

Factors Associated with Milk Production on Automatic Milking
System Farms

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Table of Contents

Acknowledgements.....	i
List of Tables	iv
List of Figures	v
Chapter 1	1
INTRODUCTION	1
AUTOMATIC MILKING SYSTEMS	2
MILK PRODUCTION IN AMS.....	5
FEEDING MANAGEMENT FACTORS AFFECTING MILK PRODUCTION	8
MILKING INTERVAL	16
SUMMARY	24
Chapter 2.....	26
SUMMARY	26
INTRODUCTION	27
MATERIALS and METHODS.....	29
<i>Farms and Data Collection</i>	29
<i>Data Processing and Statistical Analysis</i>	30
RESULTS and DISCUSSION	33
<i>Descriptive Statistics</i>	33
<i>Days in Milk</i>	34
<i>Number of Milkings per Day</i>	35
<i>Cows per Robot</i>	36
<i>Feeds and Feeding Management</i>	37
CONCLUSIONS.....	42
Chapter 3	43
SUMMARY	43
INTRODUCTION	44
MATERIALS and METHODS.....	46
<i>Farms and Data Collection</i>	46
<i>Data Processing and Statistical Analysis</i>	47
RESULTS and DISCUSSION	48
<i>Descriptive Statistics</i>	48
<i>Milking Interval Variability</i>	49
<i>Milking Failures</i>	50
<i>Interaction of MI CV and Failures</i>	51
<i>Weeks in Milk</i>	52
<i>Interaction of MI CV and Weeks in Milk</i>	52

<i>Parity</i>	53
<i>Interaction of MI CV and Parity</i>	54
CONCLUSIONS.....	55
REFERENCES	56
APPENDIX I – Tables.....	73
APPENDIX II – Figures	77

List of Tables

Table 1. Example of one farm’s feed table in an AMS.....	73
Table 2. Association of feeding management factors with milk production	74
Table 3. Associated milk production change by week compared to week one.....	75
Table 4. Association between the interaction of week and milking interval variability and milk production per cow per day	76

List of Figures

Figure 1. Average milk per cow per day by average days in milk..	77
Figure 2. Average milk per robot per day by average days in milk.....	78
Figure 3. Average milk per cow per day by average number of milkings per day.....	79
Figure 4. Average milk per robot per day by average number of milkings per day.	80
Figure 5. Average milk per cow per day by average concentrate intake..	81
Figure 6. Average milk per robot per day by average concentrate intake.	82
Figure 7. Average milk per robot per day by average number of cows per robot..	83
Figure 8. Average milk per cow per day by average neutral detergent fiber content of the partial mixed ration..	84
Figure 9. Average milk per robot per day by average neutral detergent fiber content of the partial mixed ration.	85
Figure 10. Average milk per cow per day by amount of feed material on the middle screen of the Penn State Particle Separator.....	86
Figure 11. Average milk per cow per day by feed intensity management category.....	87
Figure 12. Average milk per cow per day by milking interval coefficient of variation .	88
Figure 13. Average milk production per cow per day and average weekly milking failures.....	89
Figure 14. Interaction of milking interval coefficient of variation and average milking failures per week with associated milk production per cow per day	90
Figure 15. Average milk production per cow per day by week in milk.	91
Figure 16. Milk production change per cow per day, by week, as compared to week 1.	92
Figure 17. Average milk per cow per day by parity.	93
Figure 18. Interaction of milking interval coefficient of variation and parity with associated milk production per cow per day.....	94

Chapter 1

Literature Review

INTRODUCTION

The benefits of technological advancements on dairy farms are varied and oftentimes substantially benefit producers and dairy cows. Adopting new technologies can be challenging for producers (Eastwood et al., 2017), as they must transition from “experiential decision-making to data-driven processes.” Learning how to utilize the large amount of data that technology affords a dairy producer may also represent a daunting and overwhelming task (Butler et al., 2012). Research on how data from on-farm implemented technologies can be utilized effectively commercially is an important factor in the success of the dairy industry.

One technology which has been successfully implemented on many commercial dairy farms across multiple continents is the Automatic Milking System (AMS). Automatic milking systems provide a substantial amount of data for producers to utilize when making management decisions. Milk production, including yield and components, is an area that many dairy producers pay close attention to in AMS because it tends to be a primary measure of profitability. Along with increased milk production, reduced labor costs are also a potential benefit of AMS; thereby lowering total production costs and increasing profits for producers (de Koning, 2004). Furthermore, AMS provides the opportunity for owners and managers to utilize the wide range of data afforded by the systems to increase total farm efficiency.

Many questions exist around improving the efficiency of milk production in AMS. Our team wanted to investigate the impact of feeding management metrics and milking interval variability on milk production within AMS because research on these topics, to our knowledge, has been limited. Research specifically addressing the regularity of milking interval and any subsequent effects on milk production has only been reported in one study. More specifically with regard to feeding management practices, research on feed table management in AMS has not been published. Answers to the aforementioned questions around feeding management and milking interval variability will not only add to the body of literature around milk production associations in AMS but could also give consultants and producers more strategies to consider related to milking management settings, efficiency of production, feeding management practices and more.

AUTOMATIC MILKING SYSTEMS

A technology that continues to gain interest and utilization in the US is Automatic Milking Systems. Defined as an incentive-based robotic system in which cows choose to be milked in a voluntary or semi-voluntary manner without direct participation of an operator, AMS have numerous potential advantages (de Koning, 2010). Potential, but somewhat contested, advantages of the AMS include improved dairy cattle welfare, increased milk production by up to 12% and decreased labor by up to 18% (Jacobs and Siegford, 2012). Much of the AMS research is contradictory regarding welfare and productivity improvements. It has been suggested that on-farm management and other farm-specific variables could be affecting the outcomes of these studies, causing the

contradicting results (Jacobs and Siegford, 2012). Management practices and the ability of dairy producers to implement, monitor and alter AMS can impact the success and profitability of the technology substantially (Tse et al., 2017).

Dairy producers around the world continuously strive to produce the most milk, as efficiently as possible, while optimizing animal welfare in order to maximize their satisfaction and profitability. Indeed, the objective of dairy farming is to produce quality milk for consumers, maintain animal welfare standards, return a profit to the producer and practice environmental stewardship (von Keyserlingk et al., 2013). Automatic milking systems could potentially provide opportunities in all the aforementioned areas. Bach and Cabrera (2017) suggested an increase in milking frequency and precise, individual-cow-focused feeding management in AMS may be the solution to creating a highly profitable production system. The efficient conversion of feed nutrients into milk, potentially through the utilization of AMS features which allow greater individuality in feeding regimens, could help meet the economic objectives of dairy farming (de Ondarza and Tricarico, 2017).

Holloway et al. (2014) stated, “the installation of robotic milkers clearly establishes new forms of relationships between cows, technologies and dairy farmers.” These new forms of relationships have the potential to improve the life of dairy cows and dairy producers alike. AMS offer an opportunity for dairy producers to reduce labor for milking activities, which are typically on a highly regimented schedule not allowing for freedom of time for the producer. The labor-intensive nature of milking dairy cows in a

conventional setting can reduce a farmer's time spent on overall farm management, with family or on activities they enjoy (Rotz et al., 2003). It is conceivable, therefore, that a farmer adopting AMS technology could experience enhanced quality of life.

Dairy producers and consumers have increasingly sought answers to dairy cattle welfare questions and concerns. The possibility of technology like AMS being utilized to enhance welfare is an ongoing area of research. Stress responses in dairy cows can be caused by a fear or apprehension of humans, which in turn can inhibit milk production and reduce welfare (Rushen et al., 1999). AMS offers the possibility to reduce human-animal interaction which could allow the cow to encounter fewer of those stress-inducing experiences. Allowing cows to choose when they want to be milked, via an AMS, could have health and welfare benefits (Holloway et al., 2014). Specific welfare enhancing factors of AMS include: multiple individual cow adaptations, consistent routines, improved udder health, and even increased milk production in some studies (Svennersten-Sjaunia and Pettersson, 2008).

Consumers desire a consistent supply of safe and inexpensive food but are concerned about animal welfare, particularly on farms of certain size and scale (Wolf et al., 2016). While dairy cattle welfare is a primary concern for consumers, numerous other concerns also exist and include, but are not limited to, environmental impacts of dairy farming, safety of dairy products and social implications of dairy food product choices (Olynk et al., 2013). Dairy industry stakeholders continue to search for ways to maintain and increase efficiencies on farm and meet consumer demands, one of which may be AMS.

Adoption of AMS technology is aided by quality research and experience guiding producers to the system which best fits their motivations. AMS provides producers and other stakeholders a substantial amount of automatically collected data (Jacobs and Siegford, 2012). Successful implementation and use of an AMS can be heavily dependent upon the producer's access to and use of the data the systems provide (Butler et al., 2012).

MILK PRODUCTION IN AMS

Bach et al. (2020) indicated that, "The dairy industry has focused on maximizing milk yield, as it is believed that this maximizes profit mainly through dilution of maintenance costs." For dairy producers, maximizing milk production per AMS unit is a primary goal from an economic viewpoint (Sonck and Donkers, 1995). In order to maximize milk production in AMS, both milking frequency and milking speed must increase and the number of robots per pen must be optimized (Tremblay et al., 2016). Indeed, revenue maximization can only occur when the number of daily milking events and quantity of milk collected in each event are both maximized (Masía et al., 2020).

Milk yield in AMS versus milk yield in conventional milking systems is a much-debated topic. According to Wade et al. (2004), an increase in milk production of 5-10% with the introduction of AMS would be reasonable given previous research and economic modelling experiences. Interestingly, a study by that same group found an increase of only 2% in milk production with the introduction of AMS, after correcting for yearly

production increases from genetics and management (Wade et al., 2004). Without those annual corrections, the AMS yielded 12.4% more kilograms of milk and 10% fat and protein content (Wade et al., 2004).

Many factors can impact milk production in AMS. Free traffic flow, a greater number of robots per pen, and newly built versus retrofitted barns have been found to positively impact milk production (Tremblay et al., 2016). Cow-level factors such as genetics, health status, age and others play a major role as well in milk production responses (Sova, et al., 2014). Pasture-based grazing systems, partial grazing systems and fully housed or indoor-based systems utilizing AMS may all have different milk production responses as well. A study comparing milk production effects of 50% roughage intake from pasture with limited indoor roughage versus cows in an exercise paddock with ad libitum forage indoors in an AMS found greater milk yield in exercise paddock cows (Kismul et al., 2019). Many examples exist in published literature referencing the association of feeding management with milk production responses in conventional milking systems with far fewer such studies examining similar metrics within AMS.

Literature has previously suggested increasing milk yield is more efficacious than increasing herd size when attempting to maximize AMS capacity (Bach and Cabrera, 2017). In fact, Tremblay et al. (2016) found that a small reduction in herd size, selecting high milking speed cows in particular, increased milk production due to an increase in the number of milking events and decrease in time spent milking. Voluntary milking behavior and potential milk yield are also factors which can have an impact on milk

production in AMS (Løvendahl and Chagunda, 2011). As the number of milking events per day increases, the daily milk yield per cow increases (André et al., 2010). To maximize profitability of the AMS unit, maximizing efficiency of milk production per cow as well as per robot is crucial (de Koning and Ouweltjes, 2000; André et al., 2010). Milk harvested per AMS, a function of the number of milkings per day and the amount of milk collected each milking, has been found to be a critical factor to farm profitability (Salfer et al., 2017).

The efficiency of milk production is not as commonly researched a topic as the many ways that exist to maximize milk yield (Bach et al., 2020). Milk production efficiency is defined as the amount of milk output per resource inputs (Bauman and Capper, 2008). Improving efficiency of milk production can positively affect profitability by increasing income while lowering cost and environmental impacts by utilizing more nutrients for milk, thus diluting maintenance requirements (Bach et al., 2020). Bach et al. (2020) indicated that, “efficiency of milk production is influenced by the quantity and quality of heifers, the quantity and quality of dry cows, the length of the dry period, the length of the rearing period, the quality and adequacy of the feeds provided, the environmental conditions, the longevity of the cows, the amount of feed that is wasted or spoiled, and still other factors.”

One of the many other factors which can influence the efficiency of milk production, and thus profitability of commercial dairy producers, is milking interval (MI). Milking interval is defined as the time, generally hours, between two consecutive milking events

(Penry et al., 2018). Automatic milking systems could provide the data and opportunities which allow producers and managers the ability to become more efficient utilizing metrics like MI, among others.

As previously mentioned, feeding management likely plays a major role in milk production responses in AMS. Knoblauch et al. (2012) reported that 50 to 60% of the operating expenses on a dairy farm can be accounted for by feed, thus making this a major area of efficiency improvement discussions. Specifically, the efficiency of nutrient utilization within the cow can be a major influencer of milk production response and efficiency (de Ondarza and Tricarico, 2017).

FEEDING MANAGEMENT FACTORS AFFECTING MILK PRODUCTION

Because feed represents 50 to 60% of the cost of producing milk, dairy producers have continually put their focus on ways to mitigate feed cost (Knoblauch et al., 2012). It has been well documented that both the management of feedstuffs as well as the specific nutrient utilization at the cow-level of those feedstuffs play a large role in milk production responses in dairy cows (Allen, 1996; Oba and Allen, 1999; Heinrichs and Kononoff, 2002; Gaillard et al., 2017; Haselmann et al., 2019; Western et al., 2020). Many dairy producers have altered their feeding management practices in an effort to improve their overall farm efficiency and profitability. Differences exist in feeding practices between conventional production systems and AMS; thus, research findings in one type of system cannot necessarily be extended to the other. For example, feeding management practices that have an effect on milk production in conventional settings

include the use of a total mixed ration (TMR), amount of feed offered, frequency of feed delivery, composition of the total diet and others (de Ondarza and Tricarico, 2017).

Meanwhile, some of the feeding management practices which potentially have an effect on milk production in AMS include, but are not limited to: types of concentrate feed offered in the AMS as well as formulation of the partial mixed ration (PMR), quantity offered, location(s) feed is offered, times per day fresh PMR is delivered, frequency and method of feed push up, and so on (Bach et al., 2007; Tremblay et al., 2016; Bach and Cabrera, 2017).

In conventional production systems, one feeding management practice producers utilize to improve efficiency is the TMR, as a way to deliver a completely balanced ration to each cow. The use of a TMR helps ensure each cow receives and consumes a diet formulated to optimize their milk production potential (Coppock et al., 1985). However, within AMS, cows receive feed in two areas generally: within the robotic milking unit, typically as a concentrated pellet; and in the bunk as a PMR. As such, the amount and even type of AMS concentrate fed can be individualized to each cow's unique needs to help minimize fetching and maximize production (Bach and Cabrera, 2017). Current studies have reported mixed milk production responses to increases in concentrate quantities fed in the AMS (Schwanke et al., 2019).

The amount of concentrate fed and/or consumed in the AMS and its relationship with milk production is an ongoing debate and continues to be of significant interest to researchers and producers. It has been suggested that the feed offering in the AMS is

associated with the frequency of visits to the robot (Paddick et al., 2019). With the frequency of visits to the AMS being a prominent factor when determining the efficiency of production as previously stated, it is imperative that research in this area continues. No milk production differences were found when two different amounts of concentrate were offered in an AMS (Bach et al, 2007). Similarly, Paddick et al. (2019) found no milk production response with an increase in the quantity of pellets fed in the AMS. In contrast, a greater AMS concentrate offering was associated with increased milk yield in an observational study of 33 commercial AMS farms (Siewert et al., 2018).

While diet composition is extremely variable from farm to farm, there are a number of nutrients of interest and importance when formulating rations, no matter where the dairy cow lives or in what type of system she is milked. Crude protein (CP), fiber metrics such as neutral detergent fiber (NDF) and neutral detergent fiber digestibility (NDFD), and starch among many others are all common nutrients which need to be analyzed in order to formulate an adequate diet. From forage proportion variation (Li et al., 2020), to the ratio of different types of fatty acids (Western et al., 2020), to different sources of protein (Gaillard et al., 2017) and more; a plethora of published information relating to diet composition and subsequent milk production responses exists in conventional milking systems; however, less research has been conducted on AMS farms.

Forage typically makes up the majority of a dairy cow's diet dry matter and of particular importance, as it relates to fiber intake, is the NDF and NDFD fraction of that forage. NDF is associated with chewing and fiber mat formation in the rumen and is analyzed

carefully due to its impact on rumen function (Balch, 1971). Digestibility of the NDF fraction is determined by measuring the amount of NDF remaining after in-vitro ruminal fluid incubation for a specific amount of time. NDFD has been suggested to be influenced by large amounts of concentrate in the AMS (Miron et al., 2004; Halachmi et al., 2006). Researchers continue to speculate that the feeding behavior changes related to the use of AMS could affect rumen health in multiple ways, but there is a lack of published data to support specific conclusions (Paddick et al., 2019).

Physically effective fiber is also a metric often used to help determine if a diet has an adequate amount of fiber to stimulate chewing (NRC, 2001). Usually physically effective fiber is combined with NDF as physically effective NDF (peNDF) in an effort to explain fiber's physical characteristics through particle length alongside NDF content (Mertens, 1997). Researchers have suggested peNDF and particle size are used interchangeably oftentimes to help describe how adequately a diet will provide the necessary fiber fraction, but go on to say that those terms are not truly interchangeable (Caccamo et al., 2014). Research into the specific association of particle length and peNDF has found a decrease in peNDF as particles decrease in size, thus decreasing chewing time and rumen pH (Woodford and Murphy, 1988).

Since NDF, nor NDFD, nor peNDF can completely explain particle length alone, researchers came up with a simple and repeatable method to define adequate particle length by using the Penn State Particle Separator, abbreviated as PSPS (Lammers et al., 1996). This device allows the separation of feed particles by size utilizing three or

sometimes four screens and appropriate shaking of a specified TMR amount. Heinrichs and Kononoff (2002) suggested (in a three-screen PSPS) that 2-8% of material left on the top screen, 30-50% on the middle screen and 30-50% material on the bottom screen are the needed amounts to maintain adequate chewing and prevent TMR sorting. Knowing and understanding the amount of feed left on each screen allows for the ability to estimate the effectiveness of the fiber portion of a dairy cow's diet (Heinrichs and Kononoff, 2002). One study determining the effect of particle size on milk production found little effect (Caccamo et al, 2014), while another study found a negative effect of increasing particle size on milk yield (Li et al., 2020). No specific research yet exists evaluating the relationship between particle size distribution and milk production responses on AMS farms, but a few studies have investigated sorting behaviors within a larger study objective (Hare et al., 2018; Schwanke et al., 2019).

Crude protein content has been well studied with varying results over the past several decades. One study conducted on an AMS farm found increasing CP in the PMR from 14% to 16% had a positive impact on milk yield and feed intake (Gaillard et al., 2017).

Starch is another nutrient of great importance in dairy diets as it provides much needed energy for milk production, but can also have negative impacts on rumen health and productivity of the cow. Haisan and Oba (2020) evaluated the effects of a high starch AMS pellet in combination with a high fiber PMR compared to a low starch AMS pellet in combination with a high starch PMR and found no milk production difference between treatments; however, rumen fermentation and feed intake patterns were altered,

suggesting the formulation of the PMR should be more of a focus instead of the pellet fed in the AMS (Haisan and Oba, 2020). Interestingly, a study conducted with 27 conventional milking herds in Italy found the effects of starch and NDF are more pronounced than the effects of CP on milk production (Caccamo et al., 2012).

While research in the aforementioned areas of specific nutrient impacts on milk production in conventional settings is substantial, in AMS settings far less research has been published in the literature. The limited research that has been performed in AMS is focused primarily on ingredient substitutions such as fava beans in place of soybean meal (Hansen et al., 2021), barley replaced with glycerol (Gaillard et al., 2017), concentrate limitations (Karlsson et al., 2020), and concentrate to forage ratios in the PMR versus the AMS unit (Menajovsky et al., 2018). For example, high versus low forage PMR showed trends for each treatment to improve milk yield, while increasing day to day variability in AMS concentrate consumption or reducing ruminal pH, and increasing holding area time, leading the authors to no clear answers (Menajovsky et al, 2018). In another study, no difference was found in milk production for early lactation cows fed molasses-based liquid feed in the AMS (Moore et al., 2020). No research has been published, to our knowledge, assessing the relationships of specific nutrients such as crude protein, starch, fiber, etc. or particle size distribution with milk production in AMS.

Feeding behaviors can also influence milk production. Frequency of feed delivery in conventional milking systems has been found to impact feeding behaviors such as dry matter intake (DMI) and TMR sorting with resulting impacts on milk yield (Dado and

Allen, 1994). In fact, increasing feeding frequency from once to twice daily in conventional milking systems increased milk yield linearly in a study conducted by Sova et al. (2014). In contrast, a recent study in a conventional setting investigating feeding frequency of once, twice, or four times daily, found no milk production or milk component responses (Benchaar and Hassanat, 2020). However, these data cannot necessarily be transferred to an AMS farm due in part to having two feeding sources, the PMR and concentrate feeding system, rather than the TMR as the only feed available. Additionally, it has been suggested that AMS cows may act more asynchronously compared to those milked in conventional settings, leading to significant differences in their feeding and lying behaviors and different milk production responses to challenges like overstocking (Witaifi et. al, 2018).

Another feeding management factor with the potential to affect milk yield in AMS is feed push-up style and frequency. Farms that utilized automatic TMR push-up were found to produce more milk per robot and milk per cow than farms who pushed-up feed manually (Siewert et al., 2018). This association could be due to the number of times the feed is pushed up by an automatic pusher versus manually pushing, with automated pushing being more frequent. Otherwise, the frequency of feed push-up has not been found to be associated with milk production in AMS when being pushed up manually between 2 and 8 times per day on average (Deming et al., 2013; King et al., 2016).

Feed tables are optional parameters which can be set within the AMS software where managers are able to utilize individual cow data to determine the amount of concentrate

offered when a specific cow walks in the milking box. In general, feed tables are established on specific days in milk (DIM) and milk production parameters. These tables are one way in which AMS are able to cater to individual cow's nutrient needs, thereby utilizing precision feeding techniques. Precision feeding enables producers and consultants to customize rations for individual cows based on their needs at a certain time in their lactation and can therefore improve the efficiency of milk production (Bach and Cabrera, 2017). The adjustment of feed tables has been suggested by researchers as a way to manage negative energy balance in early lactation cows (King et al., 2018).

Interestingly, researchers have admitted that the use of precision feeding techniques such as frequent feed table changes alongside multiple concentrate options within AMS units, are only a profitable endeavor if substantial variability exists between individual responses and if the economic environment is favorable at that time (André et al., 2010). Those same researchers went on to say that during early lactation those aforementioned criteria are oftentimes met, thus justifying the use of this type of precision feeding during that time, but other stages of lactation did not show the same results (André et al., 2010).

Published research that includes any information on feed tables within AMS is extremely limited. In fact, published research specifically determining the effects or associations of feed table management on milk production does not yet exist to our knowledge. One study utilized three different feeding strategies within the AMS to determine body weight changes and mobilization or deposition of body reserves as well as milk production responses over an extended period of time (Bossen and Weisbjerg, 2009). In the aforementioned study, multiparous cows were suggested to have a slower milk yield

decrease post peak when fed an ad libitum high energy ration in early lactation and then switched to a lower energy ration after a specific body weight increase due to “mobilization metabolism” (Bossen and Weisbjerg, 2009). However, that study changed feeding strategies once a particular body weight was achieved, not with a particular DIM or milk production level as is found in current feed table management practices commercially.

Multiple researchers suggest feed tables are limited by the substitution effect, where cows will not increase overall DMI when offered more concentrate in the AMS; they will simply substitute the feed by eating less PMR and more concentrate in the AMS (Bach et al., 2007; Hare et al., 2018; Menajovsky et al., 2018). These researchers have suggested that a precision feeding strategy may not, in fact, be meeting individual cow’s nutrient requirements as expected. Siewert et al. (2018) found that high producing cows were typically offered larger quantities of concentrate within the AMS versus their lower producing counterparts, but their study did not determine cause and effect of increased concentrate offered and increased milk production. With such limited research on feed tables and their specific role, associations, and/or effects in precision feeding strategies, researchers and producers alike should continue looking for answers and solutions.

MILKING INTERVAL

The amount of time between two consecutive milking events is commonly referred to as milking interval (MI) (Penry et al., 2018). Milking intervals are generally fixed in conventional milking systems where human labor is utilized to milk cows at the same time or times every day. In AMS, milking intervals are not fixed because the system

utilizes a voluntary attendance strategy, which means individual cows choose when they visit the milking box. Variation in MI is substantial in AMS and can be affected by individual cow variation as well as management practices (Hogeveen et al., 2001; Løvendahl and Chagunda, 2011). Ouweltjes (1998) said, “Milk production in terms of milk production per hour is dependent on the milking interval.” Conventional research has found daily milk yield increases with the number of milkings per day, but this statement cannot necessarily be transferred to AMS due to capacity limitations of the milking box (de Koning and Ouweltjes, 2000).

One key to understanding MI is understanding the difference between MI and milking frequency (MF). Milking frequency is the number of times a cow is milked within a 24-hour period. It is therefore possible that, in an AMS, two cows could have very different MI, but have the same average MF. Essentially, MI is the inverse of MF, so a longer MI would be associated with a lower MF and so on (Lyons et al., 2014). The shortcoming of using MF as a factor in AMS research, when attempting to answer questions about milk production as it relates to time spent milking in a 24-hour period, can be found in an example Lyons et al. (2014) shared. These authors stated, “In AMS it is possible that an individual cow milked 2 times within a 24-hour period has both MI of 20 hours, which results in the opportunity for a short MI to compensate for a long MI being removed. Thus, a proportionally lower daily milk yield would be expected in AMS than conventional milking systems, if milking frequency is comparable” (Lyons et al., 2014).

Milking frequency and its correlation with milk yield has been relatively well-studied in conventional milking systems with particular emphasis on the impact of increasing frequency from two to three times daily milkings on milk production. The milk yield response of 3.5 kg/day (7.7 lb/d) was found to be fairly fixed when reviewing published studies which increased MF from two to three times per day, both for primiparous and multiparous cows (Erdman and Varner, 1995). Alternatively, milk yields may be reduced when decreasing the MF, potentially due to a decline in the uptake of nutrients in the mammary gland, thus upsetting the efficiency of the conversion of nutrients into milk components (Davis et al., 1999; Rémond and Pomies, 2005; Delamaire and Guinard-Flament, 2006). Interestingly, in a study by Bernier-Dodier et al. (2010), reduction in MF resulted in lower milk lactose concentrations and rapidly increased bovine serum albumin concentrations. These findings led authors to suggest the lowered MF led to an increase in the permeability of the mammary epithelium, perhaps caused by an increase in the leakiness of the tight junctions (Bernier-Dodier et al., 2010). Those same authors also found increased rate of mammary cell proliferation and a net loss of mammary tissue with reduced MF and suggested this is likely the cause for the negative effect of lowered MF on lactation persistency (Bernier-Dodier et al., 2010).

Some potential dependent factors on the milk production response to MF changes both within AMS and conventional milking systems include but are not limited to: stage of lactation, duration of MF change, and parity (Lyons et al., 2014). The previously stated positive correlations of milk production and MF are typically observed more frequently in early lactation, and as lactation progresses, the response decreases (Pettersson et al.,

2011). It has also been suggested that a late lactation higher MF could compensate for an early lactation lower MF (Svennersten-Sjaunja and Pettersson, 2008).

Changes in MF can also depend on parity, potentially due to udder capacity among other factors (Pettersson et al., 2011). Lower milk yield increases have been reported with higher MF for older cows (Maltz et al., 2003). Similarly, Clark et al. (2006) found milking once per day reduced milk production more for primiparous cows than for multiparous cows. Within AMS, some studies have suggested primiparous cows may achieve higher MF simply due to their inclination to gather around the AMS (Spörndly and Wredle, 2004; Borderas et al., 2008). Milking frequency has also been found to decrease more rapidly in multiparous versus primiparous cows (Pettersson et al., 2011).

While MF has been studied more robustly than MI, which is clearly evident by the examples given within this review, as stated previously they are not interchangeable metrics. Many possibilities exist for MF to be mistaken when not accounting for MI. Possibilities like the aforementioned irregularity of milking intervals leading to somewhat skewed milking frequency data make MF an unreliable source when it is the only metric studied in AMS with regard to when and how often cows are milked. To that end, a few studies have been published looking at both MF and MI and any associations or correlations within. One AMS study found a MF of 2.6 milking events/cow per day, but almost 10% of the milkings had MI less than 6 hours and just over 4% had MI longer than 16 hours (Hogeveen et al., 2001). Another study that investigated milk yield responses to MF of once versus twice daily as well as MI differences of as little as 2.5

hours and as much as 21.5 hours found significantly less milk yield for lower MF and decreasing milk yield for MI less than 5 hours and more than 19 hours (Rémond et al., 2009). Those results, among a handful of others, suggest that a focus needs to be placed on MI rather than only MF, due to the variation found in MI in AMS and the potential for cows to easily adapt to short and long MI (Ouweltjes, 1998; Hogeveen et al., 2001; Rémond et al., 2009; Laurs et al., 2010).

Variability exists between herds but also between individual cows with respect to the effect of MI on milk yield (André et al., 2010). Milk yield will peak at a specific MI, and any interval outside of that peak is likely to result in decreased yield for that milking, decreased total daily milk yield, and will limit the ability of individual cows to meet their full production potential (André et al., 2010). Determining an individual cow's optimal MI along with the optimal MI on each farm could potentially increase milk yield and improve udder health (André et al., 2010; Jensen et al., 2018). It is also possible to consider that cows who exhibit longer MI while still maintaining high daily milk yield could be deemed more efficient (Molfino, 2018).

Some negative effects on milk yield (Lyons et al., 2013a) and udder health (Hammer et al., 2012) have been found with MI longer than 16 hours in AMS. Additionally, research agrees that consistently long MI lead to decreased milk yields (Ipema et al., 1997) and milk flow rate (Hogeveen et al., 2001) in both AMS and conventional settings. Rémond et al. (2009) reported that only extreme variation in MI, such as 3 hours and 21 hours, when compared to a more regular MI like 11 hours and 13 hours, reduced daily milk

yield by 11.5%, suggesting consistent MI could be key to optimizing milk production.

Udder health can also be compromised with a MI less than 8 hours, as teats potentially do not have enough time to recover fully and could be susceptible to more teat damage and/or risk of bacterial invasion (Neijenhuis et al., 2001).

Lyons (2014) suggested that the relationship between milk yield and MI is non-linear therefore, milk production would be reduced after a certain milking interval timeframe and the effect of that reduction related to the animal's stage of lactation and initial milk yield. The exact point in time where that phenomenon occurs has yet to be firmly established, although some older research suggested a MI between 12 hours and 18 hours is where milk yield and MI plateau (Knight et al., 1994; Stelwagen et al., 2008).

Some studies have found higher producing cows visit the AMS more often than lower producing cows, leading to a decreased MI and increased MF (Klaas et al., 2003; König et al., 2006). In a study of MI, milk production, and flow rate in AMS, Hogeveen et al. (2001) found that shorter MI had a larger effect on the increase in milk production per hour in high producing multiparous cows, depending on total milk production of the cow.

Other factors that can impact MI, and the variability of MI, include milking failures and the number of cows, and number of times, cows must be fetched. Milking failures are defined as a time when a cow visits the milking station but fails to be milked due to a lack of attachment or potentially some other error (Siewert et al., 2019). Alternatively, when cows do not visit the milking robot within the amount of time set for them, an alert

is sent and that cow must be manually fetched and pushed to the unit. It would be conceivable to consider a cow with a great number of milking failures may lose interest in the milking unit and extend her milking interval and perhaps become more variable in her milking intervals. Similarly, if a cow must be fetched often her milking interval will be artificially consistent. No research has yet been conducted, to our knowledge, in either of the above-mentioned specific areas.

Many negative effects of changes in milking frequency, and also milking interval, have been found in conventional milking systems. Milk composition changes (Davis et al., 1999), udder discomfort (Gleeson et al., 2007) and reduced persistency of lactation (Hickson et al., 2006) are just a few of the negative outcomes found when MF and MI are increased. As previously stated, as MF and MI are altered, many changes occur within the mammary gland which can lead to both positive and negative outcomes for the cow, production, and overall efficiency. Changing milking intervals clearly has consequences on cow health and milk production in both conventional milking systems and AMS and deserves thorough attention by researchers, consultants and producers alike.

While it remains evident that no single ideal MI exists to maximize milk yield on every AMS farm, it is still a factor which producers and consultants should consider when looking at the efficiency of an operation. With research suggesting a significant impact of MI changes and to further consider how MI impacts the efficiency of milk production, the variability or regularity of MI may be a relevant factor. It has been suggested that sudden changes to a cow's milking interval in AMS could be an indication of underlying

health issues with the animal (Hogeveen et al., 2001; Nixon et al., 2009) thus giving producers and consultants another tool in early detection of potential health problems. In order to utilize this metric in AMS, however, the variability of MI must be known, considered and understood in the context of individual farms and cows.

Only two studies could be found on the topic of variability of MI and its impact on, or association with, milk production in AMS. Bach and Busto (2005) conducted a study on a small AMS farm in Spain where they evaluated the weekly coefficient of variation (CV) of MI as potential determinant for the variability within, or regularity of, cow's milking schedules and whether MI CV had an impact on milk yield. They found a negative impact of an increased weekly MI CV on milk production, meaning the more regularly the cows were milked successfully in the AMS, the more milk they produced (Bach and Busto, 2005). Those authors suggested the highest variability in MI may compromise milk synthesis at the mammary level potentially due to decreased cell proliferation or a decrease in the metabolic activity of secretory cells within the mammary gland (Bach and Busto, 2005). Additionally, high variability in MI was associated with high somatic cell counts in another study comparing 100 farms in the Netherlands (Mollenhorst et al., 2011). Both the aforementioned studies suggest the variability of MI can be associated with milk yield and/or cow health and therefore it is a metric that could provide significant value to consultants and dairy farmers. With only one small published study on MI variability and its effects on milk yield specifically, much research is still necessary to fully understand this topic.

Understanding how the variability of MI is associated with milk production is, as previously stated, an area of very little current research. Evaluating the variability in MI was suggested by Siewert et al. (2019) who used only MF, but did not include MI, in their study with AMS farms. Milking interval variability could have the potential to give researchers, consultants and producers yet another metric in their toolbox when making decisions about milking permission settings, culling decisions, grouping strategies and more. A thorough understanding of the relationships, associations and ultimately the cause and effect of MI and variability of MI on milk production would help continue improving the efficiency of milk production on AMS farms.

SUMMARY

Today's dairy producer is constantly evolving to meet changing and often increasingly difficult consumer demands, environmental concerns, feed and milk price fluctuations and more. In order to maximize milk production and profitability, producers and consultants must determine ways to improve on-farm efficiencies. Improving the efficiency of milk production through feeding management and milking permissions within AMS in particular are paramount to profitability AMS dairy herds. While much research exists in the aforementioned areas in conventional milking systems; far less can be found in the AMS literature.

Previous research in the area of milk production responses to feeding management factors have existed primarily in conventional milking settings, with very little in AMS settings found within the literature. The area of feed table management within AMS has been

avoided altogether in published literature. Meanwhile, some research has been conducted in AMS on MI and subsequent milk production response, yet only two peer reviewed published papers (Bach and Busto, 2005; Mollenhorst et al., 2011) are available, to our knowledge, which specifically discuss the variability of MI. It is important to investigate these areas which have a limited body of research in an attempt to further our knowledge and understanding as well as to facilitate future research.

Therefore, the objective of this thesis is to evaluate the association of AMS feeding management practices (Study 1) and milking interval variability (Study 2) with milk production responses in AMS. Such information provides background and a potential foundation for future research development in these areas. Additionally, these studies allow dairy producers and consultants to further their knowledge on what feeding and milking management practices could have an impact on milk production in AMS herds.

Chapter 2

Association between feeding management practices and milk production on automatic milking system farms

SUMMARY

The impact of feeding management practices on milk production responses on automatic milking system (AMS) dairy farms has not been widely investigated and oftentimes inconclusive or conflicting results have been reported in the literature. Therefore, the objective of the current study was to investigate the association between various feeding management practices and milk production per cow and per robot on AMS farms. Thirty-nine commercial farms in MN and WI using Lely Astronaut AMS were visited over the summer of 2018. Farm managers answered a questionnaire, samples of just-mixed partial mixed ration (PMR) were collected for analysis, and daily cow data for the 30 days prior to farm visit were obtained from the AMS software. Using mixed linear regression models, we found negative associations between average days in milk (DIM) and milk per cow ($P = 0.002$) and milk per robot ($P = 0.002$) as well as average number of cows per robot and milk per cow ($P < .0001$) and milk per robot ($P < .0001$). In contrast, we found positive associations between average number of milkings per day and milk per cow ($P < .0001$) and milk per robot ($P < .0001$) as well as average robot concentrate intake and milk per cow ($P < .0001$) and milk per robot ($P < .0001$). Nutrient composition of the partial mixed ration (PMR) had a negative association for neutral detergent fiber (NDF) content and milk production both per cow and per robot ($P = 0.01$ and $P = 0.03$,

respectively). The amount of material remaining on the middle screen of the Penn State Particle Separator (PSPS) had a slightly positive trend with milk production per cow only ($P = 0.07$). The use of a liquid feed in the AMS also had a negative association with milk production per cow ($P = 0.01$) and per robot ($P = 0.04$). The intensity of feed table management was analyzed using low, moderate and high intensity categories and resulted in a negative association of milk production with less intense management. Low and moderate feed table management intensity were both associated with less milk per cow ($P = 0.004$ and $P = 0.02$, respectively) and per robot ($P = 0.009$ and $P = 0.02$, respectively). Results indicate that some feeding management practices are associated with milk production on AMS farms.

INTRODUCTION

The dilution of maintenance costs by maximizing milk yield is generally considered the primary way to optimize profitability on a dairy farm (Bach et al., 2020). Dairy producers have continually put their focus on ways to mitigate those maintenance costs in particular (Knoblauch et al., 2012). Many dairy producers have altered their feeding management practices in an effort to improve their overall farm efficiency and profitability thanks to substantial research in this area in conventional milking systems. However, many differences exist in feeding practices between conventional production systems and automatic milking systems (AMS), thus research findings in one type of system cannot necessarily be extended to the other.

Box system AMS cows are offered feed in two areas - within the robotic milking unit typically as a concentrated pellet and in the feed bunk as a partial mixed ration (PMR). As such, the amount and type of concentrate fed can be individualized to each cow's unique needs through the use of precision feeding techniques like feed table management to help minimize fetching and maximize production (Bach and Cabrera, 2017; King et al., 2018). In general, feed tables are established on specific days in milk (DIM) and milk production parameters.

Current studies have reported mixed milk production responses to increases in concentrate quantities fed in the AMS with some having found inconclusive results or no differences (Bach et al., 2007, Paddick et al., 2019; Schwanke et al., 2019) and one study having found an improvement in milk yield with an increase in concentrate offered (Siewert et al., 2018). Along with the quantity of concentrate fed, the nutrient and ingredient composition of concentrate and PMR being fed can also affect milk production responses. Limited to no research has been published, to our knowledge, assessing the relationship of specific nutrients such as crude protein (CP), starch, fiber, etc. with milk production in AMS. Similarly, literature including any information on feed tables within AMS is extremely limited and published research specifically determining the effects or associations of feed table management on milk production does not yet exist. One study utilized three different feeding strategies within the AMS to determine body weight changes and mobilization or deposition of body reserves as well as milk production responses over an extended period of time (Bossen and Weisbjerg, 2009). However, Bossen and Weisbjerg's (2009) study changed feeding strategies once a particular body

weight was achieved, not with a particular DIM or milk production level as is found in current feed table management practices commercially. Multiple researchers suggest feed tables are limited by the substitution effect, where cows will not increase overall DMI when offered more concentrate in the AMS; they will simply substitute the feed by eating less PMR and more concentrate in the AMS (Bach et al., 2007; Hare et al., 2018; Menajovsky et al., 2018).

In addition to the limited number of studies in the literature on feeding management practices within AMS, those that have been published oftentimes have conflicting or inconclusive results. Combined, the mixed results of feeding management practices with the lack of publications investigating feeding table management altogether, suggest further research in these areas is warranted. Therefore, the objective of the current study was to investigate the association between feeding management practices and milk production per cow and milk production per robot on commercial AMS farms in the Upper Midwest USA.

MATERIALS and METHODS

Farms and Data Collection

Thirty-nine dairy farms with a total of 3,742 cows located in Minnesota and Wisconsin using AMS as the only milking system were enrolled in the current observational study. Potential farms were identified with the help of extension educators, consultants, equipment dealers, and producers. After identification of farms, producers were contacted and participation in this study was voluntary. Farms used Lely Astronaut (Lely Industries

NV, Maassluis, the Netherlands) robots to milk their cows. Herds enrolled in this study were comprised of Holsteins and all the cows were housed in freestall barns with no access to pasture.

Each farm was visited once for data collection during the summer of 2018, when a standardized questionnaire was answered by the farm manager, which included questions on feeding practices, such as: number of daily feedings (1, 2 or 3+), feed push-up method (automated or manual), number of concentrate feeds offered in the AMS box, and offer of a liquid feed for fresh cows (yes/no). At the same farm visit, feed table information was collected; an example of a feed table can be found in Table 1. Prior to the farm visit, farm personnel were contacted and asked to collect a sample of just-mixed partial mixed ration (PMR). The PMR samples were sent for NIR analysis to Dairyland Laboratories, Inc. (Saint Cloud, MN) and were also used in the 3-sieve Penn State particle size separator (PSPS, Lammers et al., 1996) to analyze particle size distribution.

Retrospective daily farm data were collected from the AMS software (T4C, Lely Industries), including animal identification, date, lactation number, days in milk, milk production (kg), number of milkings, and concentrate intake in the AMS (kg).

Data Processing and Statistical Analysis

All post-collection data management procedures and statistical analyses were performed in RStudio (R Core Team, 2020). Software data were aggregated into farm daily values to create the daily average lactation number, days in milk, number of milkings, milk

production, and concentrate intake per cow. The final data set had 30 daily observations per farm (1,170 total observations), which represented the 30 days prior to the farm visit.

Multiparous cow's feed tables were used for analysis. The tables were categorized into management styles of low, moderate and high intensity. The criteria for categorizing feed tables in AMS was developed by the study authors as there is no prior reference in the literature. Categorization was done by visually inspecting each farm's feed table data and determining the number of different feed options (pellets/concentrate) offered (1, 2 or 3) within the AMS, the maximum number of concentrate amounts offered coinciding with milk production levels (up to 12) and the maximum number of times feed tables were changed during an entire lactation (up to 12). The maximum number of varying concentrate amounts offered was determined by counting the number of different concentrate amounts that a farm had established on their feed tables for specific milk production levels. For example, one farm offered seven different amounts of concentrate during early through mid-lactation (according to cow production level), during other times of lactation the farm only offered one concentrate amount. Thus, that farm's maximum number of amounts offered is seven. This farm only offered one pellet option in the AMS box and they changed their feed tables three times throughout an entire lactation. An example of a feed table representing this farm is shown in Table 1. This farm was categorized as low feeding intensity.

Once all of those criteria for each farm were documented, the numbers were added together to determine a complete feed table management intensity total. Using the above

farm's example, we took the maximum number of feed options (pellet/ration) of 1, plus the maximum number of concentrate amounts offered of 7, plus the number of times the feed table was changed throughout a lactation of 3 to equal a total feeding management intensity of 11 which coincided with a "low" categorization of their feeding management intensity. Using this method, all farms ended up with a calculated total that coincided with the intensity of their management. The number of farms within each management intensity style and the range of totals is as follows: low intensity management included 14 farms with an intensity range of 9-12, moderate intensity management included 18 farms with a range of 13-16, and high intensity management included 7 farms with a range of 17-21.

To assess the association between feeding management practices and average milk production per cow and per robot, the lmer function of the lmerTest package (Bates et al., 2015) was used to generate mixed linear regression models for those two outcomes of interest. The initial models included all explanatory variables at once; farm was included as the random effect. Correlations between explanatory variables were assessed and were all under 0.7, which has been suggested as a threshold for inclusion in the same model (Dormann et al., 2012; Tremblay et al., 2016).

Initial explanatory variables were: PMR dry matter, crude protein, neutral detergent fiber (NDF), NDF digestibility at 240 hours in vitro (NDFD 240), and starch content; PSPS top sieve, middle sieve, and bottom pan amounts; number of daily PMR feedings; feed push-up method (automated or manual); feed table management intensity (high, moderate,

low); number of concentrate feeds offered in the AMS box (1, 2, or 3); and offer of an AMS liquid feed for fresh cows (yes/no). These factors were chosen due to being deemed by the authors as related to feeding and/or feeding management practices.

Farm average lactation number (parity), average days in milk, average number of milkings, and number of cows per AMS were included in the initial multivariable model as covariates.

Backward elimination was used to remove non-significant factors until all remaining variables had a $P < 0.1$. Model fit was assessed by visual observation of residual plots. The denominator degrees of freedom were estimated using Satterthwaite's method. The Tukey P -value adjustment was used for pairwise comparisons. Statistically significant differences were declared with probability values equal to or less than 0.05 and trends were declared with probability values greater than 0.05 and equal to or less than 0.10. Ninety-five percent confidence intervals (95% CI) along with P values were reported for all findings of significance.

RESULTS and DISCUSSION

Descriptive Statistics

We used 1,170 total observations in the current study, representing 30 days of daily data per farm prior to the on-farm visit. Milk production per cow averaged 37.9 ± 4.4 kg/day and milk production per robot averaged $2,286 \pm 345$ kg/day. Average lactation across all farms was 2.28 ± 0.22 . DIM averaged on all farms was 171 ± 25 . Number of daily

milkings on average was 2.82 ± 0.22 . Concentrate intake in the AMS averaged 5.3 ± 0.8 kg/cow per day. Number of cows per robot averaged 60.2 ± 5.6 . Dry matter of the PMR averaged $45.7 \pm 6.5\%$, crude protein averaged $15.8 \pm 1.1\%$, NDF content averaged $34.3 \pm 2.9\%$, NDFD240 averaged $64.9 \pm 4.5\%$ and starch content of the PMR averaged $22.8 \pm 3.9\%$. The 3-sieve Penn State particle separator results had an average of $23.9 \pm 12.4\%$ on the top screen, $36.8 \pm 9.4\%$ on the middle screen and $39.1 \pm 6.8\%$ on the bottom screen.

Parity was eliminated in the backward elimination steps of our analysis. We believe this is due to our data being at the farm level, versus at the cow level, thus most farms could be very similar in parity average and responses across our dataset. Other factors that were eliminated included PMR dry matter, PMR crude protein, PMR NDFD 240, and PMR starch, PSPS top sieve and bottom pan, number of daily feedings, feed push-up method, and number of concentrate feeds offered in the AMS box.

Table 2 shows the results for all significant associations for milk production per cow and milk production per robot mixed models.

Days in Milk

Average DIM was 170.7 ± 25.5 across all farms. Days in milk was negatively associated with milk production per cow and per robot (Figures 1 and 2, respectively). Every one day increase in DIM was associated with 0.02 kg/cow less milk per day (95% CI: -0.03, -0.008, $P = 0.002$) and 1.3 kg/robot less milk per day (95% CI: -2.09, -0.5, $P = 0.002$).

These findings are in agreement with other studies, although some studies were conducted using cow instead of farm as experimental unit. A study utilizing 13 AMS herds with cows averaging 144 ± 92 DIM found that milk yield per cow was negatively associated with DIM (Deming et al., 2013). Another study utilizing 33 AMS dairy farms where the median DIM was 173 also found a negative association between average DIM and milk yield per cow, but not for milk per robot (Siewert et al., 2018). In another study with a large dataset, where average DIM was 177.7 ± 27.9 , a similar negative association existed with increasing DIM decreasing milk per cow and milk per robot (Tremblay et al., 2016).

Number of Milkings per Day

The average number of milkings per day was 2.82 ± 0.22 . A positive association was found between number of average milkings per day and milk production per cow and milk production per robot (Figures 3 and 4, respectively). For every additional milking per day, milk production per cow increased 2.98 kg/day (95% CI: 2.45, 3.51, $P < .0001$) and milk production per robot increased 157.7 kg/day (95% CI: 125.9, 189.52, $P < .0001$). These findings are in agreement with other studies which also showed positive associations between number of milkings per day and milk production (Løvendahl and Chagunda, 2011, Tremblay et al., 2016; Siewert et al., 2018). In fact, the milk yield response of 3.5 kg/day (7.7 lb/d) was found to be fairly fixed when the number of milkings was increased from two to three times per day in a conventional milking system (Erdman and Varner, 1995).

It is important to note that the number of milkings per day can be affected by many factors. Lower milk yield increases have been reported with a higher number of milkings per day for older cows compared to younger cows in conventional settings (Maltz et al., 2003). Similarly, Clark et al. (2006) found milking once per day dropped milk production more for primiparous cows than for multiparous cows. Within AMS, some studies have suggested primiparous cows may achieve higher number of times milked per day simply due to their inclination to gather around the AMS unit (Spörndly and Wredle, 2004; Borderas et al., 2008). Milking frequency has also been found to decrease more rapidly in multiparous compared to primiparous cows (Pettersson et al., 2011).

Cows per Robot

Number of cows per robot averaged 60.2 ± 5.6 across all farms and was negatively associated with milk per cow but was positively associated with milk per robot (Figures 7 and 8, respectively). For every additional cow per robot, milk per cow decreased 0.13 kg/day (95% CI: $-0.18, -0.09$, $P < .0001$). This milk production response, while significant, is likely not biologically relevant due to the size of response. Meanwhile, for every additional cow per robot, milk per robot increased 29.2 kg/day (95% CI: 26.63, 31.82, $P < .0001$). Siewert et al. (2018) averaged 55.8 ± 6.1 cows per AMS and found positive associations between the number of cows per robot and milk yield per cow per day. Tremblay et al. (2016) averaged 51 cows per robot and found positive associations between the number of cows per robot and milk production per cow but did not find an association for milk production per robot. Our results could indicate that as the number of cows per robot increases, cows might not be as productive individually; but with more

cows per robot the total production for each robot could conceivably still increase, despite individual cow production being lowered.

Feeds and Feeding Management

Concentrate intake in the robot averaged 5.31 ± 0.79 kg/cow per day in our study.

Average concentrate intake was positively associated with milk production per cow and per robot (Figures 5 and 6, respectively). For every additional kg of concentrate eaten, milk production per cow increased 1.78 kg/day (95% CI: 1.41, 2.17, $P < .0001$) and milk production per robot increased 110.7 kg/day (95% CI: 88.17, 133.65, $P < .0001$). Siewert et al. (2018) found a positive association between average concentrate offered/cow per day and daily milk yield per cow in a study with 33 AMS farms. Interestingly, another study of 635 North American dairy farms found an increase in AMS concentrate was associated with decreased milk production, although the authors suggested that was due to the dataset being from a geographical region with poor quality forage options requiring large quantities of concentrates in the robot then leading to increased refusals and failures (Tremblay et al., 2016). These findings along with other research also showing inconsistent results continues to suggest there is a point of diminishing returns on the amount of concentrate feed offered in the robot, with milk production increasing up to a certain point but then decreasing (Bach et al., 2007; Bach and Cabrera, 2017; Hare et al., 2018; Paddick et al., 2019). The exact threshold for this response has yet to be determined and additional research is warranted.

The use of a liquid feed in the robot had a negative association with milk production both per cow and per robot. If a liquid feed was used, milk per cow decreased 4.08 kg/day (95% CI: -6.89, -1.27, $P = 0.01$) and milk per robot decreased 201.7 kg/day (95% CI: -374.73, -28.61, $P = 0.04$). Only 6 of the 39 farms in this study used a liquid feed, therefore it is possible the small sample size impacted our results. Moore et al. (2020) found that offering a molasses-based feed supplement to cows in the AMS had no impact on milk yield, but their results suggested supplementation could help maintain energy balance in early lactation and reduce incidence of ketosis. No further published research could be found specifically detailing liquid feed supplementation in the AMS and any associations or impacts on milk production. We suggest it is possible a negative association was found in the current study due to the type, nutrient composition, amount and palatability of the liquid feed along with the impact of the liquid on the pellet offered with the liquid in the AMS. It is conceivable that a liquid feed could alter the physical characteristics of the pellet offered within the AMS, potentially making it less palatable to the cow, thus decreasing intakes and lowering milk yield. It would also be possible that the liquid feed offered was not as nutrient dense as the other feeds offered, thus cows receiving a liquid supplement would be eating a less dense nutrient profile as those not receiving the liquid.

Low and moderate feeding intensity had negative associations with milk production per cow and per robot (Figure 11). Farms with low feeding management intensity had 4.62 kg/day less milk per cow (95% CI: -7.38, -1.86, $P = 0.004$) and 262.61 kg/day less milk per robot (95% CI: -438.40, -86.61, $P = 0.009$) than their high management counterparts.

Meanwhile farms with moderate feeding management intensity had 3.54 kg/day less milk per cow (95% CI: -6.61, -0.78, $P = 0.02$) and 228.86 kg/day less milk per robot (95% CI: -404.12, -53.52, $P = 0.02$). The aim of the feeding management intensity analysis specifically was to determine if a more precise feeding regimen, geared toward more individual cow feeding approaches, had an association with milk production. The answer to that question could assist producers and consultants with management decisions and where a herd manager's time could be most impactful. Our results suggest taking a more individual cow-focused approach with feed tables, with higher management and time allocated to this area, in an effort to impact milk production positively. However, this is the first research of its kind to our knowledge and should be interpreted carefully.

Inclusion of NDF and NDFD240 in our model was due to the impact fiber has on chewing stimulation, saliva production, ruminal pH, rate of passage and fiber mat formation which are important to explore (Balch, 1971). NDF content of the PMR averaged $34.3 \pm 2.9\%$ in our study. Our results showed a negative association of NDF content with milk production per cow and per robot (Figures 9 and 10, respectively). For every additional percent of NDF, milk production per cow decreased 0.45 kg/day (95% CI: -0.76, -0.13, $P = 0.01$) and milk production per robot decreased 24.8 kg/day (95% CI: -44.86, -4.68, $P = 0.03$). These results coincide with many studies as NDF has been found to limit DMI and/or increase satiety due to its rumen fill effects (Mertens, 1987; Allen, 1996; Oba and Allen, 1999). One study suggested feeding a higher NDF PMR reduced the amount of time the rumen pH was below 5.8 and did not have an effect on intake, unfortunately it also had no effect on milk production; however, the healthier rumen

could certainly be seen as advantageous (Haisan and Oba, 2020). A limitation of the current study includes no available data for physically effective NDF content, which could be an impactful metric due to its potential impact on milk yield versus NDF alone (Cao et al., 2021). We had data on NDFD240 which describe the digestibility of NDF at 240 hours of in vitro fermentation by measuring the amount of indigestible NDF remaining, a metric that has been found to significantly impact milk production, and it was determined to not be significant in our model (Oba and Allen, 1999; Fustini et al., 2017).

The amount of material on each screen of the 3-sieve PSPS had a positive associative trend in our milk per cow model, and only for the middle screen (Figure 10). For every additional percent of material on the middle screen, milk per cow increased by 0.10 kg/day (95% CI: 0.0001, 0.21, $P = 0.07$). This relatively small finding suggests milk production responses to the PSPS could be quite different from cow to cow, since the milk per robot model was not significant. Similar findings of positive associations with particle size and milk production in conventional milking systems have been found. Li et al. (2020) found that an increase in particle size as determined by retention of particles on the top two sieves of the PSPS decreased milk yield and Haselmann et al. (2019) also found that smaller particle sizes increased milk yield. Some AMS studies investigated sorting behavior utilizing a PSPS, but none specifically determined an association or cause and effect of specific amounts of material on each screen with milk production (Paddick et al., 2019; Schwanke et al., 2019; Haisan and Oba, 2020). Much difficulty exists in comparing AMS research that looks at PSPS data due to the variation in nutrient

composition of the diets and amount of concentrate fed in the robot. If cows are fed a large quantity of a high starch feed in the robot, they would likely sort in favor of longer particles in the PMR in an effort to fulfill their effective fiber needs. This theory was found to be true in one study where cows fed a larger quantity of a high starch pellet in the AMS ate more PMR and sorted in favor of longer particles (Hare et al., 2018).

The average percent of material left on the PSPS in our dataset was 23.9% on the top which coincides with the longest particles, 36.9% on the middle screen, and 39.1% on the bottom screen which are the finest particles. Heinrichs and Kononoff (2002) suggested 2-8% of material left on the top screen, 30-50% on the middle screen and 30-50% material left on the bottom screen to maintain adequate chewing; thus, our farms appear to have longer particles on average than is suggested, which was expected since AMS farms feed a PMR where all forage is included, and concentrate is supplemented in the AMS box. With higher NDF having a negative impact on milk production as well as the middle screen of the PSPS having a trend for slightly more milk per cow, our results indicate the more indigestible portion of fiber may be driving the negative milk production association. However, some longer particle sizes (middle screen) which also have lower digestibility than concentrate, were slightly associated with higher milk production. These findings help confirm the importance of fiber in a diet and how the digestibility and particle sizes can have an impact on milk production both positively and negatively.

CONCLUSIONS

The associations with milk production found within this study could contribute to the body of literature and assist future researchers as many areas are novel. Optimizing feeding management by altering feed tables more often to account for more individualized feeding strategies could increase milk production per cow and per robot. Using a liquid feed within the robot and an increase in the NDF content of the PMR decreased milk production per cow and per robot. A trend was found for a positive association of amount of PMR on the middle screen of the PSPS and milk production per cow but not milk production per robot. Additional research is necessary to determine cause and effect of these variables.

Association between milking interval regularity and milk production on automatic milking system farms

SUMMARY

The amount of time between two consecutive milking events is commonly referred to as milking interval (MI). In contrast to conventional milking systems, in automatic milking systems (AMS), MI are not fixed due to the voluntary nature cows are milked. Limited research has been conducted on the variability of a cow's MI in AMS. Therefore, the objective of this study was to determine the association between the variability of MI and milk production per cow per day on commercial AMS farms in the Upper Midwest US. Fifty-two farms using AMS as their only milking system were enrolled in the study. Cow-level MI (hours), average number of milking failures per week (0, 1 or >1), parity (1, 2, 3+), and average milk production per day (kg) were obtained for each cow's first 91 days in milk corresponding with weeks in milk (WIM) 1 through 13 post-calving. To determine the variability of MI, coefficient of variation (CV) was mathematically calculated for each cow per day. Mixed linear regression models found that the association of MI CV on milk production per cow per day was dependent upon average milking failures, parity and week in milk. A negative association with milk production was found as MI CV increased for cows averaging 1 failure per week ($P < .0001$) and for parity 3 and greater cows ($P < .0001$). Alternatively, a positive association with milk production per cow per day ($P = 0.009$) as MI CV increased was found for parity 1 cows.

Milk production per cow per day had an associated, but non-linear, decrease each week in milk ($P < .0001$ for weeks 2 through 9, $P = 0.001$ for week 10, $P = 0.0003$ for week 11) except for weeks 12 and 13 when only trends existed ($P = 0.10$ and $P = 0.05$ respectively) as MI CV increased. A limitation of this study was lack of detailed cow fetching data. Our results suggest a negative association between MI CV and milk production exists but is dependent upon failures, parity and week in milk and should be further examined to determine cause and effect.

INTRODUCTION

The amount of time between two consecutive milking events is commonly referred to as milking interval (MI). Milking intervals are generally fixed in conventional milking systems where human labor is utilized to milk cows at the same time(s) every day, whereas in automatic milking system (AMS) farms MI are variable due to the voluntary nature of the system. Individual cow variation as well as management factors can impact the variation of MI in AMS (Hogeveen et al., 2001; Løvendahl and Chagunda, 2011). Research in conventional milking systems has found daily milk yield increases with greater number of milkings per day, but this statement cannot necessarily be transferred to AMS due to capacity limitations of the box (de Koning and Ouweltjes, 2000).

Milking frequency (MF), the inverse of MI, is the number of times a cow is milked within a 24-hour period (Lyons et al., 2014). Milking frequency and its correlation with milk yield has been relatively well studied in conventional milking systems with particular emphasis on increasing frequency from two to three times daily milkings and

the impact on milk production. The milk yield response of 3.5 kg/day (7.7 lb/day) was found to be fairly fixed when reviewing published studies that increased MF from two to three times per day milking in both primiparous and multiparous cows (Erdman and Varner, 1995). Alternatively, milk yields can be reduced when decreasing the MF potentially due to a decline in the uptake of nutrients in the mammary gland and thus upsetting the efficiency of the conversion of nutrients into milk components (Davis et al., 1999; Rémond and Pomiès, 2005; Delamaire and Guinard-Flament, 2006).

Determining the optimal MI for each cow and farm could potentially increase milk yield and improve udder health (André et al., 2010; Jensen et al., 2018). It is also possible to consider that cows with longer MI while still maintaining high daily milk yield could be deemed more efficient (Molfino, 2018).

Rémond et al. (2009) determined only extreme variation in MI, such as 3 hours and 21 hours, when compared to a more regular MI like 11 hours and 13 hours, reduced daily milk yield by 11.5%, suggesting consistent MI could be key to optimizing milk production. Only two studies could be found in the literature on the topic of variability within milking interval. High variability in MI was associated with high somatic cell counts in a study comparing 100 farms in the Netherlands (Mollenhorst et al., 2011). Bach and Busto (2005) conducted a study on a small AMS farm in Spain where they evaluated the weekly coefficient of variation (CV) of MI as potential determinant for the variability within, or regularity of, cow's milking schedules and found a negative impact on milk production of an increased weekly MI CV. Both the aforementioned studies

suggest the variability of MI can have an impact on milk yield and/or cow health; however, both studies were the first of their kind and warrant further investigation. Therefore, the objective of this study was to investigate the association between the variability of milking interval and milk production per cow per day on commercial AMS herds in the Upper Midwest US.

MATERIALS and METHODS

Farms and Data Collection

Fifty-two dairy farms located in Minnesota and Wisconsin using AMS as the only milking system were enrolled in the current observational study. Potential farms were identified with the help of extension educators, consultants, equipment dealers, and producers. After identification of farms, producers were contacted and participation in this study was voluntary. Farms used Lely Astronaut (Lely Industries NV, Maassluis, the Netherlands) or DeLaval VMS (DeLaval International AB, Tumba, Sweden) systems to milk their cows. Herds enrolled in this study were comprised of Holsteins and all the cows were housed in freestall barns with no access to pasture.

Retrospective daily cow data (n = 5,728 cows) were collected from the respective AMS software for one year or six months (T4C, Lely Industries; DelPro, DeLaval International, respectively), including animal identification, date, lactation number, days in milk, milk production (kg), milking interval in hours (MI), number of milkings and number of milking failures.

Cows on all farms had milking permissions set to allow milking every 4 hours at a minimum during the lactation period analyzed in the current study (91 days in milk), allowing cows to visit often and not substantially limiting milking interval. Fetching generally occurred after a cow had not been to the milking unit for the previous 12 hours. Unfortunately, we did not have access to specific cow-level daily fetching data in order to add that metric to our model.

Data Processing and Statistical Analysis

All post-collection data management procedures and statistical analyses were performed in RStudio (R Core Team, 2020). Cow-level MI (hours), category of milking failures per week (0, 1 or >1), parity (1, 2, or 3+), and average milk production per day (kg) were obtained for each cow's first 91 days in milk corresponding to weeks 1 through 13 post-calving. To determine the association of milk production over time in this study, we chose to use weeks in milk (WIM) in the model. The final dataset was comprised of 74,464 total observations.

To assess our objective of determining the association between milking interval variability and average milk production per cow per day, the lmer function of the lmerTest package (Bates et al., 2015) was used to generate mixed linear regression models. Milk production per cow per day was the outcome variable, and all explanatory variables were added to the initial model at once; farm was included as a random effect. Variables included milking interval coefficient of variation (MI CV), category of milking failures per week (0 failure, 1 failure or >1 failure), parity (1, 2 or 3 and greater), and

week in milk (WIM, 1 through 13). Milking interval coefficient of variation was determined by the mathematical equation for coefficient of variation: standard deviation of MI divided by the mean of MI. Milking failures were recorded by the AMS software (T4C, Lely Industries). Cows were categorized to one of three levels of failures corresponding with the number of average failures they had weekly: 0 failures, 1 failure, or greater than 1 failure. In addition, the interactions of MI CV and failures, MI CV and parity, as well as MI CV and week were analyzed. Model fit was assessed by visual observation of residual plots. The denominator degrees of freedom were estimated using Satterthwaite's method. Statistically significant differences were declared with probability values equal to or less than 0.05 and trends were declared with probability values greater than 0.05 and equal to or less than 0.10. Ninety-five percent confidence intervals (95% CI) along with *P* values were reported for all findings of significance.

RESULTS and DISCUSSION

Descriptive Statistics

Averages across all observations are as follows: average milking interval was 8.30 ± 2.22 hours, average milking interval coefficient of variation was 0.24 ± 0.10 , average milk per day was 43.9 ± 11.9 kg. Zero weekly average milking failures comprised 69.9% of the dataset, 1 average weekly milking failure comprised 27.3% and more than 1 failure per week on average was just 2.8% of the observations. Parity 1 comprised 28.7% of the data, parity 2 cows were 36.8% of the data and parity 3+ cows were 34.5%.

Milking Interval Variability

We found a negative association of average milk production per cow/day and MI CV as seen in Figure 12. For every unit increase in the MI CV, milk production decreased 4.51 kg/day (95% CI: -5.26, -3.76; $P < .0001$). This finding is in agreement with the only other published study on the association of MI CV with milk production (Bach and Busto, 2005). However, the latter study grouped MI CV into four categories coinciding with the four quartiles of their respective distributions and found the milk yield decreased most drastically for cows in the highest MI CV category ($>27.3\%$) (Bach and Busto, 2005). We used MI CV as a continuous variable and our average was 0.24 (the equivalent of 24%) with a standard deviation of 0.10 (the equivalent of 10%), our median 0.22 (the equivalent of 22%), range of 0.02 to 1.33 (2.0 to 133% equivalent). Bach and Busto (2005) had categories ranging from $< 18.5\%$ to $> 27.3\%$, which would be extremely similar to our findings. However, their average milking interval was 12.06 ± 4.07 hours whereas we found a shorter MI of 8.30 ± 2.22 hours. In our study we used data from only the first 91 days in milk, when cows visit the AMS more frequently. In addition, Bach and Busto (2005) used one herd of 83 cows in Spain over a 10-month period of time and our dataset consisted of 52 farms and over 5,000 cows in the US where management practices may differ.

It is very important to note that this main effect is to be taken with strong consideration of the interaction effects. The association of MI CV and milk production per cow was dependent on a number of other factors as described below. Statisticians appear to have opposing opinions on the use or not of main effects when interactions are present

(Finney, 1948; Bradu and Gabriel, 1974; Gelman and Vehtari, 2021), thus we chose to include both main and interaction effects in our results and discussion.

Milking Failures

The inclusion of failures in the dataset was important since an incomplete milking could lead to or have an impact on many other factors, such as a lower-than-normal MI and higher-than-normal MI CV. If a cow is incompletely milked her udder capacity might be limited and she could need to visit the robot again sooner than had her udder been completely emptied. Failures have been found to disturb a cow's time budget as she will likely return several hours sooner to the robot than her fully milked counterparts (Stefanowska et al., 2000).

Milking failures can happen for a number of reasons and once they occur, they could potentially have lasting consequences. Milk leakage due to milking failure is one concern that can lead to an increased mastitis risk (Stefanowska et al., 2000; Persson Waller et al., 2003). Oftentimes primiparous cows in particular have a difficult time adjusting to the robot which can lead to a higher level of failures due to kicking and other behaviors in the robot (Miller et al., 1995).

A negative association with milk production per cow per day was found when comparing average milking failures of zero to more than 1 milking failure (Figure 13). More than 1 failure on average per week was associated with 2.98 kg/day less milk (95% CI: -3.97, -1.99; $P < .0001$). Interestingly, 1 failure compared to zero failures was associated with a

slight increase in milk per cow per day of 0.48 kg (95% CI: 0.13, 0.83; $P = 0.006$).

However, an interaction between failures and MI CV was found and as such this main effect result should be interpreted carefully.

Interaction of MI CV and Failures

A negative interaction of MI CV and milk production per day was found when cows had an average of 1 failure per week. As seen in Figure 14, for every unit increase in MI CV when cows experienced an average of 1 milking failure per week, there was a decrease of 4.62 kg/day (95% CI: -5.89, -3.36; $P < .0001$) in milk production. The association of high milking failures (> 1 failure per week on average) and MI CV on milk production was not significant ($P = 0.18$).

Interestingly, our main effect association was between cows averaging more than 1 failure per week but our interaction association is between cows averaging 1 failure per week. This tells us that MI CV is impacted more by cows averaging 1 failure per week than cows who average a higher number of failures. Perhaps this is because cows with high failures end up needing to be fetched often, giving them a more consistent MI CV than otherwise. It is also possible this result was due to our sample size since our dataset was comprised of zero failures representing 69.9% of the dataset, 1 failure at 27.3% and more than 1 failure was just 2.8% of the observations.

Although Bach and Busto (2005) evaluated milking failures in their study, they did not include failures in their milking interval variability model and thus we have no data to

compare. Any milkings with failures and the two milkings thereafter were removed from the dataset to avoid potential confounding effects (Bach and Busto, 2005).

Weeks in Milk

Milk production by weeks in milk can be seen in Figure 15. Comparing to week 1 in milk, which would coincide with days 1 through 7 post-calving, all weeks thereafter had a positive association with milk production per cow/day (Table 3 and Figure 16). Week 2 had the smallest positive association with milk production as would be expected, compared to week 1, with an increase of 11.21 kg/day (95% CI: 10.50, 11.92; $P < .0001$). The largest associated milk production increase was at week 6 compared to week 1, with an increase of 20.72 kg/cow per day (95% CI: 19.96, 21.47; $P < .0001$). These linear results are to be expected as milk production is well known to slowly increase over time until peak lactation is reached somewhere between 40 and 60 or more days in milk (García and Holmes, 2001; Macciotta et al., 2005).

Interaction of MI CV and Weeks in Milk

The association of milk production per cow per day and variability of MI was dependent upon WIM. For every unit increase in MI CV, a negative milk production response was found for each week (Table 4). These results are in agreement with previous studies that also found negative responses with increasing MI CV (Bach and Busto, 2005; Mollenhorst et al., 2011). It is possible that the variability of MI had a negative impact on the time budgets of cows who are generally creatures of habit and the result was reduced milk production. Our results indicate the greatest reduction in milk production

with increasing MI CV occurred within the first 6 weeks in milk, suggesting perhaps cows who work themselves into a consistent daily pattern early in lactation could produce more milk. Research agrees that cows prefer consistency in their routines and that consistency can lead to calmer, more productive cows (Rasmussen and Frimer, 1990; Moran and Doyle, 2015; Beggs et al., 2018).

The largest associated milk production per cow/day decrease occurred specifically during week 3, where for every unit increase in MI CV milk production decreased 13.00 kg (95% CI: -15.65, -10.34; $P < .0001$). One possible explanation for this large milk production loss in week 3 could be worsening health conditions that are associated with an increase in MI CV. Cows who are sick might be more variable in their MI as they are potentially not willing to eat as often or stand for a longer period of time waiting to enter the AMS unit. In general, research has shown that resting time is lengthened and physical activity as well as feeding behaviors are more limited when cows are sick (Broom, 2006; Harden et al., 2015; Dittrich et al., 2019).

Parity

Parity 3 and greater cows (Figure 17) produced 4.78 kg/day more milk than their parity 2 counterparts (95% CI: 4.43, 5.14; $P < .0001$). Meanwhile, parity 1 cows produced 12.53 kg/cow per day less than parity 2 cows (95% CI: -12.89, -12.16; $P < .0001$). Other recent AMS research coincides with our findings of increased milk yield for multiparous cows compared to primiparous cows (Siewert et al., 2019; Adriaens et al., 2021). Interaction

effects are present for parity and MI CV and so these results should be carefully considered.

Interaction of MI CV and Parity

For parity 3 and greater cows compared to parity 2 cows (Figure 18), every unit increase in MI CV was associated with a decrease in milk production per cow of 4.62 kg/day (95% CI: -7.49, -4.73; $P < .0001$). This finding is in agreement with Bach and Busto (2005) who found a reduction in milk production with increasing MI CV in multiparous cows in their study.

In contrast, for parity 1 cows compared to parity 2 cows, every unit increase in MI CV was associated with a milk production increase of 1.95 kg/day per cow (95% CI: 0.48, 3.42; $P = 0.009$). Bach and Busto (2005) found primiparous cows with high MI CV decreased milk production, but no other significant responses existed with those animals. Our results are interesting and would benefit from further analysis to determine cause and effect; however, with the observational information available to us, we suggest this result may be due to fetching rates being high for parity 1 cows compared to older cows. Primiparous cows, who are new to the AMS, usually take a longer time to learn and require frequent fetching particularly in the first few weeks of lactation. Previous research found parity 1 cows were more likely to be fetched than parity 2 or parity 3+ cows (King et al., 2017). If cows must be fetched their MI CV would be artificially consistent and it is conceivable to us that primiparous cows with more consistent but longer milking intervals could potentially make less milk due to udder capacity among

other reasons. One of the limitations of this study is our lack of individual daily cow-fetching data, thus this hypothesis cannot be substantiated with our dataset.

Another reason may be primiparous cows, due to their lower social ranking, are more stressed approaching the robot and so they avoided it for consistently longer periods of time creating consistent milking intervals, but those animals who were less afraid to approach were able to maximize their production potential. This thought would be in agreement with previous AMS research which suggested primiparous cows experience more stress than multiparous cows, due to social pressures, in approaching AMS units (Persson Waller et al., 2003).

CONCLUSIONS

This novel research found several associations between MI and milk production per cow per day which can be used to further our knowledge and to create a foundation for future research. Results suggest a negative association between MI CV and milk production exists but is dependent upon failures, parity and week in milk and should be further examined to determine cause and effect.

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APPENDIX I – Tables

Table 1. Example of one farm’s feed table in an AMS

Days in Milk (DIM)¹	Milk Yield (kg)²	Concentrate amount offered (kg)³
0		16.5
7		23.2
8		20.9
	110.3	23.2
	176.4	23.2
	220.5	27.6
	264.6	30.9
	308.7	34.2
	396.9	40.8
75		13.2
	110.3	18.7
	176.4	23.2
	220.5	27.6
	264.6	30.9
	308.7	34.2
	396.9	40.8

¹ The days in milk in which changes are made to a cow’s concentrate allotment within the milking robot. For example: a cow from 0 to 6 days in milk is offered 16.5 kg of concentrate, with no other options. However, once that cow reaches 8 days in milk, the concentrate amount she is offered will depend on her milk yield, as seen in the second column.

² The amount of milk a cow produces per day. For example, a cow between days 8 and 75 in milk, producing 185 kg of milk will be in the “176.4” row and receive 23.2 kg of concentrate in the robot

³ The amount of concentrate offered in the robot unit, per cow, per visit.

Table 2. Association of feeding management factors with milk production

Per cow model¹	Change in milk production for every 1-unit increase (kg) ²	<i>P</i> value	2.5% Confidence Interval	97.5% Confidence Interval
DIM	-0.02	0.002	-0.03	-0.008
Cows per robot	-0.13	<.0001	-0.18	-0.09
PMR NDF content	-0.45	0.01	-0.76	-0.13
Low feeding management intensity	-4.62	0.004	-7.38	-1.86
Moderate feeding management intensity	-3.54	0.02	-6.61	-0.78
Use of a liquid feed in the box	-4.08	0.01	-6.89	-1.27
Number of milkings per day	2.98	<.0001	2.45	3.51
Concentrate intake	1.78	<.0001	1.41	2.17
% PMR remaining on middle screen of PSPS	0.1	0.07	0.0001	0.21
Per robot model¹				
DIM	-1.3	0.002	-2.09	-0.5
PMR NDF content	-24.77	0.03	-44.86	-4.68
Low feeding management intensity	-262.6	0.009	-438.4	-86.6
Moderate feeding management intensity	-228.9	0.02	-404.1	-53.5
Use of a liquid feed in the box	-201.7	0.04	-374.7	-28.6
Number of milkings per day	157.7	<.0001	125.9	189.5
Concentrate intake	110.7	<.0001	88.2	133.7
Cows per robot	29.18	<.0001	26.63	31.82

¹ Referencing the statistical model utilized for these findings

² For every one-unit increase in the feeding management factor in the first column, the milk production changed by the amount shown

Table 3. Associated milk production change by week compared to week one

Week in Milk	Milk production change (kg/cow per day)¹	<i>P</i> value	2.5% Confidence Interval	97.5% Confidence Interval
Week 2	11.21	<i>P</i> <.0001	10.50	11.92
Week 3	16.84	<i>P</i> <.0001	16.10	17.58
Week 4	19.01	<i>P</i> <.0001	18.26	19.76
Week 5	20.36	<i>P</i> <.0001	19.60	21.12
Week 6	20.72	<i>P</i> <.0001	19.96	21.47
Week 7	20.10	<i>P</i> <.0001	19.35	20.85
Week 8	20.15	<i>P</i> <.0001	19.40	20.89
Week 9	20.11	<i>P</i> <.0001	19.36	20.86
Week 10	19.41	<i>P</i> <.0001	18.67	20.15
Week 11	19.08	<i>P</i> <.0001	18.34	19.82
Week 12	18.09	<i>P</i> <.0001	17.34	18.84
Week 13	17.69	<i>P</i> <.0001	16.95	18.43

¹Milk production change as compared to week 1 in milk

Table 4. Association between the interaction of week and milking interval variability and milk production per cow per day

Week in Milk	Milk production change for every unit increase in MI CV (kg/cow per day)¹	<i>P</i> value	2.5% Confidence Interval	97.5% Confidence Interval
Week 2	-7.58	<i>P</i> <.0001	-9.96	-5.19
Week 3	-13.00	<i>P</i> <.0001	-15.65	-10.34
Week 4	-10.48	<i>P</i> <.0001	-13.26	-7.71
Week 5	-9.97	<i>P</i> <.0001	-12.81	-7.13
Week 6	-8.52	<i>P</i> <.0001	-11.39	-5.64
Week 7	-5.18	<i>P</i> = 0.0005	-8.08	-2.27
Week 8	-5.28	<i>P</i> = 0.0004	-8.19	-2.36
Week 9	-6.02	<i>P</i> <.0001	-8.95	-3.10
Week 10	-4.82	<i>P</i> = 0.001	-7.73	-1.91
Week 11	-5.45	<i>P</i> = 0.0003	-8.37	-2.52
Week 12	-2.53	<i>P</i> = 0.10 ²	-5.52	0.45
Week 13	-2.92	<i>P</i> = 0.05 ²	-5.81	-0.04

¹ Change in milk production per cow per day for each one-unit increase in milking interval coefficient of variation (MI CV), as compared to week one in milk

² Denotes trend with *P* value ≥ 0.05 and ≤ 0.10

APPENDIX II – Figures

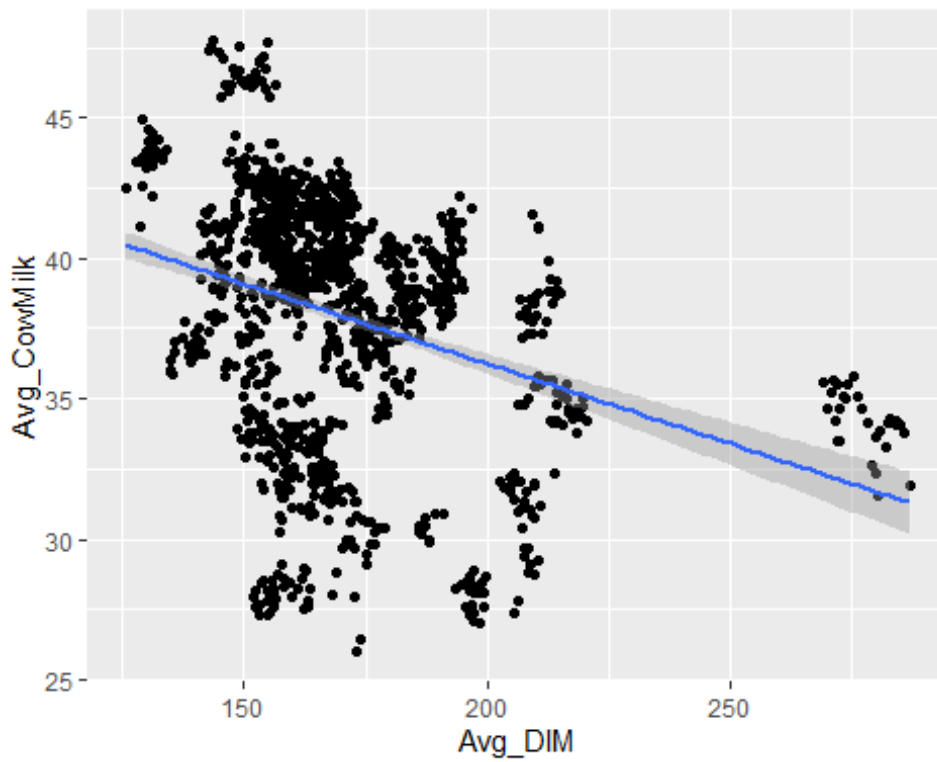


Figure 1. Average milk per cow per day (kg) by average days in milk (DIM). Average DIM was 170.7 ± 25.5 across all farms. Days in milk was negatively associated with milk production per cow. Every one day increase in DIM was associated with 0.02 kg/cow less milk per day (95% CI: -0.03, -0.008, $P = 0.002$).

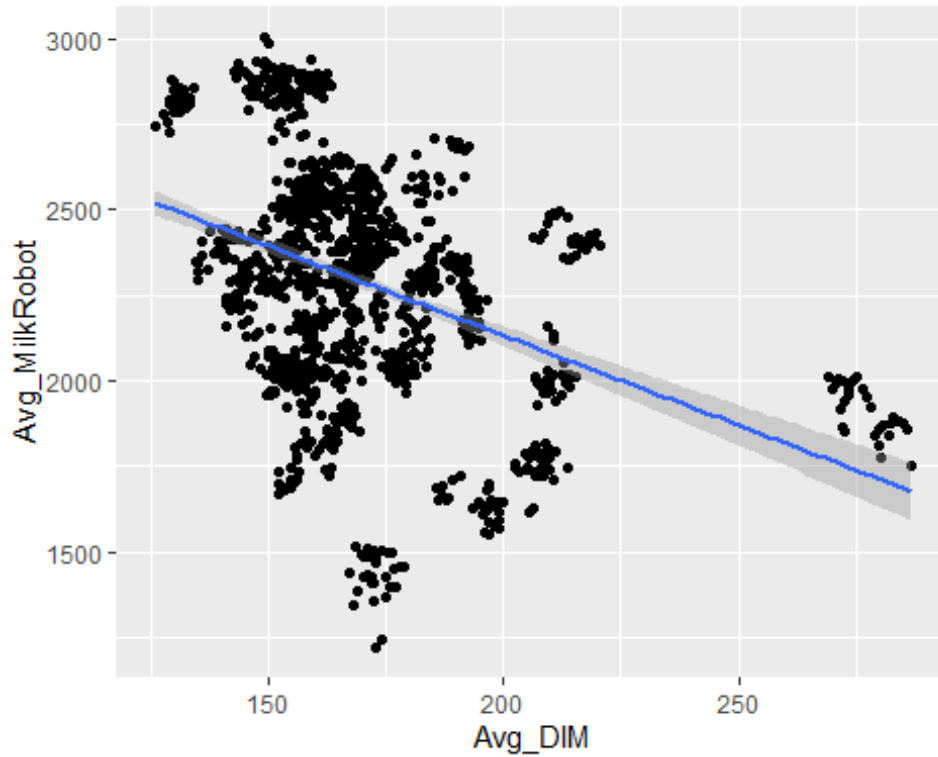


Figure 2. Average milk per robot per day (kg) by average days in milk. Average DIM was 170.7 ± 25.5 across all farms. Days in milk was negatively associated with milk production per robot. Every one day increase in DIM was associated with 1.3 kg/robot less milk per day (95% CI: $-2.09, -0.5$, $P = 0.002$).

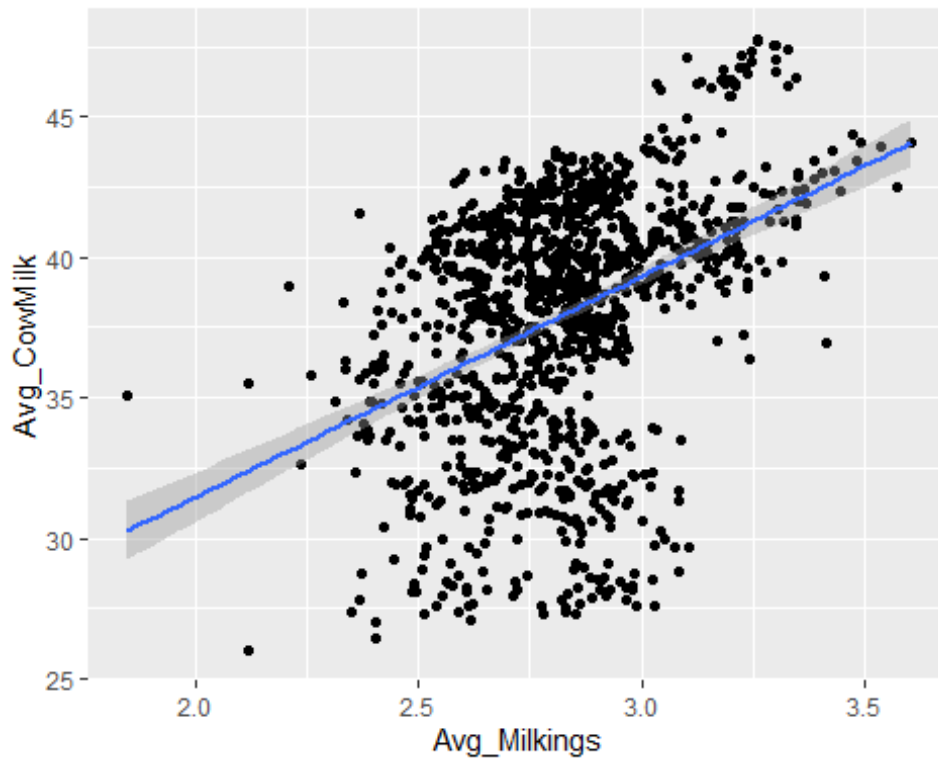


Figure 3. Average milk per cow per day (kg) by average number of milkings per day. The average number of milkings per day was 2.82 ± 0.22 . A positive association was found between number of average milkings per day and milk production per cow. For every additional milking per day, milk production per cow increased 2.98 kg/day (95% CI: 2.45, 3.51, $P < .0001$).

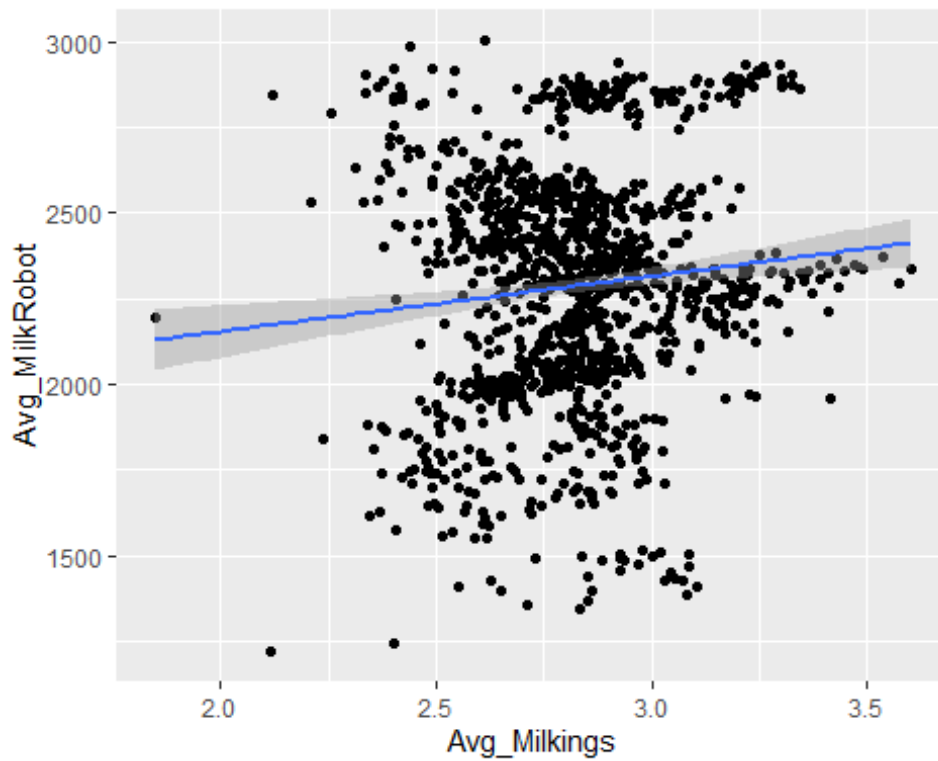


Figure 4. Average milk per robot per day (kg) by average number of milkings per day. The average number of milkings per day was 2.82 ± 0.22 . A positive association was found between number of average milkings per day and milk production per robot. For every additional milking per day, milk production per robot increased 157.7 kg/day (95% CI: 125.9, 189.5, $P < .0001$).

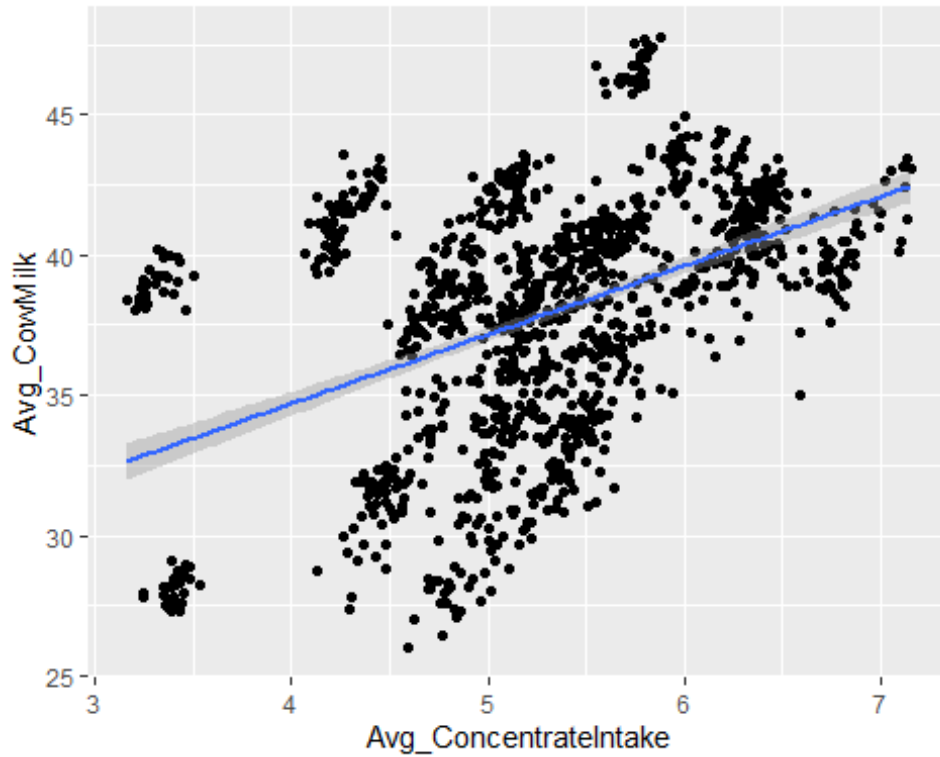


Figure 5. Average milk per cow per day (kg) by average concentrate intake (kg). Concentrate intake in the robot averaged 5.31 ± 0.79 kg/cow per day in our study. Average concentrate intake was positively associated with milk production per cow. For every additional kg of concentrate eaten, milk production per cow increased 1.78 kg/day (95% CI: 1.41, 2.17, $P < .0001$).

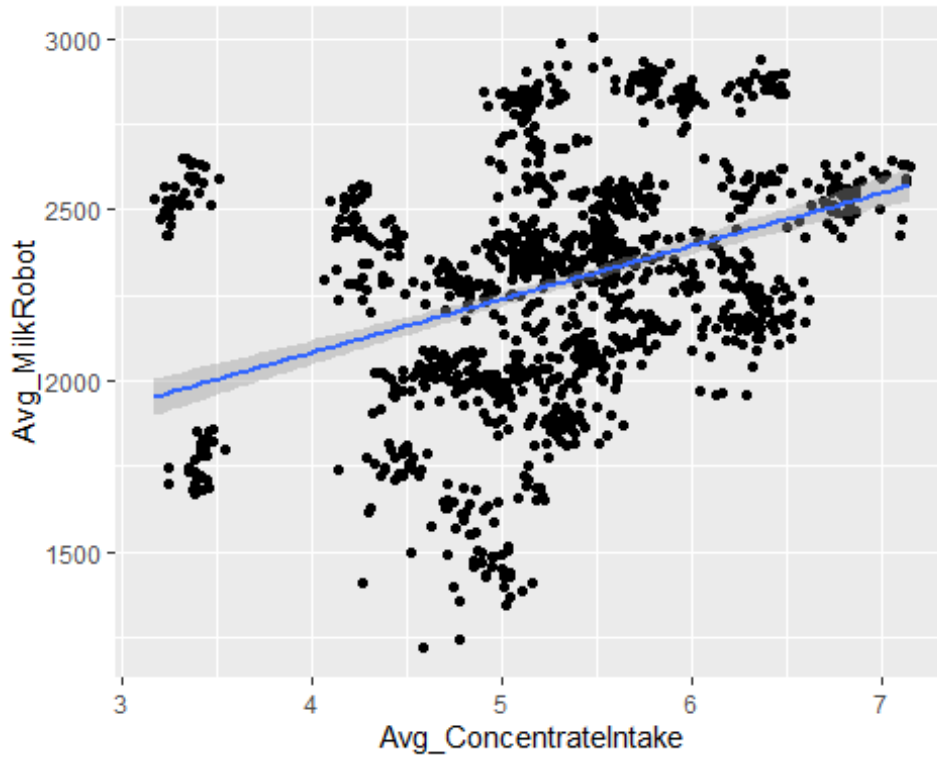


Figure 6. Average milk per robot per day (kg) by average concentrate intake (kg). Concentrate intake in the robot averaged 5.31 ± 0.79 kg/cow per day in our study. Average concentrate intake was positively associated with milk production per robot. For every additional kg of concentrate eaten, milk production per robot increased 110.7 kg/day (95% CI: 88.17, 133.65, $P < .0001$).

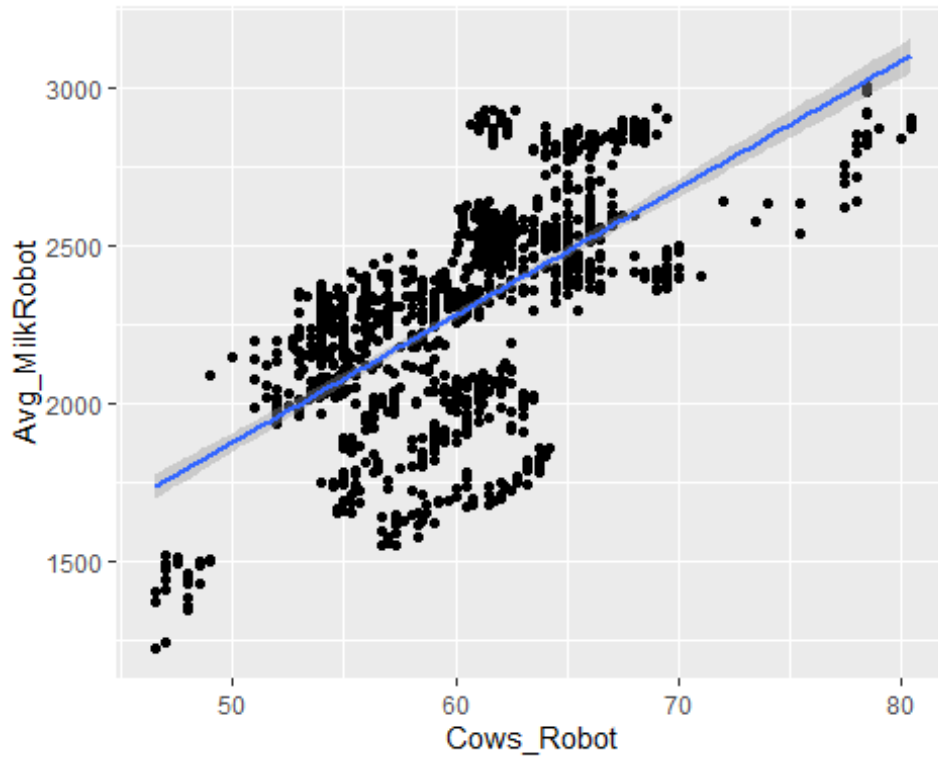


Figure 7. Average milk per robot per day (kg) by average number of cows per robot. Number of cows per robot averaged 60.2 ± 5.6 across all farms and was positively associated with milk per robot. For every additional cow per robot, milk per robot increased 29.2 kg/day (95% CI: 26.63, 31.82, $P < .0001$).

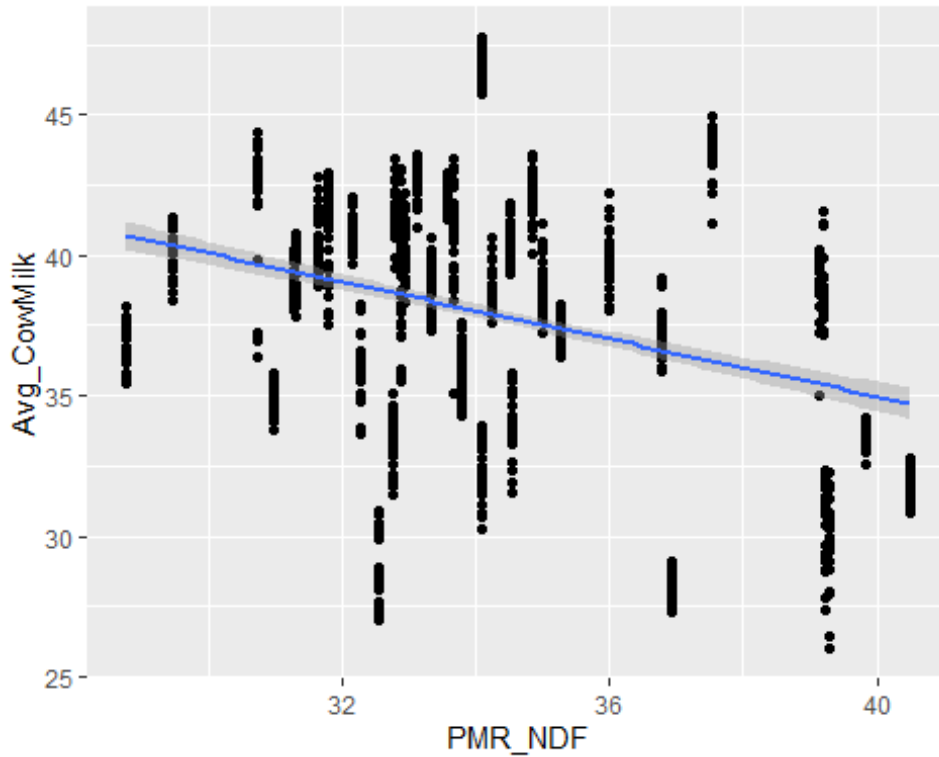


Figure 8. Average milk per cow per day (kg) by average neutral detergent fiber (NDF) content of the partial mixed ration (PMR, %). Our results showed a negative association of NDF on milk production per cow. For every additional percent of NDF within the PMR milk production per cow decreased 0.45 kg/day (95% CI: -0.76, -0.13, $P = 0.01$).

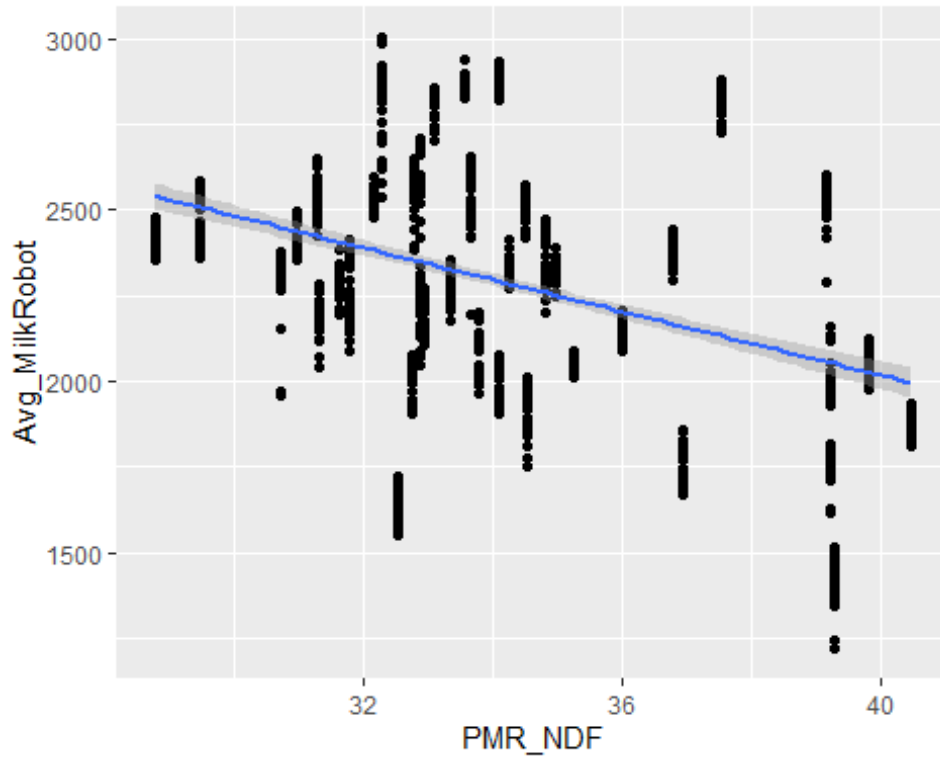


Figure 9. Average milk per robot per day (kg) by average neutral detergent fiber (NDF) content of the partial mixed ration (PMR, %). Our results showed a negative association of NDF on milk production per robot. For every additional percent of NDF within the PMR milk production per robot decreased 24.8 kg/day (95% CI: -44.9, -4.7, $P=0.03$).

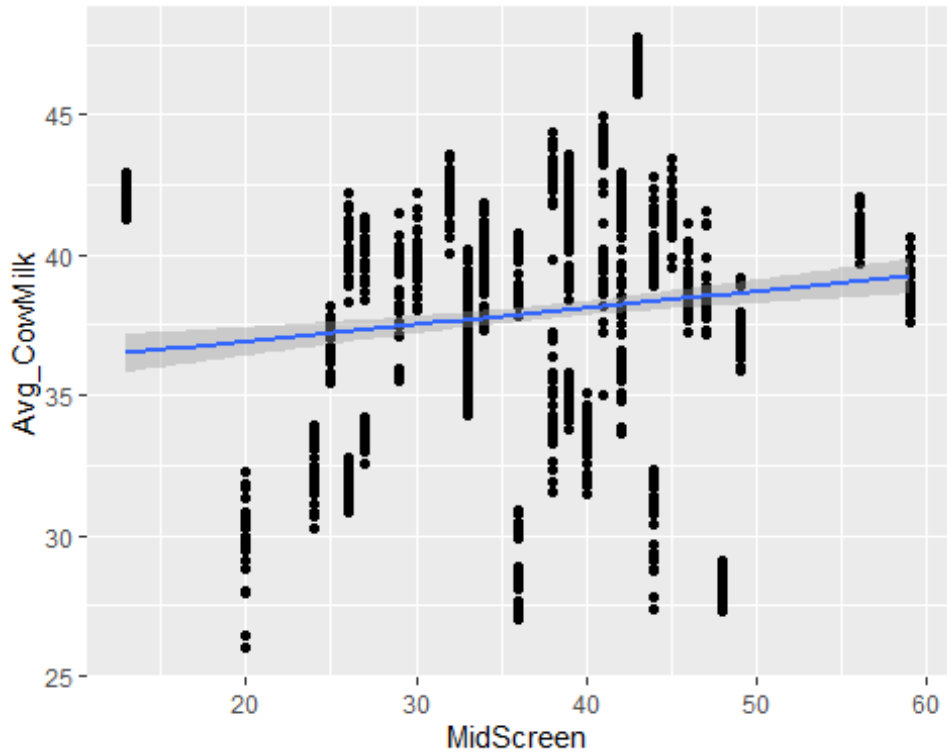


Figure 10. Average milk per cow per day (kg) by amount of feed material on the middle screen of the Penn State Particle Separator (PSPS, %). The amount of material on each screen of the 3-sieve PSPS had a positive associative trend only in our milk per cow model, and only on the middle screen as shown in this figure. For every additional percent of material on the middle screen, milk per cow increased by 0.10 kg/day (95% CI: 0.0001, 0.21, $P = 0.07$).

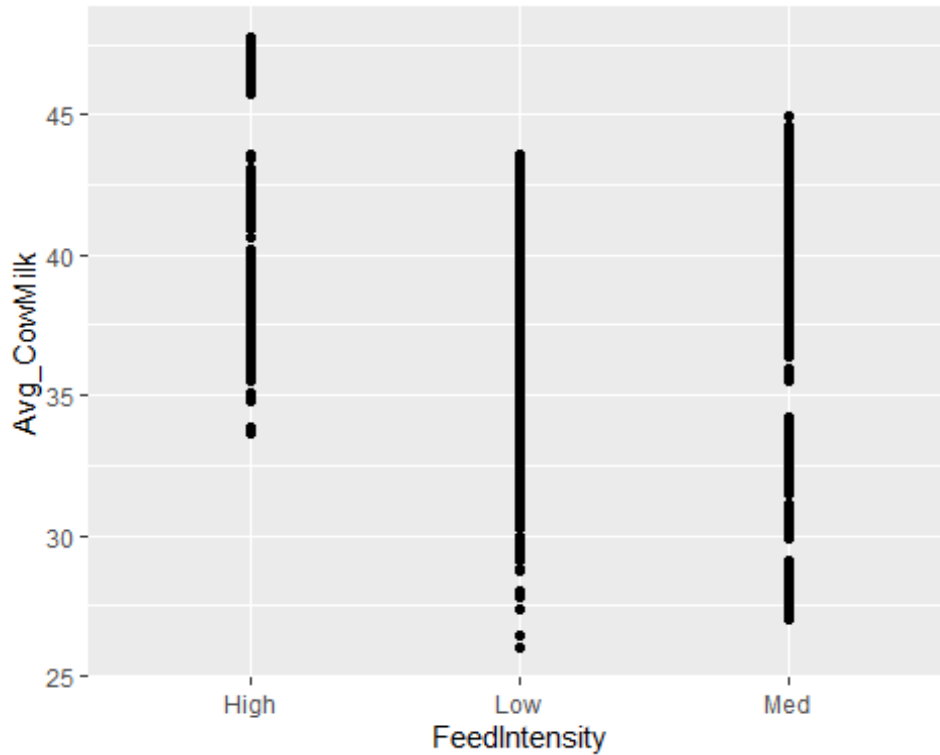


Figure 11. Average milk per cow per day (kg) by feed intensity management category. Low and moderate feeding intensity had negative associations with milk production per cow and per robot. Farms with low feeding management intensity had 4.62 kg/day less milk per cow (95% CI: -7.38, -1.86, $P = 0.004$) and 262.6 kg/day less milk per robot (95% CI: -438.4, -86.6, $P = 0.009$) than their high management counterparts. Meanwhile farms with moderate feeding management intensity had 3.54 kg/day less milk per cow (95% CI: -6.61, -0.78, $P = 0.02$) and 228.9 kg/day less milk per robot (95% CI: -404.1, -53.5, $P = 0.02$).

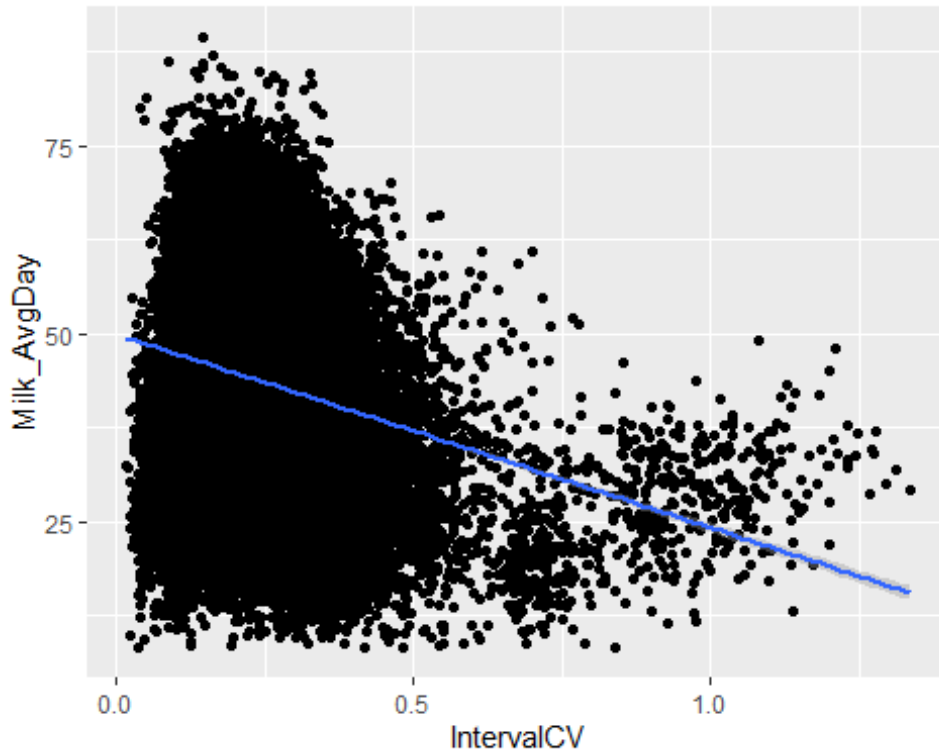


Figure 12. Average milk per cow per day (kg) by milking interval coefficient of variation (MI CV). For every unit increase in the MI CV, milk production decreased 4.51 kg/day (95% CI: -5.26, -3.76; $P < .0001$).

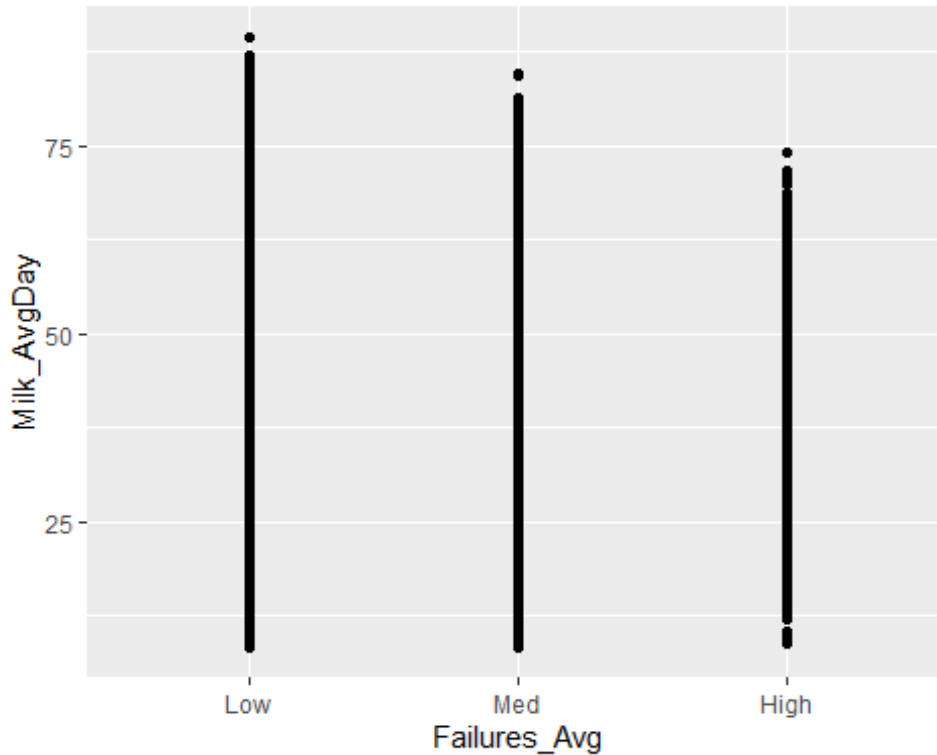


Figure 13. Average milk production per cow per day and average weekly milking failures. Comparing average milking failures of zero (LOW) to more than 1 milking failure (HIGH), a negative association with milk production per cow per day was found. Cows with more than 1 failure on average per week (HIGH) were associated with 2.98 kg/day less milk (95% CI: -3.97, -1.99; $P < .0001$). Cows averaging 1 failure (MED) compared to zero failures (LOW) were associated with a slight increase in milk per cow per day of 0.48 kg (95% CI: 0.13, 0.83; $P = 0.006$).

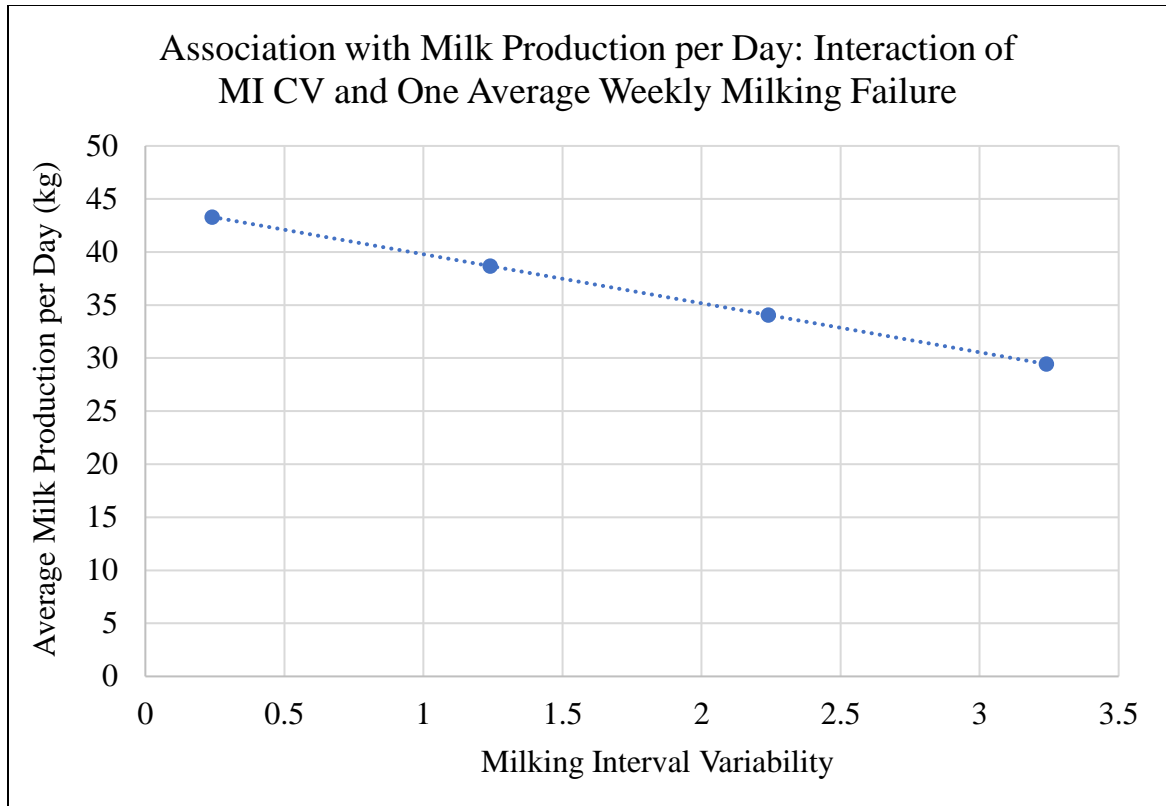


Figure 14. Interaction of milking interval coefficient of variation (MI CV) and average milking failures per week with associated milk production per cow per day (kg). When a cow experienced 1 failure per week, a negative interaction of MI CV and milk production per day was found. For every unit increase of MI CV for cows when they experienced an average of 1 milking failure per week, the associated milk production decrease was 4.62 kg/day (95% CI: -5.89, -3.36; $P < .0001$). The association of high milking failures (> 1 failure per week on average) and MI CV on milk production was not significant ($P = 0.18$).

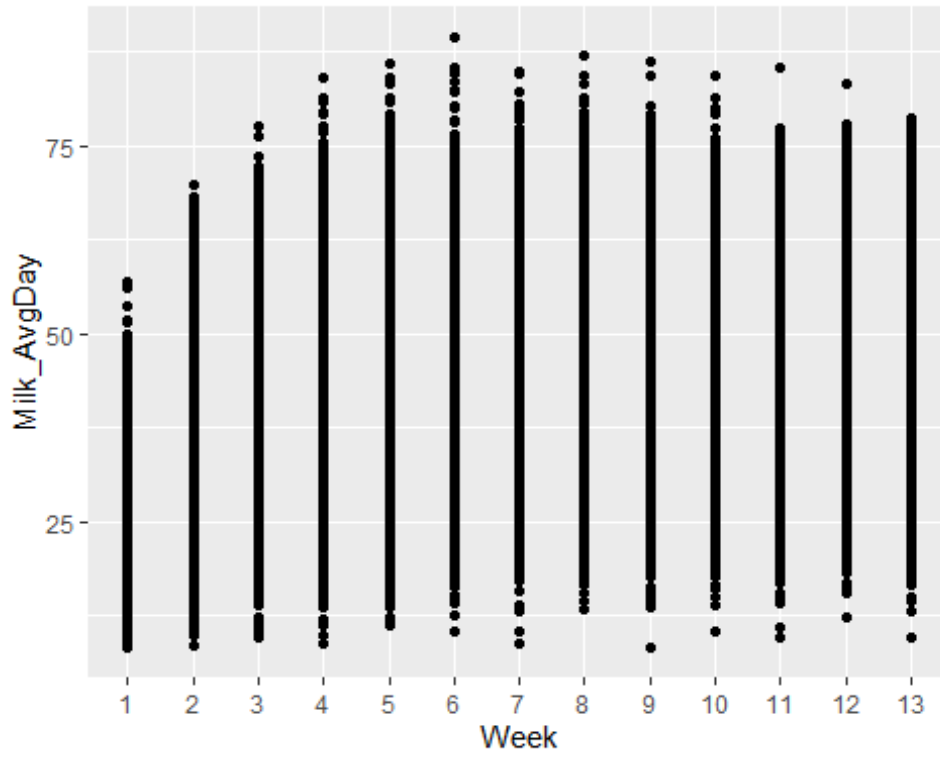


Figure 15. Average milk production per cow per day (kg) by week in milk.

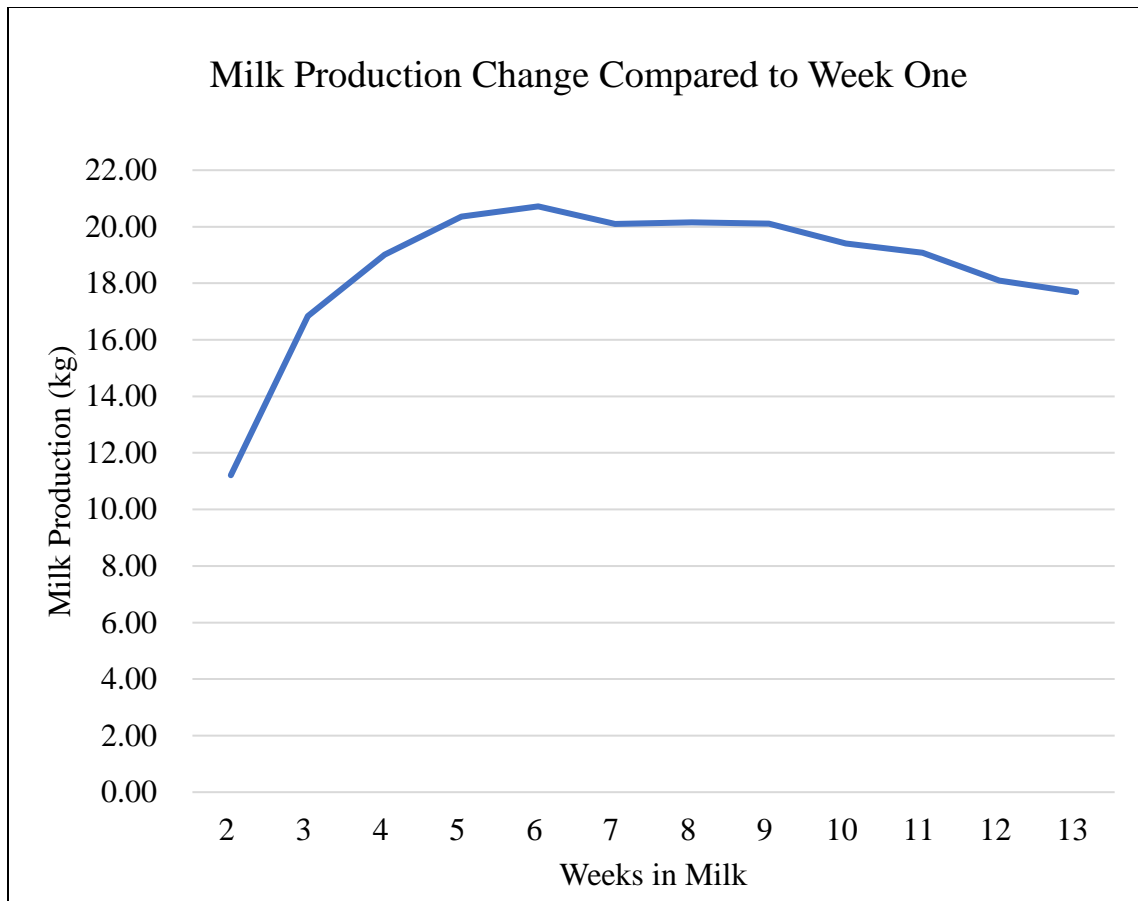


Figure 16. Milk production change per cow per day, by week, as compared to week 1. Comparing to week 1 in milk, which would coincide with days 1 through 7 post-calving, all weeks thereafter had a positive association with milk production per cow/d. Week 2 had the smallest positive association with milk production as would be expected, compared to week 1, with an increase of 11.21 kg/day (95% CI: 10.50, 11.92; $P < .0001$). The largest associated milk production increase was at week 6 compared to week 1, with an increase of 20.72 kg/cow per day (95% CI: 19.96, 21.47; $P < .0001$).

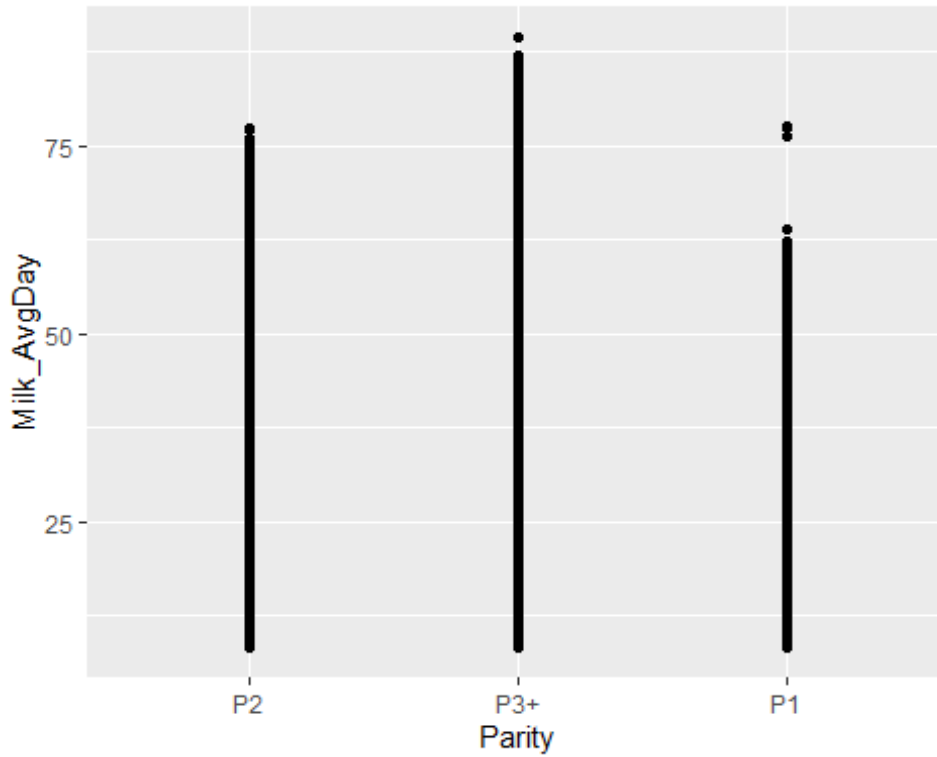


Figure 17. Average milk per cow per day (kg) by parity. Parity 3 and greater cows were associated with 4.78 kg/day more milk than their parity 2 counterparts (95% CI: 4.43, 5.14; $P < .0001$). Parity 1 cows were associated with a decrease in milk production of 12.53 kg/cow per day compared to parity 2 cows (95% CI: -12.89, -12.16; $P < .0001$).

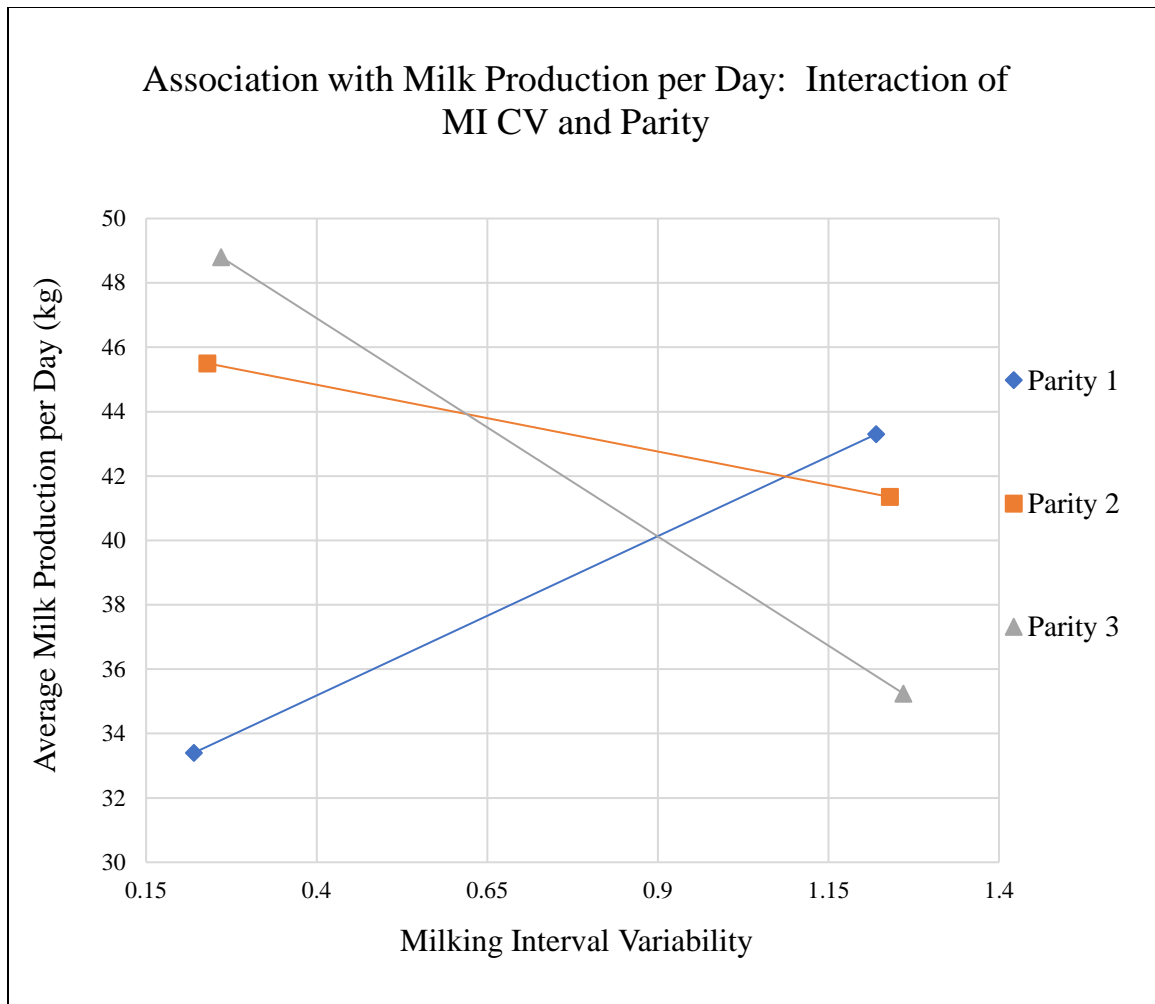


Figure 18. Interaction of milking interval coefficient of variation and parity with associated milk production per cow per day (kg). For parity 3 and greater cows compared to parity 2 cows, every unit increase in MI CV was associated with a decrease in milk production per cow of 4.62 kg/day (95% CI: -7.49, -4.73; $P < .0001$). For parity 1 cows compared to parity 2 cows with every unit increase in MI CV an associated milk production increase of 1.95 kg/day per cow was found (95% CI: 0.48, 3.42; $P = 0.009$).