EFFECTS OF THE AMOUNT OF CONCENTRATE OFFERED IN AN AUTOMATED MILKING SYSTEM ON DRY MATTER INTAKE, MILK YIELD, MILK COMPOSITION, RUMINAL FERMENTATION, AND BEHAVIOUR OF PRIMIPAROUS HOLSTEIN COWS FED ISO-CALORIC DIETS

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By

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ABSTRACT

The objective of this study was to determine if the quantity of concentrate provided in an automated milking system (AMS) affects dry matter intake (DMI), attendance to the AMS, milk and milk component yield, feeding behaviour, cow activity and ruminal fermentation of lactating dairy cows fed iso-caloric diets. Eight ruminally-cannulated primiparous Holstein cows were used in a replicated 4 × 4 Latin square design with 28-d periods. Cows were housed in a free-stall facility with a guided-traffic (feed-first) flow barn-design. Cows were offered 0.5, 2.0, 3.5, or 5.0 kg/d DM of pellet in the AMS, with an equivalent reduction of the same pellet in the partial mixed ration (PMR). Day 21 to 24 of each treatment period were used for DMI, milking performance, behaviour, and ruminal pH determination, while d 25 to 28 were used for ruminal short-chain fatty acid (SCFA) and ammonia concentrations, as well as total tract digestibility. As imposed, consumption of AMS pellet linearly increased ($P < 0.01$), equating to 0.50, 2.00, 3.49, and 4.93 kg/d. Correspondingly, the standard deviation in AMS pellet intake among days linearly increased from 0.06 to 0.85 kg/d as the quantity of concentrate in the AMS increased from 0.5 to 5.0 kg ($P < 0.01$). The PMR DMI decreased linearly with increasing AMS concentrate allocation ($P < 0.01$), but total DMI (PMR + AMS concentrate) was not affected (25.3 kg/d, $P = 0.40$). As AMS concentrate allocation increased, the selection against particles retained on an 18-mm sieve linearly increased ($P = 0.02$) and selection against particles retained on the bottom pan decreased ($P < 0.01$). Milking frequency (3.22 milkings/d, $P = 0.82$), milk yield (37.5 kg/d, $P = 0.59$), milk fat yield (1.43 kg/d, $P = 0.46$), and milk protein yield (1.22 kg/d, $P = 0.42$) were not affected; however, milk urea nitrogen concentration decreased linearly with increasing AMS concentrate ($P = 0.02$). Ruminal pH averaged 6.18 and was not affected by AMS concentrate ($P = 0.62$). Total ruminal SCFA concentration was greatest when 3.5 kg of concentrate was allocated in the AMS and ruminal ammonia decreased linearly with increasing AMS concentrate ($P = 0.01$). Time spent lying, the number of lying bouts, and average bout duration were not affected by treatment ($P \geq 0.11$). These data indicate that increasing the quantity of concentrate in the AMS increases daily variability in AMS concentrate intake while decreasing PMR intake, and increasing AMS pellet provision, under isocaloric dietary settings, is not likely to affect voluntary visits to the AMS, milk and milk component yield, or ruminal fermentation.

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LIST OF ABREVIATIONS

ADF	Acid detergent fibre
AMS	Automatic milking system
$aNDF_{OM}$	Neutral detergent fibre measured using α-amylase and sodium sulfite and subsequently corrected for organic matter content
BCS	Body condition score
BW	Body weight
Ca	Calcium
CNCPS	Cornell Net Carbohydrate and Protein System
CP	Crude protein
DIM	Days in milk
DM	Dry matter
DMI	Dry matter intake
EE	Ether extract
ECM	Energy corrected milk
F:C	Forage-to-concentrate ratio
$iNDF_{OM}$	Indigestible neutral detergent fibre measured using α-amylase and sodium sulfite and subsequently corrected for organic matter content
HOT	Hepatic oxidation theory
LRCpH	Lethbridge Research Centre pH logger
MUN	Milk urea nitrogen
N	Nitrogen
NDF	Neutral detergent fibre
NDS	Nutritional Dynamic System
NEFA	Non-esterified fatty acid
OM	Organic matter
P	Phosphorus
PMR	Partial mixed ration
PSPS	Pennsylvania State Particle Size Separator
SCC	Somatic cell count
SCFA	Short-chain fatty acid

SD	Standard deviation
SEM	Standard error of the mean
TCA cycle	Tricarboxylic acid cycle
TMR	Total mixed ration
VMS	Voluntary milking system

1.0 GENERAL INTRODUCTION

Since the first installation of the automatic milking system (**AMS**) 26 years ago, the global adoption rate has increased dramatically (Jacobs and Seigford, 2012). It is believed that AMS have such a quick adoption due to the efficiencies and flexibility in labour for producers when compared to conventional milking systems (Rotz et al., 2003). There are several cow traffic designs that can be integrated with the AMS, including free-flow, guided-flow and forced-flow (Jacobs and Seigford, 2012). Independent of cow traffic design, all facilities equipped with AMS require cows to voluntarily enter the individual milking stall. The motivation for cows to be milked with AMS corresponds to the food reward located inside the AMS (Prescott et al., 1998). Thus, nutritional strategies with the AMS require feeding both a concentrate in the AMS and a partial mixed ration (PMR) at the feed bunk to meet nutrient requirements (Bach and Cabrera, 2017).

The optimal feeding strategy for cows being milked with AMS is still unclear. Industry recommendations suggest supplying upwards of 8.0 kg/d of concentrate in the AMS (DeLaval Inc., 2018; Lely, n.d.; Brouk, 2017; Rodenburg, 2011). Evidently, producers are following these recommendations. In survey-based studies, it was determined that producers were feeding 6.0 kg of concentrate for cows producing 38.0 kg of milk (primarily free-flow barns; Tremblay et al., 2016) and 0.9 to 11.4 kg in free-flow traffic designs or 0.9 to 5.5 kg in guided-traffic designs (Salfer and Endres, 2014). Many recommendations suggest a precision feeding approach, often with the use of feed tables. This approach attempts to provide quantities of concentrate or different concentrates in the AMS that correspond to an individual cow's milk production (or perhaps DIM), attempting to meet nutrient requirements on an individual basis (Bach and Cabrera, 2017). The concept of feeding large quantities of concentrates in the AMS is thought to encourage a greater motivation for cows to enter the milking stall, resulting in an increased number of visits per day and daily milk yield. That said, previous research has demonstrated negative implications when feeding large quantities (> 5.0 kg) of concentrate in the AMS (Bach et al., 2007; Halachmi et al., 2005).

Few studies have investigated the impact of AMS feeding strategies on both the AMS concentrate and the partial mixed ration (**PMR**) intake; however, those that have, demonstrated that when cows are offered greater quantities of concentrate in the AMS, a reduction in PMR intake is observed (Bach et al., 2007; Hare et al., 2018; Menajovsky et al., 2018). Thus, cows are not necessarily consuming a greater total DMI (AMS concentrate + PMR). Further, it is evident in previous studies that when a greater day-to-day quantity of concentrate is provided in the AMS there is greater variability in AMS concentrate intake (Hare et al., personal communication; Menajovsky et al., 2018). Another challenge when feeding large quantities of concentrate in the AMS is the difficulty with meeting target consumption amounts. Bach et al. (2007) targeted feeding 3.0 and 8.0 kg of concentrate in the AMS; however, only achieved 2.6 and 6.8 kg of concentrate consumed in the AMS, while Halachmi et al. (2005) provided either 1.2 kg/milking or 7.0 kg/d of concentrate in the AMS and achieved 5.2 kg/d in the latter treatment. Hare et al. (2018) and Menajovsky et al. (2018) have demonstrated that achieving target concentrate intakes in the AMS requires greater quantities to be programmed into the AMS computer software. Last, when greater quantities of concentrate are supplied in the AMS, milk and milk component yields and visits to the AMS have not demonstrated a positive response (Bach et al., 2007; Hare et al., 2018). Thus, previous research findings do not support the current AMS feeding recommendations.

There are some notable limitations with previous AMS studies attempting to determine accurate concentrate recommendations with the AMS. All previous studies have only analyzed 2 quantities of concentrate in the AMS (Bach et al., 2007; Halachmi et al., 2005; Hare et al., 2018; Menajovsky et al., 2018) and thus the direction and severity of response variables have not yet been thoroughly investigated. In addition, only one previous study has examined ruminal fermentation and total tract digestibility parameters (Menajovsky et al., 2018). The current study attempted to provide an insight to improving AMS feeding management strategies. Specifically, the intention was to determine the effects of feeding varying quantities of concentrate in the AMS under iso-caloric conditions on attendance to the AMS, milk and milk component yield, DMI of the PMR, ruminal fermentation and total tract digestibility.

2.0 LITERATURE REVIEW

2.1 General Principles of Automated Milking Systems (AMS)

Since the start of the twentieth century, several advancements in the dairy production industry have occurred including the invention of automated milking machines that have accommodated the growing efficiency of producing larger milk volumes with fewer cows (Jacobs and Siegford, 2012). The concept of automatic milking systems (**AMS**), also called voluntary milking systems (**VMS**), or robotic milking systems, originated in the Netherlands in the early 1980's (Hyde and Engel, 2002); however, the first installation occurred in 1992 (Jacobs, 2011). Since this first installation, AMS have become increasingly popular among dairy farms around the world. By 2011, it was estimated that there were more than 10,000 farms worldwide using AMS (de Koning, 2011). Of these AMS farms, approximately 90% were located in Europe, while an estimated 9% were located in Canada (de Koning, 2010), primarily in Ontario and Quebec (Canadian Dairy Information Center, 2018). Currently, it is estimated that 6.6 % of all dairy herds in Canada are using AMS, with 0.11 % of those being located in Saskatchewan (Canadian Dairy Information Center, 2018). This translates into 7.5 % of Saskatchewan dairy farms using AMS (Canadian Dairy Information Center, 2018).

There may be benefits of adopting AMS far beyond those of conventional milking procedures (Rotz et al., 2003). Some of these advantages may include increasing production through increasing milking frequency and promoting a more flexible lifestyle for dairy producers (de Koning, 2010). It has also been suggested that AMS may improve cow health, reduce labour, and the routine activities that conventional producers are faced with (Jacobs and Seigford, 2012; Woodford et al., 2015). As AMS continue to advance, it is predicted that the use and efficiencies will continue to grow; triggering the transition from conventional milking systems to become increasingly more common in Canada.

Dairy facilities with AMS require a different type of management strategy than that of conventional parlour or tie-stall milked herds. Facilities with an AMS require cows to voluntarily enter the AMS to encourage frequent milkings and minimize labour associated with handling cows. It should be noted that cows milked with an AMS, do not visit the AMS the same number of times or at the same time of day on a regular basis (Bach et al., 2007). This results in an inconsistent number of daily milking events and milk production could be expected to vary more among days than in conventional milking systems. Encouraging voluntary visits to the AMS is partially accomplished through feeding management (Bach et al., 2007).

Feeding programs with AMS are very different than that of conventional milking systems. Conventional systems in North America usually involve a total mixed ration (**TMR**) provided in feed bunks where the cows are supplied a feed that is well mixed and balanced to meet all nutrient requirements at a specified daily DMI. In contrast, AMS feeding programs involve the use of a partial mixed ration (**PMR**) delivered to the feed bunk and a concentrate that is delivered in the AMS. The PMR is designed to supply a portion of the nutritional requirements with the AMS concentrate completing the provision of dietary nutrients to meet requirements. Thus, the ability to meet nutrient requirements depends on accurate predictions of DMI for both the concentrate provided in the AMS and the PMR.

The primary motivating factor encouraging voluntary attendance at the AMS has been determined to be through the provision of concentrate in the AMS and less towards the process of relieving the udder pressure (Prescott et al., 1998). The concentrates provided in the AMS must be highly palatable, thereby enhancing voluntary attendance by creating a rewarding experience for the cows (Jacobs, 2011). Therefore, the more superior the motivation for cows to enter the AMS, the greater probability of AMS attendance (Wagner-Storch and Palmer, 2003; de Jong et al., 2003), resulting in a greater milk yield simply due to a greater number of visits (Prescott et al., 1998; Rodenburg, 2011).

Various feeding programs have been implemented within AMS; however, a consensus over optimal strategies has yet to be achieved. One dominating concept is precision feeding. Precision feeding management strategies attempt to improve productivity and efficiency by adapting nutritional programs to meet individual cow requirements (Cerosaletti et al., 2004; Gehman, 2011), rather than formulating and delivering diets based on herd averages. Formulating diets based on herd averages generally overestimates nutrient requirements and supply in lower producing cows, while underestimating nutrient requirements and supply for high producing cows. The over and under supply of nutrients leads to inefficiencies with single TMR-based feeding programs (Bach and Cabera, 2017). Precision feeding management strategies attempt to offer individual cows different quantities or types of concentrate in the AMS based on production, stage of lactation, or parity (Bach and Cabera, 2017). This idea reintroduces the concept of component feeding, where a portion of the nutrients are supplied in separate locations for free-stall herds or separate times within the facility for tie-stall dairy herds.

Originally, component feeding was designed to offer concentrates within the parlour to allow for easier handling of cows at the time of milking, but concentrates were later removed from the parlour because it was recognized that concentrates were not necessary to improve movement of cows toward the parlour (Schingoethe, 2017). Removal of concentrates in the parlour may have initiated the provision of concentrate in automatic feeders within a free-stall pen. Similar to AMS, the concentrate feeders allowed producers to provide quantities of concentrate to individual cows based on milk production. It has been reported that some producers supplied 1 kg of concentrate in an automatic feeder for every 3 kg of milk produced (Schingoethe, 2017). In addition, component feeding also offered mineral supplement and forage components separately.

Component feeding was a historic way of feeding in parlour-based and tie-stall dairies as the TMR was not designed until 1952 (Harshburger, 1952); however, TMR were not officially introduced in scientific literature until 1966 (McCoy et al., 1966). Since then, TMR-feeding has dominated feeding programs in North America and parts of Europe. Simultaneous with TMR adoption, were observations that component feeding strategies may be disadvantageous relative to TMR including increased risk for insufficient fibre intake, unpredictable changes in the dietary forage to concentrate ratio (**F:C**), induction of milk fat depression (Coppock, 1977), increased risk for digestive upset particularly during the transition period, and more bouts where cows were off feed (Hernandez-Urdandeta et al., 1976). In contrast, TMR nutritional programs provide cows with fewer opportunities to select for individual components within the diet and theoretically provide a nutritionally complete and uniform feed (Coppock, 1977). Total mixed ration feeding strategies strive to maintain a constant nutrient composition to encourage milk production and a more accurate understanding of nutrient consumption (Coppock, 1977). Further, the more stable F:C consumed with a TMR allows for greater stability and efficiency of rumen microbial populations (DeVries et al., 2007). The TMR also allowed for incorporation of the vitamin and mineral component of the diet allowing producers to overcome challenges with vitamin and mineral intake due to unpalatability and poor acceptance of these supplements by cows (Bach and Cabrera, 2017). That said, proper TMR mixing and order of feed ingredient inclusion into the mixer is important for an even distribution of nutritional components (Schingoethe, 2017).

While AMS are relatively new, as stated above, AMS feeding programs can be compared to the older component feeding programs because concentrate is provided in the AMS and PMR is offered at the feed bunk. This concept reintroduces issues associated with component feeding and provides challenges with creating the optimal feeding programs due to the variability of intake of AMS concentrate and substitution of PMR with the AMS concentrate. Controversially, one scientist suggests that as research continues to grow regarding feeding programs with AMS, it may be possible to avoid feeding concentrates in the AMS entirely in the future, allowing for better nutritional management, mimicking what occurred with concentrates in the parlour (Schingoethe, 2017).

2.2 Motivating Factors Encouraging Cows to Enter the AMS

Voluntary attendance is crucial to optimize the labour requirements, milk production, and corresponding economic efficiencies associated with AMS (Bach and Cabrera, 2017). Attendance to the AMS for individual cows equating to less than 2 visits per day could result in a reduced milk yield due to autocrine feedback inhibition of alveolar milk secretion (Wilde and Peaker, 1990) and potentially an increase in risk for mastitis (Hillerton and Winter, 1992). Thus, maintaining voluntary visits to the AMS is essential.

Cows strongly motivated to voluntarily enter the AMS require fewer alternative rewards such as feed in the AMS (Prescott et al., 1998). There are many hypotheses to suggest that cows receive rewards through the milking procedure. First, milking is believed to relieve the discomfort that the cow experiences from having a large distended udder (Rathore, 1982). This discomfort and motivation to be milked may be emphasized during early lactation until peak lactation due to the hyperplasia and hypertrophy of mammary cells (Knight and Wilde, 1993) and related high milk yield at this time. Second, cows may be motivated to be milked for psychological reasons (Prescott et al., 1998). The process of being milked may, in some way, satisfy a cow’s natural behaviour to nurse her calf; however, this motivation may decline as lactation progresses simulating the natural weaning process (Phillip, 1993). Last, cows may be motivated to be milked because of the stimulation of oxytocin released during milk let down.

Physical stimulation of teats may also cause arousal in some animals (Andersonhunt and Dennerstein, 1995).

Despite these theories suggesting reasons why cows may be stimulated to milk, feed rewards are generally used to promote voluntary attendance to the AMS. Prescott et al. (1998) set out to determine the effects of motivation based on stage of lactation, udder fill, and feed as a reward and how they affect voluntary attendance to an AMS. Results of that study demonstrated that the motivation for cows to be milked without a feed reward was relatively weak, highly variable, and weakly dependant on stage of lactation. High producing cows (early lactation cows) were more motivated to be milked than lower producing cows (late lactation cows) suggesting that motivations to be milked may be stronger and more rewarding earlier in lactation. In addition, when cows were trailed through a Y-maze every 3.5 h, but not supplied with a feed reward, most high yielding cows and some low yielding cows chose to be milked at this interval, even though only small quantities of milk would likely be stored in the udder at this frequent of interval. The authors hypothesized this response was due to the milking process providing some sort of reward for the cows (tactile teat stimulation, arousal from oxytocin release, or a psychological satisfaction); however, they noted high motivation variability, suggesting that some cows may find being milked a negative experience. When cows were given the option to receive a feed reward or to be milked, cows chose to eat a concentrate supplied in a feeder at the end of the Y-maze. When high producing cows were supplied with 4 kg of concentrate in an AMS, they attended the AMS more frequently than low producing cows fed 2 kg of concentrate. It was therefore suggested that the level of food reward may influence attendance to the AMS. Though this study only utilized 12 cows, conclusions clearly demonstrated that feed rewards provided in the AMS significantly improved motivation and voluntary attendance.

Providing a concentrate in the AMS increases motivation to visit the AMS; however, determining the amount and type of concentrate to provide within the AMS is more complex. Theoretically, increasing visits to the AMS will allow for a greater consumption of concentrate (Halachmi et al., 2005). A handful of studies comparing different quantities of concentrate in the AMS to determine the effect on AMS visits, milk and milk component yields have been conducted. For example, Halachmi et al. (2005) offered 1.2 kg of concentrate per milking or 7.0 kg/d of concentrate in the AMS within a guided-traffic barn design. The formulation of PMR in both treatments were identical; therefore, the only change in each treatment was the quantity of concentrate provided in the AMS causing the F:C of the total diet (AMS + PMR) of each treatment to be different. Though significance levels were not outlined in this study, results demonstrated that when greater energy dense diets (PMR + AMS) were supplied, milk production increased. Despite this, no differences in the number of AMS visits were observed among treatments, suggesting that 1.2 kg/milking of concentrate was sufficient to encourage motivation to enter the AMS. Bach et al. (2007) offered 3.0 or 8.0 kg of concentrate in an AMS under iso-caloric dietary (PMR + AMS) conditions in a free-flow barn design. Authors observed no differences in voluntary attendance to the AMS, milk yield, milk component yield, and fetching concluding that there was no benefit of providing large quantities of concentrate in the AMS. Similarly, Hare et al. (2018) offered 0.5 or 5.0 kg of concentrate in the AMS under iso-energetic dietary conditions in a guided-traffic barn design and reported no differences in visits to the AMS, milk yield, or milk component yield. Most recently, Menajovsky et al. (2018) tested 4 different dietary treatments in a guided-flow traffic design: 1) a low F:C PMR with 6.0 kg of concentrate in the AMS; 2) a low F:C PMR with 2.0 kg of concentrate in the AMS; 3) a high F:C PMR with 6.0 kg of concentrate in the AMS; and 4) a high F:C PMR with 2.0 kg of concentrate in the AMS. In this study, the authors observed no differences among treatments for frequency of milking events; however, with increasing energy density in the total diet (high AMS concentrate or the PMR with a low F:C) a tendency for an increased milk yield was observed. Last, they observed greatest milk protein concentration and a tendency for milk fat concentration to be greatest when 2.0 kg of concentrate was provided in the AMS. Together, these studies suggest that increasing the quantity of concentrate with provided in the AMS will likely not positively influence voluntary attendance to the AMS, milk yield, or milk composition with iso-energetic diets; however, milk yield and milk protein concentration may be increased when using the AMS concentrate to increase the energy density of the diets (AMS + PMR). That said, Menajovsky et al. (2018), demonstrated that these slight increases in milk yield and milk protein concentration may also occur through increasing the energy density of the diet through the PMR. Although the exact quantity of concentrate necessary to maintain AMS attendance is uncertain, it is evident that not all concentrate provided in the AMS is consumed (Bach and Cabrera, 2017). As the quantity of concentrate allowance in the AMS increases, the quantity of concentrate that is refused or not offered in the AMS also increases (Figure 2.1; Bach and Cabrera, 2017).

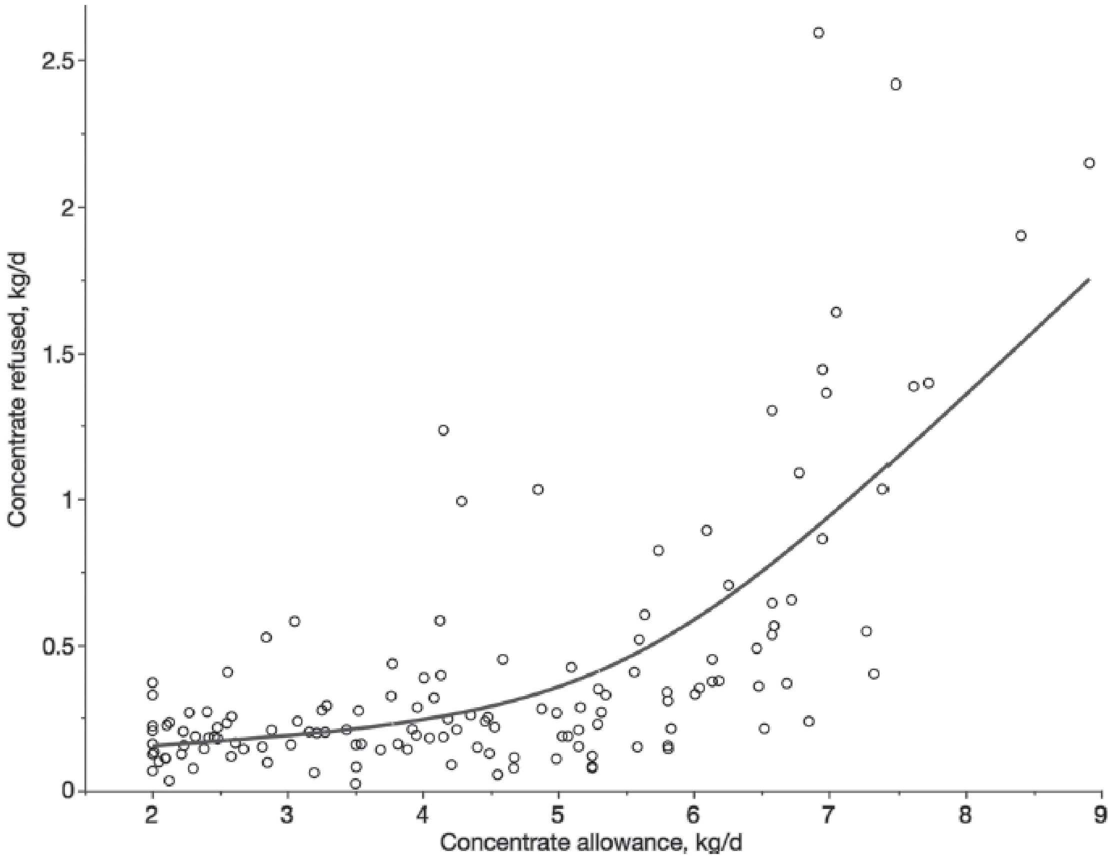

Figure 2.1. Evolution of amount of concentrate consumed (average over an 8-d period) relative to the amount of concentrate offered (averaged over a 7-d period) in an automatic milking system. Figure sourced from Bach and Cabrera (2017), with permission from the publisher.

This creates further challenges with optimal feeding strategies associated with the AMS and will be discussed below (2.3.2 Implications of Precision Feeding).

The physical and chemical characteristics of the concentrate provided in the AMS may influence milk production and voluntary attendance to the AMS. The recommended physical form of concentrate is a pellet because it is preferred by cows over mash in the AMS (Spörndly and Ashberg, 2006). A pellet with minimal fines and extreme hardness is optimal because fewer negative effects on intake have been reported with hard pellets (Rodenburg et al., 2004). However, pellets formulated to be high in starch may influence appetite, feeding related behaviours, ruminal pH, and NDF digestibility in a manner that may negatively influence milk composition, production, and increase the risk of lameness (Oba and Wertz-Lutz, 2011). In AMS herds, minimizing lameness is critical because lame cows visit the AMS less frequently (Bach et al., 2006; Borderas et al., 2008) and increase labour associated with fetching cows (Bach et al., 2007). Miron et al. (2004) observed a tendency for an increased milk protein concentration when cows were offered a high starch concentrate in the AMS, while a concentrate high in digestible fibre increased milk fat yield when provided 8.0 kg of concentrate/d. Alternately, Halachmi et al. (2006) offered 3.0 kg of concentrate in the AMS with pellets that contained 25% starch or 49% starch. Halachmi et al. (2006) observed no differences in voluntary attendance to the AMS, or milk or milk component yields among treatments. These data suggest that when only small concentrate allowances (< 3.0 kg/d) are provided in the AMS, composition has less of an impact on voluntary attendance and milk and milk component yield; however, at higher concentrate allocations, production responses may be influenced. Masden et al. (2010) determined that cows prefer pellets based on a barley-oat mix or a wheat-based pellet over either a corn or barley-based pellet. Flavours may also be used in the pellet composition, but their effectiveness has not been consistent across studies. Migliorati et al. (2010) found that when various flavours were added to pellets, AMS attendance increased, even when low quantities of pellet were offered in the AMS (1.5 to 3.5 kg/d). In contrast, when vanilla or fenugreek were added to AMS pellets, there were no differences in intake, visits to the AMS, or in visit patterns among treatments (Harper et al., 2016). Physical and chemical characteristics of AMS concentrate may demonstrate direct effects on milking frequency and milk and milk component yield; however, further investigation regarding concentrate form and composition are necessary to fully understand precision management strategies and its relationship with the PMR formulation and cow feeding behaviours.

There is substantial controversy for whether minerals and vitamins should be added to the AMS pellet. Generally, vitamins and mineral supplements are poorly accepted by cows due to low palatability and thus are primarily offered through the PMR. One concern with supplying mineral supplements in the PMR with AMS systems is that as cows increase in milk yield, the concentrate allowance in the AMS also increases; however, mineral intake might not meet requirements because the increase in pellet intake may drive an increase in DMI or that increasing pellet intake in the AMS decreases PMR intake (Bach and Cabera, 2017). For example, as the amount of concentrate provided in the AMS increases, there is a substitution effect decreasing intake of PMR (Bach et al., 2007; Hare et al., 2018; Menajovsky et al., 2018). Therefore, exact concentrate composition still requires further investigation to optimize nutrient delivery, voluntary attendance to the AMS, and milk and milk component yield.

2.3 Precision Feeding

2.3.1 AMS Feeding Recommendations

Current feeding recommendations for AMS begin with the concept of maintaining attendance to the AMS. As the number of AMS visits increases, milk production may correspondingly increase (Tremblay et al., 2016). Motivation to enter the AMS is primarily driven by the feed reward (explained previously in section 2.2; Prescott et al., 1998). Thus, it is perceived that the more concentrate provided in the AMS, the greater the motivation for cows to attend the AMS. The previous suggestion has led to the recommendation of feeding high quantities of concentrate in the AMS (Rodenburg, 2011). Many of these recommendations are upwards of 7.72 kg/d (Rodenburg, 2011), 8.0 kg/d (Lely, n.d.) or even 12.0 kg/d (DeLaval Inc., 2018). These recommendations are often based on individual farm surveys, failing to represent the industry on a larger scale and failing to implement control populations. Several AMS manufacturers (DeLaval Inc., Lely, GEA Farm Technologies) have developed AMS feeding strategies, utilizing feed tables (Table 2.1). DeLaval Inc. recommends a default of between 2.27 and 2.72 kg of concentrate in the AMS per visit (Brouk, 2017) equating to approximately 6.8 to 8.2 kg of concentrate supplied in the AMS per day. Adjustments to this default value are made based on an individual cow's milk production, and thus up to 12.0 kg of concentrate could be supplied in the

Table 2.1. Example of a feed table used to allocate concentrate (kg as fed/d) in the AMS based on parity and milk production.

	Production, kg/d				
Parity	20	30	40	50	60
Primiparous	5	6	7	8	9
Multiparous	6	7	8	9	10

AMS/d. Lely (n.d.) recommends supplying between 2.0 and 8.0 kg of concentrate in the AMS/d, again using feed tables to adjust for the differences in milk production. GEA Farm Technologies (2014) suggests a minimum of 0.5 kg/visit with a maximum of 2.0 kg/visit, or a daily maximum of 5.0 to 6.0 kg dependent on milk production. It is believed that offering greater than 2.0 kg of concentrate in the AMS/visit negatively influences ruminal pH dropping it into a critical range for a longer duration (GEA Farm Technologies, 2014). Further, GEA Farm Technologies also suggests that greater than 2.0 kg of concentrate in the AMS per visit will cause cows to visit the AMS less frequently and will not provide sufficient time to consume all the concentrate offerings during the milking event.

Many industry recommendations also include a feeding strategy where the PMR is formulated to balance for 6.0 to 8.0 kg of milk production below the herd average milk production (Lely, n.d.; DeLaval Inc., 2018; Rodenburg, 2011), or formulating a PMR to account for 80% or 90% of the total DMI and milk production in free-flow systems and guided-flow systems, respectively (Brouk, 2017). However, the basis behind these recommendations is not apparent and recent research has demonstrated high variability in AMS concentrate consumption across days when greater quantities of concentrate are supplied in the AMS (Hare et al., 2018; Menajovsky et al., 2018) thereby challenging these recommendations.

As a result of manufacturer and nutritionist recommendations, the quantity of concentrate supplied in the AMS is highly variable among farms (de Jong et al., 2003, Tremblay et al., 2016). One study examining 10 and 15 AMS farms in the USA and Canada, respectively, demonstrated that 78% of farms offered less than 5.0 kg/cow/d while 22% of farms offered greater than 5.0 kg/cow/d of concentrate in the AMS (de Jong et al., 2003). More recently, when 635 North American Lely AMS farms were analyzed (93% were free-flow), it was determined that for cows producing between 35 and 45 kg of milk per day, 5.6 to 7.1 kg of concentrate was provided in the AMS (Tremblay et al., 2016). However, there was a high degree of variation among farms (average SD = 2.2; Tremblay et al., 2016). That study also demonstrated a negative relationship between milk production and the quantity of concentrate allocation in the AMS. Evidently, farms are generally following the current industry recommendations, despite having limited scientific evidence to evaluate them.

Cows can consume TMR or PMR at a rate ranging from 50 to 150 g/min (Bach et al., 2007; Bach et al., 2009; DeVries et al., 2009), while they are able to consume pelleted concentrates at rates ranging from 250 to 400 g/min (Kertz et al., 1981). The average length of AMS milking event (box-time) is about 7 min (Castro et al., 2012) allowing cows to consume a maximum of 2.8 kg of concentrate per visit (Bach and Cabrera, 2017) or possibly even as low as 1.8 kg/visit (GEA Farm Technologies, 2014). If the previously mentioned values are extrapolated, the maximum amount of concentrate that could be consumed/day would average 8.4 kg if cows are visiting the AMS 3 times per day (Deming et al. (2013), 2.8 milkings per day; Wagner-Storch et al. (2003), 2.4 milkings/d; Bach et al. (2009): 2.2 milkings/d). Table 2.2 further illustrates the relationship between the targeted concentrate allocation and maximal concentrate allocation as affected by milking frequency. Thus, recommendations at or above 9.0 kg/d are not likely to be achieved, create a larger discrepancy between the formulated and consumed diet, and do not positively influence milking-related parameters.

There are currently three major options for cow traffic: free-flow; guided-flow; and forced-flow (Ketelaar-de Lauwere et al., 1998; Hermans et al., 2003; Bach et al., 2009). Free-flow systems allow the cows to roam freely throughout the barn visiting the feed bunk and AMS as they choose. Forced-flow systems direct cows in a unidirectional fashion with feed-first or milk-first orientations. The milk-first system refers to a traffic design where cows enter the AMS holding area prior to gaining access to the feeding area where PMR is offered, while the feed-first traffic flow refers to a system where cows enter the PMR feeding area prior to entering the AMS holding area. Guided-flow systems are an amalgamation of free-flow and the forced-flow systems and utilize pre-selection gates to guide cows through the barn. Thus, dependant on a cow's specific production information and activity, cows are permitted access to different areas of the facility (Jacobs and Seigford, 2012). It should be noted that guided-flow barns still impose a unidirectional movement as cows pass through pre-selection gates to guide them into the holding area prior to entering the AMS or other areas of the barn such as the free-stalls or feed bunk areas. Guided-flow barns are further described as milk-first or feed-first as described previously for forced-traffic barns.

Feeding strategy recommendations with the AMS seem to be dependent on the traffic flow design. Survey data from the University of Minnesota suggest that producers with free-flow

Table 2.2. Representation of pellet allocation qduantities and the arising potential concentrate delivery in the AMS based on milking frequency. This example assumes equal inter-milking intervals, consistent milking frequency, a maximum meal size of 2.5 kg/visit, and does not incorporate carry-over concentrate from the previous day.

	Target concentrate, kg/d				Target concentrate, kg/d				Target concentrate, kg/d			
	3	6	9	12	3	6	9	12	3	6	9	12
Milking Frequency	Required amount offered, kg/milking				Potentially offered, kg/milking				Maximum offered, kg/d			
2.0	1.50	3.00	4.50	6.00	1.50	2.50	2.50	2.50	3.00	5.00	5.00	5.00
2.5	1.20	2.40	3.60	4.80	1.20	2.40	2.50	2.50	3.00	6.00	6.25	6.25
3.0	1.00	2.00	3.00	4.00	1.00	2.00	2.50	2.50	3.00	6.00	7.50	7.50
3.5	0.86	1.71	2.57	3.42	0.86	1.71	2.50	2.50	3.00	6.00	8.75	8.75
4.0	0.75	1.50	2.25	3.00	0.75	1.50	2.25	2.50	3.00	6.00	9.00	10.00

AMS facility designs are feeding between 0.91 and 11.36 kg of concentrate in the AMS/cow/d, averaging 5.09 kg (Endres and Salfer, 2016). It was also observed that facilities with free-flowing AMS traffic are balancing the PMR for 4.55 to 13.64 kg below the average production within the herd (Endres and Salfer, 2016). Alternately, the same survey demonstrated that producers have the opinion that feed-first guided traffic AMS facilities are very similar to that of free-flow; however, milk-first guided traffic AMS designs have a very different feeding philosophy than that of free-flow and feed-first guided traffic-flow barn designs. The perceived differences between feed-first and milk-first guided traffic-flow barns is that the quantity of concentrate to be provided in milk-first AMS is simply offered to entice cows to visit the AMS and the PMR is formulated to offer a greater DMI (Endres and Salfer, 2016). Some believe that in milk-first guided flow systems, the main motivator of cows to enter the AMS is the PMR (Rodriguez, 2013). The main motivating factor for producers to construct guided traffic AMS barns are simply to save on the cost of the AMS concentrate because less of it can be offered in the AMS (Endres and Salfer, 2016). Thus, producers are offering between 0.91 to 5.45 kg of concentrate in the AMS per cow per day, averaging 3.49 kg. The quantity of concentrate offered per visit was denoted to be 0.68 to 1.36 kg (Endres and Salfer, 2016). Similarly, Rodenburg (2011), GEA Farm Technologies (2014), and DeLaval Inc. (2018) recommend providing greater quantities of concentrate in the AMS for free-flow traffic facilities. Brouk (2017), suggests that the composition of the pelleted concentrate supplied in the AMS and composition of the PMR are dependent on cow-traffic design. Under free-flow conditions, a lower protein and energy (from non-forage sources) based PMR is recommended to improve dependency of protein and energy on the AMS pellet, thereby encouraging visits to the AMS. Alternately, under guided-flow designs, greater energy (from non-forage sources) and protein densities in the PMR are recommended and there is less dependency of protein and energy from the AMS concentrate (Brouk, 2017). Rodenburg (2017) even suggests offering alternative feed sources in the AMS under guided-flow barn conditions, rather than the traditional hard concentrate pellet. While this is practiced in industry, there are currently no controlled studies testing this concept.

Irrespective of the barn traffic flow system, the goal of an AMS feeding program is to efficiently meet nutritional requirements while increasing voluntary visits to the AMS (reducing fetching) resulting in an optimum milking frequency and milk yield. It is often conceptualized that greater quantities of concentrate need to be supplied in free-flow AMS traffic-designed facilities because cows require greater rewards in the AMS to encourage motivation (Rodenburg, 2011; Brouk, 2017). Despite the differences in AMS feeding strategies for different traffic-flow, there is no scientific evidence to support that increasing concentrate provision improves milk or milk component yield in guided (Halachmi et al. 2005; Hare et al., 2018; Menajovsky et al., 2018) or free flow (Bach et al., 2007; Trembley et al. 2016) designs. In fact, Tremblay et al. (2016) reported a negative relationship between milk production in free-flow facilities and the quantity of concentrate allocated in the AMS. As such, while small differences may be present in feeding strategies among the cow traffic designs, many concepts including AMS intake variation among days, substitution of PMR for AMS concentrate, and cows increasing their refusals in the AMS as concentrate provision in the AMS increases are relevant irrespective of cow traffic.

2.3.2 Implications of Precision Feeding

In a survey-based study, AMS feeding management was ranked as the number one on-farm challenge (Salfer and Endres, 2018). Four important goals of AMS feeding programs include meeting nutritional needs, maintaining herd health, optimizing milk and milk component yield, and creating labour and economically efficient feed and feed delivery programs (Salfer and Endres, 2018). Precision feeding programs have the potential to improve the productivity and production efficiency by meeting the nutrient requirements on an individual cow basis (Cerosaletti et al., 2004; Gehman, 2011). As mentioned briefly, TMR feeding programs associated with parlour milking systems are potentially inefficient because diets are formulated to meet the nutrient requirements at a static level of production; however, some cows may not be meeting their nutrient requirements and others may be receiving more nutrients than they need (Bach and Cabrera, 2017). Though this feeding strategy is simple and time efficient, cows sort their feed throughout the day (Leonardi and Armentano, 2003), altering the TMR composition (Kononoff and Heirichs, 2003). Alternately, precision strategies with the AMS aim to meet the nutrient requirements for each individual cow, with the possibility of increasing the efficiency of production (Bach and Cabrera, 2017). This feeding strategy requires estimating the expected milk yield responses associated with any given concentrate supplementation and the corresponding change in PMR nutrient intake. Two studies (Maltz et al. 1991; Maltz et al., 1992) evaluated feeding strategies for concentrate supplementation when compared to TMR feeding. These studies included 2 feeding strategies including: 1) 1 kg of concentrate for every 2-kg of milk produced; or 2) accounting for BW changes on top of milk yield (Bach and Cabrera, 2017).

Unfortunately, results of both studies were inconclusive at determining individual cow responses to concentrate supplementation.

Milk production changes significantly throughout lactation, in response to different environmental conditions, concentrate composition, and by parity making it challenging to derive predictive models (André et al., 2011). Prototypes predicting production responses of concentrate intake were created and evaluated by both Duinkerken et al. (2003) and André et al. (2010). Utilizing precision feeding strategies may only seem effective when variation across cows is great; however, André et al. (2010) suggests that precision feeding strategies are justified based on individual milk production variation in response to concentrate supplementation from 3 weeks post-partum until late lactation. That said, future research is necessary to thoroughly evaluate the dynamic approach to concentrate feeding throughout the full duration of lactation and include the measurement of roughage intake and substitution rate determination (André et al., 2010).

Another challenge with precision feeding strategies is that most AMS are equipped with only one feed bin. Precision feeding strategies may require two or more feed bins to accommodate different pellet formulations (e.g. energy source and a protein source) or feed ingredients to meet nutrient requirements for cows depending on production, BW, and stage of lactation (Bach and Cabrera, 2017). An alternative approach may be to formulate a different pellet for early lactation cows, compared to the pellet provided throughout the remaining days of lactation. The pellet formulation for early lactation cows may contain expensive ingredients such as rumen protected choline and niacin to reduce the risk of ketosis and high non-esterified fatty acid (NEFA) concentration for fresh cows in a negative energy balance (Pires and Grummer, 2008). The potential advantages of precision feeding strategies to meet nutrient requirements for individual cows can be achieved only if there is precision in predicting nutrient intake and having cows achieve that nutrient intake (Bach and Cabrera, 2017).

Formulating rations for individual cows in an AMS comes with several complications. First, the increase in refused concentrate in the AMS as the concentrate allocation increases causes any unconsumed feed to be either discarded (if AMS is equipped with refusal removal system) or remain in the AMS feeder and be consumed by another cow (Bach and Cabrera, 2017). Inability to measure concentrate refused in the AMS causes an inaccurate representation of the quantity of consumed feed by individual cows in the AMS software. Further, achieving the targeted AMS allocation has been demonstrated to be challenging. Bach et al. (2007) targeted 3.0 or 8.0 kg/d of concentrate in the AMS; however, they only achieved 2.6 and 6.8 kg, respectively. Similarly, Halachmi et al. (2005) targeted treatments of 1.2 kg of concentrate per milking event or 7.0 kg/d with the later treatment only achieving 5.2 kg/d (Figure 2.2). To closely achieve the targeted quantities of concentrate in the AMS, a greater amount of AMS concentrate relative to the target should be offered (Hare et al., 2018; Menajovsky et al., 2018). Alternatively, the actual intake can be used within diet re-formulation to ensure that the diet formulated, and diet consumed are similar. It is also important to regularly calibrate the AMS feeder to ensure accurate concentrate quantities are being provided (GEA Farm Technologies, 2014; Brouk, 2017).

Increasing concentrate allocation in the AMS likely will not stimulate DMI. Studies that have monitored PMR and AMS intake consistently report a substitution of PMR for AMS concentrate intake. Bach et al. (2007), Hare et al. (2018), and Menajovsky et al. (2018) reported that for every 1 kg increase in AMS concentrate DMI, PMR DMI was reduced by 1.14, 1.58, and 0.84 kg, respectively. Thus, increasing AMS concentrate did not affect total DMI (Table 2.3). This substitution effect is important to recognize; however, further investigation is necessary to fully understand the direction of the affect because of inconsistencies across studies. The substitution effect has implications on the ability to use feed tables. Feed tables allow for an automatic adjustment of the concentrate offered based on milk production. The use of feed tables to supply AMS concentrate should be used with caution until the substitution effect has been comprehensively evaluated and is predictable. The limitation with feed tables is because an overall increase in total DMI is not obtained with increasing the quantity of concentrate in the AMS (Bach et al., 2007; Hare et al., 2018; Menajovsky et al., 2018) and thus feed tables may not be sufficiently meeting individual cow nutrient requirements: the primary goal of this feeding strategy. Alternatively, a PMR that closer represents a TMR and utilizes concentrate in the AMS to entice cows to voluntarily enter the AMS may be an appropriate strategy at this time (Salfer and Endres, 2018). In addition, maximum meal sizes/visit with AMS feeders are usually set between 2.0 (GEA Farm Technologies, 2014) and 2.5 (DeLaval Inc., 2018). Thus, cows visiting the AMS 3 times per day are unlikely to be able to consume greater than a feed table formulated value of 8.0 kg/d (Table 2.2).

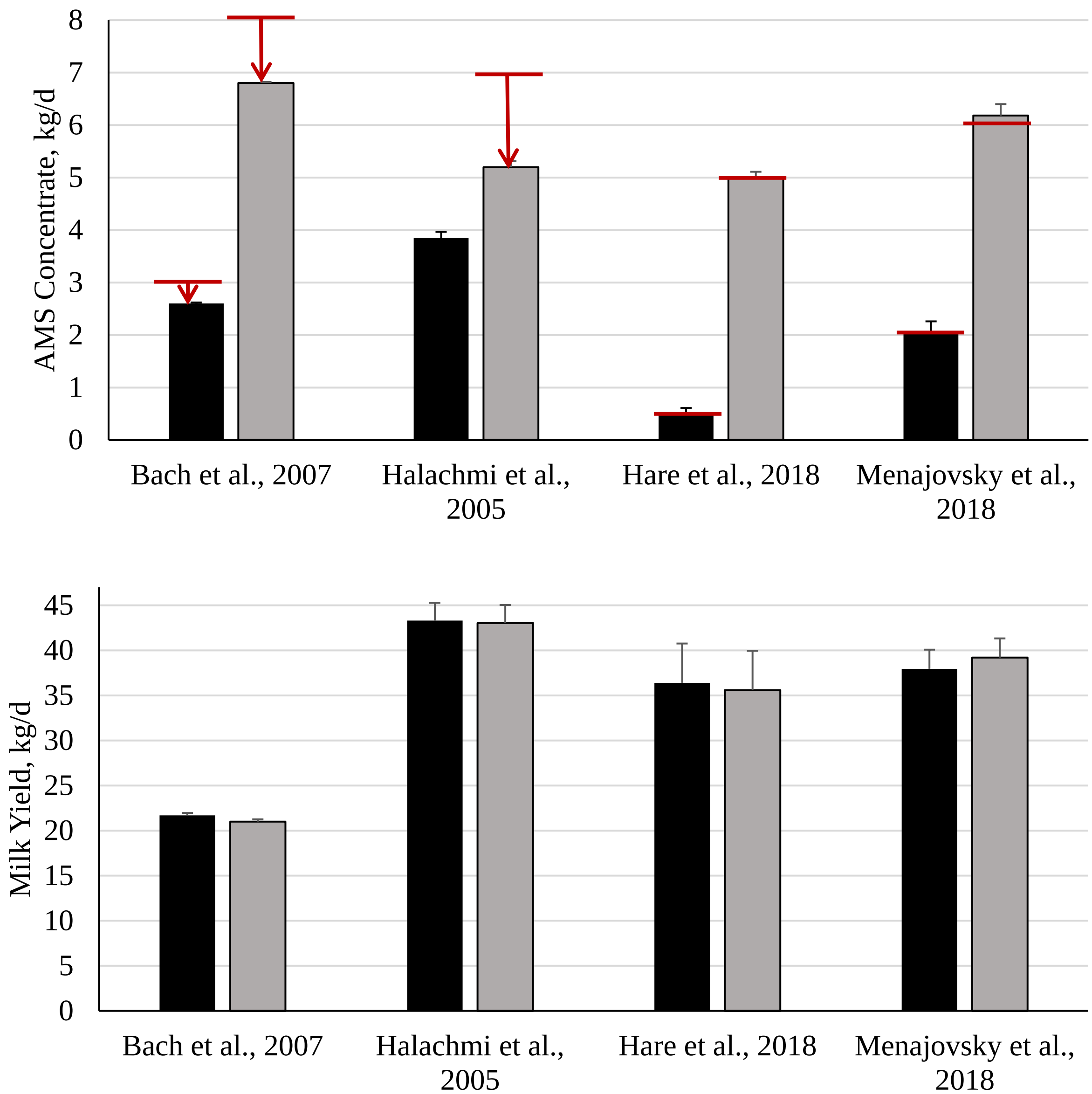

Figure 1.2. Targeted and actual concentrate delivery for cows in AMS feeding systems and their subsequent milk yield. The black bars indicate low AMS concentrate provision in each study and the grey bars indicate the high AMS concentrate provision. The red bar with an arrow indicates the target quantity for the AMS concentrate. Figure adapted from Penner et al. (2017).

Table 2.3. Substitution effect for previous AMS studies that also considered PMR intake. Substitution effect is defined as the kg of PMR intake reduction in response to a 1 kg (DM basis) increase in AMS concentrate intake.

Study	Treatment design	Substitution Effect (kg)
Bach et al., 2007	Equal energy densities	1.14
Hare et al., 2018	Equal energy densities	1.58
Menajovsky et al., 2018	High Forage PMR	0.78
	Low Forage PMR	0.89

When considering the available quantity of concentrate in the AMS at a specific visit, many things should be considered. Although the maximum meal size per visit and the target concentrate allocation are entered into the computer system, the actual quantity of concentrate delivered per visit is dependent on the inter-milking interval and total AMS visits per day for each cow (DeLaval Inc., 2018). In situations where cows attend the AMS less frequently than their previous 7-d average, there is “carry-over” concentrate from the previous day that is available to be allocated the following day in addition to the targeted amount (DeLaval Inc., 2018). Similarly, when cows attend the AMS more frequently than their 7-d average, less concentrate may be available the following day. Thus, greater or lower daily allocations of concentrate relative to the targeted quantity may be provided with infrequent milking intervals and variable milking frequency among days. In addition, infrequent inter-milking intervals within a day may cause different quantities of milk to be stored within the udder (Friggens and Rasmussen, 2001; Larsen et al., 2012) and provide different quantities of concentrate in the AMS across milking events. This “carry-over” effect and the dispense rate of the concentrate into the AMS feeder should be considered when developing feeding programs and therefore, maintaining frequent AMS visits should be targeted to limit the likelihood of over or under achieving the AMS concentrate target.

Feeding consistency of both the PMR and AMS pellet and understanding the goals of the farm are important to optimize utilization with the AMS (Salfer and Endres, 2018). Having consistent DM, mixing, delivery (time and number of deliveries), frequent and regular push-ups, and a palatable PMR are important to stimulate traffic to the AMS. Palatability and consistency of the AMS concentrate are also important factors for regulating AMS traffic (Salfer and Endres, 2018). Poor PMR composition, including poor forage quality and large variation in particle size may reduce palatability and PMR intake. Some suggest that when PMR palatability is poor, it may drive cows to visit the AMS more frequently, increasing their consumption of concentrate in the AMS, however, sorting of the PMR may be increased (Salfer and Endres, 2018). An increased AMS concentrate intake and PMR sorting may significantly decrease rumination time and increase susceptibility for sub-acute ruminal acidosis (DeVries et al., 2008). Thus, proper management, including storage (Borreani et al., 2018; Miller-Cushon and DeVries, 2017), chop length at harvest (Kononoff and Heirichs, 2003), and DM (Endres and Salfer, 2016) of feeds are important to maintain intake of PMR.

2.4. Transition Cow Feeding with AMS

Generally, the transition period for dairy cow refers to a 6-wk interval extending from 3-wk prior to calving until 3 wk post calving. Cows within the transition period experience significant physiological change as they progress from a pregnant and non-lactating state to a non-pregnant lactating state (Drackley, 1999). The transition period requires cows to adapt the new physiological state, but also to adapt to a new diet, develop a new social hierarchy, and be motivated to voluntarily enter the individual milking stall. In most cases, AMS require cows to perform relatively unnatural behaviours because cows are gregarious animals and often prefer to synchronize their behaviours with smaller groups within a herd (Benham, 1982). Conventional parlours allow for social milking: groups of cows are milked side-by-side. Thus, adjusting a herd from a conventional milking system or individual cows going through physiological changes surrounding the onset of lactation, require special attention to encourage voluntary visits to the AMS.

Various recommendations suggest providing concentrate to fresh cows in the AMS at a rate of 2.0 kg/d, while steadily increasing this quantity daily (Rodenburg, 2011; DeLaval Inc., 2018; Lely, n.d.) or even providing between 1.92 to 3.18 kg/d and increasing by 0.18 to 0.45 kg/d for the first 28 d (Salfer and Endres, 2018). Evidently, AMS feeding management of fresh cows is relatively unexplored and requires substantially more research to derive proper recommendations. Despite this, it is known that fresh cows are at the greatest risk of exhibiting metabolic diseases (Drackley et al., 1999) and ruminal acidosis (Penner et al., 2007; Penner et al., 2009). Rapidly transitioning fresh dairy cows to a greater diet fermentability does not enhance DMI or milk production (Dieho et al., 2016).

Precision feeding strategies with AMS parallel component feeding strategies that have been previously reported to increase sorting behaviour often reducing the F:C of the total diet and allowing for greater risk of digestive upset in early lactation (Coppock, 1977). A plausible hypothesis for feeding fresh cows (identified as cows 0 to 28 DIM; Salfer and Endres, 2018), may consist of feeding lower quantities of concentrate in the AMS, when compared to mid-lactation cows (Kokkonen et al., 2004). Reducing the quantity of concentrate supplied to fresh cows may lower the likelihood of diet-related metabolic diseases. After the initial 28 d post-partum (or in some recommendations 7 d; André et al., 2010), concentrate in the AMS may increase slowly at a predetermined rate. Early lactation cows (29 to 100 DIM) are often lead fed (Salfer and Endres, 2018). This means that more concentrate is provided in the AMS than is necessary, meeting requirements for greater milk production than current production quantities (Salfer and Endres, 2018). This concept is thought to encourage higher production assuming that nutrient supply helps drive milk production.

André et al. (2010) attempted to predict individual milk yield variation in response to concentrate intake in transition cows up to 3 wk post-partum. This study analysed data from 4 research herds with AMS, 2 of which were conventionally managed and 1 was an organic farm, and 1 research herd that fed concentrate in a conventional parlour milking system. A total of 5,629 records from 299 cows, comprised of 102 primiparous cows and 197 multiparous cows were used in this study. At calving, 1 to 3 kg/d of concentrate was provided. On 2 farms, concentrates slowly increased linearly over a 2 to 3-week period, until it reached a maximum, based on parity, while the other 2 farms increased concentrate linearly until d 10, where concentrate supply plateaued. Milk yield continued to increase, despite the concentrate allocation remaining constant (Figure 2.3). A model was developed to predict individual milk yield variation in response to concentrate intake. The model classified early lactation as a non-linear dynamic system, where daily milk yield and body weight change were response variables and concentrate intake was a controllable variable, linearly increasing from parturition over time. The predictive model considered the increase in milk yield over time following parturition with the fixed effect of parity and random effects of the individual variation in milk yield and the response to concentrate. Figure 2.4 represents the fitted individual milk yield response to concentrate intake by farm and parity within each farm. Model-based predictions for milk yield to concentrate intake in early lactation are not accurate as several parameters and interactions can influence the prediction, including feed utilization, mobilization rate and ensuring sufficient data is available can be challenging (André et al., 2010; Tess and Greer, 1990). This study suggests that an individual dynamic approach is only useful if there is sufficient variation between individual response variables. Although high levels of concentrate are not normally recommended during the transition period to prevent metabolic and digestive upset (Owens et al., 1998; De Brabander et al., 1999), this study suggest that high concentrate allocations may be applicable with individual and dynamic approach, if milk yield continues to respond,

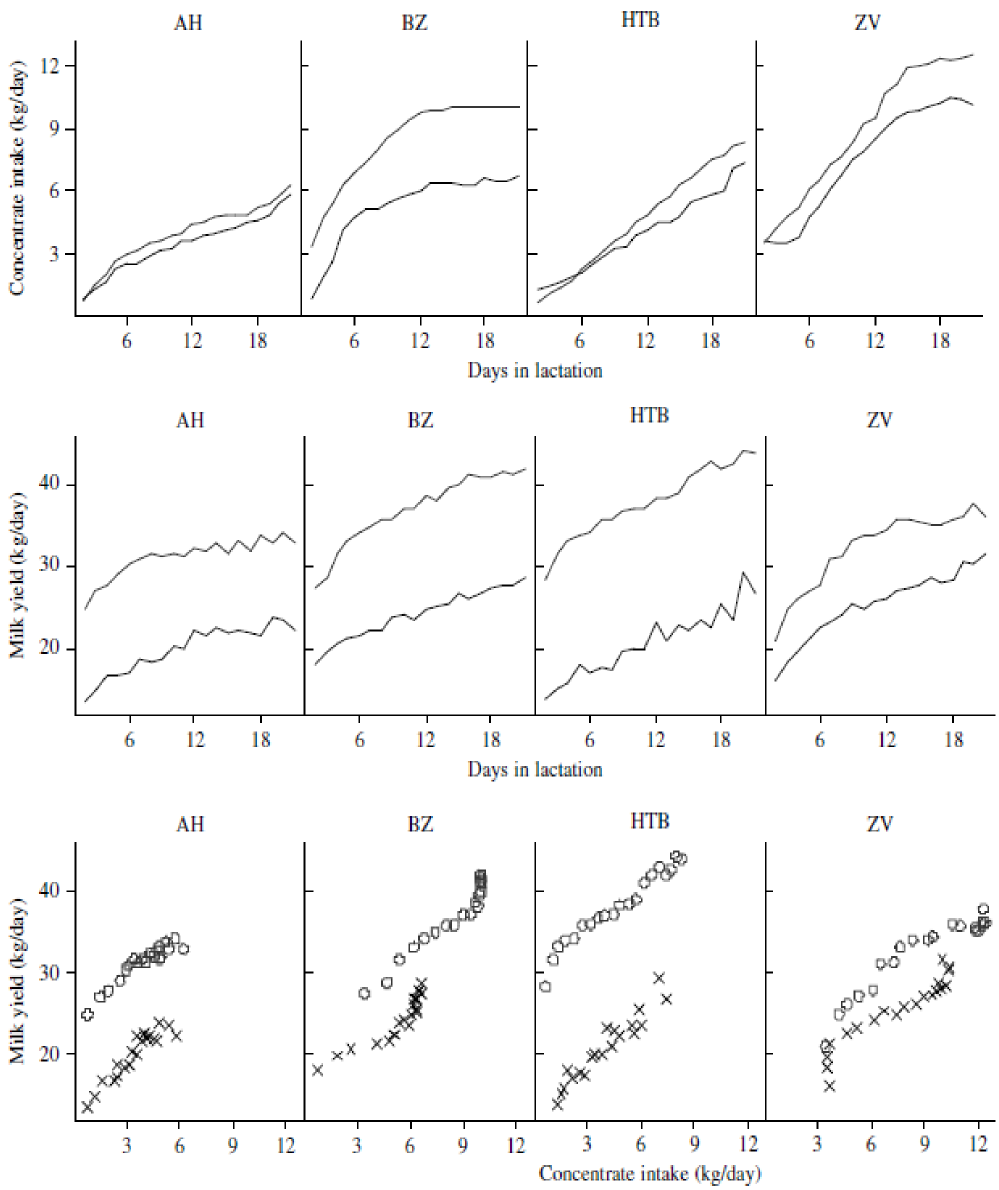

Figure 2.2. Averaged concentrate intake vs. days from calving (first row), averaged daily milk yield vs. days from calving (second row). Averaged milk yield vs. averaged concentrate intake at different days after calving (third row). Upper lines and symbols (o) are multiparous cows and lower lines and symbols (x) are primiparous cows. Farm locations are AH (Aver Heino), BZ (Bosma Zathe), HT (High-tech), and ZV (Zegveld). Figure sourced from André et al. (2010) with permission from publisher.

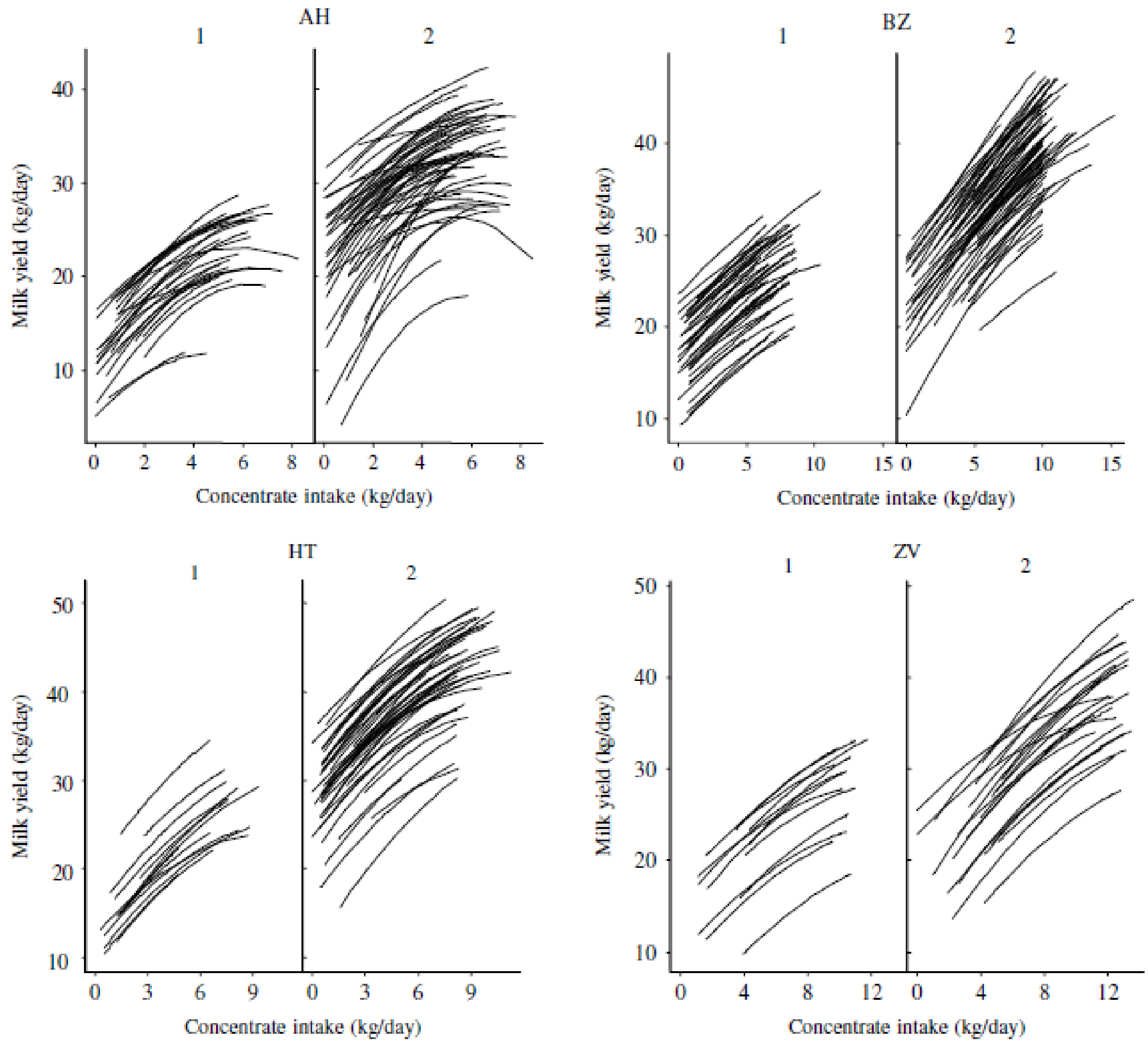

Figure 2.3. Fitted individual milk yield response curves vs. concentrate intake/farm. Different lines represent different cows (1=primiparous cows and 2=multiparous cows). Farm locations are AH (Aver Heino), BZ (Bosma Zathe), HT (High-tech), and ZV (Zegveld). Figure sourced from André et al. (2010) with permission from publisher.

with no digestive upset. Despite this consideration, caution should be used for previous outlined explanations regarding the unpredictable substitution affect and intake variabilities with high concentrate allocations.

Heifer management and training on the AMS is also important. Generally, heifers are trained on the AMS post-calving. This process includes trimming hair on the udders and tail, manually bringing cows to the AMS, proper feeder and rear plate placement determination, and calibrating the AMS to locate each individual teat for future AMS milking events for that cow. Usually it takes a few days of fetching heifers to the AMS for them to adapt to attending the AMS on their own. Siewart et al. (2017) analyzed data from 32 American AMS dairy farms and observed that primiparous cows voluntarily attend the AMS less frequently (Figure 2.5; Salfer and Endres, 2018) and thus regular fetching is more important. Primiparous cows should be fetched regularly to improve consistency and prevent early dry off or lower their realized peak milk production and lactation persistency (Salfer and Endres, 2018). Bach et al. (2007) demonstrated in multiparous cows that when more concentrate was provided in the AMS, fetching was reduced only for cows with low voluntary motivation to the AMS. This study concluded that cows frequently fetched cows are often being fetched for an alternate reason (lameness or illness) rather than lack of motivation to enter the AMS to be milked (Bach et al., 2007). Some recommendations for feeding management with AMS suggest training heifers on the AMS 2 to 3 weeks prior to their first lactation to reduce the number of stressors post-calving (Salfer and Endres, 2018; GEA Farm Technologies, 2014). Allowing heifers to regularly (some suggest 2 to 3 times daily) enter the AMS prior to calving and get used to the location, sounds, surroundings of the machine, and reducing the items to be learned post-calving may improve the transition (Salfer and Endres, 2018). Moreover, it is common perception that primiparous cows are over-dominated or intimidated by multiparous cows (Wierenga, 1990). Primiparous cows may also visit the AMS less frequently and consume less PMR at the feed bunk (NRC, 2001). Thus, it is recommended for herds using AMS, to house primiparous cows separately from multiparous cows (Grant and Albright, 1995). Bach et al. (2006), reported that primiparous cows housed with only other primiparous cows visited the AMS more frequently than when compared to primiparous cows housed with a group containing 70% multiparous cows and 30%

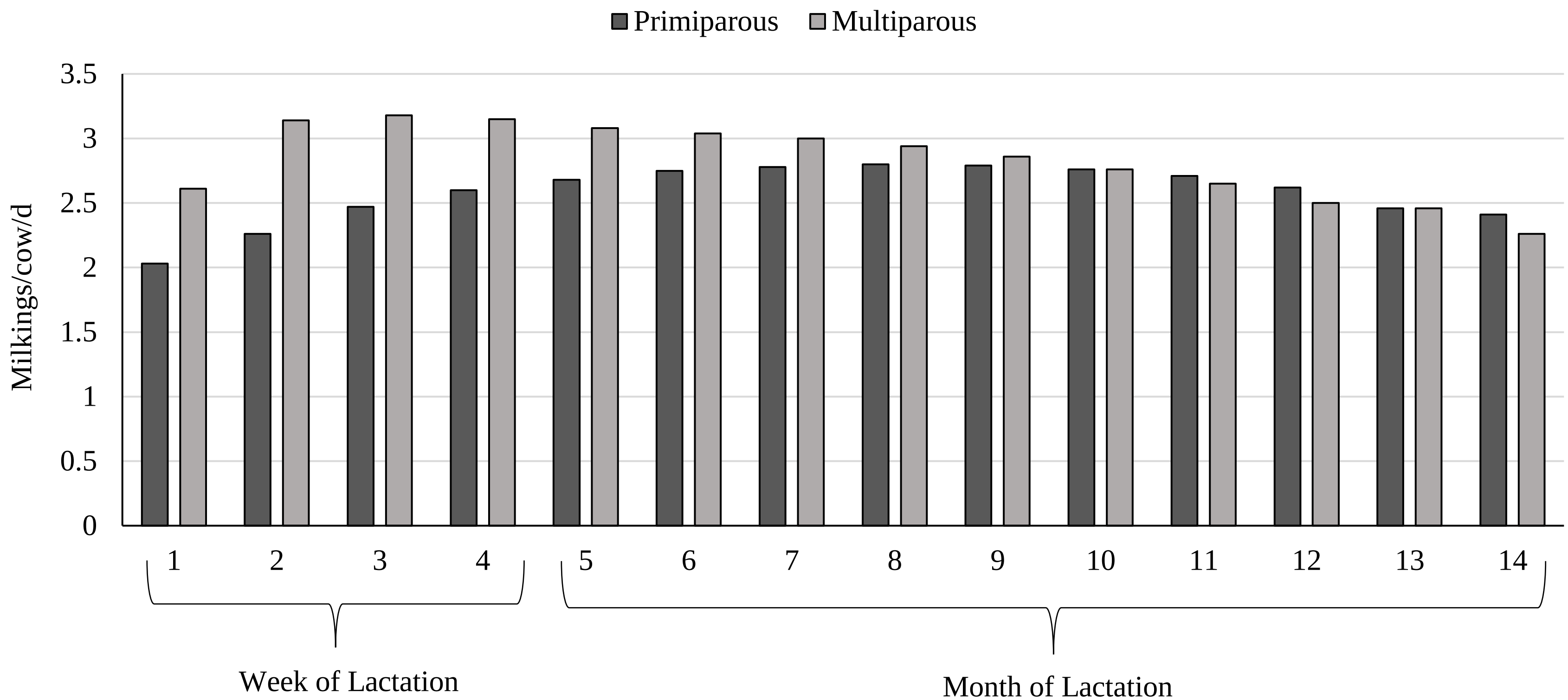

Figure 2.5. Visits of primiparous and multiparous cows to the AMS from 32 American AMS farms. Endres, University of Minnesota, personal communication (2018).

primiparous cows. Although the results of this study observed greater AMS visits in primiparous cows housed with only other primiparous cows, there was no differences in milk production between cows in the grouping strategies and primiparous cows housed with only other primiparous cows had one more PMR meal per day than primiparous cows housed with multiparous cows. This study suggests that primiparous cows in a free-flow traffic design may be not be as intimidated by the multiparous cows as once initially believed.

2.5. Feeding High Producing Cows Milked with an AMS

Generally, cows reach peak milk yield between 40 and 70 d post-partum (Olori et al., 1999). There are 2 main feeding strategies currently being practiced to encourage high milk production. These consist of AMS feed tables using either a “push” or “pull” strategy. First, using the concentrate in the AMS to push milk yield or by using the concentrate in the AMS to pull milk yield to reach a greater peak yield. The later, often includes lead feeding, which encompasses feeding greater quantities of concentrate in the AMS than necessary for current milk production in an attempt to increase peak milk yield and encourage persistency. Once peak yield has occurred, feeding according to production by stabilizing concentrate in the AMS is often recommended (Bach and Cabrera, 2017; Salfer and Endres, 2018; André et al., 2010). To accomplish a “push” feeding strategy, feed tables are formulated to provide concentrate in the AMS based on DIM or average milk production from the previous 7-d; however, they may also be formulated to provide concentrate in the AMS based on age and maximum milk yields from previous 7-d (DeLaval Inc., 2018). Considerations with heifer growth and maturation if using AMS feed tables to manage production should ensure they account for growth and production (Salfer and Endres, 2018). One study (Siewart et al., 2017) demonstrated that high producing cows are generally allocated greater quantities of concentrate in the AMS than lower producing cows suggesting compliance with existing feed tables. However, this associative data does not confirm that feeding additional grain improves milk production. Alternately, a study analyzing data from 635 North American Lely AMS farms reported a negative association between quantities of concentrate in the AMS and milk production (Tremblay et al., 2016). Feed tables designed to offer more concentrate to higher producing cows, a realization that AMS constraints are very limiting. Maximum meal sizes of concentrate in the AMS, combined with the number of daily visits to the AMS, may prevent cows from consuming greater than 6 kg of concentrate per day (Table 2.2). Therefore, further investigation of optimal feed tables for high production cows milked with an AMS is required to determine whether animals at different stages of lactation or production levels require different quantities of concentrate in the AMS.

2.6. Late Lactation and Dry Off Feeding with AMS

Generally, cows in late lactation are slowly reduced offering of concentrate in the AMS, in response to a decreased milk yield with advancing DIM (Bach and Cabrera, 2017; Salfer and Endres, 2018; Andre, 2010). There are two main feeding strategies for late lactation cows milked with AMS. First, some cows may still be producing large quantities of milk close to their dry-off period. Creating a feed table to help reduce milk production as the dry-off period approaches is a common mitigation strategy and is accomplished using feed tables based on DIM (Endres and Salfer, 2016). A complication that may occur with cows in late lactation is a reduction in milking frequency and an increase in the labour associated with fetching these cows. One strategy to combat this challenge is to improve the farm reproductive program to ensure high milk production through the end of lactation occurs (Endres and Salfer, 2016). That said, offering 2.7 vs. 0.5 kg/d of concentrate in the AMS in late lactation (207.9 DIM ± 8.54) increased milk yield with decreased inter-milking intervals (Shortall et al., 2018). Another strategy that is currently being recommended is to alter the milking permissions of cows in later lactation, while reducing the concentrate offered in the AMS to help slow down milk production to around 10 L/d and decrease voluntary AMS visits (Lely, n.d). While some suggest that cows milked twice daily in the week prior to dry-off produce more milk with no indications of behavioural discomfort after dry-off, it is believed that lower milk yields at dry-off reduces the risk of intra-mammary infections during the early dry period and at calving and thus may prove to be beneficial (Dingwell et al., 1999; Rajala-Schultz et al., 2005). Further investigation regarding optimal feeding strategies associated with cows later lactation preparing for dry-off need to be considered for accurate recommendations.

2.7. Conclusions

Optimal feeding strategies associated with AMS are an important consideration for high AMS efficiency and production; however, they are still not fully understood. It is evident that the primary motivating factor for cows to voluntarily attend the AMS is the feed reward provided at the AMS feeder (Prescott et al., 1998), usually in the form of a pelleted concentrate. Current industry recommendations generally suggest that providing large quantities (> 5.0 kg) of concentrate in the AMS will provide a stronger motivation for cows to enter the AMS voluntarily and result in an improved milk yield and milk composition, through the improved visits and precision feeding approaches (Rodenburg, 2011; DeLaval Inc., 2018; Lely, n.d.). Generally, these recommendations are developed using feed tables based on milk production and DIM. The exact range for the quantity of concentrate that should be provided to maintain attendance and milk performance has not yet been determined; however, greater AMS concentrate allocations cause greater variability in AMS concentrate intake (Hare et al., 2018; Menajovsky et al., 2018) and may not positively affect milk production (Halachmi et al., 2005; Bach et al., 2007; Tremblay et al., 2016; Hare et al., 2018; Menajovsky et al., 2018). In addition, a substitution effect occurs where PMR intake is reduced for every 1 kg increase in AMS concentrate intake (Bach et al., 2007; Hare et al., 2018; Menajovsky et al., 2018). Despite this substitution effect, it is not clear what factors drive the direction and extent of reduced PMR consumption. Further, as greater quantities of concentrate are allocated in the AMS, the ability to achieve these targets is significantly reduced (Halachmi et al., 2005; Bach et al., 2005). All studies to date, have demonstrated results of 2 different quantities of concentrates offered in the AMS. Therefore, more research is required to gain understanding to provide accurate, science-based recommendations to optimize milking frequency, milk and milk component yield.

2.8. Hypotheses

I hypothesized that increasing the quantity of concentrate provided in the AMS would result in reduced PMR consumption, greater variability in ruminal pH, and not positively affect voluntary attendance to the AMS or milk and milk component yield.

2.9. Objectives

The global objective of this research was to contribute to an improved understanding of AMS feeding management. Specifically, the objective was to evaluate how the quantity of concentrate provided in the AMS affects PMR DMI, voluntary attendance to the AMS, ruminal fermentation, total tract digestibility, and milk and milk component yield. This objective was accomplished by supplying varying amounts of concentrates in the AMS, while maintaining a constant energy level throughout the treatment groups by adjusting the PMR. The purpose of maintaining a constant energy level was to be able to determine an exact quantity of concentrates supplied in the AMS that provides sufficient motivation for cows to enter the AMS while maintaining a stable ruminal pH.

3.0 LOCATION OF CONCENTRATE ALLOCATION FOR COWS MILKED IN A GUIDED FLOW AMS ON PMR INTAKE, MILK AND MILK COMPONENT YIELD, RUMINAL FERMENTATION AND COW BEHAVIOUR

3.1 Introduction

Achieving efficient production with AMS requires frequent voluntary visits. It is thought that providing palatable concentrates in the AMS motivates voluntary attendance (Prescott et al., 1998); however, feeding management for AMS cows must consider both the AMS concentrate and the PMR. Feeding management strategies that stimulate voluntary attendance could increase milking frequency (Bach et al., 2007), improve AMS attendance (Wagner-Storch and Palmer, 2003), and result in greater milk yield (Tremblay et al., 2016; Bach and Cabrera, 2018). Although AMS adoption is increasing (Tse et al., 2017), few controlled studies have evaluated feeding management strategies.

Most published studies have only evaluated AMS concentrate provision (Halachmi et al., 2005; Melin et al., 2005; Tremblay et al., 2016). The focus on AMS concentrate has been based on the premise that increasing the quantity of concentrate may enhance voluntary visits to the AMS and may allow for precision feeding strategies (Bach and Cabrera, 2017). One study examined 10 American and 15 Canadian AMS farms and determined that 78% of farms offered less and 22% of farms offered greater than 5.0 kg/cow/d of concentrate in the AMS (de Jong et al., 2003). More recently, Tremblay et al. (2016) evaluated data from 635 North American AMS farms (primarily free-flow traffic) and reported that cows producing 35 to 45 kg/d of milk received a mean AMS concentrate allocation of 5.6 to 7.1 kg/d on an as fed basis. Notably, a large variation was reported across farms. Salfer and Endres (2014) noted that producers were feeding between 0.9 to 11.3 kg (free-flow traffic) or 0.9 to 8.2 kg (guided or forced-flow) of concentrate per cow per day in the AMS. Providing greater quantities of AMS concentrate is believed to improve milk yield and milk composition due to increased visits from increased motivation to attend the AMS. While survey data provides useful information of current production practices, results are association-based and cannot be used to compare or derive feeding management strategies.

Despite the commercial practice to feed large quantities of concentrate in the AMS, controlled studies evaluating concentrate provision in the AMS have not supported the suggestion that increasing the quantity of concentrate in the AMS will improve AMS concentrate intake, voluntary visits, or milk and milk component yield (Bach et al., 2007; Hare et al., 2018). Past research has also demonstrated that attempts to increase AMS concentrate provision are often unsuccessful, as the targeted AMS concentrate amounts are seldomly reached. Bach et al. (2007) targeted either 3.0 or 8.0 kg/d of concentrate in the AMS; however, only 2.6 and 6.8 kg/d of concentrate were achieved. Similarly, Halachmi et al. (2005) targeted 1.2 kg of concentrate per milking or 7.0 kg/d of concentrate in the AMS and achieved 3.5 and 5.2 kg/d, respectively. The discrepancy between the targeted quantity of AMS concentrate and that consumed suggests that cows were not consuming the formulated diet. Moreover, as AMS concentrate intake increases, PMR intake decreases in an unpredictable manner. Bach et al. (2007), in a free-flow design, and Hare et al. (2018) and Menajovsky et al. (2018), both in guided-flow systems, determined that for every 1 kg increase in concentrate consumed in the AMS, PMR intake was reduced by 1.14, 1.58, and 0.84 kg DM, respectively. While substitution rates are commonly evaluated in grazing research (Bargo et al., 2003), substitution effects with PMRs are not well understood. Continued research is needed to understand how AMS and PMR formulation strategies affect intake of dietary components (PMR and AMS) and production responses.

I hypothesized that increasing the quantity of concentrate in the AMS would reduce PMR consumption, increase variability in ruminal pH, without affecting voluntary attendance to the AMS or milk and milk component yield. The objective of this study was to evaluate whether AMS concentrate allocation affects PMR DMI, voluntary attendance to the AMS, feeding behaviour, cow activity, ruminal fermentation, total tract digestibility, and milk and milk component yield under iso-caloric dietary settings.

3.2 Materials and Methods

3.2.1 Animal Husbandry and Experimental Design

This study took place at the University of Saskatchewan's Rayner Dairy Research and Teaching Facility (Saskatoon, SK, Canada). Eight primiparous Holstein cows, previously fitted with silicone elastomer ruminal cannulas (Robyn Williams, Mount Evelyn, Victoria, Australia), were used in this study in a replicated 4 × 4 Latin square design. All animal use was preapproved by the University of Saskatchewan Research Ethics Board (protocol #20100021). At the start of the study cows averaged (mean ± SD) 90.6 ± 9.8 DIM and the 7-d milk yield prior to starting the study was (mean ± SD) 37.9 ± 6.0 kg/d. Cows were housed in a free-stall barn with 12 stalls and the barn was designed as a feed-first guided-traffic flow design with an AMS (DeLaval, Tetra Laval Group, Tumba, Södermanland, Sweden). All cows had permission to enter the AMS every 4 h or if the predicted milk yield was greater than 9.0 kg. A one-way gate prevented cows from entering the free-stall once in the feed bunk area. The feed bunk area contained 8 Insentec Feed Bunks (Hokofarm Group, Marknesse, Flevoland, The Netherlands) with an individual cow assigned to each bunk. Cows were trained on the Insentec bunks and AMS 6 weeks prior to the start of the study. For cows to return to the free-stall area from the feed bunk area, they had to pass through a pre-selection sort-gate. This gate directed cows toward the AMS when milking permission was granted or toward the free stall area when milking permission criteria were not met. If cows did not voluntarily enter the milking stall within 12 h, they were fetched and placed in the holding pen to be milked. Fetching times were restricted to 0400, 1030, 1730, and 2230 h daily and fetching activity was recorded. However, only 1 cow in 2 periods required fetching for 2 milking events.

Within each Latin square, cows were randomly assigned to 1 of 4 treatments with the sequence of treatments balanced to avoid carry-over effects. Periods were designed to consist of 19 d for dietary adaptation (1 to 19 of each period), a 4-d measurement phase for feeding behavioural and cow activity data collection (d 20 to 23 for each period), 1 d for device removal (d 19 for each period), and a 4-d phase for measurement of ruminal fermentation and total tract digestibility (d 25 to 28 of each period). Periods were designed to be 28 d in duration; however, periods 2, 3, and 4 were extended as the AMS required repairs during collection periods. As such, the actual duration of periods 1, 2, 3, and 4 were 28, 30, 32, and 40 d, respectively. Despite the extended periods, all data collected allowed for 4 consecutive days of behavioural measurements and 4 consecutive days for ruminal fermentation and total tract digestibility, as originally planned, with the adaptation phase extended.

3.2.2 Feeding Management and Experimental Treatments

In the present study, diets were formulated to be equal in macro- and micro-nutrient content. As such, cows in each treatment received the same total dietary nutrient provision when considering the sum of the PMR and the AMS concentrate. Thus, treatment groups differed in the amount of concentrate allocated in the AMS with targets of 0.5, 2.0, 3.5 or 5.0 kg/day (DM basis; Table 3.1). As the AMS concentrate target increased, there was an equal and corresponding reduction in the quantity of concentrate offered in the PMR. To avoid confounding effects, the pellet provided in the AMS was the same as that offered in the PMR and the forage-to-concentrate ratio (**F:C**) for each treatment (AMS concentrate + PMR) was 50:50. Diets were formulated for a 580 kg cow with an expected milk yield of 36 kg containing 4% fat and 3.2% protein using the CNCPS (6.55) platform of NDS (The RUM&N Company, Reggio Emilia, Italy).

Cows were provided their PMR in Insentec Feed Bunks (Hokofarm Group) with 1 cow assigned to each bunk to allow for measurement of PMR intake and feeding behaviour. The PMR was fed twice daily with 60% of the daily PMR allowance provided at 1100 h and 40% at 2230 h. The quantity of PMR refused was recorded at 1030 h daily and refusals were removed from the feed bunk. The PMR was provided for ad libitum consumption with refusals targeted to be between 5 and 10% (as is basis) of the total PMR offered. To achieve the specified DM provision of the AMS concentrate, the amount of concentrate offered in the AMS was monitored daily and adjustments were made every 4th day based on the mean intake of the previous 3 d. The amount of AMS concentrate eligible for each cow exceeded the target (0.51, 2.02, 3.52, and 5.03 kg for the 0.5, 2.0, 3.5, and 5.0 kg/d treatments, respectively, on a DM basis) to ensure that the target AMS consumption was achieved. The AMS feeder was calibrated weekly. To calibrate, the AMS feeder was cleaned, and 4 calibration samples were obtained directly from the feeder. The first sample was discarded to ensure material dislodged during the cleaning process did not affect the calibration outcome. The last 3 samples were weighed and an average of the 3 weights were entered into the computer system (Delpro 4.5, DeLaval).

To ensure each treatment contained the targeted F:C, forage components were sampled twice weekly, and concentrate component were sampled once weekly. Samples were used for DM determination (described below) and DM coefficients were updated as necessary.

3.2.3 Data and Sample Collection

The BW of each cow was measured on 2 consecutive days at the start of each period and at the end of the final period. Body weight was measured (0730 h) prior to the PMR feeding; however, it is important to note that time since milking, and the last AMS concentrate meal

Table 3.1. Ingredient and chemical composition of the total diets (PMR + AMS concentrate) used to test whether the location of concentrate provision affects DMI, milk production, ruminal fermentation and cow behaviour.

	Quantity of concentrate in the AMS			
Variable	0.5	2.0	3.5	5.0
Ingredient, % DM				
Barley silage	37.0	37.0	37.0	37.0
Alfalfa hay	17.4	17.4	17.4	17.4
Barley grain	15.2	15.2	15.2	15.2
PMR pellet[1]	27.0	20.4	13.9	7.4
Palmitic acid[2]	1.3	1.3	1.3	1.3
AMS pellet[1]	2.2	8.7	15.2	21.7
Chemical composition[3]				
DM, %	62.3	62.6	62.5	62.2
OM, % DM	92.3	92.3	92.3	92.3
CP, % DM	17.5	17.5	17.5	17.5
$aNDF_{OM}$[4], % DM	30.5	30.4	30.4	30.4
ADF, % DM	19.8	19.8	19.8	19.8
Starch, % DM	25.4	25.5	25.5	25.5
Ether extract, % DM	4.9	4.9	4.9	4.9
Ca, % DM	0.9	0.9	0.9	0.9
P, % DM	0.4	0.4	0.4	0.4
NE_L, Mcal/kg	1.69	1.72	1.72	1.72

[1]Premix supplied by CFRC which was incorporated into pellet contained: Crude protein, 0.7%, Calcium, 12.6%, Phosphorus, 1.8%, Magnesium, 4.1%, Potassium, 0.05%, Sodium, 12.4%, Chlorine, 10.0 %, Sulfur, 4.2%, Vitamin A, 126 800.9 IU/kg, Vitamin D_3, 49 592.3 IU/kg, Vitamin E, 1 087.6 IU/kg, Manganese, 29.7 ppm, Copper, 248.7 ppm, Iron, 1 635.3 ppm, Zinc, 363.7 ppm, Iodine, 23.1 ppm, Cobalt, 1.0 ppm, Selenium, 8.2 ppm, Ether extract, 0.005%. Pellet supplied by CFRC contained: Crude protein, 26.9%, Calcium, 1.5%, Phosphorus, 0.8%, Magnesium, 0.6%, Potassium, 1.0%, Sodium, 1.1%, Chlorine, 1.0%, Sulfur, 0.7%, Vitamin A, 11 484.1 IU/kg, Vitamin D_3, 4 488.6 IU/kg, Vitamin E, 98.4 IU/kg, Manganese, 5.4 ppm, Copper, 24 ppm, Iron, 179 ppm, Zinc, 38.6 ppm, Iodine, 2.1 ppm, Cobalt, 0.1 ppm, Selenium, 0.8 ppm, Ether extract, 5.2%.

[2]Source of palmitic acid was Energizer Rumen Protected (RP10) (Scothorn Nutrition, Grand Pré, NS).

[3]Average of the chemical composition from the metabolic measurement phase from each period

[4]$aNDF_{OM}$ treated with amylase and sodium sulfite and corrected for ash content.

varied. An average of the 2 BW measurements was calculated. Body condition score was collected independently by 3 trained personnel on d 1 of each period and at the end of the last period, using the 5-point scale described by Wildman et al. (1982). The individual scores were averaged to yield the value used for statistical analysis.

Feed intake, on an as fed basis, was recorded daily throughout the experiment. Data collected during the 4-d behavioural measurement phase and the 4-d ruminal fermentation and digestibility measurement phases were used for determination of PMR DMI. To calculate PMR DMI, individual ingredients and PMR refusals were collected daily for each cow. For refusals, 20% of the daily refusals were combined to form a composite prior to DM analysis. The silage sampling procedure involved collection of grab samples located throughout the face of the silage pit. The sample was mixed, and a 1-kg sub-sample was used for DM determination. Hay samples were collected from a pile of ground hay with grab samples taken from numerous regions of the pile. The individual grab samples within commodity type were composited, mixed, and sub-sampled. In addition, a 750-g sample was collected from each of the concentrates used in the diets. Samples were stored in a freezer at -20°C. The composited samples collected during the 4-d behavioural measurement period were used to determine DM and particle size distribution (Kononoff et al., 2003; described below). Composited samples from the ruminal fermentation and digestibility phase were analyzed for DM and chemical analysis. Dry matter was determined by placing a 500-g sample into a forced-air oven at 55˚C until the weight was constant. Subsequently, concentrate samples were ground through a 1-mm sieve using an Ultra Centrifugal Mill Type ZM 200 (Retsch GmbH & Co. KG, Haan, North Rhine-Westphalia, Germany), while silage and hay samples were ground using a Christy Norris grinder (Christy Norris Ltd., Chelmsford, Essex, England) equipped with a 1-mm sieve. The ground composites from the ruminal fermentation and digestibility phase were sent to Cumberland Valley Analytical (CVAS Ltd., Waynesboro, PA) for analysis of DM, OM, CP, NDF, $aNDF_{OM}$, ADF, ether extract, starch, $iNDF_{OM}$, Ca, and P. Analyses were completed as explained below.

3.2.3.1 Milk and milk component yield.

Milk yield was measured during the behavioural collection phase of each period using the AMS along with DelPro 4.5 (De Laval). The average milk yield across the 4-d collection period was used. In addition, the milk yield per visit, number of visits, milking duration (box-time), incomplete milkings on each quarter, quarters where the milker was kicked-off, and milkings where the milking machine was unable to find teats were recorded. Samples from each milking for each cow from the 4-d behavioural collection period were obtained via a sampling system connected to the AMS and a daily 40-mL composite (proportional to yield) was prepared for each cow in containers containing a Bronopol Microtab preservative (Dairy Herd Improvement Laboratory, Edmonton, Alberta). To minimize the duration samples were sitting at barn temperature, samples were retrieved from the sampling device every 4 h and transferred to a refrigerator for storage at 4˚C. After compositing, daily milk samples were sent to the Dairy Herd Improvement Laboratory (Edmonton, AB, Canada) for analysis of protein, fat, lactose, SCC, total solids, and milk urea nitrogen (**MUN**). Fat, protein, lactose, solids and MUN were determined using mid-infrared spectroscopy, while SCC was determined using flow cytometry. Samples were stored at 4˚C prior to submission.

3.2.3.2 Feeding behaviour responses.

The Insentec feed bunks that contained the PMR were connected and controlled via computer software (RIC Management Software, The Hokofarm Group). The software recorded the date, time, duration, and size of each PMR visit for each cow during the behavioural measurement phase within each period. These data were processed to remove visits to the feed bunk where no feed was consumed. The inter-meal intervals between each visit were then calculated and $\log_{10}$ transformed (Tolkamp et al., 1998). The transformed data were fit to normal distributions to determine appropriate meal criteria for each cow within each period using the procedure explained by DeVries et al. (2003) and the MIXDIST package (MacDonald and Green, 1988) of the R Statistical Analysis Software (The R Foundation, Adelaide, South Australia, Australia). The meal criteria were defined as the minimum time interval away from the feed bunk to identify a new meal. These data were then used to determine the number of meals (no./d), length of meals (min/meal), size of meals (kg), and rate of consumption (kg/min) using the procedure explained in Tolkamp et al. (1998). Once the daily determination of each variable was calculated, values were averaged among the 4-d behaviour collection period.

Feed sorting behaviour was analyzed during the behavioural measurement period of each treatment period. Partial mixed ration sorting behaviour was measured with the Pennsylvania State Particle Separator (**PSPS**), using the procedure described by Leonardi and Armentano (2003). All particle size measurements were conducted in duplicate (for each ingredient and refusals) for the composited samples according to Kononoff et al. (2003). The PSPS contained aperture openings of 19, 8, and 4 mm, with the remaining material caught on a pan.

3.2.3.3 Cow-activity budget responses.

Accelerometers (HOBO Pendant ® G Data Logger, Onset, Bourne, MA) were placed on the hind right leg of each cow the day prior to the start of the behavioural phase, with placement following the protocol described by Zobel and Chapinal (2013). Devices were removed after 4-d of continuous data collection for each period and the data were downloaded onto a computer. The number of standing and lying bouts and the duration of each bout were determined using SAS (SAS Institute Inc., Cary, NC), as described by Zobel and Chapinal (2013) with the algorithms of Ledgerwood et al. (2010). Data were summarized by cow and period

3.2.3.4 Ruminal fermentation and total tract digestibility.

Ruminal pH was measured during the behavioural measurement phase of each period to ensure that ruminal pH values were not affected by the ruminal digesta sampling protocols (described below) and resulting changes in activity. Ruminal pH was measured using the Lethbridge Research Centre Ruminal pH Measurement System (**LRCpH**; Penner et al., 2006). The LRCpH was inserted through the ruminal cannula into the ventral sac of the rumen, to enable 96 consecutive h of ruminal pH data collection. The LRCpH was programmed to log data every 1 min. Prior to insertion into the rumen and following removal from the rumen, the LRCpH was maintained at 39˚C for standardization in pH buffers 7 (RICCA Chemical Company, Arlington, TX) and 4 (Fisher Chemical, Battle Ground, WA). Upon retrieving the LRCpH, mV data were downloaded to a computer. The relationship between mV and pH derived from the starting and ending standardizations were used to convert the recorded mV values to pH units assuming a linear offset between the starting and ending regressions. Data were summarized to determine the daily minimum, mean, maximum, and duration and area that pH was less than 5.8, as described by Penner et al. (2007).

Ruminal digesta and fecal samples were collected over 4 consecutive days during the metabolic phase of each period. Samples were collected at 12 h intervals with a 3 h offset among days to represent a 24-h cycle. At each time point, 250-mL of ruminal digesta were collected from each the cranial, central, and caudal regions of the rumen fluid/rumen mat interface. The mixed digesta (750 mL) was strained through two layers of cheesecloth, filtrate was mixed, and sub-samples of ruminal fluid filtrate were obtained. One 10-mL sample was added to a 15-mL vial with 2 mL of 25% meta-phosphoric acid for the analysis of short-chain fatty acid (**SCFA**) concentration and the second 10-mL sample was added to a 15-mL vial with 2 mL of sulfuric acid that was subsequently analyzed for ammonia concentration. These samples were sealed and stored at -20˚C until analysis.

Corresponding to the time of ruminal fluid sampling, 200 g of feces was collected directly from the rectum of each cow. Following collection, the fecal sample was thoroughly mixed, and 125 g was added at each collection time-point to a plastic container to form a 1000-g composite per cow. The fecal samples were stored at -20˚C until thawed to prepare duplicate 500 g samples. These duplicate samples were placed in a 55˚C forced-air oven to determine DM as previously described. Fecal samples were then ground using the Ultra Centrifugal Mill ZM 100 grinder (Retsch GmbH & Co. KG, Haan, North Rhine-Westphalia, Germany) to pass through a 1-mm sieve. The ground samples were sent to Cumberland Valley Analytical Services for determination of DM, OM, CP, $aNDF_{OM}$, ADF, starch, ether extract, iNDF, and ethanol soluble carbohydrates (described below).

3.2.3.5 Sample Analyses

Feed samples collected during the behavioural phase (d 20 to 23) and the feed, refusal, and fecal samples from the metabolic phase (d 25 to 28) were dried and ground (previously described) to pass through a 1-mm sieve. Forage samples were dried at 105˚C for 3 h (National Forage Association recommendations, 2002) and concentrate samples and palmitic acid were analysed for DM by drying in an oven at 135˚C for 2 h (method 930.15, AOAC 2000). Crude protein was analysed by nitrogen combustion (method 990.03, AOAC 2000) with a Leco FP-528 Nitrogen Combustion Analyser (Leco, MI, USA). Neutral detergent fibre and ADF were analysed using Whatman 934-AH glass micro-filters with 1.5 um particle retention (method 973.18, AOAC 2000). The NDF analysis was conducted with the addition of sodium sulfite and α-amylase and was corrected for ash (**$aNDF_{OM}$**) by ashing the sample in a furnace (535˚C) for 2 h. Indigestible NDF (**iNDF**) was determined by measuring the remaining NDF after 240 h of incubation in ruminal fluid in vitro. Samples from the behavioural phase were used to determine diet composition for d 20 to 23, while samples from the metabolic phase were used for diet composition and nutrient digestibility determination. Ether extract was analysed (method 2003.05, AOAC 2006) using a Tecator Soxtec System HT 1043 extraction unit (Tecator, Foss NA, Eden Prairie, MN). Starch was analysed using the method described by Hall (2009). Ash was analysed by heating a 1.5-g sample to 550ºC for 4 h (method 942.05, AOAC 2000) and OM was calculated by subtracting the ash concentration from 100%. Calcium was determined using a dry-ash procedure (method 927.02, AOAC 2000) followed by atomic absorption (Perkin-Elmer, Model 2380, CN, USA). Phosphorus was determined using a dry-ash procedure (method 965.17, AOAC 2000) and concentration was read on a spectrometer at 410 nm (Pharmacia, LKB-Ultrasepc®III, Stockholm, Södermanland, Sweden).

Ruminal fluid samples, preserved with 25% meta-phosphoric acid, were thawed overnight at 4˚C and composited (equal volume basis) the following morning to yield 1 sample/cow/period. Sample preparation for gas chromatography followed the protocol described by Khorasani et al. (1996). The concentration of SCFA was measured using an Agilent gas chromatograph (6890 series with FID, Agilent Technologies Inc., Santa Clara, CA). Samples were injected using a 17:1 split ratio at 170˚C. The column was a Phenom FFAP (Agilent Technologies Inc., Santa Clara, CA) and the initial oven and detector temperatures were 90˚C and 250˚C, respectively. The oven temperature increased at a constant rate of 10˚C/min. Ruminal fluid samples that were frozen with sulphuric acid were thawed overnight at 4˚C. These samples were then composited as described previously for SCFA and analysed using the procedure described by Fawcett and Scott (1960). Briefly, the supernatants from the centrifuged samples were transferred in duplicate into glass test tubes with standard solutions (sodium phenate, nitroprusside and hypochlorite) and a standard curve was prepared using distilled water with the standard solutions. After a 1-h incubation period, these samples were analyzed in a spectrophotometer (SPECTRAmax®PLUS^{384}, Molecular Devices Corporation, San Jose, NC). The values determined by the spectrometer were used in calculations to determine the concentration of ammonia. If the duplicate samples had greater than a 7% coefficient of variation, they were prepared and re-run.

3.2.4 Statistical Analysis

The PROC UNIVARIATE (SAS Institute Inc., Cary, NC) procedure was used to determine if the data and residuals were normally, identically, and independently distributed prior to further analysis. All data were normally distributed. Statistical analyses were completed using the MIXED procedure of SAS 9.4 (SAS Institute Inc., Cary, NC). The model included the fixed effects of treatment and period, and the random effect was cow within Latin square.

Polynomial contrasts were used to evaluate linear, quadratic, or cubic effects of treatments. Significance was declared when $P \leq 0.05$ and trends were declared at $0.05 < P \leq 0.10$.

Data for AMS concentrate intake, PMR intake, and total DMI were also analyzed using repeated measures with the day as the repeated variable. Covariance error structures were tested to determine which one yielded lowest AIC and BIC values. The covariance structure that best suited the data was compound symmetry. The same statistical model was used to evaluate AMS concentrate intake, PMR intake, and total DMI data, with the exception that the model included the fixed effects of day and the day × treatment interaction.

A two-tailed T-test was used to evaluate if PMR sorting behaviours were different from 100 for each particle length within each treatment. If the sorting index was greater than 100, this indicated selective sorting for that particle length, while a sorting index less than 100 indicated selective avoidance for that particle length.

3.3 Results

3.3.1 Dry Matter Intake and Feeding Behaviour

The BW (648 kg) and BCS (3.33) of cows were not affected by treatment ($P_{linear} \geq 0.23$, Table 3.2). The quantity of AMS concentrate consumed for each treatment was 0.50, 2.00, 3.49 and 4.93 kg DM/d ($P_{linear} < 0.001$) with quantities consumed being similar to the targets of 0.5, 2.0, 3.5 and 5.0 kg/d, respectively. The standard deviation of AMS concentrate intake across days linearly increased as the quantity offered increased ($P_{linear} < 0.001$). Intake of PMR decreased linearly ($P_{linear} < 0.001$) as the quantity of concentrate in the AMS increased. The slope of the linear regression between PMR and AMS concentrate intake indicated that for every 1.00 kg increase in AMS concentrate consumed, PMR intake decreased by 0.97 kg DM. The standard deviation of PMR intake across days was not different among treatments (1.45 kg/d; $P_{linear} \geq 0.69$). Total DMI was not affected by treatment, averaging 25.3 kg/d ($P_{linear} \geq 0.96$). Energy intake of AMS concentrate increased linearly ($P_{linear} < 0.001$) from 0.91 to 8.86 Mcal/d, while energy intake of the PMR decreased linearly ($P_{linear} < 0.001$) from 42.93 to 34.41 Mcal/d, as the quantity of concentrate in the AMS increased. Total energy intake did not differ across treatments (44.02 Mcal/d; $P_{linear} = 0.88$).

The number of PMR meals (6.7 meals/d) and size of PMR meals (3.48 kg/meal) did not differ among treatments (Table 3.2; $P_{linear} \geq 0.23$). The duration of PMR meals tended to increase linearly ($P_{linear} = 0.061$) from 32.3 min/meal for cows fed 0.5 kg of concentrate in the AMS to

Table 3.2. Partial mixed ration (PMR) intake, intake of concentrate in the automated milking system (AMS), total DMI, and feeding behaviour of cows fed increasing quantities of concentrate in the AMS with a concurrent and equal reduction the proportion of concentrate in the PMR.

	Quantity of Concentrate in the AMS[5]					*P*-value		
Variable	0.5	2.0	3.5	5.0	SEM	Linear	Quadratic	Cubic
AMS concentrate intake, kg/d	0.50	2.00	3.49	4.93	0.08	< 0.001	0.80	0.93
Standard deviation in AMS intake, kg	0.06	0.24	0.51	0.85	0.08	< 0.001	0.28	0.94
PMR intake, kg/d	24.7	23.6	21.3	20.5	1.20	< 0.001	0.30	0.14
Standard deviation in PMR intake, kg	1.60	1.25	1.70	1.25	0.33	0.69	0.88	0.26
Total DMI, kg/d[1]	25.2	25.6	24.8	25.5	0.60	0.96	0.66	0.10
Energy Intake, Mcal/d								
AMS Energy Intake	0.91	3.59	6.29	8.86	0.10	< 0.001	0.69	0.91
PMR Energy Intake	42.93	40.91	37.09	34.41	1.26	< 0.001	0.74	0.19
Total Energy Intake	43.84	44.50	43.35	44.37	1.28	0.88	0.78	0.18
PMR feeding behaviour[2]								
PMR Meals, no./d	7.1	6.9	6.3	6.6	0.41	0.23	0.53	0.39
PMR Meal size, kg	3.6	3.4	3.5	3.2	0.18	0.29	0.87	0.40
PMR Meal duration, min	32.3	34.9	35.7	45.8	4.60	0.061	0.43	0.60
PMR consumption rate, g/min	110	98	100	84	6.30	< 0.001	0.61	0.06

Table 3.2 Continued. Partial mixed ration (PMR) intake, intake of concentrate in the automated milking system (AMS), total DMI, and feeding behaviour of cows fed increasing quantities of concentrate in the AMS with a concurrent and equal reduction the proportion of concentrate in the PMR.

	Quantity of Concentrate in the AMS[5]					*P*-value		
Variable	0.5	2.0	3.5	5.0	SEM	Linear	Quadratic	Cubic
PMR sorting index[3], %								
Particles > 19 mm	102.78	94.58	96.19	86.90	8.69	0.015	0.89	0.24
Particles 8 to 19 mm	104.29	104.03	108.48	105.13	5.69	0.49	0.49	0.22
Particles 4 to 8 mm	110.78[z]	110.65[z]	117.33	111.91[y]	6.22	0.41	0.34	0.13
Pan (< 4 mm)	80.88[z]	93.71	94.50	101.60	5.49	< 0.001	0.32	0.16
Body condition score[4]	3.30	3.33	3.33	3.35	0.12	0.36	0.89	0.76
Body weight, kg	645	646	651	650	22.20	0.23	0.82	0.45

[z]Significant difference from a sorting index of 100 ($P \leq 0.05$)

[y]Tendency ($P < 0.10$) for the sorting index to be different from 100

[1]Total DMI includes PMR and AMS intake

[2]Calculated using inter-meal intervals following procedure in DeVries et al. (2003).

[3]Sorting index was calculated using description in Leonardi and Armentano (2003).

[4]Body condition was measured using the 5-point scale defined by Wildman et al. (1982).

[5] All total diets (AMS concentrate + PMR) were isocaloric. Pellet offered in AMS was the same as the pellet in the PMR and thus a corresponding decrease in PMR pellet with increasing AMS concentrate.

45.8 min/meal for cows fed 5.0 kg treatment. Consumption rate of the PMR linearly decreased from 110 g/min to 84 g/min as the AMS concentrate was increased from 0.5 kg/d to 5.0 kg/d ($P_{linear} < 0.001$).

As the quantity of AMS concentrate increased, selection of particles retained on the 19-mm sieve linearly decreased ($P_{linear} = 0.015$) from 102.8 to 86.9 %, and cows linearly increased sorting against particles retained on the pan (Table 3.2; $P_{linear} < 0.001$). However, based on a two-tailed t-test, the sorting index was only different from 100 % for cows fed 0.5 kg/d AMS and particles retained on the bottom pan (80.9 %, $P_{linear} = 0.044$). In addition, cows fed 0.5 and 2.0 kg/d AMS concentrate (110.8 and 110.7 %, respectively) sorted for particles retained on the 4-mm sieve ($P_{linear} \leq 0.039$).

3.3.2 Activity Budget

There were no differences ($P \geq 0.173$; Table 3.4) for the number of standing bouts (10.2 bouts/d), average duration of a standing bout (79.9 min), the number of lying bouts (10.3 bouts/d), or the average duration of a lying bout (71.2 min). No differences ($P \geq 0.11$) among treatments were also observed for total standing duration (12.4 h/d) or the total duration cows spent lying each day (11.6 h/d).

3.3.3 Voluntary Milkings, Milk Yield, and Milk Composition

Milking frequency (3.2 visits/d), inter-milking interval (449 min) milk yield (37.4 kg/d), milk fat yield (1.43 kg/d), and milk protein yield (1.22 kg/d) were not different among treatments (Table 3.3, $P_{linear} \geq 0.42$). However, MUN linearly decreased as the quantity of concentrate provided in the AMS increased ($P_{linear} \geq 0.017$). While yields were not affected, the concentration of milk protein was affected cubically ($P_{cubic} = 0.002$) with the greatest concentration of milk protein occurring when 2.0 kg of concentrate was provided in the AMS. Milk fat concentration increased and then decreased as the quantity of AMS concentrate increased ($P_{quadratic} = 0.039$) with the greatest concentration of milk fat (3.98%) occurring when 3.5 kg of concentrate was provided in the AMS. Likewise, the concentration of total milk solids was quadratically affected as the quantity of concentrate in the AMS increased ($P_{quadratic} = 0.016$) with the greatest concentrations occurring for cows fed 2.0 and 3.5 kg of AMS concentrate.

The average length of a milking event (box-time; 7.03 min/milking) and total time spent being milked/day (22.42 min/d) were not affected by treatment (Table 3.3; $P_{linear} \geq 0.78$). Milking events that consisted of at least one quarter being incompletely milked (8.53%) or at

Table 3.3. Milk yield, milking frequency, voluntary attendance, and milk composition for cows fed increasing quantities of concentrate in the automated milking system (AMS) with a concurrent and equal reduction the proportion of concentrate in the partial mixed ration.

	Quantity of Concentrate in the AMS[1]					*P*-values		
Variable	0.5	2.0	3.5	5.0	SEM	Linear	Quadratic	Cubic
Milking frequency, no./d	3.2	3.3	3.1	3.3	0.18	0.82	0.26	0.14
Inter-milking interval, min	448.9	443.6	474.6	432.7	29.14	0.81	0.29	0.16
Milk yield, kg/d	37.7	37.6	37.3	37.0	2.64	0.59	0.96	0.97
Fat, kg/d	1.4	1.4	1.5	1.4	0.06	0.46	0.53	0.56
Protein, kg/d	1.2	1.3	1.2	1.2	0.06	0.42	0.64	0.30
Milk composition								
Fat, %	3.87	3.89	3.98	3.81	0.19	0.70	0.039	0.12
Protein, %	3.26	3.38	3.26	3.28	0.09	0.54	0.040	0.002
Lactose, %	4.71	4.68	4.71	4.68	0.04	0.28	0.93	0.12
MUN, mg/dL	17.4	16.9	17.1	16.1	0.55	0.019	0.38	0.17
Total Solids, %	12.9	13.0	13.0	12.8	0.29	0.34	0.016	0.85
Total kickoffs, no./d	0.1	0.1	0.2	0.2	0.06	0.49	0.61	0.37
Kick-offs, % of milkings/d	4.17	2.60	5.99	4.17	1.97	0.71	0.95	0.27
Total incomplete milkings, no./d	0.2	0.3	0.3	0.4	0.14	0.20	0.89	0.66
Incomplete milkings, % of milkings/d	5.21	8.85	9.11	10.94	4.34	0.28	0.80	0.75
Milkings with teats not found, no./d	0.16	0.09	0.06	0.03	0.07	0.19	0.82	0.92
Average box time, min/d	7.0	7.2	7.1	6.9	0.34	0.78	0.58	0.85
Total box time, min/d	22.2	22.9	21.8	22.8	1.71	0.84	0.84	0.39

[1]All total diets (AMS concentrate + PMR) were isocaloric. The pellet offered in AMS was the same as the pellet in the PMR and thus a corresponding decrease in PMR pellet with increasing AMS concentrate.

Table 3.4: Lying budgets of cows fed increasing quantities of concentrate in the automated milking system (AMS) with a concurrent and equal reduction the proportion of concentrate in the partial mixed ration.

	Quantity of concentrate in the AMS[2]					*P*-value		
Variable[1]	0.5	2.0	3.5	5.0	SEM	Linear	Quadratic	Cubic
Lying time, h/d	11.1	11.3	11.9	12.0	0.62	0.11	0.89	0.63
Lying bouts, no./d	10.8	10.6	9.9	9.9	0.91	0.47	0.86	0.24
Average lying bout duration, min/d	65.1	74.8	66.9	77.8	6.31	0.17	0.90	0.10
Standing time, h/d	12.9	12.7	12.1	12.0	0.62	0.11	0.89	0.63
Standing bouts, no./d	10.6	9.9	10.7	9.7	0.91	0.47	0.85	0.21
Average standing bout duration, min/d	77.2	85.5	73.5	83.5	10.51	0.84	0.91	0.20

[1]Variables calculated using procedure outlined by Zobel and Chapinal (2013) and coding of Ledgerwood et al. (2010).

[2]All total diets (AMS concentrate + PMR) were isocaloric. The pellet offered in AMS was the same as the pellet in the PMR and thus a corresponding decrease in PMR pellet with increasing AMS concentrate.

least one teat cup being kicked-off during a milking event (4.23%) were also not different among treatments ($P_{linear} \geq 0.28$).

3.3.4 Ruminal Fermentation and Digestibility

Minimum (5.65), maximum (6.68) and mean (6.18) ruminal pH were unaffected by treatment ($P_{linear} \geq 0.62$). The duration that ruminal pH was < 5.8 tended to be cubically affected with the numerically greatest duration when fed 2.0 kg of AMS concentrate (Table 3.5; $P_{cubic} \geq 0.068$). The standard deviation of the daily average ruminal pH decreased as the quantity of concentrate increased ($P_{linear} = 0.048$). Total SCFA concentration in ruminal digesta was affected by a cubic response as the quantity of concentrate provided in the AMS increased; total ruminal SCFA concentration was least when 3.5 kg of concentrate was provided in the AMS (111.62 m*M*, $P_{cubic} = 0.025$). Molar proportions of acetate (62.8%), propionate (22.4%), butyrate (11.2%), valerate (1.4%) and isovalerate (1.2%) were not affected by the quantity of concentrate offered in the AMS ($P_{linear} \geq 0.10$). Ruminal concentration of isobutyrate was affected cubically as the amount of concentrate provided in the AMS increased ($P_{cubic} = 0.021$), with the greatest concentration of isobutyrate occurring in the 0.5 and 3.5 treatments. The molar proportion of caproate linearly increased as the quantity of AMS concentrate increased ($P_{linear} = 0.011$). Ruminal ammonia concentration linearly decreased as allocation of AMS concentrate increased ($P_{linear} = 0.011$).

Apparent total tract digestibility of CP (70.71%), starch (95.51%) and OM (70.73%) were not affected by treatment (Table 3.6; $P \geq 0.144$). However, total tract digestibility of DM ($P_{linear} = 0.096$) and $aNDF_{OM}$ ($P_{linear} = 0.036$) decreased linearly as AMS concentrate increased, while ADF digestibility tended to be quadratically affected by quantity of AMS concentrate ($P_{quadratic} = 0.059$). Apparent total tract digestibility of ether extract increased linearly with increasing AMS concentrate ($P_{linear} < 0.001$).

Qualitatively, variation in ruminal pH, AMS visits and PMR meals varied significantly across days during the behaviour measurement period for both the 0.5 kg treatment (Figure 3.1) and the 5.0 kg treatment (Figure 3.2). Substantial variation within individual cows among days for daily feeding activities and ruminal pH were observed. In addition, it is apparent that cows do not always consume the PMR prior to visiting the AMS, as the feed-first guided traffic design would suggest.

Table 3.5: Average daily rumen fermentation results including rumen pH, ammonia and SCFA concentration from cows fed increasing quantities of concentrate in the automated milking system (AMS) with a concurrent and equal reduction the proportion of concentrate in the partial mixed ration (PMR).

	Treatment[3]					*P*-value		
Variable	0.5	2.0	3.5	5.0	SEM	Linear	Quadratic	Cubic
Ruminal pH[1]								
Minimum pH	5.68	5.67	5.67	5.57	0.06	0.75	0.17	0.12
Maximum pH	6.69	6.63	6.72	6.69	0.05	0.65	0.69	0.16
Average pH	6.19	6.11	6.21	6.19	0.06	0.62	0.51	0.17
Duration pH <5.8 (min)	197	269	129	141	66.00	0.12	0.48	0.068
Area pH <5.8 (pH × min)	47.8	45.0	20.9	25.1	20.07	0.15	0.80	0.43
Standard deviation of average daily ruminal pH	0.11	0.12	0.07	0.06	0.02	0.047	0.57	0.38
Rumen ammonia[2], mg/dL	12.1	11.2	10.3	10.2	0.70	0.011	0.33	0.540
Total concentration[2], m*M*	116.9	116.9	111.6	116.1	2.58	0.21	0.10	0.025
Acetate, %	62.47	62.58	63.07	62.97	0.73	0.10	0.68	0.41
Proprionate, %	22.72	22.83	21.87	22.18	0.53	0.13	0.78	0.16
Isobutyrate, %	0.79	0.76	0.79	0.75	0.01	0.14	0.96	0.021
Butyrate, %	11.18	11.06	11.39	11.26	0.41	0.53	0.98	0.31
Isovalerate, %	1.21	1.22	1.23	1.16	0.06	0.49	0.37	0.69
Valerate, %	1.37	1.31	1.34	1.36	0.03	0.86	0.10	0.33
Caproate, %	0.26	0.25	0.31	0.32	0.03	0.011	0.48	0.15

[1]Ruminal pH measures were collected every 1 minute during the 4-d behavioural measurements for each period. Daily averages are represented.

[2]Rumial ammonia and SCFA concentration determination were collected during the 4-d metabolic measurements for each period. Daily averages are represented.

[3]All total diets (AMS concentrate + PMR) were isocaloric. The pellet offered in AMS was the same as the pellet in the PMR and thus a corresponding decrease in PMR pellet with increasing AMS concentrate.

Table 3.6: Apparent total tract digestibility for DM, OM, CP, NDF, ADF, and starch for cows fed increasing quantities of concentrate in the automated milking system (AMS) with a concurrent and equal reduction the proportion of concentrate in the partial mixed ration (PMR).

	Treatment[1]					*P*-value		
Digestibility	0.5	2.0	3.5	5.0	SEM	Linear	Quadratic	Cubic
DM, %	69.22	69.38	68.85	68.34	0.81	0.096	0.41	0.69
OM, % DM	70.82	70.82	70.42	70.05	0.75	0.17	0.67	0.83
CP, % DM	71.20	71.31	70.41	69.92	1.03	0.22	0.72	0.70
Starch, % DM	95.15	95.19	96.10	95.60	0.51	0.14	0.42	0.14
$aNDF_{OM}$, % DM	48.34	48.36	47.22	46.31	1.14	0.036	0.51	0.66
ADF, % DM	39.62	41.18	40.00	37.98	1.41	0.14	0.059	0.63
Ether extract, % DM	94.98	98.11	99.07	99.33	0.24	<0.001	<0.001	0.18

[1]All total diets (AMS concentrate + PMR) were isocaloric. The pellet offered in AMS was the same as the pellet in the PMR and thus a corresponding decrease in PMR pellet with increasing AMS concentrate.

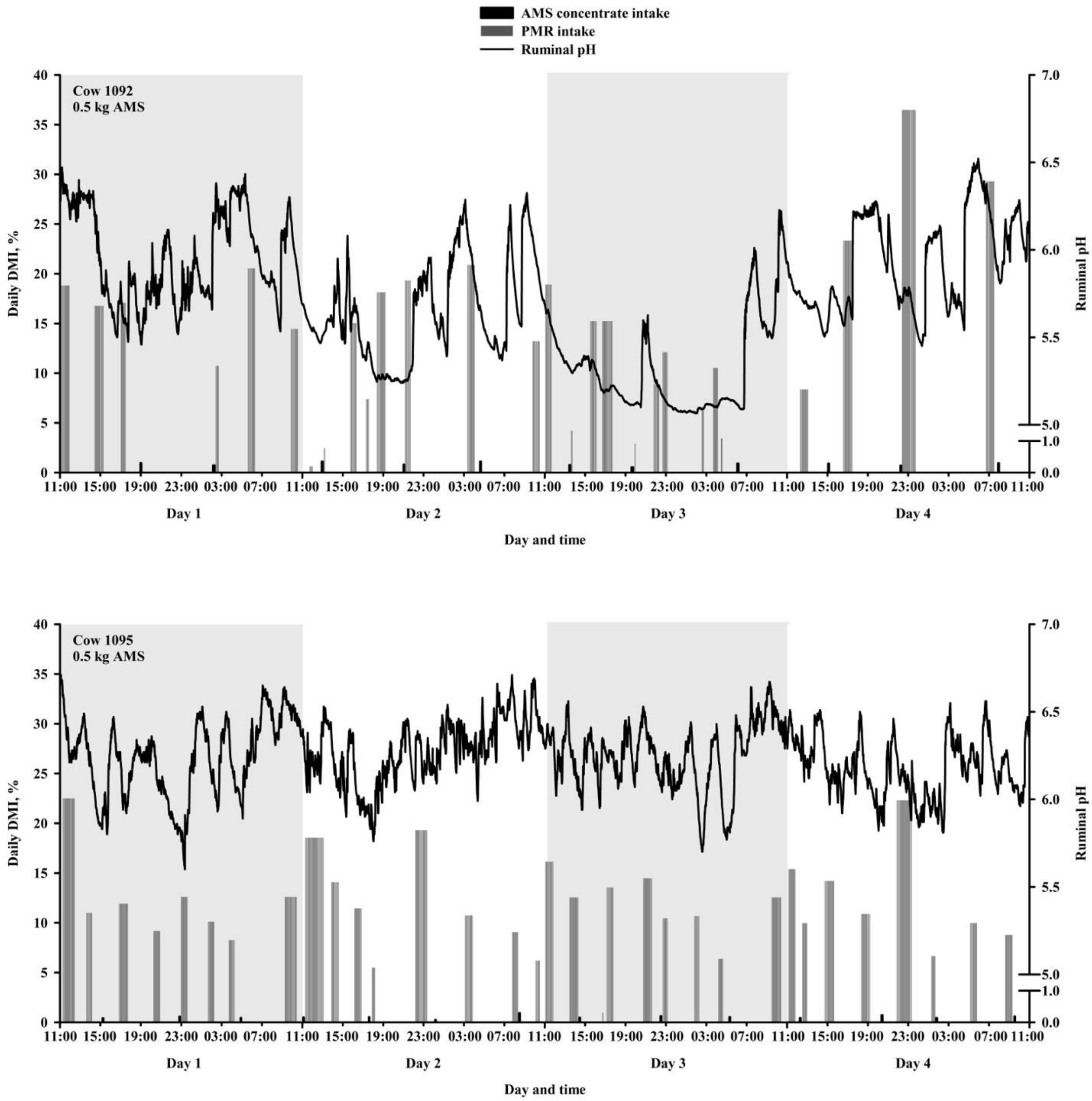

Figure 3.1. Day-to-day variation in feeding behaviour, visits to the AMS, and ruminal pH for individual cows and between selected cows when fed 0.5 kg of concentrate in the AMS.

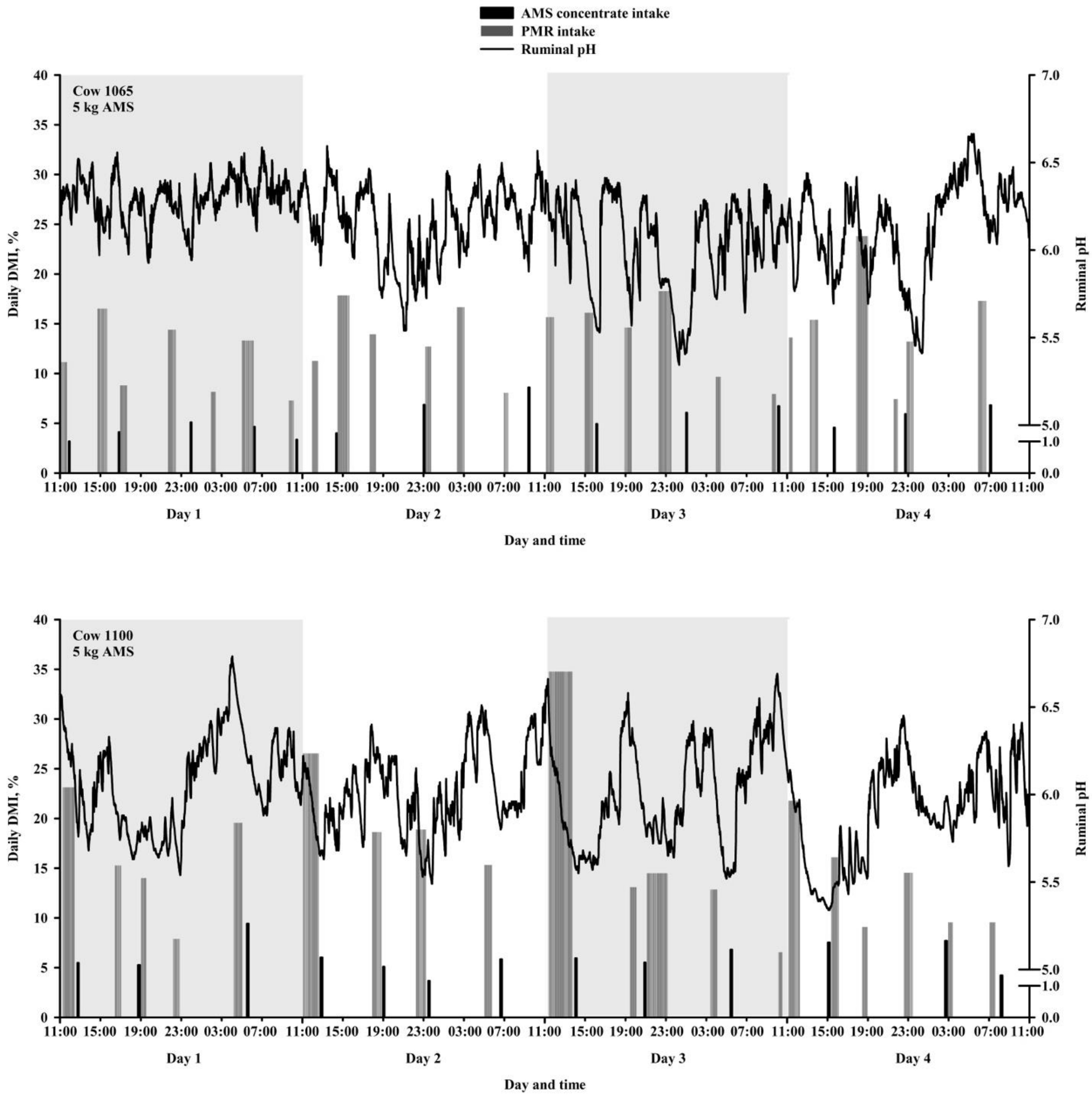

Figure 3.2. Day-to-day variation in feeding behaviour, visits to the AMS, and ruminal pH for individual cows and between selected cows when fed 5.0 kg of concentrate in the AMS.

3.4 Discussion

Feeding management strategies associated with cows milked using AMS are becoming increasingly important as the adoption of AMS continues to increase worldwide (Jacobs and Siegford, 2012; Tse et al., 2017). Researchers have previously evaluated the effect of the quantity of concentrate offered in the AMS when maintaining an iso-caloric diet (Bach et al., 2007; Hare et al., 2018) or using the concentrate to increase the nutrient density (Halachmi et al., 2005; Tremblay et al., 2016; Menajovsky et al., 2018). Regardless of the dietary strategy, a consistent response is that increasing the quantity of concentrate offered in the AMS does not necessarily stimulate an improved voluntary attendance to the AMS, milk or milk component yield, or DMI. While studies have been conducted to evaluate the effect of concentrate allocation under iso-caloric settings, previous studies have been limited to a 2-treatment approach (Bach et al., 2007; Hare et al., 2018) and, as such, are unable to adequately characterize the response to increasing AMS concentrate provision. The linear increase in AMS concentrate allocation, along with a concurrent and equal reduction in concentrate provision in the PMR in the present study had no effect on total DMI, visits to the AMS, or milk and milk component yield. These findings suggest that when dietary nutrient composition is equal, increasing AMS concentrate provision (at the expense of concentrate provision in the PMR) in a feed-first guided traffic design does not affect production responses. Findings in the present study are generally supported by previous research including Bach et al., (2007) and Hare et al. (2018).

A challenge with most previous AMS studies is that the targeted concentrate consumption in the AMS was not achieved (Halachmi et al., 2005; Bach et al., 2007). In the current study, the computer-programmed concentrate allocation was slightly greater than the targeted concentrate consumption to ensure consumption was similar to the target. For example, the programmed values of 0.51 (SD = 0.014), 2.02 (SD = 0.041), 3.52 (SD = 0.085) and 5.03 (SD = 0.142) kg DM were entered into the computer software system to achieve the 0.5, 2.0, 3.5 and 5.0 kg targeted DM amounts, respectively, with the resulting consumption being 0.50, 2.00, 3.49 and 4.93 kg/d in the AMS. In addition to requiring a computer-programmed AMS allocation that exceeded the target consumption, feeding greater quantities of AMS concentrate increased variation in AMS concentrate intake among days. In fact, the standard deviation for the mean concentrate intake among days increased from 0.06 to 0.85 kg/d when the AMS concentrate target increased from 0.5 to 5.0 kg/d. Only 1 previous study, known by the authors, has reported variability in AMS concentrate intake among days (Menajovsky et al., 2018). In that study, Menajovsky et al. (2018) reported that when 6.0 kg of concentrate was provided in the AMS, the standard deviation for mean AMS concentrate intake among days was 0.85 kg/d and was greater than when compared to feeding 2.0 kg of concentrate in the AMS (SD = 0.25). Although not presented in the publication of Hare et al. (2018), data obtained from the authors demonstrate greater variation in AMS concentrate intake among days when 5.0 kg (SD = 0.78) were offered in comparison to 0.5 kg (SD = 0.07) of concentrate in the AMS. The variability in AMS concentrate intake among days when providing larger quantities of concentrate in the AMS is expected to reduce consistency of nutrient intake and uniformity in the diet consumed.

The majority of studies focusing on AMS feeding management have only reported AMS concentrate intake (Halachmi et al., 2005; Melin et al., 2005; Tremblay et al., 2016) while a few have reported both AMS and PMR intake (Bach et al., 2007; Hare et al., 2018; Menajovsky et al., 2018). In the present study, total DMI did not differ among treatments when considering both AMS concentrate and PMR intake. The lack of response for total DMI occurred as cows linearly decreased PMR intake in response to a linear increase in AMS concentrate intake. In fact, I observed that for every 1 kg DM increase in AMS concentrate intake, PMR DMI decreased by 0.97 kg DM. Previous studies reporting substitution ratios (the reduction in PMR intake for every kg increase in AMS concentrate) in AMS do not appear to be consistent and ratios as low as 0.78 (Menajovsky et al., 2018) and as great as 1.58 (Hare et al., 2018) have been reported. In another study (Bach et al., 2007), a reduction of 1.14 kg DM in PMR intake was observed for every 1 kg increase in AMS concentrate consumed. While a nearly 1:1 substitution rate was observed in the present study, the inability to predict the substitution rate among the AMS concentrate and PMR preclude the ability to impose precision feeding management strategies. In pasture-based situations, a reduction in pasture DMI is often observed when cows are provided supplements (Kellaway and Porta, 1993). This substitution rate is believed to explain variations in milk response to supplementation (kg milk/kg supplement; Bargo et al., 2003) as it has been observed that when the rate of substitution is high, milk response to supplementation is low (Bargo et al., 2003). That said, relative to grazing scenarios where cows are provided concentrate supplementation, the substitution rates observed in AMS are relatively high and, in some cases, represent the potential for negative effects on DMI (Bargo et al., 2003). It has been hypothesized that the substitution rates and variation in milk responses in pasture-based systems may be influenced by pasture-related parameters, cow genetics, individual milk production, and stage of lactation (Bargo et al., 2003). However, factors affecting the substitution ratio are not well defined for dairy cattle milked with AMS and require further investigation.

As stated above, understanding how cows adjust their PMR feeding behaviour is important for feeding management in AMS. Hare et al. (2018) and Menajovsky et al. (2018) have reported that increasing the quantity of concentrate in the AMS reduces PMR consumption and alters the sorting behaviour, eating rate, and meal size of the PMR. For example, Hare et al. (2018) reported that when 5.0 kg of concentrate was provided in the AMS, cows sorted for particles retained on the 8-mm sieve and against particles retained on the pan, when compared to 0.5 kg of concentrate allocated in the AMS. Those researchers also found greater PMR meal durations, feeding rates, and meal sizes when cows were fed 0.5 kg of concentrate in the AMS. Alternately, Menajovsky et al. (2018) reported that when 6.0 kg of concentrate was offered in the AMS, cows discriminated more against particles retained on the 8-mm sieve, while when 2.0 kg of concentrate was offered in the AMS, cows increased sorting against particles retained on the pan. Menajovsky et al. (2018) also observed shorter daily eating durations (min/d) for cows fed a greater quantity of concentrate in the AMS; however, no other differences in PMR feeding behaviours were observed. In the present study, increasing the quantity of concentrate in the AMS (and correspondingly decreasing the concentrate in the PMR) resulted in selective avoidance for particles retained on the top sieve of the PSPS. In addition, a linear increase in sorting against particles collected on the pan was observed as the proportion of concentrate in the AMS decreased. The sorting responses corresponded to a linear increase in meal duration and a reduction in eating rate as the quantity of concentrate consumed in the AMS increased. The increased meal duration and reduced eating rate likely provided a mechanism to allow for greater sorting. It is not clear why cows in the present study increased sorting against long particles and reduced their sorting against fine particles as the proportion of AMS concentrate increased. However, because the PMR of the 0.5 kg treatment contained the lowest F:C, cows had a greater ability to sort against long particles (DeVries et al., 2007). Such sorting behaviour can be expected to further alter the composition of the diet consumed relative to that formulated and delivered and has implications for feeding management (Miller-Cushon and DeVries, 2017). Thus, feeding strategies must consider both the AMS and PMR as the AMS concentrate provision influences feeding behaviours associated with the PMR.

As AMS feeding strategies impose a component feeding system, understanding the impact of the site where concentrate is offered (AMS vs. PMR) on ruminal fermentation is critical. Only one previous study has reported ruminal pH, SCFA concentrations, and ruminal ammonia concentrations for cows milked in AMS (Menajovsky et al., 2018). In the present study, I did not observe any treatment effects on the minimum, mean, or maximum ruminal pH. The lack of response was surprising given the marked changes in the quantity of concentrate offered in the AMS and PMR, the changes observed for feeding behaviour (meal duration and eating rate), and PMR sorting characteristics. However, when considering the AMS and PMR components collectively, the dietary treatments were formulated to be equivalent suggesting that detectable but subtle changes in eating patterns do not necessarily have marked impacts on ruminal pH when total DMI is consistent. Moreover, the maximum meal size offered in the AMS was 2.5 kg. It is likely that constraining the maximum meal size limits the impact of dietary concentrate on ruminal fermentation. In support of the present study, Maekawa et al. (2002) observed no differences in ruminal pH when comparing component feeding to TMR-feeding when the diet composition was the same; however, those authors did conclude that component feeding is likely to alter the F:C allowing cows to consume more concentrate than was intended. Menajovsky et al. (2018) reported that increasing the dietary energy density by reducing the F:C of the PMR reduces ruminal pH, while increasing the dietary energy density by increasing the AMS concentrate allocation did not affect the ruminal pH response. This suggests that with the maximum meal size imposed in the AMS, changing the site of concentrate allocation would likely not affect ruminal pH.

While ruminal pH was not affected, ruminal SCFA concentration was least when 3.5 kg of concentrate was provided in the AMS. There is no clear explanation to justify the change in ruminal SCFA concentration, but it may be possible that the response is related to differences in activity patterns among cows and treatments. In the present study, ruminal digesta samples were collected in attempt to obtain a good representation over a 24-h cycle; however, the timing of the PMR meals and AMS visits relative to the timing of sampling may not have been consistent among treatments and periods as the day-to-day movement patterns of cows varies. In addition, cows in AMS systems may not follow a 24-h pattern (Bach and Cabrera, 2017). Thus, changes in ruminal SCFA concentrations may be partially affected by activity patterns of cows in addition to the dietary treatments.

Ruminal ammonia concentrations decreased linearly with increasing AMS concentrate provision. As with total ruminal SCFA concentrations, I cannot rule out the fact that activity patterns may have influenced this response. In a previous study, Menajovsky et al. (2018) reported no difference in ruminal ammonia concentrations across treatments when different quantities of concentrate were allocated in the AMS. However, in the present study, total tract digestibility of DM, $aNDF_{OM}$, and ADF tended to be reduced and ether extract linearly increased with increasing amounts of concentrate provided in the AMS supporting the concept that ruminal fermentation may have been altered. Also, despite total dietary formulations in the current study being equivalent, Idid observe increased variability for AMS concentrate intake among days as the allocation of concentrate in the AMS increased. It is hypothesized that this linear increase in variability of AMS concentrate intake and changes in sorting behaviour of the PMR influenced ruminal ammonia concentrations and negatively affected DM, $aNDF_{OM}$, and ADF digestibility. I am unable to explain the cause for improved ether extract digestibility.

The results for reduced digestibility of DM, $aNDF_{OM}$, and ADF are not consistent with the literature when supplementation is provided, at least when the supplement has a greater nutrient density than the basal forage or PMR (Bargo et al., 2003). Menajovsky et al. (2018) reported greater DM digestibility when 6.0 kg of concentrate was provided in the AMS, while ADF digestibility was greatest when 2.0 kg of concentrate was allocated in the AMS and ether extract and $aNDF_{OM}$ were not different among treatments. Although the digestibility responses observed in the present study are inconsistent with that of Menajovsky et al. (2018) and grazing applications (Bargo et al., 2003), those studies generally imposed diets that increased the total energy density thereby explaining the improvement in digestibility. Previous research has emphasized that the consumption of large amounts of concentrate in the AMS consistently influences the feeding behaviour and feed intake of the PMR (Hare et al., 2018; Menajovsky et al., 2018) and may also influence the rate and extent of NDF digestibility (Miron et al., 2004; Halachmi et al., 2005). While I cannot attribute the reduction in DM, $aNDF_{OM}$, and ADF digestibility to low ruminal pH, I speculate that the component feeding strategy imposed in AMS and the resultant changes in feeding behaviour may impact ruminal residence time and hence digestibility. To date, factors affecting the digestibility of nutrients for cows milked with AMS are relatively unexplored and further investigation is necessary.

Last, I observed that altering AMS concentrate allocation did not affect AMS attendance, milk or milk component yield, or lying behaviour. Given the diets were equivalent in nutrient supply, it is not surprising that milk and milk component yields were not different. These results parallel previous studies in both free-flow (Bach et al., 2007) and guided-flow barn designs (Hare et al., 2018; Menajovsky et al., 2018). In a retrospective study, Tremblay et al. (2016) indicated that feeding larger quantities of concentrate in the AMS was associated with reduced milk yield. Thus, the data in the present study are consistent with other published findings to support that greater concentrate allocations in the AMS do not positively influence milk production parameters; however, this contradicts current AMS feeding management recommendations (Salfer and Endres, 2014).

In addition, I observed no effect of AMS concentrate allocation on the yield of milk fat and protein. These findings are consistent to that of Menajovsky et al. (2018) and Bach et al. (2007). However, I did observe changes in milk composition with milk fat concentration being greatest when fed 3.5 kg of concentrate in the AMS and greatest milk protein when fed 2.0 kg of concentrate. Considering that component yield was not affected, these changes are difficult to explain. Hare et al. (2018) reported no differences in either milk fat, protein, or total solid concentrations when the total diet nutrient composition did not differ. In contrast, Menajovsky et al. (2018) reported greater milk fat concentration and a tendency for greater milk protein concentration when 2.0 kg of concentrate was provided in the AMS relative to 6.0 kg of concentrate in the AMS without a concurrent change in energy density of the PMR. Moreover, in the present experiment, MUN was reduced as the concentrate allocated in the AMS increased, corresponding to the linear reduction in ruminal ammonia concentration. Menajovsky et al. (2018) also demonstrated reduced MUN when 6.0 kg of concentrate was provided in the AMS compared to 2.0 kg. Like the treatment responses associated with total SCFA and rumen ammonia concentration, the variability of AMS concentrate intake among days in addition to the sorting behaviour associated with the PMR likely help explain these responses.

4.0 GENERAL DISCUSSION

4.1 Novel Research Approach and Results

The broad objective of this research was to contribute data that will enhance the understanding of feeding management of cows milked with AMS. This objective was accomplished by evaluating how the quantity of concentrate provided in the AMS affects DMI of the PMR, voluntary attendance to the AMS, ruminal fermentation, total tract digestibility, and milk and milk component yield. To achieve these objectives, 0.5, 2.0, 3.5, and 5.0 kg of a pelleted concentrate was supplied in the AMS. All treatment diets contained the same energy density and the pellet provided in the AMS was the same to that of the PMR. Thus, as the AMS concentrate allocation increased with treatment, a corresponding reduction of the same pellet in the PMR occurred. To address the objectives, Imeasured the amount of concentrate delivered to each cow, PMR intake, ruminal fermentation, and total tract digestibility. Measuring both AMS concentrate and PMR intake has only been reported in 3 published papers (Bach et al., 2007; Hare et al., 2018; Menajovsky et al., 2018). Only 1 previous study examined ruminal fermentation and total tract digestibility (Menajovsky et al., 2018) for cows in AMS. As such, the experimental approach of this study provides novel findings that can be used to refine feeding management strategies for cows milked with AMS.

4.1.1. Increasing concentrate delivery in the AMS increased the day-to-day variability in AMS concentrate intake

While the diets in the present study were formulated to be isocaloric, as the quantity of concentrate provided in the AMS increased, the variability in AMS concentrate intake also increased. To our knowledge, only 1 other study has reported such a finding. Menajovsky et al. (2018) reported a deviation among days of 0.25 and 0.85 when cows were provided 2.0 and 6.0 kg of concentrate in the AMS, respectively. Similarly, the current study demonstrated a deviation across days of 0.06, 0.24, 0.51, and 0.85 when 0.5, 2.0, 3.5, and 5.0 kg of concentrate were provided in the AMS, respectively. Precision feeding attempts to meet nutrient requirements on an individual cow basis through the AMS concentrate provision (Bach and Cabrera, 2017). Thus, some cows may be receiving more concentrate in the AMS when compared to other cows, depending on production or DIM in later lactation. Under precision feeding conditions, attempting to achieve target quantities of concentrate in the AMS will prove to be increasingly more difficult with cows receiving greater quantities of concentrate. The increased variability in day-to-day concentrate intake with increasing AMS concentrate allocation poses a challenge for using precision feeding strategies in AMS, making it problematic for nutritionists and producers to accurately understand total nutrient consumption. However, it should be acknowledged that precision feeding may involve more than simply increasing or decreasing the quantity of pellet offered in the AMS as in some cases, more than 1 pellet can be fed (Bach and Cabrera, 2017).

Although voluntary visits and milk and milk component yields were not affected by treatment, the greater variability in AMS concentrate intake among days for cows offered a greater quantity of AMS concentrate is speculated to have influenced the ruminal ammonia and MUN concentrations as both measurements linearly decreased with increasing concentrate. There is a strong correlation between blood urea N and MUN with dietary N intake (Nousiainen et al., 2004, Preston et al., 1965). Although blood urea N was not measured in the present study, lower concentrations of ruminal ammonia are generally correlated to lower concentrations of MUN. Further, energy intake and carbohydrate availability in the rumen are contributing factors to microbial protein yield (Rohr, 1986; Lebzien et al., 1993) and are used in the rumen in combination with ammonia produced from protein digestion (Kohn, 2007). Hristov et al. (2005), determined that as carbohydrates entering the rumen increase, ruminal ammonia is reduced through the inhibition of ammonia production and through the increase in uptake of ammonia for microbial protein synthesis. Similarly, Hristov and Ropp (2003), determined that when more ruminally available starch and sugars are available, ammonia capture by ruminal bacteria is enhanced. In the present study, meal size did not demonstrate to be an influencing factor of minimum, mean, or maximum ruminal pH, however, increasing the meal size of the concentrate in the AMS reduced both ruminal ammonia and MUN.

4.1.2. Increasing the AMS concentrate causes a reduction in PMR intake and alters PMR feeding behaviour

The quantity of concentrate provided in the AMS influenced the PMR intake and PMR feeding behaviour. As could be predicted, when the quantity of concentrate provided in the AMS linearly increased, intake of PMR was linearly reduced, creating a substitution effect. In this study, the combined effect of increased AMS concentrate and decreased PMR intake did not affect total DMI. However, substitution ratios have been inconsistent across studies. For example, for every 1 kg DM increase in AMS concentrate, PMR intake has been found to decrease by 1.14 and 1.58 kg DM in Bach et al. (2007) and Hare et al. (2018), respectively. Menajovsky et al. (2018) had a substitution rate of 0.89 and 0.78 kg DM for the high forage and low forage dietary treatments, respectively. This compares to the 0.97 kg reduction in PMR DMI in the present study for every 1 kg DM increase in AMS concentrate provided. Therefore, as identified in Table 4.1, it is evident that the substitution ratio is highly variable across studies.

Substitution ratios are not commonly reported when evaluating TMR or PMR feeding systems. However, substitution effects are reported for grazing applications for both beef and dairy cattle (Fieser and Vanzant, 2004; Horn and McCollum, 1987; Chase and Hibberd, 1987; Martin and Hibberd, 1990; Hess et al., 1996; Galloway et al., 1993; Kellaway and Porta, 1993; Depies, 1994; Reis and Combs, 2000). It has been noted that supplement type may influence the substitution ratio (Galloway et al., 1993); however, other studies have found little differences when evaluating the type of supplement (Fieser and Vanzant, 2004) even when comparing starch and digestible fibre-based supplements (Garces-Yepez et al., 1997; Elizalde et al., 1998). Generally, when supplements are provided to dairy cows on pasture, their DMI of pasture is reduced (Kellaway and Porta, 1993). This may be explained by the variation in milk response (kg milk/kg supplement) and thus when the milk response is low, the substitution rate is high (Bargo et al., 2003). Further, voluntary intake can be considered an indicator of overall forage quality (Reid, 1961). The quality of the forages has been an identified factor influencing the substitution ratio of forages for supplement (Fieser and Vanzant, 2004). Generally, larger substitution ratios are noted with higher quality forages (Fieser and Vanzant, 2004; Horn and McCollum, 1987). It was hypothesized by Fieser and Vanzant (2004), that the relationship between a greater substitution ratio and higher forage quality may be related to the bodies energy-satiety intake control mechanisms. That said, more research is necessary to predict substitution ratios and associative effects to estimate performance (Fieser and Vanzant, 2004).

In addition to nutritional factors, physiological factors such as stage of lactation, parity, and energy demand may have an influence on this substitution ratio. However, the mechanisms for the substitution ratio have not been elucidated. A predicted explanation for the extent of substitution may be causative of the hepatic oxidation theory (**HOT**) during early lactation, gut

Table 4.1. Experimental considerations of studies considering both AMS and PMR for substitution ratio determination.

Study	DIM (Average ± SD)	Parity	Traffic Design	Dietary Strategy	Substitution Ratio (kg DM)
Bach et al., 2007	191 ± 2.13	69 Primiparous 46 Multiparous	Free-flow	Isocaloric	1.14
Hare et al., 2018	227 ± 25 123 ± 71	5 Multiparous 3 Primiparous	Guided-flow	Isocaloric	1.58
Menajovsky et al., 2018	141 ± 13.6	Multiparous	Guided-flow	Low Forage High Forage	0.89 0.78
Present study	90.6 ± 9.8	Primiparous	Guided-flow	Isocaloric	0.97

fill during mid-lactation and a drop in DMI and energy corrected milk (**ECM**) during late lactation. Cows earlier in lactation, have higher levels of circulating NEFA from adipose tissue storage and higher propionate produced in the rumen due to the transition to higher energy-based diets (Allen et al., 2009). In the liver, propionate is an intermediate for the tricarboxylic acid cycle (**TCA** cycle) and accelerates the complete oxidation of the acetyl-CoA pool (Allen et al., 2009). Resultantly, the hepatic energy status is increased, the firing of the hepatic vagal afferent nerve is decreased; thereby, enhancing the satiety signals to the hypothalamus that cause a cessation of feed intake. Thus, the substitution rate during early lactation can be expected to be greater (> 1), where as during mid lactation, the substitution may be less severe (< 1) and less associated to HOT and more towards gut fill. Distension caused by physical fill (volume and weight of rumen contents; Schettini et al., 1999) of the rumen stimulates stretch receptors in the reticulo-rumen wall, which signal the satiety centres in the brain, ending meals (Grovum, 1987). The influence of gut fill is also related to an increase in total DMI lagging milk production. During later lactation, cows are maintaining a positive energy balance and thus DMI decreases along with production, however, the substitution rate is expected to increase (>1) because of the reduction in DMI and ECM. Table 4.1 demonstrates previous studies that have analyzed AMS concentrate and PMR intake to determine substitution affect. Being able to predict the substitution ratio is necessary to implement precision feeding strategies with AMS.

Sorting, meal duration, and consumption rate of the PMR were influenced by the concentrate allocation in the AMS. While the specific changes in the eating behaviours are not entirely consistent with previous studies examining these parameters in AMS (Hare et al., 2018, Menajovsky et al., 2018), the studies collectively demonstrate the importance of understanding the effect of the AMS allocation on PMR intake and PMR feeding behaviour. In the current study, the duration of PMR meals linearly increased, while the consumption rate of PMR linearly decreased as the AMS concentrate allocation increased. The sorting index of particles retained on the top sieve (> 19 mm) linearly decreased while the sorting index of particles retained on the bottom pan (< 4 mm), increased as the quantity of concentrate provided in the AMS increased. Alternately, Hare et al. (2018) reported a greater time spent eating, larger meal sizes and faster consumption rate of the PMR when cows were offered 0.5 kg of concentrate in the AMS, when compared to 5.0 kg of concentrate in the AMS. Like the current study, Hare et al. (2018) also reported that when 5.0 kg of concentrate is provided in the AMS, cows are more likely to sort against particles retained on the bottom pan of a PSPS (< 4 mm) in the PMR. Another study reported that when cows were provided 2.0 kg of concentrate in the AMS, they spend more time eating PMR per day than cows provided 6.0 kg of concentrate in the AMS. This study also determined that when cows were provided 6.0 kg of concentrate in the AMS, they sorted more against particles retained on the 8-mm sieve and sorted more for particles retained on the 4 mm sieve. The results of all 3 of these studies demonstrate that the quantity of concentrate provided in the AMS influence PMR feeding behaviours. Implications of these results may provide challenges with precision feeding strategies which attempt to meet nutrient requirements on an individual-cow basis based on production or even DIM. In addition, estimation of consumed nutrients is difficult to represent when cows are fed PMR in group housing situations because cows provided different AMS concentrate allocations will consume their PMR in a different manner.

4.1.3. Increasing the AMS concentrate allocation, while maintaining isocaloric diets, does not affect voluntary attendance or milk and milk component yield.

The results of this study indicate that that decreasing the concentrate allocation in the AMS does not affect attendance to the AMS, milk production, and milk components yield. This finding is consistent with other research experiments under isocaloric conditions (Bach et al., 2007, Hare et al., 2018) and those of increasing energy density (Halachmi et al., 2005, Menajovsky et al., 2018) and may prove to have a large economic impact for producers. Although the present study utilized the same pellet formulation for both the AMS and PMR pellets, producers are often sold on the concept of a high-quality and palatable pellet that may also include added flavours (Bach and Cabrera, 2017). Industry costs of AMS pellet may vary from $270 to $595/mT , depending on composition, however most commonly average between $375 and $450 /mT. As such, the AMS pellet is often costlier for producers than the PMR supplement and thus with lower AMS concentrate allocations, producers may be able to reduce their feed cost without affecting milk yield revenue.

While not investigated in the present study, feeding modest quantities of concentrate in the AMS may also create some opportunity regarding ingredients used in the pellet or rather the form of the supplement provided. It is currently recommended to feed a pelleted concentrate in the AMS because it is preferred by cows (Spördingly and Asberg, 2006). In addition, the consumption rate of pelleted forms of concentrate are between 250 and 400 g/min (Kertz et al.,

1981) are generally greater than that of ground barley (128 and 143 g/min; Spördingly and Asberg, 2006) or ground alfalfa (66 g/min; Spördingly and Asberg, 2006). The faster rate of consumption of pellet is often desired because of the high levels of concentrates offered in the AMS (> 5 kg/d) in the short duration they are in the milking stall (7 min/milking; Castro et al., 2012). However, when small or modest quantities of concentrate are offered in the AMS, a pelleted form may not be necessary. Therefore, the optimal form of concentrate with lower AMS concentrate allocations is still undetermined and requires further investigation and is out of scope of the research within this thesis.

4.1.4 The PMR rather than the quantity of concentrate offered in the AMS may have a greater influence on ruminal pH.

A more thorough understanding of the day-to-day variation in ruminal pH of cows milked with AMS is necessary for health and nutritional implications. It is commonly assumed that the greater quantities of concentrate provided in the AMS at a given visit will decrease ruminal pH more severely than when lower quantities of concentrate are provided (GEA Farm Technologies, 2014). However, ruminal pH measurements have only been collected from one study previous to the current study (Menajovsky et al., 2018). The assumption of greater quantities of concentrate provided in the AMS reducing ruminal pH are derived from research outcomes of supplying a lower F:C in the TMR or PMR or providing larger concentrate meals in automatic feeders of component fed cows. These assumptions may not prove to be accurate in AMS situations. Limitations with the AMS including the maximum meal sizes per AMS visit and time since last milking may prevent ruminal pH from being significantly reduced. Maximum meal sizes are usually between 2.0 and 2.8 kg/milking (GEA Farm Technologies, 2014; DeLaval Inc., 2018, Lely, n.d.). The allocation of concentrate to be dispensed each visit is dependant on the inter-milking interval and the carry-over from the previous day (explained in section 2.5). Although the current study observed no differences in daily minimum, mean, or maximum pH, a linear decrease in the variation in mean pH across days as the quantity of concentrate provided in the AMS increased was detected. A hypothesis to explain this observation may be due to the PMR having a greater influence on ruminal pH than that of the concentrate consumed in the AMS. When cows were provided lower quantities of concentrate in the AMS, the composition of PMR contained more concentrate in the PMR. The decreased F:C in the PMR may contribute to increased variation in daily mean ruminal pH. That said, large variation in ruminal pH among and within days were observed (Figure 3.1 and Figure 3.2) across all treatments. In addition, when greater quantities of concentrate were offered in the AMS, cows consumed PMR meals slower which may provide a greater buffering affect. Further, cows allocated greater quantities of concentrate in the AMS may not consume the full allocated amount. Maximum meal sizes programmed in the AMS may also contribute to the lack of variation in mean ruminal pH for cows fed larger quantities of concentrate. Therefore, additional research is necessary to fully understand the factors influencing ruminal pH of cows milked with AMS.

4.2 Inherent Limitations with AMS

4.2.1 The AMS does not weigh the quantity of concentrate provided and the amount of concentrate offered may not be equal to that consumed.

The AMS determines the quantity of concentrate delivered based on the number of auger rotations. Calibrations of the auger allow for accurate determination of concentrate weight for the revolutions. Factors that may influence the quantity of concentrate that is delivered in the AMS may be the density, DM, and physical form or consistency of the concentrate. Seldomly, research facilities have a concentrate refusal removal system and weigh scale engineered into the AMS feeder to remove any feed left behind by cows to accurately determine consumption of the concentrate (Figure 2.1; Bach and Cabrera, 2017).

While the AMS records the quantity of concentrate delivered, it is currently assumed to be equal to that consumed. In the current study, this assumption was also imposed as there was no mechanism to allow for measurement of AMS concentrate refusal. As a contingency approach, a camera was used to attempt to record whether cows were consuming their allocated meal; however, challenges determining a qualitative refusal arose when the AMS feeder was not completely empty prior to a new cow entering the AMS. Despite the inability to measure concentrate refusals and to determine whether treatment affected the quantity of pellet refused, it is believed that the assumption that delivered concentrate equals consumed concentrate is accurate. The latter assumption is supported with the nearly 1 to 1 substitution rate of PMR intake for AMS concentrate observed. Moreover, there were no production responses detected suggesting that energy intake was similar among treatments. Collectively the equal reduction in PMR intake with increased AMS concentrate intake and lack of production responses can be used as supportive information to suggest that cows consumed their AMS concentrate allocation.

4.2.2. Cows do not consume the quantity of AMS concentrate programmed in the computer.

The target quantity of concentrate programmed into the computer software to be delivered in the AMS, does not equal the quantity of concentrate offered in the AMS, rather the programmed amount indicates the maximal quantity that can be provided without considering carry-over effects. The quantity of concentrate offered in the AMS at any given visit is determined by the inter-milking interval and any carry-over concentrate from the previous day. For example, if a cow visits the AMS less frequently than the previous 7-d average, she will receive a greater allocation in the AMS the following day, due to the carry-over of concentrate that should have been allocated the previous day. Specific to DeLaval Inc. (2018), when the daily target concentrate allocation to be delivered in the AMS is 3.0 kg, 0.125 kg of concentrate will be saved per hour elapsed since the previous milking. Thus after 6 h, 0.750 kg of concentrate will be available and after 12 h, 1.5 kg will be available. With a 7-d milking frequency averaging 3.0 visits/d, 1.0 kg of concentrate will be provided in the AMS per visit. With an irregular milking frequency of 2.0 visits/d, 1.0 kg of concentrate will be carried over to the next day. Thus, 4.0 kg of concentrate will be available and will be divided among AMS visits, based on the time since previous milking. As the inter-milking interval increases, a greater quantity of concentrate will be saved. As such, it can be recognized that as the quantity of concentrate provided in the AMS increases, the quantity of refused feed in the AMS may also increase (Bach and Cabrera, 2017). Despite this limitation, commercial facilities, including the system used in the present study, do not have the ability to weigh the AMS concentrate added or refused.

As previously mentioned, as the quantity of concentrate programmed into the computer software is increased, the variability in concentrate delivery in the AMS increased among days, making it more challenging to accurately determine the amount of consumed concentrate in the AMS. Generally, greater quantities of concentrate must be programmed into the computer software to achieve the targeted amount (Hare et al., 2018; Menajovsky et al., 2018); however, in the present study only a slightly greater quantity (0.51, 2.02, 3.52, and 5.03 kg DM) of concentrate had to be programmed in the AMS to achieve 0.50, 2.00, 3.49 and 4.93 kg DM. Hare et al. (2018) programmed 0.54 and 5.20 kg DM to achieve 0.50 and 5.00 kg DM in the AMS; while Menajovsky et al. (2018) programmed 2.07 and 6.55kg DM to achieve 2.04 and 6.18 kg

DM in the AMS. The DM of the concentrate and possibly the time of year (season) may influence the computer programmed quantity relative to the quantity offered. Hare et al. (2018) and Menajovsky et al. (2018) were both conducted during the summer months, were as the current study took place in the winter. Cows have greater nutrient requirements in the winter (NRC, 2001) for maintenance energy. In addition, it has been determined that cows in hot climates have a decreased DMI, decreased milk yield (West et al., 2003; West, 2003; Bava et al., 2012) are less physically active (West, 2003) and spend less time lying (Bava et al., 2012). During the winter, more gate-passing events may be observed (Clark et al., 2014) due to an increase in activity level from the lack of heat stress present. Based on this information, it may be hypothesized that during the present study lower levels were programmed into the computer software when compared to previous studies (Hare et al., 2018; Menajovsky et al., 2018) to achieve the targeted amount because cows were more likely to consume the actual offered amount, suggesting cows may be visiting the AMS slightly more frequently during the winter. Further investigation is necessary to fully understand seasonal implications with AMS feeding and management considerations with precision feeding strategies. Therefore, from a nutritional standpoint the ability to predict concentrate intake and formulate rations considering both the AMS concentrate and the PMR are difficult when large quantities of concentrate are targeted in the AMS.

4.2.3. Conventional management imposes a 24-d, but a 24-h interval may not represent the biological pattern for cows in an AMS.

Moreover, data management with AMS utilize a traditional 24-h representation when determining milk yield, milk component yield, and feed intake (AMS concentrate and PMR). In the current study, ruminal fluid and fecal samples were collected in attempt to represent this 24-h cycle. In addition, milk samples were composited by day assuming a 24-h duration. These procedures were conducted with the notion that cows in an AMS follow a 24-h cycle similar to cows managed in conventional milking systems and because the AMS concentrate allocation re-sets at midnight each day (DelPro 4.5, Delaval Inc., Tetra Laval Group, Tumba, Sodermanland, Sweden). Despite this, intervals between AMS visits, timing and duration of the PMR meals, and ruminal pH were highly variable among cows and within a cow across days (Figure 3.1 and Figure 3.2).

Additionally, the composition of milk at each milking is dependant on inter-milking interval and the milk yield per milking event (Friggens and Rasmussen, 2001). It could also be speculated that milk composition may also depend on feeding times and meal size throughout the day (Sahana et al., 2008). Friggens and Rasmussen (2001) determined that though single milk sampling may prove to moderately represent milk protein, lactose, and urea content, single milk sampling inaccurately represents milk fat and SCC content. In another study, it was observed that milk fat content varied significantly depending on milk yield and inter-milking interval; while, milk fatty acid composition did not vary within individual cows between milking events (Larsen et al., 2012). The methodology in the present study of compositing individual milk samples from each milking overcomes the limitations associated with analysis of individual samples, but variability in daily patterns still may have influenced the results. Further research is necessary to evaluate the relationship between PMR consumption, milk yield, and inter-milking interval to determine if a 24-h day is an accurate representation for cows milked with AMS. Alternatively, a mechanism to determine the duration of the natural biological patterns for each cow may have utility in evaluating management strategies with AMS.

4.2.4. Data management in AMS.

There is a large quantity of data recorded by the AMS software (Jacobs and Siegford, 2012). Some of these data include milk yield per visit, daily milk yields, milk conductivity by quarter, presence of blood by quarter, incomplete milking events, milking kick-off events, concentrate consumption by visit and by day, and cow activity monitoring data. Traditionally farmers detect estrous, mastitis, and other health- and management-related issues through involvement in the milking procedure and frequent interactions with cows; however, relying on the AMS data reports to understand issues in the herd may be challenging for producers with limited experience with computers and data analysis. Thus, data recorded by the AMS can easily be misinterpreted (Jacobs and Siegford, 2012) if not appropriately summarized and presented. Proper organization and an understanding for how the information is collected is important for the appropriate interpretation. Default and custom report summaries including individual cow or herd-based data for the specified information can be collected to assist with the interpretation of the collected AMS data. Specifically, to the DelPro 4.5 (DeLaval) computer software program, the monitor board contains a glimpse of average AMS milkings/cow/d, concentrate intakes (kg/cow/d), incomplete milkings (%), milk production (kg/cow/d) among other information. As noted in the current experiment, cows milked with AMS have a highly variable milking pattern among days, within cows and among cows. The information represented in the monitor board should be considered with caution because it is a representation of the previous 24 h and not necessarily the last day (0000 to 2400). Fluctuations in the numbers represented may occur throughout the day depending on which cows have milked and when.

Further, changes in udder conformation may occur throughout a lactation or among lactations (Jacobs and Siegford, 2012). Frequent training of new cows on the AMS and re-training of cows must be done to optimize performance of the AMS (Jacobs and Siegford, 2012). Genetic selection for proper teat placement is important for the efficiency and production of the AMS and generally cull rates will increase if these parameters are not considered (Rodenburg, 2002). In addition, frequent trimming of tail and udder hairs are necessary for efficient milking procedures with AMS (Salfer and Endres, 2018).

While the number of AMS in Canada is rapidly growing (de Koning, 2010), the ability to adapt to increasing quota allocation and to expand may be challenged with AMS. It has been reported that the cost of herd expansion for herds with AMS must include the price of additional AMS unit(s) with estimates ranging from $150,000 to $200,000 US per unit (Jacobs and Seigford, 2012). This cost is in addition to that required for quota, other infrastructure (building costs), additional cows, and related nutrition and veterinary costs. For AMS producers to expand their herds, the expansion may be needed in increments of 60 cows because it is estimated that 1 AMS can milk 60 cows (or 80 to 90 kg of fat in Canada). Thus, the cost of expansion is large and, in many cases, impractical for some herds as economic drivers for AMS require it to occur in AMS units. Therefore, special consideration in management practices should be considered with AMS herd.

4.3 Industry Applications

Current AMS feeding recommendations suggest that feeding greater quantities of concentrate in the AMS will increase the motivation of cows to voluntarily enter the individual milking stall, with the prediction that the improved AMS attendance will increase milk production and reduce fetching on farm (Rodenburg, 2011). Despite these recommendations, scientific experimentation including the present study, suggest that greater quantities of concentrate are not required in the AMS to maintain AMS visits or milk and milk component yields (Bach et al., 2007; Halachmi et al., 2005; Hare et al., 2018; Menajovsky et al., 2018). In addition to the recommendations mentioned above, it is the belief of producers that greater quantities of concentrate should be offered to cows in the AMS in facilities with a free-flow traffic design, when compared to guided-flow systems (Endres and Salfer, 2016). Although the exact quantities of concentrate in free-flow and guided-flow traffic designs are unsettled at this time, it is likely that they may be farm specific, rather than dependant on traffic design. Bach et al. (2008), prepared and fed a common TMR and delivered it to 47 dairy herds once daily. All farms utilized in this study were located within a 50-km radius of each other and had similar genetic profiles. This study used survey-based data (owner profiles, management practices, facility design information, and animal information), to analyze farm performance over an 8-month period Results of this study demonstrate that despite the similarities between farms and common TMR, average milk production varied between 20.6 and 33.8 kg/d/cow. Non-dietary factors including age at first calving, level of feed refusals, stocking density, and feed push ups accounted for 50% of the variation in milk production. Moreover, many industry recommendations are derived from studies with no control populations or from survey-based data. Bach et al. (2008), demonstrates high variations and the uncertainties among farms therefore survey-based recommendations should be considered with caution.

Regardless of the traffic flow design imposed in the current research (feed-first guided-traffic design), several concepts from the current study are applicable to all cow-traffic flow designs. First, the reduction in PMR intake with increasing AMS concentrate allocation and increased AMS concentrate intake variability among days with increasing AMS concentrate are both applicable and likely independent of cow-traffic design. Both the unpredictable reduction in PMR intake and the increased variability in AMS concentrate intake with increasing quantity of AMS concentrate provided challenge the ability to impose precision feeding strategies. Precision feeding strategies with the AMS attempt to meet individual cow nutrient requirements through the concentrate allocations in the AMS (Bach and Cabrera, 2017). In addition, the lack of milk and milk component yield responses in free-flow (Bach et al., 2007), and guided-flow traffic designs (current study) suggest that there may be less of a difference in feeding strategies for the different cow traffic designs. Further research is necessary to fully understand if precision strategies can be implemented into AMS feeding management to improve nutrient delivery and production. Therefore, revisions of recommendations should be considered to correspond to applications of the current research regarding quantity of concentrate provided in the AMS.

5.0 CONCLUSIONS

For cows in a feed-first guided-traffic flow design, increasing the quantity of concentrate in the AMS, while maintaining total dietary nutrient density by altering the PMR, does not affect voluntary attendance to the AMS, milk yield, and milk component yield. Feeding greater quantities of concentrate in the AMS increases the day-to-day variation in AMS concentrate intake thereby reducing the ability to implement precision feeding strategies. Increasing the quantity of concentrate offered in the AMS, and consequently decreasing the concentrate in the PMR, also affects PMR sorting and feeding behaviour and may reduce total tract DM, $aNDF_{OM}$, and ADF digestibility. Therefore, feeding smaller quantities (< 5 kg) of concentrate in the AMS should minimize variability in nutrient intake, allow cows to achieve target AMS consumption, while not affecting AMS visits and the yield of milk and milk components.

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