Choline: A Limiting Nutrient for Transition Dairy Cows

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Introduction

Choline has been shown to be a required nutrient for many animals including rats, mice, dogs, pigs, guinea pigs, chickens, and trout. Choline is often referred to as a vitamin, however, it doesn't fit any of the classical definitions for a vitamin. It is not a co-factor in enzymatic reactions, it can be synthesized endogenously as phosphatidylcholine (PC), and it is required in larger amounts than vitamins. The ability to synthesize choline endogenously does not mean it is a dispensable or non-essential nutrient. Deficiency symptoms include suppressed growth rates, renal dysfunction, and development of fatty liver. Choline is crucial for normal function of all cells. The most common form of choline in biological systems is PC, a phospholipid that is a component of all cell membranes and lipoproteins that function to transport lipids through the circulatory system. Choline is a source of methyl groups, therefore, it can spare methionine and have interactions with other nutrients involved in one-carbon metabolism (e.g. folate). Choline is also a component of acetylcholine, an important neurotransmitter.

The NRC (2001) wrote: "The establishment of a choline requirement, either for lactating dairy cow, or a transition cow in the late dry period and in early lactation, will require more extensive feeding experiments than available at the time of this publication." It has now been 15 years since publication of the last NRC. Since publication of the last NRC, numerous studies have been conducted to examine the effects of feeding ruminally protected choline to dairy cows, particularly as they transition from the dry period to early lactation. In light of new research it seems appropriate to initiate discussion on whether choline should be considered a required nutrient in dairy diets.

Transition Cow And Choline Biology

Several studies have shown 50 to 60% of transition cows experience moderate to severe fatty liver (Bobe et al., 2004). These studies have been conducted in numerous countries across different genetic lines of cattle, different feedstuffs, and varying management systems and the data were not generated from a population of problem cows or herds . The consistency amongst these studies suggests that development of fatty liver is a "normal" part of the cow's biology. Because fatty liver is a classic deficiency symptom for choline, it is reasonable to question if transition cows are typically deficient in choline.

At calving there are hormonal changes that trigger an intense period of lipid mobilization from adipose tissue and as a result, blood nonesterified fatty acid (NEFA) concentrations typically increase 5- to 10-fold (Grummer, 1993). NEFA remain elevated, although to a lesser extent, during early lactation when cows experience negative energy balance. Blood flow to the liver doubles as a cow transitions from the dry period to lactation (Reynolds et al., 2003). NEFA concentration and blood flow are the two biggest factors affecting how much NEFA is taken up

by the liver. As a result, daily fatty acid uptake by the liver increases and estimated 13-fold at calving, from approximately 100 to 1300 g/day (Overton, unpublished). Not all of the fatty acids taken up by the liver will be stored and contribute to fatty liver. However, Drackely (2001) estimated that during peak blood NEFA concentration, approximately 600 g might be deposited in 24 hours, which would correspond to an increase in liver fat of 6-7% by weight. As a reference, fat above 5% in the liver (wet basis) is considered by the veterinary community to be moderate to severe fatty liver. It is important to understand that this dramatic increase in NEFA uptake by the liver is part of the normal biology of transition cows and is not restricted to fat cows, poorly fed cows, or cows housed in suboptimal environments.

The most desirable fate of fatty acids entering the liver would be complete oxidation to provide energy to the liver or reesterification and export as triglyceride from the liver as part of a very low density lipoprotein (VLDL). Hepatic oxidation increases approximately 20% during the transition period (Drackley et al., 2001). This increase does not represent a strategic move by the cow's liver to cope with the sudden surge of NEFA uptake at calving. It occurs because the liver becomes metabolically more active. Unfortunately, the increase in oxidation is not sufficient to cope with the increased load of fatty acid being presented to the liver. Research conducted nearly 25 years ago at the University of Wisconsin (Kleppe et al., 1988) and Michigan State University (Pullen et al., 1990) revealed that ruminants have a low capacity to export triglyceride from the liver as very low density lipoprotein (VLDL) as compared to nonruminants. This and the inability to markedly increase fatty acid oxidation is why transition dairy cattle develop fatty liver when experiencing elevated blood NEFA.

It is now apparent that choline deficiency is a limiting factor for VLDL triglyceride export from the liver. It has been shown in many species, using a wide variety of experimental approaches, that rate of VLDL export is highly related to the rate of hepatic PC synthesis (Cole et al., 2012). Models include monograstrics fed choline deficient diets, isolated hepatocytes cultured in choline and methionine deficient media, and in knock out mice for genes involved in PC synthesis (Cole et al., 2012).
Interestingly, there is no evidence that synthesis of any other phospholipid is required for hepatic VLDL assembly and secretion. In addition to direct PC synthesis from dietary choline, there is endogenous hepatic synthesis of PC via methylation of phosphotidylethanolamine (PE). Sharma and Erdman (1988) demonstrated dietary choline is extensively degraded in the rumen of dairy cows and very little is available to the small intestine for absorption. Choline flow to the duodenum increased less than 2 g/day, even when free choline intake was increased to more than 300 g/d. Therefore, ruminants are more highly dependent than nonruminants on endogenous synthesis of PC from PE. Is endogenous synthesis of PC from PE sufficient during the transition period or do cows require choline supplementation? The high proportion of transition cows developing moderate to severe fatty liver during the transition period suggests that endogenous synthesis is not sufficient in many cows.

Evidence for a Choline Deficiency in Transition Dairy Dows

The first piece of evidence that transition cows are deficient in choline is the development of fatty liver during the periparturient period (Grummer, 1993; Bobe et al., 2004). More compelling evidence is the alleviation of fatty liver when supplying cows with choline that is protected from ruminal degradation (Cooke et al., 2007; Zom et al., 2011). Dutch researchers (Goselink et al., 2013) recently demonstrated greater gene expression for microsomal triglyceride transfer protein (MTTP) in liver of transition cows supplemented with rumen-protected choline (RPC). MTTP is an important protein required for hepatic VLDL synthesis. Recently, it was shown that choline, but not methionine, increases VLDL secretion from primary bovine (McCourt et al., 2015). This provided solid evidence that choline limitation is a causative factor for inadequate fat export out of the liver.

The reduction in liver fat content when feeding transition cows RPC is accompanied by improved health and production. Lima et al. (2012) observed reduced incidences of clinical ketosis, mastitis, and morbidity when feeding RPC from 25 days prepartum to 80 days postpartum. It has been known for years that elevated fat in the liver is associated with poor reproductive performance (Bobe et al., 2004). First service conception rate was increased by feeding RPC in one study (Oelrichs et al., 2004) but not another (Lima et al., 2012). I (Grummer, 2012) completed a meta-analysis for 13 studies that fed RPC to transition cows. Feed stability or evidence of bioavailability of choline source was not a criterion for study selection. Studies were not screened for "soundness" of research. Treatment means and sample size (standard error of the mean) had to be available for the analysis. Ten of the thirteen trials were published in peer-reviewed journals. For studies to be included in this analysis, RPC had to be fed ***prior*** to calving. Time when RPC supplementation was started varied between 28 to 7 days prior to expected calving. RPC supplementation was terminated anywhere from the day of calving (one study) to 120 days in milk. Response variables included DMI, milk yield, energy corrected milk yield, fat %, protein %, and fat and protein yield. Insufficient data was available for analysis of liver fat or energy-related blood parameters. Analysis revealed a significant increase of 4.9 lb milk/day and 1.6 lb of dry matter intake/day (Table 1). Milk fat and protein percentage were not significantly affected by treatment but yields were (Table 1). These studies were conducted in several countries under a variety of management conditions and they did not target problem herds or cows. This implies that benefits to supplementing protected choline can be realized by a wide variety of herds. Alleviating a choline deficiency not only reduces liver fat but also improves parameters that are economically important to dairy producers.

Table 1. A Meta-analysis of 13 studies examining the effects of feeding RPC to transition cows on dry matter intake and milk.

	Control	RPC	SEd	P =
DMI, lb/d	39.98	41.60	.46	.0042
Milk, lb/d	70.88	75.75	.75	<.0001
ECM, lb/d	76.87	82.78	1.33	.0038
Fat yield, lb/d	2.788	3.042	.086	.021
Protein yield, lb/d	2.300	2.467	.053	.010

Can Protected Methionine Substitute For Protected Choline?

Protected methionine has often been suggested as a possible alternative to protected choline for supplementation to transition dairy cows. Methionine and choline both serve as methyl donors. Methionine methyl groups can be used for endogenous synthesis of PC from PE. As an amino acid, methionine is needed for the synthesis of apolipoproteins. Therefore, there is a conceptual basis for methionine substitution for choline. Six feeding trials have been conducted to examine the effects of rumen-protected methionine or methionine analog on liver total lipid or triglyceride content and none of them showed a reduction (Socha, 1994; Bertics et al., 1997; Piepenbrink et al., 2004; Preynat et al., 2010; Osorio et al., 2013; Zhou et al., 2016).

The reason for methionine's failure to prevent fatty liver in transition cows is not known. One explanation may be that the studies cited above employed insufficient doses of protected methionine or methionine analog. Choline contains three methyl groups while methionine only contains one methyl group. When differences in molecular weight between choline and methionine are accounted for, choline by weight is 4.3 times more "potent" than methionine as a methyl donor. Therefore, assuming equal bioavailability of the rumen-protected products being fed, one could speculate that one would need to feed 64.5 g/d of methionine during the transition period to obtain a similar amount of methyl groups as when feeding 15 g/d of choline. As previously mentioned, choline, but not methionine, increases VLDL secretion from primary bovine hepatocytes (McCourt et al., 2015).

Conclusions

Since the last NRC (2001) publication, a significant body of evidence has accumulated to support choline being a required but limiting nutrient in transition cow diets. There is overwhelming evidence that feeding transition dairy cows 15 g choline/day in a form that is protected from ruminal degradation will alleviate choline's classic deficiency symptom and lead to improvements in health and performance.

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Hepatic methyl metabolism: influencing success during the transition to lactation

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SUMMARY

- Adipose tissue mobilization during negative energy balance results in increased hepatic NEFA uptake.
- NEFA can be completely oxidized to energy, incompletely oxidized to ketones, or esterified to triglycerides for storage or export as VLDL.
- VLDL export from ruminant livers is limited, primarily because of limited phosphatidylcholine.
- Use of a cell culture models confirms that increasing choline concentrations can increase VLDL export from hepatocytes. Increasing choline concentrations also tended to reduce oxidative stress associated with a fatty acid challenge.
- Choline can be used to donate a methyl group for methionine regeneration and may have supported gluconeogenesis in hepatocytes.
- Increasing concentrations of methionine decreased the need for endogenous regeneration of methionine and may have supported fatty acid oxidation within hepatocytes. Increasing concentrations of methionine did not change VLDL export or oxidative stress.
- The lack of interaction between methionine and choline in cell culture models supports separate mechanistic roles for methionine and choline within the hepatocyte.

INTRODUCTION

The transition to lactation period is characterized by negative energy balance (NEB) which reflects decreased feed intake and increased energy and glucose demands associated with lactation. During NEB, stored body fat is mobilized in an attempt to compensate for the energy deficit and transported to the liver in the form of nonesterified fatty acids (NEFA) and glycerol. While the mobilized NEFA provide critical fuel sources during the transition to lactation period, inability of the liver to metabolize them can lead to ketosis and fatty liver which have negative effects on productivity and animal health.

HEPATIC UPTAKE AND METABOLISM OF NEFA

During periods of NEB, triglycerides (TG) are mobilized from adipose stores and are transported to the liver to aid in alleviating NEB (Dole, 1956; Gordon and Cherkes, 1956). Hepatic update of NEFA is reflective of blood flow and blood NEFA concentration, both of which are increased after

calving. It has been well characterized that blood NEFA concentration increases after calving, reflective of adipose tissue mobilization, and can increase to 1 mmol/L or greater (Grummer et al., 1993; Reynolds et al., 2003). Additionally, blood flow nearly doubles from one week precalving to one and a half weeks postcalving (Reynolds et al., 2003), increasing exposure of the liver to nutrients and metabolites, including NEFA. Glycerol can be used as a gluconeogenic precursor after hepatic uptake. Conversely, NEFA are β-oxidized to acetyl-CoA units with four possible fates: complete oxidation through the TCA cycle, incomplete oxidation through ketogenesis to ketones, TG synthesis and packaging as very-low density lipoprotein for export from the liver (minimal in ruminant animals), or TG synthesis for storage as liver lipids (reviewed by Grummer, 1993). When available acetyl-CoA exceeds the capacity of the TCA cycle, there are increases in production of ketones and deposition of TG, leading to the onset of ketosis and fatty liver syndrome (White, 2015). The progression of these disorders is the response to poor adaptation to the challenges associated with the transition to lactation period.

During this period of NEFA mobilization, the capacity of the liver to completely oxidize fatty acids to energy is only limitedly increased (Grum et al., 1996) and thus, more acetyl-CoA are metabolized through the alternative pathways including ketogenesis and synthesis of TG for storage or export. Capacity of the liver to synthesize TG from acetyl-CoA is increased by 188% at +1 vs. -21 days relative to calving, highlighting the capacity of the liver to store fatty acids that cannot be immediately oxidized (Grum et al., 1996). Accumulation of liver lipids during early lactation can be as high as 500 g/d and it is predicted that 60% of dairy cows have severe or clinical fatty liver, defined as a liver lipid content greater than 10% on wet weight basis (Drackley, 1999; Bobe et al., 2004).

VLDL EXPORT

Just as in nonruminants, export of very low density lipoproteins (VLDL) can prevent accumulation of fat within the liver and can allow for transport of lipid fuel sources to other tissues, including the mammary gland. Although the capacity of the liver to synthesize TG is increased during the transition to lactation, the ability of the ruminant liver to export TG as VLDL is not proportionately high. Components of VLDL includes TG, apolipoproteins (ApoB and ApoE, specifically), cholesterol, and phosphatidylcholine and have been well studied in nonruminant models. Generation of phosphatidylcholine can either be de novo (methylation of phosphotidylethanolamine) or dietary (choline) and depletion of methyl donors from rodent diets significantly increases liver TG accumulation (Rinella et al., 2008; Cole et al., 2012). In ruminants, the component limiting VLDL export is phosphatidylcholine. Supplementation of dairy cows with rumen-protected choline reduces liver TG concentrations during the transition to lactation period (Zom et al., 2011; Goselink et al., 2013). Examination of genes involved in fatty acid transport and VLDL assembly are increased in cows supplemented with rumen-protected choline, suggesting that the decreased liver TG accumulation is due to increased VLDL export (Goselink et al., 2013). Less is know about the interaction of the two pathways to generate phosphatidylcholine in ruminants.

METHYL DONOR METABOLISM

Methyl donors, including choline, methionine, betaine, and folate, are essential for DNA methylation, prevention of oxidative stress, energy metabolism, and protein synthesis; however, because of rumen fermentation, lactating ruminants are deficient in methyl donors (Pinotti et al., 2002). While the role of methyl donors has been extensively studied in nonruminants, less is understood regarding their action and mechanism in ruminants. In order to elucidate the mechanism of methyl donor metabolism, a bovine primary hepatocyte cell culture model was used to examine the role of two methyl donors, choline and methionine, in hepatic metabolism. Cells were exposed to increasing doses of choline and methionine in the absence or presence of a fatty acid cocktail designed to mimic the profile of fatty acids in circulation at calving (Chandler et al., 2015).

Given that methionine is a required amino acid essential for body and milk protein synthesis, regeneration of methionine is a vital role of methyl donors within liver cells. Increasing concentrations of methionine decreased endogenous regeneration of methionine suggesting that endogenous methionine regeneration is a hepatic priority when methionine concentrations are low (Chandler et al., 2015). Increasing choline concentrations increased methionine regeneration, suggesting that choline may serve a role as a methyl donor for methionine regeneration (Chandler et al., 2015).

Examination of pathways that support fatty acid oxidation via TCA cycle oxidation, and glucose production via gluconeogenesis also indicated specific regulatory roles of choline and methionine within the liver cells. The balance of carbon flux through these two cycles is influenced by the activity of the rate limiting enzymes, pyruvate carboxylase (PC) and phosphoenolpyruvate carboxykinase (PEPCK). Supplementation with choline tended to increase both these genes, suggesting that more carbon may be used to generate glucose (Chandler et al., 2016). Conversely, supplementation with methionine increased PEPCK, without altering PC, which may suggest that more carbon were oxidized via the TCA cycle (Chandler et al., 2016).

Quantification of VLDL in ruminants is difficult due to differences in lipid profiles of the VLDL between ruminants and nonruminants. An antibody-based assay was validated and used to quantify VLDL secreted into the cell culture media in cells exposed to choline and methionine in the presence of a fatty acid challenge. Increasing choline concentrations increased VLDL export from the hepatocytes (McCourt et al., 2015). No change in VLDL export was observed as methionine concentrations were increased. This was supported by no differences in PEMT, the enzyme that catalyzes the methylation of phosphatidylethanolamine to phosphatidylcholine (Chandler et al., 2015).

Oxidation of fatty acids is critical for energy production in the liver; however, it also results in oxidative stress within the cells. Given this relationship, accumulation of reactive oxygen species (ROS) were examined in the cell culture model described above. Increasing concentrations of

choline, but not methionine, tended to decrease ROS released into the cell culture media (Chandler et al., 2015).

CONCLUSIONS

Negative energy balance and adipose tissue mobilization are well-characterized hallmarks of the transition to lactation period in dairy cows. The ability of the liver to metabolize NEFA and glycerol are not only essential to meeting the demands of lactation, but to avoiding metabolic disorders. Recent attention to methyl donors and their role in maintaining hepatic health and optimizing hepatic function has necessitated a better understanding of their mechanism in the liver. Use of cell culture models aid in understanding specific mechanisms and suggests a biological priority for methyl donor use. The lack of interaction between methionine and choline in cell culture models supports separate roles for methionine and choline within the hepatocyte. It is clear that the requirement for methionine needs to be met, either by dietary sources or by endogenous regeneration. Choline can provide methyl groups for regenerating methionine, but is also involved in increasing VLDL export and may decrease oxidative stress.

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Insulin Resistance in Transition Dairy Cows: Friend or Foe?

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Introduction

An alternative title for this presentation could be: Fat Reserves- How to Manage a Valuable Resource? Most transition dairy cows experience periods of intense fat mobilization. Mobilized fat supports lactation, but if not managed properly it may lead to metabolic disorders such as fatty liver and ketosis. At calving and in early lactation, hormonal changes are important in governing fat mobilization. Discussing all the endocrine changes in this presentation is not possible; therefore, the focus will be on Insulin. Insulin is a key hormone that regulates nutrient metabolism in dairy cows. It is known as an anabolic hormone because it signals to tissues that the nutritional state is favorable and nutrients can be stored. Results from these signals include increasing glucose storage in the liver (as glycogen) and stimulation of fat synthesis and storage in adipose tissue and inhibition of fat mobilization from adipose tissue.

During the transition period, cows become "insulin resistant". Simply defined, this means that insulin has less of an affect than normal. If insulin is less effective, it means that liver glucose storage is decreased and fat mobilization from adipose tissue is increased. It is important to note that insulin resistance is not an all or nothing proposition. The magnitude of resistance is a sliding scale, so the degree of fat mobilization can vary and does not only occur at a maximum rate or not at all. Mobilization of fat helps support lactation. If mobilization of fat is too extensive, metabolic disorders such as fatty liver and ketosis can result. This begs the question, is insulin resistance a friend or a foe?

Insulin resistance: A Friend

In one very important respect, insulin resistance is a "friend". Cows purposely undergo insulin resistance as a means to support pregnancy and lactation. It is a normal biological process and a classic example of homeorhesis. Homeorhesis is a term coined by Dale Bauman, Cornell University (Bauman and Currie, 1980), and is defined as: the orchestrated or coordinated control in metabolism of body tissues to support a physiological state. In the case of the transition cow, glucose uptake by insulin sensitive tissues (muscle, adipose tissue) is decreased. Therefore, extra glucose is available to be channeled to the fetus or mammary gland, which are insulin insensitive (not affected) tissues. Additionally, fatty acid mobilization is increased when the cow becomes more insulin resistant. Once lipolysis takes place and the nonesterified fatty acids (NEFA) enter the blood, they can be diverted to the growing fetus or mammary gland to serve as an energy source or as a precursor for milk fat synthesis. The process of becoming insulin resistant is very important, especially to just fresh cows because they typically can't consume enough feed to meet the nutrient needs of lactation. The coordinated shift of nutrients from reserves to the mammary gland is instrumental in getting the cow through the

transition period and to a period of time when feed intake can provide sufficient nutrients to support lactation.

Insulin Resistance: A Foe

It is my impression that for most people, insulin resistance has a negative connotation and is perceived as a deleterious event. Can it be a foe? A potential negative aspect of insulin resistance is *excessive* NEFA mobilization from adipose tissue. About one third of the NEFA mobilized from adipose tissue is taken up by the liver (Emery et al., 1992). Liver uptake of NEFA is influenced primarily by NEFA concentration in blood and blood flow to the liver. NEFA concentration in blood may increase 5-7 fold at calving and blood flow to the liver is increased two-fold (Reynolds et al., 2003). NEFA daily uptake by the liver can increase by as much as 10-15-fold as the cow transitions from the dry period to lactation (Overton, unpublished). This dramatic increase presents a tremendous challenge to the liver. Ideally, these extra fatty acids will either be completely oxidize to CO_2 to provide energy to the liver or they will be exported as part of a very low density lipoprotein and be available to the mammary gland as an energy source or precursor for milk fat synthesis. If the capacity to utilize the fatty acid for those purposes is exceeded, then the fatty acids may be stored in the liver as triglyceride or be converted to ketones. In other words, the cow may experience fatty liver and subclinical or clinical ketosis. Additionally, elevated NEFA concentrations have been linked to depressed feed intake, suppressed immune function, and decreased risk of pregnancy and other maladies of transition cows. So yes, insulin resistance can be a foe.

At what point does insulin resistance become a foe? Unfortunately, this is a very difficult question to answer. Researchers have tried to measure blood NEFA or beta-hydroxybutyrate (BHBA, a ketone) or liver triglyceride and correlate it to production, health or reproduction (e.g. Chapinal et al., 2012). The goal of this research is to find "cut-off levels" or concentrations that are predictive of when herd performance or health is at risk of being impaired. Liver triglyceride is not a practical measure because it requires a liver biopsy, which is too invasive. However, blood measures such as NEFA or BHBA can be useful tools. Suffice it to say, interpretation of these tests can be very tricky. For example, do "one size fits all" cut offs from large epidemiological studies apply to all herds?

Is Reducing NEFA (Reducing Insulin Resistance) Always Good?

The following are three examples that argue that reducing NEFA (reducing insulin resistance) may not always be beneficial. Example 1. Genetically superior cows for milk production have higher blood NEFA and BHBA concentrations during the first 3 weeks postpartum (Harrison et al., 1990). This occurs because milk production increases faster relative to feed intake in genetically superior cows compared to cows with lower genetic potential to produce milk. Evidence also indicates that genetically superior cows my experience greater insulin resistance (Chagas et al., 2009). Example 2. There is compelling evidence that overfeeding energy to cows during the dry period leads to lower liver triglyceride and blood NEFA and BHBA concentrations (Janovick et al., 2011, Richards, 2011, Mann et al., 2015). This evidence has resulted in the promotion of feeding "controlled energy" diets in which dry cows are fed to meet energy requirements during the dry period. It has been hypothesized that overfeeding energy creates cows that are similar to human type 2 diabetics and have increased insulin resistance.

However, in some but not all studies, feeding a controlled energy diet led to a reduction in milk production, milk fat percentage, or energy corrected milk production compared to cows overfed energy (e.g. Janovick et al., 2011). This probably occurs because these cows mobilize less fat to support lactation. Example 3. Niacin, if fed in a form that protects it from degradation in the rumen, affects adipose tissue directly and suppresses fat mobilization. Consequently, blood NEFA and liver triglyceride is reduced (Yuan et al., 2012). Once again, the reduction in NEFA corresponded to nearly a 20 lb/d reduction in energy-corrected milk during the first week postpartum.

Strategies for Managing Insulin Resistance and Fat Mobilization

On one hand, we desire insulin resistance and fat mobilization to support lactation; on the other hand it may potentially compromise liver health and function. How can nutritionists balance the act?? Historically, the main strategy has been to reduce lipid mobilization during the transition period (Figure 1). Options include feeding controlled energy diets during the dry period (Janovick et al., 2011), feeding protected niacin (Yuan et al., 2012), shortening the dry period (Rastani et al., 2005), and drenching with propylene glycol (Studer et al. 1993). As previously discussed these strategies risk a loss of milk or milk fat yield. John Newbold stated it very nicely in the proceedings from the Nottingham Nutrition Conference (2005): *"Nutritional restriction to adipose tissue mobilisation might be necessary, but there is a philosophical problem. We have selected cows that have increased reliance on mobilised body reserves as a source of nutrients for milk production. The farmer has paid the geneticist for this- are we now going to ask him to pay the nutritionist to work in the opposite direction? We have our priorities wrong. We should explore what can be done to help the liver deal with mobilised fatty acids before considering whether we need to try to reduce the amount of fatty acid supplied to the liver."* Feeding rumen-protected choline is the only proven strategy to assist the liver during times of elevated NEFA. Choline in feedstuffs is degraded in the rumen, therefore insufficient quantities are absorbed from the intestine. Fatty liver is a symptom of choline deficiency. Choline is required for phosphatidylcholine synthesis, which in turn is required for VLDL assembly and fat export from the liver. Feeding rumen protected choline to transition cows reduces severity of fatty liver and ketosis (Lima et al., 2012) and increases milk production and energy-corrected milk production (Grummer et al., 2012).

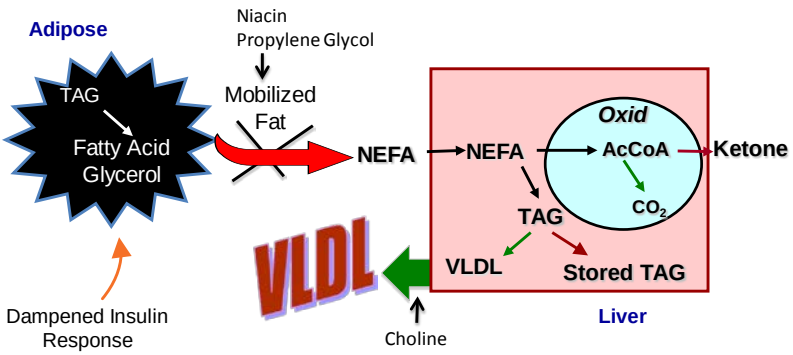

Figure 1. Strategies to manage insulin resistance: Agents such as niacin and propylene glycol reduce fat mobilization; choline enhances triglyceride (TAG) export from the liver as part of very low density lipoproteins (VLDL). NEFA= nonesterified fatty acids, AcCoA=acetylCoA.

This article should raise many questions. Are elevated NEFA or BHBA always bad and do one size fits all "cut off" values for alarm levels in blood serve us well? May alarm levels vary depending on the herd's genetic potential for milk production? What do you tell a dairy producer if "too many" cows are testing above cut-offs for blood NEFA or BHBA but the cows are milking like crazy? How will you manage fat mobilization? Perhaps a combination of antilipolytic compounds prepartum (e.g. feeding rumen-protected niacin or drenching propylene glycol) and rumen-protected choline pre- and postpartum to enhance liver fat export to the mammary gland may be most effective? By feeding antilipolytic compounds prepartum, the surge in blood NEFA that occurs at calving may be reduced (Yuan et al., 2012). By feeding rumen-protected choline prepartum, the liver will be able to process the fatty acids mobilized during the surge at calving. By feeding postpartum, choline will facilitate transfer of fatty acids from adipose tissue to the mammary gland during negative energy balance.

Bottom line: Insulin resistance and mobilization of fat reserves as NEFA are essential for cows to successfully transition from the dry period to lactation. Nutritional tools are available to manage insulin resistance so that there is a greater chance that it is a friend rather than a foe.

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GOLDILOCKS MEETS CHOLINE, TOO HOT? TOO COLD? JUST RIGHT?

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Background. Choline has been identified as a required nutrient for many species including humans, chicks, and pigs. Choline is found in low concentrations in most feeds, ranging from 0.04% in corn silage and alfalfa hay to 0.3% in protein sources such as soybean meal and cottonseed meal (DM basis). Its low concentrations in feeds are indicative of the low amounts required by livestock (e.g. 3 g/day for a lactating sow). Although the bovine requirement for choline has yet to be established, the supplementation of choline to dairy cows in transition usually improves milk production and often aids in the reduction of fat in the liver. However, in order for choline to be absorbed in the small intestine of ruminants, the choline must have some protection from degradation by ruminal microbes which degrade dietary choline to methane and acetic acid. Several ruminally-protected choline (**RPC**) products are being marketed commercially to the dairy industry across the world. ReaShure (Balchem Corp., New Hampton, NY) is such a product containing approximately 25% choline chloride. In 16 experiments published since 2003, dairy cows supplemented with an RPC product starting in late gestation (~ 3 weeks prepartum) produced an average of 4.4 lb/day more milk or fat-corrected milk compared with cows not supplemented with RPC. Ten of the 16 studies reported a significant increase in milk due to RPC supplementation. This increase in milk due to choline supplementation often has been explained through choline's role to improve lipid metabolism by the liver. The liver of the modern dairy cow accumulates fat (triacylglycerol, **TAG**) in the early weeks after calving because of the massive mobilization of adipose tissue for energy use during the extensive period of negative energy balance (**NEB**). The efficiency of the excessively fat liver to manufacture glucose for milk synthesis is compromised resulting in reduced milk yield. The liver can export some of the fat with the aid of choline. Removing choline from the diet is a way that researchers often create fatty liver in nonruminant species. When animals are pregnant or lactating, concentrations of choline in the liver decrease dramatically (Zeisel, 2000).

Proper body condition at calving is important for optimal milk yield. Thin cows lack the energy reserves to support needed energy for milk synthesis during the inevitable negative energy state whereas fat cows are often poor eaters and experience greater states of negative energy postpartum resulting in even greater fatty liver, reduced milk yield, and poor reproductive performance. Excessive fat reserves are oftentimes hidden from view; fat that is stored in the abdomen around the intestines and the kidneys is not considered when one is condition-scoring cows. Excessive energy intake during the dry period can build up this abdominal (visceral) fat without changing the overall body condition of the cow (Drackley et al., 2014). Nonlactating and nonpregnant Holstein cows were fed a lower energy diet (0.61 Mcal/lb) of 41% wheat straw and 28% corn silage or a higher energy diet (0.735 Mcal/lb) of 0% wheat straw and 50% corn silage (DM basis) in ad libitum amounts. After 8 weeks on the 2 diets, body condition was the same, 3.47 vs. 3.52, respectively. Upon slaughter, it was discovered that the cows fed the lower energy diet had 56 fewer pounds of abdominal fat (70 vs. 126 lb). Feeding

prepartum diets that better match the energy requirement for maintenance and pregnancy is the "just right" (a.k.a. the "Goldilocks") approach. Eating the porridge at the right temperature, sitting in the chair that is the right height, and sleeping in the bed that is the most comfortable is not extreme to either side. Likewise, underfeeding or overfeeding energy to pregnant cows during the whole dry period ends up damaging cow performance postpartum. In the last 2 decades of research in this area of transition cow feeding, formulating well-balanced diets containing substantial proportions of low quality forages such as wheat straw has resulted in better health of the postpartum cow (Drackley, 2016). Too often, cows are overfed during the dry period because a dairy will feed the TMR refused by the lactating cows. This approach may not appear to be harmful because body condition appears "normal" but research indicates that dangers lurk like the fury of a moma bear.

Experimental hypothesis and approach. Because overfeeding energy during the dry period often leads to fatty liver and because choline plays a key role in improving the liver's management of fat, it was hypothesized that choline supplementation would most benefit those dairy cows overfed energy during the dry period.

Ninety-six pregnant, nonlactating multiparous Holstein cows (University of Florida) were assigned to 1 of 4 dietary treatments on the day of 'dry off' (~7 weeks prior to expected calving date). Dietary treatments were arranged in a 2 × 2 factorial. One factor was RPC (ReaShure, Balchem Corp., New Hampton, NY) top-dressed once daily at 0 or 60 g/day per cow from 21 days prior to expected calving date through 21 days postpartum. The second factor was diets of 0.74 (excess energy) or 0.64 (maintenance energy) Mcal of NE_L/lb of dietary DM fed in ad libitum amounts for the whole dry period. Therefore, the 4 treatments were maintenance energy intake without RPC (**MNE**) or with RPC (**MNE+C**) and excess energy intake without RPC (**EXE**) or with RPC (**EXE+C**). Chopped wheat straw (< 2 inches), corn silage, and triticale silage were adjusted to formulate to the targeted energy density of the prepartum diets. Wet brew was added to the TMR (16.7% of dietary DM) to minimize sorting by the cows managed in a Calan gate system. At the time of enrollment, parity (1.9), 305-day mature equivalent milk production (26,701 lb), body condition score (3.55), or body weight (1622 lb) did not differ between the 4 groups of cows. After calving all cows were fed the same basal diet (0.76 Mcal NE_L per lb and 16.0% CP, DM basis) through 15 weeks postpartum when the trial ended. Diets were formulated to have methionine at 2.3% of metabolizable protein and a lysine-to-methionine ratio of 2.9 prepartum and 3.1 postpartum. Measurements taken included intake of feed, body weight and condition, yield and IgG content of colostrum, health disorders, milk production and composition, triacylglycerol content of liver via biopsy, uterine health assessments, selected metabolites and immune responses in blood, and pregnancy to timed artificial insemination.

Data were analyzed using the MIXED procedure of SAS version 9.4 (SAS/STAT, SAS Institute Inc., Cary, NC). The REPEATED statement was used for dependent variables measured over time. Models included the fixed effects of energy intake prepartum (excess vs. maintenance), RPC (with vs. without), interaction between energy intake prepartum and RPC, day or week of measurement, and all 2- and 3-way interactions. Cow was nested within treatment and was the

error term for testing the effects of treatment. Data were transformed to achieve normality if needed before analyses. Binary data were analyzed by logistic regression using the GLIMMIX procedure of SAS. Time to event such as interval to pregnancy by 210 DIM was analyzed with Cox's proportional hazard regression model using the PHREG procedure of SAS. Statistical significance was considered at $P \leq 0.05$ and tendency was considered at $0.05 < P \leq 0.10$.

Experimental results and discussion. For nearly every dependent variable, the influence of each main treatment effect was independent. That is, the effect of choline was the same if the cow was fed the lower energy diet or the greater energy diet prepartum. Likewise, the effect of prepartum energy intake was the same regardless of whether the cow was supplemented with choline. Therefore, the main effects of prepartum energy intake and choline will be presented separately.

Effects of prepartum energy intake.

Prepartum responses. Body condition score from dry-off to calving was unchanged. Mean DM intake during the last 15 days of gestation (mean of 23.7 lb/day) did not differ due to energy density of the diets. However intake of energy did differ between the 2 groups as planned. Two weeks prior to calving, cows fed the EXE diet were consuming energy at 140% of their requirement for maintenance and pregnancy whereas cows fed the MNE diet were eating at 110% of their requirement (NRC, 2001). The pattern of NE_L intake over the last 2 weeks of gestation also differed [$P < 0.01$, energy diet by day interaction (Figure 1)]. As reported by many others, intake of energy decreased as parturition approached. However, the intake of NE_L by cows fed the EXE diet decreased at twice the rate compared to that by cows fed the MNE diet, dropping the equivalent to 0.6 vs. 0.3 lb per day or a total of 9 (34%) and 4.5 lb (20%), respectively. As a result of the greater NE_L intake prepartum, mean concentration of nonesterified fatty acids (**NEFA**) tended to be lower (252 vs. 295 µEq/mL, $P < 0.10$) and that of glucose was greater (66.4 vs. 63.5 mg/100 mL, $P < 0.05$) in plasma of cows fed the EXE compared with the MNE diet although values were within the normal range for well-managed prepartum dairy cows.

Postpartum responses. Cows fed the EXE diet prepartum consumed 2.7 lb less feed DM ($P < 0.01$) during the 15-week postpartum period compared with cows fed the MNE diet (50.4 vs. 53.1 lb/day, respectively). This response is rarely significant although numerically lower postpartum DM intake by cows overfed energy prepartum has been reported previously (Holcomb et al., 2001; Dann et al., 2006; Zhang et al., 2015). However, mean production of milk over the first 15 weeks postpartum was not different (91.9 vs. 95.1 lb/day of uncorrected milk yield [$P = 0.25$] and 93.9 vs. 96.2 lb/day of energy-corrected milk yield [$P = 0.38$] for cows consuming EXE and MNE diets, respectively). Concentration of fat (3.88 vs. 3.78%) and true protein (2.95 vs. 2.97%) in milk were not affected by prepartum energy intake. The gross efficiency of converting feed DM into ECM almost reached a significant tendency favoring cows fed the EXE diet (1.90 vs. 1.84 lb of milk per lb of feed intake, $P = 0.11$). This improved gross efficiency of milk from feed came at the cost of body reserves. After body weight of both groups of cows hit a low after 4 weeks of lactation, cows from the EXE treatment simply maintained their body weight the rest of the way whereas cows from the MNE treatment

started gaining weight until they put on ~70 lb at 15 weeks postpartum. This greater reliance on body reserves for the milk that was produced by cows fed EXE diets prepartum is reflected in greater mean concentrations of circulating beta-hydroxybutyric acid (**BHBA**; 0.52 vs. 0.43 mmol/L, $P < 0.05$) and NEFA (502 vs. 453 µEq/mL, $P < 0.10$). As a result of greater fat circulating in the blood, the liver of cows fed EXE diets accumulated more TAG fat at 7 (11.1 vs. 8.7% of DM) and 21 (10.1 vs. 7.6% of DM) days in milk compared with cows fed MNE diets prepartum.

Fatty liver is often associated with ketosis and reduced reproductive performance. Incidence of health disorders were recorded although the study lacked sufficient numbers of cows to adequately test the effect of prepartum energy intake. Incidence of diseases/disorders that reached a probability of significance of ≤ 0.20 due to feeding EXE diets included ketosis (16.9 vs. 10.2%) and uterine infection at 40 days in milk (15.2 vs. 7.1%). However excess energy intake prepartum did not influence ovarian cyclicity postpartum either at 26 (45.1 vs. 60.6%) or at 40 (78.7 vs. 82.5%) days in milk compared to cows fed MNE diets as determined by the presence of a corpus luteum detected using ultrasonography. Pregnancy at first AI was 32% for both treatment groups.

<u>Effects of choline supplementation.</u>

Prepartum responses. Although pregnant cows began RPC supplementation at 21 days prior to expected calving date, cows consumed supplemental RPC for only the last 17 days of gestation on average because they calved earlier than expected. Supplementing RPC did not change mean DM intake during the last 15 days (23.1 vs. 24.2 lb DM/day for –RPC and +RPC-fed cows, respectively). Body condition score of cows averaged 3.51 and did not differ due to RPC feeding. Blood concentrations of NEFA and BHBA also were unaffected by RPC supplementation.

Postpartum responses. Yield of colostrum was not affected by RPC supplementation (18.8 vs. 21.8 lb) but colostrum from cows fed RPC had a greater concentration of immunoglobulin G (**IgG**; 78 vs. 57 g of IgG/L). The source of colostrum that was fed to the calves born from the cows on this study was not controlled. Nevertheless, the growth of the calves over the following 12 months of life was affected by being exposed to RPC in utero. Calves born to dams supplemented with RPC tended to be 4.6 lb lighter at birth (84.5 vs. 89.2 lb, $P < 0.10$) but were 31 lb heavier at 12 months of age (739 vs. 7089 lb, $P < 0.05$) thus growing at 0.1 lb/day faster compared to calves born from unsupplemented dams (1.97 vs. 1.87 lb/day). Apart from the colostrum, all calves were managed the same during this time period. Choline has been helpful in the diet of nonruminant animals during pregnancy to improve offspring performance (Newberme et al., 1970; Zeisel, 2006). Whether this may be true for dairy calves should be investigated in the future.

As occurred in the prepartum period, intake of feed DM postpartum was not affected by RPC supplementation (52.3 vs. 51.1 lb/day) although the 1.2 lb/day numerical increase due to RPC supplementation is the same increase as that reported by Grummer (2012) using a meta-analysis of RPC-feeding studies with lactating dairy cows. Cows supplemented with RPC tended ($P < 0.10$) to produce more milk during the first 15 weeks of lactation (95.9 vs. 91.0 lb/day). This

tendency for increased milk yield detected during the first 15 weeks continued for 40 weeks of lactation (81.7 vs. 77.1 lb/day, $P < 0.10$; Figure 2). Holstein cows produced nearly 5 more pounds per day of milk for 40 weeks of lactation when supplemented with 15 g of choline chloride for approximately 5.5 weeks over the transition period. This milk increase is similar to that reported by Elek et al. (2008), Janovick et al. (2006), and Lima et al. (2012) and to that reported in the meta-analysis by Grummer (2012). Although concentration of fat (3.82 vs. 3.84%) and true protein (2.95 vs. 2.97%) in milk were not affected by RPC, the yield of both fat and protein tended to be greater by cows fed RPC due to their tendency for greater milk yield. Greater milk yield without a significant increase in feed intake resulted in a greater mean NEB of cows fed RPC over the 15 weeks (-1.18 vs. -0.53 Mcal/day). The pattern of NEB over the 15 weeks postpartum also differed between groups. Cows fed RPC were experiencing a more NEB in weeks 2 (-11.4 vs. -8.9 Mcal/day) and 3 (-8.7 vs. -6.6 Mcal/day) postpartum. No difference in energy balance occurred between groups after cows moved past week 6 (RPC by week interaction, $P < 0.10$). Despite a greater NEB, loss of body weight from calving to week 4 postpartum was not different (101 vs. 83 lb). In addition, mean concentrations of NEFA and BHBA in blood were not affected.

Treatment for ketosis was the only disease/disorder that reached a probability of significance of ≤ 0.20 due to feeding RPC (18 vs. 9% for +RPC vs. –RPC, respectively). Diagnosis of ketosis was based upon ketostix classification of urine BHBA as 'moderate' (~40 mg/100 mL) or 'large' (80 mg/100 mL). In a field study using more cows (n = 369), primiparous and multiparous cows were fed 15 g/d of RPC from 25 days prepartum to 80 days postpartum (Lima et al., 2012). Yield of fat-corrected milk increased 4 lb/day (98.3 vs. 94.3 lb/day) due to RPC feeding. Cows fed RPC had less morbidity, especially less clinical ketosis (4.7 vs. 13.9% for primiparous cows and 3.5 vs. 9.8% for multiparous cows). Other measures that are indicators of cow health suggest a positive influence of RPC in the current study. Rectal temperature measured at 4, 7, and 12 days in milk decreased linearly from 101.8 to 101.2°F for RPC-supplemented cows whereas that for –RPC cows increased linearly from 101.6 to 101.9°F. A concentration of < 8.5 mg of total Ca/100 mL of blood plasma was used as a definition of subclinical milk fever in blood samples collected at 0, 1, 3, and 7 days in milk (Chapinal et al., 2012; Martinez et al., 2012). Cows fed RPC had greater mean concentrations of Ca across measurement days (8.72 vs. 8.46 mg/100 mL) and the prevalence of subclinical milk fever (using any of the 4 days of measurement) was reduced ($P < 0.05$) from 52.1 to 31.6%.

The pattern and the mean concentration of TAG in liver over 7, 14, and 21 days in milk was not affected by RPC supplementation (8.2 vs. 7.4% TAG DM basis for +RPC and –RPC, respectively). This lack of effect of RPC on liver TAG is in agreement with Zahra et al. (2006) and Piepenbrink and Overton (2003). However several studies have reported reduced TAG concentrations in the liver of lactating dairy cows in the early postpartum period including Elek et al. (2013), Santos and Lima (2009), and Zom et al. (2011). The TAG values in the current FL study were quite low and may have been less susceptible to TAG reduction by RPC.

The greater NEB of cows fed RPC did not influence the proportion of cows cycling at 26 and 40 days in milk based upon a detectable corpus luteum. However pregnancy at first insemination

tended to favor cows fed RPC (41.3 vs. 23.6%, *P* < 0.10) although the proportion of cows pregnant by 40 weeks postpartum did not differ (69.8 vs. 62.5%). In a study conducted at a commercial dairy in California using both primiparous and multiparous cows (Lima et al., 2012), pregnancy rate after the first and second insemination was numerically but not significantly better due to feeding RPC from 25 days pre-calving to 80 days post-calving (59.8 vs. 52.7%).

Summary. Supplementing ruminally protected choline chloride at 15 g/day from approximately 17 days prepartum to 21 days postpartum resulted in greater (P < 0.10) yield of milk (4.9 lb/day) and milk components through 40 weeks of lactation, greater NEB at 2 and 3 weeks postpartum without changing TAG in liver, greater concentration and yield of IgG in colostrum, greater pregnancy at first insemination, and better daily gains in body weight by calves from those dams regardless of the amount of energy consumed during the entire dry period. Compared with feeding to maintenance, overfeeding energy by 40% during the dry period resulted in a greater decrease in DM intake as day of calving approached, lower intake of DM (2.5 lb/day) with no change in milk yield, greater concentrations of fat in blood and liver, and greater delay in gaining body weight postpartum.

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Figure 1. Effect of Prepartum Diet on Energy Intake

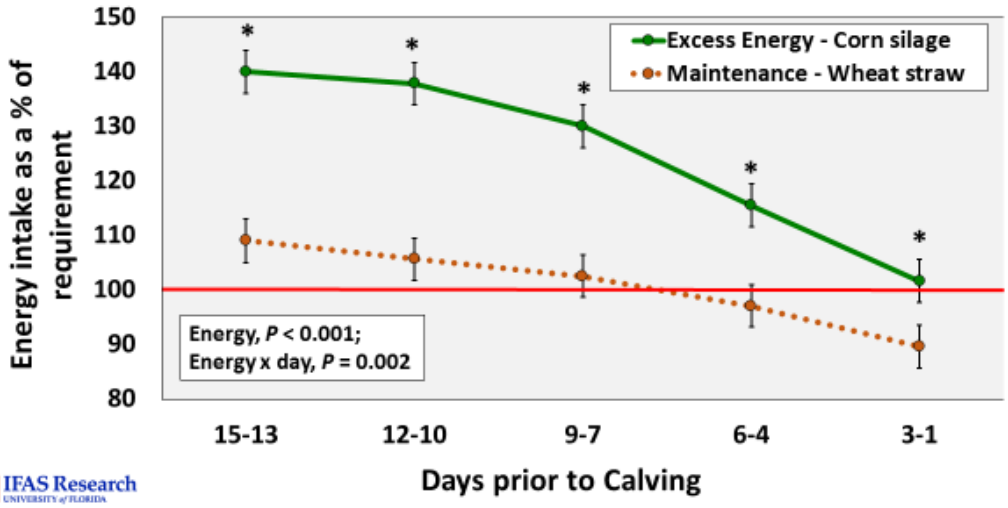
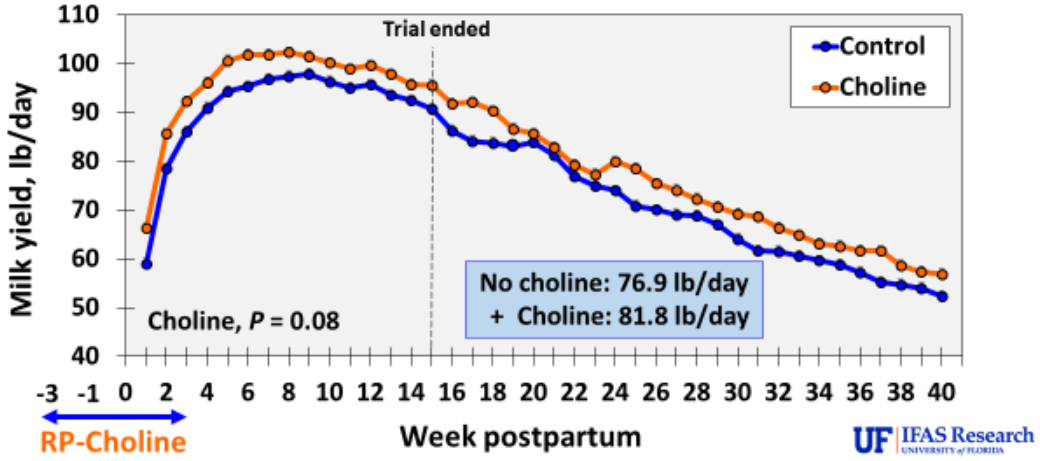


Figure 2. Positive Benefits of Ruminally-Protected Choline Continued After Supplementation Ceased – 40 Weeks



Transition cow nutrition and metabolic health.

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SUMMARY

- Supporting hepatic metabolic function is necessary to meet the energy and glucose demands of lactation.
- Supplementation with conjugated linoleic acid can marginally reduce milk fat synthesis and increase milk volume synthesis.
- Detecting and treating metabolic disorders is critical to enable transition cows to reach genetic milk production potentials. Many tools are available to detect hyperketonemia at the individual cow and herd level and can be integrated into management and nutrition strategies.
- Prefresh body condition score and body condition score changes across the transition to lactation period may increase postcalving blood BHBA.
- Previous lactation residual feed intake did not change hyperketonemia incidence or blood BHBA postcalving.

INTRODUCTION

The transition to lactation in the dairy cow lifecycle is a metabolically challenging period that reflects a coordinated response to calving and the onset of lactation. Central to a successful transition period is the ability of the liver to make enough glucose to support milk production, specifically during a period characterized by reduced feed intake. When nutrition and metabolism cannot keep up with the demands of milk production, cows fail to reach their genetic potential for milk production and develop metabolic disorders. Included in these metabolic disorders, ketosis effects 40 to 60% of early-lactation dairy cows and costs an average of $289 per case (McArt et al., 2012, 2014). Ketosis, sub-clinical or clinical, results in decreased milk production, increased risk of displaced abomasum, and decreased reproductive efficiency. Often developing alongside ketosis, fatty liver is associated with decreased milk production, decreased lifespan, increased veterinary costs, and longer calving intervals and affects 60% of dairy cows. Therefore, strategically feeding the transition dairy cow provides an opportunity to maximize milk production and improve metabolic health.

USE OF CONJUGATED LINOLEIC ACID

One strategy for sparing energy during the transition to lactation period is to marginally depress milk fat in order to spare energy from milk fat synthesis to milk volume synthesis. It is well documented that *trans*-10, *cis*-12 CLA is a potent inhibitor of milk fat synthesis and reduces milk fat yield up to 50% during supplementation; however, the depression is recovered shortly after

supplementation is ended (Baumgard et al., 2000; 2001; 2002; Peterson et al., 2002). Milk fat is the most energetically expensive component of milk, representing 50% of milk energy and can account for up to 35% of net energy intake in early lactation (Bauman & Currie, 1980; Kay et al., 2006). A decrease in milk fat excretion could spare energy for other uses such as milk production, production of other milk components, or body growth. In cases of early lactation CLA supplementation, reduction of milk fat content was accompanied by increased milk production (Bernal-Santos et al., 2003; Moallem et al., 2010; Schlegel et al., 2012). These studies were university research settings where nutrition and management were closely controlled.

In a recent study on a commercial dairy, primiparous and multiparous cows were supplemented with CLA during the transition to lactation period (Chandler et al., 2015c,d). Supplementation was provided for 21 days before calving (in the prefresh pen) to 30 days after calving for multiparous cows and 70 days after calving for primiparous cows (through automatic milk system robots). Cows supplemented with CLA had increased milk production both during the supplementation period and an increased milk production over 100 days in milk (Chandler et al., 2015a,b). Milk fat percent was decreased as expected; however, because milk volume also increased, milk fat yield was not altered. This study demonstrated that supplementation of CLA was effective at reducing milk fat concentration but concomitantly increased milk yield and therefore did not change fat yield in multiparous and primiparous animals in a commercial setting.

Overall, feeding rumen-protected CLA mixtures containing *trans*-10, *cis*-12 CLA may have beneficial applications, including strategies to increase milk yield, and maintain performance during the transition to lactation period. This study demonstrated that supplementation of CLA was effective at reducing milk fat concentration but concomitantly increased milk yield and therefore did not change fat yield in multiparous and primiparous animals in a commercial setting.

DETECTION OF HYPERKETONEMIA

A final strategy is reducing the risk for, and early detection and treatment of, metabolic diseases. The negative impacts of ketosis, specifically decreased milk production, can be ameliorated by early detection and treatment (McArt et al., 2014). Ketosis can be detected on farm with urine, milk, and blood testing; however, these methods vary in accuracy and can be both labor intensive and expensive. Milk fat:protein ratios have historically been used as an indicator of ketosis on farm; however, this ratio is only weakly correlated with ketosis incidence.

An alternative strategy is to monitor herd level ketosis prevalence and employ individual cow testing when prevalence exceeds the goal. Regression analysis of milk component data and cow information collected from herd data management software is used to predict herd prevalence of ketosis with 90% accuracy (Chandler et al., 2015b). This tool, the KetoMonitor (AgSource, Verona, WI), aids in management decisions by allowing for monitoring prevalence over time and across intentional and unintentional changes. Furthermore, using a herd-level prevalence tool can reduce unnecessary individual cow testing when herd prevalence is low.

NUTRITIONAL INFLUENCES ON HYPERKETONEMIA

A recent study examined the relationship between body condition score, residual feed intake (as a measure of feed efficiency), and hyperketonemia in transition cows that were sampled twice weekly postcalving. Animals with a BCS of 4 or greater prior to calving, or those that lost one ormore body condition unit over the transition period, were more likely to develop HYK, emphasizing the importance of avoiding over conditioning of cows in the dry period and excessive BCS loss throughout the transition period. There was no relationship between RFI and HYK which supports the continued selection for efficient cattle without the increased risk of HYK onset in subsequent lactations.

CONCLUSIONS

Together these strategies provide valuable tools to improve transition cow health and productivity. Understanding liver metabolism and the role of these strategies in improving liver health can guide nutritional decisions on commercial dairies.

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Precision Dairy Monitoring Technology Opportunities & Challenges

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Introduction

Technologies are changing the shape of the dairy industry across the globe. This rapid introduction of new technologies should come as no surprise given the technological culture shift in every facet of our society. In fact, many of the technologies applied to the dairy industry are variations of base technologies used in larger industries such as the automobile or personal electronic industries. Undoubtedly, these technologies will continue to change the way that dairy animals are managed. This technological shift provides reasons for optimism for improvements in both cow and farmer well-being moving forward. Many industry changes are setting the stage for the rapid introduction of new technologies in the dairy industry. Across the globe, the trend toward fewer, larger dairy operations continues. Dairy operations today are characterized by narrower profit margins than in the past, largely because of reduced governmental involvement in regulating agricultural commodity prices. Consequently, small changes in production or efficiency can have a major impact on profitability. The resulting competition growth has intensified the drive for efficiency resulting in increased emphasis on business and financial management. Furthermore, the decision making landscape for a dairy manager has changed dramatically with increased emphasis on consumer protection, continuous quality assurance, natural foods, pathogen-free food, zoonotic disease transmission, reduction of the use of medical treatments, and increased concern for the care of animals. Lastly, powers of human observation limit dairy producers' ability to identify sick or lame cows or cows in heat.

Precision Dairy Farming

Precision Dairy Farming is often used to describe many technologies aimed at improving dairy management systems. Bewley (2010) described Precision Dairy Farming as the use of technologies to measure physiological, behavioral, and production indicators on individual animals to improve management strategies and farm performance. Eastwood et al. (2004) defined Precision Dairy Farming as "the use of information technologies for assessment of fine-scale animal and physical resource variability aimed at improved management strategies for optimizing economic, social, and environmental farm performance." Spilke and Fahr (2003) stated that Precision Dairy Farming, with specific emphasis on technologies for individual

animal monitoring, "aims for an ecologically and economically sustainable production of milk with secured quality, as well as a high degree of consumer and animal protection." With Precision Dairy Farming, the trend toward group management may be reversed with focus returning to individual cows through the use of technologies (Schulze et al., 2007). Technologies included within Precision Dairy Farming range in complexity from daily milk yield recording to measurement of specific attributes (e.g. fat content or progesterone) within milk at each milking. The main objectives of Precision Dairy Farming are maximizing individual animal potential, early detection of disease, and minimizing the use of medication through preventive health measures. Precision Dairy Farming is inherently an interdisciplinary field incorporating concepts of informatics, biostatistics, ethology, economics, animal breeding, animal husbandry, animal nutrition, and engineering (Spilke and Fahr, 2003). The ideal Precision Dairy Farming technology explains an underlying biological process that can be translated into meaningful action with information readily available to the farmer and a reasonable return on investment. Additionally, the best technologies a flexible, robust, reliable and demonstrated effective through research and commercial demonstrations.

The list of Precision Dairy Farming technologies used for animal status monitoring and management continues to grow. Because of rapid development of new technologies and supporting applications, Precision Dairy Farming technologies are becoming more feasible. Many Precision Dairy Farming technologies including daily milk yield recording, milk component monitoring (e.g. fat, protein, and SCC), pedometers, automatic temperature recording devices, milk conductivity indicators, accelerometers for monitoring lying behavior, rumination monitors, automatic estrus detection monitors, and daily body weight measurements are already being utilized by dairy producers. Despite its seemingly simplistic nature, the power of accurate milk weights should not be discounted in monitoring cows, as it is typically the first factor that changes when a problem develops (Philpot, 2003). Other new Precision Dairy Farming technologies have been introduced to measure jaw movements, ruminal pH, reticular contractions, heart rate, animal positioning and activity, vaginal mucus electrical resistance, feeding behavior, biological components (enzymes, antibodies, or microorganisms), odor, glucose, acoustics, progesterone, individual milk components, color (as an indicator of cleanliness), infrared udder surface temperatures, gain analysis, and respiration rates. Unfortunately, the development of technologies tends to be driven by availability of a technology, transferred from other industries in market expansion efforts, rather than by need. Relative to some industries, the dairy industry is relatively small, limiting corporate willingness to invest extensively in development of technologies exclusive to dairy farms. Many Precision Dairy Farming technologies measure variables that could be measured manually, while others measure variables that could not have been obtained previously.

Realistically, the term "Precision Dairy" should not be limited to monitoring technologies. Perhaps a more encompassing definition of Precision Dairy Management is the use of automated, mechanized technologies toward refinement of dairy management processes, procedures, or information collection. This definition incorporates monitoring technologies, automated milking systems, automated calf feeding systems, and precision feeding systems. Automated milking systems have already been widely adopted in Europe.

Adoption rates in North American have increased in recent years. The introduction of robotic milking components to rotary parlors will increase mechanization of milking in larger farms in the near future. Automated calf feeding systems have created a paradigm shift in how to raise dairy calves. Despite initial concerns of increased disease transmission, the benefits to automated calf feeding seem to outweigh the drawbacks when managed properly. New options for monitoring total mixed ration delivery and consumption will also improve how lactating dairy animals are fed. This is a particularly important economic and social concern given increased feed prices and concern for dairy efficiency and greenhouse gas emissions.

Benefits

Perceived benefits of Precision Dairy Farming technologies include increased efficiency, reduced costs, improved product quality, minimized adverse environmental impacts, and improved animal health and well-being. These technologies are likely to have the greatest impact in the areas of health, reproduction, and quality control (de Mol, 2000). Realized benefits from data summarization and exception reporting are anticipated to be higher for larger herds, where individual animal observation is more challenging and less likely to occur (Lazarus et al., 1990). As dairy operations continue to increase in size, Precision Dairy Farming technologies become more feasible because of increased reliance on less skilled labor and the ability to take advantage of economies of size related to technology adoption.

A Precision Dairy Farming technology allows dairy producers to make more timely and informed decisions, resulting in better productivity and profitability (van Asseldonk et al., 1999). Real time data can be used for monitoring animals and creating exception reports to identify meaningful deviations. In many cases, dairy management and control activities can be automated (Delorenzo and Thomas, 1996). Alternatively, output from the system may provide a recommendation for the manager to interpret (Pietersma et al., 1998). Information obtained from Precision Dairy Farming technologies is only useful if it is interpreted and utilized effectively in decision making. Integrated, computerized information systems are essential for interpreting the mass quantities of data obtained from Precision Dairy Farming technologies. This information may be incorporated into decision support systems designed to facilitate decision making for issues that require compilation of multiple sources of data.

Historically, dairy producers have used experience and judgment to identify outlying animals. While this skill is invaluable and can never be fully replaced with automated technologies, it is inherently flawed by limitations of human perception of a cow's condition. Often, by the time an animal exhibits clinical signs of stress or illness, it is too late to intervene. These easily observable clinical symptoms are typically preceded by physiological responses evasive to the human eye (e.g. changes in temperature or heart rate). Thus, by identifying changes in physiological parameters, a dairy manager may be able to intervene sooner. Technologies for physiological monitoring of dairy cows have great potential to supplement the observational activities of skilled herdspersons, which is especially critical as more cows are managed by fewer skilled workers (Hamrita et al., 1997). Dairy producers with good "cow

sense" are the ones who will benefit the most from technology adoption. Those who view technologies as a way to do something they don't like to do will likely struggle.

Adoption

The list of Precision Dairy Farming technologies used for animal status monitoring and management continues to grow. Despite widespread availability, adoption of these technologies in the dairy industry has been relatively sparse thus far (Gelb et al., 2001, Huirne et al., 1997). Perceived economic returns from investing in a new technology are always a factor influencing technology adoption. Additional factors impacting technology adoption include degree of impact on resources used in the production process, level of management needed to implement the technology, risk associated with the technology, institutional constraints, producer goals and motivations, and having an interest in a specific technology (Dijkhuizen et al., 1997, van Asseldonk, 1999). Characteristics of the primary decision maker that influence technology adoption include age, level of formal education, learning style, goals, farm size, business complexity, increased tenancy, perceptions of risk, type of production, ownership of a non-farm business, innovativeness in production, average expenditure on information, and use of the technology by peers and other family members. Research regarding adoption of Precision Dairy Farming technologies is limited, particularly within North America.

To remedy this, a five-page survey was distributed to all licensed milk producers in Kentucky (N=1074) on July 1, 2008. Two weeks after the first mailing, a follow-up postcard was mailed to remind producers to return the survey. On August 1, 2008, the survey was resent to producers who had not returned the survey. A total of 236 surveys were returned; 7 were omitted due to incompletion leaving 229 for subsequent analyses (21%). The survey consisted of questions covering general farm descriptive demographics, extension programming, and decision making behavior. With regard to Precision Dairy Farming the following question was presented to survey participants: *"Adoption of automated monitoring technologies (examples: pedometers, electrical conductivity for mastitis detection) in the dairy industry has been slow thus far. Which of the following factors do you feel have impacted these modest adoption rates? (check ALL that apply)."* Data were entered into an online survey tool (KeySurvey, Braintree, MA). Statistical analyses were conducted using SAS® (Cary, NC). Surveys were categorized by herd size, production system, operator age, and production level. Least squares means among categories were calculated for quantitative variables using the GLM procedure of SAS®. Statistical differences were considered significant using a 0.05 significance level using Tukey's test for multiple comparisons. For qualitative variables, χ^2 analyses were conducted using the FREQ procedure of SAS®. Statistical differences were considered significant at a 0.05 significance level.

Among the 229 respondents, mean herd size was 83.0 ± 101.8 cows and mean producer age was 50.9 ± 12.9. Reasons for modest adoption rates of Precision Dairy Farming technologies and dairy systems software are presented in Table 1. The reasons selected by the highest percentage respondents were (1) not being familiar with technologies that are available (55%), (2) undesirable cost to benefit ratios (42%) and (3) too much information provided without knowing what to do with it (36%%). The high percentage of producers who indicated they were unfamiliar with available technologies indicates that marketing efforts may improve technology

adoption. Actual or perceived economic benefits appear to influence adoption rates demonstrating the need for economic models to assess technology benefits and re-examination of retail product prices. As herd size increased, the percentage of producers selecting "poor technical support/training" and "compatibility issues" increased ($P < 0.05$), which may be reflective of past negative experiences. In developing technologies, manufacturers should work with end-users during development and after product adoption to alleviate these customer frustrations. Few significant differences were observed among age groups, though the youngest producers were more likely to select "better alternatives/easier to accomplish manually." Prior to technology development, market research should be conducted to ensure that new technologies address a real need. Utilizing this insight should help industry Precision Dairy Farming technology manufacturers and industry advisors develop strategies for improving technology adoption. Moreover, this information may help focus product development strategies for both existing and future technologies.

Table 1. Factors influencing slow adoption rates of Precision Dairy Farming technologies

Factor	N	Percent
Not familiar with technologies that are available	101	55%
Undesirable cost to benefit ratio	77	42%
Too much information provided without knowing what to do with it	66	36%
Not enough time to spend on technology	56	31%
Lack of perceived economic value	55	30%
Too difficult or complex to use	53	29%
Poor technical support/training	52	28%
Better alternatives/easier to accomplish manually	43	23%
Failure in fitting with farmer patterns of work	40	22%
Fear of technology/computer illiteracy	39	21%
Not reliable or flexible enough	33	18%
Not useful/does not address a real need	27	15%
Immature technology/waiting for improvements	18	10%
Lack of standardization	17	9%
Poor integration with other farm systems/software	12	7%
Compatibility issues	12	7%

Outlook

Though Precision Dairy Farming is in its infancy, new Precision Dairy Farming technologies are introduced to the market each year. As new technologies are developed in other industries, engineers and animal scientists find applications within the dairy industry. More importantly, as these technologies are widely adopted in larger industries, such as the automobile or personal computing industries, the costs of the base technologies decrease making them more economically feasible for dairy farms. Because the bulk of research focused on Precision Dairy Farming technologies is conducted in research environments, care must be taken in trying to transfer these results directly to commercial settings. Field experiments or simulations may need to be conducted to alleviate this issue. Because of the gap between the impact of Precision Dairy Farming technologies in research versus commercial settings, additional effort needs to be directed toward implementation of management practices needed to fully utilize information provided by these technologies. To gain a better understanding of technology adoption shortcomings, additional research needs to be undertaken to examine the adoption process for not only successful adoption of technology but also technology adoption failures. Before investing in a new technology, a formal investment analysis should be conducted to make sure that the technology is right for your farm's needs. Examining decisions with a simulation model accounts for more of the risk and uncertainty characteristic of the dairy system. Given this risk and uncertainty, a stochastic simulation investment analysis will represent that there is uncertainty in the profitability of some projects. Ultimately, the dairy manager's level of risk aversion will determine whether or not he or she invests in a technology using the results from this type of analysis. Precision dairy farming technologies provide tremendous opportunities for improvements in individual animal management on dairy farms. In the future, Precision Dairy Farming technologies will change the way dairy herds are managed.

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Describing aNDFom Digestibility in Multiple Pools and Predictions for Rumen Function

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Introduction

Fiber digestibility and indigestibility are critical factors when assessing forage quality and formulating diets. Digestion characteristics of NDF influence feeding and rumination behavior, rate of particle breakdown, ruminal turnover and fill, dry matter intake, and overall efficiency of milk component output. Traditionally, nutritionists have focused on measures of NDF digestibility at specific time points and assumed that NDF was a relatively homogenous fraction. However, recently the focus has included indigestible fiber as well because of the recognition of its importance establishing the digestible portion or pool of NDF which leads to the extent of digestion and influences the rate(s) of fiber fermentation in the rumen. For purposes of nutritional modeling, indigestible NDF is required as the end point for fermentation to allow accurate estimation of the potentially digestible NDF fraction and its rate(s) of digestion. Measuring true NDF indigestibility would require infinite time, especially in aerobic systems, so in the actual rumen of a dairy cow or in an artificial rumen system, true indigestibility is never achieved. The standard nomenclature throughout the literature is “indigestible NDF (iNDF)” (Mertens, 1993; Huhtanen et al., 2006); however, to improve the accuracy of the standard terminology used to describe fiber fermentation dynamics, Mertens (2013) coined the term “undigested NDF (uNDF)” as the laboratory measure (typically in vitro or in situ) of indigestible NDF at a specified fermentation time. You will see both terms used, and for the most part, they are interchangeable as long as you know the method and time point used to determine the NDF digestion endpoint. However, moving forward, we will standardize our terminology to uNDF. To achieve iNDF requires estimations out to infinite time and that estimated residue might not be consistent with the interactive behavior of the forage and feed with rumen function and retention time.

This undigestible fraction is analyzed in laboratories with long term in-vitro fermentation and defined uNDF (Cotanch et al., 2014). Furthermore the component available to microbial digestion is defined as digestible NDF (dNDF) and can be obtained by subtracting the uNDF from total aNDFom (dNDF = aNDFom - uNDF). Previous work demonstrated that the dNDF of forages can be composed of two digestible fractions (Van Soest et al., 2005), both fractions following first order behavior but with different digestion rates and defined as fast and slow digesting pools (Raffrenato and Van Amburgh, 2010); whereas in plant by-products the dNDF is identifiable as one fraction and disappearing with a first order behavior (Cotanch et al., 2014). The size of the

various fractions or pools of dNDF and the associated digestion rates, when combined to create differences with a total mixed ration, might affect feed intake and rumination behavior, milk production and feed efficiency.

Why Should We Use uNDF?

Determination of uNDF should be included in routine forage and feed analysis because undigestible NDF is a uniform feed fraction with a predictable digestibility (i.e. zero). By contrast, NDF is a non-uniform feed fraction; it contains multiple pools that digest predictably as a function lignification and cross-linking (Grabber et al. 2009; Van Soest, 1994).

Undigestible NDF is the functional fiber fraction that influences physical effectiveness, gut fill, and digestion/passage dynamics of forages. Undigested NDF is important biologically because:

- it can be used to estimate potentially digestible NDF(pdNDF) (NDF - uNDF),
- the uNDF fraction together with earlier time points of fermentation can be used to estimate the fast and slow pools of NDF digestion and their digestion rates (Raffrenato and Van Amburgh, 2010),
- measures of NDF pools and rates of digestion based on uNDF can help explain feeding and ruminating behavior, especially when chemical composition (i.e. ADL, NDF, ADF) are similar,
- estimates of the slow pool of NDF and its rate of digestion plus the uNDF are related to dry matter intake and passage from the rumen since the fast pool disappears faster than can pass,
- uNDF plays a critical role in maintaining the ruminal digesta load, and
- uNDF predicts forage quality because of the relationship between uNDF and OM digestibility (Nousiainen et al., 2003).

At any given time, rumen fiber fill is a function of dietary uNDF, slowly fermenting NDF, and undigested fast-pool NDF. The rumen space resulting from turnover of the fast fiber together with the slow fiber and uNDF allows for more dry matter intake. The more rapidly rumen space is made available (i.e. the greater the turnover), the higher the intake that can be attained. The total mass of uNDF within the rumen can be thought of as a “baseline” of fill which constrains the possible NDF flux. We propose that there is a maximum and minimum amount of ruminal uNDF to avoid limits on feed intake and to maintain proper ruminal health, respectively. Undigested NDF can improve the precision of estimating dry matter intake by telling us, for example, how much uNDF in a TMR that a cow can consume before filling her rumen, and conversely, how much uNDF must be consumed to maintain rumen fill and digestive efficiency.

In fact, there may be an optimal mass of digesting NDF within the rumen; above this amount, fill limits intake while below this amount, intake could increase further although possibly at the expense of feed efficiency (Weakley, 2011). Although the effect on dry matter intake of adjusting dietary NDF is 2 to 3 times greater than changing the NDF digestibility (Mertens, 2009), in many practical feeding situations where dietary NDF has reached the maximum fill potential in high-

producing cows, then NDF digestibility (or undigestibility) becomes most important (Weakley, 2011). We believe that uNDF measured at 240 hours of in vitro fermentation ($uNDF_{240}$) is a forage fraction that accurately assesses the indigestible component of NDF (Raffrenato et al. 2009).

Updating the analysis of NDF to aNDFom

One other related aspect of uNDF and NDF in general is the use of organic matter correction. Biogenic ash (ash integral to plant development) is soluble in NDF solution, so that is properly accounted for during the assay, however, soil ash is not soluble in NDF solution and if not removed or accounted for will falsely inflate the NDF values and the same is true for the uNDF. Moving forward, both the NDF and the uNDF should be ash corrected to remove any potential confounding by soil contamination. Management approaches that take advantage of practices like "hay in a hurry" along with large, high horsepower choppers will impact the amount of soil that is found in the forages. In addition, based on region of the country that forage is produced or sourced will also affect the level of contamination. More sandy soils and irrigation practices such as flood irrigation can cause soil to be adhered to the plant. The easiest way to account for the contamination is to ash the residue after both the NDF and uNDF to correct the value. This also reduces bias in the estimation of rates of digestion since organic matter correction provides a more correct value for the true available NDF content. Thus, aNDFom analyses (NDF with sodium sulfite, amylase and ash correction) will provide nutritionists with more accurate information and in some cases significantly lower values.

There are no changes in the targets for aNDFom intake and in many cases, under reformulation, the amount of forage fed will increase 2-3% once the ash content of the NDF is accounted for. Under conditions where there was significant ash contamination, the amount of forage required to meet the typical dietary levels (e.g. 32%) can be increased by over 10% to maintain adequate aNDFom levels for normal rumen health. It is possible in certain situations, that inconsistent intakes, changes in rumination and rumen pH along with manure scores that are inconsistent can be an outcome of underfeeding forage and fiber because the NDF content of the diet was underestimated due to ash contamination. This most likely happens in the regions of the country where flood irrigation and sandy soils are more prevalent but it is still a possibility in the Northeast due to larger equipment, wide-swathing and variable field conditions.

How do we measure uNDF?

The approach for estimating iNDF within the structure of the Cornell Net Carbohydrate and Protein System (CNCPS; Tylutki et al., 2008) has been through the use of acid detergent lignin (ADL) and a fixed (static) factor of 2.4 calculated as ADL*2.4/NDF (Chandler et al., 1980). For other applications the approach most often used is that of Conrad et al. (1984) where a surface area relationship is described by a power function ($(1- lignin^{0.67}/NDF^{0.67})$ was used to describe the relationship between lignin and NDF to characterize the unavailable NDF. These "static calculations" or use of a fixed factor for calculating uNDF are used in many of the net energy equations by commercial laboratories and the 2001 NRC (NRC, 2001) and that can create errors

in energy predictions due to the lack of a constant relationship between lignin and carbohydrate and digestibility.

More recently, uNDF has been estimated through long-time in vitro or in situ fermentations. The method recommended by the Cornell group requires 240 hours of in vitro fermentation using a Tilley-Terry system with modifications described by Raffrenato and Van Amburgh (2010). The fermentation end point *per se* is not important – it will vary with fermentation system. For example, the in situ approach published by Huhtanen et al. (2007) uses 288 hours to reach a similar fermentation endpoint to measure uNDF. The goal is to reach a point where the residue weight does not change significantly with additional hours of fermentation – this will be a measure of uNDF and the estimate of indigestible NDF for estimation of rates and extent of digestion. For commercial laboratory application and routine model inputs, we prefer the use of an in vitro approach which allows for sample submission from nutritionists and development of an adequate-sized database to develop NIR equations that will reduce the cost and increase the speed of sample analysis.

Examples of the chemistry related to NDF and NDF digestibility in four corn silages along with the calculated in-digestibilities based on Chandler et al., and Conrad et al., are found in Table 1. The data in the table demonstrate the subtle differences that can be observed when analyzing for aNDFom compared with aNDF. The average difference among this very small sampling is 0.9 units of NDF, a very modest amount. However, we have analyzed or dealt with samples that were up to 10 units different after ashing, so again, it depends on where the sample is from and the agronomic conditions it is grown under. The uNDF as measured at 240 hr averages 24.8 %NDF whereas the lignin (%NDF)*2.4 value averages 41.9% and the power function of Conrad et al. (1984) averages 20.7%. The differences between the actual measurement and the calculations are significant and will result in biased estimations of total digestibility, rates of digestion and energy predictions. Overall, this small example demonstrates that the values estimated by the previous methods using fixed factors as a function of the chemical measurement of lignin miss the potential interaction (cross-linking) between lignin and carbohydrate that actually impact the digestion capacity of the plant.

Table 1. Corn silage fiber chemistry, 240 in vitro indigestibilities (uNDF), and estimations of indigestible fiber by Chandler et al. (1980) (lignin (%NDF) x 2.4) and Conrad et al., 1984.

Corn silage	aNDF, %DM	aNDFom, %DM	Lignin, %NDF	uNDF, %NDF	Chandler et al. 1980	Conrad et al., 1984
1	38.1	37.5	6.61	23.6	42.3	16.4
2	39.5	38.9	6.46	25.6	39.2	16.89
3	41.5	40.9	7.47	27.3	43.4	17.7
4	43.7	41.9	7.51	22.8	42.8	31.8

Similar observations have been made for the non-forage fiber sources. Byproducts like beet pulp and citrus pulp that have good nutrient value and can be routine sources of energy for lactating dairy cattle have digestion behavior that is not dissimilar from forages. Data were generated to better understand when the uNDF is identified in non-forage fiber sources and that is in Table 2. For most non-forage feeds, the uNDF can be measured after 120 hr of in vitro

digestion provided the samples are filtered on the appropriate filter paper (Whatman AH934 or equivalent). The only feed that had behavior more similar to forages was citrus pulp where the uNDF of the sample represented below was only identified at 240 h of fermentation.

Once the uNDF was identified and understood, it was important to evaluate the measured values from these non-forage fiber sources in a similar manner to the forages to better understand if the static calculations for uNDF and the measured uNDF were similar. The data in Table 3 demonstrate that the measured uNDF is both over- and under-predict for the feeds represented in this table and these inconsistencies will impact the estimation of digestible NDF and will also affect energy predictions from this group of feeds. Static values as a function of the lignin to NDF relationship do not adequately account for the digestibility and uNDF of non-forage fiber feeds in a similar manner as forages, however it is expected that the variation in non-forage fiber feeds will not be as great as the forages due to the lack of agronomic conditions affecting their development.

Table 2. The aNDFom (%NDF) residues of feeds after 96, 120, and 240h of fermentation

	Time (h)				
	96	120	240	SEM	P-value
Beet pulp	22[a]	19[b]	17[b]	0.01	0.004
Canola meal	40	41	41	0.01	0.79
Citrus pulp	21[a]	20[a]	16[b]	0.01	0.002
Corn Gluten feed	16[a]	14[ab]	13[b]	0.01	0.028
Corn distiller	16	16	14	0.01	0.50
Corn germ	34	29	27	0.03	0.74
Flaked corn	14	14	12	0.02	0.73
Rice hulls	94	93	93	0.01	0.61
Soybean meal	11	9	9	0.01	0.95
Soy hulls	10[a]	9[ab]	8[b]	0.01	0.022
Wheat distiller	28	26	25	0.01	0.20
Wheat middling	36[a]	31[b]	30[b]	0.01	0.001

[a,b]Values with different letters are statistically different

Implications and Applications

Data being generated on lactating dairy cattle indicate the cow can “identify” with the values related to the uNDF measurements along with the rest of the pools (fast and slow digesting NDF pools) and these measurements are in some manner related to rumen fill, eating speed and ultimately, dry matter intake. Data generated in a forage digestibility study at Miner Institute with high and low forage inclusion levels demonstrated that the cow consumes approximately the same amount of uNDF as she excretes in her feces every day. The precision of the relationship was surprising as showing in Table 4. The relationship between uNDF intake and uNDF excretion was 1:1 and coupled with the relationship between the rumen contents of uNDF and the intake of uNDF suggests that if we understand the uNDF, we can directly estimate the rumen fill of total NDF and further, we should be able to predict intake among differences in TMR uNDF values.

Table 3. The neutral detergent fiber, acid detergent lignin and comparison of three methods of estimation of uNDF based on 120 hr fermentation, the Chandler equation or the Conrad equation, respectively.

Feed	aNDFom (%DM)	ADL (%DM)	uNDF (%aNDFom)	2.4 x ADL (%aNDFom)	$ADL^{2/3}/NDF^{2/3}$ (%aNDFom)
Beet pulp	47	5.4	19	28	24
Canola meal	29	8.8	41	73	45
Citrus pulp	25	1.94	20	19	53
Corn gluten feed	37	2.27	14	15	4
Corn distiller	41	4.4	16	26	23
Corn germ	63	5.9	29	23	21
Flaked corn	13	1.4	14	26	23
Rice hulls	71	0.8	93	20	5
Soybean meal	9	0.85	1	23	21
Soy hulls	72	1.3	9	10	7
Wheat distillers	38	3.8	26	29	22
Wheat middlings	45	4.9	31	17	23

Table 4. Intake of NDF and uNDF and rumen fill for Miner study

Item	LF-LD	HF-LD	LF-HD	HF-HD
NDF_{om} intake				
kg/d	8.87	8.95	8.48	9.88
% of BW	1.32	1.33	1.27	1.47
Rumen NDF_{om}				
kg	8.50	8.58	7.82	8.48
% of BW	1.27	1.28	1.17	1.27
$uNDF_{240om}$ intake				
kg/d	2.39	2.63	2.03	2.21
% of BW	0.36	0.39	0.30	0.33
Rumen $uNDF_{240om}$				
Kg	3.82	4.16	3.20	3.46
% of BW	0.57	0.62	0.48	0.52
Fecal uNDF, kg/d	2.41	2.64	2.04	2.24
Ratio rumen/intake uNDF	1.60	1.58	1.58	1.57
Ratio intake uNDF/fecal uNDF	1.0	1.0	1.0	0.99

Summary

Studies are underway to evaluate the concept of aNDFom pools, chewing and rumination and feed intake. The data generated to date suggests that predictions for energy, rates of digestion, microbial yield and dry matter intake will be improved through the application of uNDF and the pool approach to defining NDF digestion. This is exciting and gives new life to an

old topic, and might help explain differences in feeding behavior that nutritionists and others have observed but have never been able to quantify.

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FINANCES AND RETURNS FOR ROBOTIC DAIRIES

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Introduction

Over 35,000 robotic milking systems (RMS) units are operational on dairy farms around the world. The main reasons dairy producers install milking robots are to improve their lifestyle and to expand without hiring additional labor.

What drives robot profitability?

Milk production per cow, milk produced per robot per day, labor savings, and length of useful life are the main factors affecting RMS profitability. The primary disadvantage is the capital investment of $150,000 to $200,000 per robot that will milk 50 to70 cows each. Most historical data shows milking robots are less profitable than conventional milking systems. Advances in robotic technology, improved management skills, and higher labor costs may change these results.

Labor efficiency

USDA (2016) reports that wages paid to livestock workers increased 3% in 2014 and 4% in 2015. Reported RMS labor savings vary. Researchers have reported from no savings up to 29% savings with RMS. Barn design and management may explain much of this variation. Farm Management Records (Finbin, 2016) showed that Upper Midwest RMS farms averaged 2.2 million lb of milk per full time worker compared to 1.5 million lb for similar sized herds milking in parlors (Table 1). Our survey of 53 Minnesota and Wisconsin robot farms showed that even when total labor is similar, time saved from milking is used for activities, such as improving animal health, analyzing records, improving reproduction, and more timely forage harvest.

Another factor affecting the decision to install robots is the future availability of labor for milking cows. A 2014 survey indicated that 51% of all farm labor was immigrant labor (Adcock et al., 2015). The future availability of immigrant workers may be reduced if less foreign workers choose to work on farms or if tighter immigration laws are passed in the US.

Milk production change when transitioning to robots

The primary driver for the change in milk production with RMS is a change in milking frequency. de Koning (2010) found that robotic herds had production increases of 5 to 10% compared to milking 2X, but production decreased 5 to 10% compared to milking 3X. In our survey, the average RMS milking frequency was 2.8 with a range of 2.4 to 3.2. To optimize efficiency, the goal is to have high milking frequency in early lactation and lower milking frequency in later

lactation. The primary factors that affect individual cow and herd average milking frequency include:

1. Number of cows per robot
2. Milking permission settings
3. Palatability and quality of partial mixed ration and robot box feed
4. Robot free time (time robot is idle)
5. Cow fetching policy
6. Barn design and walking distance (a major factor for grazing herds)

Robotic milking systems compared to conventional parlor systems

Bijl et al. (2007) compared the economic performance of Dutch farms using RMS to closely matched conventional farms milking 2X. Because of higher costs for the RMS, conventional farms were more profitable. However, the labor requirement was 29% lower on the RMS farms resulting in more milk production and income per worker. They concluded that investing in RMS allows farms to milk more cows and produce more milk with less labor.

Farm management records collected by the University of Minnesota show a similar pattern (Table 1). Herds utilizing RMS had higher milk production and gross margin, but costs were higher, resulting in slightly lower net farm income.

Table 1. Robot and parlor farm profitability, 2011-2015, Upper Midwest[1]

Item	Robot	Parlor	Difference
Milk/cow/yr	23,532 lb	21,528 lb	+2004 lb
Gross margin/cow/yr	$4564	$4254	+$310
Feed cost/cow/yr	$2251	$2206	+$45
Direct cost/cow/yr[2]	$3261	$3190	+$71
Overhead cost/cow/yr[3]	$899	$581	+$318
Net farm income/cow/yr	$185	$230	-$45
Milk sold/Full time worker/yr	2,206,107 lb	1,542,874 lb	+663,233 lb
Depreciation + interest/cow/yr	$547	$249	+$298

[1]Finbin, University of Minnesota www.finbin.umn.edu

[2]Feed, vet, supplies, bedding, fuel repairs, marketing and hired labor

[3]Building and machinery depreciation, building leases, insurance, utilities, interest

We developed a web application to compare the profitability of robots and parlors: http://z.umn.edu/RobotParlor. This tool was used to compare the economics of RMS and parlor systems on farms with 120, 240 and 1,500 lactating cows over a 20-year pay-back time. Milking labor costs were set at $16/hr with a milk price of $17/cwt. We assumed milk production would increase 5 lb/day per cow with RMS compared to milking 2X and decrease 2 lb/day compared to 3X milking. The per cow barn investment is higher for the RMS, reflecting the additional cost to install labor savings features typical in RMS barns. We inflated labor costs at 1, 2, or 3%

annually. Net annual impact refers to the net present value of projected differences in RMS cash flows converted to an annuity.

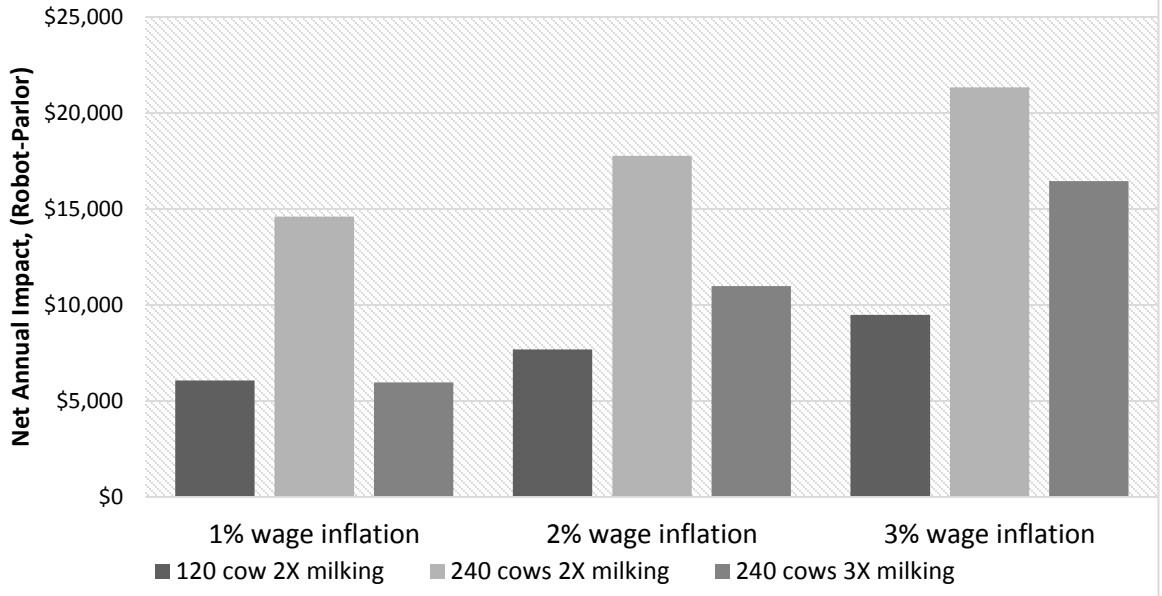
The 120 and 240 cow RMS systems had higher net annual impact compared to a double 8-parlor system (Figure 1). Labor cost inflation and milk production per cow had a large impact on profit. For each pound change in daily production per cow, the net annual impact changed by $931.

The 1,500-cow parlor system was more profitable than RMS. A 1% annual wage inflation resulted in a $162,672 (3X milking) and $51,177 (2X milking) more profit for the parlor. The difference was $130,570 (3X milking) and $32,395 (2X milking) at 3% wage inflation. Using similar milk production and 3% wage inflation the parlor had $80,672 higher annual impact.

The primary reason for the differences in profit is the more intensive use of the milking system. The RMS assumed full utilization at 60 cows per robot across all herd sizes. The parlor was only being used four hours per day with the 120-cow system. In the 240-cow simulations, the parlor was being used 8 and 12 hr/day in the 2X and 3X respectively. For the 1,500-cow herd, both the robot and parlor were at near maximum utilization.

Milk production and labor assumptions between the systems greatly affect the profitability projections. More research is needed to understand the economics of how these systems perform with different herd sizes and management practices.

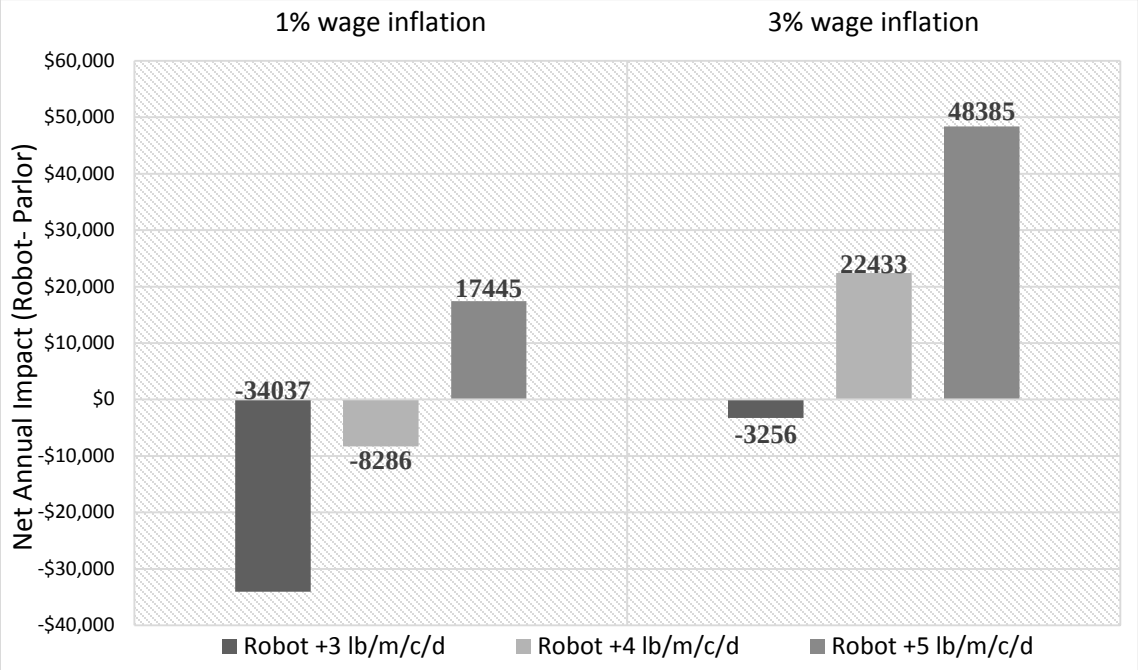
Figure 1. Net annual impact of robot system compared to a parlor system



Breakeven labor rate. Since the 1,500-cow robot system was less profitable than the parlor system at $16 labor, we determined the breakeven labor rate at which the two systems would have similar annual incomes. At the wage inflation rate of 1% and a 2 lb lower milk production with the robot system, the breakeven labor rate is $32.30 per hour. If the farm is able to achieve similar milk production between the two systems and wage inflation averages 3% over the 20-year time horizon, the breakeven wage rate drops to $22.91 per hour.

Breakeven milk production. We also examined how higher milk production in RMS would affect the profit comparison (Figure 2). If the robot system can get 3 lbs more milk/cow per day than a parlor with 3% annual wage inflation, the annual income is only $3256 higher for the parlor. At 5 lbs more milk, the RMS is more profitable under both 1% and 3% wage inflation rates. Current research indicates that RMS on average do not achieve as high of milk production as a parlor milking 3X, but as robot management and facility design improve, this may change. Another potential advantage is that cows in robot barns can be managed and milked in stable groups within the pens. Cows have access to resources (feed, water, beds, milking) at all times. More precise feeding management can potentially increase milk per cow.

Figure 2. Net annual impact of a 1,500-cow dairy with 25 robots compared to a double-24 parlor at different milk production levels and wage inflation rates



Milk per robot

Total daily milk per robot is an important characteristic to maximize profit. Using a four robot system with a 2% annual wage inflation rate and a 20-year time horizon, net annual income increases approximately $4,100 for every 500 lb increase in daily milk production per robot. Currently a small number of US farms are consistently achieving in excess of 6,000 lb of milk per robot per day. Many factors can influence milk per AMS unit. The most important factors include:

1. Milking permission settings and strategies that get the correct cows milked at the correct times
2. High daily milk production per cow
3. Reduced box time per cow
4. RMS in top working condition

Effect or RMS Dairy Enterprise Profitability

We also examined how the economic life, labor efficiency and milk yield change affects the profitability of RMS. For 180 cow dairy we considered two scenarios: RMS retrofitted in an existing facility and a RMS in combination with a new high technology free stall barn.

Robot retrofit

This is retrofitting 3 robots in an existing 180 cow barn. We included increased annual payments for the 3 new robots ($63,000) for 10 years and increases in insurance ($2,700) and maintenance ($9,000/robot per year). The annual impact is the return compared to a current milking system with no debt payments. Table 2 shows how annual impact varies with different milk production, milking labor and robot lifespan. Our survey of producers indicated that well designed (automatic manure removal, split entry pens), well managed free flow barns average about 45 minutes of daily milking type labor per robot. It is slightly lower with similar guided flow barns. Dairy producers need to reduce daily milking labor to about 45 minutes per robot and achieve a small increase in milk yield for the RMS to have a higher annual impact than the previous milking system. Robots must also last for 10 to 14 years to breakeven.

Table 2. Net annual impact of installing a robot in an existing facility[1]

Milking labor (min/d/robot)	Robot lifespan (years) 8	10	13	15	17	20
I. Milk yield increase of 2.0 lb/cow/d						
75	($7,469)	($144)	$7,169	$10,747	$13,723	$17,461
60	($4,581)	$2,821	$10,251	$13,910	$16,966	$20,828
45	($1,693)	$5,786	$13,334	$17,073	$20,210	$24,196
30	$1,195	$8,751	$16,417	$20,235	$23,454	$27,563
II. No change in milk yield						
75	($17,516)	($10,394)	($3,387)	($14)	$2,754	$6,181
60	($14,628)	($7,429)	($305)	$3,148	$5,998	$9,548
45	($11,740)	($4,464)	$2,778	$6,311	$9,242	$12,915
30	($8,852)	($1,499)	$5,861	$9,474	$12,485	$16,283
III. Milk yield decrease of 2.0 lb/cow/d						
75	($27,615)	($20,696)	($13,998)	($10,832)	($8,271)	($5,158)
60	($24,727)	($17,731)	($10,915)	($7,669)	($5,027)	($1,791)
45	($21,839)	($14,766)	($7,832)	($4,506)	($1,783)	$1,577
30	($18,951)	($11,801)	($4,749)	($1,344)	$1,460	$4,944

[1]Net Annual Impact compared to the status quo with 13.5 h/d milking labor.

Robot with a New Barn

To achieve the maximum benefit of robots it is preferable to design them into a new, high technology, low labor requirement facility. This facility would include various upgrades such as wider and more frequent crossovers to facilitate cow movement, automated manure removal, automated feed pushers, and a more temperature-controlled environment. This totally new facility resulted in annual payments of about $101,000 over 20 years for the 180-cow farm. A 10 lb per day increase in daily production is required before robots are consistently more profitable than the previous system.

Table 3 shows the results by projected milk yield increase and milking labor. Much higher milk production and labor savings must be achieved to recuperate the large investment. We assumed a 15-year economic life for the robots and 30 years for the barn. This could be different depending on farmer's desire to upgrade the robots and potential barn time horizon.

Table 3. Net annual impact with robots as part of a new facility.[1]

Milking labor	Milk yield increase (lb/cow/d)					
(min/robot/d)	4.0	8.0	10.0	12.0	14.0	16.0
30 year time horizon; 15 year AMS economic life[2]						
75	($17,581)	($5,375)	$6,768	$18,847	$30,862	$42,814
60	($13,784)	($1,577)	$10,565	$22,644	$34,659	$46,611
45	($9,986)	$2,220	$14,362	$26,441	$38,457	$50,408
30	($6,189)	$6,017	$18,160	$30,239	$42,254	$54,205

[1] Annual Impact compared to the status quo with 13.5 h/d milking labor.
[2] Robot is replaced twice over the 30-yr time horizon

CONCLUSIONS

Most previous simulations and observational studies have shown that RMS are not as profitable as parlors. Our understanding of robotic facility design, feeding, and management will continue to improve, resulting in decreased labor requirements and higher milk production of cows milked with robots. The main management factors affecting whether robots are more profitable than parlors are increased milk production per cow, labor wages and labor savings. Another major factor is years of economically useful life. For comparing the relative return of robots versus parlors, the producer needs to understand how their management ability and future wage inflation affect potential future net income.

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Enhancing the Efficiency of Nutrient Utilization in Cattle

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Why It Is Important to Enhance Nutrient Utilization Efficiency in Cattle

The efficiency of conversion of dietary nutrients, in particular nitrogen (N), into milk or meat protein is low in ruminants. In dairy cattle, less than 30% of dietary N is captured into milk protein (VandeHaar and St-Pierre, 2006). Similarly, up to 90% of feed N is excreted in urine and feces, with only a small fraction (10 to 20%) being converted into meat protein in beef cattle (Satter et al., 2002). Since feed is the major contributor to total operational costs, and protein is typically the most expensive component of most diets, the capture of only a small proportion of dietary N into saleable product reduces on-farm profitability (VandeHaar and St-Pierre, 2006). This is especially critical today because of the rising and volatile cost of traditional protein ingredients including soybean meal and the current low milk and cattle prices, which are severely eroding profit margins.

Besides its impact on economic returns, the high excretory losses of N, in particular, urine urea-N, can also compromise ecological health. Compared to fecal N, urine urea-N is more prone to being broken down and altered into various compounds including ammonia and nitrates that can pollute the atmosphere, and ground and surface water (VandeHaar and St-Pierre, 2006). Although cattle producers in the U.S. do not currently face direct costs associated with the loss of N in urine and feces, the implementation of taxes on N excretion on farms in some European countries including the Netherlands is a harbinger of the future changes in environmental policies and regulation (Higgs et al., 2012). Moreover, the recent lawsuits alleging nitrate pollution of groundwater from agricultural activities in the Yakima Valley, WA, and the three rural counties neighboring Des Moines, IA, are a strong indication of the rising tide of change with regards to environmental accountability in the U.S. (Scheider, 2015). Therefore, as nutrient management policies and regulations will become more stringent, and as feed costs continue to rise, viability of both the dairy and beef sectors is dependent on the implementation of dietary strategies that will enhance the efficiency of N utilization.

Nitrogen Metabolism in the Rumen and Urea-N Recycling

The indiscriminate nature of dietary protein degradation by the rumen microbes is one of the major causes of the low efficiency in N use by cattle (Calsamiglia et al., 2010). Dietary protein, is broken down to peptides, amino acids and ammonia, which can all be used to support microbial growth (synthesis of microbial protein) as long as fermentable energy is available. However, excess peptides and amino acids are further broken down to ammonia, and the bulk of ammonia that is not used for microbial growth is then lost from the rumen through

absorption into blood. This loss of ammonia from the rumen is one of the major reasons for the low efficiency in N use. Since it is neurotoxic at high blood concentration, absorbed ammonia is converted to urea-N in the liver. Thereafter, the urea-N is released into blood (blood urea-N), and can then either be recycled back to the rumen or excreted in urine (urine urea-N). It is estimated that between 40 and 80% of urea-N that is synthesized in the liver can be recycled back to the rumen and used to support microbial growth (Lapierre and Lobley, 2001). The proportion that is recycled and used for microbial growth is dependent on a number of factors, including the supply of fermentable energy from the diet. The remainder (20 to 60%) is what is then irreversibly lost to the environment as urine urea-N and can potentially compromise ecological health.

Dietary Strategies to Enhance Nitrogen Utilization Efficiency in Cattle

Limiting the amount of ammonia that is lost from the rumen and increasing the proportion of blood urea-N that is recycled back to the rumen could potentially improve the efficiency of N usage in cattle (Calsamiglia et al., 2010). In one distillers grains study (Chibisa and Mutsvangwa, 2013), we attempted to reduce the amount of ammonia that is lost from the rumen by feeding a low- compared to a high-protein diet (15.2 vs. 17.3% CP of diet DM) to lactating cows. As expected, cows fed the low-protein diet consumed less N, which resulted in a lower concentration of ammonia in the rumen compared to cows fed the high-protein diet (Table 1). In addition, the amount of urea-N that was synthesized in the liver and blood urea-N concentration were also lower in cows that were fed the low-protein diet suggesting that reducing dietary CP concentration also limited the amount of ammonia that was lost from the rumen. Moreover, cows that were fed the low- compared to the high-protein diet also excreted 21.8% less total N and 30.1% less urine urea-N (Table 1), which is desirable from an environmental stewardship standpoint. Others (Spek et al., 2013; Arriola Apelo et al., 2014) reported similar findings when feeding low-protein diets to lactating cows, which is a strategy that can also reduce feed costs.

Although it is effective in limiting the excretion of urea-N in urine, reducing dietary CP content can also potentially cause a decrease in milk and milk component yields, which is undesirable for economic reasons. This loss in production results in from the low dietary protein content causing a deficiency in ruminally-degradable protein (RDP), which compromises rumen function and reduces dry matter intake (DMI), microbial protein synthesis or the supply of key amino acids required for milk protein production (Giallongo et al., 2015). Ultimately, there is a decrease in performance due to the metabolizable protein and energy requirements of the lactating cow not being met. In our study (Chibisa and Mutsvangwa, 2013), although feeding diets containing 15.2 compared to 17.3% CP did not impair DMI, it resulted in a decrease in milk and milk protein yield (3.6 and 0.14 kg/d, respectively; Table 2). Therefore, this decrease in production performance was possibly caused by a deficiency in dietary RDP that resulted in a decrease in rumen ammonia concentration and compromised microbial growth. Kristensen et al. (2010) and Spek et al. (2013) also made similar observations, and this makes the feeding of low-protein diets to lactating cows an economically risky strategy to adopt on farms.

Urea recycling is an important evolutionary adaptation mechanism that enables ruminants to survive on low-protein diets as a greater proportion of blood urea-N is returned back to the

rumen instead of loss in urine (Lapierre and Lobley, 2001). Therefore, there has been interest in determining whether it is possible to fully harness this "N salvage" mechanism so as to prevent a deficiency in RDP supply when lactating cows are fed lower protein diets. In our study (Chibisa and Mutsvangwa, 2013), we had anticipated that feeding a low compared to a high-protein diet (15.2 vs. 17.3% of diet DM) would increase the amount of urea-N recycled back to the rumen. This would have then buffered the rumen from the decrease in dietary RDP supply, thereby preventing a decrease in milk and milk protein yield due to compromised microbial growth. However, although the amount of recycled urea-N that was used for microbial growth was greater in cows fed the low- compared to the high-protein diet (11.5 vs 8.5%), it was not substantial enough to prevent a decrease in microbial growth (Fig 1 and 2). Ultimately, this contributed to the compromised production performance in our study (Chibisa and Mutsvangwa, 2013). Similarly, although the amount of urea-N that was recycled back to the rumen was greater (23.2 vs. 8.1%) in cows fed a low compared to a high-protein diet (12.9 vs. 17.1% of diet DM), it did not prevent a decrease in milk yield (36 vs. 42 kg/d; Kristensen et al., 2010). Spek et al. (2013) also reported a 3.4 kg decrease in milk yield despite a greater proportion of urea-N synthesized in the liver being recycled back to the rumen in cows fed diets containing 11.6 compared to 15.4% CP (DM basis).

On the contrary, feeding a low compared to a high-protein diet (14.9 vs. 17.5% of diet DM) to lactating cows did not compromise milk production (Mutswangwa et al., 2016). This was attributed to a greater tendency for the use of recycled urea-N for microbial growth in cows fed the low- compared to the high-protein diets. However, although not reported, the low-protein diets contained a higher amount of starch compared to the high-protein diets as a result of the higher inclusion level of barley grain (38.3 vs. 30.8%). Therefore, this greater supply of readily fermentable energy in cows fed the low- compared to high-protein diet could have enhanced the recycling of urea-N to the rumen and its use to support microbial protein synthesis. However, Recktenwald et al. (2014) did not observe an increase in the capture of recycled urea-N for microbial growth in the rumen after increasing the amount of dietary starch (28.7 vs. 22.0% of diet DM) in diets containing 15.2 compared to 16.7% CP. Given these contradictory responses, more research is warranted to shed more light on regulation of urea-N recycling in dairy cows and whether this "N salvage" mechanism could be fully harnessed to prevent production losses associated with feeding low-protein diets.

Another strategy that has received considerable interest in recent years, is the supplementation of rumen-protected amino acids in low-protein diets (Giallongo et al., 2015). Supplementation compensates for the deficiency in metabolizable protein supply by providing specific additional amino acids to support milk and milk protein synthesis. To date, most of the research on amino acid supplementation have primarily focused on lysine, methionine and histidine. Lysine and methionine are the most limiting amino acids for milk protein production in typical corn-alfalfa forage-based diets (NRC, 2001), whereas histidine has been reported to be a limiting amino acid in corn-silage based diets that are deficient in metabolizable protein (Lee et al., 2012). Provision of these rumen-protected amino acids (lysine, methionine and histidine) has been shown to be effective in preventing a decrease in milk and milk protein yield when lactating cows are fed low-protein diets (Lee et al., 2012; Arriola Apelo et al., 2014; Giallongo et al., 2015). Therefore, this strategy is beneficial in reducing N loss to the environment while also maintaining

production performance; and depending on the cost of including supplemental rumen-protected amino acids in the diet, it could also be economically prudent.

Currently, there is a growing body of research on the dietary inclusion of compounds that can modify/reduce the rate and extent of digestion of protein in the rumen and, therefore, limit the loss of ammonia into blood. Tannins, which are plant secondary metabolites, are an example of such compounds that can potentially improve protein utilization when included in moderate amounts (up to 4% of diet DM) in the diet (Makkar, 2003). Tannins bind dietary protein to form a tannin-protein complex that is stable in the rumen (pH 3.5 to 7.5) and cannot be degraded by the rumen microbes. However, the tannin-protein complex breaks down in the abomasum as a result of the low pH (< 3.5) to release the protein for potential enzymatic digestion (Jones and Mangan, 1977). In addition, tannins also reduce the growth and activity of the microbes that degrade protein in the rumen. Ultimately, this reduces the concentration of ammonia in the rumen and its loss into blood, which in turn reduces the synthesis and loss of urea-N in urine.

Although beneficial in improving N utilization, feeding an excessive amount of tannins can also potentially compromise production performance. For instance, feeding up to 1.8% (of diet DM) quebracho-chestnut tannin extracts resulted in a decrease in urine urea-N excretion (Aguerre et al., 2016). However, Aguerre et al. (2016) also noted a decrease in dry matter intake, nutrient digestibility and milk protein yield as the tannin inclusion level increased (0.45, 0.9 and 1.8% of diet DM), which led them to recommend limiting the amount to 0.45% to prevent a decrease in lactation performance. On the contrary, inclusion of up to 3% quebracho or chestnut tannin extracts did not compromise dry matter intake, nutrient digestibility or milk yield and composition (Dschaak et al., 2011; Liu et al., 2013). Since the reasons for this discrepancy are still not known, there is a need for additional research to fine-tune the use of this strategy such that the decrease in urine urea-N excretion is not accompanied by a reduction in milk and milk component yield. In addition, besides the commercially-sourced tannin extracts (primarily from countries like Argentina and Brazil), there is a need for more research on the use of local sources of tannins including the tannin-rich forages (e.g., sainfoin and birdsfoot trefoil) and by-products (e.g., grape pomace), which have also been reported to improve N utilization in a handful of studies (Hymes-Fecht et al., 2013; Ishida et al., 2015; Huyen et al., 2016).

Conclusion:

Given the rising cost of feed, mounting consumer pressure to reduce the environmental cost of milk and meat production, and the current low milk and cattle prices, it is imperative that producers adopt feeding strategies that enhance the efficiency of conversion of dietary nitrogen into saleable products (milk or meat). Several strategies including reducing dietary protein concentration and feeding compounds that limit protein degradation in the rumen (e.g., tannins) have been shown to be effective in reducing feed costs and urinary N excretion; however, they can also potentially compromise production performance. Therefore, besides fine-tuning the existing ones, there is a need for development of other novel strategies to effectively address the productivity, profitability and environmental sustainability-related challenges that dairy and beef producers in the U.S. are facing today.

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Table 1. Rumen ammonia and blood urea-N concentration, and urine N and urine urea-N excretion in lactating cows fed a low- compared to a high-protein diet

	Diet			
Item	**15.2% CP**	**17.3% CP**	**SEM**	***P*-value**
Rumen ammonia, mg/dL	10.9	12.7	0.95	0.04
Blood urea-N, mg/dL	15.9	19.0	1.12	<0.01
Urine N, g/d	212	271	18.9	<0.01
Urine urea-N, g/d	79	112	6.5	<0.01

Table 2. Dry matter intake, milk and milk protein yield in lactating cows fed a low- compared to a high-protein diet

	Diet			
Item	**15.2% CP**	**17.3% CP**	**SEM**	***P*-value**
Dry matter intake, kg/d	28.7	29.2	0.85	0.29
Milk yield, kg/d	40.3	43.9	1.53	<0.01
Milk protein yield, kg/d	1.22	1.36	0.049	<0.01

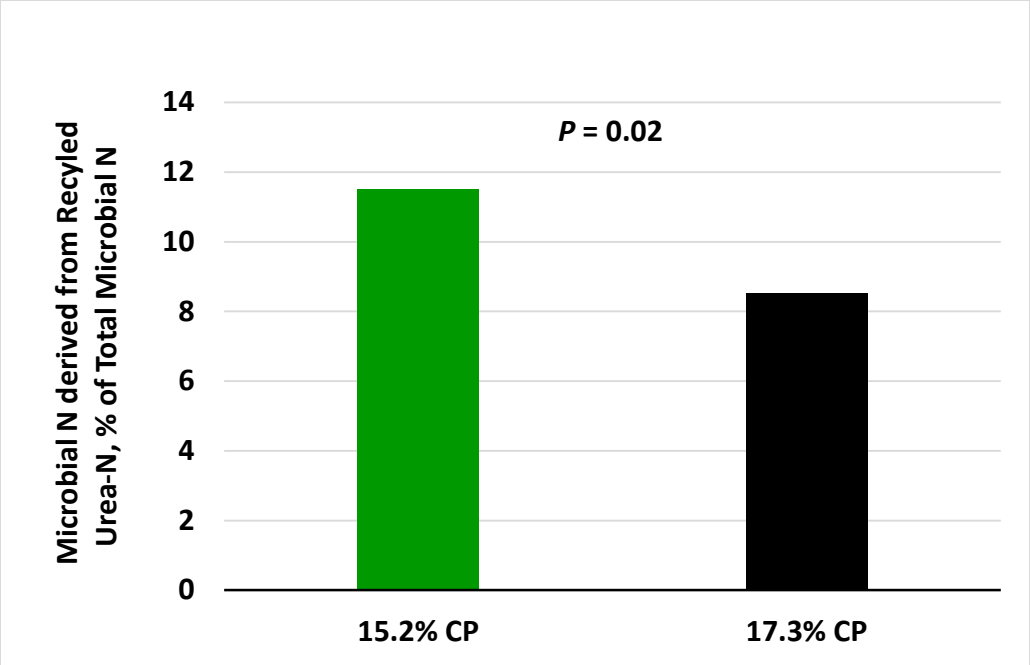

Fig 1. Use of recycled urea-N for microbial growth in lactating cows fed a low- compared to a high-protein diet

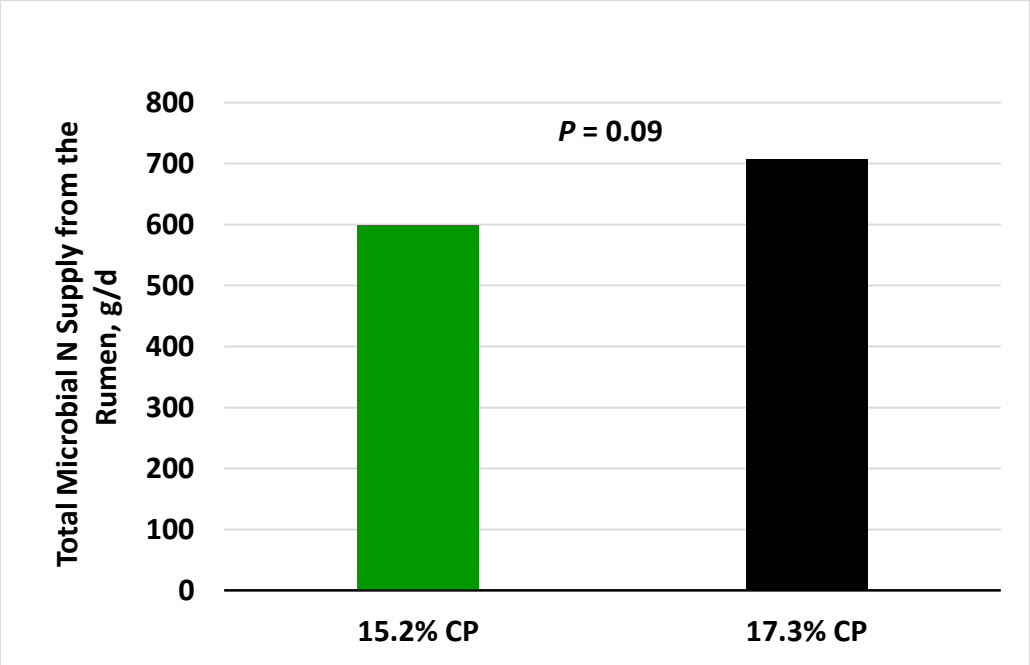

Fig 2. Microbial N flow out of the rumen in lactating cows fed a low- compared to a high-protein diet

Rumen-protected histidine supplementation; nutrient driven beef improvement

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The objective of this study was to determine the effect of supplementation with rumen-protected (RP) histidine in finishing cattle on growth, feed to gain ratio, and carcass traits/product quality. This project tested three levels of daily RP-histidine (control, low dose, and high dose) over a 55-d finishing period and implemented an aggressive 120 d implant (Revalor-XS). Crossbred beef steers were vaccinated, dewormed, blocked by body weight, and randomized into pens of six (eight pens total). The cattle were fed using Calan gates from an average starting BW of 355kg to a finishing LW of 615kg. Cattle were fed twice daily and the morning feed was top-dressed with the RP histidine according to treatment group: control (no RP-histidine), low dose (50g/hd each d), or high dose (100g/hd each d). Each steer received A,D&E injectable vitamin supplementation 80 d prior to initiation of treatments. Individual intakes were recorded, and feed samples and orts were analyzed every 5 d. On d 56, the steers were harvested at a USDA inspected facility, chilled, carcass data recorded at 24 h post-mortem, and fabricated 48 h post-mortem. One *longissimus lumborum* (Striploin: LL) and one *gluteus medius* (Top Sirloin: GM) was obtained from each animal, aged under vacuum (LL: 21 d, GM: 14 d), and cut into 2.54 cm steaks. One steak was used for retail display (9 d) for which subjective color scores as well as objective color scores (using Hunter Mini-Scan: l*, a*, and b*) were recorded on d 0,1,3,5,7, and 9. All samples were analyzed on d 0 and 9 using T-BARs to quantify lipid oxidation. Another steak was weighed and cooked to evaluate purge and cooking loss, and then tenderness was assessed using Warner-Bratzler Shear Force (WBSF). Consumer taste panels were conducted (1 panel for each muscle, 2 total) to determine consumer acceptability.

RP-histidine supplementation improved the lean color, uniformity, and brightness of the product throughout retail display, and decreased surface discoloration, browning, and purge compared with control. Consumers preferred RP treated product over control based upon improved juiciness, increased overall satisfaction and willingness to purchase the product. A trend for increased growth and improved feed to gain was observed in the treated animals. There were no negative effects of RP-treatment in terms of carcass quality (quality grade and yield grade), tenderness, cook loss, texture, flavor, incidence of off-flavor, or pH. Overall, 55 d of pre-harvest RP-histidine treatment positively impacts consumer perception and may optimize product quality and marketability.

Effects of Serum Concentration of Beta-Carotene at Artificial Insemination on Productive and Reproductive Parameters in Lactating Holstein Cows.

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The objective of this study was to determine the effects of beta-carotene concentration in serum at the moment of artificial insemination (AI) on Holstein cows. A total of 497 lactating dairy cows were enrolled. All animals were assigned to a timed AI protocol (CIDR+estradiol benzoate+GnRH-7d-PGF-2d-CIDR-out+PGF+ECP-2d-timed AI). Blood samples and body condition score (BCS) were collected at the moment of AI. Serum beta-carotene was quantified in a single step denaturation and extraction into a solvent, followed by measurement using a portable spectrophotometer (iCheck; BioAnalyt, GmbH, Teltow, Germany). Milk production and herd health records were collected for the entire experimental period, and pregnancy diagnosis was performed by ultrasound 31 d post-AI. Data was analyzed using the MIXED and GLIMMIX procedures of SAS. Animals with BCS ≤ 2.75 had lower ($P < 0.01$) concentration of beta-carotene compared with cows with BCS ≥ 3.0 (3.82 ± 0.09 μg/ml and 4.16 ± 0.06μg/ml, respectively). Multiparous cows had greater concentrations of beta-carotene compared with primiparous cows ($P < 0.01$). There was no correlation between milk production and concentration of beta-carotene ($r < 0.01$; $P > 0.10$), but a quadratic correlation between pregnancy per AI and concentration of beta-carotene ($P = 0.03$) was found. When serum beta-carotene was categorized as low (≤3.21μg/ml), intermediate (> 3.21 and ≤ 4.82 μg/ml) and high (> 4.82μg/ml), cows with intermediate and high concentrations had higher pregnancy per AI (21.6%, 32.4% and 31.6%, respectively; $P = 0.05$). In conclusion, the concentration of beta-carotene was affected by BCS and parity. Animals with higher concentrations of serum beta-carotene had greater pregnancy per AI, suggesting it may be possible to use beta-carotene as a marker for fertility in lactating dairy cows.

Effect of rumen protected vitamin B complex on metabolic parameters, milk production and d 15 conceptus and endometrial outcomes

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Keywords: dairy, cow, reproduction, embryo, vitamin B complex, rumen protection, milk production, BHBA, endometrial gene expression

The aim of this project was to determine the effects of a rumen-protected vitamin B complex supplementation (VIT B) compared with a control diet containing no supplement (CON) on: milk production and components, concentrations of BHBA, haptoglobin and progesterone in plasma, ovarian dynamics and day 15 conceptus and endometrial outcomes. Fifty-one multiparous Holstein cows from the herd at the UBC Dairy Education and Research Centre were enrolled into the study 3 weeks prior to parturition and were randomly assigned to one of the two treatments. Blood samples (2/week), weekly milk samples and daily feed intake were collected. Cows were enrolled onto a double-ovsynch protocol at 33±3 days post-partum and inseminated by timed Artificial Insemination (AI). Ovarian structures were monitored and measured using *per rectum* ultra-sonography. The uterus was flushed on day 15 post AI for conceptus collection and endometrial samples were collected at the same time. Data was analyzed by ANOVA using the GLM procedure of SAS. Overall, 42 cows were flushed and 13 embryos were collected (recovery rate = 31%). Vitamin B supplementation had no affect on the size of the embryo (*P*=0.49), ovulatory follicle size (*P*=0.51) or CL size at embryo collection (*P*=0.51). However, cows with third or higher parity had significantly larger embryos compared to second parity cows (9.39±1.44 vs. 1.73±1.76, *P*<0.05). Milk production (*P*=0.33) and milk fat (*P*=0.81) values were also similar between the two groups. BHBA and haptoglobin levels between the two groups were also identical. Analysis of transcripts related to embryo development, immune system, adhesion and regulation of Vitamin B molecules showed that *OXTR* (*P*=0.04) *MUC5B* (*P*=0.05), *MUC1* (*P*=0.02), *IL1β* (*P*=0.05) *SPP1* (*P*=0.03), *TRD* (*P*=0.03), *FZD8* (*P*=0.05) and *FOLR1* (*P*=0.05) genes were significantly upregulated in the VIT B group. *SELL (P*=0.10), *PLAU* (*P*=0.10) and *MYH9* (*P*=0.10) genes showed a tendency to be more upregulated in the endometrium of VIT B group compared to CON group. In conclusion, strategic dietary vitamin B supplementation during the transition and early lactation did not affect major outcomes of production and reproduction in lactating dairy cows. However, benefits of vitamin B in fertility might potentially be linked to endometrial and conceptus gene expression.