over the first 8 days of lactation. Across 4 commercial dairy farms this effect was substantiated by Kaufman et al., (2016). Assessing rumination time on a weekly basis, multiparous animals suffering sub-clinical ketosis demonstrated significant (P < 0.10) reductions in rumination time compared with healthy animals though this trend was not observed in primiparous animals except where SCK was accompanied by a second disease diagnosis. This variability in association between SCK and rumination in the post-partum period was demonstrated further by Schirmann et al., (2016), who reported no association between the diagnosis of SCK and rumination time despite serum BHB measurements being taken 3 times per week for the first 2 weeks postpartum. However, the small sample size (n =80) and low incidence of SCK (n=9) means these results should be interpreted with care.

The reduction in rumination in response to clinical transition disease appears to be a well-established and repeatable one. Larger studies examine the response of rumination to various levels of subclinical disease states would be of benefit; however, it appears that rumination time has some level of sensitivity to the calcium and ketone body concentration in the transition cow.

Activity

At its most basic, an animal's locomotor activity portrays its ability or willingness to move, a measure which can reflect the animal's response to changes in physiological status (Broom, 2006). These effects can be additive, for instance, a severe case of mastitis may lead to discomfort while walking, thereby reducing activity. The resulting fever and upregulation of inflammatory cytokines may reduce appetite and thus the drive to actively seek food, decreasing activity further (Broom, 2006, Tizard, 2008). Activity levels generally increase in the hours prior to calving, thereafter, a decrease from peak is observed with activity

levels generally stabilising early in lactation (Jensen, 2012). Decreased activity levels have been associated with the diagnosis of clinical transition diseases such as LDA, pneumonia, and metritis (King, et al., 2017). The extent of this decrease varies by condition, with animals suffering LDA demonstrating a 45% reduction in activity per-day from 12 days prior to diagnosis (King, et al., 2017). For comparison, animals diagnosed with pneumonia recorded a 34% daily decrease for the 5 days prior to diagnosis. Steensels et al., (2017) reported a general reduction in activity between healthy and diseased animals over the first 21 days post-partum, unlike King, et al., (2017) however, no significant difference between individual clinical diseases was observed. These results are bolstered by a relatively large study examining the response of activity to clinical disease. Analysing approximately 1,500 records across 3 farms Edwards & Tozer, (2004) demonstrated reduced activity in clinically ill cows (ketosis, retained placenta, and milk fever, LDA, indigestion, reduced feed intake, traumatic gastritis, acidosis, and bloating) when compared with healthy herd mates. This change in activity level was found to be significant (all P-values <0.002) for days -2 to +1 relative to diagnosis.

The precision with which activity reflects more subtle changes in physiological status was explored by Najm et al., (2020). Across a small sample set (n = 75) a statistically significant reduction in animals diagnosed with subclinical ketosis (SCK+) (n=6) compared with healthy herd mates was observed on day 6-12 post-partum (P < 0.001) with SCK+ animals recording lower than the group average activity level over these days. Liboreiro et al., (2015) also reported a weak but significant (P < 0.01) negative correlation between BHB concentration and activity level (r = -0.14). Interestingly, this study found no significant correlation between calcium concentration and activity, though it did tend towards significance (r = 0.09, P-value 0.17). This is in keeping with the observed activity of sub-clinically hypocalcaemic (SCH) animals reported by Barraclough et al., (2020), (SCH+ = 30, SCH- = 6) who reported significant reduction in the case of clinical hypocalcaemia but not sub-clinical hypocalcaemia.

The association of activity with physiological status broadly follows that of rumination in that significant disturbances in health are accompanied by a reliable response in activity levels. Disturbance at the sub-clinical level is clearly less reliable, with the absence of significant reduction in sub-clinical hypocalcaemia of particular note given this disease's direct effect on muscle tissue.

Visit Behaviour

Under free flow AMS conditions dairy cows may approach the milking robot throughout the day. Cow-robot interactions are recorded as Milking Visits; where a complete milking is carried out and concentrate feed provided, Refusals; where milking is refused due to an animal representing for milking too quickly following a previous milking visit, and Failure; Where milking is not successfully completed (see Section 2.3.1 for further details). As access to the concentrate feed provided by the

robot is the primary motivation for robot visits, the number and nature of each cow's interaction with the robot serves as an indicator of the animal's appetite and ability to access the robot and thus may provide information on the animal's physiological status (Bach et al., 2007, King et al., 2018).

Similar to that observed in studies examining the response of rumination and activity to changes in physiological status in transition cows, the literature describing the changes in visit behaviour is composed of small scale, often single herd studies. Within these, the response of visit behaviour seen to cases of lameness predominates, demonstrating its associated reduction in robot visits (Miguel-Pacheco et al., 2014, Steensels et al., 2016). Milking visits have been demonstrated to reduce in response to mastitis, a response hypothesised to be a protective action due to increased pain during milking (King et al., 2018). Within the same population, animals diagnosed with an LDA recorded 0.062 fewer milkings/d (P = 0.009). while those diagnosed with sub-clinical ketosis also tended to record a reduction in visits when compared with their healthy herd mates, though this difference was not statistically significant. The unique nature of robot visit metrics, not only to herds utilising AMS but to those operating a free-flow housing system means large scale studies investigating their association with transition disease are limited. However, considering the relevance of the factors which drive voluntary robot visits they remain an intriguing prospect for incorporation into a TMP.

1.6.3 Conclusions

The data generated by precision dairy technology as applied in AMS represents a significant opportunity for the development of transition cow monitoring programs. Production and behaviour data has a demonstrated ability to reflect both profound and subtle changes in physiological status during transition. In contrast to the previously applied monitoring techniques, such as the use of manual observation or metabolic indicators, this approach offers an objective, early and crucially, automated assessment of transition health. However, the potential this technology holds to facilitate improved transition cow management has yet to be fully explored. As demonstrated above, the majority of research within this field has focused on the association of production and behaviour data with the occurrence of transition disease in its clinical or sub-clinical form. There remains an opportunity to apply transition cow monitoring programs in a more holistic sense, focused on the broad cost and consequence of poor transition health in place of the diagnosis of specific disease states.

1.7 The Future of Transition Monitoring Programs

1.7.1 Defining the Cost and Consequences of Poor Transition Health

Transition disease, in its clinical or subclinical form, has invariably been the target of transition monitoring programs, initially using manual observation of clinical health and laterally via precision dairy technology. However, the cost and consequence of transition failure are broad and far reaching. Screening programs focused on the identification of specific disease states have an inherently narrow scope. This approach may therefore be ill-suited to the assessment of a complex physiological process such as that which takes place during the transition period.

An alternative approach may be one which utilises a more holistic assessment of the outcomes related to transition success or failure. One which is reflective of, as defined by LeBlanc, (2010), the cost and consequence of transition disease, compromised production and welfare. An approach focused on these broader outcomes may allow for greater flexibility in the assessment of transition health, accounting for both the complex nature of the challenges faced as well as the resilience of each individual cow.

Beyond the increased incidence in disease, the cost and consequence of poor transition health may be broadly viewed through its deleterious effects on dairy cow survival, milk production, and fertility. Assessed in early lactation, performance in these areas is reflective of the success with which the transition cow has adapted to the challenge of the initiation of lactation (Probo et al., 2018, Stevenson et al., 2020, Pascottini et al., 2022). Their incorporation into TMPs may more fully encompass the complete spectrum of transition cow health, allowing producers to monitor performance based on broad outcomes with demonstrated economic, welfare and social impacts within their industry (De Vries & Marcondes, 2020).

Of further value to producers is the potential for the development of TMPs, which can predict these long-term performance metrics, facilitating intervention to prevent or mitigate losses. A prognostic approach to TMPs would stand in contrast to the retrospective, diagnostic approach which has been predominant within this field to date. The following sections reviews the existing literature relating to the use of transition cow data in the inferential and predictive modelling of removals in early lactation, milk production, and reproductive performance. The following sections review these three metrics of transition success.

1.7.2 Removals in Early lactation

Herd removals can be classed as voluntary or involuntary. Voluntary removal represents the planned culling of animals which the producer deems to have reached the end of their productive life or, the sale of animals for continued milk production elsewhere. Involuntary removals are those which occur through unplanned culls or on farm mortality. The financial impact of involuntary removals and its effect on the economic sustainability of the dairy industry is well documented (Kerslake et al., 2018). However, they also affect the social sustainability of dairy farming and are widely used as a welfare assessment within the UK (RSPCA Welfare Standards, 2023). The ability to identify animals at risk

of early lactation removal may provide producers with an opportunity to intervene and reduce the risk of loss.

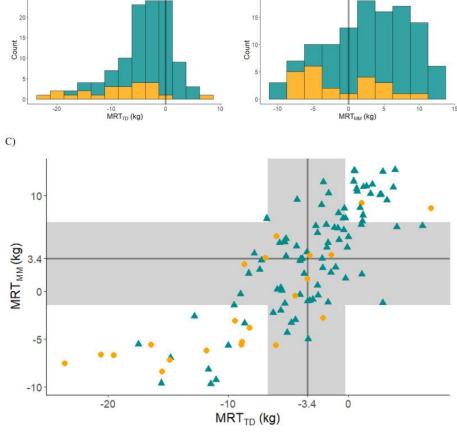
Early lactation is a high-risk period for dairy cow mortality. In a large survey of dairy cow mortality in Denmark from 1990 to 2001, 30% and 41% of mortality was recorded within 30 days of calving for parities < 3 and 3+ respectively (Thomsen et al., 2004). While the association between clinical transition disease and increased cull risk has been well described (Table 1-1), metabolic indicators, specifically BHB, NEFA and calcium in peri-parturient cows also have a well-established association with cull risk in early lactation. Through an amalgamation of data collected from four clinical trials, Roberts et al., (2012) constructed a dataset of 5.979 lactations from 69 farms. BHB concentration of >1.2mmol/L in the first week post-partum, returned an odds ratio of 1.8 for culling within 60 DIM (p= <0.001). Calcium concentration (<2.2 mmol/L) and NEFAs (>0.8 mmol/L) also returned significant association, with odds ratios of 1.5 and 2.0 respectively. These results are corroborated widely across a range of studies. Seifi et al., (2011) found significant associations between serum calcium and NEFA concentration in the first two weeks post-partum and cull risk by 60 DIM. More recently, reports by Venjakob et al., (2018) and Menta et al., (2021) has strengthened this association with the authors reporting significant association between early lactation cull risk and periparturient calcium and BHB respectively. These studies demonstrate the relationship between physiological status in early lactation, as assessed by clinical diagnosis and metabolic indicators, and the subsequent risk of removal from the herd. However, there has been no investigation of the association between early lactation production and behaviour parameters with early lactation cull risk.

The prediction of cull risk using transition cow data has received limited attention within the literature. Lukas et al., (2015) investigated the utility of daily milk yield in the first week post-partum for the prediction of cull risk by 100 days in milk. The analysis of 807 lactations over 3 herds evaluated daily milk yield, rate of increase in milk yield and the difference between recorded and expected milk yield – termed Transition Success Measure (TSM). TSM was found to be the strongest and most reliable predictor of cull risk across all herds. Following development of 3 farm specific predictive models using TSM, the Area Under the Receiver Operator Curve (AUC ROC) for the predicted risk of cull when assessed using a withheld test dataset was 0.82, 0.86 and 0.73 for herds 1, 2 and 3 respectively. As the authors did not report the performance of these models when applied on a previously unseen herd, their generalisability remains unknown. However, this study does demonstrate the potential of production data for the prediction of cull risk. To date no investigation of the predictive power of sensor data such as rumination or activity, for the likelihood of cull has been carried out.

1.7.3 Milk Production

Reduced milk yield is one of the most common and economically consequential effects of poor transition health (Liang et al., 2017). Its use in the monitoring of transition cows as part of a monthly test-day milk recording schemes and the inherent disadvantages of this approach have been descried in Section 1.3. A more appropriate alternative may be the use of yield deviation. An animal's yield deviation represents the difference between observed and expected yield over a given time. Its calculation involves the generation of an expected yield for each individual animal based on their prior production levels. Observed yield is then compared with expected and the difference regarded as a deviation. In contrast to the reliance on comparison with producers' expectations, or the production of herd mates as described in Section 1.3 and displayed in Figure 1-1, the use of yield deviation aims to utilise the cow as her own control. This approach aims to provide a more accurate reflection the animal's physiological status and thus, a more useful assessment of transition health.

Deviation from expected yield has been investigated as both a marker of resilience in dairy cows (Elgersma et al., 2018) and a tool for the retrospective monitoring of transition cow health (Nordlund, 2006). The assessment of deviation form expect first test-day yield was developed as a commercial transition monitoring tool, the Transition Cow Index (TCI) (Nordlund, 2006). As part of the validation process the relationship between deviations identified by the TCI and clinical disease within 7 days of the first test day record was examined. Across 18,814 lactations from 30 herds animals recording cases of metritis, ketosis, lameness or left displaced abomasum demonstrated a negative deviation when compared with healthy animals. Similarly, Salamone et al., (2024) found early lactation health and metabolic status to be associated with yield deviation, with deviations observed to be significantly more negative in diseased vs non-diseased animals (Figure 1-2). Within a multivariable model using the same dataset a significant negative association between serum NEFA concentration and yield deviation was also observed (P = < 0.001). Yield deviation therefore, represents a viable method for the assessment of physiological status in the transition cows. As applied within the TCI the use of yield deviation offers a valuable, but retrospective method of transition health assessment. To date no investigation of the accuracy with which yield deviation in early lactation might be predicted has been carried out. Where this could be achieved and incorporated into a TMP, it may provide valuable information to producers and offer an opportunity to mitigate or prevent production losses.



B)

Figure 1-2 Graphical demonstration of the relationship between Yield deviation from expected yield and the diagnosis of clinical disease. Description from source (Salamone et al., 2024). (The terms Yield Deviations and Yield Residual are equivalent.) Panels A and B show the milk yield residuals in the transition period (MRT) for test-day (MRTTD) and milk meter (MRTMM) distribution. The colours distinguish between non clinically diseased (green) and diseased (orange). Both colours are stacked on top of each other. The bin widths in panels A and B are 2.5. In panel C, the relation between the MRTTD and MRTMM is plotted. A distinction is made between the clinically diseased (•) and non-clinically diseased (•). The axes are located on the median of each MRT. Distribution bands were also plotted to represent the interquartile ranges.

1.7.4 Reproduction

A)

As our understanding of the physiological processes influencing reproduction in the dairy cow has grown, the importance of transition health has become increasingly apparent. The negative effects of clinical transition disease, particularly those associated with the reproductive tract, on subsequent fertility have been well described (Table 1-1) (Gilbert, 2019). The association between early lactation subclinical disease, as assessed by a wide range of metabolic indicators, and subsequent reproductive performance has also been investigated (Butler, 2013, Nigussie, 2018). The interpretation of associations between these metabolic indicators and fertility are complicated by the variance in sample timing and cut off selection, as

well as the varied application of hormonal protocols across studies. However, some broad conclusions on this relationship can be drawn.

Across the majority of the available literature, negative energy balance in the transition period has been demonstrated to exert a detrimental effect on subsequent fertility. A meta- analysis carried out by Raboisson et al., (2014) on the effect of subclinical ketosis (BHB > 1.4mMol/L, NEFA >0.4mMol/L pre- or >1.0 post- partum) on subsequent reproductive performance found an odds ratio of 0.67 for conception to first service. A longer calving to first service and calving to conception interval were also observed. As regards the effect of NEB on cyclicity, the odds ratio of experiencing early ovulation (< 33 DIM) were found to be 5.72 for healthy animals when compared with those diagnosed with ketosis within the first 60 days (Stevenson et al., 2020). In the case of hypocalcaemia, the association of calcium status with reproduction is not consistent across the literature (Couto Serrenho et al., 2021). However, two large multi-herd studies demonstrated reduced odds of conception to first AI for animals diagnosed with hypocalcaemia within the first 3 weeks post-partum (Chapinal et al., 2012, Venjakob et al., 2018).

The association of production parameters and fertility performance was assessed over 312 UK herds by Hudson & Green, (2018). Protein percentage and milk yield at first test-day returned significant positive associations with the risk of conception, (OR= 1.05 and 1.16 respectively) between days 20 and 150 in milk. Butter fat returned a slight but significant negative association (OR= 0.98). However, these factors, in combination with production parameters from test day 2 failed to account for a substantial portion of the variance in conception risk observed within this dataset (R²= 0.22). These results broadly agree with those of (Madouasse et al., 2010), which observed the effect of first test-day protein percentage and fat on the probability of conception (DIM 20-145) across 2,128 UK herds. Though milk yield at first test-day was not found to be significant within this dataset. The association of milk constituents in early lactation on fertility within an Irish spring calving system was assessed over 87,227 cow lactations by Carty et al., (2020). Within the first 30 days post-partum milk protein and milk yield were associated with increased hazard of pregnancy for animals submitted to service, a single unit increase in each yielding a 12% and 1% increase in pregnancy hazard. A quadratic relationship between milk fat and pregnancy hazard was observed over this period, with a positive association reported up to 4.6%. Overall, however, milk constituents in this study were found to exert only a modest effect on the hazard of pregnancy. These three large studies highlight a statistically significant but generally small effect which milk production parameters in early lactation exert on subsequent fertility performance.

In comparison to reports investigating the relationship between, metabolic or production parameters and subsequent fertility, those exploring transition cow behaviour parameters are limited and small in sample size. A single cohort study which analysed the effect of milking frequency on reproductive performance within a herd utilising AMS

found no significant association between visit frequence within the first 100 days and probability of submission or conception-based metrics (Talukder et al., 2015). Another single herd study reported a positive association between rumination time in the 21 days pre-calving and subsequent time to pregnancy, though this was not replicated for rumination in the post-partum period. The association of activity levels in early lactation has received greater research attention, largely focused on the recording of oestrus via wearable activity sensors. Bretzinger et al., (2023) demonstrated that the expression of oestrus in DIM 7-60 was associated with subsequent intensity of heat at AI and risk of becoming pregnant within DIM 200. A finding which was echoed by Borchardt et al., (2021) investigating the recording of automatically detected oestrus from days 7-40 post-partum. Those which failed to record an oestrus had significantly reduced reproductive performance as measured by hazard of insemination within 100 DIM, and time to pregnancy. These studies provide some insight into the relationship between transition cow behaviour and reproductive performance; however, this remains an area in need of further research. Of particular benefit would be the examination of behaviour based solely within the transition period evaluated on a multi-herd basis.

Similarly, the prediction of reproductive performance using transition cow behaviour data remains largely unexplored. To date predictive models have focused on data from later in lactation, often within the voluntary waiting period or in close proximity to the time of insemination. For example, the intensity and duration of increased activity at the time of insemination has been used to predict the likelihood of conception (Marques et al., 2024). While this is an understandable approach, the demonstrated association between transition performance and fertility (Roche et al., 2018), suggests the predictive power of behaviour parameters during this time is worthy of further investigation. Where this could be established, the ability to identify animals likely to experience reduced reproductive performance may be incorporated into TMPs to facilitate pre-emptive action.

The ability to predict reproductive performance in early lactation offers great potential for the implementation of targeted reproductive management (TRM). This field of research seeks to categorise animals by their expected reproductive performance and thus facilitate bespoke management strategies to maximise reproductive efficiency (Giordano et al., 2022). This approach has been applied in the selective use of sexed semen (Berry, 2021). as well as a targeted administration of exogenous hormones (Gonzalez et al., 2023). However, this remains a novel field of study in which the applications of data derived from AMS has yet to be investigated.

1.8 Conclusions and Objectives

Transition cow monitoring programs have seen several significant developmental shifts since first implemented. The first was the

progression from the manual, subjective assessment of physiological status, to the use of objective markers such as monthly test-day milk records, and metabolic health indicators. Buoyed by rapid progression in the field of metabolic analysis, and an ability to examine large number of markers from a single sample, the investigation of metabolomics for the assessment of the physiological status of the transition cow under research conditions expanded rapidly. Despite this however, its application on farm remained limited due to the labour-intensive nature of sampling and the limited availability of cow-side tests. Therefore, despite a large volume of published work documenting the association of various metabolic indicators with physiological status, their integration into TMPs has been limited.

The slow pace with which metabolic analysis was adopted on farm stands in contrast to that of precision dairy technology. Sensors capable of monitoring dairy cow production and behaviour have recorded rapid uptake across the dairy industry in the past decade. In further contrast to metabolic analysis, this adoption of PDT was arguably achieved without sufficient publication of validation studies or development of the tools necessary for data interpretation (Stone, 2020). The rapid increase in the availability of this data does mean however, that where progress in the interpretation and integration of this data can be made, on farm impact may be possible in a shortened time frame as the mean of data collection are already widely in use.

A much slower development in the field of TMPs has been the shift in the outcomes of interest from diagnosis of transition disease to the prediction of long-term outcomes following transition. The pace of development in this regard is understandable. Transition disease, particularly its clinical form, presents a clear and obvious welfare concern to all stakeholders within the dairy industry. Their negative effect on the social, economic, and environmental sustainability of the industry ensures that their prompt diagnosis and treatment will remain a vital aspect to transition cow management. It remains the fact however, that despite the development seen in our ability to monitor transition cows, the incidence of transition disease has remained static for the past two decades. Therefore, further consideration of alternative approaches to TMPs, those which focuses on early identification of animals expected to experience the cost and consequence of poor transition health would appear warranted.

In the use of long-term outcomes reflective of transition failure, predictive models provide an opportunity to employ management strategies in early lactation which may reduce potential losses. To achieve this requires the development of models which accurately predicted each animal's expected performance. The potential value in such programs, developed using production and behaviour data as collected by AMS, lies not only in the volume and variety of automatically collected data, but in the infrastructure already in place to facilitate interpretation, integration and crucially, action at the farm level.

The aim of this thesis was to investigate the relationship between production and behaviour data as collected by AMS in the early post-

partum period and, subsequent performance assessed using risk of removal, milk production and reproductive performance in early lactation. An emphasis was placed on the predictive power of this data and its potential utility within a prognostic transition cow monitoring program. This study was conducted following ethical approval from the School of Veterinary Medicine and Science, University of Nottingham, Committee for Animal Research and Ethics (Reference No. 3404 210708).

Chapter 2 General Introduction to Material and Methods.

2.1 Introduction to the Study Data

The data analysed throughout this thesis was collected from herds utilising Lely AMS in the UK and Republic of Ireland between January 2016 and June 2023. It is comprised of a combination of automatically collected sensor data and manually entered farm records. These were remotely accessed and extracted from Lely's on farm software Time 4 Cows ® (T4C), via Lely's third-party application programming interface (API) (https://api-integration.lely.com/index.html). Data was collected from participating herds at regular intervals over the lifetime of the project and as such, no single "master dataset" was utilised for analysis. Rather, each individual piece of analysis used the most current dataset available at its initiation. This chapter aims to describe the herd recruitment and data extraction process. Data handling procedures specific to each individual analysis are described in their respective chapters.

2.1.1 Farm Recruitment

Lely International headquarters are based in Masluaus, the Netherlands. From here, regional management is disseminated to areas termed "Clusters". Farms within the UK and Republic of Ireland are managed under the Atlantic Cluster, based in Birmingham UK. Operating within each cluster are individual Lely Centres. These serve as direct points of contact for producers utilising Lely AMS. Each centre employs Farm Management Support (FMS) advisors who are responsible for customer care. Throughout this project all contact with Lely clients was carried out by FMS advisors exclusively.

Prior to the commencement of recruitment, criteria for herd eligibility were established. Herds were deemed eligible for inclusion if they were based in the U.K. or Republic of Ireland, have been utilising at least 2 Lely Astronaut Milking Robots ® exclusively for at least two years, under a "free flow" traffic system (Munksgaard et al., 2011) and were utilising SCR® collar rumination and activity monitoring technology (Rebranded by Lely as Lely Qwes-HR collars, Lely International N.V.).

These criteria were applied to maximise the volume and consistency of data available from recruited herds. The requirement that each herd have two robots, operating over two years was put in place to ensure an adequate number of animal records. The requirement for SCR animal monitoring technology was to standardise monitoring technology across the dataset (see Section 2.3.2 for further discussion).

Facilitated by both Lely Head Quarters and the Atlantic Cluster, all Lely centres in the U.K. and ROI were approached to request assistance in the recruitment of farms for participation in this study. At this time, 8 Lely centres were operational within the Atlantic cluster. All, with the exclusion of Lely Centre Ayre, agreed to participate in the study. Of the seven participating centres two were in the Republic of Ireland; Mitchelstown and Mullingar, one in Scotland; Kilmarnock, one in Northern Ireland; Eglish, and the remaining 3 in England; Birmingham, Holsworty and Yeovil. A map detailing the geographical location of each participating Lely centre is presented in Figure 2-1. Details relating to the numbers of herds recruited by each centre are presented in Table 2-1.

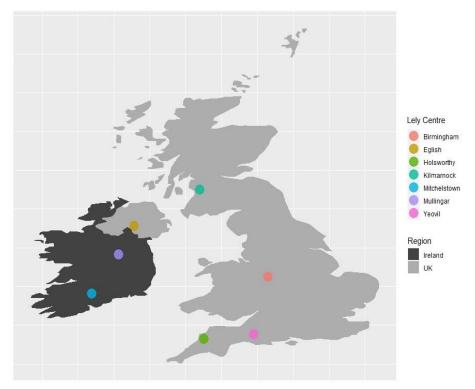


Figure 2-1 Location of participating Lely Centres in the UK and Republic of Ireland.

Table 2-1 Number of recruited by each participating Lely centre

| Region | Lely Centre | No. of herds recruited |
|---------------------|--------------|------------------------|
| Republic of Ireland | | |
| • | Mullingar | 9 |
| | Mitchelstown | 3 |
| Norther Ireland | | |
| | Eglish | 6 |
| England | - | |
| - | Birmingham | 6 |
| | Holsworty | 6 |
| | Yeovil | 12 |
| Scotland | | |
| | Kilmarnock | 4 |

The process of herd recruitment was carried out over approximately 6 months from January 2021 to July 2021 and proceeded as follows. A brief overview of the project including all criteria for eligibility was disseminated to FMS advisors. FMS advisors were asked to identify farms which satisfied all the eligibility criteria and approach eligible herds to assess their willingness to participate. A consent form, authorising the use of their data in the study was distributed to all herds which agreed to participate (Appendix 10.1).

In total, consent forms for 46 herds were returned. The herd recruitment procedure utilised represents a non-probabilistic convenience sample. The eligibility criteria set out above is biased against those within the first two years of operation and those utilising a single robot. A further source of bias is the voluntary nature of participation. As all data generated on farm remains property of the producer, the data utilised in this study could not be accessed unilaterally thus, the need for volunteers was unavoidable. However, as participation in this study did not require extensive input from the producers (for example, the requirements to complete an interview or survey) the cost of participation was low. This may have reduced the effect of volunteer bias in this study. A third source of selection bias is the role of FMS in the selection of farms for participation. As the researcher had no direct contact with producers, the choice of those invited to participate was at the discretion of FMS; which may have been subject to some selection biases based on working relationships. An alternative approach to recruitment considered at the outset of the project was the random selection of herds fulfilling the selection criteria. An ordered list of farms for recruitment would then be passed to FMS advisors with the request to approach each farm in turn until a set number were successfully recruited. Following consultation with management at Lely Atlantic Cluster this approach was not pursued due to the extra workload placed on FMS advisors volunteering their time to recruit farms. By simplifying the recruitment process it was possible to increase the number of farms

recruited within the study period, however these biases need to be considered in the interpretation of results.

2.1.2 Data Extraction

Following recruitment to the study, data retrieval was initiated. This was carried out on an individual herd basis as and when herds were recruited. The researcher established a sharing licence within Lely's third-party API. This licence defined the type of data available from recruited herds and the time span for which access would be granted.

Upon agreement to participate and the return of a signed consent form, a herd identifier was provided by the FMS advisor to Lely International and the herd added to the sharing licence. This enabled an option to activate an API link on each farm's T4C software. FMS advisors were then asked to access T4C for each recruited farm in person, or via TeamViewer® to activate this link. Once activated a farm key was generated. This was provided to the research team and served to allow herd access via Lely's API.

Data retrieval was carried out using R statistical software (R Core Team 2021). The "POST" function within the JSONlite package (Ooms, 2014). was used to offer licence security information in addition to the farm key to Lely's API. This provided access to each farm's data individually. Thereafter, the "GET" function was used to request specified API data. For each data scrape a total of 14 separate requests were made per herd (Table 2-2).

2.1.3 Data Handling

Following completion of each herd scrape, the data retrieved was converted from JavaScript object notation (JSON) to a vector. Thereafter, the herd was assigned a random numerical identifier, all animals were assigned a numerical identifier within their respective herd and all cow-lactation assigned an identifier comprised of the cow identifier, herd identifier and lactation number. All data which might be used to identify the herd (e.g., Government ear tags numbers, Lely herd identifier) was then deleted. An animal lactation and herd identifier as well as a date and a Days in Milk (DIM) indicator was associated with each individual observation across all data frames. With each successive data scrape, new data was amalgamated with prior scrapes on a herd basis. When a dataset was required for analysis the most up to date herd datasets were merged into a multi-herd dataset. This was then brought forward for data assessment, manipulation and analysis.

Table 2-2 API requests used for data extraction

| Lely API Address | General Description |
|--------------------------------|---------------------------------------|
| /api/animals | Animal identifiers, Date of birth, |
| | Gender, Herd identifier |
| /api/milkvisitrobotdata | Data relating to milking time, Dead |
| | time, Milking speed and teat |
| | position, reported on a per visit |
| | basis |
| /api/milkvisits | Milk weight and animal weight on |
| | a per visit basis |
| /api/milkdayproductionsquality | Daily total of milk production, Fat & |
| | protein indications, Successful |
| | milking visits, Refusals and |
| | Failures |
| /api/milkvisitsquality | Conductivity and milk temperature |
| | on a per visit basis |
| /api/feedvisits | Concentrate dispensed per robot |
| | visit |
| /api/calvings | Calving Dates |
| /api/dryoffs | Dry off Dates |
| /api/heats | Date of heats as detected by |
| | activity monitoring |
| /api/inseminations | All insemination records |
| /api/pregnancies | Date and result of last pregnancy |
| | check, Date of last insemination |
| /api/currentreprostatus | General reproductive data on a |
| | cow basis including calving date, |
| | current status, date of last |
| | insemination, number of |
| | inseminations, date of last heat |
| /api/activities | Activity data reported on a two |
| | hourly basis |
| /api/ruminations | Rumination data reported on two- |
| | hourly basis |

2.2 Data Quality

2.2.1 Data Access Limitations

Technical issues relating to the generation of a farm key prevented one Scottish herd from being added to the project's sharing licence. An API connection could not be established for two herds in the Republic of Ireland and two in England. This was assumed to be the results of poor internet connectivity with the on-farm computer. Finally, API data was absent for two herds, one in the Republic of Ireland and one in England. The cause of this was not established and the herds were excluded from analysis. Of the 46 herds for which a signed consent forms received; data was available for thirty-nine.

Lely's third-party API places a restriction on access to rumination data. In general, historical herd data is retained within Lely T4C for several years following recording. However, rumination data is retained for the past 365 days only, thereafter records are deleted. This restricted access to rumination data to one year prior to the day on which the first data scrape was carried out and created a large disparity in data volume between rumination data and all other data.

As outlined in section 2.1.1, eligibility for inclusion in this study required the use of SCR monitoring of rumination and activity. Over the course of this project rumination and activity data for a total of 10 farms became unavailable for analysis. The cause was, in the majority of cases established as the adoption of new NEDAP® monitoring technology in place of SCR. This resulted in subsequent data being unusable as no comparison between monitoring devices could be established. Where this occurred rumination and activity data prior to the change in monitoring technology was retained and utilised.

2.2.2 Data Overview

Data handling and missingness for the variables utilised in each analysis are discussed within their respective chapters. However, as a general introduction to the data utilised in this thesis, a brief overview is supplied here.

Following a data scrape completed across all available herds on 6/8/23 production records for 33,813 lactations from 12,736 cows across 39 herds were available for analysis. The percentage of those herds located in the England, Scotland, Wales, Republic of Ireland and Northern Ireland were 36%,3%,8%, 38%, and 15% respectively (Table 2-3). Thirty eight percent were spring block calving with the remainder calving all year round. AMS milking events were recorded from 2016 to 2023 with the majority recorded in 2021 (Figure 2-2) The average herdlevel 305-day milk yield ranged from 5,759 to 14,762 kg (Figure 2-3). Parity demographics of the dataset are presented in (Figure 2-4). Thirty percent of the dataset related to records from primiparous animals, 24% to second lactation animals and 46% to animals in third lactation or greater. Rumination records for 12,477 lactations from 7,554 cows across 39 herds were available. Recording dates ranged from 2020 to 2023 with the majority recorded in 2022 (Figure 2-5). Activity records for 19,632 lactations from 7,986 cows across 39 herds ranging from 2018 to 2023 were available (Figure 2-6).

Table 2-3 Descriptive statistics for the 39 herds with accessible data

| Variable | No. [range] |
|------------------------------------|---------------------------|
| No. of Farm | 39 |
| Mean No. of Milking cows per farm | 148 |
| Mean No. of AMS units per farm | 3 |
| Mean Milk Production/Cow/Year (Kg) | 10,184 [5,759–14,762] |
| Calving Pattern | % of Dataset (Herd Level) |
| All Year Round | 62 |
| Seasonal | 38 |
| Geographical Region | |
| England | 36 |
| Scotland | 3 |
| Republic of Ireland | 38 |
| Northern Ireland | 15 |
| Wales | 8 |

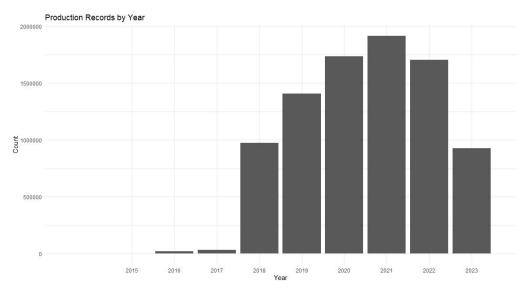


Figure 2-2 No. of successful milking events recorded by year across all herds

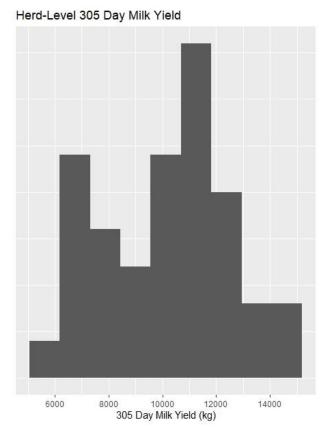


Figure 2-3 Mean Herd-Level 305-day milk yield across all 39 herds. Calculated as an average of cumulative 305-day yields recorded in 2020 and 2021

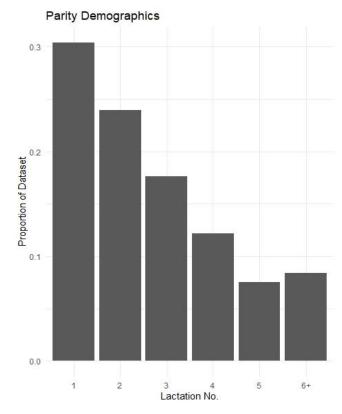


Figure 2-4 Parity demographics for all available lactations across all herds from 2016-2023

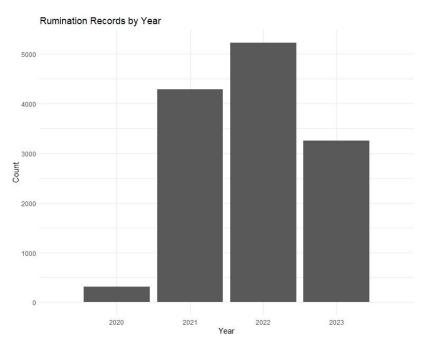


Figure 2-5 Cow-Lactations recording rumination events by year across all herds

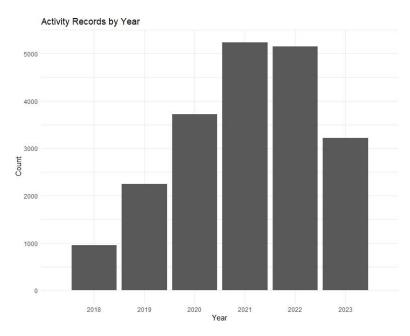


Figure 2-6 Cow-lactations recording activity events by year across all herds

2.3 Production and Behaviour Data

Production and behaviour data available for analysis within our dataset can be broken down into 2 broad categories; data generated through cow-robot interactions and data generated via wearable technology - in this case - neck mounted accelerometers. Production and behaviour parameters extracted from Lely's API during this project are detailed below. An overview of the API requests utilised is available in Table 2-2. Graphical representation of production and behaviour data from days 1-21 for all available lactations are presented in Figure 2-7 to Figure 2-15.

2.3.1 Cow-Robot Data

Data generated from cow-robot interactions is comprised of those relating to milk quantity, milk quality, feed dispensed, robot visit frequency as well as milk fat and protein indications. These are recorded via their respective sensor, stored in T4C before being made accessible via Lely's API.

Robot visits are recorded when the transponder on the cow's collar is identified within the robot. Lely's API presents robot visits as; Successful, Failed, Incomplete, or Refused. Each are recorded via T4C as totals per cow per day. A successful milking is defined as one in which over 80% of the expected yield is harvested. A failure is a visit where milking was attempted but not initiated (for example due to a failure to attach all four milking cups). Incomplete milking visits occur where less than 80% of expect yield is harvested (for example where clusters are kicked off prior to completion). Finally, a refusal is recorded where an animal is denied a milking permission due to their representing at the robot too quickly following a previous successful milking visit.

Some variation may exist between farms in relation to the definition of these visits. For instance, Lely offer standard definitions within their software for when an animal should be refused a milking. General recommendations are that animals not expected to yield at least 8kg of milk at presentation should be refused in order to improve the robot availability. However, producers have the ability to change this setting and therefore, the definition of refusal for their herd in comparison to others. Field experience suggests that this rarely occurs.

Farms recruited to this project operated what is known as a "free flow" traffic system which allow cows constant access to the milking robot throughout the day. All milking visits recorded within our dataset were treated as voluntary visits. In practice however, a proportion of these are likely to be involuntary visits for which the cow was manually brought to the robot or "fetched". Fetching cows is commonly required in the early post-partum period, particularly in first lactation animals (Drach et al., 2017). While the ability to differentiate between visit types would be of interest, at present no differentiation between voluntary or fetch visit can be made using Lely's software.

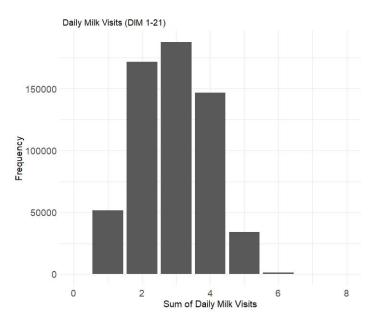


Figure 2-7 Distribution of successful milking visits recorded from days in milk 1-21 across all herds from 2016-2023

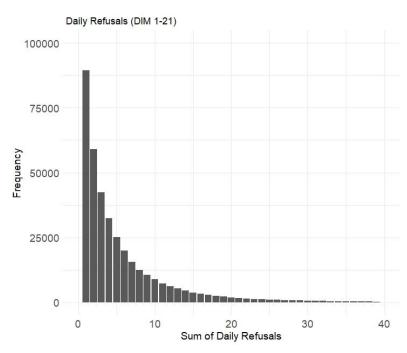


Figure 2-8 Distribution of refusals recorded from days in milk 1-21 across all herds from 2016-2023

Milk yield is measured on a quarter level via electronic milk meters and reported in kilograms (Kg) at the cow-level from midnight to midnight on a daily basis. This data is accessible via Lely's "/api/milkdayproductionsquality" API request. The distribution of daily milk yield record overs days 1-21 in milk for all available lactation is presented in Figure 2-9.

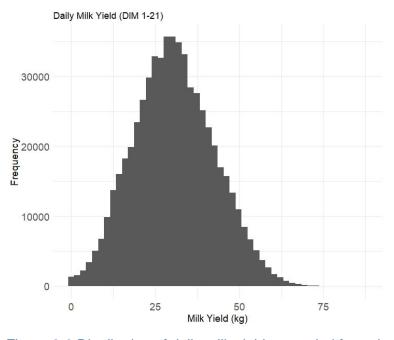


Figure 2-9 Distribution of daily milk yields recorded from days in milk 1-21 across all herds from 2016-2023

Conductivity is measured on a quarter level at intervals of 5 seconds across the milking process. A single quarter is then recorded as an arbitrary unit representing a mean of all readings across the milking event. This is reported at the quarter level on a per-visit basis and available via the "/api/milkvisitsquality" API request. Any quarter exceeding 80 (AU) across a milking event is flagged as a possible case of mastitis. Data relating to milk temperature is retrieved under the same API request as conductivity and is reported on a per visit basis in Degrees Celsius. Temperature is measured by in-line sensor. As milk must travel through approximately 1 meter of milk line prior to reaching these sensors, measurements are likely to vary with the volume of milk produced and environmental temperature at the time of milking. Prior investigation of the correlation between milk temperature as measured by Lely AMS and vaginal temperature reported only moderate correlation (r= 0.52) (Pohl et al., 2014). The distribution of temperatures available for analysis are presented in Figure 2-10

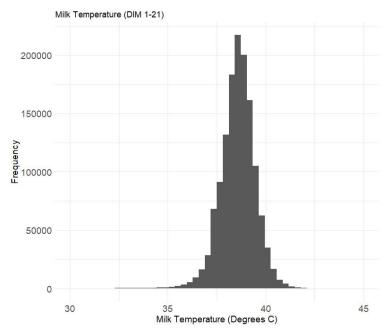


Figure 2-10 Distribution of milk temperature recorded from days in milk 1-21 across all herds from 2016-2023

Fat and protein indications are provided by in-line sensors based on analysis of milk fat globule size and are available via Lely's "/api /milkdayproductionsquality" API request. When compared with monthly test-day regimes this analysis has demonstrated moderate correlation (Fadul-Pacheco et al., 2018). However, sensor calibration is required to optimise the accuracy of indications. No information was available on the frequency with which farms in this study calibrated their sensors. Milk fat and protein indications for all available lactations is presented in Figure 2-11 and Figure 2-12 respectively.

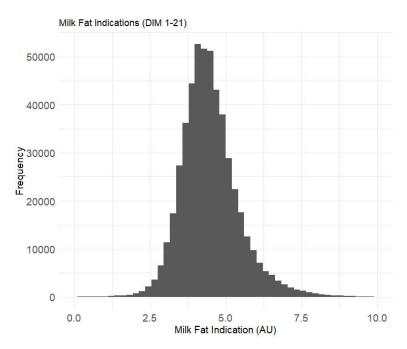


Figure 2-11 Distribution of milk fat indications recorded from days in milk 1-21 across all herds from 2016-2023

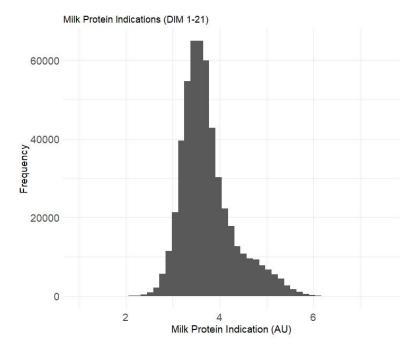


Figure 2-12 Distribution of milk protein indications recorded from days in milk 1-21 across all herds from 2016-2023

The weight of concentrate dispensed by the AMS is retrieved via the "/api/feedvisits" API request and is reported on a per cow per day basis. The level of concentrate dispensed is at the discretion of the producers, though Lely provide recommended feed tables which are automatically adopted by T4C. No information relating to the feed tables in use on any participating herd was available. While the weight of concentrate

dispensed is recorded, no indication of the proportion consumed by the cow is available. The distribution of concentrate dispensed per cow per day over days 1-21 is displayed in Figure 2-13.

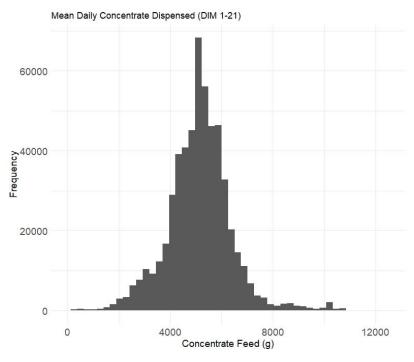


Figure 2-13 Distribution of concentrate feed dispensed recorded from days in milk 1-21 across all herds from 2016-2023

2.3.2 Activity and Rumination

Wearable technology, incorporated into Lely T4C software is necessary to allow cow identification upon entering the robot. However, an increasing number of farms are opting to pair this technology with rumination and activity sensors. Lely customers wishing to employ rumination and activity monitoring have a choice of two suppliers, SCR and NEDAP. These are distinct pieces of technology utilising proprietary hardware and software. To standardise the wearable technology used across the dataset, only farms using SCR collars were eligible for inclusion in this project. This decision was based solely on the larger proportion of farms within the Atlantic Cluster employing SCR when compared with NEDAP.

Activity and Rumination were detected using a previously validated neck mounted accelerometer (Elischer et al., 2013). Under Lely's API, rumination is reported in 12 two-hour blocks per day. This represents the sum of rumination over the past 24 hours. No indication relating to eating time was available. Activity readings are collected using a tri-axial accelerator capable of detecting motion in the orthogonal planes and reported as an arbitrary unit. Acceleration associated with upward movement of the cow's head, such as when it bobs during walking is translated using proprietary algorithm into activity with higher readings reflective of higher activity levels. No specification of activity type is available. For example, no distinction was made between walking, standing or lying.

For both metrics, data is summarised in two-hour intervals and stored for a maximum of 22 hours in a data logger housed within the collar. A reader placed within the robot allows the data logger to upload this data and reset memory availability during milking visits. Where a connection between the collar and stationary reader is not established for over 22 hours the first 2 hours reading within the data logger is overwritten. This overwriting procedure will continue iteratively until a connection is made. Rumination and activity data were accessed via Lely's "/api/ruminations" and "/api/ activities" API requests respectively. The distribution of 2-hour activity and rumination records are presented in Figure 2-14 and Figure 2-15 respectively.

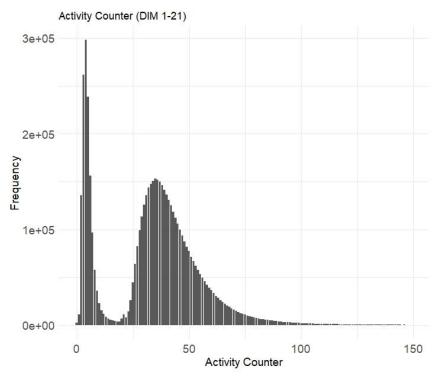


Figure 2-14 Distribution of 2-hour activity records for days in milk 1-21 across all herds from 2020-2023

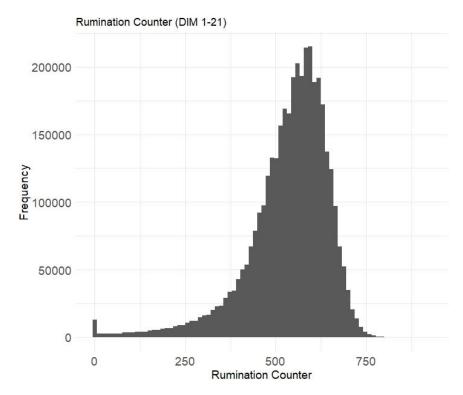


Figure 2-15 Distribution of 2-hour rumination records for days in milk 1-21 across all herds from 2020-2023

2.4 Introduction to Methods

This project explores the utility of production and behaviour data as recorded by AMS for inclusion in automated transition monitoring programs. This was achieved using two distinct approaches to statistical analysis. The goal of the first was inferential, to gain an understanding of the association between these relatively novel data sources and subsequent transition cow performance. The second was predictive, to establish the accuracy with which this data could be used to predict performance and pre-emptively identify animals likely to suffer from poor transition health.

Each of these approaches carried a differing set of priorities. In the case of the inferential studies model simplicity and ease of interpretation for stakeholders, both within the project and without, was of importance. In the development of predictive model, accuracy of prediction and potential for generalisation across a broad range of herds was considered key.

2.4.1 Inferential Analysis

The investigation of the relationship between production and behaviour data as collected via AMS, and transition performance within a large, multi-herd study has not been previously undertaken. Our goal was to quantify the association between these variables and long-term indicators of transition success. To achieve this, we conducted a series of observational, retrospective cohort studies. The structure of the data analysed within this study is hierarchical. Observations may be

clustered at several points, geographically by country or region, by cows within herds, or lactations within individual cows. Mixed-effects models are an extension of traditional regression models which provide a means though which non-independence within clusters such as these can be controlled (Brown, Prescott, 1999). It achieves this by allowing the incorporation of random effects (for example that attributable to the herd of origin) alongside the fixed effects. Furthermore, it allows for the variance observed within the model attributed to each of the random and fixed effects to be quantified, easing interpretation. Our interest lay in deciphering the influence of physiologic status of the transition cow, as captured by production and behaviour data during transition on subsequent performance. The clustering of data may be safely ignored where this is likely to have little or no effect on the outcomes of interest. However, the outcomes by which performance was judged in this thesis (for example, cull risk in early lactation) are likely to be influenced by on-farm management factors. As such, an ability to account for the influence of herd of origin was necessary. Mixed-effect models provide a well-established means to achieve this (Duffield et al., 2002). Assessment of model fit was carried out using pseudo-R² as calculated by the MuMIn package (Bartoń K, 2024). This statistic is reported as marginal pseudo-R², representing the variance attributable to the fixed effects, and conditions pseudo-R² which denotes the variance attributable to the combined fixed and random effects.

2.4.2 Predictive Modelling

The goal of the predictive modelling presented in this thesis was the development of an accurate, generalisable model capable of predicting an animal's production, fertility and survival. As such, interpretation of the model itself was of little importance. Machine learning has demonstrated an ability to outperform traditional statistics in predictive modelling (Eicker et al., 2002) and we apply a range of machine learning algorithms in Chapters 4, 6, and 7. The application of a variety of algorithms in the development stage is advocated by the No Free Lunch Theorem (Ausiello et. al., 2012). The premise of this theorem is that in the search for an optimised algorithm no one approach will outperform any other when applied across a wide range of problems. This encourages the examination of a range of algorithms for each individual problems to assess their performance. Within this thesis the utility of mars models, neural networks and support vector machines are investigated (Kuhn and Johnson, 2013). However, random forest and decision tree algorithms, which consistently outperformed these approaches, form all final models reported.

2.4.3 Decision Trees and Random Forests

Decision trees are a widely used supervised learning algorithm which have been applied for both regression, survival and classification problems (James et. al., 2013). The basic premise of this approach is to iteratively partition data from the initial root node until a stopping criterion is reached at which point decision the tree is complete. In the case of binary classification (for example predicting success or failure of

conception to first insemination), splitting is carried out with the goal of increasing homogeneity within each sub-node. Thus, splitting will lead to each subsequent node containing an increasing proportion of a specific class than the node that preceded it. This is formalised using the Gini Index, defined for a two-class problem as:

$$2(P_1 \times P_2)$$

Where P_1 and P_2 are the class probabilities for each respective class. The goal when splitting any node is to minimise the Gini index within each sub node, which can be achieved by minimise any one of the class probabilities. Tree based approaches are simple and easily interpretable however, they have a number of limitations. Of these, two primary examples are their tendency to create highly complex models and their sensitivity to changes in the data used for construction (James et. al., 2013). Models which tend toward complexity are prone to overfitting. This term is used to describe a model which performs very well on training data but cannot replicate these results when applied to previously unseen data (James et. al., 2013). Overly complex models are particularly prone to this. Techniques which reduce the complexity of trees are therefore an important factor in their construction. Designation of a stopping criteria based for example on tree depth, or minimum reduction in the Gini Index can be used to limit tree complexity. In addition to this a process of "pruning" can be applied. This serves as a form of regularization in which a complexity penalisation value is added to the Gini Index. The weight of this penalty can be adjusted to control the trade-off between a decrease in Gini index and tree complexity allowing the best sized tree for a particular problem to be identified.

The second issue surrounding the use of decision tress is related to their sensitivity to changes in the data used in their construction. Slight changes to data used to build decision tress can lead to significant changes in final decision tress size and structure. This has been addressed using decision trees as an ensemble learner as set out by (Breiman, 2001). This approach involves the construction of a large number of decision trees before utilising a majority vote across all trees to classify each observation. Each decision tree is built using a random selection of the original data to a designated stopping criteria as detailed above. This ensemble approach leverages a weakness of decision tress, their unstable nature leading to large variance in construction under different datasets, into one of the key strengths of Random Forest.

Random forests have been an extremely successful algorithm (Biau & Scornet, 2016) and have been applied across a wide range of industries. Within the dairy industry their utility in the predictions of outcomes such as milk yield and fertility has been previously demonstrated. The nextMILK model developed by Salamone et al., (2024) aims to predict first test-day milk yield exclusively using data generated in the animals' prior lactation and achieved this with a MAPE of 13%. This approach compared favourably when compared benchmarking models built using traditional lactation curve methods

such as the MilkBot model (Salamone et al., 2022). A similar method of prediction was adopted by Dallago et al., (2019) applied to predict the first test day yield in maiden heifers. Once again Random Forest performed well with no statistically significant difference between predicted and observed values when assessed using a cross validated test set. Though in this case the performance of the Random Forest model was inferior to that achieved by a neural network algorithm.

Random Forest algorithms have also been applied in the prediction of fertility outcomes, most notably by Shahinfar et al., (2014). Utilising individual animal health and production data in conjunction with herd data for over 100,000 breeding events, insemination outcomes were predicted with an AUC-ROC of 75.6 and 73.6, in primiparous and multiparous animals respectively. This represented a significant improvement in predictive performance compared with achieved by naïve bayes, bayesian networks and bootstrap aggregation. In the case of predicting the occurrence of a culling event in early lactations the use of random forest is limited, however this is likely reflective of the relative lack of reported models of any kind for this particular outcome. Random forests have however, been applied for the prediction of mortality risk in feedlot animals over the six weeks post- arrival on farm. The reported accuracy from these models was poor and no comparison with alternative model types was made within the analysis (Wisnieski et al., 2022). This limits our ability to assess the relative merits of this analytical approach for the prediction of culling events.

2.5 Model Generalisability and Deployment

Historically, the geographical distribution of AMS usage has been centred in northern Europe. This is reflective of the high cost of labour, small farm size and indoor housing systems common to this region - conditions which lends themselves to the adoption of robotic milking. However, over the past decade there has been a sustained effort by companies such a Lely to expand the use of AMS. This has seen the development of Lely's grazing system and introduction of Lely Grazeway ®, to increase uptake of AMS in areas where the maximisation of grazed grass is a priority. Likewise, the Lely XL® initiative has developed housing and management solutions to increase the efficiency of robotic milking in large herds which traditionally favoured conventional milking systems using large rotary parlours. This global expansion of AMS poses two challenges in the development of decision support models.

The first is the development of models which can be deployed on as broad a range of Lely farms as possible. Key to achieving this is limiting the number of data sources required by the model to deliver an accurate prediction. A wide range of sensors capable of monitoring dairy cow production and behaviour are available to Lely customers, however, their uptake is not universal. As a result of economic constraints or producer preference, auxiliary sensors such a neck mounted accelerometer, or novel monitoring technology may not be in use across all holdings. The development of models which rely on such

sensors will therefore limit the range of holdings on which they can be deployed and should where possible be avoided.

The second challenge is the development of generalisable models which, once deployed, perform effectively across a range of management, geographical and environmental conditions. The term generalisability is applied in machine learning to describe a model's ability to perform adequately when presented with a previously unseen dataset (Yang et al., 2022). Assessment of generalisability can be performed by examining performance when the model is applied to a dataset other than that used for model training. This serves to give some indication of how this model may perform when deployed in the real world and is a key indicator of the potential value a model can provide. As a rule of thumb, the generalisability of a simple model will exceed that of more complex model and should be pursued. Three techniques employed to improve the generalisability of the predictive models reported in this thesis are described below.

2.5.1 Feature Selection

The reduction in the number of variables incorporated with a model serves to increase the generalisability and boarded potential application. As the volume of data generated by AMS continues to increase, this is becoming a vital aspect of model development. Feature selection can be carried out using a wide range of approaches broadly categorised into filter or wrapper methods (Kuhn and Johnson ,2013). Wrapper methods assess variables concurrently, with the aim of finding the combination of predictors which maximise model performance. By comparison filter methods assess each variable individually. Only after fulfilling specific criteria are variables then added to the final model.

Within this thesis a wrapper approach, specifically recursive feature elimination is utilised in feature selection for predictive models. Recursive feature elimination is a modified backward step elimination procedure (Kuhn and Johnson ,2013). The model is first fitted using all available variables and performance assessed using a pre-specified metric (E.g. Area Under the Receiver Operator Curve). Ranking of variable importance is then carried out and the model is iteratively refitted without the least important variables. Model performance over the range of subsets analysed can then be assessed and that achieving the highest level of performance used in the final model. This serves to allow the development of a parsimonious model which increases the range of farms over which it can be deployed by reducing the number of sensors required.

2.5.2 K-fold Cross Validation

K-fold Cross Validation is a commonly applied resampling technique designed to reduce overfitting and increase generalisability (James et. al., 2013). In this approach, K represents the number of folds or partitions used to segregate the data. In the case where K is set to 10, the data is randomly broken into ten subgroups. The first is withheld while the model is built using folds 2-10 and evaluated on fold 1.

Thereafter fold 2 is withheld and the model built using groups 1 and 3-10 and evaluated on fold two. This is repeated iteratively, and the final error estimated attained by averaging the error across each fold. K-fold cross validation can be applied within the feature selection, model tuning, and model evaluation phases as required to provide an indication of expected model performance when applied to novel data.

2.5.3 External Validation

The evaluation of model performance on training data, with or without cross validation is not likely to be truly indicative of model performance when deployed in the real world. This is particularly true in the case of the predictive models investigated here, which are not only required to perform when applied to previously unseen animals but to previously unseen animals from previously unseen herds. Therefore, for the assessment of all predictive models a train and test split was applied on a herd basis to all data. The objective of this is to allow the model to be built using a random selection of herds while a small subset of herds is withheld. The model is built and assessed using cross validation before being tested on previously unseen herds. It is the performance in this test dataset by which the model's utility is then judged. This has the disadvantage of reducing the training set available for model development but the distinct advantage of providing a more realistic assessment of model performance under real world conditions (Fenlon et al., 2018).

2.6 Measuring Predictive Performance

2.6.1 Classification Models

The metrics used in the assessment of any classification model can be broadly grouped into those defining its capability for discrimination and calibration. Discrimination is based on the model's ability to correctly separate observations into their respective class, (e.g., alive at 100 days in milk vs culled by 100 days in milk). Metrics such as, accuracy, sensitivity, specificity are often used in this regard. Discriminatory metrics offers a simple assessment of model performance and are of great use in a diagnostic setting, where the outcome of the individual is paramount. However, they provide no information regards the bias of the model (Fenlon et al., 2018). Calibration metrics allow us to define the individual predicted probability of belonging to a specific class (e.g., each individual animals' probability of being culled by 100 DIM). This offers an assessment of model fit and which can be assessed visually using a calibration plots.

2.6.2 Graphical Assessment

The receiver operator curve plots a model's true positive rate against its false positive rate across all possible classification thresholds. Assessment on the curve allows us to quickly assess how the models balance the trade-off between detection of true positives while avoiding false positive. A related metric – the area under the receiver operator

curve (AUC-ROC) provides us with a single figure describing the performance of the model over all classification thresholds and allows rapid comparison of models. The AUC-ROC represents the probability that the model will rank a randomly chosen positive outcomes higher than a random chosen negative outcome. This ranges from 0-1, where a value of 1 represents perfect, and a random guess will yield an AUC-ROC of 0.5.

As the receiver operator curve offers a means of visual assessment for model discrimination, calibration plots offer an efficient visual assessment of calibration performance. This is often carried out following binning of observations into groups by predicted probability (e.g., Deciles). The mean predicted probability of the event within each group is then plotted against the proportion of actual events observed within the group. For instance, for observations binned with the 0/9-1.0 probability, we would expect a perfect model to have predicted probability of 0.95. Where this perfect calibration is repeated across all bins, a straight 45-degree line is returned.

2.7 Discussion

Within the study design for this project a number of decisions were made which prioritised data quantity and accessibility over data quality. As an industry partnered project, these decisions were motivated in large part by our focus on the development of generalisable TMP which could be broadly deployed across Lely's customers in the UK and ROI. To this end, our goals were to maximise the number of recruited herds in order to develop a dataset which represented a diverse range of farm management systems within this region. And second, to exclusively utilise data which was readily available for integration through Lely's current software. When applied to this project's inferential and predictive aims, this approach offered a number of advantages but also some limitations.

A defining feature of this project is the exclusive use of data available via Lely's third-party API. Data accessible by this API is, for the most part, automatically collected sensor data, available in real time and presented in a uniform format across all Lely herds. These traits lend themselves exceptionally well to integration of API data within automated transition cow monitoring programs. However, the scope of data available is limited. Of particular note is the absence of farm-specific environmental and management factors many of which have previously demonstrated significant association with the outcomes examined within this thesis. It stands to reason therefore, that their inclusion within this dataset has the potential to improve the performance of any models investigated. However, in line with our prioritisation of data quantity and availability, the acquisition of such information was not pursued within this project but is an area for future development and research to consider.

Information relating to environmental and management factors are not readily available on commercial herds utilising Lely AMS. Their

inclusion, therefore, would serve only to reduce the generalisability of our models within Lely's client base. In addition to this, acquiring this data was considered likely to reduce the quantity of data available for analysis. The addition of an extra barrier to participation, such as a survey relating to selected management practices, may have reduced the willingness of both FMS advisors and producers to engage with this project. Hence, similar to our decision to allow FMS advisors free choice in the farms approached for participation (Section 2.1.1), we chose to simplify the recruitment process by relying solely on API data with the goal of maximising the quantity of data available for analysis.

Despite these decisions however, the number of farms recruited was below the target of 10 herds per participating Lely Centre. This is likely reflective of the time commitment required of FMS advisors to complete the recruitment process. While every effort was made to streamline this procedure, it remained a time-consuming task requiring multiple interactions with both the producer and the research team. The limitations this placed on recruitment and hence data quantity was compounded by data access issues which became apparent within the data extraction phase, particularly those relating to the data generated by the neck mounted accelerometers.

In the pursuit of generalisable and widely deployable models, our study design placed a focus on the use of data generated by the milking robot. In contrast to the wearable technology utilised on Lely farms, which are manufactured by a third-party and can be replaced at any time, sensors within the robot offer greater consistency and uniformity of data. The loss of data relating to wearable technology which occurred over the course of this project vindicated our prioritisation of data generated by the robot. However, the utility of rumination and activity data for the assessment of physiological status cannot be overlooked and represents an intriguing point of investigation. Therefore, while its inclusion within selected models resulted in a reduction in the volume of data available for analysis, this was considered a worthy compromise.

Despite the barriers encountered in herd recruitment and data extraction, the results remain the assembly of a large, multi-herd dataset comprised of a wide range of cutting-edge sensor data from farms in 5 different countries. While any results must be viewed in light of the sample size and biases, it provides a great opportunity for the investigation of AMS data and its application within a TMP. Crucially, it is reflective of the data readily available within commercial Lely farms and therefore provides insight into the potential this data holds to make meaningful improvements to transition cow health.

Chapter 3 The Association of Production and Behaviour Data with Yield Deviation in Transition Cows on Automatic Milking Systems.

3.1 Introduction

The three-weeks pre- and post-partum (commonly referred to as the transition period) represents a hugely influential time for the health and production of the modern dairy cow. The physiological challenges associated with the initiation of lactation require significant adaptive responses by the metabolic, endocrine and immune systems (Ingvartsen et al., 2003). Failure to respond to these challenges appropriately is a major cause of dairy cow morbidity with approximately 75% of all disease occurring in the first month post-partum (LeBlanc et al., 2006)

Within the transition period, calving is a critical inflection point. In the days immediately post-partum the modern dairy cow experiences more significant endocrine changes than at any point during the lactation cycle (Grummer et al., 2004). In addition to this, cows experience a four-fold increase in calcium demand on the day of calving (Horst et al., 1997), while glucose requirements triple by day 4 post-partum (Grummer et al., 2004). Under modern management conditions, these physiological changes generally coincide with a change in housing, social group and diet, further increasing the adaptive responses required. The transition period represents a challenging time in the cow's production cycle, however, at no point is this more extreme than in the days immediately after calving.

Despite this, objective means of monitoring transition cows during this time have seen limited uptake within the commercial diary industry. Research aimed at increasing our ability to identify animals likely to experience poor transition has investigated a range of metabolic indicators and described their association with subsequent health and production. Several have been incorporated into transition cow monitoring programs designed to assess physiological status in the immediate post-partum period including the assessment of serum Beta-Hydroxy-Butyrate (BHB), non-Esterified-Fatty-Acids (NEFA), and Calcium concentration (LeBlanc et.al., 2006; Goff et.al., 2008; McArt et al., 2012). These programs seek to provide information relating to transition cow health while facilitating targeted intervention at the level of the individual. However, their application on commercial dairies remains limited (Espadamala et al., 2016; König et al., 2023). The labour-intensive nature of such monitoring programs is a likely barrier to their adoption particularly in areas when skilled agricultural labour is in short supply, such as the UK and Republic of Ireland (RADBF, 2017: Kelly et al., 2020). An alternative means of monitoring the physiological status of transition cows in the day immediately post-partum is therefore required, ideally one which can be applied with minimal labour input. The proliferation of automatic milk systems (AMS) within the dairy industry may offer means to achieve this through the collection of high frequency, cow-level production and behaviour data.

Automatic milking systems offers a wide range of benefits to producers. Key amongst these is the ability to automatically and objectively monitor the physiological status of animals within the milking herd using modern sensor technology. Production and behaviour data, harvested by the milking robot from the day of calving provides information relating to milk quality, quantity, as well the nature and frequency of cow-robot interactions. This data may provide a viable alternative to traditional transition cow monitoring programs and facilitate the automated monitoring of transition health in the immediate post-partum period.

One of the most common and economically consequential effects of disease during the transition period is a reduction in early lactation milk production (Liang et al., 2017). This has led to its utilisation as a proxy measure for transition health (Nordlund, 2006; Lukas et al., 2015; Caixeta, 2021). Commercially, this has been implemented as the Transition Cow Index® (TCI), (https://agsource.com/fresh-cow-summary/) which aims to utilise the disparity between expected and observed first test-day milk yield to assess the effectiveness of a given transition cow program and improve long-term profitability. Deviation from expected yield in early lactation has a demonstrated association with transition cow disease and metabolic status. (Nordlund, 2006; Salamone et al., 2024). While this has been proven to be a useful metric for the assessment of transition cow health, its association with AMS metrics assessed in the days immediately post-partum has not been investigated.

The objective of this study was to test the hypothesis that AMS production and behaviour data from days 1-3 post-partum is significantly associated with subsequent transition cow health assessed using yield deviation over the first 30 days in milk. This investigation serves to further our understanding of the relationship between AMS data collected in the immediate post-partum period and animal performance over the ensuing month of lactation. Furthermore, it may provide direction as to the utility of these variables for incorporation into an automated transition cow monitoring program.

3.2 Materials and Methods

3.2.1 Study Population

A convenience sample of 46 herds was recruited on a voluntary basis from the UK and Republic of Ireland as described in Chapter 2. Criteria for inclusion was the use of Lely Astronaut milking robots (Lely International, N.V.) under free flow traffic conditions (Munksgaard et al., 2011), in conjunction with rumination and physical activity monitoring technology (Lely Qwes-HR collars, Lely International N.V.). Year-round and seasonal calving patterns were represented within the dataset. At

the time of analysis complete or partial production records from 33,813 lactation across 39 herds spanning the years 2016 to 2023 were available.

Data Analysis

All data analysis was conducted using R statistical software (R Core Team, 2021). A three-step data analysis procedure was undertaken. In Step 1 the expected cumulative yield for days 1-30 DIM (ECY) for each cow-lactation was established using a mixed-effects model. In Step 2, ECY was compared with the observed yield and a Yield Deviation established. This served to capture each animal's cumulative yield performance over the first 30 days relative to expected. In Step 3, a multivariable mixed-effect model was built using production and behaviour data recorded over the first 3 days post-partum. Our aim was to assess the relationships between data collected in the immediate post-partum period and subsequent Yield Deviation.

3.2.2 Step 1: Modelling of Expected Yield

To quantify YD, it was necessary to establish an expected 30-day cumulative milk yield. For each multiparous lactation six independent variables were utilised to model ECY: Herd ID, parity, year of calving, season of calving, previous production, and herd production. Categorical variables were created for Herd ID, parity; (2 and 3+), year of calving and season of calving; winter (December, January, February), spring (March, April, May), summer (June, July, August), autumn (September, October, November). Milk vield utilised in the previous production and herd production variables were assessed over 1-30 DIM where the day of calving was designated day zero. Animals for which complete production records for days 1-30 in milk could not be established were removed. Previous production, which sought to quantify each animal's level of production prior to the lactation under analysis was then calculated using the animals' mean 30-day cumulative yield from all available lactations prior to the lactation in question. Lactations for which prior production could not be calculated were removed. The Herd Production variable, which sought to quantify the within-herd production level at the time of calving was calculated as the mean 30-day cumulative yield for all cows calved within the same herd, year, and season as the lactation in question. Following data cleaning and feature engineering an approach similar to that utilised by Nordlund (2006) and Lukas (2015), was used to model ECY. This approach models expected yield on a per cow-lactation basis utilising herd and animal-level data and is detailed below.

The outcome of interest in Step 1, ECY, represented the total expected milk production over the first 30 days of lactation under a given set of individuals, herd and environmental conditions.

A mixed-effects linear regression model was built within the lme4 package (Bates et al., 2015) and took the form;

$$Y_{ij} = \mu + \beta_1 X_{1ij} + \beta_2 X_{2ij} + \beta_3 X_{3ij} + \beta_4 X_{4ij} + \beta_5 X_{5j} + v_j + \epsilon_{ij},$$

$$[v_j] \sim N(0, \Omega_v)$$

$$[\epsilon_{ii}] \sim N(0, \Omega_e)$$

Where subscript i and j denote the ith cow-lactation in the jth herd; Y represents the response variable of Expected Cumulative 30-day Yield (Kilograms); μ represents the intercept value; β_1 to β_4 represent the coefficients for cow-lactation fixed effects X_{1ij} to X_{4ij} , these being Year of Calving, Season of Calving, Parity, and Previous Production; β_5 represents the coefficient for the herd-level fixed effects X_{5j} of Herd Production; v_j represents a random effect for Herd (assumed to have a normal distribution of mean = 0, and variance = Ω_v) and ε_{ij} represents the residual model error (assumed to have a normal distribution of mean = 0, and variance = Ω_e). Residuals were examined graphically to assess their distribution.

3.2.3 Step 2. Assessment of 30-Day Yield Deviation

Expected 30-Day Cumulative Yield, estimated in Step 1, was used in Step 2 to quantify YD for each multiparous cow-lactation. This was calculated as the difference between the observed 30-day cumulative yield and ECY and expressed as a percentage of ECY. Therefore, similar to the TCI, YD represents the degree to which each animal under or over-produced relative to expected. However, in contrast to the use of a single monthly test-day yield, we chose to account for each animal's cumulative milk yield over the first 30 days of lactation and thus reduce the inaccuracies associated with a single measurement.

3.2.4 Step 3: Inferential modelling of Yield Deviation and Early Post-partum Variables

For each cow-lactation analysed the day of calving was designated day zero and data from days 1-3 used to engineer independent variables. Milk quantity was assessed as Mean Milk Yield (kg); the mean of daily milk yields over days 1-3, and Mean Yield Acceleration; the mean of the change in daily milk yield from days 1–2, and 2–3. Milk quality was assessed using milk conductivity and constituent data. Conductivity was assessed as Mean Conductivity: the mean udder-level conductivity of milk recorded over days 1-3 and, Conductivity Alert; the total number of instances where quarter-level conductivity exceeded 80 units. Maximum Temperature (Degree Celsius); the maximum mean daily milk temperature as averaged across all successful milking visits within a given day. Milk Fat and Protein indications, as recorded once daily by Lely's MQC-C ® (Fadul-Pacheco et al., 2018) were utilised as Mean Fat and Mean Protein; the mean of recorded values across days 1-3 for their respective constituents. A Mean Fat-to-Protein ratio (FPR) was subsequently calculated. Mean Concentrate Dispensed; the mean grams of concentrate feed dispensed by the robot to each cow per day. Visit behaviour was assessed using milking visits and milking refusals (where milking permission is denied due to an animal re-presenting a

short time after a previous milking visit). These were averaged over days 1-3 and reported as Mean Milkings and Mean Refusals. From amongst the variables a selection of interaction terms were investigated based on biological plausibility to influence Yield Deviation including Mean Milk Yield with Mean Concentrate Dispensed, Yield Acceleration, Protein, Milkings and Refusals. The interaction between Mean Milkings and Mean Yield Acceleration was also examined. Animals which failed to record milk quantity and quality data for at least one robot visit for each of the days 1-3 post-partum were removed. Finally, herds with less than 100 lactations were removed. Retained cow lactations which had a corresponding Yield Deviation from Step 2 formed the final dataset brought forward for analysis.

To explore the relationship between early lactation AMS data and Yield Deviation, a mixed-effect linear model was constructed with herd as a random effect using the Ime4 package (Bates et al., 2015). Prior to modelling, non-linearity was assessed using multivariable regressive splines (Friedman, 1991). Correlation between independent variables was assessed using a Pearson's correlation matrix. Decisions relating to deletion or retention of highly correlated variables made based on the theoretical importance of each variable. Both Mean Fat, which demonstrated a high correlation (r >0.75) with Mean FPR, and Mean Conductivity, which demonstrated high correlation with Conductivity Alert were removed to avoid collinearity (Dohoo et al., 2003). Mean FPR was retained as it provides a built in assessment of milk fat in addition to milk protein, while conductivity alert was chosen over mean conductivity as this represents a more practical and widely utilised indicator of udder health on farms utilising Lely robots. Mean Refusals, and Conductivity Alert which demonstrated a highly right-skewed distribution was transformed using a cubic transformation. All variables excluding Parity were centred by subtracting the column means of each variable from their corresponding columns and scaled by dividing each variable by their standard deviations (van den Berg et al., 2006).

A multivariable mixed-effect model was constructed using a manual backward step procedure (Dohoo et al., 2003). All candidate variables were screened using univariable analysis and brought forward for inclusion in a multivariable model where a P-value of ≤ 0.20 was observed. A random intercept to account for the clustering of data at herd-level was included (Dohoo et al., 2003). Variables were retained in the final model where a *P*-value of ≤0.05 was observed. Variables which formed a significant interaction term were forced into the final model. On completion of the backward step, all independent variables removed were re-entered into the final model to test for significance. The inclusion of Cow as a random effect nested within Herd was also evaluated, as was the possible confounding factor of Calving Season (Year-Round or Seasonal). As their inclusion led to a negligible change in coefficient estimates (<5%) and resulted in no change in parameter significance, these were excluded for model parsimony. Parity (2, 3+) was evaluated and retained as a confounder within the final multivariable model. Goodness-of-fit was evaluated by the graphical

assessment of residuals and the assessment of marginal and conditional R² values (Nakagawa et al., 2017).

3.3 Results

Step 1

Complete or partial production records from 33,813 lactation across 39 herds were available for analysis. Due to the sparsity of data in the years prior to 2018 all cow-lactations starting prior to this point or, for which production records were found to be incomplete were removed (n =9,712). All those for which prior production could not be established were removed (n= 11,507) including all first lactations animals (n =6,799). Thereafter, herds with less than 100 lactations available for analysis were removed (8 herds containing 427 lactations). In total, 12,295 lactations from 30 herds remained for analysis. Coefficients of the final linear mixed model used to estimate ECY are presented in Table 3-1. Residuals were examined graphically and found to be normally distributed (Figure 3-1). The mean absolute error and mean absolute percentage error were 138 kilograms and 12% respectively.

Table 3-1 Linear mixed model for Expected Cumulative 30-day Yield for 12,295 cow-lactations from 30 herds between 2017 and 2023 as described in Step 1

| Intercept 1118 1005–1127 Herd Production¹ 96.9 90–104 Previous Production² 97.7 92.7–103 Autumn Reference - Winter 4.6 -6–15 Spring 8.6 -2–20 Summer 2.6 -9–14 2017 Reference - 2018 -23.0 -77–32 |
|---|
| Previous Production² 97.7 92.7–103 Autumn Reference - Winter 4.6 -6–15 Spring 8.6 -2–20 Summer 2.6 -9–14 2017 Reference - 2018 -23.0 -77–32 |
| Autumn Reference - Winter 4.6 -6–15 Spring 8.6 -2–20 Summer 2.6 -9–14 2017 Reference - 2018 -23.0 -77–32 |
| Winter4.6-6-15Spring8.6-2-20Summer2.6-9-142017Reference-2018-23.0-77-32 |
| Spring 8.6 -2–20 Summer 2.6 -9–14 2017 Reference - 2018 -23.0 -77–32 |
| Summer 2.6 -9-14 2017 Reference - 2018 -23.0 -77-32 |
| 2017 Reference - 2018 -23.0 -77_32 |
| 2018 -23.0 -77_32 |
| |
| 0040 |
| 2019 32.9 -16–82 |
| 2020 34.5 -14_83 |
| 2021 41.6 -7–90 |
| 2022 45.1 -3_94 |
| 2023 38.0 -11_88 |
| Parity 2 Reference - |
| Parity 3+ -42.0 -5232 |
| Random Effects Variance Std. Dev |
| Herd ID (Intercept) 3775 61.4 |
| Residual 36129 190 |

¹The mean 30-day cumulative yield for all animals within the herd of origin, in the year and season for a given lactation. ² The mean 30-day cumulative yield for all available lactations prior to the lactation in question. CI = confidence interval

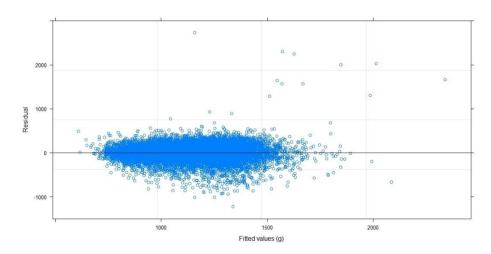


Figure 3-1 Scatter Plot of residuals plotted against fitted values for modelling of expected yield for Expected Yield Model in Step 1

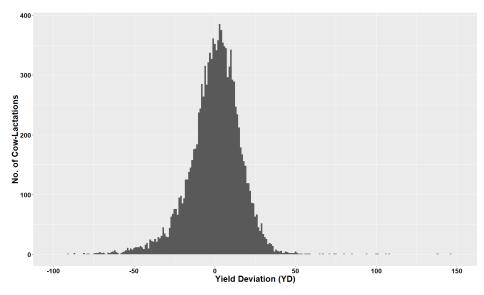


Figure 3-2 Histogram of Yield Deviation, the difference between cumulative expected and observed yield at DIM 30, as calculated in Step 2.

STEP 2:

Across the entire dataset the range of YD was -91% to 146% with a mean of 0.0% and median of 1.2% (Figure 3-2). Mean YD ranged from -1.0% to 0.4% on a herd basis.

Step 3

Incomplete milk quantity and quality records led to the removal of 12,029 cow-lactations. Following data cleaning AMS data relating to 15,532 cow-lactations remained, of which 7,417 from 27 herds had a corresponding Yield Deviation available from Step 1. These were brought forward for inferential modelling. The mean Yield Deviation for the final dataset was 0.1% ranging from -81 to 108% on a cow-lactation basis. Descriptive statistics for the herds included in the final model are presented in Table 3-2. The average number of cow-lactations per herd was 277 ranging from 80 to 794. Thirty-one percentage of all cow-lactations were classed as parity 2 with those remaining classed as parity 3+. The median and interquartile range for all independent variables examined are presented in Table 3-3

Following determination of the final model, residuals were examined and found to follow an approximately normal distribution. Yield Deviation was modelled with a mean absolute error of 8.13 and root mean square error of 11.10. Coefficient of determination was established as a marginal R² of 47% and condition R² of 60%. Results of the final multivariable model are presented in Table 3-4, Figure 3-3 and Figure 3-4. Having accounted for clustering of data at the herd-level and the confounding effect of parity, significant (*P*- value <0.5) positive association with Yield Deviations were observed for the variables Mean Milk Yield, Yield Acceleration, Refusal and Milk Protein Percentage with co-efficient of 13.3, 4.44, 1.2, and 1.29 respectively. Mean Milking and

Fat-to-protein Ration had a negative association with coefficients of - 0.63 and -0.70 respectively.

Table 3-2 Descriptive statistics for herds included in the final multivariable model in Step $\bf 3$

| Variable | No. [range] |
|---|---------------------------|
| No. of Farms | 27 |
| Mean No. of Cow-Lactations/Herd | 274 [80–794] |
| Mean Cow-Lactation Yield Deviation | 0.10 [-81–108] |
| Mean Herd Yield Deviation | 0.15 [-2.8–2.5] |
| Mean Milk Production/Cow/Year (Kg) ¹ | 10,280 [6,668–15,202] |
| Calving Pattern | % Of Dataset (Herd Level) |
| All Year Round | 70 |
| Seasonal | 30 |

¹ Calculated using mean figures from the years 2020 and 2021.

Table 3-3 Descriptive statistics for independent variables examined in Step 3.

| Variable | Median | IQR |
|--------------------------------|--------------|------------|
| Mean Milk Yield (kg) | 23 | 9 |
| Mean Yield Acceleration (%) | 16 | 13 |
| Mean Concentrate Dispensed (g) | 3343 | 893 |
| Mean Refusals | 1.7 | 1.3 |
| Mean Milkings | 2.6 | 1 |
| Mean Protein | 4.84 | 0.55 |
| Mean Fat | 4.87 | 1.34 |
| Mean Fat-to-Protein Ratio | 1.02 | .3 |
| Mean Conductivity | 73.25 | 4.5 |
| Conductivity Alert | 0 | 2 |
| Milk Temperature (Degrees C) | 38.2 | 1.1 |
| | Percentage c | of Dataset |
| Temp >40 | 1% | |
| Parity 2 | 31% | |
| Parity 3+ | 69% | |

Table 3-4 Linear mixed model for 30-day Yield Deviation for 7,417 cowlactations from 30 herds between 2017 and 2023 as described in Step 3

| Variable | Estimate | 95% CI | <i>P</i> -value |
|----------------------------|-----------|-----------|-----------------|
| Intercept | 3.58 | 0.9_6.18 | < 0.001 |
| Mean Milk Yield | 13.3 | 12.9_13.7 | < 0.001 |
| Mean Yield Acceleration | 4.44 | 4.1_4.7 | < 0.001 |
| Mean Refusals | 1.20 | 0.8_1.5 | < 0.001 |
| Mean Milkings | -0.63 | -1.00.2 | < 0.001 |
| Mean Protein | 1.29 | 0.9_1.6 | < 0.001 |
| Mean Fat-to-Protein Ratio | -0.70 | -1.00.4 | < 0.001 |
| Parity 2 | Reference | - | - |
| Parity 3+ | -3.26 | -3.82.6 | < 0.001 |
| Mean Yield X Mean Yield | | | |
| Acceleration | 1.23 | 0.9_1.4 | < 0.001 |
| Mean Yield X Mean Milkings | -1.10 | -1.40.8 | < 0.001 |
| Mean Yield X Mean Refusals | 1.04 | 0.7_1.3 | < 0.001 |

The interaction term Mean Milk Yield X Mean Yield Acceleration had a positive association with YD. Within this term an initial negative association between Mean Milk Yield and YD was apparent when recorded in conjunction with highly negative Yield Acceleration. This relationship became increasingly positive as yield acceleration increased (Figure 3-3). Mean Milk Yield X Mean Refusals record a positive association and demonstrated a consistent relationship with YD throughout their ranges. Finally, Mean Milk Yield X Mean Milkings demonstrated a negative association with YD. Within this term a strongly positive relationship between Mean Milk Yield and YD was observed for lower values of Mean Milkings, becoming progressively weaker as the number of milkings increased (Figure 3-4)

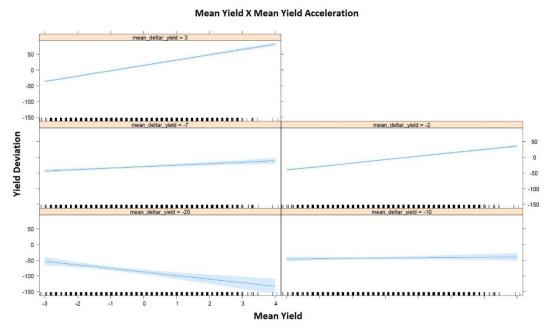


Figure 3-3 Effects plot for the interaction term Mean Milk Yield X Yield Acceleration assessed overs days 1-3 post-partum as retained in the final multivariable model for Yield Deviation at DIM 30.

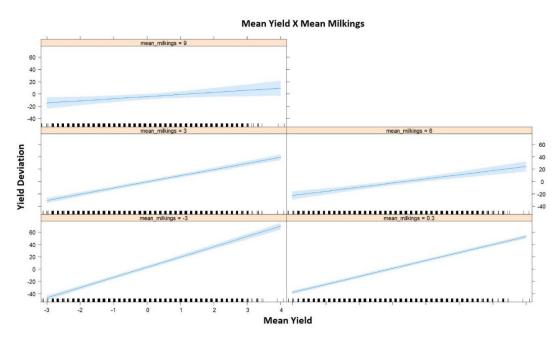


Figure 3-4 Effects plot for interaction term Mean Milk Yield X Mean Milkings assessed overs days 1-3 post-partum as retained in the final multivariable model for Yield Deviation at DIM 30.

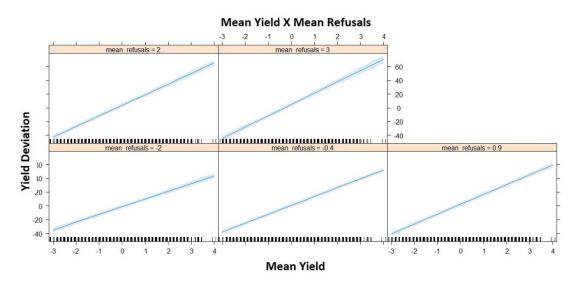


Figure 3-5 Effects plot for interaction term Mean Milk Yield X Mean Refusals assessed overs days 1-3 post-partum as retained in the final multivariable model for Yield Deviation at DIM 30.

3.4 Discussion

Production and behaviour data collected by AMS in days 1-3 postpartum demonstrated statistically significant association with transition cow health as assessed by Yield Deviation over the first 30 days in milk. The significant statistical associations and substantial coefficient of determination observed within our final multivariable model emphasise the critical nature of the immediate post-partum period for transition success. Furthermore, it highlights the potential this data may hold for the development of an automated transition cow monitoring program.

The positive association between yield parameters retained in the final model and subsequent YD highlight the importance of the transition cow's ability to successfully initiate and accelerate milk production in the days immediately post-partum. While dairy cows at this point in lactation are not producing milk at levels near their peak potential, they are, in relative terms, increasing their production level at an extremely high rate (Ingvartsen et al., 2003). Prior research examining the characteristics of yield acceleration post-partum has highlighted the physiological stress which this rapid rate of acceleration places on the transition cow, with several authors proposing a potential link between increased milk yield acceleration and increased risk of transition disease (Ingvartsen et al., 2003; Hansen et al., 2006). This hypothesis is based on the physiological challenges posed by milk production as well as the correlation between the increased rate of production and incidence of clinical disease seen in the post-partum period. However, it has yet to be adequately investigated (Ingvartsen et al., 2003). While clinical disease was not directly assessed within our study, a larger milk yield and a higher yield acceleration were associated with a positive yield deviation in early lactation, an outcome indicative of good health (Nordlund, 2006). Rather than being at increased risk of poor transition health, our results may indicate that animals capable of producing higher levels of milk and supporting higher rates of acceleration

represent a group which have adapted well to the stressors of calving and thus are positioned to fulfil their production potential over the first month of lactation.

The variance in relationship between Mean Milk Yield and YD, demonstrated within the Mean Milk Yield X Yield Acceleration interaction term is notable (Figure 3-3). The negative relationship between Mean Milk Yield and YD when observed in conjunction with highly negative Yield Acceleration may be due to animals registering a large volume of milk on the first day post-partum, followed by a precipitous decline. Such yield patterns would be in keeping with the development of disease and may explain the tendency for these animals to under-perform relative to expected over the first month of lactation despite recording a relatively high yield over the first 3 days.

Examination of the relationship between milk production and health in early lactation is complicated by the biological prioritisation of milk production to provide nutrition for the calf (Bruckmaier & Gross, 2017). This can lead to the continued production of milk in the face of disease and negative energy balance (Rajala & Gröhn, 1998). Considering this, further validation of these results while incorporating clinical disease data would certainly be worthwhile. However, our findings do indicate that both yield and yield acceleration parameters, assessed over days 1-3 post-partum, have potential as a means of monitoring transition cow health in the immediate post-partum period.

Milk constituents have been previously demonstrated to reflect the energy balance of early lactation dairy cows (Friggens et al., 2007; Gross & Bruckmaier, 2019). Furthermore, indications of negative energy balance, such as elevated FPR and reduced milk protein percentage have been associated with reduced milk yield and the occurrence of transition disease (Heuer et al., 1999; Kaufman et al., 2018). However, these studies have typically been carried out using monthly test-day milk sampling in weeks 2-3 post-partum. While the utility of milk constituent sampling at day 7 in milk for the assessment of energy status has been previously demonstrated (Toni et al., 2011), the degree to which this association translates to the days 1-3 post-partum remains unclear.

Within our study, both increased protein percentage and decreased fatto-protein ratio, traits generally linked with a more positive energy balance (de Vries & Veerkamp, 2000), resulted in an increase in Yield Deviation, patterns which broadly agree with those previously demonstrated using monthly test-day milk samples. This consistency of relationship between milk constituents and early lactation performance demonstrates the potential value for AMS milk quality data to provide to transition cow monitoring programs in the day immediately following calving.

The use of in-line sensors to monitoring fat and protein indications within AMS remains a relatively novel application. When compared with monthly test-day regimes this analysis has demonstrated only moderate

correlation (Fadul-Pacheco et al., 2018). Though this may not affect the utility of these sensors, the requirement for producers to calibrate their sensors limits their interpretability. Within our study no information relating to the frequency or accuracy with which sensors were calibrated was available. Hence, while our results are encouraging for the potential use of in-line fat and protein indications, extrapolation of these results beyond the dataset investigated here should be done with caution.

Interestingly, a negative association between Mean Milkings and YD was observed within this dataset. This is an unexpected, and counter intuitive finding. Reduced milking visits have been reported in association with the diagnosis of lameness (Bach et al., 2007), mastitis, and left displaced abomasum (King et al., 2018). We would therefore expect those animals recording a higher number of milking visits to represent a healthier cohort and thus deliver a more positive YD. compared with those recording a lower number of visits. In addition to this, the positive effect of increased milking frequency in early lactation on subsequent yield has been well described (McNamara et al., 2008). The interaction between Mean Milk Yield and Mean Milkings add further detail to this association as the positive effect of increased yield on YD weakens as the number of milk visits increases (Figure 3-4). A clear explanation for these findings is not apparent. However, it is worth considering the confounding effect of management on robot visits. Within the first 3 days post-partum the practice of manually fetching fresh cows to the robot is common and may artificially alter the number of robot visits recorded per cow. Within our dataset, no information was available as to the frequency of manual fetching practiced at farm level and so this finding should be interpreted with caution.

Prior investigation of the herd-level relationship between refusals and milk production have reported a negative association (Tremblay et al., 2016; Siewert et al., 2018). However, as these were assessed using cows from all stages of their lactation these are likely skewed by the prevalence of late lactations animals. As highlighted by Beck, (2014), refusals are not created equally. Those recorded by animals in the initial and peak phases of lactation may demonstrate a positive association with production parameters. The opposite may be observed for animals in late lactation. Within free-flow AMS systems refusals are, in moderation, desirable as they demonstrate the cow's appetite for the concentrate supplied by the robot, her ability to access the robot area and the availability of the robot itself. As our study investigated refusals in days 1-3 in milk exclusively, the positive associations reported here is in keeping with expectations. Notably, within the interaction term Mean Milk Yield X Mean Refusals (Figure 3-5), is the positive association of increasing yield was held across all levels of Refusals. This finding should be interpreted in light of the relatively low number of refusals recorded within this dataset, a median of 1.7 and IQR of 1.3 (Table 3-3.). While refusals in early lactation animals are desirable, excessive refusal may be expected to result in reduced cow performance (Tremblay et al., 2016), though this level of elevated refusal does not

appear to have been reached within this dataset. In contrast to milking visits, refusals represent truly voluntary visits and serve to provide an indication of an animal's desire to access the robot which is less vulnerable to the effects of management practices. As a single metric, reflective of physiological, environmental and management conditions, the significant associations with Yield Deviation demonstrated here highlight its potential for use in transition cow monitoring.

Within the conditional R² of 60% reported for the final model, the fixed effects explained 47% of the observed variance while the random effect of herd explained a further 13%. This suggests that our model encompassed a substantial proportion of the variance seen within this dataset. The relatively small effect of herd suggests that herd specific environmental and management factors play an important but limited role in determining Yield Deviation when compared with the fixed effects investigated.

Considering the narrow window over which AMS data was assessed (DIM 1-3), the degree to which it can explain subsequent Yield Deviation is noteworthy. This emphasises the critical importance of transition cow health in the immediate post-partum period and highlights the value data collected during this time may provide to transition cow monitoring programs. The success with which each cow navigates the challenges associated with the initiation of lactation will largely determine the success of the ensuing lactation (Drackley, 1999). Where the extent of this success or failure can be established by day 3 post-partum there exists an opportunity to implement management strategies aimed at mitigating potential losses. Such a system would represent a valuable tool for the management of transition health. The production and behaviour data investigated here, have demonstrated their potential utility within TMPs, not only though the strength of its relationship with Yield Deviation, but the means by which this data is collected and made available

This is the first study to report the association between data collected by AMS and subsequent yield deviation. In contrast to previously reported, labour intensive methods of transition cow monitoring all variables assessed in this study are collected automatically from the day of calving and available for integration into a TMP in real time. This represents an intriguing prospect the development and deployment of automated transition cow monitoring programs within the Lely client base.

An inherent limitation of the analysis described within this study is the use of an expected cumulative yield as a benchmark of animal performance. The advantages of Yield Deviation as employed here are clear (ref) however, it remains reliant on the accuracy of expected cumulative production as initially modelled though the ECY model. Deviation from expected yield may therefore represent true deviations from expected performance, inaccurate initial modelling of expected performance or a combination of both. Caution must therefore be

applied to the interpretation of the biological variables demonstrating statistical association with Yield Deviation as differentiation between these sources of variation is not possible within our analysis.

A further limitation of our analysis is the reliance on prior production data for the calculation of an expected cumulative 30-day yield and subsequent Yield Deviation. This forced the exclusion of primiparous animals from our analysis. The challenges of transition in primiparous animals are somewhat unique when compared with multiparous. Having not yet reached mature weight, heifers experience additional energy requirements associated with growth and carry an increased risk of dystocia (Duffield et al., 2009). However, they also suffer important clinical diseases such as hypocalcaemia at a significantly lower rate than older animals. A range of physiological assessments have demonstrated conflicting associations with subsequent performance in primiparous and multiparous animals (Ospina et al., 2010; Burfeind et al., 2014; Goff et al., 2020). The investigation of the association between AMS data and subsequent yield deviation within primiparous animals would therefore be of great interest. Modelling expected first test-day yield in maiden heifers has been investigated using a range of herd and cow level parameters available at calving such as body weight and the occurrence of twinning (Dallago et al., 2019). This approach may allow the establishment of YD in primiparous animals and allow examination of its association with AMS data in the future.

3.5 Conclusions

Our results indicate that production and behaviour data collected by AMS over DIM 1-3 have significant association with Yield Deviations over the first month of lactation. The production parameters, Mean Milk Yield and Mean Yield Acceleration both recorded a positive association with YD as did Milk Protein Percentage and Mean Refusals. Both Milk fat-to-protein ratio and Mean Milkings recorded a negative association with a Yield Deviation. The final multivariable model accounted for a substantial proportion of the variance in observed Yield Deviation, with the fixed effects accounting for 47% and the random effect of herds a further 13%. This highlights the importance of the calving period in successful transition and the potential utility of data collected during this time for the assessment of transition health. The demonstrated relationship between automatically collected sensor data and Yield Deviation may offer a means by which dairy producers utilising AMS can pre-emptively identify animals likely to experience poor transition health. Of particular interest, where the development of an automated TMP is concerned, is the investigation of the predictive power which AMS data may hold for Yield Deviation.

Chapter 4 Predictive Modelling of Deviation from Expected Milk Yield in Transition Cows on Automatic Milking Systems

4.1 Introduction

The transition period is a pivotal time in the production cycle of the dairy cow. It is estimated that between 30 to 50% of all cows experience metabolic or infectious disease during this time (LeBlanc, 2010). Moreover, those affected by disease often demonstrate reduced production, fertility, and survival for the remainder of the lactation when compared with healthy herd mates (Carvalho et al., 2019). Despite recent advances in the field of transition cow management (Mezzetti et al., 2021), preserving the health and production potential of dairy cows during this period remains one of the largest challenges facing the industry (Redfern et al., 2021).

The assessment of yield deviation, generally defined as the disparity between expected and observed yield, is an objective means of assessing transition success. To date, its most notable application has been in the development of the Transition Cow Index® (https://agsource.com/fresh-cow-summary/). This transition cow monitoring tool utilises the disparity between expected and observed first test-day milk yield to assess transition performance (Nordlund, 2006; Schultz et al., 2016). A key advantage of this approach is the range of clinical and sub-clinical disease which may be reflected in yield deviation (Nordlund, 2006). However, the TCI is limited by its retrospective nature. While it provides an objective assessment of transition cow health, it fails to provide an opportunity to manage animals likely to suffer a negative deviation in a proactive manner.

A model capable of predicting deviation from expected production may allow producers to prevent or mitigate the impact of poor transition through early intervention (LeBlanc, 2010). Applied as part of a prognostic transition cow monitoring program (TMP) such a model may allow for the classification of animals into groups based on their predicted yield deviation. Resources such as increased monitoring or diagnostic testing could then be preferentially allocated to those groups predicted to suffer negative deviations (Guterbock, 2004; Jensen et al., 2018).

Data routinely collected by automatic milking systems (AMS) present a unique opportunity to develop such a model. A range of production metrics recorded by AMS are increasingly available in conjunction with behavioural data recorded by wearable sensors. This represents a powerful combination of data capable of providing detailed physiological information at the cow level. The ability of AMS software to integrate this data and provide producers with decision support tools relating to cow management has been previously demonstrated (Wetering, 2019).

To date however, the utility of these metrics for the prediction of deviation in early lactation milk yield has not been reported.

The objective of this study was first, to explore the accuracy with which production and behaviour data collected on AMS from 1–3 days in milk (DIM) can predict deviation from expected 30-day cumulative milk yield in multiparous cows. And second, to assess the accuracy with which predicted Yield Deviations could be used to classify cows into groups which may facilitate improved transition management.

4.2 Materials and Methods

4.2.1 Study Population

A convenience sample of 46 commercial dairy farms was recruited on a voluntary basis from the United Kingdom and Republic of Ireland, as described in Chapter 2. Briefly, criteria for inclusion was the use of Lely Astronaut milking robots (Lely International, N.V.) under free flow traffic conditions (Munksgaard et al., 2011), in conjunction with rumination and physical activity monitoring technology (Lely Qwes-HR collars, Lely International N.V.). Year-round and seasonal calving patterns were represented within the dataset (Table 4-1).

Following the provision of written consent by participating herd owners, AMS data were accessed via Lely's third-party application programming interface. Records describing the frequency of robot visits as well as the weight, conductivity, and temperature of milk harvested were available from January 2016 to January 2022 for a total of 22,301 lactations. This data formed the Production Dataset (PDS). Due to a shorter duration of data retention applied to rumination and physical activity records, these were available from October 2020 to January 2022 and formed the Behaviour Dataset (BDS). This represents a subset of the PDS and totalled 5,961 lactations.

Table 4-1 Descriptive statistics for recruited herds

| Variable | No. [range] |
|---|---------------------------|
| No. of Farm | 31 |
| Mean No. of Milking cows per farm ¹ | 148 [31–335] |
| Mean No. of AMS units per farm ¹ | 3 [2–8] |
| Mean Milk Production/Cow/Year (Kg) ¹ | 10,280 [6,668–15,202] |
| Calving Pattern | % of Dataset (Herd Level) |
| All Year Round | 58 |
| Seasonal | 42 |
| Geographical Region | |
| England | 52 |
| Republic of Ireland | 22 |
| Northern Ireland | 16 |
| Wales | 10 |

¹ Calculated using mean figures from the years 2020 and 2021.

4.2.2 Data Analysis

To assess the accuracy with which deviation from expected cumulative 30-day yield could be predicted at day 3 of lactation a three-step analytic procedure was conducted. In Step 1, a mixed-effect linear regression model was used to calculate an expected 30-day cumulative yield (ECY) for each multiparous cow-lactation in the PDS. In Step 2, 30-Day Yield Deviation (YD) was calculated as the difference between the observed and expected 30-day cumulative yield. In Step 3, production, rumination, and physical activity data from 1–3 DIM was used to predict YD using machine learning models. These steps are described in detail below. All analysis was conducted using R statistical software (R Core Team, 2021).

Step 1: Modelling Expected 30-Day Cumulative Yield

To quantify YD, it was necessary to establish an expected 30-day cumulative milk yield. For each multiparous lactation six independent variables were utilised to model ECY: Herd ID, parity, year of calving, season of calving, previous production, and herd production. Categorical variables were created for Herd ID, parity; (2 and 3+), year of calving and season of calving; winter (December, January, February), spring (March, April, May), summer (June, July, August), autumn (September, October, November). Milk yield utilised in the previous production and herd production variables was assessed over 1-30 DIM where the day of calving was designated day 0. Where over 10% (3 days) of daily milk records were absent, the entire lactation was removed (n= 5,054) Where under 10% of yield data was absent, daily yield was imputed using the Predicted Mean Matching method of the MICE package (van Buuren & Groothuis-Oudshoorn, 2011). A 30-day cumulative yield was calculated by summing daily yield over 1-30 DIM. Previous production, which sought to quantify each animal's level of production prior to the lactation under analysis was then calculated

using the animals' mean 30-day cumulative yield from all available lactations prior to the lactation in question. Lactations for which prior production could not be calculated, including all first lactation animals, were removed (n = 8,646). The herd production variable, which sought to quantify the within herd production level at the time of calving was calculated as the mean 30-day cumulative yield for all cows calved within the same herd, year, and season as the lactation in question.

Following data cleaning and feature engineering a modelling approach similar to that utilised in Section 3.2 was used to model ECY. This approach models expected yield on a per cow-lactation basis utilising herd and animal-level data and is detailed below. The outcome of interest in Step 1, ECY, represented the total expected milk production over the first 30 days of lactation under a given set of individual, herd, and environmental circumstances.

A mixed-effects linear regression model was built within the lme4 package (Bates et al., 2015) and took the form;

$$\begin{split} Y_{ij} = \ \mu + \beta_1 X_{1ij} + \ \beta_2 X_{2ij} \ + \ \beta_3 X_{3ij} + \beta_4 X_{4ij} \ + \ \beta_5 X_{5j} \ + v_j + \ \epsilon_{ij}, \\ [v_j] \sim \ N(0, \Omega_v) \\ [\epsilon_{ij}] \sim \ N(0, \Omega_e) \end{split}$$

Where subscript i and j denote the ith cow-lactation in the jth herd; Y represents the response variable of Expected Cumulative 30-day Yield (Kilograms); μ represents the intercept value; β_1 to β_4 represent the coefficients for cow-lactation fixed effects X_{1ij} to X_{4ij} , these being Year of Calving, Season of Calving, Parity, and Previous Production; β_5 represents the coefficient for the herd-level fixed effects X_{5j} of Herd Production; v_j represents a random effect for Herd (assumed to have a normal distribution of mean = 0, and variance = Ω_v) and ϵ_{ij} represents the residual model error (assumed to have a normal distribution of mean = 0, and variance = Ω_e).

Step 2: Assessment of 30-Day Yield Deviation

For each cow- lactation the difference between the observed 30-day cumulative yield and ECY calculated is Step 1 was expressed as a percentage of ECY and labelled Yield Deviations (YD). Therefore, YD represents the degree to which each animal under or over-produced relative to expected cumulative milk yield over the first 30 days of lactation.

Following assignment of YD, its distribution was examined and two cut points for group classification selected. These cut points were selected on a biological basis while also considering the practical application of the predictive model. The first cut point was placed at 0% deviation from expected to separate animals expressing positive and negative yield deviation. Animals suffering negative deviations were further subdivided to allow differentiation of animals suffering large or moderate negative deviations. Within this dataset a standard deviation of 16% YD was observed. A negative 15% deviation was selected as the second cut

point representing approximately a single standard deviation below expected. This resulted in the formation of three groups. Cow-lactations with a negative YD of at least 15% were classified RED, cow-lactations with a YD between -14% and 0% were classified AMBER, those with a positive YD were classified GREEN.

Step 3: Predicting 30-Day Yield Deviation

In Step 3, machine learning models were constructed to predict YD using production, rumination, and physical activity data from 1–3 DIM. Our aim was to investigate the accuracy with which deviation from expected 30-day cumulative yield could be predicted within three days post-partum.

Data Preparation and Feature Engineering

Production and behaviour data from days 1–3 in lactation were utilised for model construction. For all animals assigned a YD in Step 2 milking visit frequency, the weight, conductivity, and temperature of milk harvested from the first 3 days in lactation was accessed from the PDS. For the same time period daily rumination and physical activity data were collated from within the BDS. Animals which failed to record a milk vield, conductivity or temperature reading for days 1-3 of lactation were removed (n = 293). Milk harvested at each visit was summed to create a daily yield for each cow. Daily rumination and activity levels were available as an arbitrary unit (AU) in 12 two-hour blocks (Elischer et al., 2013). Where between 1 and 3 blocks were missing, the mean value for all available blocks for the day in question were used to impute missing blocks. Where over 3 blocks in a single day were missing, the entire lactation was removed (n = 785). Following data preparation, 4 herds for which less than 20 lactations were available were removed. Thereafter 2,462 multiparous lactations from 27 herds remained for analysis.

Using this data, ten features were selected based on their biological plausibility to reflect the production potential of early lactation cows. These were: Maximum Yield (Kilograms); the highest daily yield, Minimum Activity (AU); the lowest daily activity, Maximum Rumination (AU); the maximum daily rumination, Maximum Temperature (Degree Celsius); the maximum mean daily milk temperature as averaged across all successful milking visits within a given day, Maximum Conductivity (AU); the maximum mean daily milk conductivity, as averaged across all successful milking visits for a given day, Total Refusals; the sum of refusals (where milking is refused due to an animal re-presenting a short time after a previous milking visit), Total Milkings; the sum of successful milking visits. To assess the change in yield, rumination, and physical activity levels over days 1–3, Yield Acceleration, Delta Rumination and Delta Activity were calculated. These represented the summed daily rate of change for their respective metrics from days 1-2 and 2-3 in lactation. For example, Yield Acceleration for any cow-lactation was calculated as the change in yield from days 1–2, expressed as a percentage of day 1 yield, summed with, the change in yield from days 2-3, expressed as a percentage of day 2 yield.

Model construction

Data were randomly split on a herd basis into a training dataset (21 herds) and test dataset (6 herds) comprising 74% and 26% of available lactations respectively. The mean number of lactations per farm was 91 with no single herd contributing more than 13% of total lactations. Random forest regression (Breiman, 2001), elastic net regression (Zou & Hastie, 2005), and multivariate adaptive regression splines (Friedman, 1991) were used to create predictive models for ECY. All 10 features were offered to each model. Model performance was evaluated using the minimisation of mean absolute error (MAE) from 5-fold cross validation repeated 10 times on the training dataset (Hastie et al., 2009) Random forest regression achieved the lowest MAE of all models and was used for construction of the final predictive model.

Final Model Training

Recursive feature elimination within the CARET package (Kuhn, 2008) was used to identify an optimal sub-set of variables for inclusion in the final model. Following feature selection, final hyperparameter tuning was carried out using a grid search (Kuhn, 2008). MAE was minimised with hyperparameters of 1000 and 2, representing the number of trees used and number of variables considered at each split point respectively.

Final Model Assessment

Final model performance and validity was assessed using the test dataset. Predictions from the test dataset were compared with observed outcomes and MAE calculated as a measure of model performance. The accuracy with which cow-lactations were assigned to their appropriate category, RED, AMBER or GREEN, was assessed by calculating sensitivity, specificity as well as positive predictive values (PPV) and negative predictive values (NPV). Due to the imbalanced distribution of lactations between groups, balanced accuracy, which accounts for class imbalance was utilised over the traditional metric of accuracy (Jiao & Du, 2016).

4.3 Results

4.3.1 Modelling Expected 30-Day Cumulative Yield

Model residuals were examined graphically and found to be normally distributed (Figure 4-1). Coefficients of the final linear mixed model used to estimate ECY are presented in Table 4-2. The mean absolute error and mean absolute percentage error were 140 kilograms and 15% respectively.

Table 4-2 Linear mixed model for Expected Cumulative 30-day Yield for 8659 cow-lactations from 31 herds between January 2017 and January 2022

| Fixed Effects | Estimate | 95% CI |
|----------------------------------|----------|-----------|
| Intercept | 1089.1 | 1005_1127 |
| Herd Production ¹ | 93.4 | 84_103 |
| Previous Production ² | 85.5 | 79_92 |
| Autumn | Baseline | - |
| Winter | 6.3 | -5_18 |
| Spring | 24.3 | 11_37 |
| Summer | 10.9 | -2_23 |
| 2017 | Baseline | - |
| 2018 | 47.7 | 11_83 |
| 2019 | 34.2 | 3_65 |
| 2020 | 62.8 | 8_70 |
| 2021 | 42.5 | 11_73 |
| 2022 | 62.8 | 28_98 |
| Parity 2 | Baseline | - |
| Parity 3+ | -37.9 | -5026 |
| Random Effects | Variance | Std. Dev |
| Herd ID (Intercept) | 3775 | 61.4 |
| Residual | 36129 | 190 |
| 1= | | |

¹The mean 30-day cumulative yield for all animals within the herd of origin, in the year and season for a given lactation. ² The mean 30-day cumulative yield for all available lactations prior to the lactation in question. CI = confidence interval

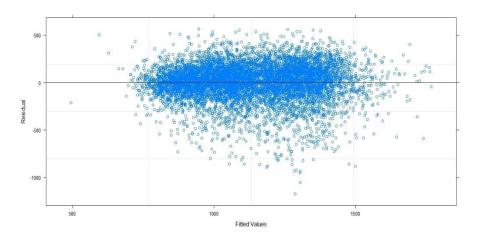


Figure 4-1 Scatter Plot of residuals plotted against fitted values for modelling of expected yield for Expected Yield Model in Step 1

4.3.2 Assessment of Yield Deviation

Across the entire dataset the range of YD was -91 to 84% with a mean of -0.02% and median of 1.6%. Mean YD per herd ranged from -6.0 to

4.9%. RED, AMBER, and GREEN groups comprised 15, 30 and 55 percent of all lactations respectively Figure 4-2.

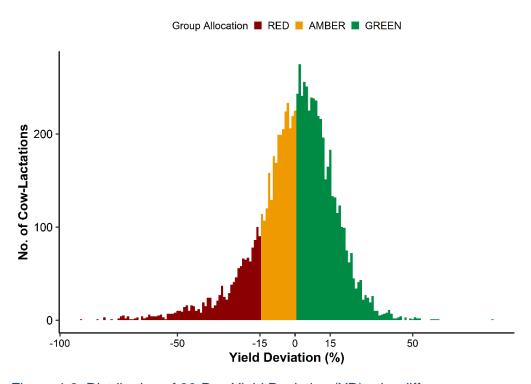


Figure 4-2 Distribution of 30-Day Yield Deviation (YD) - the difference between actual and expected 30-day cumulative yield expressed as a percentage of expected, for 8659 cow-lactations from 31 herds between January 2017 and January 2022. RED Group (< /= -15% YD), AMBER Group (-14%-0% YD), GREEN Group (> 0 YD).

Descriptive statistics for independent variables examined in Step 3 are presented in Table 4-3. Results of the initial random forest regression, elastic net regression and multivariate adaptive regression splines analyses are presented in Table 4-4. Random forest regression achieved an MAE of 8.5%, the lowest among models considered and was selected for further training.

Table 4-3 Descriptive statistics for independent variables examined in Step 3.

| Variable | Median | IQR |
|----------------------|--------|------|
| Peak Yield | 29 | 10 |
| Maximum Rumination | 6253 | 1410 |
| Yield Acceleration | 36 | 25 |
| Minimum Activity | 471 | 171 |
| Delta Rumination | 19 | 29 |
| Delta Activity | -6 | 20 |
| Maximum Temperature | 38.4 | 1.1 |
| Maximum Conductivity | 76 | 6 |
| Total Refusals | 12 | 23 |
| Total Milkings | 9 | 3 |

Table 4-4 Regression Performance for Elastic Net, Random Forest Regression and ¹Multivariable Adaptive Regression Splines

| | Mean Absolute Error | | | |
|--------------------------|---------------------|--------|------|------|
| Model | Min. | Median | Mean | Max. |
| Elastic Net Regression | 8.4 | 9.1 | 9.2 | 10.2 |
| Random Forest Regression | 7.9 | 8.5 | 8.5 | 9.5 |
| MARS ¹ | 8.2 | 9.0 | 9.0 | 10.0 |

Feature Selection and Model Tuning

Variable importance scores for the final random forest regression model, as calculated by recursive feature elimination, are presented in Table 4-5. The top six features, representing a compromise between model parsimony and performance were chosen for inclusion in the final model. These were, in descending order: Maximum Yield, Maximum Rumination, Yield Acceleration, Minimum Activity, Delta Rumination, and Delta Activity.

Table 4-5 Variable importance following recursive feature elimination of the final random forest model as selected for the minimisation of MAE in the model training process

| | Variable |
|----------------------|--------------------|
| | Importance |
| Variable | Score ¹ |
| Peak Yield | 132063 |
| Maximum Rumination | 30366 |
| Yield Acceleration | 24651 |
| Minimum Activity | 23271 |
| Delta Rumination | 22711 |
| Delta Activity | 18591 |
| Maximum Temperature | 17804 |
| Maximum Conductivity | 17363 |
| Total Refusals | 17188 |
| Total Milkings | 11474 |

¹Variable importance score as calculated by the VarImp function of the CARET package

Final Model Assessment

Assessment of the final model was carried out using the test dataset comprised of 628 cow-lactations from 6 herds. Across all cow-lactations within the test dataset, observed YD averaged 1.5%, ranging from -65.2 to 59.3%. Our model predicted YD with an MAE of 9%. At the herd-level, MAE ranged from 3% in Herd 45 to 14% in Herd 14 Table 4-6.

Table 4-6. Regression performance for the prediction of Yield Deviation within the test dataset as achieved by the final random forest model, selected for minimisation of MAE in the model training process

| Herd ID | 1 | 14 | 18 | 20 | 40 | 45 |
|---------|----|--------|--------|--------|------|----|
| | Re | gressi | ion Pe | erform | ance | |
| MAE (%) | 8 | 14 | 13 | 9 | 6 | 3 |

MAE = Mean absolute error

Classification performance across the entire test dataset is presented in Table 4-7. Within the test dataset, RED, AMBER, and GREEN categories accounted for 13, 30 and 57% of the population respectively. Calibration plots in which group classification, assigned using predicted YD, are plotted against observed YD for the entire test dataset and individual herds are presented in Figure 4-3 and Figure 4-4 respectively. Animals with a negative YD (RED and AMBER Groups) were classified with a sensitivity of 81%, NPV of 73%, specificity of 67% and a PPV of 76%. Animals with a positive and negative YD were differentiated with a balanced accuracy of 74% across the test dataset. This ranged from 93% in Herd 45, to 65% in Herd 14 (Table 4-8). Classification of animals within the RED group was achieved with a balanced accuracy of 67%, specificity of 99%, a PPV of 91%, sensitivity

of 35%, and a NPV of 90%. At the herd-level, PPV ranged from 83% in Herds 20 and 40, to 100% in Herds 45,14 and 18. Sensitivity ranged from 23% in Herd 1 to 100% in Herd 14. Of RED animals incorrectly classified, 76% were misclassified as AMBER and 24% misclassified as GREEN.

Table 4-7 Classification Performance for the final random forest model as selected for the minimisation of MAE in the model training process, of RED, RED + AMBER, and GREEN groups within the test dataset

| | RED | RED + AMBER | GREEN |
|----------------|-----|----------------|-------|
| Prevalence (%) | 13 | 43 | 57 |
| Sensitivity | 35 | 67 | 81 |
| Specificity | 99 | 81 | 67 |
| PPV | 91 | 73 | 76 |
| NPV | 90 | 76 | 73 |
| Balanced | 67 | 74 | 74 |
| Accuracy | | | |

PPV = Positive predictive value, NPV = negative predictive value. RED Group (< /= - 15% Yield Deviation), AMBER Group (-14%– 0% Yield Deviation), GREEN Group (> 0 Yield Deviation)

Table 4-8 Classification performance of the final random forest model as selected for the minimisation of MAE in the model training process, for the RED, RED + AMBER and GREEN Groups within the test dataset

| Herd ID | 1 | 14 | 18 | 20 | 40 | 45 |
|--------------------------|-------|---------------------------------------|-------------|---------|----------|-----|
| | Class | Classification Performance: RED Group | | | | ١ |
| Prevalence (%) | 13 | 5 | 32 | 19 | 10 | 8 |
| Sensitivity | 23 | 100 | 43 | 28 | 63 | 67 |
| Specificity | 99 | 100 | 100 | 99 | 99 | 100 |
| PPV | 90 | 100 | 100 | 83 | 83 | 100 |
| NPV | 89 | 100 | 79 | 85 | 96 | 97 |
| Balanced Accuracy | 61 | 100 | 71 | 63 | 81 | 83 |
| | Class | sification I | Performa | nce: RE | ED + AME | BER |
| | Grou | ps | | | | |
| Prevalence (%) | 40 | 29 | 45 | 46 | 56 | 50 |
| Sensitivity | 46 | 83 | 70 | 70 | 93 | 95 |
| Specificity | 92 | 47 | 83 | 80 | 42 | 92 |
| PPV | 80 | 38 | 78 | 75 | 67 | 92 |
| NPV | 72 | 88 | 77 | 76 | 83 | 94 |
| Balanced Accuracy | 69 | 65 | 77 | 75 | 68 | 93 |
| | Class | sification I | Performa | nce: Gl | REEN Gr | oup |
| Prevalence (%) | 60 | 71 | 55 | 54 | 44 | 50 |
| Sensitivity | 92 | 47 | 83 | 80 | 41 | 92 |
| Specificity | 46 | 83 | 70 | 70 | 93 | 95 |
| PPV | 72 | 88 | 77 | 76 | 83 | 94 |
| NPV | 80 | 38 | 78 | 75 | 67 | 92 |
| Balanced Accuracy | 69 | 65 | 77 | 75 | 68 | 93 |
| D = D O / / / / / - | A | | · • • • • • | | | |

RED Group (< /= -15% Yield Deviation), AMBER Group (-14%–0% Yield Deviation), GREEN Group (> 0% Yield Deviation)

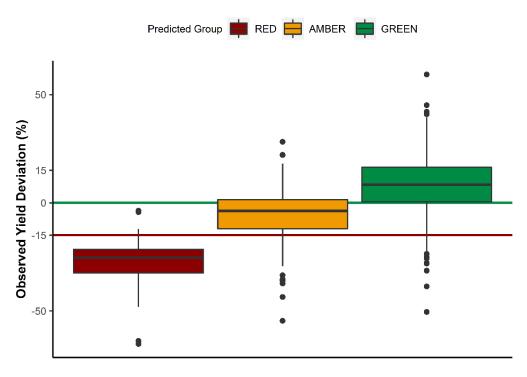


Figure 4-3 Calibration plot for class membership of the test dataset comprised of 628 lactations from 6 herds. Cow-lactations were classified using Predicted 30-day Yield Deviation (YD) into three groups: RED Group (</= -15% YD), AMBER Group (-14%–0% YD), GREEN Group (> 0% Yield Deviation).

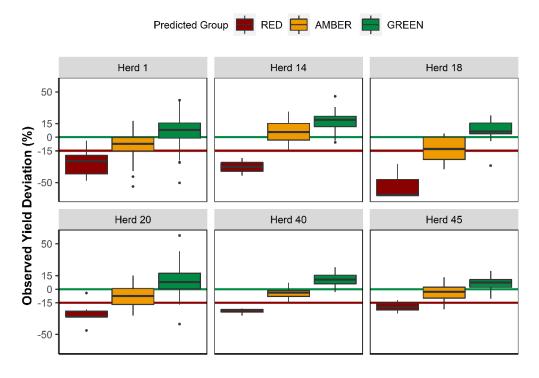


Figure 4-4 Calibration plots for the prediction of group membership for all 6 herds within the test dataset. Cow-lactations were classified using Predicted 30-day Yield Deviation into three groups: RED Group (< /= -15% YD), AMBER Group (-14%–0% YD) or GREEN Group (> 0% Yield Deviation)

4.4 Discussion

These results suggest that milk yield, rumination and physical activity patterns expressed by dairy cows in the first 3 days post-partum have utility in the prediction of deviation from expected 30-day cumulative yield. However, these predictions currently lack the accuracy required to classify cows reliably and completely into groups which may facilitate improved transition cow management.

Our final model predicted YD with an MAE of 9% which suggests good accuracy across individual animals and generalisability across herds. To assess the practical application of this model, the accuracy with which predicted YD could classify animals into three pre-defined management groups was examined. Where accurate classification could be achieved this may facilitate the allocation of resources, such as veterinary examination or disease screening, towards those predicted to underperform and away from those expected to meet expectations (Guterbock, 2004).

Of particular interest was the accuracy with which animals recording a large negative deviation (RED group) could be classified. The PPV and specificity for the classification of the RED group was excellent across the entire test dataset. However, on average across the test dataset the sensitivity for classification of animals within the group was poor. A large variation in sensitivity across herds was also recorded, ranging from 100% in Herd 14 to 23% in Herd 1. This has practical implications for the utility of our model, in particular where poor sensitivity is the result of the misclassification of RED animals as GREEN. Where resources are to be allocated based on group classification, misclassification of this type is undesirable. Interestingly, only half the herds within the test dataset (Herds 1,18 and 20), misclassified any RED animals as GREEN. Of those three herds, Herd 1 accounted for 80% of all those misclassified. The number of RED animals misclassified limits the potential benefit of this model at present. Further research is therefore required to understand the reasons for the observed between-herd variance in the classification accuracy within the RED group.

Despite the diverse composition of our test dataset, MAE showed very little variance at the herd level. This highlights the consistency with which yield, rumination and physical activity data can predict YD. This may be attributed to the inclusion of relative metrics such as Yield Acceleration, Delta Rumination and Delta Activity in our model. As opposed to metrics utilising absolute terms, these relative metrics use the cow's prior performance as a benchmark. The inclusion of such metrics has previously demonstrated importance in the development of generalisable models when utilising animal sensor data (Stangaferro et al., 2016).

The six parameters retained in our final model were comprised of three absolute and three relative metrics stemming from three sources: milk yield, rumination, and physical activity. Our findings are in agreement with prior work demonstrating the utility of early lactation milk yield as a

predictor of production in the remainder of the lactation (Grzesiak et al., 2006; Njubi et al., 2010). While rumination in the early lactation period has been associated with subsequent milk production its predictive values has not been previously reported (Peiter et al., 2021). Our results also highlight the utility of features stemming from physical activity. In the immediate post-partum period physical activity has not been previously reported to carry any direct association with subsequent milk yield, though its association with transition cow health has been cited (Kaufman et al., 2016). An investigation of the association between physical activity in the days immediately post calving and subsequent yield performance may be therefore warranted, in particular as it relates to production in free flow AMS herds.

This study is the first to report the development of a predictive model for yield deviation in early lactation dairy cows. While we believe this model has demonstrated potential utility for the improvement of transition cow management, there are several limitations associated with our modelling approach. For instance, we were unable to assign a predicted yield deviation to primiparous animals in our study. This stems from the reliance on performance in prior lactations for the calculation of expected yield and subsequently, yield deviation. The practical consequences of this may be mitigated by the lower risk for poor transition health which first lactation animals represent when compared with their multiparous herd mates (Lee Ji-Yeon Kim III-Hwa, 2006). However, it may be possible for future analysis to incorporate the prediction of first lactation yield in order to investigate the accuracy of YD predictions in primiparous animals (Dallago et al., 2019). A further limitation of this study relates to the method by which YD was calculated. As this was measured by cumulative 30-day yield, animals which were removed from the herd prior to 30 DIM were not included in this study. Therefore, additional research to include the utility of early lactation data for the prediction of cull risk in early lactation would be beneficial as these animals form a group which would likely benefit from early management intervention.

4.5 Conclusions

The application of predictive modelling in transition management programs a relatively novel concept. Our results advance this field by demonstrating the utility of yield, rumination, and physical activity metrics from 1-3 DIM for the prediction of deviation from expected milk yield over the first month of lactation. When used to differentiate animal groups based on Yield Deviation, our model classified those suffering large negative deviations with excellent specificity but poor sensitivity. This lack of sensitivity limits the current utility of this model to inform transition management on farm. However, these results highlight the potential in automatically collected AMS data for the prediction of deviation in early lactation milk yield.

Chapter 5 The Association of Production and Behaviour Data with Fertility Performance

5.1 Introduction

The efficiency of reproductive management has a significant impact on the economic sustainability of the dairy herd. When managed appropriately, the direct costs associated with generating pregnancies can be minimised, while indirect benefits such as increased milk production and reduced involuntary culls can generate substantial economic gains (Cabrera et. al., 2014; Shalloo et al., 2014). The reproductive performance of the UK dairy herd has improved over the past decade. Since 2010 the median conception rate has increased by 3% to 35%. Over the same period the median submission rate has increased from 27% to 40% (Hanks, 2021). Positive trends have also been reported within the Irish dairy industry with a 9% increase in sixweek calving rate reported between 2012 and 2020 (NDC, 2021) However, inefficiencies remain within the UK and Irish industries as reflected by their median calving intervals of 400 and 388 days respectively (NDC, 2021; Hanks, 2021). Moving forward, continued advancements in reproductive performance will play a key role in both industry's pursuit of increased economic sustainability (Diavão et al., 2023).

The chain of events which comprise a successful fertility cycle is complex (Roche et al., 2018). Common to a large proportion of these however, is that they occur during the transition period. For many cows, resumption of cyclicity, uterine involution, as well as the maturation and selection of the follicle which will ultimately be fertilised will all occur during this time. Reproductive success is therefore linked with the health of the cow during transition (Roche et al., 2018; Stevenson et al., 2020; Consentini et al., 2021; Pascottini et al., 2022). Adaptation to the challenges of transition requires an acute homeorhetic response across three broad pathways, these being energy metabolism, mineral metabolism and immune function. Insufficient or inappropriate responses to these challenges has a detrimental effect on subsequent fertility (Pascottini et al., 2022). In the case of energy metabolism, the severity and duration of negative energy balance experienced directly influences the resumption of cyclicity by modulating the concentration of both luteinising hormone and follicle stimulating hormone (Roche et al., 2018). Follicles which mature and ovulate under states of negative energy balance tend to be larger, more aged and consequently less fertile, yielding lower conception rates (Roche et al., 2018). A direct effect of insufficient immune function on reproductive performance is seen in the occurrence of metritis, which has demonstrated significant association with delayed resumption of cyclicity (Vercouteren et al., 2015), and reduced conception to first insemination (Elkjær et al., 2013). For the transition cow, challenges relating to mineral metabolism

are dominated by the risk of hypocalcaemia. By suppressing the function of both smooth muscle and immune cells, hypocalcaemia is associated with reduced uterine involution and the retention of foetal membranes. This has also been demonstrated to lead to delays in resumption of cyclicity, as well as reduction in conception rate to first service (Caixeta et al., 2017).

The means by which commercial dairy producers can monitor the health of transition cows as they adapt to these challenges has grown exponentially in the past two decades. The assessment of energy and mineral metabolism using commercially available serum tests (Geishauser et al., 2001; Townsend, 2011; Seifi & Kia, 2017) as well as their association with subsequent reproductive performance (Walsh et al., 2007; Seifi & Kia, 2017) have been well described. These investigations demonstrate the association between transition health indicators and subsequent fertility. Furthermore, they highlight the potential such indicators may have for the identification of animals likely to suffer poor reproductive performance. By facilitating the pro-active management of sub-fertile animals, transition cow health monitoring programs may represent a powerful tool for the improvement of reproductive efficiency. However, the uptake of such monitoring approaches remains limited due to the labour-intensive nature of sampling required (König et al., 2023).

Modern automatic milking systems (AMS) offer an alternative means to monitoring transition cow health. In contrast to the traditional means of transition cow monitoring, they provide an automated assessment of production and behaviour data, available in real time from the point of calving. To date the association of this data with subsequent reproductive performance has not been investigated. An improved understanding of this relationship may inform future work directed towards the use of AMS data within an automated transition cow monitoring program. The objective of this study was to test the hypothesis that production and behaviour data collected by AMS over day 1-21 post-partum would demonstrate significant association with subsequent detection of oestrus and conception to first insemination.

5.2 Materials and Methods

5.2.1 Study Population

A convivence sample of 46 herds was recruited from the UK and Republic of Ireland as described in Chapter 2. Briefly, the criteria for inclusion were the use of Lely Astronaut milking robots (Lely International, N.V.) under free flow traffic conditions (Munksgaard et al., 2011), in conjunction with rumination and physical activity monitoring technology (Lely Qwes-HR collars, Lely International N.V.). Year-round and seasonal calving patterns were represented within the dataset. Data relating to milk quantity, milk quality and the frequency of cowrobot interactions, as well as reproductive management records were accessed. No information pertaining to the use of fixed time insemination was available. To assess the degree to which such

practices may have been applied within our dataset, insemination records were examined as previously described by Barden et al. (2024). In brief, for each herd, the proportion of inseminations on each day, of each week were calculated. Thereafter, the binomial standard deviation for a uniformly distributed proportion for all inseminations within each week was calculated. Any day for which the proportion of inseminations reported was in excess of 2 standard deviations for the week was identified and all inseminations on that day marked as potentially fixed time (PFT) inseminations. Within the final conception to first insemination (CFI) dataset,10% of inseminations were identified as potential fixed time inseminations.

5.2.2 Data Preparation

Two outcomes of interest were selected for investigation. The first was Expression of Oestrus or Insemination (EOI). An animal was classed as EOI+ where an oestrus or insemination event was recorded between days 22 and 65 post-partum. This time frame was selected in order to encompass an oestrus cycle before and after the start of breeding where a traditional 42-day waiting period is applied. An oestrus event was defined as three consecutive 2-hour periods of increased activity compared with each animal's pre-determined baseline as detected by a neck mounted activity monitor. Insemination records were assessed via the on-farm management system, Lely T4C. The second outcome of interest was Conception to First Insemination (CFI). Animals were classed as CFI+ where they received their first and only insemination between days 22 and 80 post-partum and were subsequently recorded as pregnant on the farm management system. This time frame was selected to examine the relationship between transition cow data and fertility in the initial stages of the breeding period.

Independent variables were engineered using data from the first 21 days post-partum. Milk quantity was assessed as Mean Milk Yield (kg); the mean of daily milk yields over days 1-21. Yield Acceleration (%); was defined as the change in daily milk yield over consecutive days expressed as a percentage of the first. For example, Yield Acceleration for each cow-lactation was calculated as the change in yield from days 1–2, expressed as a percentage of day 1 and so on for days 1-21. This was utilised as Mean, Minimum and Maximum Yield Acceleration, where Mean Yield Acceleration was the mean of Days 1-21 and Minimum and Maximum the highest and lowest daily FPR recorded over days 1-21. Milk Quality was assessed using milk temperature, conductivity and constituent data. Two measures of milk temperature were employed, Maximum Temperature; the maximum quarter-level temperature recorded and, Temperature > 40; a binary indicator for animals recording a milk temperature of over 40 Degree Celsius. Conductivity was assessed as Mean Conductivity (AU); the mean udder-level conductivity recorded over days 1-21 and Max Conductivity; the highest mean udder-level conductivity recorded over days 1-21. Milk fat and protein indications, as recorded once daily by Lely's MQC (Fadul-Pacheco et al. 2018) were utilised as Mean Fat and Mean

Protein: the mean of recorded values across days 1-21 for their respective constituent. For each day a fat-to-protein ratio was subsequently calculated. This was utilised as Mean Fat-to-Protein ratio (FPR); the mean daily fat-to-protein ratio achieved across days 1-21, and a Maximum FPR; the highest daily FPR recorded across days 1-21. Mean Concentrate Dispensed (g) was calculated as the mean grams of concentrate dispensed by the robot to each animal per day. Robot visit behaviour parameters consisted of milking visits and milking refusals (where milking permission is denied due to an animal re-presenting a short time after a previous milking visit). These were averaged over days 1-21 and reported as Mean Milkings and Mean Refusals. To be retained in the final dataset, measurements for each of these parameters were required for at least 16 of the first 21 days postpartum. Where these were not available the entire cow-lactation was removed. Parity was assessed as 1 and 2+. Within the CFI dataset two additional variables were assessed. Potential Fixed Time Insemination as a binary variable, and Days in Milk at First Service. A selection of interaction terms were investigated based on their biological plausibility to influence reproductive performance. The interaction between Mean Milk Yield and Yield Acceleration, Mean Concentrate Dispensed, Mean Protein, Mean Milkings and Mean Refusals was assessed, as was the interaction between Mean Milkings and Mean Yield Acceleration. Prior to modelling, independent variables for each outcome of interest were screened for correlation using a Pearsons correlation matrix (Kirch, 2008). Variable distributions were assessed, and transformations applied as necessary. All numeric variables were centred by subtracting the column means of each variable from their corresponding columns and scaled by dividing each variable by their standard deviations (van den Berg et al., 2006).

5.2.3 Model Construction

For each outcome of interest, a multivariable mixed-effect logistic model was constructed within the lme4 package (Bates et al., 2015) using a manual backward step procedure as described in Section 3.2. All candidate variables were screened using univariable analysis and brought forward for inclusion in a multivariable model where a *P*-value of ≤ 0.20 was observed. Multivariable models were constructed using the logit link function and included a random intercept to account for the clustering of data at herd level (Dohoo et. al., 2003). Variables were retained in the final model where a *P*-value of ≤.05 was observed. Any variables which formed a significant interaction term were forced into the final model. On completion of the backward step, all main effects which were removed were re-entered and tested for significance in the final model. The effect of nesting cow-lactations within Cow and Year, as well as cows within herd was assessed. The inclusion of Calving Pattern as a random effect within the final model were also evaluated. Parity was retained as a confounder within the final EOI and CFI models. Model residuals were assessed for normality of distribution and goodness-of-fit was evaluated using marginal and conditional R² values (Nakagawa et al., 2017). Model performance was assessed following

internal validation using the area under the receiver operator curve (AUC-ROC), sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV).

The final multivariable mixed-effect logistical models took the general

form:

$$log\left(\frac{P_{ij}}{1 - P_{ij}}\right) = \beta_0 + \beta_1 X_{1ij} + \dots + \beta_m X_{mij} + v_j + \epsilon_{ij}$$
$$[v_j] \sim N(0, \Omega_v)$$
$$[\epsilon_{ij}] \sim N(0, \Omega_e)$$

where the subscript i refers to the ith cow and the subscript j refers to the jth herd. Each β represents a fixed effect for EOI and CFI models, v_j represents a random effect for Herd (assumed to have a normal distribution of mean = 0, and variance = Ω_v) and ϵ_{ij} represents the residual model error (assumed to have a normal distribution of mean = 0, and variance = Ω_e). Parameter P_{ij} represents the probability of EO or CFI for the ith cow in the jth herd.

5.3 Results

Complete or partial production records from 33,592 cow-lactations across 39 herds spanning the years 2016 to 2023 were available for analysis. Records describing oestrus expression between days 22 and 65 post-partum were available for 18,652 cow-lactations, 46% of which expressed oestrus. Of the 26,293 first services recorded between days 21-80 for which an outcome could be established, 40% resulted in conception. Following the removal of lactations with incomplete AMS data, 16,486 cow-lactations remained for pairing with a corresponding EOI and CFI outcome. A total of 9,638 lactations from 25 herds remained in the final EOI dataset, 48% of which were classed as EOI+. Of these, 96% were classed EOI+ following an automatically detected heat event and 4% following an insemination event. In the final CFI dataset 8.635 cow-lactations from 23 herds remained, of which 41% conceived and 10% were listed as PFT inseminations. Descriptive statistics for the herds within EOI and CFI datasets are presented in Table 5-1 and Table 5-2 respectively. Frequency distribution for EOI and CFI events are presented in Figure 5-1 and Figure 5-2 respectively. The voluntary waiting period for each herd was estimated using the 5th percentile of days in milk at first services. On a herd basis the mean DIM at first services for the earliest 5% of services ranged from 27 to 56 DIM. The herd level distribution for this metrics is presented in Figure 5-3

Median values and interquartile ranges for all independent variables are provided for the EOI and CFI datasets in Table 5-3 and Table 5-4 respectively. To normalise their distribution, a log transformation (van den Berg et al., 2006) was applied to Mean Refusals, Conductivity Alert, Max Yield Acceleration, and Mean Yield Acceleration. Mean Fat Percentage, Min Yield Acceleration and Max Conductivity were removed due to correlation of over 0.75, with Mean FPR (r = 0.89), Mean Yield Acceleration (r = 0.87) and Mean Conductivity (r= 0.78) respectively. Coefficients for the final EOI and CFI multivariable models are presented in Table 5-5 and Table 5-7 respectively. The odds ratios reported reflect the change in the odds of an animal being classed as positive (i.e., EOI+ or CFI+) in response to an increase of one standard deviation within their respective independent variable. To aid interpretability, model outputs are described using the value of one standard deviation within each variable's original unit of measurement.

Table 5-1 Descriptive statistics for herds included in the final EOI multivariable model

| Variable | No. [range] |
|-------------------------------------|----------------|
| No. of Cow- Lactations | 9,638 |
| No. of Farms | 25 |
| Calving Pattern | |
| All Year Round | 16 |
| Seasonal | 9 |
| Mean No. of Cow-Lactations/Herd | 385 [152–952] |
| Mean Peak Yield¹ – Herd Level | 46kg [32-63kg] |
| Oestrus Expression | |
| Total | 48% |
| Parity 1 | 56% |
| Parity 2+ | 44% |
| Percentage EOI+ - Herd Level | 47% [24–76%] |
| Mean DIM at EOI+ event – Herd Level | 50 [45-53] |

¹Calculated using maximum single day yield recorded in the first 100 days in milk.

Table 5-2 Descriptive statistics for herds included in the final CFI multivariable model

| Variable | No. [range] |
|--|----------------|
| No. of Cow- Lactations | 8,635 |
| No. of Farms | 23 |
| Calving Pattern | |
| All Year Round | 16 |
| Seasonal | 7 |
| Mean No. of Cow-Lactations/Herd | 375 [174–869] |
| Mean Peak Yield1 – Herd Level | 47kg [32-62kg] |
| Conception Rate to First Insemination | |
| Total | 41% |
| Parity 1 | 45% |
| Parity 2+ | 39% |
| Mean Conception Rate – Herd Level | 42% [23–61%] |
| Percentage of first services recorded | 65% [45-90%] |
| within 22-80 DIM – Herd Level | _ |
| Median DIM at First Service – Herd Level | 60 [53-70] |

¹Calculated using maximum single day yield recorded in the first 100 days in milk.

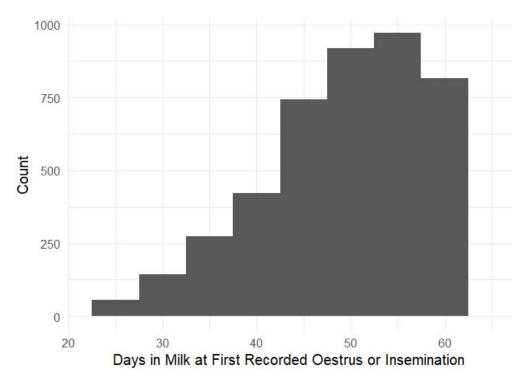


Figure 5-1 Frequency distribution for the timing of oestrus or insemination events across the 9,638 lactations in the EOI dataset.

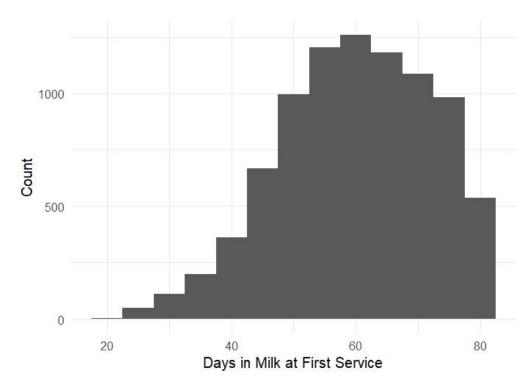


Figure 5-2 Frequency distribution for the timing of first insemination events across the 8,635 lactations in the CFI dataset.

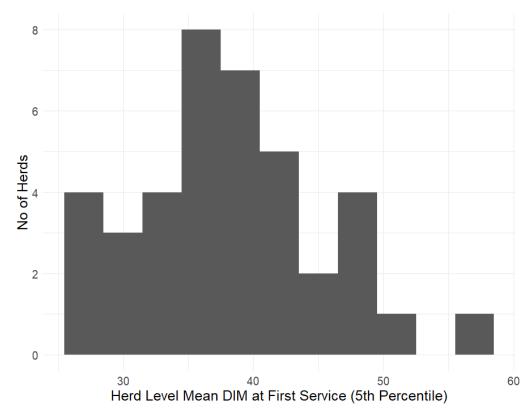


Figure 5-3 Frequency distribution for the estimated VWP adopted on each herd as calculated by the mean DIM at first service for the earliest 5% of services.

Table 5-3 Descriptive statistics for independent variables in final EOI dataset.

| Variable | Median | IQR |
|--------------------------------|--------------------------|------|
| Mean Milk Yield (kg) | 30 | 14 |
| Mean Yield Acceleration (%) | 3.6 | 2.1 |
| Min Yield Acceleration (%) | 28 | 19 |
| Max Yield Acceleration (%) | -10 | 20 |
| Mean Concentrate Dispensed (g) | 5168 | 1416 |
| Mean Refusals | 2.4 | 4.3 |
| Mean Milkings | 2.9 | 1.4 |
| Mean Protein | 3.74 | 0.37 |
| Mean Fat | 4.43 | 0.84 |
| Mean Fat-to-Protein Ratio | 1.18 | 0.23 |
| Max Fat-to-Protein Ratio | 1.4 | 0.32 |
| Mean Conductivity (AU) | 69.4 | 3.9 |
| Max Conductivity (AU) | 75.8 | 5.3 |
| Conductivity Alert (AU) | 0 | 3 |
| Milk Temperature (Degrees C) | 38.0 | 1.2 |
| | Percentage of Dataset | |
| Temp >40 | 1% | |
| Parity 1 | 33% | |
| Parity 2+ | 67% | |
| Seasonal Calving Pattern | 36% | |
| All Year-Round Calving Pattern | 64% | |

IQR = Interquartile range

Table 5-4 Descriptive statistics for independent variables in final CFI dataset.

| Variable | Median | IQR |
|--------------------------------|--------|------|
| Mean Milk Yield (kg) | 30 | 14 |
| Mean Yield Acceleration (%) | 3.6 | 2.2 |
| Min Yield Acceleration (%) | -10 | 20 |
| Max Yield Acceleration (%) | 28 | 20 |
| Mean Concentrate Dispensed (g) | 5262 | 1391 |
| Mean Refusals | 2.0 | 3.7 |
| Mean Milkings | 2.9 | 1.2 |
| Mean Protein | 3.73 | 0.35 |
| Mean Fat | 4.87 | 0.83 |
| Mean Fat-to-Protein Ratio | 1.17 | 0.22 |
| Max Fat-to-Protein Ratio | 1.4 | 0.31 |
| Mean Conductivity (AU) | 69.4 | 3.9 |
| Max Conductivity (AU) | 75.8 | 5.5 |
| Conductivity Alert | 0 | 3 |
| Milk Temperature (Degrees C) | 38.1 | 1.1 |
| DIM at First Service | 60 | 18 |
| | | |

| | Percentage of Dataset |
|--------------------------------|--------------------------|
| PFT Insemination | 10% |
| Temp >40 | 1% |
| Parity 1 | 34% |
| Parity 2+ | 66% |
| Seasonal Calving Pattern | 30% |
| All Year-Round Calving Pattern | 70% |

IQR = Interquartile range, DIM= Days in milk, PFT= Potential fixed time

5.3.1 EOI Model

The output of the final multivariable EOI logistic regression model is presented in Table 5-5. Mean Milk Yield, Mean FPR, Mean Conductivity, Conductivity Alert and Parity were all negatively associated with the odds of EOI. Each 8kg increase in Mean Milk Yield was associated with a 37% decrease in the odds of EOI (OR: 0.63, CI: 0.58-0.69). An increase of 0.2 in Mean FPR returned a 14% decrease in the odds of EOI (OR: 0.86, CI: 0.81-0.90). Mean Conductivity and Conductivity Alert both returned a 6% decrease in the odds of EOI in response to a 5.0 and 5.2 increase respectively (OR: 0.94, CI: 0.90-0.99). Multiparous animals demonstrated a 22% decrease (OR: 0.78, CI: 0.67-0.90) in the odds of EOI compared with primiparous animals.

Two variables recorded a positive association with the odds of EOI. An increase of 0.29 in Mean Protein (OR: 1.07, CI: 1.02-1.14) and 0.8 increase in Mean Milkings (OR: 1.26, CI: 1.18-1.36) was associated with an 8% and 26% increase in the odds of EOI respectively.

Classification performance for the final multivariable mixed-effect logistic model is presented in Table 5-6. Following internal validation, sensitivity and specificity of the final model was 67% and 66% respectively. A PPV 65%, NPV 68% and AUC-ROC of 0.72 was achieved. The final model explained 15% (conditional R²) of the variation in the expression of oestrus observed with 6% (marginal R²) attributable to the fixed effects.

Table 5-5 Fixed effects retained in multivariable mixed logistic regression model assessing the association between early lactation AMS production and behaviour data and the risk of expression of oestrus or insemination (EOI). SD = Standard deviation for respective variable.

| | | Odds | | | |
|--------------------|-------------|-------|-------------|-----------------|------|
| Variable | Coefficient | Ratio | 95% CI | <i>P</i> -value | SD |
| Intercept | 0.04 | 1.05 | 0.89 -1.37 | 0.7 | - |
| Mean FPR | -0.14 | 0.86 | 0.81-0.90 | < 0.001 | 0.20 |
| Mean Protein | 0.07 | 1.07 | 1.02-1.14 | 0.007 | 0.29 |
| Mean Milkings | 0.23 | 1.26 | 1.18-1.36 | <0.001 | 8.0 |
| Mean Conductivity | -0.06 | 0.94 | 0.90-0.99 | 0.012 | 5.0 |
| Mean Milk Yield | -0.45 | 0.63 | 0.58-0.69 | < 0.001 | 8 |
| Conductivity Alert | -0.06 | 0.94 | 0.90-99 | 0.014 | 5.2 |
| Parity 1 | Reference | - | - | - | - |
| Parity 2+ | -0.25 | 0.78 | 0.0.67-0.90 | < 0.001 | |

Table 5-6 Performance metrics for EOI and CFI Mixed-effect Multivariable Logistic Models assessed through internal validation

| | EOI | CFI |
|-------------|-------|-------|
| | Model | Model |
| AUC-ROC | 0.72 | 0.62 |
| Sensitivity | 67 | 59 |
| Specificity | 66 | 58 |
| PPV | 65 | 49 |
| NPV | 68 | 67 |

AUC-ROC= Area under the receiver operator curve, PPV = positive predictive value, NPV = negative predictive value

5.3.2 CFI Model

The output for the final CFI model is presented in Table 5-7 and Figure 5-4. Accounting for parity and DIM at first service, all statistically significant effects retained in the final model demonstrated a negative association with the odds of conception to first insemination. An 8Kg increase in Mean Milk Yield was associated with an 11% decrease in the odds of conception (OR: 0.89, CI: 0.82-0.96). A 0.2 increase in Mean FPR results in a 7% decrease (OR: 0.93, CI: 0.88-0.98). Multiparous animals returned a 10% decrease in the odd of conception verse primiparous animals (OR: 0.90, CI: 0.78-1.04) and finally, the interaction term Mean Milk Yield X Mean Concentrate Dispensed returned an odds ratio of 0.95. Within this interaction term, the association between Mean Milk Yield and the odds of conception became increasingly negative as Mean Concentrate Dispensed increased (Figure 5-4). Classification performance of the final CFI model is presented in Table 5-6. A sensitivity of 59% and specificity of 58%, PPV of 49%, NPV 67% and AUC-ROC of 0.62 were achieved. A conditional R² of 5% with a marginal R² of 1% was returned.

Table 5-7 Fixed effects retained in multivariable mixed logistic regression model assessing the association between early lactation AMS production and behaviour data and the risk of conception to first insemination (CFI). SD =Standard deviation for each respective variable.

| | | Odds | | | |
|----------------------|-------------|-------|-------------|-----------------|------|
| Variable | Coefficient | Ratio | 95% CI | <i>P</i> -value | SD |
| Intercept | -0.26 | 0.77 | 0.63 -0.93 | 0.006 | - |
| Mean FPR | -0.07 | 0.93 | 0.88-0.98 | 0.002 | 0.20 |
| Mean Milk Yield | -0.11 | 0.89 | 0.82-0.96 | 0.002 | 9.7 |
| Mean Milk Yield X | | | | | - |
| Mean Conc. Dispensed | -0.05 | 0.95 | .9099 | 0.043 | |
| Mean Conc. Dispensed | 0.01 | 1.01 | 0.95-1.08 | 0.69 | 1340 |
| Parity 1 | Reference | - | - | - | - |
| Parity 2+ | -0.10 | 0.90 | 0.78-1.04 | 0.15 | - |
| DIM at Service | 0.10 | 1.11 | 1.06 – 1.16 | < 0.001 | 12 |

FPR = Fat-to-protein ratio, Conc. = Concentrate feed, DIM = Days in milk, CI = Confidence interval

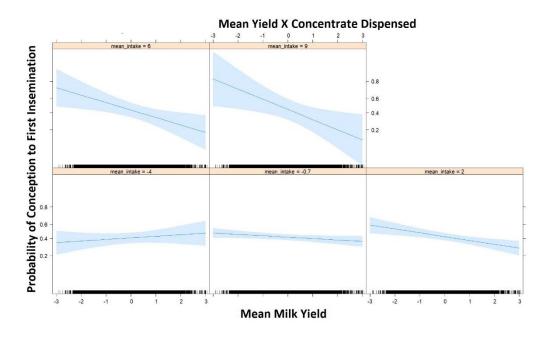


Figure 5-4 Effects plot for the interaction term Mean Milk Yield X Mean Concentrate Dispensed assessed overs days 1-21 post-partum as retained in the final multivariable model for Conception to First Insemination (CFI).

5.4 Discussion

Expression of oestrus in early lactation and conception to first insemination represent economically important aspects of reproductive management. Here, we report significant association between data collected by AMS over days 1-21 post-partum and both these outcomes. Of note, is the consistency with which the associations between AMS data and reproductive success reflects those previously demonstrated using traditional means of transition cow monitoring. These results emphasise the importance of transition cow health for the management of reproductive efficiency and demonstrate the potential utility of AMS production and behaviour data within transition cow monitoring programs.

The association between traditional indicators of energy metabolism and subsequent reproductive performance is clearly defined within the current literature. Energy balance, as assessed through body condition score in early lactation has demonstrated a consistent association with both conception to first insemination (Patton et al., 2007; Santos et al., 2009; Bedere et al., 2018) and resumption of cyclicity (Santos et al., 2009; Monteiro et al., 2021). Similarly, the relationship between metabolic indicators of NEB such as beta-hydroxy-butyrate and non-esterified-fatty acids with the resumption of cyclicity (Miqueo et al., 2019; Chin et al., 2024) and conception to first insemination (Garverick et al., 2013) have also been well established. Within our dataset, assessment of energy balance was carried out using milk fat and protein indications automatically collected via in-line sensors. Indications of a negative energy balance (i.e., increased Mean FPR and

decreased Mean Milk Protein (Toni et al., 2011)) were associated with reduced odds of oestrus expression within the final EOI model. This is in keeping with previous studies which reported significant positive association between milk protein percentage assessed over the first 100 days post-partum and the odds of resumption of cyclicity (Opsomer et al., 1999), while increase FPR had been associated with an increased risk of ovarian cysts (Heuer et al., 1999). The effect of FPR within the CFI model is also in keeping with prior reports with both Loeffler et al., (1999) and Heuer et al., (1999) reporting a negative association between test-day FPR in early lactation and the risk of conception to first insemination. This is the first study to report the relationship between milk fat and protein indications as measured via in-line sensors and subsequent reproductive performance. Our results indicate this may be a viable alternative to traditionally employed, labour-intensive assessments of energy balance such as body condition scoring and analysis of serum samples.

Two conductivity-based variables were retained within the final EOI model, both demonstrated a reduced odds of oestrus expression with deteriorating udder health. Susceptibility to mastitis is highest in the periparturient period, in part due to the compromised immune function commonly experienced during this time (Sordillo, 2005). Animals with SCC >500,000 in the first month post-partum demonstrated significantly higher incidence of delayed resumption of cyclicity when compared with animals recording an SCC of <500,000 (Isobe et al., 2014). A similar effect of clinical mastitis was demonstrated by (Rial et al., 2022) with mastitis diagnosed in the two weeks post-partum leading to a delay in both the resumption of cyclicity and detection of first oestrus. While there has been no prior investigation of the relationship between milk conductivity and subsequent fertility, our findings substantiate the previously describe relationship between indicators of udder health and reproduction.

Mean Milkings returned the largest positive effect size within the final EOI model highlighting an interesting relationship between this AMS metric and the odds of oestrus expression. Milking visits have been reported to decrease in response to disease, including ketosis and mastitis (King et al., 2018), and so to some extent may reflect the status of both the immune and metabolic systems. A more direct effect may relate to the demonstrated negative impact of lameness on milking visits (Borderas et al., 2008). Given the negative effect of lameness on the intensity of oestrus expression (Morris et al., 2011), this may account for the associations demonstrated here. However, the sparsity of reports detailing the association between metrics unique to AMS, such as milking visits, with reproductive performance means validation of these findings would be beneficial.

Across both EOI and CFI models, an increase in Mean Milk Yield was associated with a reduction in the odds of reproductive success. The relationship between milk yield and fertility is complex with conflicting results commonly reported. Bedere et al., (2018) demonstrated a significant association between the time to first observed oestrus and

milk yield with each extra kilogram of milk produced at peak associated with a delay in oestrus detection of 1.1 days. In contrast, Santos et al., (2009) demonstrated a positive association between milk yield over the first 90 DIM, and cyclicity at 65 DIM, while other studies have failed to demonstrate any association between these variables (Bulman & Lamming, 1978; Opsomer et al., 1999, Gümen et al., 2003). The relationship between milk yield and the probability of conception to first insemination is similarly equivocal with conflicting reports of a positive (Domecq et al., 1997), negative (Grimard et al., 2006; Bedere et al., 2018), or complete lack of association (Santos et al., 2009) between milk yield and conception.

The lack of consistency seen across these investigations may be somewhat explained by the variance in both the dependant and independent variables used across these studies (e.g. peak yield vs 305-day yield, resumption of cyclicity as defined by blood progesterone vs detection of oestrus). Larger, standardised studies are therefore required to more clearly define this relationship. Within this study we have demonstrated a significant negative association between milk production over the first 21-days and both expression of oestrus and conception to first insemination. While these results should be interpreted with caution considering the variance seen within the literature, it remains that across this large multi-herd dataset Mean Milk Yield exerted the largest effect of all variables examined. This highlights the importance of this variable and its potential utility within a transition monitoring program.

For both final models the coefficients of determination demonstrate that the fixed effects assessed in this analysis account for a very small proportion of the variance in outcome observed. This indicates that factors outside of those considered in this analysis are responsible for the vast majority of variance in reproductive performance seen in this dataset. Given the restricted period over which independent variables were analysed this was not an unexpected result. By limiting our analysis to variables from days 1-21 we excluded data relating to the animal's physiological status likely to be highly influential on reproductive performance. For example, indicators of udder health (Santos et al., 2004) and the level milk production (Rutten et al., 2016) in the days surrounding an animals' first insemination have a demonstrated association with the risk of conception. Inclusion of factors such as these would likely increase the degree to which any model may explain reproductive performance. However, the purpose of this study was to examine the association between data generated during the transition period and subsequent fertility. While our results must be interpreted in light of the low coefficients of determination observed, this does not negate the potential benefits an improved understanding of these associations may deliver.

We have demonstrated that variables recorded by AMS overs days 1-21 post-partum have a statistically significant association with fertility. As the first study of its type, our results advance this field by assessing the relationship between metrics unique to AMS (e.g., Milking Visits and

Refusals), and subsequent fertility. Furthermore, we report the association of AMS data relating to milk production, energy balance and udder health as important factors in reproductive performance. These findings agree with much of the prior literature demonstrating that data automatically collected by AMS has potential as an alternative means of transition of cow monitoring. While the low coefficient of determination, small effect sizes and moderate AUC-ROC observed must temper the potential this data holds for the prediction of reproductive outcomes within a TMP, this does warrant further investigation due to the benefits of automated collection of data by AMS and the opportunity to predict performance outcomes in a timely manner for intervention. The advent of targeted reproductive management programs, designed to allow bespoke management of fertility in line with expected performance (Giordano et al., 2022), offers a means to prevent or mitigate expected reproductive losses. Where a TMP could be developed to work in tandem with targeted reproductive management, there exists great potential to limit the effects of poor transition health on reproductive efficiency.

A major limitation of our study is the absence of data relating to herdlevel reproductive management practices. Within the herds analysed a large variance in the percentage of first services occurring between days 22 and 80 was evident between herds. Mean herd level percentage was 65% but ranged from 45 to 90%. Similar when analysed by calving pattern, a large variance in proportion of animals served over this time was seen with 55% and 70% of first service being recorded within this time for seasonal and all year-round calving herds respectively. This highlights the difference in approach to fertility management which exists between these systems and the difficulty associated with the interpretation of data collected across multiple production systems. In addition to this, there exists the potential for the use of exogenous hormones to influence both outcomes of interest analysed. While the use of fixed time insemination with our dataset appears to be limited, this remains a major limitation of our study. Similarly, our reliance on farm records relating to insemination and pregnancy diagnosis event must be considered in the interpretation of our findings. Finally, as an assessment of reproductive performance, the detection of oestrus is reliant on both the resumption of cyclicity, and the expression of heat with sufficient intensity and duration to allow detection. As opposed to the use of serum progesterone or ovarian ultrasound examination for the assessment of cyclicity, our approach may overestimate the prevalence of anoestrus due to its failure to detect animals expressing silent oestrus (Gautam, 2023).

5.5 Conclusion

This is the first study to assess the relationship between data collected by AMS in days 1-21 post-partum and subsequent reproductive performance. Production and behaviour data collected over this time demonstrated significant association with expression of oestrus in days 22-65 and conception to first insemination over days 22-80. Our findings relating to milk production, energy balance and udder health agree with reports utilising traditional means of transition cow monitoring. This demonstrates the potential AMS data holds as an alternative means of assessing transition success. Further research is required to assess the accuracy with which this data may predict reproductive performance within a TMP and the value this may provide when used in tandem with a targeted reproductive management strategy.

Chapter 6 Predictive Models for the Implementation of Targeted Reproductive Management in Multiparous Cows on Automatic Milking Systems

6.1 Introduction

Targeted reproductive management (TRM) aims to improve the fertility efficiency of the dairy herd by applying bespoke group-level management strategies based on expected reproductive performance (Giordano et al., 2022). This is achieved through a three-step process. First, the development of models for the prediction of reproductive performance, second the classification of animals into groups based on these predictions and finally, the implementation of targeted management strategies designed to optimise reproduction. Examples of its application include the targeted use of reproductive hormones in animals at reduced risk of oestrus expression in early lactation (Rial et al. 2022; Gonzalez et al. 2023) and the preferential use of sexed semen in animals deemed highly likely to conceive to artificial insemination (Berry, 2021). Models capable of predicting performance in these areas would be of value in the implementation of TRM.

The influence of the transition period on the subsequent fertility of the dairy cow is well documented (Walsh et al., 2011, Chapinal et al. 2012). The success with which the cow adapts to the stressors of early lactation has a demonstrated effect on expression of oestrus (Vercouteren et al. 2015; Banuelos and Stevenson 2021) and risk of conception (Elkjær et al. 2013; Caixeta et al. 2017; Mohtashamipour et al. 2020). As the adoption of sensor technology on commercial dairy farms continues to increase, so too does the opportunity to harvest data reflective of the cow's physiological status during transition. These data may prove to have utility for the development of the predictive models which form the basis of TRM.

Dairy farms employing automatic milking systems (AMS) offer a unique opportunity to collate transition cow data for use in such models. A wide range of variables relating to milk quantity, quality, concentrate dispensed and robot visit behaviour are automatically generated from sensors incorporated into the milking robot. In addition to this, auxiliary data sources such as neck mounted accelerometers as well as historical cow-level data are often readily available. The integration of a wide range of data sources in model development has the potential to improve the accuracy of predictions by providing a more detailed representation of each animal, however, an increased quantity of data does not guarantee improved model performance (Berisha et al., 2021). Furthermore, as the number of data sources utilised increases, the ease with which the model can be deployed in a commercial setting is

reduced (Leff et al., 2021). The development of predictive models should therefore aim to balance model accuracy with parsimony by minimising the number of data sources used.

The objective of this study was first, to assess the accuracy with which the likelihood of expression of oestrus and conception to first insemination could be predicted using data collected by AMS from days 1-21 in milk. A second objective was to assess the change in model performance following the addition of two auxiliary data sources.

6.2 Materials and Methods

Forty-six commercial AMS herds from the United Kingdom and Republic of Ireland were enrolled as described in Chapter 2. Criteria for inclusion was the use of a Lely Astronaut Milking Robot (Lely International N.V.) under free flow traffic conditions (Munksgaard et al., 2011), in conjunction with rumination and activity monitoring technology (Lely Qwes-HR collars, Lely International N.V.). Participating herd data from January 2016 to August 2023 was available via Lely's third-party application programming interface. Data relating to milk quantity and quality, the frequency of cow-robot interactions, rumination and activity, as well as reproductive management records were accessed. No information pertaining to the use of oestrus synchronisation or fixed time insemination was available for animals within this dataset. All analysis was carried out using R statistical software (R Core Team 2021).

6.2.1 Data Preparation

Two outcomes of interest were selected for investigation. The first was an Expression of Oestrus or Insemination Event (EOI). An animal was classed as EOI+ where an oestrus event or an insemination event was recorded between days 22 and 65 post-partum. An oestrus event was defined as three consecutive 2-hour periods of increased activity compared with each animal's pre-determined baseline as detected by a neck mounted activity monitor. Records relating to insemination events were obtained from the on-farm management system. The second outcome of interest was Conception to First Insemination (CFI). Animals were classed as CFI+ where they received their first and only insemination between days 22 and 80 post-partum and were subsequently recorded as pregnant on the farm management system.

For each outcome of interest, two datasets were constructed. The first was comprised solely of data provided by the AMS and named the RBT dataset. The second was comprised of AMS data in conjunction with two auxiliary data sources: RBT+ dataset. The aim of this study was to compare the accuracy with which EOI and CFI could be predicted using their respective RBT and RBT+ datasets. To allow the comparison of RBT and RBT+ using identical subjects, cow-lactations were assessed for missingness of data across all three data sources. Only cow-lactations with sufficient data across all three were retained. Following feature engineering, fertility data for all retained cow-lactations were

assessed and an outcome for EOI and CFI assigned. Those without sufficient data to allow determination of their fertility performance were removed. For each of the outcomes EOI and CFI, an RBT and RBT+ dataset were brought forward for model development. This data preparation process is described in detail below and displayed in Figure 6-1.

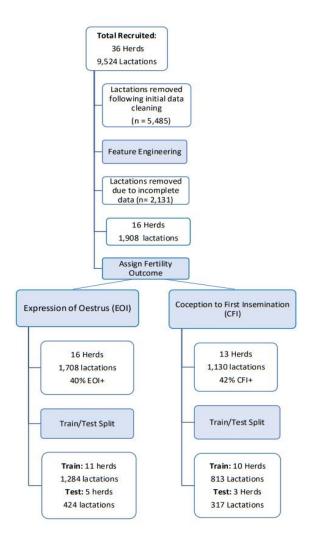


Figure 6-1 Workflow for data cleaning, feature engineering and preparation of the final datasets used for model construction and external validation.

6.2.2 Feature Engineering: AMS Data

The day of calving was designated day zero, data collected by the AMS from days 1-21 was used to engineer 10 features for inclusion in the RBT dataset. Milk quantity was assessed as Mean Milk Yield (kg); the mean of daily milk yields over days 1-21, and Mean Yield Acceleration (%); the mean change in daily milk yield from consecutive days

expressed as a percentage of the first. For example, Mean Yield Acceleration for each cow-lactation was calculated as the change in yield from days 1-2, expressed as a percentage of day 1, averaged with, the change in yield from days 2-3, expressed as a percentage of day 2 yield and so on. Milk Quality was assessed using milk conductivity and constituent data. Two measures of milk conductivity were employed, Mean Conductivity (AU); the mean udder-level conductivity recorded over days 1-21 and Conductivity Alert; the total number of instances where quarter-level conductivity exceeded 80 units. Milk fat and protein indications, as recorded once daily by Lely's MQC (Fadul-Pacheco et al. 2018) were utilised as Mean Fat and Mean Protein; the mean of recorded values across days 1-21 for their respective constituent. A Mean Fat-to-Protein ratio (FPR) was subsequently calculated. Mean Concentrate Dispensed (g) was calculated as the mean grams of concentrate dispensed to each animal by the robot per day. Robot visit behaviour parameters consisted of milking visits and milking refusals (where milking permission is denied due to an animal re-presenting a short time after a previous milking visit). These were averaged over days 1-21 and reported as Mean Milkings and Mean Refusals. To be retained in the final dataset, measurements for each of these ten parameters were required for least 16 of the first 21 days post-partum. Where these were not available the entire cow-lactation was removed.

6.2.3 Feature Engineering: Auxiliary Data Sources

Rumination and activity data derived from neck mounted accelerometers were used to engineer 7 variables (Schirmann et al., 2009). Animals which failed to record complete rumination and activity values for at least 16 of the 21 days analysed were removed from the analysis. Engineered variables were Mean Rumination (Arbitrary Unit, AU); the mean daily rumination recorded, Mean Delta Rumination; the mean change in daily rumination, and Mean Variation Rumination; the variance in Mean Rumination across the 21-day period. These three metrics were also calculated for data relating to daily activity. In addition, the sum of heats recorded by the neck mounted accelerometers over days 1-21 post-partum was assessed as the Transition Heat Count.

Cow-level historical data as recorded by the on-farm management system were used to engineer 7 variables. For each cow-lactation, records describing milk quantity, quality and fertility performance for the prior lactation were assessed. Milk yield in the prior lactation was assessed as PV-Mean Milk Yield; Daily milk yield as averaged across the entire lactation, PV-Transition Yield; mean milk yield for the first 30 days of the previous lactation and PV-Yield at Dry; mean milk yield for the last 10 days prior to drying off. PV-Dry Conductivity represented the conductivity of milk in the 10 days before dry-off. The days in milk at dry-off and the number of days dry were assessed as PV-DO and PV-DD respectively. Finally, days in milk at conception in the prior lactation as well as age at first calving were assessed as PV-DC and AFC

respectively. The necessity of a prior lactation for the assessment of these variables resulted in the removal of all primiparous animals from the dataset. Prior to modelling all numeric variables were centred by subtracting the column means of each variable from their corresponding columns and scaled by dividing each variable by their standard deviations (van den Berg et al., 2006).

Following feature engineering, an EOI and CFI status was assigned to retained cow-lactations. For each outcome of interest, animals for which no definitive status could be established (i.e., EOI+ or EOI- and CFI+ or CFI-) were removed. In the case of EOI, animals without an oestrus or insemination event which also failed to log complete activity records for days 22-65 were removed. For example, an animal for which no oestrus or insemination event was recorded but, for whom a complete activity record was not available was removed from analysis as we could not rule out the occurrence of oestrus on the days for which activity records were absent. In the case of CFI, animals which did not receive a first insemination between 22 and 80 DIM and those inseminated after the end of our observation window were removed. Following assignment of EOI and CFI status, herds with less than 50 cow-lactations available for each outcome were removed. Thereafter, the datasets for both EOI and CFI were split into RBT and RBT+, comprised of 10 and 25 variables respectively. The former was comprised solely of data provided by the AMS, the latter comprised of AMS data in conjunction with both auxiliary data sources. These four datasets were brought forward for model construction. The EOI and CFI datasets contained 1,708 multiparous lactations from 16 herds, and 1,130 multiparous lactations from 13 herds respectively.

6.2.4 Model Construction and Evaluation

To assess the accuracy with which the RBT dataset could predict the likelihood of both EOI and CFI, predictive models for each outcome of interest were constructed and externally validated using the procedure described below. The dataset was split randomly by herd into a train and test dataset. All features within the training dataset were offered to a random forest, neural network and support vector machine models and classification accuracy assessed over five-fold cross validation repeated 10 times. Random forest, which achieved the highest AUC-ROC of all evaluated models was brought forward for further investigation. A random forest recursive feature elimination (RFE) model (Section 2.5.1) was used in feature selection. The optimal subset of variables was assessed by the effect of their inclusion on the area under the receiver operator curve (AUC-ROC) (Kuhn, 2018). Retained variables were selected based on examination of recursive feature elimination plots generated by the VarImp function in the CARET package (Kuhn, 2008) with the goal of balancing parsimony and accuracy. Those retained were brought forward to a final random forest model (Breiman, 2001) trained to maximise AUC-ROC over 5-fold cross validation repeated 10 times. The scaled variable importance for features retained in the final model was assessed using the VarImp

function. This model building procedure was repeated using the RBT+ dataset.

The final predictive models EOI-RBT, EOI-RBT+, CFI-RBT and CFI-RBT+ were evaluated by their predictive performance using the test dataset. Model performance was assessed using classification accuracy, sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV) and AUC-ROC. Within each outcome of interest, the classification accuracy for the RBT and RBT+ models were compared using McNemar's test (Dietterich, 1997). The null hypothesis was that no difference in classification accuracy exists between the RBT and RBT+ models, a significance level of 0.1 was employed. To investigate the accuracy with which these models might be applied on farm, test dataset predictions were ranked by probability within their respective farms and segregated into quartiles (Q). Those in Q1 represented animals least likely within their respective herds to record a positive outcome (an oestrus or insemination event in the case of EOI, conception to first insemination in the case of CFI). Those in Q4 represented the most likely. Of particular interest in our study were Q1 and Q4 as they represented key groups which may be selected for targeted reproductive management. For example, animals classified as Q1 for EOI may receive blanket hormone treatment while those in Q4 may be observed for natural oestrus without hormonal intervention. Similarly, those in Q4 for CFI may be selected for insemination with sexed semen while those in Q1 may receive beef semen. Model calibration was assessed via calibration plots following the binning of model-predicted probabilities into 10 equidistant bins. Furthermore, to allow direct comparison between models expected calibration error (ECE) (Guo et al., 2017) was calculated.

6.3 Results

Complete or partial records relating to 9,524 lactations from 36 recruited herds were assessed. Initial data cleaning involved the removal of primiparous lactations, those which failed to reach 21 days post-partum and those which failed to record any corresponding milk quality, rumination, or activity data. A total of 4,039 multiparous lactations from 34 herds were brought forward for feature engineering. Incomplete milk quality, rumination, or activity data led to the removal of 2,131 lactations. From the remaining 1,908 lactations, those without corresponding fertility records and those originating from herds with less than 50 cow-lactations available for analysis were removed (Figure 6-1). Descriptive statistics for herds retained in the final datasets are presented in Table 6-1.

Table 6-1 General descriptive statistics for herds which contributed cowlactations to EOI and CFI datasets.

| Variable | No. [range] |
|-------------------------------------|-----------------------|
| Total No. of Herds | 16 |
| Calving Pattern | |
| All Year-Round Calving, | 13 |
| Spring Calving | 3 |
| Geographical Region | |
| England | 9 |
| Republic of Ireland | 3 |
| Northern Ireland | 3 |
| Wales | 1 |
| Mean No. of Milking Cows/Herd | 215 [57 - 419] |
| Mean No. of AMS Units/Herd | 3 [2-7] |
| Mean 305 Day Yield – Herd Level | 8438kg [5216-12825kg] |
| Mean % of animals inseminated by 80 | 74% [46–95%] |
| DIM – Herd Level | |

A total of 1,708 cow-lactations from 16 herds formed the final EOI dataset of which 40% were classed as EOI+. The herd-level mean DIM at which an EOI event was recorded was 51, ranging from 45-55 DIM. The final CFI dataset was comprised of 1,130 cow-lactations from 13 herds of which 42% were classed as CFI+. The herd-level mean DIM at first insemination was 61, ranging from 54 to 70 DIM. Descriptive statistics for the cow-lactations retained in the final EOI and CFI datasets are presented in Table 6-2 and Table 6-3 respectively. Frequency distributions for EOI and CFI events are presented in Figure 6-2 and Figure 6-3 respectively. Descriptive statistics for variables included in the RBT and RBT+ datasets are presented in Table 6-4 and Table 6-5 respectively.

Table 6-2 Descriptive statistics for cow-lactations retained in the final EOI and EOI+ random forest models for the prediction of expression of oestrus between DIM 22 and 65.

| Variable | No. [range] |
|---|----------------|
| No. of Cow- Lactations | 1,708 |
| No. of Farms | 16 |
| Calving Pattern | |
| All Year-Round Calving | 13 |
| Spring Calving | 3 |
| Mean No. of Cow – Lactations/Herd | 106 [52-352] |
| Mean Peak Yield ¹ – Herd Level | 54kg [38-69kg] |
| Expression of Oestrus (EOI+) | |
| Total | 40% |
| Parity 2 | 45% |
| Parity 3+ | 36% |
| Mean % of EOI +Animals – Herd Level | 42% [20–73%] |
| Mean DIM at EOI+ event – Herd Level | 51 [45-55] |

¹ Calculated using maximum single day yield recorded in the first 100 days in milk. DIM = Days in milk

Table 6-3 Descriptive statistics for cow-lactations retained in the final CFI and CFI+ random forest models for the prediction of conception to first insemination between DIM 22 and 80.

| Variable | No. [range] |
|--|--------------|
| No. of Cow- Lactations | 1,130 |
| No. of Farms | 13 |
| Calving Pattern | |
| All Year-Round Calving | 10 |
| Spring Calving | 3 |
| Mean No. of Cow-Lactations/Herd | 87 [30–219] |
| Mean Peak Yield ¹ – Herd Level | 54kg [39– |
| | 69kg] |
| Conception Rate to First Insemination | |
| Total | 42% |
| Parity 2 | 43% |
| Parity 3+ | 41% |
| Mean Conception Rate to first insemination | 46% [21–69%] |
| Herd Level | |
| Mean DIM at first insemination – Herd | 61 [54-70] |
| Level | |

¹ Calculated using maximum single day yield recorded in the first 100 days in milk. DIM = Days in milk

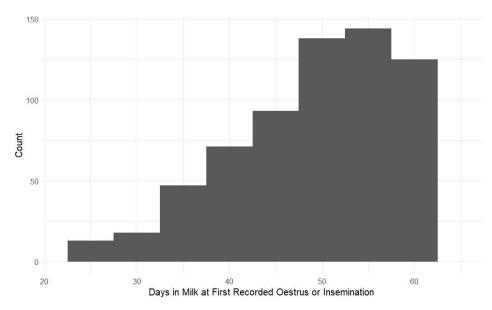


Figure 6-2 Frequency distribution for days in milk at EOI event (oestrus detection or insemination) within the final EOI dataset.

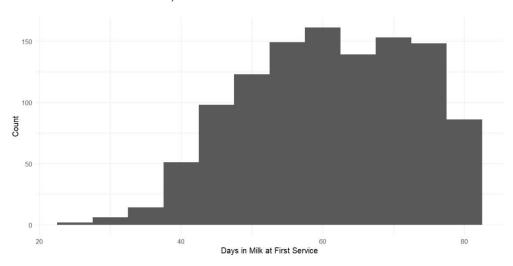


Figure 6-3 Frequency distribution for days in milk at first insemination within the final CFI dataset.

Table 6-4 Variables analysed within the RBT datasets

| | EOI Dataset | | CFI Dat | aset |
|--------------------------------|-------------|------|---------|------|
| Variable | Median | IQR | Median | IQR |
| Mean Milk Yield (kg) | 38.8 | 10.8 | 38.0 | 11.7 |
| Mean Yield Acceleration (%) | 3.9 | 1.8 | 3.8 | 1.8 |
| Mean Conductivity (AU) | 69.6 | 3.9 | 69.7 | 4.0 |
| Conductivity Alert | 0 | 2.0 | 0 | 2.0 |
| Mean Milk Fat-to-Protein Ratio | 1.17 | 0.2 | 1.23 | 0.25 |
| Mean Protein (AU) | 3.72 | 0.32 | 3.71 | 0.36 |
| Mean Fat (AU) | 4.87 | 8.0 | 4.56 | 0.84 |
| Mean Concentrate Dispensed (g) | 5392 | 817 | 5301 | 1073 |
| Mean Refusals | 2.9 | 4.2 | 3.0 | 4.5 |
| Mean Milkings | 3.57 | 0.9 | 3.5 | 0.9 |

IQR= Inter Quartile Range

Table 6-5 Additional variables analysed within the RBT+ Datasets

| EOI Dataset | | CFI Dataset | |
|-------------|--|--|---|
| Median | IQR | Median | IQR |
| 559.23 | 88.7 | 554.33 | 90.4 |
| 0.5 | 1.7 | -0.05 | 1.6 |
| 2968 | 4024 | 2801 | 4032 |
| 40 | 8.4 | 39.7 | 8.2 |
| -0.92 | 1.5 | -0.93 | 1.4 |
| 14.95 | 28.2 | 13.2 | 25.0 |
| 0 | 1.0 | 0 | 1.0 |
| 34.16 | 13.1 | 32.9 | 12.9 |
| 33.12 | 15.3 | 33.1 | 14.7 |
| 21.79 | 10.5 | 19.4 | 11.9 |
| 365 | 55.0 | 368 | 60.0 |
| 54 | 14.0 | 54 | 16.0 |
| 0 | 0.1 | 0 | 0.1 |
| 1427 | 715 | 1460 | 757 |
| | Median 559.23 0.5 2968 40 -0.92 14.95 0 34.16 33.12 21.79 365 54 0 | Median IQR 559.23 88.7 0.5 1.7 2968 4024 40 8.4 -0.92 1.5 14.95 28.2 0 1.0 34.16 13.1 33.12 15.3 21.79 10.5 365 55.0 54 14.0 0 0.1 | Median IQR Median 559.23 88.7 554.33 0.5 1.7 -0.05 2968 4024 2801 40 8.4 39.7 -0.92 1.5 -0.93 14.95 28.2 13.2 0 1.0 0 34.16 13.1 32.9 33.12 15.3 33.1 21.79 10.5 19.4 365 55.0 368 54 14.0 54 0 0.1 0 |

IQR= Inter Quartile Range, PV= Previous Lactation, DIM = Days in Milk. ¹DIM at dry-off in prior lactation. ²Days dry in prior lactation, ³ Milk conductivity at dry-off in prior lactation, ⁴ Age at first calving.

6.3.1 Expression of Oestrus and Insemination Events

The EOI dataset was divided into a training and test dataset of 1,284 lactations from 11 herds and 424 lactations from 5 herds respectively. Following assessment of the recursive feature elimination plot, five

variables, the combination of which resulted in the highest AUC-ROC were retained in the final EOI-RBT model. These were, Mean Milk Yield, Mean Concentrate Dispensed, Mean Protein, Mean Refusals and Mean Conductivity. The scaled variable importance for each is presented in Table 6-6. Animals were classified with an AUC-ROC of 0.6 (Figure 6-4) sensitivity of 38%, specificity of 79%, PPV, NPV, and accuracy of 60% (Table 6-7). An expected calibration error of 0.10 was observed.

Table 6-6 Scaled Variable importance for the EOI-RBT Model, a random forest model for the prediction of Expression of Oestrus from DIM 22-65 (EOI) utilising AMS data exclusively

| Variable | Variable Importance Score ¹ |
|----------------------------|--|
| Mean Concentrate Dispensed | 100 |
| Mean Milk Yield | 90 |
| Mean Conductivity | 51 |
| Mean Refusals | 26 |
| Mean Protein | 0 |

¹Variable importance score as calculated by the VarImp function of the CARET package.

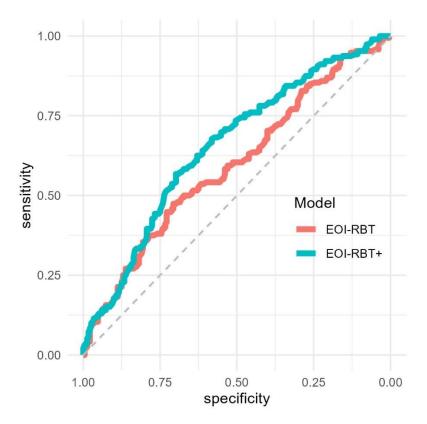


Figure 6-4 ROC for EOI-RBT and EOI-RBT+ random forest models for the predictions of expression of oestrus between days 22 and 65 post-partum as evaluated on the test dataset

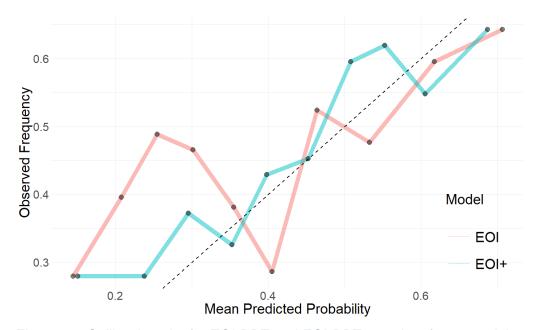


Figure 6-5 Calibration plot for EOI-RBT and EOI-RBT+ random forest models for the predicted probability of expression of oestrus or insemination (EOI) between DIM 22-60 DIM as evaluated on the test dataset

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Table 6-7 Classification performance of all random forest models built utilising AMS data exclusively (RBT) and AMS data in conjunction with auxiliary data (RBT+) for the prediction of Expression of Oestrus or Insemination from DIM 22-65 (EOI) and Conception to First Insemination from DIM 22-80 (CFI)

| | Model | | | |
|-----------------|----------|-----------|----------|-----------|
| | EOI- RBT | EOI- RBT+ | CFI- RBT | CFI- RBT+ |
| AUC-ROC | 0.60 | 0.65 | 0.56 | 0.62 |
| Accuracy | 60 | 57 | 62 | 59 |
| Sensitivity | 38 | 15 | 25 | 25 |
| Specificity | 79 | 91 | 88 | 80 |
| PPV | 60 | 59 | 53 | 44 |
| NPV | 60 | 57 | 64 | 63 |
| McNemar P-value | 0.9 | | 0.5 | |

DIM= Days in Milk, AUC-ROC= Area under the receiver operator curve, PPV = Positive predictive value, NPV = negative predictive value.

In the final EOI-RBT+ model, 8 variables including at least one from each of the 3 data sources were retained following RFE; Mean Concentrate Dispensed, Mean Variation Activity, PV-Mean Milk Yield, Mean Activity, Mean Milk Yield, AFC, Mean Protein, and PV-DC. Scaled importance for all variables retained in the final model is presented in Table 6-8. Animals were classified with an AUC-ROC of 0.65, sensitivity of 15%, specificity of 91%, PPV of 59%, NPV and accuracy of 57%. An ECE of 0.056 was observed.

Table 6-8 Scaled variable importance for the EOI-RBT+ Model, a random forest model for the prediction of Expression of Oestrus from DIM 22-65 (EOI) utilising AMS and Auxiliary data.

| Variable | Variable Importance Score1 |
|--------------------|----------------------------------|
| Mean Concentrate | 000.01 |
| Dispensed | 100 |
| Variation Activity | 93 |
| PV-Mean Milk Yield | 84 |
| Mean Activity | 82 |
| Mean Milk Yield | 70 |
| AFC | 60 |
| Mean Protein | 3 |
| PV-DC2 | 0 |

1 Variable importance score as calculated by the VarImp function of the CARET package. 2 DIM at conception in prior lactation.

Table 6-9 Classification Accuracy (%) per quartile of all random forest models for the prediction of Expression of Oestrus from DIM 22-65 (EOI) and Conception to First Insemination from DIM 22-80 (CFI) as assessed on the test dataset.

| Model | Q1 | Q2 | Q3 | Q4 |
|-----------|----|----|----|----|
| EOI- RBT | 61 | 55 | 53 | 60 |
| EOI- RBT+ | 72 | 62 | 45 | 48 |
| CFI- RBT | 63 | 60 | 70 | 54 |
| CFI- RBT+ | 70 | 72 | 49 | 42 |

1Quartiles based on the within farm predicted likelihood of a positive outcome. Q1 representing the least Q4 the most likely. DIM = Days in milk.

6.3.2 Conception to First Insemination

The CFI dataset was divided into a training set of 813 lactations from 10 herds and test set of 317 lactations from 3 herds. Seven variables were retained in the CFI-RBT model following RFE: Mean Delta Yield, Mean Concentrate Dispensed, Mean Conductivity, Mean Milk Yield, Mean Fat-to-Protein Ratio, Mean Protein and Mean Milkings. Scaled variable importance for each is presented in Table 6-10. Animals which conceived to first insemination were classified with an AUC-ROC of 0.56 (Figure 6-6), sensitivity of 25%, specificity of 86%, a PPV of 53%, NPV of 64% and accuracy of 62% (Table 6-7). An ECE of 0.095 was observed.

Table 6-10 Scaled variable importance for the CFI-RBT Model, a random forest model predicting conception to first insemination from DIM 22-80 utilising AMS data exclusively (CFI-RBT)

| | Variable |
|--------------------|--------------------|
| | importance |
| Variable | score ¹ |
| Mean Delta Yield | 100 |
| Mean Concentrate | |
| Dispensed | 94 |
| Mean Conductivity | 69 |
| Mean Milk Yield | 57 |
| Mean Milk FPR | 43 |
| Mean Protein | 11 |
| Mean Milkings | 0 |
| 4) / 1 1 1 1 | , |

¹Variable importance score as calculated by the VarImp function of the CARET package.

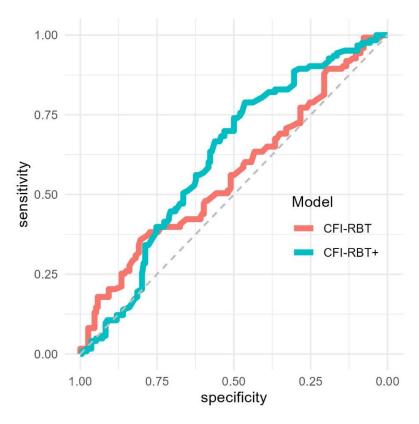


Figure 6-6 ROC for CFI-RBT and CFI-RBT+ random forest models for the prediction of conception to first insemination (CFI), between days 22 and 80 post-partum as evaluated on the test dataset

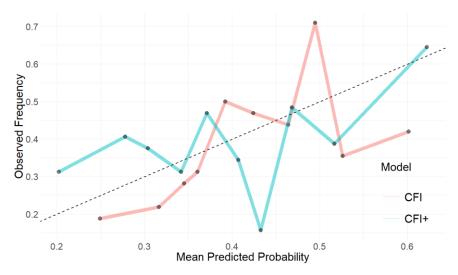


Figure 6-7 Calibration plot for the predicted probability of conception to first insemination (CFI), between days 22 and 80 post-partum as evaluated on the test dataset.

Eleven variables were retained in the final CFI-RBT+ model: Mean Conductivity, Mean Concentrate Dispensed, Mean Delta Yield, Mean Milkings, Mean Fat-to-Protein Ratio, Mean Protein, Mean Rumination, Mean Delta Rumination, PV-DO, PV-Dry Yield and PV-Mean Milk Yield. Scaled variable importance for each is presented in Table 6-11 Animals were classified with an AUC-ROC of 0.62 Figure 6-6, sensitivity of 25%, specificity of 80%, PPV of 44%, NPV of 63% and accuracy of 59% (Table 6-7). When the accuracy of RBT and RBT+ models were compared using McNemar's test, a *P*-value of 0.5 was obtained and the null hypothesis accepted. The accuracy with which RBT and RBT+ models predicted animals within Q1 was 63% and 70% respectively. Those in Q4 were classified with an accuracy of 42% by RBT and 54% by RBT+ (Table 6-9). An ECE of 0.10 was observed.

Table 6-11 Scaled variable importance for the CFI-RBT+ Model, a random forest model predicting conception to first insemination from DIM 22-80 utilising AMS and auxiliary data.

| Variable | Variable importance score ¹ |
|----------------------------|--|
| Mean Concentrate Dispensed | 100 |
| Mean Delta Yield | 75 |
| PV-Mean Milk Yield | 67 |
| Mean Rumination | 63 |
| Mean Conductivity | 37 |
| Mean Milk FPR | 37 |
| Mean Delta Rumination | 37 |
| PV-Yield at Dry | 25 |
| PV-DO2 | 19 |
| Mean Protein | 2 |
| Mean Milkings | 0 |

¹Variable importance score as calculated by the VarImp function of the CARET package. ² DIM at dry-off in prior lactation.

6.4 Discussion

This is the first study to report the utility of transition cow data collected by AMS for the prediction of reproductive performance. By comparing these models with those utilising two additional data sources, we also report the marginal utility provided by rumination and activity data as well as historical cow-level data. Though the performance described is comparable with previously reported models, their current utility for the implementation of TRM is limited by poor classification accuracy within key groups. Of note within this study is the failure of the addition of auxiliary data sources to increase the accuracy of prediction over models built using AMS data alone.

The early resumption of cyclicity post-partum is key to subsequent reproductive performance. Approximately 25% of cows are reported to be acyclic by day 50-60 post-partum (Walsh et al. 2007; Santos et al. 2009; Pinedo et al. 2020). Animals suffering this delayed resumption of cyclicity experience decreased rates of insemination (Borchardt et al. 2021) and conception (Galvão et al. 2010) in early lactation.

The detection of oestrus during the voluntary waiting period is a commonly used proxy for the assessment of ovarian cyclicity (Fricke et al. 2014; Rial et al. 2022). We chose to assess cyclicity using oestrus or insemination events between days 22 and 65 post-partum. Within a TRM strategy, the ability to predict the likelihood of EOI may be used to minimise the number of acyclic animals at the start of their breeding period through targeted pre-breeding screening or hormone treatment (Rhodes et al., 2003). EOI-RBT and EOI-RBT+ models classified animals with an accuracy of 60% and 57% respectively, demonstrating

poor model discrimination. When assessed by quartile, the accuracy with which the RBT+ model classified animals least likely to record an oestrus event (Q1) was moderate at 72%. However, this model failed to classify those in Q3 or Q4 with an accuracy above 50%, limiting its ability to reliably inform management decisions. Assessment of model calibration (Figure 6-5) revealed moderate calibration across the range of range of observed frequencies within the EOI-RBT+ model. This stands in contrast to the large calibration error seen in the EOI model, particularly in sub-groups with an observed frequence of oestrus or insemination below 50%. This is reflected in the ECE returned for both models with the EOI- RBT model recording an expected error twice that of the EOI-RBT+ model.

The ability to predict the likelihood of conception to first insemination may facilitate the selective breeding of animals, such as the targeted use of sexed semen in animals with a high probability of conception. We investigated the likelihood of conception to first insemination within the first 80 days post-partum. This incorporates the first 6 weeks of the breeding season assuming a 42-day voluntary waiting period, a time where the targeted use of sexed semen is commonly advocated (Butler et al., 2014). CFI models demonstrated poor discriminative power. though the accuracy of classification for Q1 and Q2 within the CFI-RBT+ model was moderate, 70 and 72% respectively. However, both CFI models demonstrated a lack of predictive power for Q4, limiting the confidence with which this highly fertile sub-group could be targeted with sexed semen. Examination of model calibration demonstrated similar shortcoming. As presented in Figure 6-7, calibration was found to be poor across the sub-groups investigated for both the CFI-RBT and CFI- RBT+ models. This was further demonstrated by both models returned very similar ECE.

The utility of transition cow data for the prediction of subsequent fertility within our dataset is limited, though the accuracy of CFI models is comparable with models previously reported (Hempstalk et al., 2015; Shahinfar et al., 2014, Barden et al. 2024). While the potential value which TRM strategies may provide cannot be measured solely in the accuracy of their predictive models (Harris, 2017), they do serve as the foundation of their utility. For the on-farm implementation of TRM relating to EOI and CFI, the accuracy of classification within the groups at both extremes of reproductive performance is key. None of the four models investigated here demonstrated satisfactory accuracy across both Q1 and Q4 (Table 6-9). While we believe these models have demonstrated potential for the prediction of fertility, improved classification performance will be required prior to their implementation within a targeted reproductive management strategy.

Developing a more detailed representation of each animal through the integration of additional data sources is often cited as a potential means of improving model performance (Shahinfar et al., 2014; Dallago et al., 2019; Ha et al., 2022). We investigated the change in performance following the addition of two auxiliary sources chosen based on their demonstrated ability to reflect transition health and influence fertility

(Eastham et al., 2018; Stevenson et al., 2020; Borchardt et al., 2021). In the final EOI-RBT+ model, data from auxiliary sources comprised 5 of the 8 variables retained following recursive feature elimination (Table 6-8). Similarly, in the final CFI+ model, data from auxiliary sources accounted for 5 of the 11 variables retained (Table 6-11). Despite this, no statistically significant difference was detected in the classification accuracy of the RBT and RBT+ models. The retention of these variables within the RBT+ models highlight the care which must be applied to feature selection and preserve model parsimony as the number of available data sources continues to expand.

While the increasing adoption of technology on commercial dairy farms has the potential to improve the accuracy of predictive models, this must be balanced with the limitations it imposes on model development and deployment. For example, rumination and activity monitoring, while commonly employed by farms utilising AMS, is not ubiquitous. Furthermore, as this technology is supplied by third parties, issues relating to the uniformity of sensors and software across farms can complicate data integration and increase data missingness. As seen within our study, incorporating such data sources may reduce the volume of data available for model development. Furthermore, it introduces additional data requirements for farms wishing to employ such models, limiting its deployment. The integration of additional data sources must therefore be justified by substantial improvement in model performance.

All data utilised within this study was accessed via Lely's third-party API. The use of data which is widely collected and remotely accessible across Lely AMS significantly increases the ease with which predictive models may be deployed commercially. However, the nature of this dataset imposed several limitations on this study. One such limitation is the absence of data relating to individual herd management practices. in particular the use of exogenous hormones and fixed time artificial insemination. In the case of the CFI models, the use of such treatments has the potential to influence the probability of conception (Fricke et al., 2022) and hence bias our predictions. The use of fixed time artificial insemination is not commonly practiced in the UK or Republic of Ireland, particularly in the first 80 days post calving. However, to assess the degree to which such practices may have been applied within our dataset, insemination records were examined as previously described by Barden et al. (2024). In brief, for each herd, the proportion of inseminations on each day, of each week were calculated. Thereafter, the binomial standard deviation for a uniformly distributed proportion (i.e., 1/7) for all inseminations within each week was calculated. Any day for which the proportion of inseminations reported was in excess of 2 standard deviations for the week was identified and all inseminations on that day marked as potentially fixed time inseminations. Within the final CFI dataset 8% of inseminations were identified as potential fixed time inseminations. This indicates that the practice of herd-level fixed time insemination is unlikely to have been used extensively within this dataset. However, we cannot conclusively state that insemination

events analysed were not influenced by such treatments and our results must be interpreted in-light of this limitation. A further limitation is our reliance on farm-level recording of insemination events and pregnancy diagnosis. It was not possible for the research team to assess the means and consistency by which these events were recorded across farms, or the effect between-farm variance may have had on our results. As these events form key aspects of our analysis, this represents a major limitation within our study. This study is the first to demonstrate the potential utility of AMS data collected during transition to develop a generalisable predictive model for subsequent reproductive performance. However, due to the small number of herds utilised and the limited number of lactations assessed within each herd. these results must be interpreted with caution. While we sought to incorporate a range of farming systems commonly found on AMS within the UK and Republic of Ireland, validation of these results utilising a larger and more diverse dataset is required to more confidently assess the generalisability of these models.

For the implementation of TRM, parsimonious predictive models which reliably deliver good to excellent classification accuracy in a wide range of commercial settings are required. As the range of novel data sources expands, the investigation of their ability to improve model performance will be key to progress within this field. Recent studies have demonstrated the potential of data sources such as genomics (Rial et al., 2024) and the in-line assessment of progesterone (Blavy et al. 2018) for the prediction of reproduction. Where such data sources were readily available for integration with AMS data, they may hold greater potential for predictive performance than the auxiliary data sources investigated here. However, future studies should not neglect the exploration of alternative methods for the improvement of model performance. For example, the clustering of farms based on environmental and management conditions may provide an opportunity to increase performance by tailoring models to each farm's specific circumstance (Ng et al., 2015). This approach has been employed on farms utilising AMS to facilitate targeted management recommendations (Tremblay et al., 2016). However, its utility for the development of predictive models remains unexplored. Such methods may reduce the need for the integration of a large number of data sources and facilitate the implementation of TRM on a broader scale across automatic milking systems.

6.5 Conclusions

These results demonstrate the limited accuracy with which AMS data collected over days 1-21 post-partum can predict subsequent reproductive performance. Within a transition monitoring program, the models investigated failed to accurately classify animals within key performance groups thus limiting the confidence with which TRM strategies could be employed. The failure of the addition of auxiliary data sources to significantly improve model accuracy highlights the care with which feature selection should be undertaken to ensure the

development of parsimonious models. Future studies should focus on the incorporation of both novel data sources and novel analytical techniques in the pursuit of TMPs with improved predictive power while maintaining parsimony and ease of on farm deployment.

Chapter 7 Factors Associated with the Risk of Removal in Early Lactation for Dairy Cows in Automatic Milking Systems

7.1 Introduction

The involuntary removal of early lactation dairy cows negatively affects the economic and social sustainability of the dairy industry. Substantial direct and indirect costs are associated with such removals, including animal disposal, replacement, and loss of milk sales (Orpin et al., 2010). Furthermore, as removals during this period are largely the result of transition cow disease (Dechow et al., 2008; De Vries et al., 2010), they represent a welfare concern for dairy producers and consumers alike (Alonso et al., 2020). In the United Kingdom, the incidence of removals in early lactation has been adopted as a key indicator of dairy cow health and welfare by the Royal Society for Prevention of Cruelty to Animals (RSPCA, 2023). Across 500 UK dairy herds surveyed in 2022, the median herd reported the removal of 5% of milking cows by 100 days in milk (DIM) (Hanks et al., 2023). This rate of removal remains largely unchanged from that reported 10 years ago (Hanks et al., 2012), indicating that despite advancements in transition cow management (Mezzetti et al., 2021), little progress has been made towards reducing the rate of removals in the first 100 days post-partum.

The initiation of lactation poses a range of challenges to the modern dairy cow. In the days immediately surrounding calving, the cow experiences more significant endocrine changes than at any other point in lactation (Grummer et al., 2004). In addition to this, the demands of milk production leads to a four-fold increase in calcium requirements on the day of calving (Caixeta et al., 2021), while glucose requirements triple by day-4 post-partum (Eicker et al., 2002.). Under modern management conditions, these physiological changes are often compounded by changes in housing, social group and diet. The significance of these challenges is reflected in 30-50% of cows suffering some form of metabolic or infectious disease around the time of calving (Leblanc, 2006), and an increase in the incidence of removals by 100 DIM which peaks in the first 30 days. (Dechow et al., 2008; De Vries et al., 2010; Pinedo et al., 2014).

Investigation of the relationship between physiological status in the days immediately post-partum and the subsequent risk of removal has been undertaken to better understand these losses. When sampled within days 1-3 in milk, serum metabolic markers have demonstrated an association with the subsequent risk of removal. Venjakob, (2018), found the hazard of removal by 60 DIM to be 1.69 times greater in cows suffering sub-clinical hypocalcaemia when compared with normocalcaemic cows. A similar effect of calcium concentration was demonstrated by Menta (2021), who reported a relative risk of culling by

60 DIM of 2.93 for animals diagnosed with sub-clinical hypocalcaemia within 3 DIM. In the same study, an association between the risk of removal and metabolic indicators of negative energy balance was also reported. Beta-hydroxybutyrate and free fatty acid concentration returned a positive association with risk of cull which tended toward significance (*P*-values of 0.05 & 0.08 respectively). These reports demonstrate an association between the success with which dairy cows respond to the metabolic challenges of calving and their subsequent risk of removal in early lactation. Monitoring of physiological status during early lactation may therefore allow for the early identification of cows at high risk of removal, potentially facilitating early intervention and loss prevention (Seifi et al., 2011; Roberts et al., 2012).

The monitoring capabilities of automatic milking systems (AMS) may offer the means to achieve this through an automated transition cow monitoring program (TMP). Variables recorded by AMS, such as milk quantity (Mansell, 2003), quality (Gross et al., 2019), as well as robot visit behaviour (King et al., 2017) have been previously demonstrated to reflect the physiological status of the dairy cow. In contrast to the labour-intensive analysis of serum metabolic indicators, these variables are collected automatically from the point of calving and represent a more convenient method of monitoring. However, their association with the subsequent risk of removal in early lactation has not been investigated. Such analysis would provide insight into the production and behaviour traits of cows at high risk of removal within the first 100 days. Furthermore, if these parameters demonstrate utility for the prediction of removals, they represent an opportunity to reduce the impact of early removal through pro-active management of high-risk animals.

The primary objective of this study was to test the hypothesis that AMS data from 1-3 DIM is statistically associated with the risk of removal from the herd by 100 DIM. A secondary objective was to assess the utility of this data for the prediction of removal using an externally validated machine learning model.

7.2 Materials and Methods

7.2.1 Study Population

Forty-six commercial dairy farms from the United Kingdom and Republic of Ireland were recruited as described in Chapter 2. Criteria for inclusion was the use of Lely Astronaut milking robots (Lely International N.V.) under free-flow traffic conditions (Munksgaard et al., 2011). Data describing milk quantity and quality as well as robot visit behaviour from 2016 to 2023 were accessed via Lely's third-party application programming interface.

7.2.2 Data Analysis

All analysis was conducted using R statistical software (R Core Team, 2021). Prior to feature engineering the initial percentage of the herd removed by DIM 100 (RD100) was assessed. Cows were classed as

RD100 where the final milking visit of the cow's final lactation occurred within the first 100 days post-partum. No information pertaining to the reason for removal was available.

Preparation of the final dataset is displayed in Figure 7-1 and described below. The day of calving was designated day zero. Data from days 1-3 was used to engineer 10 independent variables for use in both inferential and predictive models. Milk quantity was assessed as Mean Milk Yield (kg); the mean of daily milk yields over days 1-3, and Mean Yield Acceleration; the mean of the change in daily milk yield from days 1-2, and 2-3. Milk quality was assessed using milk conductivity and constituent data. Conductivity was assessed as Mean Conductivity; the mean udder-level conductivity of milk recorded over days 1-3 and, Conductivity Alert; the total number of instances where guarter-level conductivity exceeded 80 units. Milk fat and protein indications, as recorded once daily by Lely's MQC (Fadul-Pacheco et al., 2018) were utilised as Mean Fat and Mean Protein: the mean of recorded values across days 1-3 for their respective constituents. A Mean Fat-to-Protein Ratio (FPR) was subsequently calculated. Mean Concentrate Dispensed; the mean grams of concentrate feed dispensed by the robot to each cow per day. Visit behaviour was assessed using milkings and refusals (where milking permission is denied due to a cow re-presenting a short time after a previous milking visit). These were averaged over days 1-3 and reported as Mean Milkings and Mean Refusals. Finally, Parity (1 or 2+) and Calving Pattern (Seasonal or All-Year-Round) were assigned. Following feature engineering, records were assessed for missingness. Cows which failed to record milk quantity and quality data for at least one robot visit for each of the days 1-3 post-partum were removed. Thereafter, herds with less than 50 cow-lactations per parity group were removed. Following establishment of the final dataset the percentage of animals classed as RD100 within each herd was reassessed and compared with the initial incidence of removal in the first 100 DIM.

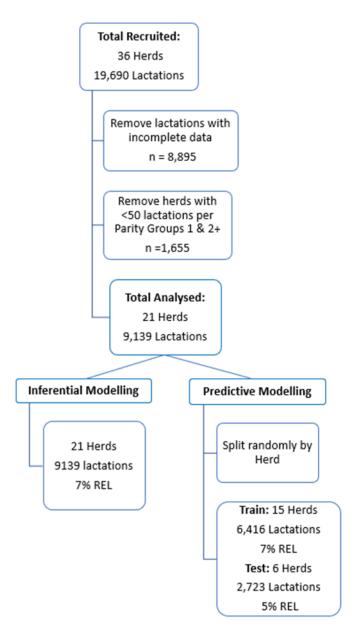


Figure 7-1 Data preparation for inferential and predictive modelling

7.2.3 Inferential Modelling

To explore the relationship between early lactation AMS data and RD100, a mixed-effect logistic model was constructed with herd as a random effect using the Ime4 package (Bates et al., 2015). In addition to the 10 main independent variables described above, seven interaction terms were selected for investigation based on their biological plausibility to influence the risk of removal from the herd. The interaction effect for Mean Milk Yield with FPR, Mean Yield Acceleration, Mean Milkings, Mean Refusals, Mean Concentrate Dispensed and Parity were assessed as well as the interaction between Parity and Mean Yield Acceleration. Prior to modelling, non-linearity was assessed using multivariable regressive splines (Friedman, 1991).

Thereafter, Conductivity Alert which demonstrated a highly right-skewed distribution was transformed using a cubic transformation. All numeric variables were centred by subtracting the column means of each variable from their corresponding columns and scaled by dividing each variable by their standard deviations (van den Berg et al., 2006).

A multivariable mixed-effect model was constructed using a manual backward step procedure (Dohoo et al., 2009). All candidate variables were screened using univariable analysis and brought forward for inclusion in a multivariable model where a P-value of ≤ 0.20 was observed. Models were constructed using the logit link function and included a random intercept to account for the clustering of data at herd-level. Variables were retained in the final model where a P-value of ≤0.05 was observed. Parity, and any variables which formed a significant interaction term were forced into the final model. On completion of the backward step, all main effects which were removed were re-entered to test for significance in the final model. The inclusion of Calving Pattern was investigated as a possible confounding variable. Its inclusion led to a 12% change in coefficient estimate for Mean Protein and was retained. Goodness-of-fit was evaluated by graphical assessment of residuals and marginal and conditional R² values (Nakagawa et al., 2017). Model performance was assessed following internal validation using the area under the receiver operator curve (AUC-ROC), sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) with and without the random effect of

The final multivariable mixed-effects logistical model took the general form:

$$log\left(\frac{P_{ij}}{1 - P_{ij}}\right) = \beta_0 + \beta_1 X_{1ij} + \dots + \beta_n X_{nij} + v_j + \epsilon_{ij}$$
$$[v_j] \sim N(0, \Omega_v)$$
$$[\epsilon_{ij}] \sim N(0, \Omega_e)$$

where the subscript i refers to the ith cow and the subscript j refers to the jth herd; β_0 represents the intercept; $\beta_1 - \beta_n$ represent fixed effects; v_j represents a random effect for Herd (assumed to have a normal distribution of mean = 0, and variance = Ω_v) and ϵ_{ij} represents the residual model error (assumed to have a normal distribution of mean = 0, and variance = Ω_e). Parameter P_{ij} represents the probability of removal by 100 days in milk for the ith cow in the ith herd.

7.2.4 Predictive Modelling

The second objective of our study was to train and externally validate a predictive model for removal from the herd within 100 DIM. Our aim was to assess the utility of AMS data from 1-3 DIM for the prediction of RD100.

To facilitate external validation, the dataset was split randomly by herd into train and test datasets. Herds were first ranked by RD100 percentage and segregated into 3 groups designed to mirror the quartiles reported by Hanks et al. (2023). Herds in Q1 reported a RD100 of <4% and Q4 a RD100 of >7%, the remaining herds were grouped as Q2/3. To ensure balanced representation, two herds from each of these 3 groups were selected randomly to form the test dataset. All remaining herds were allocated to the training dataset. To address class imbalance in the training dataset, up sampling of the minority class was carried out using the ROSE package (Lunardon et al., 2014). All 10 independent variables, in addition to Parity were offered to an extreme gradient boosting model (Chen et al., 2016). This model was trained to maximise AUC-ROC over 5-fold cross validation repeated 10 times. Model hyperparameters were optimised using a random grid search (Kuhn, 2008). Final model evaluation was carried out on the test dataset using AUC-ROC, sensitivity, specificity, PPV and NPV. Model calibration was also assessed via calibration plots and expected calibration error (ECE).

7.3 Results

In total, records relating to 19,690 complete or partial lactations from 36 herds were available for analysis. Incomplete production records led to the removal of 8.895 lactations. A further 1.656 lactations from herds with insufficient lactations per-parity group were removed. The final dataset consisted of 9,139 lactations from twenty-one herds (Figure 7-1, Table 7-1). Within the retained herds, the initial incidence of RD100 (prior to adjustment for missingness), ranged from 1.5% to 13% with a median of 6% on a herd basis. After the removal of cow-lactations with incomplete production and behaviour records, a herd-level minimum, maximum and median RD100 of 2% ,12% and 7% respectively was observed (Table 7-2, Figure 7-2). Herd-level changes in RD100 ranged from -3.5 to 2.2% with a mean of -0.47%. For all twenty-one retained herds, the initial incidence of RD100 and the incidence observed within the final dataset is displayed in Figure 7-3. Within the final dataset the incidence of RD100 was 7% across all lactations with a mean DIM at removal of 43. Descriptive statistics for the independent variables within the final dataset are presented in Table 7-3. To aid interpretability, model outputs are described using the value of one standard deviation within each variable's original unit of measurement.

Table 7-1 Descriptive statistics for herds contributing cow-lactations to the final dataset

| Variable | No. [range] |
|--|------------------------|
| Total No. of Herds | 21 |
| Calving Pattern | |
| All Year-Round Calving | 16 |
| Spring Calving | 5 |
| Geographical Region | |
| England | 10 |
| Republic of Ireland | 5 |
| Northern Ireland | 4 |
| Wales | 1 |
| Scotland | 1 |
| Mean No. of Milking Cows/Herd | 217 [57 - 438] |
| Mean No. of AMS Units/Herd | 3 [2-7] |
| Mean 305 Day Yield – Herd Level | 8431kg [4081–12,825kg] |
| Median Initial RD100 – Herd Level ¹ | 6% [1-13%] |

¹ Median percentage of cow-lactations removed by 100DIM, calculated prior to adjustment for missingness.

Table 7-2 Descriptive statistics for cow-lactations retained in the final mixed-effect logistic and XGBoost model.

| Variable | No. [range] |
|--|----------------|
| No. of Cow- Lactations | 9,139 |
| No. of Farms | 21 |
| Mean No. of Cow-Lactations/Herd | 435 [160–1021] |
| Demographics | |
| Parity 1 | 28% |
| Parity 2+ | 72% |
| Calving Pattern | |
| All Year-Round Calving | 76% |
| Spring Calving | 24% |
| Percentage Removed by 100 DIM | |
| Total | 7% |
| Parity 1 | 2% |
| Parity 2+ | 4% |
| Percentage Removed by 100 DIM – Herd Level | 7% [2-12%] |
| Median DIM at Removal Herd Level | 43 [19-75] |

¹ Calculated using maximum single day yield recorded in the first 100 days in milk. DIM = Days in milk

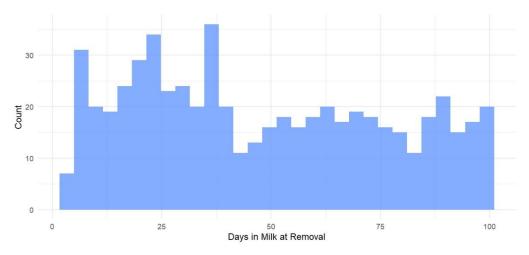


Figure 7-2 Days in Milk at removal for all 9,139 cow-lactations from 21 herds retained in the final dataset

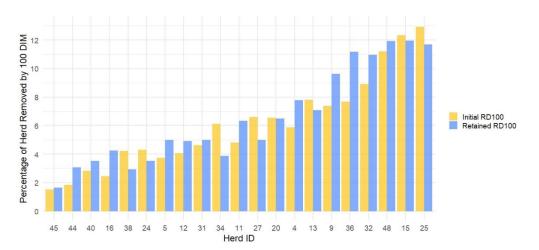


Figure 7-3 Percentage of the milking herd removed in the first 100 days of lactation (RD100) for the 21 herds retained in the final dataset. Initial RD100 (Yellow) represents the percentage removed assessed over all available lactations, Retained RD100 (Blue) represents the percentage removed from all lactations retained in the final dataset following adjustment for missingness

Table 7-3 Descriptive statistics for independent variables assessed in the mixed-effect logistic and XGBoost model.

| Variable | Median | IQR |
|----------------------------|--------|------|
| | | - |
| Mean Milk Yield | 20 | 11 |
| Mean Delta Yield | 16 | 14 |
| Mean Conductivity | 73.25 | 4.5 |
| Conductivity Alert | 0 | 2.0 |
| Mean Fat-to-Protein Ratio | 1.07 | 0.33 |
| Mean Protein | 4.74 | 0.56 |
| Mean Fat | 5.04 | 1.46 |
| Mean Concentrate Dispensed | 3135 | 977 |
| Mean Refusals | 2.0 | 6 |
| Mean Milkings | 2.0 | 1 |

IQR = Interquartile Range

7.3.1 Inferential Modelling

The output of the final multivariable logistic regression model is presented in Table 7-4 and Figure 7-4. Accounting for difference in Parity and Calving Pattern, Mean Milk Yield, Mean Yield Acceleration and Mean Refusals were all negatively associated with the odds of RD100. Each 7kg increase in Mean Milk Yield was associated with a 18% decrease in the odds of removal. An 18% increase in Mean Yield Acceleration returned a 10% decrease in the odds of removal. Mean Refusals returned a 30% decrease in the odds of removal for each additional 0.40 in Mean Refusals recorded.

Three variables were associated with an increased risk of removal. A 0.4% increase in Mean Protein and 0.3% increase in Mean FPR was associated with a 19% and 16% increase in the odds of RD100 respectively. Finally, multiparous cows demonstrated a 75% increase in the odds of removal compared with primiparous cows and animals in seasonal calving systems demonstrated a 48% decrease in the odds of removal compared with those in all-year-round calving systems.

Classification performance for the final multivariable mixed logistic model, with and without the random effect of Herd are presented i

Table 7-5. Following internal validation assessed with the random effect of Herd, the sensitivity and specificity of the final model was 65% and 64% respectively. Without the random effect of Herd, the sensitivity and specificity of the model was reduced to 56% and 57% respectively. The final model explained 13% (conditional R²) of the variation in risk of removal observed with 7% (marginal R²) attributable to the fixed effects.

Table 7-4 Results of the multivariable mixed logistic regression model assessing the association between early lactation AMS production data and the risk of removal from the herd by 100 days in milk. SD= Standard Deviation

| | | Odds | | | |
|-----------------|-------------|-------|--------|-----------------|------|
| Variable | Coefficient | Ratio | 95% CI | <i>P</i> -value | SD |
| | | | 0.03- | | |
| Intercept | -3.1 | 0.04 | 0.06 | < 0.001 | |
| | | | 1.06- | | |
| Mean FPR | 0.15 | 1.16 | 1.27 | 0.001 | 0.27 |
| | | | 1.06- | | |
| Mean Protein | 0.17 | 1.19 | 1.34 | 0.003 | 0.41 |
| | | | 0.62- | | |
| Mean Refusals | -0.36 | 0.70 | 0.78 | < 0.001 | 8.3 |
| | | | 0.73- | | |
| Mean Milk Yield | -0.20 | 0.82 | 0.93 | 0.001 | 7.5 |
| Mean Yield | 0.44 | 0.00 | 0.84- | 0.000 | 47.5 |
| Acceleration | -0.11 | 0.90 | 0.96 | 0.002 | 17.5 |
| Parity 1 | Ref. | - | - | - | - |
| | | | 1.32- | | - |
| Parity 2+ | 0.55 | 1.73 | 2.30 | < 0.001 | |
| Calving Season | | | | | - |
| - AYR | Ref. | - | - | - | |
| Calving Season | | | 0.30- | | - |
| – SE | -0.65 | 0.52 | 0.90 | 0.02 | |

CI= Confidence interval, FPR = Fat-to-protein ratio, Ref = Reference Level, AYR= All-Year-Round, Se= Seasonal

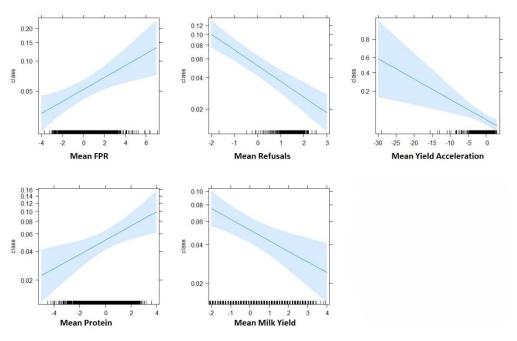


Figure 7-4 The probability of removal by 100-day post-partum (RD100) plotted against AMS production and behaviour variables retained in the final mixed-effects multivariable logistic.

Table 7-5 Performance metrics for mixed-effect multivariable logistic model assessing the association between early lactation AMS production data and the risk of removal from the herd by 100 days in milk, with and without the random effect of Herd

| | With Herd | Without Herd |
|-------------|-----------|--------------|
| AUC-ROC | 0.70 | 0.61 |
| Sensitivity | 65 | 56 |
| Specificity | 64 | 57 |
| PPV | 11 | 8 |
| NPV | 96 | 95 |

AUC-ROC= Area under the receiver operator curve, PPV = Positive predictive value, NPV = negative predictive value

7.3.2 Predictive Modelling

The training and test datasets were comprised of 6,416 and 2,723 lactations from 15 and 6 herds respectively. The prevalence of RD100 within the training dataset was increased from 7% to 50% following up sampling (Figure 7-1). Within the test dataset, the prevalence of RD100 was 5.4%. Removal by 100 DIM was predicted across the test set with an AUC-ROC of 0.52, sensitivity of 38%, specificity of 70%, PPV of 7% and NPV of 95% (Table 7-6). Herd specific prediction for each herd within the test set is presented in Table 7-7. Scaled variable importance

for independent variables in the predictive model is presented in Table 7-8. Model calibration is presented in Figure 7-5. An ECE of 0.35 was observed.

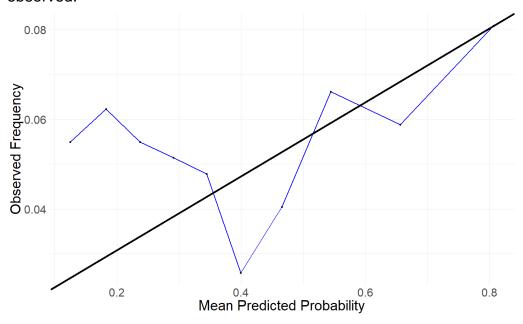


Figure 7-5 Calibration plot for the predicted probability of the risk of removal from the herd by 100 days in milk as evaluated on the test dataset.

Table 7-6 Performance Metrics for XGBoost model for the prediction of removal by 100 days post-partum as assessed on the test dataset

| AUC-ROC | 0.52 | |
|--------------------|------|--|
| Sensitivity | 38 | |
| Specificity | 70 | |
| PPV | 7 | |
| NPV | 95 | |
| AUC-ROC= Area | | |
| under the receiver | | |

under the receiver operator curve, PPV = Positive predictive value, NPV = negative predictive value.

Table 7-7 Herd-level performance metrics for the XGBoost model for the prediction of removal by 100 days post-partum as assessed on the test dataset

| Herd | 9 | 12 | 25 | 40 | 44 | 45 | |
|-------------|----|----|----|----|----|----|--|
| Sensitivity | 42 | 25 | 17 | 50 | 64 | 86 | |
| Specificity | 79 | 86 | 82 | 74 | 50 | 77 | |
| PPV | 18 | 9 | 11 | 6 | 4 | 6 | |
| NPV | 93 | 96 | 88 | 98 | 98 | 99 | |

PPV = Positive predictive value, NPV = Negative predictive value

Table 7-8 Scaled variable importance for independent variables in the XGBoost model for the prediction of removal from herd with the first 100 days post-partum

| | Variable |
|----------------------------|----------------------------------|
| Variable | Importance Score ¹ |
| Mean Fat-to-Protein Ratio | 100 |
| Mean Yield Acceleration | 89 |
| Mean Conductivity | 76 |
| Conductivity Alert | 40 |
| Mean Concentrate Dispensed | 29 |
| Mean Refusals | 28 |
| Mean Fat | 25 |
| Mean Milkings | 19 |
| Mean Milk Yield | 18 |
| Mean Protein | 14 |
| Parity | 0 |

¹Variable importance score as calculated by the VarImp function of the CARET package

7.4 Discussion

Early lactation AMS production and behaviour data collected over 1-3 DIM has demonstrated significant association with the risk of RD100. Cows recording higher Mean Milk Yield, Mean Yield Acceleration and Mean Refusals had reduced odds of removal by 100 days. Those with increased Mean Protein and Mean FPR over days 1-3 had increased odds of removal. Within the final multivariable model, these variables accounted for a relatively small proportion of the variance in risk observed. This suggests that while production and behaviour data were significant factors, variables outside of those assessed in this study have a substantial influence on the risk of removal. When applied in a predictive model, AMS data proved to have limited utility for the prediction of RD100 in previously unseen herds.

Increased Mean Milk Yield and Mean Yield Acceleration over days 1-3 in milk was associated with a reduced risk of removal within the first 100

days. Milk yield in the days immediately following calving has a demonstrated association with peri-partum metabolic health (Westhoff et al., 2024). The rate of increase in yield over this time is higher than at any other point during lactation and represents a substantial metabolic challenge (Hansen et al., 2006). Within our study, those capable of supporting higher levels of milk production and a more rapid yield acceleration may represent a group which have adapted well to the stressors of calving and hence, have a reduced risk of RD100. These findings agree broadly with those of Lukas et al. (2015), who demonstrated that cows with reduced milk production relative to expected over DIM 1-7 have an increased risk of removal by DIM 100.

Within free flow AMS, cows may present for milking as often as they wish. This offers a unique opportunity to assess physiological status through voluntary robot visit behaviour. Where a cow re-presents at the robot shortly after a successful milking visit, she is directed through the robot without being milked. This is known as a refusal. Within our study. those recording a higher number of refusals over days 1-3 were found to have reduced odds of removal. Robot visits are driven by the desire for the concentrate feed supplied during milking (Prescott et al., 1998). Cows registering increased refusals are therefore likely demonstrating a strong appetite. Furthermore, as this feed is a high value resource and the robot is subject to competition between herd-mates (Rodenburg, 2017), it is likely that cows recording a greater number of refusals represent a healthier and more robust cohort capable of exerting dominance within the herd. This is evidenced further by the reduced level of refusals associated with the incidence of both lameness and low body condition score (BCS) (King et al., 2017).

Increased FPR was associated with an increased risk of RD100. Milk fat-to-protein ratio is well established as a marker of physiological status in the transition cow (Friggens et al., 2007). In early lactation, its increase is reflective of fat mobilization in response to a state of negative energy balance (NEB). While NEB is common in the postpartum period, its severity is a key determinant of transition cow health (Macrae et al., 2019). When assessed at herd level using monthly milk test-day constituent data, Dechow et al. (2008) reported a positive association between increased FPR and the risk of cull by day 60 postpartum. Our results demonstrate that this association holds at the level of the individual in the days immediately post-partum. A reduction in early lactation milk protein percentage is generally reported in response to a state of NEB and has been associated with several transition cow diseases (Toni et al., 2011). As such, the positive correlation between protein percentage and RD100 reported here is unexpected. The analysis of milk protein in the immediate post-partum period is complicated by the variance in protein content seen as colostrum gives way to mature milk (Westhoff et al., 2024). However, increased milk protein percentage in the immediate post-partum period has been associated with cows in higher BCS as well as those experiencing extended dry periods (Pires et al., 2013; Ahmann et al., 2021). Both of these conditions are conducive to unfavourable metabolic status postpartum and have themselves been associated with an increased risk of removal in early lactation (Pattamanont et al., 2021). This offers a potential explanation for our findings; however, further research will be necessary to substantiate this association.

Overall, our final multivariable model explained a small proportion of the variance in risk of removal, returning a conditional R² of 13%. This indicates that a substantial portion of the variance observed in our dataset remains unexplained. The fixed effects investigated account for 7% of observed variance. Previous studies investigating the association between physiological status in the first three days post-partum and subsequent risk of removal fail to report a coefficient of determination (Neves et al., 2018; Venjakob et al., 2018; Menta et al., 2021). However, given the short timeframe over which animals were assessed (DIM 1-3), the low marginal R² reported here is unsurprising. While this approach allows us to specifically assess the impact of physiological status at calving, this represents a narrow timeframe when compared with the risk period analysed (DIM 4-100). Our model, therefore, likely fails to account for the development of disease leading to death or cull for which no physiological indicators were apparent within the first three days post-partum.

Just under half of the variance captured by the model was attributable to the random effect of Herd. This effect was further demonstrated by the classification performance of the model following internal validation. Moderate classification performance was achieved when the random effect of Herd was included, this decreased to poor when the effect of Herd was nullified (Table 7-5). The effect of group-level factors including the farm system and management practices on the risk of removal in early lactation have been previously demonstrated (Thomsen et al., 2006; Raboisson et al., 2011). Our study reinforces these findings, highlighting the potential for the incorporation of herd-level data to improve model accuracy.

This study furthers our understanding of removals in early lactation by demonstrating the relationship between production and behaviour traits assessed in the first 3 days post-partum and the risk of removal in the first 100 days. This highlights the detrimental effect of poor metabolic health at the point of calving and emphasises the importance of dry period management in herds reporting an elevated rate of removal. The variables retained within our final model may provide valuable information for such herds. In contrast to previous studies which have demonstrated this relationship, we investigated automatically collected variables which are available in real time via AMS. This may allow producers to efficiently incorporate this data into their assessment of transition cow health. Furthermore, where these variables demonstrate predictive power for the risk of removal, they may facilitate the development of predictive models capable of identifying cows at high risk of removal.

Although commonly misconstrued in the literature, statistically significant association does not infer predictive power (Lo et al., 2015). Associations identified by inferential models further our understanding of the biological processes under investigation and may provide direction in the search for predictive variables. However, it is through the performance of predictive models validated on previously unseen herds, that the predictive power of candidate variables should be assessed (Poldrack et al., 2020). A number of inferential studies have identified significant association between markers of physiological status in early lactation and the subsequent risk of removal (Seifi et al., 2011; Roberts et al., 2012; Venjakob et al., 2018; Menta et al., 2021). However, the predictive power of these variables was not investigated.

Our results demonstrate that despite statistically significant association, early lactation AMS data has minimal predictive power for RD100. This highlights the limitations of inferential models for the identification of predictor variables and the dangers in the use of statistical association to infer predictive power. Across the entire test dataset, an AUC-ROC of 0.52 was returned, indicating a non-informative model. On a herd basis, variance in model performance was small (Table 7-7). While sensitivity ranged from 86% in Herd 45 to 17% in Herd 25. PPV did not exceed 18% for any herd. Model calibration was poor, particularly within subgroups with a low probability of removal. This highlights the limitations of this model's ability to deliver accurate predictions for both individual and subgroups of animals. Applied on farm, this model would fail to identify the majority of cows suffering RD100 while generating a large number of false positives. Thus, while early lactation AMS data demonstrated significant association with the risk of RD100, the variables investigated had limited utility for the development of predictive models. Future inferential studies should incorporate an assessment of predictive power for any variables investigated in order to provide direction in the development of such models.

7.5 Conclusions

This is the first study to report the association between AMS data from days 1-3 in milk and the risk of removal in early lactation. By pairing this analysis with an externally validated predictive model we also report the predictive power of this data. We have demonstrated that cows recording increased Mean Milk Yield, Mean Yield Acceleration and Mean Refusals have a reduced odds of removal in the first 100 days. Conversely those with elevated Mean Protein and Mean FPR demonstrated increased odds of removal. When applied in a predictive machine learning model, AMS data demonstrated limited utility for the prediction of RD100, highlighting the danger in the use of statistical association to infer predictive power. We suggest that future analysis should increase its focus on the predictive power of early lactation data in the pursuit of an accurate, generalisable predictive model for the risk of removal in early lactation.

Chapter 8 Discussion and Conclusion

8.1 Introduction

Since the adoption of the United Nations Sustainable Development Goals, almost a decade ago, the dairy industries of the United Kingdom and Republic of Ireland have both recorded an increase in the volume of milk produced per-annum (Kelly et al., 2020; O'Mara et al., 2021; Uberoi, 2021). Concurrent with this expansion, the industry has been subjected to increasing scrutiny regarding its environmental and social sustainability (Schiano et al., 2020; Bojovic & McGregor, 2023). For the dairy industry to thrive over the coming decade, it must respond to this scrutiny by adapting its systems of production to reflect the goals of its regulators, and values of its consumers. Its future, therefore, is contingent on it fulfilling its core purpose of food production, in an environmentally, economically and socially sustainable manner. A single, but significant component of this will be the success with which the health and welfare of animals within the industry can be protected, without which true sustainability cannot be achieved.

The disproportionate influence of the transition period on the health and welfare of the dairy cow has been long established (Drackley 1999). This has motivated research seeking to further our understanding of the physiological challenges associated with transition and explore management strategies to reduce their impact. Despite this however, the current rate of morbidity and mortality in early lactation remains onpar with that reported decades previously (Mulligan et al., 2006a; Daros et al., 2022). This failure to translate our understanding of transition cow physiology into meaningful improvement in health reflects the complexity of the problem posed by transition. Under these circumstances it is imperative that stakeholders within the dairy industry seek to apply the resources at their disposal, not only to further improve our understanding of transition failure, but to develop monitoring programs which help producers reduce its cost and consequence.

The aim of this thesis was to assess the relationship between production and behaviour data as collected by AMS in the early postpartum period, and subsequent dairy cow performance. Of particular interest was the accuracy with which AMS data could predict performance and thus, lend itself to use within an automated transition cow monitoring program. Utilising data from 39 herds recruited across the UK and Republic of Ireland, mixed-effect models were employed to describe, for the first time, the relationship between transition cow production and behaviour data collected by AMS and subsequent performance. Crucial to the significance of these findings is the time at which, and methods by which, this data was collected. In Chapter 3 and 7 we demonstrate that data collected overs days 1-3 post-partum has significant association with Yield Deviation at 30 DIM, and the risk of removal from the herd by 100 DIM. Similarly, in Chapter 5, data collected prior to day 22 post-partum demonstrated significant association with reproductive outcomes up to 58 days later. These

findings emphasise the critical nature of the transition period and highlights the potential for data collected during this time to be used within a transition cow monitoring program.

Given the barriers to the adoption of labour-intensive transition cow monitoring techniques described in Chapter 1, our investigation of automatically collected data is a vital aspect of this thesis. In reporting the association between behaviour parameters, including milking visits and refusals, we advance our understanding of these AMS-specific metrics by highlighting their utility, but also their limitations for the assessment of transition success under free-flow housing conditions. In addition, we demonstrate significant association between production parameters collected via in-line sensors, and subsequent performance. This represents an important contribution to our understanding of these novel data sources and highlights the opportunity for in-line sensor data to replace the labour-intensive monitoring approaches traditionally employed within TMPs.

The development, and external validation of machine learning models for the prediction of production, fertility and survival, described in Chapters 4, 6, and 7, assess the degree to which AMS data may be leveraged into meaningful improvements in animal health through prognostic TMPs. Transition cow data demonstrated moderate predictive power for Yield Deviation at 30 DIM and reproductive performance by 80 DIM, but no predictive power for the risk of removal by 100 DIM. These findings demonstrate the potential utility of this data to identify animals likely to experience poor milk production or fertility performance in the early stages of lactation and should encourage further investigation of how this data may be applied within TMPs. The marginal increase in model performance following the incorporation of auxiliary data sources reported in Chapter 6 yielded an important finding for the future development of such models. By examining the need to balance model accuracy with generalisability and ease of deployment, we highlight a significant issue facing developers operating in an environment of rapidly increasing data complexity. Finally, Chapter 7, describes a direct comparison of inferential and predictive modelling of the risk of removal from the herd by 100 DIM. This serves to demonstrate the dangers in the use of inferential models to imply predictive power and the need for externally validated predictive models to be incorporated in the assessment of novel data sources within predictive TMPs. By providing an automated means of assessing production and behaviour data in the early post-partum period, the monitoring capacity of AMS offers great potential for the development of transition cow monitoring programs. The investigation of the relationship between AMS data and subsequent performance, as well as the assessment of its predictive power, represents an important step towards realising this potential. There remains however, the need to pair the predictive models outlined here with intervention studies to quantify the impact of early mitigating actions on the economic, social and environmental impacts of poor transition health.

8.2 Inferential Modelling

The aim of the inferential analysis within this thesis was to further our understanding of the AMS variables investigated and to provide context as to their potential utility within TMPs. As detailed in Chapter 2, the outcomes investigated within this thesis may be significantly affected by herd-level environmental and management factors (Enevoldsen et al., 1996; Windig et al., 2005; Haine et al., 2017). An unbiased examination of independent variables, therefore, requires a statistical approach which accounts for potential correlation in outcome between animals within the same herd. The multivariable, mixed-effect modelling deployed in Chapters 3, 5, and 7 allowed us to identify variables significantly associated with each outcome of interest while accounting for the random effect of Herd and relevant confounders. Furthermore, within the final models, it allowed for the segregation of the coefficient of determination between fixed and random effects. This proved to be of particular value in Chapters 5 and 7, which assessed fertility performance and cull risk respectively. Across both chapters, a substantial proportion of the conditional R² was attributable to the random effect of Herd, indicating that the clustering of outcomes at the herd level explained a large proportion of the variance observed. Given our focus on understanding the potential utility of these variables within a TMP applied across a broad range of herds, this represents a crucial point of context within our investigation. Our modelling approach allowed us to explore this further by applying backward predictions with and without the effect of Herd, demonstrating significant reductions in model performance when the effect of Herd was nullified. Despite the value this approach provides, it remains underutilised within this field. This is exemplified by the literature investigating the association between early lactation physiological markers and subsequent risk of cull. These investigations utilise mixed-effects models to highlight significant association between cull risk and early lactation parameters related to calcium and energy status, (Seifi et al., 2011; Neves et al., 2018; Venjakob et al., 2018). However, no attempt is made to quantify the explanatory power of these models or to compare that attributable to the fixed effects with that of random effects. This hinders the selection of variables with potential utility within TMPs by failing to provide a clear assessment of the strength of relationship between independent and dependant variables. Furthermore, it does not allow for meaningful comparison to be made with novel data sources which may be investigated in the future. Our approach aims to provide greater context for the variables investigated and allow for a clearer appraisal of their utility.

8.2.1 The transition period – A key inflection point

As described in Chapter 1, the application of TMPs in the dairy industry to date has focused on the diagnosis of specific disease states. This approach allows the assessment of independent variables to be targeted within a time frame immediately surrounding the outcome of interest. For example, to assess the utility of activity and rumination

data to diagnose an LDA, Stangaferro et. al., (2016) retrospectively examined neck mounted accelerometer data in the days immediately surrounding diagnosis. The time frame over which independent variables were assessed in this thesis is reflective of our prognostic aims and represents an attempt to strike a balance between the temporal relevance of data (i.e., the time between observation and outcome of interest), and the lead time required by producers to implement pre-emptive measures and mitigate associated losses. Data from days 1-3 post-partum were assessed in both Chapters 3 and 7 to facilitate intervention prior to the increased rate of morbidity and mortality generally seen in the first weeks of lactation (Ingvartsen et al., 2003; De Vries et al., 2010). Similarly, the investigation of reproductive outcomes in Chapter 5 examined data available before the end of the VWP to facilitate intervention prior to the breeding period. Of interest was the degree to which this data was associated with subsequent health and production despite the lag between observation and outcome.

Prior reports have highlighted the association between physiological status in the early post-partum period and long-term performance outcomes. This includes the demonstration of association between serum calcium and free fatty acid concentration during the first three days post-partum, and subsequent survival and reproductive performance (Venjakob et al., 2018; Menta et. al., 2021), milk constituents within the first week post-partum and subsequent production (Toni et al., 2011), and energy balance assessed over days 1-21 and reproductive success (McArt et al., 2012; Civiero et al., 2021). The results of Chapters 3, 5, and 7 align with these reports in highlighting transition cow variables, reflective of metabolic. inflammatory and immune status, which demonstrated significant associations with long-term performance outcomes. However, while these are encouraging results for the potential utility of transition period data within TMPs, the challenges of transition extend beyond the observation windows employed within this thesis. An example of such is the challenge posed by negative energy balance. We have demonstrated a significant negative association between dairy cow performance and NEB (as indicated by an increased fat-to-protein ratio) over days one to three (Chapters 3 and 7) and one to twenty-one postpartum (Chapter 5). However, positive energy balance is often not reestablished until day 50 post-partum (Grummer et al., 2010). Thus, the use of a condensed observation window cannot quantify the true depth and duration of NEB experienced, this is likely a limiting factor in the degree to which our observations can explain the outcomes of interest. This limitation is highlighted by the pattern in model fit attributed to fixed and random effects in Chapters 3, 5, and 7. When assessed via the fixed effects, a pattern of decreasing model fit concurrent with increasing lag between observation and outcomes was seen across all inferential models. The shortest lag was that of Chapter 3 which examined the association between AMS data collected overs days 1-3 and Yield Deviation at DIM 30. This model returned a marginal R² of 47%. This stands in contrast to the model reported in Chapter 7 which

returned marginal R² of 7%. It seems likely that these findings, to some extent, reflect the fact that as the lag between observation and outcome lengthens, an increasing number of physiological, environmental and management changes, with the potential to influence subsequent performance, will occur outside of the observation window. In contrast to the diagnostic TMPs, sufficient lead time to allow pre-emptive action is a pre-requisite for predictive monitoring programs, thus necessitating lag between observation and outcome. The identification of variables which are available for assessment as early as possible in lactation is therefore, an important factor in the development of these programs. The absence of reported coefficients of determination for previously investigated serum and milk-based variables (Toni et al., 2011; McArt et al., 2012; Venjakob et al., 2018; Menta et. al., 2021; Civiero et al., 2021) prohibit direct comparison with the models reported here. Nevertheless, our results advance this field by highlighting the association of variables which are readily available on AMS with subsequent performance, findings with offer much greater potential utility within TMPs. However, despite the critical nature of the transition period, the inherent lag between observation and outcome associated with a prognostic approach to monitoring, likely limits the extent to which data collected during this time can explain long-term performance outcomes.

8.2.2 AMS Technology – An alternative means of transition cow monitoring

A key barrier to adoption of TMPs has been the labour-intensive nature of the monitoring protocols required. The AMS data examined within this thesis was collected automatically using cutting edge sensor technology and includes both novel means of assessing traditional transition cow metrics (e.g., fat-to-protein ratio assessed via in-line sensors rather than test-day milk), as well as metrics unique to automatic milking systems (e.g., refusals). It was important therefore, that the relationship between these metrics and subsequent performance be assessed and where possible compared with traditional means of assessment to further our understanding of their potential utility.

Assessment of Cow-Robot Interactions

A unique aspect of data derived from free-flow AMS is the ability to monitor the frequency and nature of cow-robot interactions. These metrics have been demonstrated to reflect the animal's drive to access the concentrate feed provided by the robot as well as their physical ability to reach the robot (Steensels et al., 2016; King et al., 2018). Thus, they have the potential to serve as useful indicators of physiological status. However, outside of reported association with lameness (Bach et al., 2007; Borderas et. al., 2008; King et al., 2016) their potential use in the diagnosis of disease is limited to a small number of single herd investigations (King, et al., 2017; Steensels et al., 2016). Excluding the investigations reported within this thesis, their use in prognostic models related to long-term performance outcomes remains unexplored.

The mean number of milking visits recorded was retained as an explanatory variable in the final models for Yield Deviation and Expression of Oestrus or Insemination only (Chapters 3 & 5). Within these models it demonstrated a negative association with YD and positive association with EOI. The negative relationship between Mean Milk Visits and Yield Deviation is counterintuitive, given the previously demonstrated positive association between increased frequency of milking in early lactation and subsequent production (Siewert et al., 2019). Where YD is utilised as a proxy for health (Nordlund, 2006), these results also stand in contrast to those reporting reduced milking visits in response to clinical disease (Bach et al., 2007; King et al., 2018). The potential for manual fetching to bias milking visits means validation of these results, ideally under circumstances which facilitate the differentiation of voluntary and fetched milking visits, is required. However, given the widespread practice of manual fetching on free-flow AMS, our findings indicate that the utility of milking visits to reliably reflect the physiological status of the transition cow is likely to be limited.

Unlike milking visits, refusals are not influenced by manual fetching routines and may represent a more promising behaviour-based indicator of physiological status. The mean number of refusals recorded over DIM 1-3 demonstrated a statistically significant positive association between both Yield Deviation (Chapter 3) and negative association with the odds of removal by 100 DIM (Chapter 7). Under the assumption that animals recording an increased number of voluntary visits to the robot are likely to be a healthier, more robust cohort, these results align with our expectations. However, as this is the first report of their association with long-term transition outcomes, further validation of these results would be beneficial.

Within the data automatically collected by modern AMS, visit behaviour represents an intriguing but complex subset. Given the limited investigation of visit metrics as a means of assessing physiological status, particularly in the early post-partum period, this thesis contributes substantially to the current literature. However, gaps remain in our understanding of the relationship between visit behaviour and subsequent transition cow performance.

Assessment of Milk Quantity and Milk Quality

Of the variables used in the assessment of milk quantity and milk quality within this thesis, milk yield, milk yield acceleration, and fat-to-protein ratio consistently demonstrated significant association with the outcomes investigated. These metrics are traditionally assessed using monthly test-day regimes through which animals will receive, at most, a single reading prior to the end of the transition period. The automated in-line means of assessing these parameters offers a high frequency approach to monitoring milk quantity and quality and is available from the day of calving. Their assessment represents an interesting point of analysis as their availability is not limited to producers utilising AMS.

Modern conventional parlours offer real time assessment of yield while a range of in-line fat and protein sensors are commercially available (e.g., MSD SenseHub™ Dairy https://ie.sensehub.global/). While their uptake in the UK and ROI remains limited to-date, continued growth in their use could significantly increase the capacity for conventional dairy farmers to utilise in-line milk quality data within transition cow management. This broadens the potential applications of our findings beyond producers utilising AMS. Due to the novel nature of this technology, the number of reports documenting the relationship between these metrics and health and production outcomes is limited when compared with those utilising test-day milking data. Our findings therefore represent a considerable addition to the literature surrounding this means of data collection.

As highlighted in Section 1.6.1, conflicting reports as to the association between milk yield and subsequent performance exist, indicating that care must be exercised when interpreting the association between production parameters and subsequent performance. We have demonstrated that milk production and the acceleration in the rate of production overs day 1-3 are positively associated with YD at DIM 30 and negatively associated with the risk of removal by DIM 100, while milk production assessed overs days 1-21 was negatively associated with both EOI and CFI. When compared with monthly test-day regimes, the means to assess milk volume from the point of calving, where the rate of change in production is greatest (Ingvartsen et al., 2003), offers a more pertinent assessment of physiological status for the transition cow. However, the variability of daily milk yield is high in early lactation and may be influenced by a wide range of environmental and social factors (LeBlanc, 2010). Further to this, a number of management practices have the potential to affect recorded yields. The time between calving and introduction to the milking herd, as well as level of colostrum harvested by the calf, may all contribute to variances in milk yield recorded in the days immediately post-partum. While we attempted to reduce the effect of this variance by excluding yield from the day of calving (Day Zero), variance in daily milk yield is reported to persist for the first 10 days of lactation (Constable et al., 2010; Kessler et al., 2014). Validation of these results is therefore warranted. however, the consistency with which these production parameters demonstrate statistical association across all three outcomes investigated highlights this means of data collection as a viable alternative to test-day regimes traditionally utilised with TMPs.

Mounting an adequate and appropriate metabolic response to the increase in energy requirements experienced at the start of lactation is a crucial challenge facing all transition cows. The means to monitor the success or failure of this response, and the association with subsequent health and performance has been described for the manual assessment of body condition score (Manríquez et al., 2021), as well as metabolic indicators in serum, urine and milk (Geishauser et al., 2001, Friggens et al., 2007; Jansen et al., 2021). Modern AMS offer an alternative means of monitoring metabolic status through in-line fat and protein indications.

However, the relationship between these parameters and transition performance has not been previously examined. In Chapters 3, 5, & 7 we demonstrated a negative association between fat-to-protein ratio and subsequent production, fertility and survival. These findings are supported by previously reported association between FPR measured via monthly test-day regimes, and subsequent yield (Kaufman et al., 2018), cull risk (Dechow et al. 2008), and fertility performance (Heuer et al., 1999), re-emphasising the negative effects of excessive mobilisation of fat in the post-partum period. The consistency demonstrated between these results and those reported in this thesis, using in-line milk constituent analysis furthers our confidence in the use of this novel technology as an indicator of metabolic health post-partum. The significance of fat-to-protein ratio across these models should also encourage the investigation of additional measure of energy balance within automated transition cow monitoring programs. A number of automated systems for assessing energy balance have been commercially deployed within AMS, including automated body condition scoring (O' Leary et al., 2020) and automated BHB monitoring (Mazeris. 2010). While these variables were not available for investigation within our dataset, our findings suggest their value within TMPs is worthy of investigation.

8.3 Predictive Modelling

8.3.1 Prognostic TMPs – A viable tool for transition cow management

The Inferential models reported within this thesis further our understanding of the relationship between AMS data and subsequent dairy cow performance. Translating this understanding into improved transition cow health requires predictive models applied within a TMP. The goal of prognostic TMPs is to identify animals likely to experience the cost and consequence of poor transition health. The accuracy with which predictive models can achieve this will largely determine the value these programs can deliver on farm. However, an assessment of the true value of any prognostic TMP requires analysis of the entire program. This must include for instance, the number of animals in receipt of treatment as a result of the program as well as the effectiveness of these treatments in mitigating losses. While analysis of this kind will play a vital role in the future development of prognostic TMPs, it is beyond the scope of our research. In its absence, our assessment of the utility of AMS data within TMPs is based solely on its ability to efficiently identify animals likely to experience reduced performance in a time frame which facilitates pre-emptive intervention.

The predictive power of AMS and neck mounted accelerometer data examined in Chapters 4 and 6 represents a critically important finding in support of the premise of prognostic TMPs. While the classification accuracy reported is moderate and the need for further development of these models is clear, the results remain encouraging, particularly within groups at the extremes of the performance spectrum. For the

identification of animals experiencing a highly negative yield deviation, animals least likely to express oestrus in the VWP or least likely to conceive to first insemination, their respective predictive models achieved group level classification accuracies ranging from approximately 60 to 70%. While the novel nature of yield deviations means comparison with prior reports of predictive models is not possible, prediction accuracy relating to reproductive performance within the current literature is comparable with that reported within this thesis (Hempstalk et al., 2015; Shahinfar et al., 2014). Furthermore, these results are on-par with diagnostic TMPs currently deployed within commercial on-farm software (Lely, personal communication). Applied on farm, such models may have sufficient accuracy to improve transition health by identifying animal groups for which management intervention is likely to be beneficial. However, as this is the first report of prognostic models for use within TMPs, the foundation these results establish for the continued development of these models may be of greater importance than the performance accuracies achieved. By demonstrating the predictive power of the variables analysed, these findings set a precedent for the continued investigation of transition cow data for use within prognostic TMPs. The expanding range of variables collected on modern dairy farms offers an opportunity to enhance model performance by providing a more comprehensive assessment of physiological status during transition. Furthermore, the degree to which these results demonstrate the predictive power of data collected in the days immediately post-calving should encourage the investigation of alternative observation windows. For example, the utility of data collected during the dry period remains underexplored. The scarcity of automatically collected data during this time means investigations have focused on manually collected variables such as serum metabolic indicators as well as dry cow management data. The association of dryperiod indicators of negative energy balance, such as serum NEFAs concentration, and body condition score, with subsequent performance highlight the potential utility such assessments may have within TMPs (Ospina et al., 2013; Chebel et al., 2018). More recently, Cook et al., (2024) demonstrated significant association between stocking density and time spent in the close-up dry pen with subsequent cull risk and milk production. While this demonstrates the potential for pre-partum data to be incorporated into TMPs, the lack of an automated means of data collection remains a challenge. Perhaps more promising, is the investigation of rumination and activity monitoring during the dry period. Within a small, single herd study, rumination levels in the week prior to calving were found to be significantly associated with subsequent fertility, production and culling risk (Santos et al., 2024). This is an area in need of further research but, incorporating such data may serve to improve the predictive power of the models reported here. The predictive models reported within this thesis represent an encouraging first step in the development of prognostic TMPs. While they directly advance this field of research, they serve also to highlight the ample opportunity which exists for their performance to be enhanced through continued development.

While these results serve to demonstrate the potential for the future development of prognostic TMPs, challenges are also apparent. The absence of predictive power for the removal in early lactation reported in Chapter 7 highlights a potential limitation of this approach to transition cow monitoring, particularly where the lag between observations and outcomes is large. Across Chapters 4, 6, and 7, the group levels classification performance of predictive models decreased as the time between observation and outcome of interest lengthened. While comparison between these models cannot be used to assess the effect of lag, our results raise important questions regarding how to balance model accuracy with the minimum lead time required for predictions in order to instigate pre-emptive action. The degree to which the lead time for each individual outcomes can be lengthened while maintaining adequate predictive power is an intriguing point of development within these models. However, it highlights an inherent limitation within this approach, one which is likely to restrict the predictive power which can be achieved by models incorporating a prolonged lag between observation and outcome.

The development of accurate predictive models represents a single step within a TMP, thus our assessment of the value which these models may provide when applied on farm must be interpreted with caution. While our results highlight some inherent limitations, the predictive power demonstrated in Chapters 4 and 6 remain encouraging. These serve to support the premise of using data available from AMS at critical time periods in lactation to predict long-term outcomes. While further development of these models is required, these results, coupled with the expanding capacity for the automatic assessment of physiological status throughout the lactation cycle, offer a positive outlook for the future of prognostic TMPS.

8.3.2 Optimising model development in an environment of increasing data complexity

As the application of sensor technology within the dairy industry has continued to expand, the volume of data available for incorporation within prognostic models has increased concurrently. This technological advancement offers the means to improve the accuracy of predictive models, but also poses challenges in their development. As discussed in Chapter 2, the ease with which models can be deployed on a global scale has increased the diversity of farms on which they must deliver accurate predictions. Therefore, key to maximising the improvements in transition health these models can provide is the efficient assessment of novel data sources while maintaining model generalisability and ease of deployment.

Given the rapid developments in sensor technology, the efficient assessment of novel sensor data for utility within predictive models will play an important role in the future of TMPs. Mixed effects models offer an easily interpretable approach to inferential analysis of such data.

However, where the modelling aims are predictive, machine learning algorithms offer greater flexibility, and have been demonstrated to deliver higher predictive performance (Eicker et al., 2002). While each approach has its own advantages and limitations, when used together they complement one another by allowing the examination of both the statistical and practical significance of the relationships investigated. Despite this, the use of predictive modelling in tandem with inferential analysis is rarely undertaken within the current literature relating to TMPs. More commonly observed is the demonstration of statistically significant association without any investigation of predictive power. The risk associated with inferring predictive power from such studies has been widely reported (Heus et al., 2018). Despite this however, it is commonly encountered in research relating to precision medicine, both in human (Bzdok et al., 2021; Varga et al., 2020) and veterinary fields (Garverick et al., 2013; Neves et al., 2018; Carty et al., 2020). The lack of predictive power demonstrated in Chapter 7, despite the presence of statistically significant association serves to re-emphasise the importance of combining inferential studies with externally validated predictive models to more efficiently and appropriately assess the true utility of candidate variables. This will serve to further our understanding of novel metrics while also expediting the process by which they can be selected or rejected for use within a predictive TMPs. The approach described within this thesis, represents an efficient means of achieving this and will contribute to advancing this field of analysis.

A key element in the development of generalisable predictive models is the pursuit of model parsimony. As a rule, simple models tend to generalise to a greater extent than complex ones, though this may come at the expense of a more accurate prediction across the training set - the so-called Variance-Bias trade off (James et. al., 2013). Within this thesis we applied two approaches to feature selection. Our use of recursive feature elimination within Chapter 4 represents a commonly used "wrapper" approach which aims to identify the subset of variables which results in the desired performance output (Kuhn, 2008), in this case, minimisation of RMSE. This represents an automated, relatively efficient and, when carried out in conjunction with cross validation. robust method of feature selection (Kuhn and Johnson, 2013). However, this approach fails to differentiate between data sources (e.g., variables derived from the robot itself, or from neck-mounted accelerometers). In Chapter 6, we chose to pair RFE with a manual feature selection process which sought to quantify the marginal effects of auxiliary data sources, a technique currently underutilised within this field. The value in this approach was highlighted by the retention of features from all 3 data sources in the final model, despite it failing to demonstrate a statistically significant improvement in predictive performance over its more parsimonious counterpart. Minimising the number of data sources required serves to broaden the number of farms eligible to employ a specific model, while also reducing the risks associated with sensor malfunctions and data missingness. Feature selection applied at the level of the individual variables has an important role in model development and has been applied across all predictive

models relating to TMPs. However, our findings demonstrate its shortcomings and highlight the importance of a more considered approach where deployment on a broad scale is of importance. Ultimately, the challenges surrounding model generalisability and deployment stem from the unprecedented opportunity the expansion of precision dairy technology presents for the development of predictive models. Where these can be addressed through appropriate feature selection, there exist great potential for these models to contribute meaningfully to transition cow management. Our results highlight the importance of considering the practicalities associated with model deployment and generalisability in order to maximise potential impact.

To accurately assess expected generalisability, models intended for deployment across a wide range of herds must be tested under conditions which reflect this. External validation serves as a robust method of simulating expected model performance following deployment (Rockenschaub et al., 2023). However, attaining a dataset with sufficient quantity and diversity of herds to facilitate this is challenging. Internal validation, an approach requiring a much smaller dataset, continues to be reported (Madouasse et al., 2010; Shahinfar et al., 2014; Rutten et al., 2016;). While the use of cross-validation within this approach may somewhat limit the risks of falsely inflating predictive performance (James et. al., 2013), this does not represent an appropriate means of assessing herd-level generalisability. Variations of external validation, designed to accommodate datasets with limited herd numbers have been reported. For instance, predictive models for cull risk and reproductive performance have been trained and tested using the same herds but with data collected from different time periods (Hempstalk et al., 2015; Lukas et al., 2015; Ho & Pryce, 2020). This approach fulfils the basic tenants of external validation and represents an improvement over internal validation. However, given the influence of herd-level management factors on the outcomes investigated, the risk of falsely inflating predictive performance remains. Of importance within this thesis, was the development of models which could perform adequately across farms employing a range of environmental and management practices. Hence, an approach which split the train and test data sets by herd was deemed the most suitable. This approach allowed us to assess the predictive power of our models when presented with a previously unseen animal from a previously unseen herd - a scenario reflective of real-world deployment. The application of this approach is limited by the requirement for a large, multi-herd dataset, particularly in the investigation of novel sensor technology. However, where the goal is the development of predictive models which can deliver meaningful improvement in animal health its importance cannot be overstated.

8.4 Future Studies

A number of areas worthy of further investigation have been highlighted within this thesis. As all recruited farms were located within the UK and Republic of Ireland, the extent to which our results can be extrapolated

outside of this region is limited. Validation of these results across a broader range of farming system would be a worthwhile endeavour and provide a more robust indication of the generalisability of our findings. This is of particular importance in the case of Chapter 6 relating to fertility where the management systems applied vary significantly between geographical location. The degree to which lag between observations and outcomes affects predictive power for any specific outcomes is an intriguing and important area of investigation within TMPs, particularly where predictive power is lacking (e.g., the risk of removal from herd, Chapter 7).

Assessing the impact adoption of prognostic TMPs may exert on transition cow health, welfare and production requires a large scale, multi-herd intervention study. This study should aim to investigate the change in outcome for transition cows which receive pre-emptive treatment as a result of a TMP when compared with control animals. Research of this kind is extremely complex, particularly when carried out over multiple herds, a fact demonstrated by their scarcity within the literature. When compared with automated prognostic TMPs, as described within this thesis, the development of automated diagnostic monitoring programmes have received far greater research attention (Stangaferro et al., 2016; King, et al., 2017; Steensels et al., 2017; Paudyal et al., 2018). Despite this, only 3 reports examining the value such programs can deliver when compared with traditional monitoring techniques are available, and all are based on single herds (Silva et al., 2021; Perez et al., 2023; Rial et al., 2024b). Within the field of prognostic TMPs this knowledge gap is greater still. While a small number of recent publications have examined the relative value of targeted reproductive programs, these are limited in scope (Rial et al., 2022, Gonzalez et al., 2023). A substantial increase in the number of intervention studies examining the impact of TMPs is therefore required to assess the value such programs can deliver to producers and provide direction for future development.

While the approach to prognostic TMPs hold several advantages when compared with diagnostic TMPs, it is not designed to replace diagnostic models. Rather, it may serve as a means to improve how such models are applied. An example may be the application of the prognostic models described in Chapter 4 in conjunction with diagnostic models, similar to those described by Stangaferro et al., (2016) which demonstrate acceptable sensitivity and specificity for the automated diagnosis of transition cow disease. There exists an opportunity to develop diagnostic models which may be applied to animals deemed high risk (e.g., those predicted to suffer a large, negative yield deviation). Likewise in the case of predictions related to conception to first service at day 21, animals' classification at this early point in lactation may allow for the application of bespoke models and more accurate predictions of conception success at the point of service. This is an intriguing next step in TMPs which warrants investigation in the future.

8.5 Conclusions

The research presented within this thesis examines a novel means of transition cow monitoring, one which seeks to assess transition health using long-term performance outcomes. In contrast to traditional transition cow monitoring programs, which rely on the diagnosis of specific disease states, this approach offers a more holistic means of assessing transition health and more closely reflects our understanding of transition cow physiology.

Our inferential models have demonstrated significant statistical association between AMS production and behaviour data and subsequent yield deviation, fertility and survival. Group level classification of animals based on expected yield deviation and fertility outcomes using machine learning models demonstrated potential for incorporation into prognostic TMPs. In contrast, predictive models for the risk of removal by 100 DIM were found to be non-informative.

The development of generalisable predictive models for deployment in an increasingly interconnected agricultural environment is crucial to the utility of TMPs. We have highlighted the challenges increasing data complexity can place on model development and demonstrated the means by which these may be overcome. We believe this approach will maximise the positive impact of predictive models developed and accelerate the pace at which they can be confidently deployed commercially.

Chapter 9 References

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Chapter 10 Appendices

10.1

Study Title: Use of machine learning to predict transition success in dairy cows in an automatic milking system

Researcher:

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Purpose of the study:

I am a PhD student at the University of Nottingham. This project has been developed in partnership with the *Biotechnology and Biological Sciences Research Council and Lely Industries*.

Our research aims to utilise data generated from Lely automatic milking systems to predict transition period outcomes for dairy cows. Through this analysis we hope to increase the value Lely customers derive from their farm's data by developing tools which assist in transition cow management.

To this end I would be extremely grateful if you would be willing to contribute an anonymised version of your holding's T4C data to this study.

Consent:

Participant

This consent form is a formal indication that you agree to participate in this study and in so doing:

- Grant one-time access to your T4C software via TeamViewer
- Consent to the sharing of historical T4C data with the University of Nottingham
- Are aware that any information collected by the researchers:
 - will be anonymised and treated confidentially
 - will be used for research purposes only
 - may be used in a research publication
 - may be presented at research conferences or meetings

If you have any queries regarding this study, please feel free to contact me.

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|-----------------|------------|
| Name: | Signature: |
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To enable data sharing please provide you Lely Licence Key, TeamViewer ID, and Password

Thank you very much for participating in this study I look forward to sharing our researching findings with you in the near future.