CONTROLLING ENERGY INTAKE IN THE PREPARTUM PERIOD TO IMPROVE TRANSITION COW HEALTH

by

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ABSTRACT

A common feeding practice during the dry period is to switch dairy cows to a low forage, energy dense diet 3 weeks prepartum, but this practice has been criticized as it may lead to the overconsumption of energy and increase the risk of metabolic disease postpartum. The aim of this trial was to compare the metabolic status of transition Holstein dairy cows fed a 77% forage diet (77F) (NE_L = 1.46 Mcal/kg; NDF = 41%) versus those fed a 87% forage diet (87F) (NE_L = 1.41 Mcal/kg; 48% NDF). Approximately 60 days before calving cows were dried off and fed the 87F diet. Three weeks before expected calving cows were randomly assigned to either remain on the 87F diet (n=42) or switched to the 77F diet (n=45). After calving, all cows were fed a common lactation diet (NE_L=1.59 Mcal/kg). Dry matter intake (DMI) was measured daily from 2 weeks before to 2 weeks after calving. Blood was sampled twice-weekly prepartum and daily for 10 days postpartum. Subclinical ketosis (SCK) was diagnosed using a threshold of BHBA $\geq$ 1.0 mmol/L after calving. Metritis was determined by examining vaginal discharge. Cows on the 87F diet had lower DMI prepartum than those on the 77F diet (12.7 kg/d ± 0.3 vs. 15.4 ± 0.3, *P* <0.0001), but no difference was detected after calving (19.7 ± 5.5 kg/d; *P*=0.64). Although the calculated prepartum required energy intake was the same between treatments (15.3 ± 1.2; P=0.16), and both groups consumed in excess to these requirements, cows on the 77F diet consumed 5.1 Mcal/d more than those on the 87F diet. Postpartum BHBA levels were lower for cows fed the 87F diet prepartum (0.49 mmol/L ± 0.03 vs. 0.58 ± 0.03; P=0.003), and fewer animals on this diet were diagnosed with SCK (48.9% vs. 16.7%; *P*=0.001). We also noted a tendency for fewer cows to be diagnosed with metritis on the 87F diet (28.9% vs. 14.3%; *P*=0.1). There was no difference in daily milk production between treatments. These results indicate that feeding an 87F diet before calving can improve health in transition dairy cows.

PREFACE

A version of Chapter 2 has been submitted for publication and is currently under review: L.A. Vickers, D.M. Weary, D.M. Veira and M.A.G. von Keyserlingk. Feeding a high forage diet prepartum improves health in transition dairy cows. Lori Vickers and Drs. Dan Weary, Doug Veira and Marina von Keyerlingk designed the study collaboratively. Lori Vickers executed the research trial and collected all data. Lori Vickers was primarily responsible for data analysis and preparation of the manuscript under the supervision of the co-authors. Co-authors edited drafts.

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ADF = acid detergent fiber

BCS = body condition score

BHBA = beta hydroxybutyrate

CP = crude protein

DA = displaced abomasum

DIM = days in milk

DM = dry matter

DMI = dry matter intake

NDF = neutral detergent fiber

NEB = negative energy balance

$\mathbf{NE_I}$ = net energy intake

$\mathbf{NE_L}$ = net energy of lactation

$\mathbf{NE_M}$ = net energy of maintenance

$\mathbf{NE_P}$ = net energy of pregnancy

$\mathbf{NE_{PRE}}$ = prepartum net energy

$\mathbf{NE_{POST}}$ = postpartum net energy

NEFA = non-esterified fatty acid

NRC = National Research Council

RP = retained placenta

SARA = subacute ruminal acidosis

SCC = somatic cell count

SCK = subclinical ketosis

TG = triglyceride

TMR = total mixed ration

VD = vaginal discharge

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CHAPTER 1: INTRODUCTION

THE TRANSITION PERIOD

On a typical dairy farm in Canada a dairy cow has a calf once a year. The calf grows up, gets bred at about 14 months of age and has her first calf when she is 2 years old. Approximately 2-3 months after she calves and starts lactating, she is bred again; 2 months before she is due to have her next calf, her lactation ends (referred to as the 'dry period') and she prepares for her next lactation cycle. She has her calf and the cycle continues. In her 10 month lactation cycle, the average dairy cow produces 9,793 kg of milk (CDIC, 2011b).

The transition period, defined as 3 weeks before to 3 weeks after calving, is when the dairy cow transitions from a pregnant, non-lactating state to a non-pregnant, lactating state. During this time she is faced with numerous physiological challenges and stressors related to parturition and the onset of lactation in addition to diet changes and social regrouping. One of the main challenges is a sudden increase in nutrient requirements needed to support lactation at a time when dry matter intake (**DMI**) and nutrient supply begin to decline (Drackley, 1999). Late in pregnancy and early lactation, nutrient demand increases considerably (Ingvartsen, 2006), essentially doubling overnight once lactation starts (Drackley et al., 2005). In the week leading up to calving, DMI has been shown to decline, with a drop of approximately 30% occurring in the 24h before calving (Huzzey et al., 2007). DMI has been shown to drop by 19% on the day after calving, relative to the day of calving when cows are routinely switched to a high energy diet to support lactation needs (Huzzey et al., 2007). As a result, postpartum intakes are usually less than energy requirements (Bell, 1995); this combined with the increased energy demands

of lactation often lead the cow into a negative energy balance (**NEB**). In response to NEB, cows mobilize stored triglycerides in the adipose tissue in an attempt to meet energy demands for maintenance and milk production.

The risk of disease increases when the cow experiences an extended period of NEB (Goff, 2006), perhaps explaining the high prevalence of disease around transition. Gröhn et al. (1995) estimated a 7.4% lactation prevalence of retained placenta, a 7.6% prevalence of metritis and a 4.9% prevalence of ketosis. Jordan and Fourdraine (1993) surveyed 61 high producing dairy herds in the US and reported a 3.7% (range 0-20%) rate of ketosis, a 12.8% (range 9-66%) incidence of metritis and a 9.0% (range 0-22.6%) incidence of retained placenta in the postpartum period. Metabolic and infectious disease can result in lower milk yields (Rajala-Schultz et al., 1999a; Rajala-Schultz et al., 1999b), lower conception rates (LeBlanc et al., 2002) and increased incidences of involuntary culling (Gröhn et al., 1998).

Canada has 981,000 dairy cows, with 71,100 in British Columbia (CDIC, 2011a). On a typical dairy farm, cows calve once a year; therefore, each year, a large number of dairy cows in Canada are vulnerable to illness creating serious welfare concerns. Further, this period has frequently been shown to be of economic importance to dairy producers: cows that fail to transition successfully are generally less productive. For example, cows diagnosed with metritis in the 3 weeks following calving produced less milk in the first 20 weeks of lactation and were more likely to be culled from the herd (Wittrock et al., 2011). In addition, ketosis has been shown to have negative impacts on milk production with daily milk loss ranging from 3.0 – 5.3kg/d (Rajala-Schultz et al., 1999a). Therefore,

it is in the best interest of dairy farmers to reduce disease during the transition period for both their economic survival and for the welfare of their cows.

Nutrient Partitioning

In the first third of a cows lactation cycle, she is in a NEB energetically equivalent to approximately 9kg/d milk, and must mobilize body reserves to compensate (Bauman and Currie, 1980). In the final third of her lactation, as milk production beings to decline, she is generally fed in excess to her requirements, allowing her to replenish body stores through lipogenesis (Bauman and Currie, 1980). At this time, cows are able to build up body reserves in anticipation of the next lactation (Friggens, 2003). After eating, insulin is released from the pancreas and activates the enzyme acetyl CoA carboxylase that promotes fat storage (Drackley, 2000). In the adipose tissue, fatty acids are esterified to form triglycerides and stored; in the liver, fatty acids are esterified to phospholipids and used or incorporated into intracellular plasma membranes. In the mammary gland, lipogenesis produces long chain fatty acids which are rapidly incorporated into the milk (Drackley, 2000).

As parturition approaches and the dairy cow transitions into lactation, nutrient demand increases drastically. Requirements for glucose and metabolizable energy increase 2-3 fold from 3 weeks before to 3 weeks after calving (Drackley et al., 2001). Towards the end of gestation the fetus and mammary gland use a substantial amount of maternal nutrients; the dairy cow's ability to partition nutrients to these areas is essential for her survival and it is what allows for high milk production (Bauman and Currie, 1980). This physiological state provides a very high nutrient demand to the cow and

forces her to alter her metabolism in order to meet these high energy needs; if the cow is unable to redirect nutrients to the fetus and mammary gland efficiently, she is susceptible to metabolic disorders. Nature has given pregnancy and milk production such high biological priorities that they will continue at the expense of other metabolic processes even if a state of disease is created (Bauman and Currie, 1980).

In order to transition successfully, the cow must undergo several metabolic adaptations. She must increase hepatic gluconeogenesis (glucose generation from non-carbohydrate substances), decrease glucose usage by peripheral tissues, increase fatty acid production from adipose tissues and increase amino acid mobilization from muscle (Bell, 1995). These coordinated processes allow for increased nutrient usage by the mammary gland.

At the onset of lactation, the adipose tissue begins to mobilize body reserves through lipolysis (Bauman and Currie, 1980). Lipolysis is the mobilization of fatty acids from adipose tissue triglycerols that occurs in times of NEB or stress. Fatty acids are released from their glycerol backbone and increase the intracellular fatty acid pool; since there is no stimulus to re-esterify them, they diffuse into the blood. Free fatty acids then bind to serum albumin and circulate to various body tissues (Drackley, 2000). However, when high rates of lipolysis occur (such as in early lactation), the concentrations of blood albumin tend to be low (McNamara, 1991) causing free fatty acids to be taken up by body tissues (Drackley, 2000).

After calving, cows produce more milk than can be energetically produced from the amount of feed they are able to consume, forcing them to rely on body stores. A decrease in serum insulin and an increase in glucagon promotes oxidation of fatty acids

(Vernon, 2005). Triglycerides are mobilized to non-esterified fatty acids (**NEFA)** though the action of the enzyme lipase (Frandson et al., 2006) and either enter the mammary gland, leading to an increase in milk fat or are absorbed into the liver. Once in the liver, NEFA can either be 1) oxidized to provide energy in the liver, 2) converted to triglycerides, or 3) partially oxidized to ketone bodies and released back into the blood stream to be used for energy by other body tissues (Drackley, 1999; Grummer et al., 2004; Ingvartsen, 2006). In non-ruminant animals, NEFA conversion to ketone bodies serves as a glucose sparing strategy in time of deficits and similar adaptive processes may occur in transition dairy cows (Drackley, 1999). The heart, kidney, skeletal muscles, mammary glands and gastrointestinal tract can oxidize ketone bodies for energy (Heitmann et al., 1987); further, ketone bodies can be a substrate for fatty acid synthesis in the mammary gland suggesting ketogenesis is a strategy to compensate for insufficient glucose (Drackley, 1999) and a metabolic adaptation to hunger (Ingvartsen, 2006).

NUTRITION AND DISEASE

The highest rate of production disease occurs during the transition period (Mulligan and Doherty, 2008), with most metabolic and infectious diseases occurring within the first 2 weeks of lactation (Kehrli et al., 2006). In order to transition successfully into lactation, dairy cows must be able to make a range of metabolic adjustments, as discussed above, placing high demands on the liver (Kehrli et al., 2006). During the transition period, cows undergo dietary changes, environmental stressors (i.e. group changes) and immunosuppression; the interrelationship among production disease often leads to a cascade effect whereby metabolic diseases lead to increased incidence of

infectious disease, leading to decreased fertility and milk production (Mulligan and Doherty, 2008).

Metabolic Disease

Mobilizing fatty acids from adipose tissues to support lactation is a normal biological process for many species (Mulligan and Doherty, 2008). However, elevated ketone bodies are a sign of disturbed energy metabolism (Andersson, 1988) and increase when the liver has reached its maximum ability to oxidize NEFA (Walsh et al., 2007). If too severe, high NEFA concentrations lead to increased triglyceride accumulation in the liver cells, ultimately impairing liver function (Mulligan and Doherty, 2008). Moderate accumulation of triglyceride in the liver is tolerable, but when the ratio of stored lipids to glycogen exceeds 2:1, pathological problems may develop (Drackley et al., 2001). Liver function is impaired leading to decreased metabolic function of the liver, including decreased gluconeogenesis, variable effects on ketogenesis and β oxidation and increased rate of lipogenesis (Bobe et al., 2004). When cows are in NEB, they mobilize NEFA in excess of requirements (Bobe et al., 2004). Regardless of the concentration of NEFA circulating in the blood, the liver always takes up a constant proportion and oxidizes these in accordance with need for ATP; the rest are esterified to triglycerides creating a disposal sink (Drackley and Andersen, 2006) and a condition known as fatty liver. Fatty liver develops when lipid uptake by the liver is greater than the liver's ability to oxidize and secrete lipids leading to net storage of triglycerides (Grummer, 1993). To avoid fatty liver the energy status of the transition cow must be improved, decreasing NEFA mobilization from adipose tissues (Bobe et al., 2004).

Ruminants are not very efficient at oxidizing NEFA (Palmquist, 1994) favouring either oxidation to ketone bodies or storage to triglycerides. If excessive body fat is mobilized, NEFA that are released from body reserves essentially overwhelm the liver's ability to use them as fuel so a portion are converted to ketone bodies (Goff, 2006). Ketosis is a condition that occurs when there is high concentration of the ketone bodies acetoacetate, betahydroxybutyrate (**BHBA**) and acetone and low concentrations of glucose (Ingvartsen, 2006). The lack of glucose and insulin lead to increased fatty acid mobilization from adipose tissues, increasing hepatic ketogenesis (Kehrli et al., 2006). Eventually, blood glucose levels fall below what is needed to support nerve and brain function, leading to clinical signs of ketosis, including stumbling, head pressing and central nervous system function dysfunction (Goff, 2006).

Infectious Disease

Neutrophils are a type of white blood cell involved in the first line of defense against infection (Frandson et al., 2006). It has been reported that the function of neutrophils is impaired in transition dairy cows leading to a state of immunosuppression; neutrophil function declines prior to calving, reaches a nadir and return slowly by 4 weeks postpartum (Kehrli et al., 2006). Cows with normal uterine health show a slight reduction of the neutrophil ability to kill bacteria, while cows with puerperal metritis or subclinical endometritis exhibit marked declines in neutrophil ability to kill bacteria (Hammon et al., 2006). Further, elevated blood NEFA concentrations before calving have been linked with uterine disorders and impaired neutrophil function (Hammon et al.,

2006). Therefore, the metabolic demands of lactation can affect the cows ability to recover from immunosuppression (Kehrli et al., 2006).

NUTRITIONAL MANAGEMENT DURING THE TRANSITION PERIOD

History of Dry Cow Nutrition

The dry period has been characterized as a resting period between lactations as it is the period of lowest nutritional requirements for the dairy cow. In fact, this is not a rest period; rather, it is a time to prepare for the many physiological processes that set the stage for a successful lactation (Van Saun, 1991). Over the past 30 years, milk production has increased around 2% per year (Eastridge, 2006). Cows have been bred for high milk yields and within days of parturition they will produce significantly more milk than what is required to feed a calf (Grummer et al., 2004). Energy requirements needed to support lactation have increased more rapidly than DMI; animals have limits as to how much dry matter they can consume due to rumen fill or satiety, leading diets to be formulated with higher nutrient densities to ensure animals are getting required energy (Eastridge, 2006). Therefore, to improve transition success, it has been suggested that we must either decrease the nutrient demand on the cow or increase her nutrient intake (Grummer et al., 2004). As a result, net energy density of lactation diets has increased from 1.23-1.36Mcal/kg in 1980 to greater than 1.60 Mcal/kg in 2006 to support high producing cows (Eastridge, 2006).

Controlled Energy Diets

In the dry period, dairy cows are fed a high forage diet formulated to meet minimum energy requirements to maintain body condition. Three weeks before calving, they are switched to an energy dense, moderate forage diet formulated to nutritionally prepare them for the physiological demands of the late dry period and early lactation. This diet is designed to allow the rumen microbe population and papillae to adapt to higher energy diets that are fed postpartum and to increase her nutrient intake to meet lactation demands. However, despite the sound rationale for this diet, prevalence of production diseases remains high (Drackley and Janovick Guretzky, 2007).

There is growing evidence that restricting DMI in the dry period allows cows to increase DMI immediately postpartum, resulting in higher energy balances, and decreased body fat mobilization, evident by lower NEFA and BHBA concentrations (Dann et al., 2006). In contrast, cows fed nutrient dense prepartum diets ad libitum often greatly over consume energy and show greater body weight loss, increased mobilization of body tissues and longer periods of negative energy balance postpartum (Agenäs et al., 2003). Even though cows fed high-energy diets prepartum have higher DMI, energy intake and energy balance (Rabelo et al., 2003), they showed more drastic declines in DMI as parturition approached (Johnson and Otterby, 1981; Rabelo et al., 2003). However, when intakes were restricted to levels below minimum energy requirements for maintenance and lactation set out by the National Research Council (NRC, 2001), cows had greater NEFA and insulin concentrations, suggesting that high energy diets prepartum may lower liver triglyceride and NEFA concentrations that accumulate when cows are in NEB (VandeHaar et al., 1999).

Far off cows (i.e. cows at the start of the dry period) have been shown to over consume dry matter by as much as 60% of their daily requirements if fed ad libitum, indicating they do not regulate intake in response to energy requirements (Dann et al., 2006). Further, at the onset of lactation, cows allowed to over consume energy in the dry period show higher BHBA and NEFA levels, lower energy balance, higher total lipid and triglyceride levels at the onset of lactation, suggesting an increased risk for developing ketosis and other metabolic health issues (Dann et al., 2006). In addition, cows restricted to consuming energy slightly below NRC recommendations during the dry period show improved post calving appetites and lower body fat mobilisation, shown by lower plasma NEFA levels and less fat accumulation in the liver (Douglas et al., 2006). Further, these same cows showed an increase in NEFA concentrations 2 weeks before calving which peaked within the first week of lactation, fell and by 14 days in milk (**DIM)** were again comparable to pre-calving levels (Douglas et al., 2006). In contrast, cows allowed to over consume energy showed a delayed rise in NEFA before calving, with a comparable peak 2 weeks after calving, but these remained elevated for 7-8 weeks postpartum (Dann et al., 2006; Douglas et al., 2006). This result suggests that preventing excess intake during the far off period may improve energy status of the transition cow. In summary, it appears cows perform best when nutrient requirements are met without greatly exceeding or going under energy requirements.

Forage for Dairy Cows

Dairy cattle are ruminant animals with specialized digestive tracts, allowing them to break down forages and fibrous roughages. The rumen houses many bacteria, protozoa

and other microbes that produce cellulose capable of breaking down complex structures of plant cell walls (Frandson et al., 2006). Fermentation of high concentrate diets can cause a drop in rumen pH due to the increase in lactic acid production and in extreme cases can cause a detrimental shift in the microbial population of the rumen (Goff, 2006). A forage component is essential for a healthy functioning digestive tract of a dairy cow as forages stimulate chewing and rumination, which produce saliva that acts as a buffer against drops in pH and prevent digestive upsets (Baumont et al., 2006).

One way of achieving a controlled energy diet in the prepartum period is to dilute nutrients with the addition of high fibre forages with low energy contents. On their own low quality roughages (such as straw) are not enough to meet energy needs of a dairy cow. They are bulky feeds with low digestibility due to their high levels of cellulose and lignin, which tend to limit intake (Kellems and Church, 2001). However, they can be effectively added to a total mixed ration (**TMR**) to dilute rations in the prepartum period (Drackley and Janovick Guretzky, 2007; Kellems and Church, 2001).

OBJECTIVES

Recent work had shown the potential of low energy dry cow diets to improve transition success. Although there is no direct evidence to support claims that the high forage/low energy ration is successful in improving transition cow health and performance, there is anecdotal evidence from farmers that suggests cows on such diets have easier calving, better intakes, more consistent body condition at calving, improved general heard health and fewer incidences of metabolic disease (Beever, 2006). Currently, many cows experience decreased DMI during the transition period and fail to make a successful transition to the postpartum diet, which increases the extent of NEB,

loss of body condition and body weight loss seen around calving. A potential solution to the dry cow diet predicament was put forward by Drackley and Janvick Guretzky (2007) that involves formulating a bulky, high forage ration of low energy density (1.32- 1.41 Mcal NE_L/kg DM) that cows can consume free choice without greatly exceeding their daily energy requirement. Researchers to date have provided support for this feeding management system by showing lower BHBA and NEFA concentrations (Agenäs et al., 2003; Dann et al., 2006); however, it has not been determined whether these results translate into fewer cases of disease after calving. The objective of this study was to test the effects of feeding a higher forage diet throughout the dry period on postpartum health.

CHAPTER 2: FEEDING A HIGHER FORAGE DIET PREPARTUM IMPROVES HEALTH IN TRANSITION COWS[1]

INTRODUCTION

The dairy industry remains challenged with high rates of disease after calving, often attributed to prolonged periods of negative energy balance (**NEB**). In an effort to reduce NEB many producers feed an energy dense diet beginning approximately 3 week before calving, but this practice may lead to over consumption of energy and increase the risk of metabolic disease postpartum (Drackley and Janovick Guretzky, 2007). NEB and excessive lipid mobilization have been linked to postpartum disease, including increased risk of displaced abomasums (LeBlanc et al., 2006), fatty liver (Bobe et al., 2004), ketosis and subclinical ketosis (Drackley et al., 2001). Metabolic and infectious disease lower milk yields (Rajala-Schultz et al., 1999a), decrease conception rates (LeBlanc et al., 2002) and increase rates of involuntary culling (Gröhn et al., 1998), reducing the profitability of the farm and the welfare of the cow.

Previous work has shown a relation between prepartum and postpartum intakes, suggesting that increasing nutrient density of prepartum diets could improve DMI and reduce NEB postpartum (see review by Grummer, 1995). However, there is growing evidence that restricted feeding during the dry period may be advantageous (Agenäs et al., 2003; Douglas et al., 2006; Holcomb et al., 2001; Holtenius et al., 2003).

If given the opportunity, far off cows (~60 to 30 days before expected calving date) over consume energy by as much as 60% of their daily requirements (Dann et al.,

[1] A version of this chapter has been submitted for publication and is currently under review: L.A. Vickers, D.M. Weary, D.M. Veira and M.A.G. von Keyserlingk. Feeding a high forage diet prepartum improves health in transition dairy cows

2006). This excess energy is stored in the adipose tissue and broken down to NEFA in times of NEB; NEFA are used for energy and excess triglycerides (**TG**) are stored in the liver, potentially increasing the risk of fatty liver and health problems after calving (see review by Drackley, 1999). Cows in early lactation that consumed excess energy prepartum have higher BHBA and NEFA levels, greater NEB, and higher total liver lipid and TG levels postpartum, suggesting an increased risk of ketosis and other metabolic health issues (Dann et al., 2006). These results indicate that offering diets meeting rather than exceeding energy requirements prepartum will lower body fat mobilisation and improve health postpartum (Douglas et al., 2006).

Drackley and Janovick Guretzky (2007) presented a possible solution to the dry cow diet predicament: feeding higher forage diet with lower energy density (1.32-1.41 Mcal NE_L/kg DM) that can be consumed ad libitum without risk of excess energy intake. Recent work had shown benefits of feeding these diets in terms of improved metabolic profiles postpartum (Dann et al., 2006; Douglas et al., 2006; Holtenius et al., 2003). Anecdotal evidence from farmers claims improved health and faster return to oestrus (Beever, 2006), but no work has tested the effects of lower energy / higher forage diets on postpartum health. The objective of the current study was to test the effects of feeding a diet of 77% forage and a diet of 87% forage throughout the dry period on postpartum health.

MATERIALS AND METHODS

Animals Housing, and Diet

This experiment was conducted at the University of British Columbia's Dairy Education and Research Center (Agassiz, BC, Canada) from July 2009 – March 2010.

Cows were cared for according to the guidelines set by the Canadian Council on Animal Care (2009). During the 9-month study period, 91 multiparous Holstein cows were enrolled in the experiment with an average dry period of 55.9 ± 7.2 days (mean ± SD). Following dry off, all cows were housed in a free stall pen fitted with a post and rail feed barrier system and fed a diet with 87% forage (**87F**) (NE_L = 1.41 Mcal/kg; 48% NDF) described in Table 1 & 2. Approximately 3 week before expected parturition, cows were moved to the prepartum pen equipped with Insentec (Insentec, Markness, Holland) feed bins where they were randomly assigned to one of two dietary treatments. Half of the cows remained on the 87F diet until parturition (treatment group), while the other half (control group) were switched to a diet with 77% forage (**77F**) (NE_L = 1.46 Mcal/kg; 41% NDF; Table 1 and Table 2). Groups were balanced for BCS, parity and previous 305-d milk production. Care was taken to ensure groups were consistently balanced over time as cows entered the experimental trial.

Cows were housed in the prepartum group pen containing 24 freestalls fitted with a mattress (Pasture Mat, Promat Inc., Woodstock, Ontario, Canada) covered with 5 cm of sand bedding, 12 Insentec feed bins and 2 Insentec water troughs (see Chapinal et al., 2007 for description). Alternate bins provided the two diets; these bins were programmed to allow access only to those cows assigned to that diet, allowing us to house randomly assigned cows to dietary treatment in the same pen. At all times, 12 cows were assigned to each dietary treatment in the prepartum pen to maintain a constant stocking density; if necessary, non experimental cows in the same stage of lactation were taken from the main herd and moved into the pen to maintain the stocking density. Group composition was dynamic with cows entering and leaving the experiment

depending on their expected and actual calving dates. At signs of imminent calving (i.e., udder enlargement, milk letdown, relaxation of tail ligament) cows were moved to a maternity pen. The maternity pen consisted of a sawdust bedded pack fitted with a single Insentec feed bin to allow for continued monitoring of DMI. Cows also had free access to water from a self-filling water trough. Cows were fed their assigned diets in the maternity pen. After calving, cows were milked and moved directly to the postpartum group pen. The postpartum pen consisted of 12 stalls identical to those described above, 6 Insentec feed bins and a single Insentec water trough. All cows, regardless of prepartum treatment, were fed a common lactation diet (NE_L = 1.59 Mcal/kg; Table 1 & Table 2) and monitored for an additional 2 weeks. Cows in the postpartum pen were milked twice a day at approximately 0630 and 1700h. After the two week experimental period, cows were returned to the main milking herd and housed in a freestall pen fitted with a post and rail feed barrier system, and fed the lactation diet described in Table 1 and Table 2. If experimental cows were used in subsequent research trials, they were distributed equally among treatments.

Pre- and postpartum groups were fed at TMR twice daily at approximately 0700 and 1600h. Fresh TMR samples were taken twice weekly immediately before the morning feeding. Samples were pooled weekly to create one weekly sample. Samples were dried at 60°C for 48 h to determine DM content. Dried weekly samples were pooled into monthly composites and ground. Monthly composite samples were analyzed for contents of DM, CP, ADF, NDF, sugar, starch, crude fat and mineral analysis using wet chemistry methods (Cumberland Valley Analytical Services, Inc., Maugansville, MD; Cumberland Valley Analytical Services, 2011).

Individual Animal Factors, Health Checks and Blood Sampling

Individual DMI was measured daily using the Insentec system. Daily feed intakes were corrected weekly for DM. Body weight was measured weekly on 2 consecutive days and an average value was taken. BCS (Ferguson et al., 1994) was determined once a week for each cow starting 3 weeks before expected parturition and continuing for 2 weeks postpartum.

After calving, all cows were checked twice weekly for metritis. Cows were sorted after morning milking and restrained using a headlock feed barrier. Puerperal metritis was diagnosed using a speculum and visually examining vaginal discharge. Briefly, before inserting the speculum into the vaginal cavity, the vulva was thoroughly cleaned with a paper towel soaked with iodine solution (Prepodyne Scrub, Kane Veterinary Supplies, Edmonton, Alberta, Canada) diluted in warm water. The speculum was disinfected in an iodine solution and inserted into the vaginal cavity and any discharge was visually examined using a flashlight. Appearance and smell of the vaginal discharge was evaluated and assigned a score based on the scoring system of Urton et al. (2005): no mucus or clear mucus = 0; cloudy mucus or mucus with flecks of puss = 1; mucopurulent (≤50% pus present) and foul smelling = 2; purulent (≥ 50% puss present) and foul smelling =3; and putrid (red/brown colour, watery, foul smelling) = 4. Cows were diagnosed as having puerperal metritis if they showed a vaginal discharge (**VD**) score of 4. Cows were diagnosed with mild metritis if they showed a VD score of 2 or 3.

Blood was sampled between 900 and 1200h twice a week starting 3 weeks before expected parturition and daily postpartum for 10 days. Blood samples were collected

from the coccygeal vein using Vacutainer tubes (Vacutainer Venous Blood Collection Tubes, BD Biosciences, Franklin Lakes, NJ). BHBA analysis was performed on whole blood for postpartum cows immediately after the sample was collected using a Precision Xtra meter (MediSense, Abbott, Gurnee, Illinois) (see Iwersen et al., 2009 for description). Samples were refrigerated for 1 to 3h, allowed to clot and were then centrifuged at 2700 rpm for 15 min. Serum was recovered, divided into 2 allocates and frozen immediately at -20°C for later NEFA analysis using the microtiter procedure (Wako NEFA-HR (2)), Richmond, Virginia (see method from Johnson and Peters, 1993).

Cows were diagnosed as having subclinical ketosis (**SCK**) using a threshold of serum BHBA concentration ≥1.0 mmol/L in the first week postpartum and/or ≥1.4 mmol/L from day 7 to day 10 postpartum (Walsh et al., 2007). Retained placenta (**RP**) was diagnosed if any placenta tissue was hanging from the vulva 24h after parturition. As per standard farm practice, cows with RP were treated with penicillin for 3 consecutive days. Displaced abomasum (**DA**) was diagnosed by listening for rumen contraction and abnormal abomasum sounds. The herd veterinarian treated identified cases of DA immediately. Clinical signs of mastitis included clots, abnormal milk or a positive CMT test (Sargeant et al., 2001). Body temperatures were taken daily between 900 and 1200h starting 3 weeks prepartum until 2 weeks postpartum using a rectal thermometer (GLA M750, GLA Agriculture Electronics, San Luis Obispo, CA). After calving, all cows with parity greater than 3 were treated with a subcutaneous infusion of 250 mL of calcium borogluconate.

Daily milk weights were recorded for each cow during the morning and afternoon milking. Milk samples were taken weekly for 2 consecutive days and composite samples

were analyzed for fat, lactose, protein and SCC (Pacific Milk Analysis Lab, Chilliwack, British Columbia). In situations where milk was withheld from the tank (colostrum, sick cows) milk production was recorded manually.

In order to assess reproductive performance the interval from calving to pregnancy (days open) was recorded. The herd veterinarian diagnosed pregnancy.

Time of calving was determined by analyzing video recorded in the maternity pen with a camera (WV-BP330, Panasonic, Osaka, Japan) mounted 6m above the pen. Videos were recorded to a digital recording system (Genetec Inc., Saint-Laurent, Quebec, Canada). Time of calving was recorded as the hour in which the calf was fully expelled from the cow.

Statistical Analysis

Four cows were dropped from the study due to miscellaneous health problems (one with a prolapsed uterus; one with severe edema; one euthanized 6 d postpartum; and one with a ripped teat). Statistical analyses were performed using SAS 9.1 with cow as the experimental unit. One cow on the 77F diet calved 8d early, limiting the beginning of prepartum data collection to begin d-13 relative to calving. Feeding events were screened for outliers based on feeding rates using PROC UNVARIATE in SAS. Of the 165,998 feeding events (times the system identified an individual cow at the feeder), 3.2% were identified as falling more than 3 times the interquartile range beyond first and third quartile and were removed from analysis. Hour of calving marked the beginning of day 0 and experimental days were adjusted accordingly. Five experimental periods were used for analysis: week -2 (day -13 to day -8), week-1 (day -7 to day -1), calving (day 0),

week+1 (day 1 to day 7) and week+2 (day 8 to day 14). Previous work has shown differences in disease risk across these periods (Huzzey et al., 2007), so data were also analyzed by periods in this study. DMI data were analyzed using PROC MIXED with day treated as a repeated measure and cow as the subject and with a compound symmetry covariance matrix. Data were tested for fixed effects in this order: treatment, period and treatment by period. There was a treatment by period interaction; therefore, data were analysed by period with treatment as a fixed effect.

Cows with BHBA levels ≥ 1.4 mmol/L in week +2 or later were not included as this has been shown to be indicative of prolonged NEB and are often associated with clinical ketosis (Walsh et al., 2007). Following the removal of these animals (n=8 on 77F diet and n=4 on 87F diet), data were screened for outliers and data points falling 3 times beyond the first and third quartile were removed from analysis (0.85% of observations). BHBA data were analyzed using PROC MIXED with day treated as a repeated measure and cow as subject with a compound symmetry covariance matrix. Analysis of fixed effects was carried out in this order: treatment, day and treatment by day. In order to analyse NEFA data, the results were divided into 5 experimental periods: week -2 (day -13 to day -8), week-1 (day -7 to day -1), calving (day 0), week+1 (day 1 to day 7) and week+2 (day 8 to day 14). NEFA data were analyzed using PROC MIXED with period treated as a repeated measure, with cow as subject and a compound symmetry covariance matrix. Analysis of fixed effects was carried out in this order: treatment, week and treatment by week. The effects of periods were significant; therefore a second analysis was carried out using contrast statements to test the difference in NEFA concentration for

all cows regardless of treatment between week -2 to week -1, week -1 to calving, calving to week +1 and week +1 to week +2.

Health data were analyzed by a chi-squared analysis to compare observed frequencies with expected frequencies by chance. Cows were classified as either having a specific illness or not having it as described by the definitions above.

Milk production data were analyzed in weekly periods for 22 weeks. Difference in milk production between the two treatment groups was analyzed using a mixed model with cow as subject, period as the repeated measure and a compound symmetry covariance structure. Analysis of fixed effects was carried out in this order: treatment week and treatment by week. Averages of SCC, lactose, protein and fat were summarized into two weekly periods: week+1 (day 1 to day 7) and week+2 (day 8 to day 14) and analyzed using a MIXED model with an autoregressive covariance structure, with week as the repeated measures. Analyses of fixed effects were tested in the following order: treatment, week and treatment by week.

Energy balance was calculated for each cow using NRC calculations for energy requirements (NRC, 2001). Net energy intake (NE_I; Mcal/d) was determined by multiplying DMI by the calculated mean NE_L density of the diet. Energy requirements for maintenance (NE_M; Mcal/d) were calculated as follows: $NE_M = 0.080$ Mcal/kg x $BW^{0.75}$. Energy requirements for pregnancy (NE_P; Mcal/d) were calculated using the following: $NE_P = [(0.00318 \times D - 0.0352) \times (CBW/45)]/0.218$, where D = day of gestation and CBW is the calf birth weight in kg. Calf birth weight was measured for 51 calves (n=28 on the 77F diet; n=23 on 87F diet); therefore, these cows were used for calculations. Prepartum energy balance were determined as follows: $NE_{PRE} = NE_I - (NE_P$

+ NE_M). Net energy for milk production (NE_L; Mcal/d) = (0.0929 x fat %) + (0.0563 x protein %) + (0.0395 x lactose %) x milk yield (kg/d). Postpartum energy balance was calculated as follows: $NE_{POST} = NE_I - (NE_M + NE_L)$. Pre and postpartum energy balances were calculated for each cow in order to report their energy balances and determine the magnitude of their NEB.

Prepartum NE_I data and pre and postpartum energy balance data were analyzed using PROC MIXED with day treated as a repeated measure and cow as subject with a compound symmetry covariance matrix. Analysis of fixed effects was carried out in the following order: treatment, day and treatment by day.

In order to analyze reproductive data, cows were classified as pregnant or not pregnant at 90 DIM, 120 DIM and 150 DIM. Pregnancy rate was analyzed by a chi-squared analysis to compare observed with expected frequencies. For this analysis, experimental cows culled from the herd before 60 DIM were removed from analysis, leading to the removal of 8 cows, 4 from the 87F diet and 4 from the 77F diet.

RESULTS

DMI

Cows fed the 77F diet had higher DMI pre-calving ($P<0.0001$) than those fed the 87F diet; however, we observed no differences in DMI in the 24 hours before calving (9.1 ± 4.0 kg/d; Figure 3). Both treatment groups showed drops in DMI from week -2 to week -1 (77F diet: 16.4 ± 0.4kg/d vs. 14.8 ± 0.4kg/d; $P<0.0001$ and 87F diet: 14.0 ± 0.5kg/d vs. 12.0 ± 0.5kg/d; $P<0.0001$). We observed no difference in DMI between the 2 groups in the postpartum period with all cows consuming on average 19.7 ± 5.5 kg/d. Cows diagnosed with SCK, regardless of treatment, consumed less feed than did cows that

transitioned without any health issues in week 1 after calving (17.2 ± 0.4 kg/d vs 19.4 ± 0.6kg/d; $P<0.001$). There was a trend in the same direction week 2 after calving (19.6 ± 0.4kg/d vs 20.8 ± 0.58kg/d; $P=0.1$).

Energy Balance

Throughout the experiment, BCS did not differ between treatments, averaging (mean ± SD) 3.3 ± 0.3 in both treatments. However, we did observe a drop in BCS from week -1 to week +2 (3.4 ± 0.03 vs. 3.2 ± 0.03; $P<0.0001$) for both treatments.

With increased DMI comes increased NE_I. Cows fed the 77F diet had a higher NE_I than those fed the 87F diet in week -2 (22.4 ± 0.7 Mcal/d vs. 18.7 ± 0.8 Mcal/d; $P=0.0004$) and in week -1 (19.9 ± 0.7 Mcal/d vs. 16.0 ± 0.7 Mcal/d; $P<0.001$) (Figure 4). Calculated required energy for prepartum cows was 15.3 ± 1.2 Mcal/d. Both treatment groups consumed in excess to their requirements between d-12 and d-2 relative to calving (77F diet 29.3% in excess; 87F diet 12.8% excess). After calving, there was no difference in NE_I. Cows on the 77F diet tended to have heavier calves (45.1 ± 1.1kg vs. 42.1 ± 1.2 kg; $P=0.07$).

As expected, all cows were in a positive energy balance before calving, but the surplus was more pronounced for cows on the 77F diet compared to cows receiving the 87F diet in week -2 (9.8 ± 0.6 Mcal/d vs 3.3 ± 0.7 Mcal/d; $P<0.001$) and week-1 (6.5 ± 0.7 Mcal/d vs. 1.9 ± 0.8 Mcal/d; $P<0.001$) (Figure 5). In the weeks following parturition all cows were in a NEB (-13.9 ± 9.5 Mcal/d; $P=0.23$).

Blood Metabolites

No cows in the prepartum period displayed any obvious signs of illness (i.e. fever) and were all classified as healthy before calving. Postpartum blood BHBA levels were lower for cows fed the 87F diet prepartum compared to those on the 77F diet (0.49 ± 0.02 mmol/L vs. 0.58 ± 0.02 mmol/L; *P*=0.003) (Figure 1). In the week -1 before calving, cows on the 77F diet tended to have lower serum NEFA levels compared to cows on the 87F diet (0.72 ± 0.03 meq/L vs. 0.79 meq/L; *P* = 0.07; Figure 2). During the remainder of the study period no difference in NEFA concentrations were observed between the two treatments; however, there was an effect of week for all cows. NEFA concentrations were lowest at week -2, began increasing by week -1 (0.68 ± 0.03 meq/L to 0.76 ± 0.03 meq/L; *P*=0.04) and peaked on the day of calving (1.40 ± 0.05 meq/L). By week +1 NEFA levels once again had decreased and continued to do so into week +2 (1.06 ± 0.02 meq/L to 0.97 ± 0.03 meq/L; *P* <0.001).

Health Outcomes

Overall, more cows were classified as having SCK postpartum on the 77F diet (48.9% vs. 16.7%, *P*=0.001). There was also a trend for more cows on the 77F diet to develop metritis postpartum (28.9% vs. 14.3%; *P*=0.1). There were no treatment differences for mild metritis, RP, milk fever, mastitis or DA. More cows on the 87F diet tended to remain healthy and go through the transition period without displaying any signs of illness (13.3% vs. 27.6%; *P*=0.08; Table 3).

Milk Production, Components & Reproductive Performance

In the first week after calving, cows fed the 77F diet and the 87F diet produced 33.3 ± 0.8kg/d and 31.8 ± 0.9kg/d, respectively (P=0.2; Figure 6). There was a trend for cows on the 77F diet to produce more milk in the second week postpartum (44.6 ± 1.0kg/d vs. 41.7 ± 1.1 kg/d; P=0.06). However, over the first 22 weeks of lactation, we noted no differences in average weekly milk production for cows fed the 77F diet versus the 87F diet (48.1 ± 1.0 kg/d and 46.7 ± 1.0 kg/d respectively; P=0.33).

During the first 2 week of lactation there were no differences (P>0.05) in SCC (311,000 ± 60,000 /ml), fat (5.8 ± 0.8%), protein (3.8 ± 0.05%) or lactose (4.2 ± 0.02%) between the treatment groups.

By 120 DIM, 52.6% of cows fed the 87F diet were pregnant, versus 31.7% of those on the 77F diet (χ^2 = 3.55; P=0.06). At 150DIM 71.1% of cows fed the 87F diets were pregnant, versus 51.2% of cows fed the 77F diet (χ^2 = 3.3; P=0.07).

DISCUSSION

Feed Intake

Cows fed the 77F diet consumed more DM during the 2 week before parturition. This finding is in accordance with other work that reported that cows fed lower forage diets prepartum have higher intakes than those high forage diets (Holcomb et al., 2001). Previous work has shown that the greatest drop in DMI takes place on day -1 (Goldhawk et al., 2009; Huzzey et al., 2007). The greatest decline in DMI in both treatment groups took place 48 to 24 h before calving with those cows consuming the 77F diet showing the greatest declines; cows on the 87F diet declined on average 2.3 kg/d and those on the 77F

diet declined by 4.3 kg/d, relative to the previous 24 h.

Higher energy diets fed prepartum have been shown to cause greater drops in DMI before calving (Grummer et al., 2004), though mechanisms explaining this effect remain unclear (see review by Ingvartsen and Andersen, 2000). Preventing large drops in DMI as parturition approaches is important; these drops in DMI in the week before calving have been associated with increased risk of metritis (Huzzey et al., 2007) and increased lipid mobilization from adipose tissues increasing the risk of metabolic disorders (Bertics et al., 1992).

The lack of difference in DMI in postpartum cows was surprising given that previous reports had predicted higher postpartum DMI in cases where cows were restricted to consuming energy intakes at or below NRC recommendations in the prepartum period (Agenäs et al., 2003; Dann et al., 2006; Douglas et al., 2006). By design, cows in the present study were provided ad libitum access to diets that had similar energy contents but differed in the amount of forage (addition of rye grass straw of the treatment diet). This resulted in cows on the 77F diet and the 87F diet consuming 130% and 112%, respectively, of NRC recommendations in the 10 days before calving. In a review article, Grummer (1995) reported a positive correlation between prepartum and postpartum feed intake, suggesting it was important to maximize intake before calving. It should also be noted that individual differences among cows can play a role in driving DMI fluctuations, as cows housed in the same environments and fed the same prepartum diets show great fluctuations in the magnitude of DMI decrease around calving (Grummer, 1995). Our results show cows consuming higher DMI pre-calving did not have increased DMI in the post calving period. This result suggests that focusing on

prepartum DMI, without considering net energy intake and diet ingredients, will not improve appetite post calving.

Blood Metabolites

As cows approach parturition, body stores are mobilized leading to increased serum NEFA levels. Excess lipids are stored as TG in the liver and decrease the metabolic function of the liver (see review from Bobe et al., 2004). In this study, NEFA levels demonstrated similar patterns for both treatment groups with concentrations increasing as parturition approached. Other studies have shown that restricting intake to below NE_{PRE} requirements can lead to higher NEFA concentrations providing evidence that the cows mobilize body stores to support fetal development (Douglas et al., 2006). Cows in the present study were consuming above NE_{PRE} requirements, perhaps explaining the lack of difference in NEFA concentrations.

Postpartum BHBA levels were higher for cows fed the 77F diets; this result agrees with previous work showing that limiting energy intake in the dry period results in higher energy balance leading to lower BHBA levels postpartum (Dann et al., 2006). Interestingly, milk production and postpartum DMI did not differ among treatment groups. Cows consuming the 77F diet were likely exhibiting higher rates of ketogenesis in order to meet the energy demand of milk production, providing further evidence of the compromised metabolic state of the cows fed the 77F diet prepartum. This explanation is in agreement with other work suggesting that cows consuming less energy in the dry period are more efficient in energy utilization (Agenäs et al., 2003). We failed to see the expected response in NEFA levels, but did see, as expected, elevated BHBA

concentrations postpartum associated with postpartum illness and reduced reproductive performance as reported by other authors (Ospina et al., 2010a; Ospina et al., 2010b; Walsh et al., 2007). In non-ruminants, increased BHBA concentrations have been shown to increase insulin secretion thereby inhibiting lipolysis, leading to lower blood free fatty acid concentrations (Bates and Linn, 1976). Further, there is now growing acceptance that BHBA is anti-lipolytic in that it regulates its own production though a negative feedback mechanism that conserves body fat stores (Senior and Loridan, 1968). The lack of agreement between our study and others suggests the need for further research.

Cows fed in excess to their nutrient requirements during the dry period may develop insulin insensitivity. When the net energy intake of cows is reduced through either increasing forage content of the diet or restricting intakes of high-energy diets, cows respond by lowering insulin concentrations (Dann et al., 2006; Douglas et al., 2006; Holcomb et al., 2001). Similarly, cows allowed to consume excess energy (in this case 178% of that recommended by NRC) showed a higher insulin response following a glucose challenge in the prepartum period than those fed at 110% of the level recommended by NRC (Holtenius et al., 2003).

Health

Several authors have suggested that controlling energy intake in the prepartum period may improve postpartum health (Beever, 2006; Drackley and Janovick Guretzky, 2007). Until now, there has been no scientific evidence in support of this idea. Although Dann et al (2006) recorded health disorders they were unable to show differences in the frequency of these disorders between cows fed diets varying in net energy intake, perhaps

due to the small number of animals used. Metabolic profiling of transition cows has shown that cows with increased NEFA concentrations are at an increased risk for fatty liver and other metabolic disorders, such as ketosis (Bobe et al., 2004; Grummer, 1993). Walsh et al. (2007) observed that cows with BHBA levels ≥ 1.0 mmol/L in the first week prepartum, consistent with our definition of SCK, were 20% less likely to become pregnant after the first insemination and were 50% less likely to become pregnant. Excessive NEB, defined using BHBA threshold levels, have been used as predictors of disease and herd level problems. A BHBA concentrations ≥ 1.0 mmol/L from day 3 to day 14 postpartum is associated with increased risk of clinical ketosis and metritis (Ospina et al., 2010b). Further, cows with BHBA concentrations ≥ 1.0 mmol/L postpartum had a 13% decreased risk of pregnancy within a 70 d voluntary waiting period as well as 393 kg decrease in milk production for a 305 day lactation, illustrating the detrimental effects of excessive NEB on reproductive performance and milk production (Ospina et al., 2010a). Cows in the present study that were fed the 87F diet throughout the entire dry period were at a reduced risk of developing subclinical ketosis and therefore likely to experience fewer reproductive problems. In this study, cows fed the 87F diet tended to get pregnant quicker than those fed the 77F diet. Half the cows on the 87F diet were pregnant by 120DIM, while it took up to 150DIM for 50% of the cows on the 77F diet to become pregnant.

Cows experiencing subclinical endometritis, puerperal metritis or subclinical endometritis in the periparturient period had decreased DMI (Huzzey et al., 2007) and higher NEFA and BHBA levels (Hammon et al., 2006), all indicators of prolonged NEB in the periparturient period. In our study, cows on the 87F diet prepartum had similar

postpartum DMI to those fed a 77F diet; however, BHBA levels were lower and fewer cases of metritis after parturition were observed, suggesting improved immune status. This interpretation is consistent with results showing reduced neutrophil function in cows experiencing NEB postpartum (Hammon et al., 2006). Due to the limited number of cases of metritis and SCK on the 87F diet (6 and 7 animals, respectively), inferential comparisons were not performed (Table 4); we encourage more work using larger sample numbers.

CONCLUSION

The results from the current study are consistent with earlier work demonstrating that over consumption of energy during the dry period can be detrimental to transition cow health (Agenäs et al., 2003; Dann et al., 2006; Douglas et al., 2006; Holtenius et al., 2003). Transition cows appear unable to regulate intake to meet demands, and consume excess to requirements when provided energy dense diets (Dann et al., 2006). Controlling energy intake during the dry period by adding forages to dilute nutrient content lowers disease risk.

TABLES

Table 1: Ingredients and chemical composition (as % DM) of diets (mean ± SD).
All cows were fed the 87F diet upon dry off; 3 weeks before expected parturition half the cows were switched to the 77F diet (n=45), while the other half remained on the 87F diet until parturition (n=42). After calving, all cows (n=87) were fed the lactation diet.

	77F Diet	87F Diet	Lactation Diet
Ingredients (%)			
Corn silage	33.0	30.0	29.0
Alfalfa hay	44.0	12.0	5.0
Grass silage	-	14.0	11.0
Grass hay	-	-	4.0
Rye grass straw	-	30.0	-
Concentrate mix	23.0	13.0	51.0
Chemical			
Crude Protein, %	15.0 ± 0.6	14.2 ± 0.6	16.9 ± 0.7
Soluble Protein, % CP	37.6 ± 2.7	42.0 ± 4.0	36.8 ± 3.3
Acid Detergent Fiber, %	29.4 ± 1.9	32.6 ± 1.4	21.5 ± 1.2
Neutral Detergent Fiber, %	41.3 ± 1.7	48.4 ± 1.9	35.6 ± 1.9
Sugar, %	5.8 ± 0.9	3.9 ± 0.4	4.4 ± 0.4
Starch, %	16.3 ± 1.9	13.6 ± 1.6	22.5 ± 2.6
Crude Fat, %	2.3 ± 0.4	2.4 ± 0.4	3.4 ± 0.5
Ash, %DM	8.7 ± 0.7	8.4 ±0.4	8.2 ± 0.5
Calcium, %DM	1.1 ± 0.1	0.9 ± 0.1	1.0 ± 0.1
Phosphorus, %DM	0.3 ± 0.02	0.3 ± 0.01	0.4 ± 0.01
Magnesium, %DM	0.4 ± 0.01	0.3 ± 0.03	0.3 ± 0.03
Potassium, %DM	1.5 ± 0.2	1.9 ± 0.2	1.7 ± 0.1
Sulfur, %DM	0.3 ± 0.01	0.2 ± 0.01	0.2 ± 0.01
Sodium, %DM	0.2 ± 0.04	0.2 ± 0.03	0.4 ± 0.02
Iron, PPM	578.9 ± 172.2	452.3 ± 33.9	427.3 ± 90.4
Manganese, PPM	59.0 ± 14.0	99.1 ± 20.4	72.3 ± 10.6
Zinc, PPM	72.2 ± 7.5	71.4 ± 10.0	150.0 ± 19.2
Copper, PPM	19.4 ± 1.4	22.9 ± 3.0	24.1 ± 2.3
Chloride Ion, %DM	0.4 ± 0.6	0.5 ± 0.1	0.5 ± 0.1
Net Energy Lactation (Mcal/kg)	1.45 ± 0.02	1.41 ± 0.03	1.59 ± 0.1

Table 2: Composition of concentrate mix fed in the pre and postpartum diets
All cows were fed the 87F diet upon dry off; 3 weeks before expected parturition half the cows were switched to the 77F diet (n=45), while the other half remained on the 87F diet until parturition (n=42). After calving, all cows (n=87) were fed the lactation diet.

	77F Diet	87F Diet	Lactation Diet
Concentrate Ingredients (%)			
Soybean hulls	70.4	-	-
Barley, rolled	6	-	28
soybean meal	-	28.9	8.6
Canola meal	-	23	7.6
barley, fine ground	-	-	20
Corn gluten meal	-	5.6	-
Corn, rolled	-	-	8
Distillers corn wheat	1.8	-	8
Aminoplus[1]	12	22.5	6.8
Mill run	-	4	4.2
Dicalcium phosphate	1	-	-
Calcium sulfate	2	2.3	-
Limestone	8.8	4.1	2.1
Magnesium oxide	0.75	1.1	0.3
Salt cobalt	0.6	-	-
Vitamin E premix	1.2	-	-
A-Max yeast[2]	0.4	0.7	0.2
Molasses	0.4	1.2	1.5
Mineral and Protein mix[3]	0.075	-	-
Complete Dairy Premix[4]	0.283	-	-
Calcium chloride	0.625	-	-
Selenium	0.165	-	-
Sel-Plex 2000[5]	0.016	-	-
Niacin Premix	0.9	-	-
Dairy premix[6]	0.397	-	1.1
Pre lactation mineral	0.05	-	-
Pre lactation vitamin	0.1	-	-
Salt		1.4	0.7
Urea	-	-	0.04
Sodium bicarbonate	-	-	1.2
Smartamine Metasmart[7]	-	-	0.04
Calcium salt	-	-	1.7

[1] Ag Processing Inc, Omaha, NE
[2] Vi-Cor, Mason City, IA
[3] Alltech, Van Water and Rogers, Nicholasville, KY
[4] Complete dairy premix includes trace minerals and Vitamin ADE
[5] Alltech, Nicholasville, KY. Includes a vitamin and mineral complex with active dry yeast (60g/d)
[6] Akey Lewisberg, OH. Dairy premix is a vitamin and mineral complex formulated for stage of lactation
[7] Adisseo, Antony, France

Table 3: Incidences of postpartum disease per treatment (%).
On each treatment, a number of cows had more than one disease. All cows were fed the 87F diet upon dry off; 3 weeks before expected parturition half the cows were switched to the 77F diet (n=45), while the other half remained on the 87F diet until parturition (n=42). After calving, all cows (n=87) were fed the lactation diet.

Item	77F Diet	87F Diet	χ^2	P
RP	4.44	11.9	1.63	0.2011
Milk Fever	4.44	7.14	0.292	0.5889
Mastitis	11.11	7.14	0.4097	0.5221
DA	2.22	2.38	0.0024	0.9606
SCK	48.89	16.67	10.15	0.0014
Metritis	28.89	14.29	2.714	0.0995
Mild Metritis	46.67	61.9	2.0309	0.1541
Healthy	13.33	28.57	3.0741	0.0795

Table 4: DMI, NEFA, BHBA and milk production (mean ± SD) for cows on a 87F diet or a 77F diet prepartum.
Cows either transitioned healthy (n=12 & n=6), with SCK (n=7 & n=22) or with metritis (n=6 & n=13). All cows were fed the 87F diet upon dry off; 3 weeks before expected parturition half the cows were switched to the 77F diet, while the other half remained on the 87F diet until parturition. After calving, all cows were fed the lactation diet

	Healthy				SCK				Metritis			
	week -2	week -1	week +1	week +2	week -2	week -1	week +1	week +2	week -2	week -1	week +1	week +2
DMI (kg/d)												
87F	14.1 ± 2.5	11.9 ± 2.8	18.6 ± 5.0	22.3 ± 6.5	13.6 ± 4.3	11.0 ± 0.6	15.41 ± 6.0	16.8 ± 5.8	12.2 ± 3.9	10.6 ± 4.4	14.5 ± 6.8	20.8 ± 9.7
77F	16.9 ± 2.5	14.7 ± 3.7	21.8 ± 6.2	21.5 ± 3.8	15.1 ± 0.5	14.1 ± 3.6	17.7 ± 4.6	20.4 ± 3.0	17.8 ± 2.9	16.4 ± 4.2	20.4 ± 6.9	22.0 ± 4.3
NEFA (Meq/ml)												
87F	0.6 ± 0.1	0.8 ± 0.4	1.0 ± 0.4	1.0 ± 0.4	0.8 ± 0.3	0.8 ± 0.1	1.6 ± 0.5	1.1 ± 0.2	0.7 ± 0.3	0.9 ± 0.1	1.3 ± 0.5	1.4 ± 0.7
77F	0.8 ± 0.2	0.8 ± 0.1	1.1 ± 0.3	1.1 ± 0.3	0.6 ± 0.3	0.7 ± 0.3	1.1 ± 0.6	0.9 ± 0.3	0.8 ± 0.1	0.7 ± 0.3	1.1 ± 0.4	1.0 ± 0.2
BHBA (mmol/L)												
87F			0.5 ± 0.2				0.8 ± 0.3				0.5 ± 0.2	
77F			0.5 ± 0.2				0.7 ± 0.3				0.5 ± 0.2	
Milk production (kg/d)												
87F			39.9 ± 11.2				39.8 ± 10.6				36.5 ± 10.5	
77F			38.3 ± 10.1				34.7 ± 8.3				33.2 ± 12.4	

FIGURES

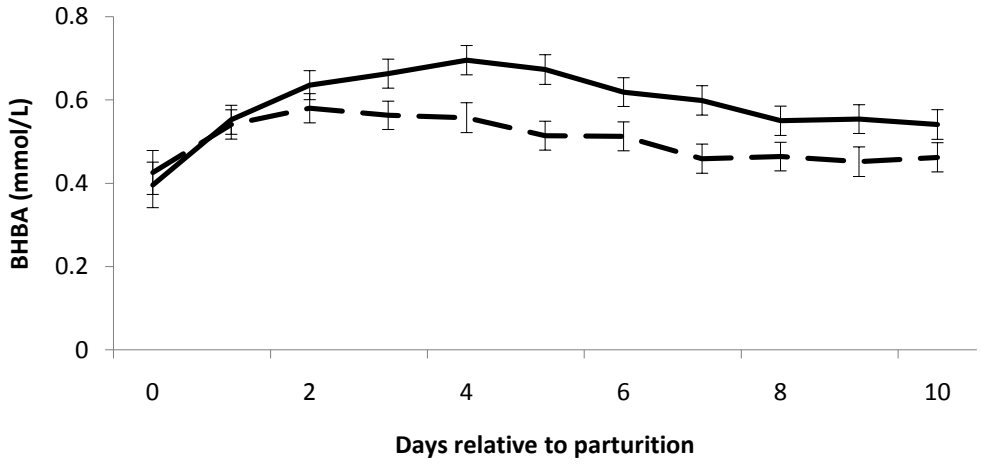

Figure 1: Least square mean (±SE) of BHBA (mmol/L).

All cows were fed the 87F diet upon dry off; 3 weeks before expected parturition half the cows were switched to the 77F diet (——— n=45), while the other half remained on the 87F diet until parturition (--------; n=42). After calving, all cows (n=87) were fed the lactation diet. Treatment differences were significant (*P*=0.003)

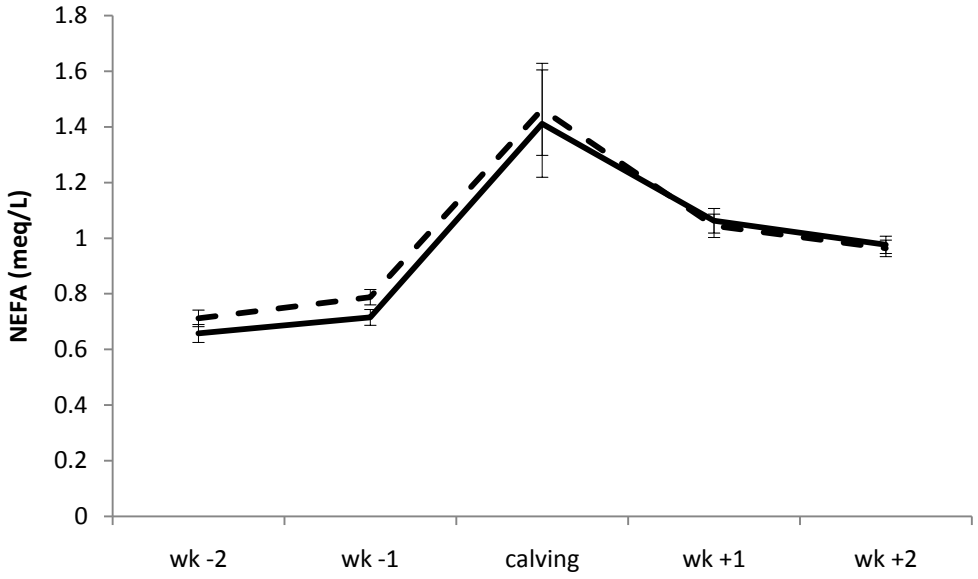

Figure 2: Least square mean (±SE) of NEFA (meq/L).

All cows were fed the 87F diet upon dry off; 3 weeks before expected parturition half the cows were switched to the 77F diet (——— n=45), while the other half remained on the 87F diet until parturition (--------; n=42). After calving, all cows (n=87) were fed the lactation diet. There was a tendency for treatments to differ in week -1 (P=0.07) but not during the other periods.

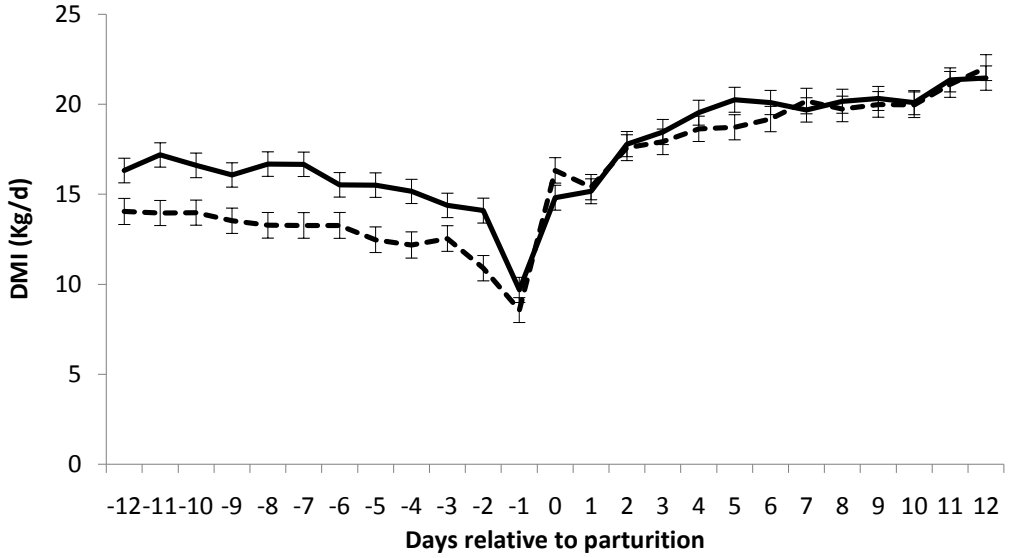

Figure 3: Least square means (±SE) of DMI (kg/d).

All cows were fed the 87F diet upon dry off; 3 weeks before expected parturition half the cows were switched to the 77F diet (——— n=45), while the other half remained on the 87F diet until parturition (--------; n=42). After calving, all cows (n=87) were fed the lactation diet. DMI varied with treatment prepartum (P<0.0001), but not postpartum.

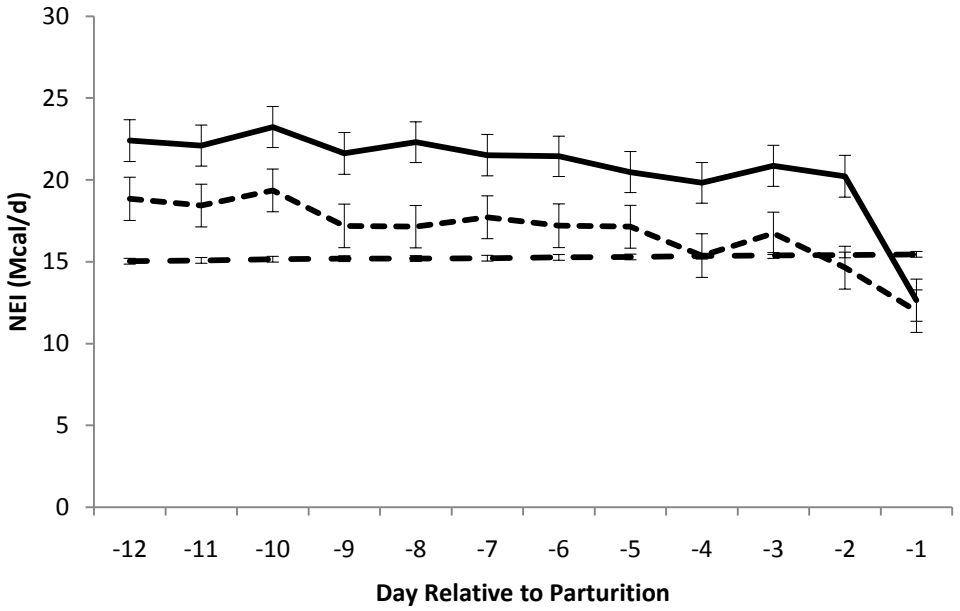

Figure 4: Least square means (±SE) for prepartum NE_I (Mcal/d).

All cows were fed the 87F diet upon dry off; 3 weeks before expected parturition half the cows were switched to the 77F diet (——— n=28), while the other half remained on the 87F diet until parturition (--------; n=23). After calving, all cows (n=51) were fed the lactation diet. Prepartum energy requirement denoted by ======. Treatment differed in week -2 (P=0.0004) and week -1 (P<0.001).

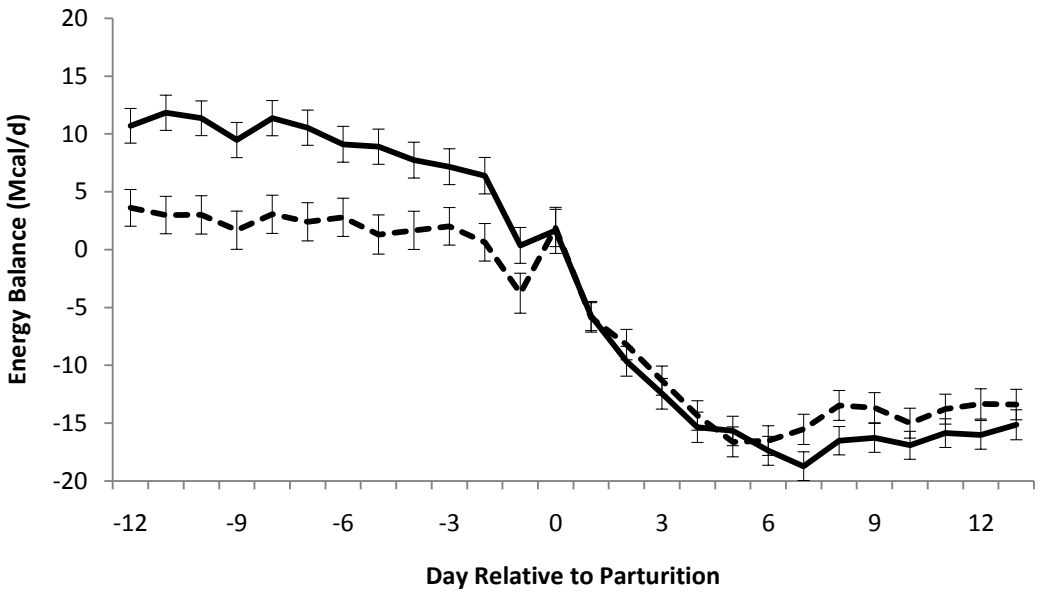

Figure 5: Prepartum and postpartum energy balance (least squared means ± SE) (Mcal/d).

All cows were fed the 87F diet upon dry off; 3 weeks before expected parturition half the cows were switched to the 77F diet (——— n=45), while the other half remained on the 87F diet until parturition (--------; n=42). After calving, all cows (n=87) were fed the lactation diet. Treatments differed in week -2 and week -1 (P<0.0001), but not postpartum (P=0.23)

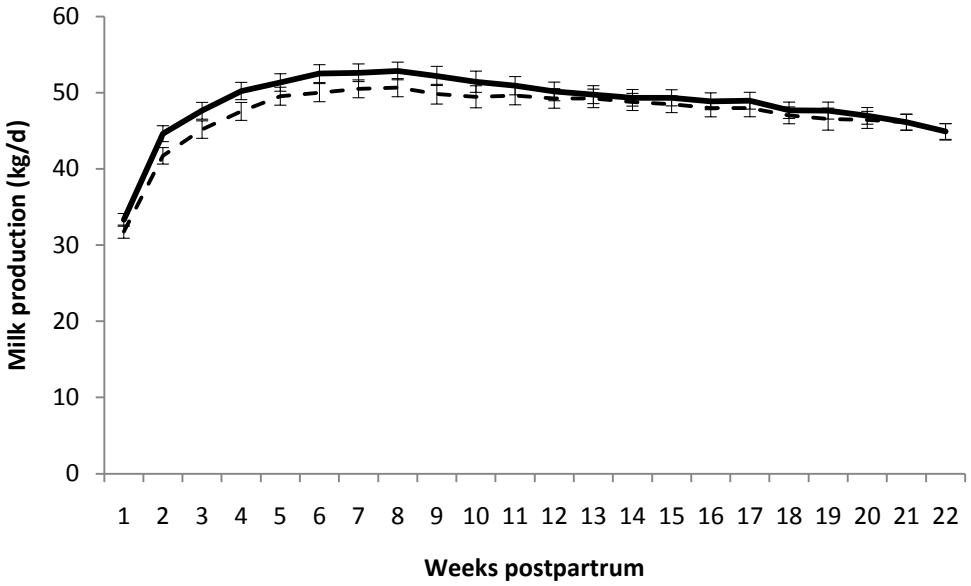

Figure 6: Least square means (±SE) for weekly milk production (kg/d).

All cows were fed the 87F diet upon dry off; 3 weeks before expected parturition half the cows were switched to the 77F diet (——— n=45), while the other half remained on the 87F diet until parturition (--------; n=42). After calving, all cows (n=87) were fed the lactation diet. In week+2 there was a trend for treatments to differ (P=0.06), but over the entire 22-week period no treatment differences were detected (P=0.33).

CHAPTER 3: GENERAL CONCLUSION

Cows that fail to transition successfully into lactation are vulnerable to a host of problems during lactation including a higher disease risk, lower milk production and compromised reproductive performance. Therefore, it is important for the cows' productivity and her welfare that she not succumb to production diseases. It is now thought that cows that are overfed in the dry period may become insulin resistant. Holtenius et al (2003) fed cows either 6, 9 or 14.5 kg/d DM and found those offered the high diet had higher insulin and glucose levels in the dry period, and showed a higher insulin peak when presented with a glucose tolerance tests suggesting a degree of insulin resistance. Even a moderate degree of insulin resistance can encourage further mobilization of NEFA from the adipose tissues and muscles in order to spare glucose for the mammary glands (Bell, 1995). There are now a number of lines of evidence indicating that controlling energy in the dry period can help cows undergo a successful transition into lactation.

There are still many knowledge gaps in transition cow research such as why vastly different nutrition and management programs produce similar transition cow results, be they good or bad (Drackley, 1999). A successful passage though the transition period is more complex than maximizing feed intake (Grummer et al., 2004). Successfully implementing a one group, high forage dry cow diet on a dairy farm takes careful management. According to Drackley and Janovick Guretzky (2007), care must be taken to ensure sorting behavior is limited, cows have continuous access to feed and that dry matter content of the feed be monitored closely. The authors recommend chopping the straw portion to less than 5cm (approximately 2") particles and closely monitoring for sorting activities; if cows are able to sort against the straw, some animals may consume more energy than the diet is formulated to provide, while other, more timid cows may be left consuming a lower quality ratio (Drackley and Janovick Guretzky,

2007). Further, as a result of the bulky nature of the diet, cows are likely to spend more time eating; therefore, it is vital to ensure ad libitum access to feed. The NDF content of the feed increase throughout the day, indicating that cows are sorting against the forage (DeVries et al., 2005). Feeding TMR twice a day leads to reduced sorting behaviour and results in a more uniform distribution of feed (DeVries et al., 2005). It is important that cows maintain intakes in the transition period and feeding systems should be designed to give animals the opportunity to do so.

Good nutrition programs on a farm will only be able to provide benefits when accompanied by good management practices. Management practices should aim to provide a low stress environment for transition cows as stressors have been linked with a higher concentration of NEFA; multiple stressors can have additive effects, perhaps shedding some light on why outbreaks of metabolic disorders occur on farms (Drackley et al., 2005). Transition cows should not be overstocked at the feed bunk. Multiparous cows overstocked at the feed bunk consume less DM in the week before calving (Proudfoot et al., 2009). Overstocking at the feed bunk leads to less time spent feeding and more time spent inactively standing within the feed area waiting to gain access to feed (Huzzey et al., 2006). Further, when cows are overstocked at the lying stall, they will spend less time lying down and compensate by spending less time standing outside of the stall (Fregonesi et al., 2007).

Dairy cows are social animals (Grant and Albright, 2001) and a newly regrouped cow must quickly find her place in the herd (Grant and Albright, 1995). For the fresh cow, a group change involves both social and nutritive stress as well as adaptation to different feeding and management practices (i.e. milking) (Grant and Albright, 2001). Moving cows between groups causes a period of increased social interactions for approximately 48 hours before the group

reaches stability (Kondo and Hurnik, 1990). Further, regrouping has been shown to decrease feeding time, lying time (von Keyserlingk et al., 2008) and rumination time (Schirmann et al., 2011). Moving cattle on a modern dairy farm is unavoidable; however, by feeding a one-group dry cow diet though the dry period, one routine group change can be eliminated from standard farm practice.

FUTURE RESEARCH

There is growing support that cows overfed in the prepartum period develop insulin resistance (Holtenius et al., 2003) but this trial did not test this concept. It would have been interesting to give cows on both treatments a glucose tolerance test in the prepartum and postpartum period. In a glucose tolerance test, the cows are infused with glucose and the clearance rate (how quickly glucose is taken up from the blood) and insulin peak levels are measured. If cows fed in excess to their energy requirements in the prepartum period show higher insulin peaks it would be an indication that the body tissues are less responsive to insulin and the cows are becoming insulin resistant. Further, there is growing support that prepartum NEB is associated with decreased neutrophil function and higher incidences of uterine health disorders (Hammon et al., 2006). Therefore, it would be interesting to compare the immune status of cows fed a high forage diet and a moderate forage diet.

This trial has provided support that controlling energy intake by increasing forage content in prepartum diets can improve transition success and postpartum health. It is worth investigating whether these concepts can be carried over to lactating cows as well. As concentrates and fermentable non-structural carbohydrates are added to the diet to increase the energy content to support lactation, the risk for subacute ruminal acidosis (**SARA**) increases

(Stone, 2004). SARA and chronic acidosis occur after the ingestion of large amounts of readily fermentable carbohydrates leading to a drop in rumen pH due to a shift in the microbial population favouring the production of lactic acid (Owens et al., 1998). Acute acidosis can result in damage to the ruminal and intestinal wall, while chronic acidosis can lead to depressed feed intake (Owens et al., 1998). High producing cows can be at an increased risk for SARA due to their higher DMI (Stone, 2004). Designing rations for optimum rumen health and high milk production is a balancing act between fermentable carbohydrates and fibre content, as fibre is associated with increased chewing and saliva production which acts to buffer the rumen against drops in pH (Allen, 1997). Therefore, it would be worth investigating whether increasing the forage content of lactation diets could improve the rumen environment and overall health of dairy cows. Cows fed higher forage diets would likely have lower milk production, but this may be offset through improved health.

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