2019

MAY 8 & 9, 2019
DOUBLETREE BY HILTON FRESNO CONVENTION CENTER
FRESNO, CALIFORNIA
IN CONJUNCTION WITH A
TECHNICAL SYMPOSIUM SPONSORED BY
KEMIN ANIMAL NUTRITION & HEALTH

2019 CANC SPONSORSHIPS

PLATINUM SPONSORS:

Adisseo	Beau Williamson	559-288-0050
Alltech	Caley Heiman	559-226-0405
Arm & Hammer Food & Production	Shawna Martinho	559-960-0654
Biomin America	Sarah DeVary	913-296-8911
Chr. Hansen Inc.	Robert McCoy	559-280-7299
DSM Nutritional Products	Ryan Barcellos	559-707-9082
EDGE Dairy Consulting	Michael DeGroot, Ph.D.	559-859-6563
Energy Feeds International	Mike Nolan	858-243-9602
Foster Commodities	Bob Myers	209-457-6520
Kemin	Charity Faber	515-290-8873
Micronutrients	JJ Degan	317-697-2638
milc group	Manuel Soares	800-308-2178
Novita Nutrition	Jill Kosbau	605-610-0975
Nutrius LLC	Patti Murphy	559-897-5862
Origination, Inc.	Jerry Marshall	209-531-3256
Pacific Elements, LLC	Kevin Swager/Janae Costa	209-541-1216
Pine Creek Nutrition Service, Inc.	Daugherty/Tully/Stroup/Brixey	209-667-4181
Rock River Laboratory, Inc.	Zachery Meyer	559-623-1911
Western Milling, LLC	Claudio Ribeiro	559-302-1078

GOLD SPONSORS:

A.L. Gilbert Company	Marit Arana, Ph.D.	209-847-1721
ADM	Gabriela Acetoze, Ph.D., PAS	559-259-2557
Ag Processing Inc.	David Gast	402-492-3309
Associated Feed & Supply Co.	Chris Benevedes	209-664-3357
Balchem Corporation	Dave Ellis	360-335-4406
DairyWorks LLC	Terry Battcher	209-380-2803
Diversified Dairy Solutions LLC	Doug DeGroff	559-280-7880
Elanco Animal Health	Jessica Davis, DVM	559-331-6270
Enzabac Advanced Products	Geoff Gates	208-736-1607
Feedworks USA Ltd.	Peter Williams	530-304-2586
Innovative Liquids LLC	Luis Rodriguez, Ph.D.	916-833-1280
Jefo	Annie St-Pierre	450-799-2000
MicroBasics	Mark Kane-Berman	858-756-9447
Milk Specialties Global	Erin Martin	952-942-7310
Phibro Animal Health Corporation	Karol Ehmen	217-592-1427
Progressive Dairy Solutions, Inc.	Matt Budine	209-847-1377
Protekta Inc./Vilofoss	Morten Jakobsen	519-357-8454
Richardson Oilseed Limited	Francis Kolla	204-934-5153
SoyBest	Charlie Macgregor, Ph.D., PAS	360-631-2112
SoyPlus/SoyChlor	Johnny Ware	602-762-6424
Westway Feed Products LLC	Dan Rigsbee	559-260-0494
William C. Loughlin Co.	Matt Kelly	559-799-0678
Zinpro Corporation	Julian Casas DVM, MPVM	559-747-4390

2019 CANC SPONSORSHIPS Continued.....

SILVER SPONSORS:

CDP KTG-North America	M.J. Bakke, PAS	559-348-3818
Global Animal Products	Ben Tarr/Dave Fischer	806-622-9600
J.D. Heiskell & Company	Amanda Flora	559-685-6100
J.R. Simplot AgriBusiness	Lori Bishop	208-780-2775
Lallemand Animal Nutrition, Inc.	Stuart Gebauer	970-397-1293
NuWest Milling	John Austel	209-883-1163
Vetagro Inc.	Enrica Catellani	773-610-2087
Virtus Nutrition	Reneé Smith	661-699-8552

BRONZE SPONSORS:

Alforex Seeds	Dawn Klawitter	831-540-8379
BioTech Associates Ltd., Inc.	Chuck Parks/Roberta Parks	559-486-1569
Gavilon	Nicole Dominguez	559-688-5151
Mission Ag Resources LLC	Deb Mize	559-591-3333
Penny Newman Grain	James Netto	559-448-8800
Perdue AgriBusiness Animal Nutrition	Detra A. Herrman	717-253-0390
Phileo Lesaffre	Clay Samper, MS	559-676-5925
Pinnacle Premix LLC	Travis Taylor/J. Verstuyft	530-908-2923
Veterinary Service, Inc.	Kelley Thurman	559-485-7474

2019 CANC Student Poster Presentation Sponsors:

BioTech Associates Ltd., Inc.	Chuck Parks/Roberta Parks	559-486-1569
Boer Commodities	Bart Pecchenino	559-253-7055
Custom Dairy Performance, Inc./ KTG-North America	M.J. Bakke, PAS	559-348-3818
James Farrell & Co.	Rick Corra	206-295-7799
Nutri-Systems, Inc.	Andy Riordan	559-298-9134

TABLE OF CONTENTS

TECHNICAL SYMPOSIUM
Sponsored by
Kemin Animal Nutrition & Health

CALIFORNIA ANIMAL NUTRITION CONFERENCE

CALIFORNIA ANIMAL NUTRITION CONFERENCE

Title & Author **Page**

CALIFORNIA ANIMAL NUTRITION CONFERENCE

Title & Author **Page**

Using Amino Acid nutrition to improve productive, health, and reproductive performance in dairy cows

Phil (F. C.) Cardoso*

*Associate Professor, Department of Animal Sciences, University of Illinois, Urbana, IL, USA 61801. E-mail: cardoso2@illinois.edu

■ Take Home Messages

- Nutritional strategies and feeding management during the pre-calving and post-calving periods affect health, productivity and fertility of high-producing dairy cows.
- Management to improve cow comfort and ensure good intake of the ration is pivotal for success.
- Rumen-protected methionine and lysine added to the diet of Holstein cows during the transition period and early lactation improves the survival rate of preimplantation embryos.
- Impacts of the transition program should be evaluated in a holistic way that considers disease occurrence, productivity, and fertility.

■ Introduction

Strategies to improve the reproductive performance of dairy cows include alteration of nutritional status. In other species, dietary supplementation with specific amino acids (AA) (e.g., arginine, glutamine, leucine, glycine, and methionine) had beneficial effects on embryonic and fetal survival and growth through regulation of key signaling and metabolic pathways (Del Curto et al., 2013). Methionine and lysine are the most limiting AA in lactating cows (NRC, 2001), but supplementation of diets with crystalline methionine and lysine has been excluded because free methionine and lysine are quickly and almost totally degraded by the microorganisms in the rumen (NRC, 2001). In contrast, supplementing rumen-protected methionine (RPM) and rumen-protected lysine (RPL) has a positive effect on milk protein synthesis in dairy cows (Ordway, 2009; Osorio et al., 2013). Although the role of methionine in bovine embryonic development is unknown, there is evidence that methionine availability alters the follicular dynamics of the first dominant follicle (Acosta et al., 2017), the transcriptome of bovine preimplantation embryos in vivo (Penagaricano et al., 2013) and the embryonic lipid content (Acosta et al., 2016) which may serve as an energy substrate, improving embryo survivability.

■ Reproduction, Nutrition, and Health

A widespread assumption is that fertility of modern dairy cows is decreasing, particularly for Holstein-Friesen genetics, in part because of unintended consequences of continued selection for high milk production. This assumption has been challenged recently (Leblanc, 2010). There is a wide distribution of reproductive success both within and among herds. For example, within five California herds encompassing 6,396 cows, cows in the lowest quartile for milk yield in the first 90 days postpartum (32.1 kg/day) were less likely to have resumed estrous cycles by 65 days postpartum than cows in quartiles two (39.1 kg/day), three (43.6 kg/day) or four (50.0 kg/day); milk production did not affect risk for pregnancy (Santos et al., 2009). Changes in management systems and inadequacies in management may be more limiting for fertility of modern dairy cows than their genetics per se.

Dairy cows are susceptible to production disorders and diseases during the peripartal period and early lactation, including milk fever, ketosis, fatty liver, retained placenta, displaced abomasum, metritis,

mastitis, and lameness (Mulligan et al., 2006; Roche et al., 2013). There is little evidence that milk yield per se contributes to greater disease occurrence. However, peak disease incidence (shortly after parturition) corresponds with the time of greatest negative energy balance (NEB), the peak in blood concentrations of nonesterified fatty acids (NEFA), and the greatest acceleration of milk yield. Peak milk yield occurs several weeks later. Disorders associated with postpartum NEB also are related to impaired reproductive performance, including fatty liver and ketosis (Mcart et al., 2012). Cows that lost > 1 body condition score (BCS) unit (1-5 scale) had greater incidence of metritis, retained placenta, and metabolic disorders (displaced abomasum, milk fever, ketosis) and a longer interval to first breeding than cows that lost < 1 BCS unit during the transition (Kim and Suh, 2003).

Indicators of NEB are highly correlated with lost milk production, increased disease and decreased fertility. However, the extent to which NEB is causative for peripartal health problems rather than just a correlated phenomenon must be examined critically. For example, in transition cows, inflammatory responses may decrease dry matter intake (DMI), cause alterations in metabolism and predispose cows to greater NEB or increased disease (Graugnard et al., 2012 and 2013). Inducing a degree of calculated NEB in mid-lactation cows similar to what periparturient cows often encounter, does not result in marked increases in ketogenesis or other processes associated with peripartal disease (Moyes et al., 2009). Nevertheless, early postpartal increases in NEFA and decreases in glucose concentrations were strongly associated with pregnancy at first insemination in a timed artificial insemination (TAI) program (Garverick et al., 2013). Although concentrations of NEFA and glucose were not different between cows that ovulated or did not before TAI, probability of pregnancy decreased with greater NEFA and increased with greater glucose concentrations at day three postpartum (Garverick et al., 2013). In support of these findings, early occurrence of subclinical ketosis is more likely to decrease milk yield and compromise fertility. Mcart et al. (2012) reported that cows with subclinical ketosis detected between three and seven days after calving were 0.7 times as likely to conceive to first service and 4.5 times more likely to be removed from the herd within the first 30 days in milk (DIM) compared with cows that developed ketosis at eight days or later.

Cows that successfully adapt to lactation and can avoid metabolic or physiological imbalance are able to support both high milk production and successful reproduction while remaining healthy. Decreased fertility in the face of increasing milk production may be attributed to greater severity of postpartal NEB resulting from inadequate transition management or increased rates of disease. Competition for nutrients between the divergent outcomes of early lactation and subsequent pregnancy will delay reproductive function. Because NEB interrupts reproduction in most species, including humans, inappropriate nutritional management may predispose cows to both metabolic disturbances and impaired reproduction. Cows must make "metabolic decisions" about where to direct scarce resources, and in early lactation, nutrients will be directed to milk production rather than to the next pregnancy.

Different nutritional strategies have been proposed to improve reproduction of the dairy cow with no detrimental effect on lactation performance. Feeding high quality forages, controlled-energy diets, or supplemental fat in the diet are some of the most common ways to improve energy intake in cows (Cardoso et al., 2013; Drackley and Cardoso, 2014). Reproduction of dairy cattle may benefit by maximizing DMI during the transition period, and minimizing the incidence of periparturient problems (Cardoso et al., 2013; Drackley and Cardoso, 2014).

■ Dietary Considerations during the transition period

Controlling energy intake during the dry period to near calculated requirements leads to better transition success (Dann et al., 2005 and 2006; Janovick et al., 2011 Graugnard et al., 2012 and 2013). Cows fed even moderate-energy diets (1.50 – 1.60 Mcal NEL/kg DM) will easily consume 40–80% more energy (net energy of lactation; NEL) than required during both far-off and close-up periods (Dann et al., 2005 and 2006). Cows in these studies were all less than 3.5 BCS (1-5 scale) at dry-off and were individually fed a total mixed ration (TMR) based on corn silage, alfalfa silage, and alfalfa hay with some concentrate supplementation. We have no evidence that the extra energy and nutrient intake was beneficial in any way. More importantly, our data indicate that allowing cows to over-consume energy even to this degree

may predispose them to health problems during the transition period if they face stressors or challenges that limit DMI (Cardoso et al., 2013).

Prolonged over-consumption of energy during the dry period can decrease post-calving DMI. Over-consuming energy results in negative responses of metabolic indicators, such as higher NEFA and beta-hydroxybutyrate (BHB) in blood and more triacylglycerol (TAG) in the liver after calving (Janovick et al., 2011). Alterations in cellular and gene-level responses in liver (Loor et al., 2006) and adipose tissue (Ji et al., 2012) potentially explain many of the changes at the cow level. Over-consumption of energy during the close-up period increases the enzymatic "machinery" in adipose tissue for TAG mobilization after calving, with transcriptional changes leading to decreased lipogenesis (fat synthesis), increased lipolysis (fat utilization) and decreased ability of insulin to inhibit lipolysis (Ji et al., 2012). Controlling energy intake during the dry period also improved neutrophil function postpartum (Graugnard et al., 2012) and so may lead to better immune function.

Allowing dry cows to consume more energy than required, even if cows do not become noticeably over-conditioned, results in responses that would be typical of overly fat cows. Because energy that cows consume in excess of their requirements must either be dissipated as heat or stored as fat, we speculated that the excess is accumulated preferentially in internal adipose (fat) tissue depots in some cows. Moderate over-consumption of energy by non-lactating cows for 57 days led to greater deposition of fat in abdominal adipose tissues (omental, mesenteric, and perirenal) than in cows fed a high-bulk diet to control energy intake to near requirements (Drackley et al., 2014). The NEFA and signaling molecules released by visceral adipose tissues travel directly to the liver, which may cause fatty liver, subclinical ketosis and secondary problems with liver function.

Data from our studies support field observations that controlled-energy dry cow programs decrease health problems (Beever, 2006). Other research groups (Holtenius et al., 2003; Vickers et al., 2013) have reached similar conclusions about controlling energy intake during the dry period, although not all studies have shown benefits (Winkleman et al., 2008). Application of these principles can be through controlled limit-feeding of moderate energy diets or ad libitum feeding of high-bulk, low-energy rations (Janovick et al., 2011; Ji et al., 2012).

Nutritionally complete diets must be fed and the TMR must be processed appropriately so that cows do not sort the bulkier ingredients. Feeding bulky forage separately from a partial TMR, or improper forage processing will lead to variable intake among cows, with some consuming too much energy and some too little. Underfeeding relative to requirements, where nutrient balance also is likely limiting, leads to increased incidence of retained placenta and metritis (Mulligan et al., 2006). Merely adding a quantity of straw to a diet is not the key principle; rather, the diet must be formulated to limit the intake of energy (approximately 1.3 Mcal NEL/kg DM, to limit intake to about 15 Mcal/day for typical Holstein cows) but meet the requirements for protein, minerals and vitamins. Reports of increased transition health problems or poor reproductive success with "low energy" dry cow diets must be examined carefully to discern whether nutrient intakes were adequate.

Less is known about diet formulation for the immediate postpartum period to optimize transition success and subsequent reproduction. Increased research is needed in this area. Proper dietary formulation during the dry period or close-up period will maintain or enable rumen adaptation to higher grain diets after calving. Failure to do so may compromise early lactation productivity. For example, Silva-del-Rio et al. (2010) attempted to duplicate the dietary strategy of Dann et al. (2006) by feeding either a low-energy far-off diet for five weeks followed by a higher-energy diet for the last three weeks before parturition, or by feeding the higher-energy diet for the entire eight-week dry period. They found that cows fed the higher-energy diet for only three weeks before parturition produced less milk than cows fed the diet for eight weeks (43.8 vs. 48.5 kg/day). However, the far-off dry period diet contained 55.1% alfalfa silage and 38.5% wheat straw but no corn silage. In comparison, the higher-energy dry period diet and the early lactation diet both contained 35% corn silage. Ruminal adaptation likely was insufficient for cows fed the higher energy diet for only three weeks.

A major area of concern in the fresh cow period is the sudden increase in dietary energy density leading to subacute ruminal acidosis (SARA), which can decrease DMI and digestibility of nutrients. Adequate physical form of the diet, derived either from ingredients or mixing strategy, must be present to stimulate ruminal activity and chewing behavior, although good methods to quantify "adequacy" remain elusive. Dietary starch content and fermentability likely interact with forage characteristics and ration physical form. Dann and Nelson (2011) compared three dietary starch contents (primarily from corn starch) in the fresh cow period for cows fed a controlled energy-type ration in the dry period. Milk production was greatest when starch content was moderate (23.2% of DM) or low (21.0% of DM) in the fresh cow diet compared with high (25.5% of DM). If SARA decreases DMI and nutrient availability to the cow, NEFA mobilization and increased ketogenesis may follow. In addition, rapid starch fermentation in the presence of NEFA mobilization leads to bursts of propionate reaching the liver, which may decrease feeding activity and DMI according the hepatic oxidation theory (Allen et al., 2009). A moderate starch content (23-25% of DM) with starch of moderate fermentability (e.g., ground dry corn rather than high-moisture corn or ground barley) along with adequate effective forage fibre may be the best strategy for fresh cows. Recent research also has demonstrated that high grain diets can lead to greater numbers of gram-negative bacteria such as *E. coli* with resulting increases in endotoxin present in the rumen, which may decrease barrier function and inflammatory responses in the cow (Zebeli and Metzler-Zebeli, 2012).

Supplemental fats have been widely investigated as a way to increase dietary energy intake and improve reproduction. A novel strategy is to use polyunsaturated fatty acid (PUFA) supplements to improve reproduction (Silvestre et al., 2011). Cows fed calcium salts of safflower oil from 30 days before to 30 days after calving, followed by calcium salts of fish oil to 160 days postpartum, had greater pregnancy rates and higher milk production. The mechanism is believed to be provision of greater amounts of linoleic acid (omega-6 PUFA) until early postpartum, which improves uterine health, followed by greater amounts of omega-3 PUFA from fish oil to decrease early embryonic loss (Thatcher et al., 2011). The effects of turbulent transitions on reproduction are established early postpartum, likely during the first ten days to two weeks postpartum (Mcart et al., 2012; Garverick et al., 2013). By eight weeks postpartum, > 95% of cows should be at or above energy balance (Sutter and Beever, 2000). Use of targeted prepartum and postpartum strategies may minimize health problems and lessen NEB, and thereby improve subsequent fertility.

■ The Importance of Amino Acids

Some AA are limiting for optimal milk production as evidenced by an increase in milk yield, and milk protein yield, and percentage after supplementation with specific, rumen-protected AA. The first two limiting AA for milk production are considered to be methionine and lysine (NRC, 2001). In addition, many AA can have positive effects on physiological processes that are independent of their effects on synthesis of proteins (Wu, 2013). Fertilization and the first few days of embryo development occur in the oviduct. By about five days after estrus the embryo arrives in the uterine horn. The embryo reaches the blastocyst stage by six to seven days after estrus. The embryo hatches from the zona pellucida by about day nine after estrus and then elongates on days 14–19. The elongating embryo secretes the protein interferon-tau that is essential for rescue of the corpus luteum and continuation of the pregnancy. By day 25–28 the embryo attaches to the caruncles of the uterus and begins to establish a vascular relationship with the dam through the placenta. During all the time prior to embryo attachment, the embryo is free-floating and is dependent upon uterine secretions for energy and the building blocks for development, including AA. Thus, it is critical to understand the changes in AA concentrations in the uterus that accompany these different stages of embryo development.

The lipid profile of oocytes and the early embryo can be influenced by the environment of the cow. Our group ran a trial to determine the effect of supplementing rumen-protected methionine on DNA methylation and lipid accumulation in preimplantation embryos of dairy cows (Acosta et al., 2016). Lactating Holsteins entering their 2nd or greater lactation were randomly assigned to two treatments from 30 ± 2 DIM to 72 ± 2 DIM: control (CON; n = 5, fed a basal diet with a 3.4:1 lysine:methionine) and methionine (MET; n = 5, fed the basal diet plus Smartamine M to a 2.9:1 lysine:methionine). Cows were superovulated (FSH) and embryos were flushed 6.5 days after artificial insemination. Embryos with stage of development four or greater were used for analysis. For lipids, fluorescence intensity of Nile Red

staining was compared against a negative control embryo (subtraction of background). Thirty-seven embryos were harvested from cows (MET = 16; CON = 21). Cows receiving MET had greater lipid accumulation (7.3 arbitrary units) compared with cows receiving CON (3.7 arbitrary units). There were no treatment effects on numbers of cells or stage of development. In conclusion, cows supplemented with methionine produced embryos with higher lipid concentration compared with CON cows; this lipid could potentially serve as an important source of energy for the early developing embryo.

The requirements for complete development of bovine embryos have not yet been determined. Current culture conditions allow development of bovine embryos to the blastocyst stage (day 7-8) and even allow hatching of a percentage of embryos (day 9); however, conditions have not been developed in vitro that allow elongation of embryos. The methionine requirement for cultured pre-implantation bovine embryos (day 7-8) was determined in studies from University of Florida (Bonilla et al., 2010). There was a surprisingly low methionine requirement (7 µm) for development of embryos to the blastocyst stage by day seven; however, development to the advanced blastocyst stage by day seven appeared to be optimized at around 21 µm (Bonilla et al., 2010). Thus, the results of these studies indicated that development of morphologically normal bovine embryos did not require elevated methionine concentrations (>21 µm), at least during the first week after fertilization. Stella (2017) reported the plasma concentration of cows fed RPM or not (CON);it seems that cows fed RPM have plasma methionine concentration greater than 20 µm.

Researchers at the University. of Wisconsin (Toledo et al., 2015) conducted a trial with 309 cows (138 primiparous and 171 multiparous) that were blocked by parity and randomly assigned to two treatments: 1) CON: cows fed a ration formulated to deliver 2500 g of metabolizable protein (MP) with 6.9% lysine and 1.9% Met (as a % of MP) and 2) RPM: cows fed a ration formulated to deliver 2500 g of MP with 6.9% lysine and 2.3% Met (as a % of MP). Cows were randomly assigned to three pens with headlocks and fed a single basal TMR twice daily. From 28 to 128 DIM, after the morning milking, cows were headlocked for 30 minutes and the TMR of CON and RPM cows were individually top dressed with 50 g of distillers dried grains (DDG) or a mix of 29 g of DDG and 21 g of Smartamine M), respectively. Following a double Ovsynch protocol, cows were inseminated and pregnancy checked at 28 days (plasma Pregnancy Specific Protein-B concentration), and at 32, 47 and 61 days (ultrasound). Individual milk samples were taken once per month and analyzed for composition. There were no statistical differences in milk production, but milk from RPM cows had a higher protein concentration. Cows fed the methionine enriched diet tended (P = 0.08) to have a lower pregnancy loss from 28 to 61 days after AI (16.7 % CON cows vs. 10.0% in RPM cows). Pregnancy losses between days 28 and 61 were not different in the primiparous cows (12.8% CON and 14.6% RPM), however, pregnancy losses were lower (P = 0.03) in multiparous cows that received the methionine enriched diet (19.6% CON vs. 6.1% RPM; Toledo et al., 2017).

Perhaps the most detrimental impact of NEB on reproductive performance is delayed return to cyclicity. Dominant follicle (DF) growth and estradiol (E2) production are key factors for a successful conception, and their impairment can be attributed to reduced luteinizing hormone (LH) pulses and decreased circulating insulin and IGF-I concentrations (Komaragiri and Erdman, 1997). Furthermore, immune function is also suppressed during the periparturient period. Negative energy balance and fatty liver syndrome have been shown to impair peripheral blood neutrophil function (Hammon et al., 2006). Acosta et al. (2017) reported that methionine and choline supplementation induced a down regulation of pro-inflammatory genes, possibly indicating lower inflammatory processes in follicular cells of the first DF postpartum.

Additionally, supplementing methionine during the transition period increased 3β-hydroxysteroid dehydrogenase (3b-HSD) expression in the follicular cells of the first DF postpartum. Higher methionine concentrations in the follicular fluid of supplemented cows can potentially affect oocyte quality. Understanding how this may affect reproductive performance in commercial farms needs to be further investigated. Batistel et al. (2017) reported that studies with non-ruminant species argue for the potential relevance of the maternal methionine supply during late gestation in enhancing utero-placental uptake and transport of nutrients. The authors hypothesized that the greater newborn body weight from cows fed RPM compared with CON (42 vs. 44 kg) could have been a direct response to the greater nutrient supply

from the feed intake response induced by methionine. The fact that certain AA and glucose induce motor signaling to different degrees is highly suggestive of "nutrient specific" mechanistic responses (Figure 1).

■ Conclusions

Formulation and delivery of appropriate diets that limit total energy intake to requirements but also provide proper intakes of all other nutrients before calving can help lessen the extent of NEB after calving. Effects of such diets on indicators of metabolic health are generally positive, suggesting the potential to lessen effects of periparturient disease on fertility. Dietary supplementation of cows with methionine during the final stages of follicular development and early embryo development, until day seven after breeding, led to lipid accumulation changes in the embryos and resulted in differences in gene expression in the embryo. Methionine supplementation seems to impact the preimplantation embryo in a way that enhances its capacity for survival because there is strong evidence that endogenous lipid reserves serve as an energy substrate. The lower pregnancy losses from cows fed a methionine enriched diets suggest that methionine favors embryo survival, at least in multiparous cows.

■ References

Acosta, D.A.V., A.C. Denicol, P. Tribulo, M.I. Rivelli, C. Skenandore, Z. Zhou, D. Luchini, M.N. Corrêa, P.J. Hansen, and F.C. Cardoso. 2016. Effects of rumen-protected methionine and choline supplementation on the preimplantation embryo in Holstein cows. Theriogenology; 85:1669-1679.

Acosta, D.A. V., M.I. Rivelli, C. Skenandore, Z. Zhou, D.H. Keisler, D. Luchini, M.N. Corrêa, and F.C. Cardoso. 2017. Effects of rumen-protected methionine and choline supplementation on steroidogenic potential of the first postpartum dominant follicle and expression of immune mediators in Holstein cows. Theriogenology. 96:1-9.

Allen, M.S., B.J. Bradford, and M. Oba. 2009. Board Invited Review: The hepatic oxidation theory of the control of feed intake and its application to ruminants. J. Anim. Sci. 87:3317-3334.

Batistel, F., A.S.M. Alharthi, L. Wang, C. Parys, Y. Pan, F.C. Cardoso, and J.J. Loor. 2017. Placentome nutrient transporters and motor signaling proteins are altered by methionine supply during late-gestation in dairy cows and are associated with newborn birth weight. The Journal of Nutrition. 147:1640-47.

Beever, D.E. 2006. The impact of controlled nutrition during the dry period on dairy cow health, fertility and performance. Animal Reproduction Science. 96:212-226.

Bonilla, L., D. Luchini, E. Devillard, and P.J. Hansen. 2010. Methionine requirements for the preimplantation bovine embryo. J. Reprod. Dev. 56:527–32.

Cardoso, F.C., S.J. Leblanc, M.R. Murphy, and J.K. Drackley. 2013. Prepartum nutritional strategy affects reproductive performance in dairy cows. J. Dairy Sci. 96:5859–71.

Dann, H.M., D.E. Morin, M.R. Murphy, G.A. Bollero, and J.K. Drackley. 2005. Prepartum intake, postpartum induction of ketosis, and periparturient disorders affect the metabolic status of dairy cows. J. Dairy Sci. 88:3249-3264.

Dann, H.M., N.B. Litherland, J.P. Underwood, M. Bionaz, A. D'Angelo, J.W. McFadden, and J.K. Drackley. 2006. Diets during far-off and close-up dry periods affect periparturient metabolism and lactation in multiparous cows. J. Dairy Sci. 89: 3563-3577.

Dann, H.M. and B.H. Nelson. 2011. Early lactation diets for dairy cattle – focus on starch. In Proceedings of the 2011 Cornell Nutrition Conference for Feed Manufacturers, Syracuse, NY, USA.

Drackley, J.K. and F.C. Cardoso. 2014. Prepartum and postpartum nutritional management to optimize fertility in high-yielding dairy cows in confined TMR systems. Animal. 8:5-14.

Drackley, J.K., R.L. Wallace, D. Graugnard, J. Vasquez, B.F. Richards, and J.J. Loor. 2014. Visceral adipose tissue mass in non-lactating dairy cows fed diets differing in energy density. J. Dairy Sci. 97:3420-3430.

Garverick, H.A., M.N. Harris, R. Vogel-Bluel, J.D. Sampson, J. Bader, W.R. Lamberson, J. N. Spain, M.C. Lucy, and R.S. Youngquist. 2013. Concentrations of nonesterified fatty acids and glucose in blood of periparturient dairy cows are indicative of pregnancy success at first insemination. J. Dairy Sci. 96:181-188.

Graugnard, D.E., M. Bionaz, E. Trevisi, K.M. Moyes, J.L. Salak-Johnson, R.L. Wallace, J.K. Drackley, G. Bertoni, and J.J. Loor. 2012. Blood immunometabolic indices and polymorphonuclear leukocyt e

function in peripartum dairy cows are altered by level of dietary energy prepartum. J. Dairy Sci. 95:1749-1758.
Graugnard, D.E., K.M. Moyes, E. Trevisi, M.J. Khan, D. Keisler, J.K. Drackley, G. Bertoni, and J.J. Loor. 2013. Liver lipid content and inflammometabolic indices in peripartal dairy cows are altered in response to prepartal energy intake and postpartal intramammary inflammatory challenge. J. Dairy Sci. 96:918-935.
Hammon D.S., I.M. Evjen, T.R. Dhiman, J.P. Goff, and J.L. Walters. 2006. Neutrophil function and energy status in Holstein cows with uterine health disorders. Vet Immunol Immunopathol. 13:21-9.
Holtenius, K., S. Agenäs, C. Delavaud, and Y. Chilliard. 2003. Effects of feeding intensity during the dry period. 2. Metabolic and hormonal responses. J. Dairy Sci. 86:883-891.
Ji, P., J. Osorio, J.K. Drackley, and J.J. Loor. 2012. Overfeeding a moderate energy diet prepartum does not impair bovine subcutaneous adipose tissue insulin transduction and induces marked changes in peripartal gene network expression. J. Dairy Sci. 95:4333-4351.
Kim, I.H. and G.H. Suh. 2003. Effect of the amount of body condition loss from the dry to near calving periods on the subsequent body condition change, occurrence of postpartum diseases, metabolic parameters and reproductive performance in Holstein dairy cows. Theriogenology. 60:1445-1456.
Komaragiri M.V. and R.A. Erdman. 1997. Factors affecting body tissue mobilization in early lactation dairy cows. 1. Effect of dietary protein on mobilization of body fat and protein. J Dairy Sci. 80:929-37.
Leblanc, S.J. 2010. Assessing the association of the level of milk production with reproductive performance in dairy cattle. Journal of Reproduction and Development 56(Suppl.), S1-S7.
Loor, J.J., H.M. Dann, N.A. Janovick-Guretzky, R.E. Everts, R. Oliveira, C.A. Green, N.B. Litherland, S.L. Rodriguez-Zas, H.A. Lewin, and J.K. Drackley. 2006. Plane of nutrition prepartum alters hepatic gene expression and function in dairy cows as assessed by longitudinal transcript and metabolic profiling. Physiological Genomics. 27:29-41.
Mcart, J.A., D.V. Nydam, and G.R. Oetzel. 2012. Epidemiology of subclinical ketosis in early lactation dairy cattle. J. Dairy Sci. 95:5056-5066.
Moyes, K.M., J.K. Drackley, J.L. Salak-Johnson, D.E. Morin, J.C. Hope, and J.J. Loor. 2009. Dietary-induced negative energy balance has minimal effects on innate immunity during a Streptococcus uberis mastitis challenge in dairy cows during mid-lactation. J. Dairy Sci. 92:4301-4316.
Mulligan, F.J., L.O'Grady, D.A. Rice, and M.L. Doherty. 2006. A herd health approach to dairy cow nutrition and production diseases of the transition cow. Animal Reproduction Sci. 96:331-353.
NRC. 2001. Nutrient Requirements of Dairy Cattle. Seventh revised edition. Washington, DC: Natl. Acad. Press.
Ordway, R.S., S.E. Boucher, N.L. Whitehouse, C.G. Schwab, and B.K. Sloan. 2009. Effects of providing two forms of supplemental methionine to periparturient Holstein dairy cows on feed intake and lactational performance. J Dairy Sci. 92:5154–66.
Osorio, J.S., P. Ji, J.K. Drackley, D. Luchini, and J.J. Loor. 2013. Supplemental Smartamine M or metasmart during the transition period benefits postpartal cow performance and blood neutrophil function. J Dairy Sci. 96:6248–63.
Penagaricano, F., A.H. Souza, P.D. Carvalho, A.M. Driver, R. Gambra, J. Kropp, K.S. Hackbart, D. Luchini, R.D. Shaver, M.C. Wiltbank, and H. Khatib. 2013. Effect of maternal methionine supplementation on the transcriptome of bovine preimplantation embryos. Plos One. 8:e72302.
Roche, J.R., A.W. Bell, T.R. Overton, and J.J. Loor. 2013. Nutritional management of the transition cow in the 21st century – a paradigm shift in thinking. Animal Production Sci. 53:1000-1023.
Santos, J.E., E.J. Depeters, P.W. Jardon, and J.T. Huber. 2001. Effect of prepartum dietary protein level on performance of primigravid and multiparous Holstein dairy cows. J. Dairy Sci. 84:213–24.
Santos, J.E., H.M. Rutigliano, and M.F. Sa Filho. 2009. Risk factors for resumption of postpartum estrous cycles and embryonic survival in lactating dairy cows. Animal Reproduction Sci. 110:207-221.
Silva-del-Rio, N, P.M. Fricke, and R.R. Grummer. 2010. Effects of twin pregnancy and dry period strategy on milk production, energy balance, and metabolic profile in dairy cows. J. Animal Sci. 88:1048-1060.
Silvestre, F.T., T.S.M. Carvalho, N. Francisco, J.E.P. Santos, C.R. Staples, T.C. Jenkins, and W.W. Thatcher. 2011. Effects of differential supplementation of fatty acids during the peripartum and breeding periods of Holstein cows: I. Uterine and metabolic responses, reproduction, and lactation. J. Dairy Sci. 94:189-204.
Stella, S.L. 2017. Uterine immune mediators and lipid profiles of the preimplantation embryo and endometrial tissue in Holstein cows supplemented with rumen-protected methionine and observed

neutrophil extracellular trap (net) formation in the bovine endometrial tissue. MS Thesis. University of Illinois, Urbana.

Sutter, F. and D.E. Beever. 2000. Energy and nitrogen metabolism in Holstein-Friesen cows during early lactation. Animal Sci. 70:503-514.

Thatcher, W.W., J.E.P. Santos, and C.R. Staples. 2011. Dietary manipulations to improve embryonic survival in cattle. Theriogenology. 76:1619-1631.

Toledo, M.Z., G.M. Baez, A. Garcia-Guerra, N.E. Lobos, J.N. Guenther, E. Trevisol, D. Luchini, R.D. Shaver, M.C. Wiltbank. 2017. Effect of feeding rumen-protected methionine on productive and reproductive performance of dairy cows. PLoS ONE 12:e0189117. https://doi.org/10.1371/journal.pone.0189117

Vickers, L.A., D.M. Weary, D.M. Veira, and M.A.G. von Keyserlingk. 2013. Feeding a higher forage diet prepartum decreases incidences of subclinical ketosis in transition dairy cows. J. Animal Sci. 91:886-894.

Winkleman, L.A., T.H. Elsasser, and C.K. Reynolds. 2008. Limit-feeding a high-energy diet to meet energy requirements in the dry period alters plasma metabolite concentrations but does not affect intake or milk production in early lactation. J. Dairy Sci. 91:1067-1079.

Wu, G, F.W. Bazer, M.C. Satterfield, X. Li, X. Wang, G.A. Johnson, R.C. Burghardt, Z. Dai, J. Wang, and Z. Wu. 2013. Impacts of arginine nutrition on embryonic and fetal development in mammals. Amino Acids. 45:241-256.

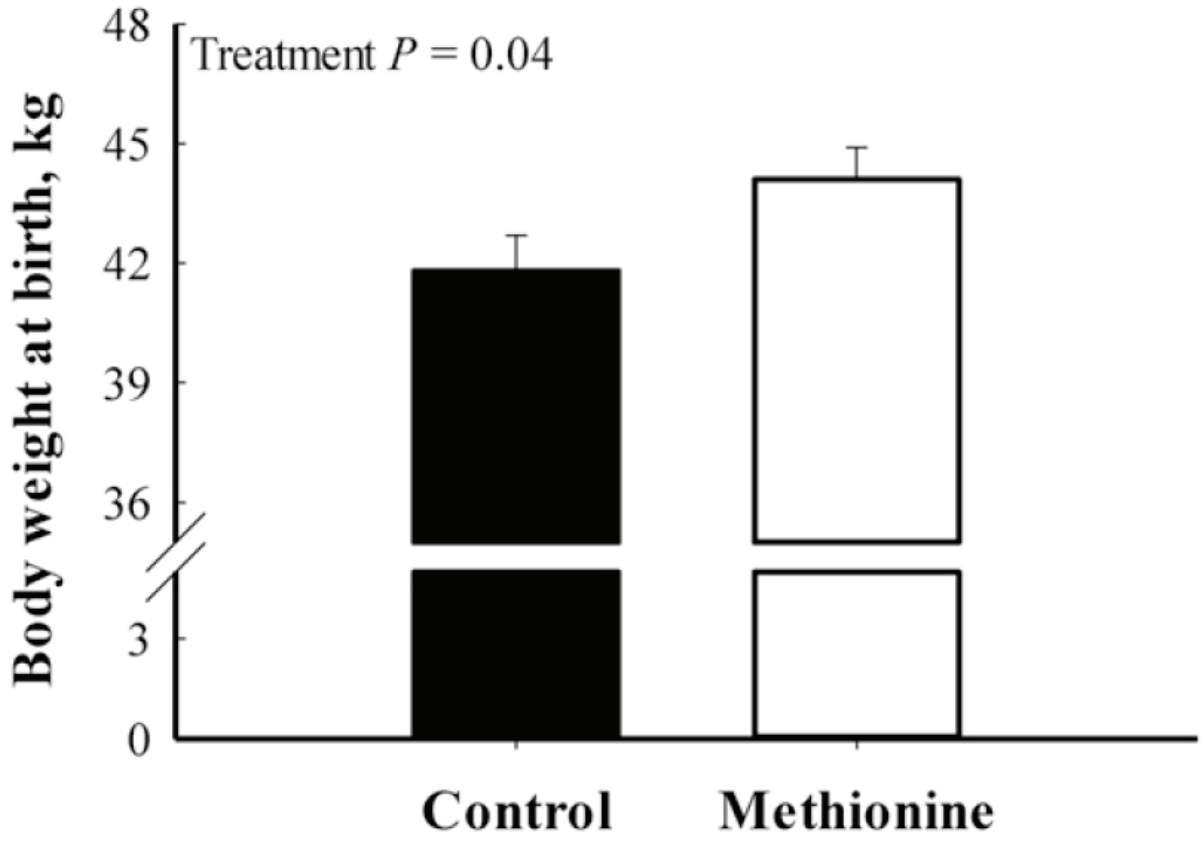

Figure 1. Calf birth body weight (control group, n = 39; methionine group, n = 42) in response to feeding cows a basal control diet or the basal diet plus ethylcellulose rumen-protected methionine (0.9 g/kg dry matter intake) during the last 28 d of pregnancy. Values are means 6 pooled SEMs.

Beyond Metabolizable Protein: Considerations about Amino Acid Nutrition

S.I. Arriola Apelo
University of Wisconsin-Madison, Department of Dairy Science

Why Amino Acid Nutrition?

Rumen undegraded protein (RUP) is the most expensive ingredient in dairy cow diets (Griswold and St-Pierre, 2018). Despite high RUP costs, almost two thirds of the nitrogen (N) consumed by a lactating dairy cow is excreted in urine and feces (Arriola Apelo et al., 2014b). Manure N is a dangerous waste product because its risk of leaching, runoff, volatilization, or denitrification. Nitrogen leakage to the environment may contribute to deterioration of air and water quality, greenhouse emissions, eutrophication of aquatic ecosystems and reduced biodiversity, and human health problems (Leip et al., 2015).

The three largest dairy regions in the U.S. (California, Upper Midwest, and Northeast) are also home to essential fresh water sources, including the California Central Valley Watersheds, Great Lakes, Chesapeake Bay, and Delaware River. With close to 9.4 million lactating cows, waste N from U.S. dairies currently reaches about 1.3 million tons yearly threatening essential water sources for millions of people and the economies around them. Furthermore, U.S. milk production has increased steadily for the last 10 years with no major improvement in N efficiency. In addition, the United Nations forecasts a constant increase in world population, reaching 9.7 billion people by 2050, and the World Health Organization projects a 68% increase in milk consumption over 30 years for South East Asia (http://www.who.int/nutrition), a highly populated region and second destination of U.S. dairy exports (http://www.usdec.org). However, along with the predicted increase in demand for dairy products, more people will need drinkable water and clean air. This perspective imposes a challenge for the dairy industry in the U.S. and worldwide to increase productivity while reducing N emissions. It also presents an opportunity for the producer to reduce dietary costs while improving environmental sustainability and public perception.

However, there are still gross limitations for nutritionists to formulate diets that allow a significant increase in N efficiency without losses in production. Dairy cows have three main N losses. First, ruminally digestible N that exceeds microbial utilization capacity (i.e. high rumen degradable protein, RDP) is absorbed as ammonia and excreted as urea in urine. Second, N from protein not digested in the small intestine is excreted in feces. And third, absorbed AAs (i.e. metabolizable protein, MP) not used by the cow for production purposes are catabolized to urea in the liver and excreted in urine (Arriola Apelo et al., 2014b).

Understanding of rumen and intestinal digestion dynamics and development of rumen protection technologies that conserve protein and AA quality have led to minimum N losses pre-absorption, such that more than 50% of N losses typically happen after AA have been absorbed and even reached the udder. Furthermore, supply of digestible AA that will form the MP pool can be predicted fairly accurately at the individual AA level, at least for the 10 essential AAs that need to be supplied in the diet (NRC, 2001, Tylutki et al., 2008, Van Amburgh et al., 2015, Roman-Garcia et al., 2016, White et al., 2017). Conversely, a limited understanding of AA metabolism postabsorption along with technical difficulties that limit data availability result in an oversimplification of the biology in post-absorptive equations that predict milk protein output. As a result, predictions of MP allowable milk carry systemic error bias, limiting the ability of nutritionist to formulate rations that make the most efficient use of digestible AA.

Four characteristics of current post-absorption equations more likely compromise digestible AA efficiency. These are 1) aggregation of individual AA into the MP pool, 2) fixed MP (or

individual AA) efficiency for milk protein production, 3) the limiting AA theory, and 4) mis-representation of energy and MP/AA interactions. Observations in monogastric species, including lactating sows, show that balancing for individual essential AAs can significantly increase AA efficiency post-absorption to as high as 85% (Baker, 1996, Huber et al., 2015). However, the empirical approach used in monogastric research with large number of observations is not feasible in ruminants. This limitation presents an opportunity for mechanistic research on AA metabolism to contribute towards improved N efficiency for milk protein production, and reduced N excretion by lactating dairy cows.

Fixed post-absorptive efficiency

The aggregated MP pool of digestible AA can be used for different metabolic purposes including maintenance, growth, gestation, reproduction, for which different efficiencies are applied (NRC, 2001). For maintenance and lactation, MP is used with an efficiency of 67%, and MP allowable milk is calculated using a broken line model such that if energy is limiting milk production, the excess MP is entirely wasted (0% MP efficiency).

Observations in vitro and in vivo, as well as comprehensive meta-analysis of the literature have demonstrated that milk protein responses to MP or individual AAs follow a diminishing return type of response (Arriola Apelo et al., 2014b, Lapierre et al., 2018). Using a broken line model results in systematic bias, underperforming models that allow MP efficiency to fluctuate.

Individual AA efficiency

In a lactating dairy cow, the mammary glands and splanchnic tissues extract more than 90% of the AAs in the MP pool (Arriola Apelo et al., 2014b). Essential AAs are classified in two groups based on their metabolism in the mammary glands beyond milk protein production (Mepham, 1982, Lapierre et al., 2012). Interestingly, these two groups of AA also present contrasting differences in affinity by the liver. This degree of complexity limits the ability of a single parameter (i.e. MP efficiency) to accurately represent AA use post-absorption and it requires deeper understanding of individual AA metabolism at systemic and individual tissue level.

Splanchnic AA metabolism. When absorbed, AA need to pass across the portal drained viscera (PDV) and the liver (i.e. splanchnic tissues) before reaching the udder where they can be used for milk protein synthesis. Splanchnic tissues will extract some AA for maintenance functions, but the largest proportion of AA extracted, particularly by the liver, is catabolized into urea for excretion in urine. However, splanchnic tissues have two sources of AAs: intestinal absorption and arterial blood supply, from recycling of AAs not used by peripheral tissues (e.g. the udder). In a high producing dairy cow, splanchnic blood flow can represent about 40% of cardiac output, contributing with 80% of total AA supply (i.e. intestinal absorption represents only 20% of AA supply) (Larsen et al., 2015). Therefore, liver extraction of AA will reflect that 80:20 ratio between artery and intestine.

In each pass, the liver extracts small amounts of Group 2 AAs (<5% on each pass). Extraction of group 1 AA is more variable, and of some AA (e.g. Met and Phe) it can take as much as 15%. However, as the main purpose of AA extraction is catabolism to control plasma levels and prevent harmful fluctuations, hepatic extraction is a fixed proportion of supply. In other words, despite the differences between AAs, hepatic extraction is considered stable with dietary chances, and hence, it could be represented with a fixed efficiency.

Mammary AA metabolism. Unlike splanchnic tissues, the mammary glands have shown some plasticity on their affinity for individual AAs (Bequette et al., 2000, Cantalapiedra-Hijar et al., 2015, Nichols et al., 2016). However, the underlying mechanisms of mammary AA metabolism to safely increase AA extraction efficiency without risking decreasing milk protein production are not well understood.

The high energetic cost of milk protein turnover (Hanigan et al., 2009) suggests that *regulating milk protein synthesis in response to nutrient availability would be a better strategy than merely relying on substrate depletion.* In line with this hypothesis, in one day the blood flow to the udder supplies several folds the amount of AA required for milk protein production. More importantly, Yoder et al. (2018) recently demonstrated that excess AA supplied in blood actually enter and leave the milk producing cells without been used, further supporting the idea that demand, not supply is the limiting factor for milk protein production.

If that is the case, increasing the demand of AAs by the udder would increase retention of AA that are already entering those cells, and it would reduce recycling to peripheral tissues and N losses. Our studies at molecular level suggest that indeed, regulation of milk protein production plays a more important role than supply itself (*unpublished data*), and it may explain all the variation in milk protein production observed in response to AA. Our observations also indicate that the sensor mechanisms that regulate protein production respond to individual AAs independently, regardless of the level of other AAs (Arriola Apelo et al., 2014c, Arriola Apelo et al., 2014d). On the other hand, these observations suggest that energy supply potentiates the effect of AA or dietary protein in milk protein production, something currently not represented in nutrient requirement systems.

Individual AA efficiencies and the limiting AA theory

Extended use of corn and soybean in dairy diets generates an AA imbalance in the MP pool that has led to the identification of Met and Lys as first limiting AAs under those situations. Particularly in high forage, European-type diets, His has been identified as a potentially third limiting AA.

Nutrient requirement systems stand on the limiting AA theory, typically represented by the barrel with staves of different lengths. Based on this theory, if an AA, for example Met, is limiting production, only supplementation with Met will increase the prediction of MP allowable milk (i.e. milk production). Meanwhile, any supplementation with another (non-limited) AA would be fully wasted (0% efficiency).

We first demonstrated in vitro that lactating dairy cow mammary tissue can increase the rate of synthesis of milk proteins in response to one AA regardless of which was the most limiting one (Arriola Apelo et al., 2014d). Later, we supplemented lactating dairy cows fed a protein deficiency diet with rumen protected Lys, Met, or Leu, or combinations of the three (Arriola Apelo et al., 2014a). When compared to a protein adequate diet, supplementation with any single AA overcame the limitation in milk protein yield imposed by dietary protein restriction. More recently, Yoder and collaborators (2018) infused either the first three limiting AA (Met, Lys, and His) or leucine and isoleucine into the jugular vein of lactating dairy cows that were experiencing a total MP deficiency of 498g (1.1lb). Both treatments increased milk protein yield, and the former with an efficiency of 110%, suggesting that this treatment increased the efficiency of other dietary AAs.

The observations described above contradict the limiting AA theory and indicate that additive effects of, at least, some essential AAs on milk protein production are possible. Haque and collaborators demonstrated that maximal levels of milk protein production could be achieved by

supplementing either 4 EAA (Lys, Met, His and Leu) or the 10 of them. This approach could be implemented in future nutritional strategies, in which only the most efficient AAs (productively or economically) are supplemented, reaching the desired milk protein production level and minimizing post-absorptive N losses.

Much research is still needed to improve N efficiency by lactating dairy cows, which is currently impacting both the producer economy and surrounding environment. In addition, the perspective for the next 30 years presses on dairy producers, nutritionists and researchers for an economically viable reduction of waste N production by lactating dairy cows.

References cited

Arriola Apelo, S. I., A. L. Bell, K. Estes, J. Ropelewski, M. J. de Veth, and M. D. Hanigan. 2014a. Effects of reduced dietary protein and supplemental rumen-protected essential amino acids on the nitrogen efficiency of dairy cows. J. Dairy Sci. 97(9):5688-5699.

Arriola Apelo, S. I., J. R. Knapp, and M. D. Hanigan. 2014b. Invited review: Current representation and future trends of predicting amino acid utilization in the lactating dairy cow. J. Dairy Sci. 97(7):4000-4017.

Arriola Apelo, S. I., L. M. Singer, X. Y. Lin, M. L. McGilliard, N. R. St-Pierre, and M. D. Hanigan. 2014c. Isoleucine, leucine, methionine, and threonine effects on mammalian target of rapamycin signaling in mammary tissue. J. Dairy Sci. 97(2):1047-1056.

Arriola Apelo, S. I., L. M. Singer, W. K. Ray, R. F. Helm, X. Y. Lin, M. L. McGilliard, N. R. St-Pierre, and M. D. Hanigan. 2014d. Casein synthesis is independently and additively related to individual essential amino acid supply. J. Dairy Sci. 97(5):2998-3005.

Baker, D. H. 1996. Advances in amino acid nutrition and metabolism of swine and poultry. Pages 41-52 in Nutrient Managment of Food Animals to Enhance and Protect the Environment. E. T. Kornegay, ed. CRC Lewis, Boca Raton, FL.

Bequette, B. J., M. D. Hanigan, A. G. Calder, C. K. Reynolds, G. E. Lobley, and J. C. MacRae. 2000. Amino acid exchange by the mammary gland of lactating goats when histidine limits milk production. J. Dairy Sci. 83(4):765-775.

Cantalapiedra-Hijar, G., I. Ortigues-Marty, and S. Lemosquet. 2015. Diets rich in starch improve the efficiency of amino acids use by the mammary gland in lactating Jersey cows. J. Dairy Sci. 98(10):6939-6953.

Griswold, K. and N. St-Pierre. 2018. Cost of nutrients and relative value of feedstuffs for Pennsylvania dairy farms. Lancaster Farming.

Hanigan, M. D., J. France, S. J. Mabjeesh, W. C. McNabb, and B. J. Bequette. 2009. High rates of mammary tissue protein turnover in lactating goats are energetically costly. J. Nutr. 139(6):1118-1127.

Huber, L., C. F. de Lange, U. Krogh, D. Chamberlin, and N. L. Trottier. 2015. Impact of feeding reduced crude protein diets to lactating sows on nitrogen utilization. J. Anim. Sci. 93(11):5254-5264.

Lapierre, H., M. Larsen, D. Sauvant, M. E. Van Amburgh, and G. Van Duinkerken. 2018. Review: Converting nutritional knowledge into feeding practices: a case study comparing different protein feeding systems for dairy cows. animal 12(s2):s457-s466.

Lapierre, H., G. E. Lobley, L. Doepel, G. Raggio, H. Rulquin, and S. Lemosquet. 2012. Triennial lactation symposium: mammary metabolism of amino acids in dairy cows. J. Anim. Sci. 90(5):1708-1721.
Larsen, M., C. Galindo, D. R. Ouellet, G. Maxin, N. B. Kristensen, and H. Lapierre. 2015. Abomasal amino acid infusion in postpartum dairy cows: Effect on whole-body, splanchnic, and mammary amino acid metabolism. J. Dairy Sci. 98(11):7944-7961.
Leip, A., G. Billen, J. Garnier, B. Grizzetti, L. Lassaletta, S. Reis, D. Simpson, M. A. Sutton, W. Vries, F. Weiss, and H. Westhoek. 2015. Impacts of European livestock production: nitrogen, sulphur, phosphorus and greenhouse gas emissions, land-use, water eutrophication and biodiversity. Environmental Research Letters 10(11):115004.
Mepham, T. B. 1982. Amino acid utilization by lactating mammary gland. J. Dairy Sci. 65(2):287-298.
Nichols, K., J. J. M. Kim, M. Carson, J. A. Metcalf, J. P. Cant, and J. Doelman. 2016. Glucose supplementation stimulates peripheral branched-chain amino acid catabolism in lactating dairy cows during essential amino acid infusions. J. Dairy Sci. 99(2):1145-1160.
NRC. 2001. Nutrient Requirements of Dairy Cattle. 7th ed. National Academy Press, Washington, DC.
Roman-Garcia, Y., R. R. White, and J. L. Firkins. 2016. Meta-analysis of postruminal microbial nitrogen flows in dairy cattle. I. Derivation of equations. J. Dairy Sci. 99(10):7918-7931.
Tylutki, T. P., D. G. Fox, V. M. Durbal, L. O. Tedeschi, J. B. Russell, M. E. Van Amburgh, T. R. Overton, L. E. Chase, and A. N. Pell. 2008. Cornell Net Carbohydrate and Protein System: A model for precision feeding of dairy cattle. Anim. Feed Sci. Technol. 143(1):174-202.
Van Amburgh, M. E., E. A. Collao-Saenz, R. J. Higgs, D. A. Ross, E. B. Recktenwald, E. Raffrenato, L. E. Chase, T. R. Overton, J. K. Mills, and A. Foskolos. 2015. The Cornell Net Carbohydrate and Protein System: Updates to the model and evaluation of version 6.5. J. Dairy Sci. 98(9):6361-6380.
White, R. R., Y. Roman-Garcia, J. L. Firkins, M. J. VandeHaar, L. E. Armentano, W. P. Weiss, T. McGill, R. Garnett, and M. D. Hanigan. 2017. Evaluation of the National Research Council (2001) dairy model and derivation of new prediction equations. 1. Digestibility of fiber, fat, protein, and nonfiber carbohydrate. J. Dairy Sci. 100(5):3591-3610.
Yoder, P., X. Huang, and M. D. Hanigan. 2018. Effects of infused leucine and isoleucine or methionine, lysine, and histidine on cow performance. in Proc. ADSA Annual Meeting, Knoxviille.

Variation of nutrient composition in feed byproducts; implications on balancing dairy rations for amino acids[1]

P.J. Kononoff
Department of Animal Science
University of Nebraska-Lincoln
Lincoln, NE
68583-0908

Introduction

Ruminant production systems, including milk production, have become more efficient over time. When comparing dairying practices and resources needed in 1944 to 2007, Capper et al. (2009) reported that over this time dairy producers used 21% less animals, 23% less feedstuffs, 35% less water, and 10% less land to produce the same one billion kg of milk. Despite this increase in efficiency, increased pressure for land use and high commodity prices over the past decade have increased feed costs for dairy farmers, challenging them to consider less costly sources of protein and fiber (Bradford and Mullins, 2012). In doing so, feed byproducts have played a major role in dairy nutrition but they also represent a means by which efficiency of human food production is improved. This is because when animals consume byproducts they are using a feed resource that from a human perspective would be considered a waste product (VandeHaar and St-Pierre, 2006).

[1]Portions of have been previously published proceedings including: Kononoff, P. J. 2018. Feed co-products: "We're one, but we're not the same." Mid-South Ruminant Nutrition Conference, August 8 and 9, 2018, Grapevine, TX.; Kononoff, P.J. and M.J. de Veth. 2015. Amino acids and dairy rations .... What is there to know about sources and costs. Proceedings of the 76th Annual Minnesota Nutrition Conference. September 16 and 17th . Prior Lake, MN. Pages 124 – 128.

To illustrate this, Karlsson et al. (2018) recently conducted an experiment in which all feedstuffs which could be considered human-edible product were removed from the diet of lactating dairy cows and replaced with feed byproducts. They examined these formulations and their effects on what is known as "human-edible feed conversion efficiency." This is an index which is determined by measuring human-edible material produced by a system minus the human-edible material used by the same system. In this study replacing human edible material (cereal grains and soybean meal) with human-inedible by-products (beet pulp, DDGS, and canola meal) resulted in a net increase in human food protein production without lowering milk production. Although often advantageous, the use of byproducts in dairy cattle diets may also be challenging because they vary in availability and in chemical composition. The objective of this work is to outline the nutritional value of common feed byproducts and to discuss how those can be effectively included in dairy rations.

Variation in the Supply of Protein

Classically speaking dairy rations are based upon some given assumptions of feed characterization. In the field assumptions may be based upon the results of a single lab assay, a collection of data from multiple assays or a default tabular value. In practice we know that the values assumed and that which is delivered to the animal may deviate and as a result it is important to understand common sources of variation. When nutritionists anticipate that the diet may contains less nutrients than the expect the may decide on a final diet that 1) does not include the feed that may vary, 2) minimize that feed, or 3) formulate a diet that contains excess nutrients or "safety factors." The consequence of each is usually

increased cost thus it is important to know the variance in feed composition and to understand the nature of it within a single farm. Several years ago the findings of a large field study were published and the goals of this study were to identify and measure sources of variation in the chemical composition of traditional analytes such as DM, NDF, and CP (St-Pierre and Weiss, 2015). Some of the findings and recommendations to these findings are summarized in Table 1. In general, wide variation in chemical composition of traditional dairy ingredients were observed in this study. As expected the farm to farm variation of forages was large and much less so for concentrate ingredients such as corn and soybean meal. Additionally, the variation of wet byproducts such as wet corn gluten feed, wet brewers grains, and wet distillers grains was also large. It was recommended that for these feeds, multiple samples be assayed at the farm level and data also summarized at the farm level. The California dairy industry make excellent use of many wet byproducts such as vegetable and fruit waste and although not tested the recommendations are likely transposable to these feeds. Currently there is very limited data that sheds light on the extent of variation in amino acids within a feed but it should be noted that the variation in DM and CP has substantial implications when formulating diets for amino acid supply. In one study which evaluation the variation in the chemical composition including amino acids, of byproducts common on California dairy farms (DeGroot et al., 2007). The variation observed in this study was similar to that of St-Pierre and Weiss (2015), Although DDGS do not vary as much as commonly believed, knowing what class of DDGS the feed is from is important (i.e. low vs high fat DDGS). Also, and as expected, when the name of the feed is less descriptive such as "bakery byproduct" variation tends to be high.

In addition to chemical composition, nutritionists should also be aware that both rumen and intestinal digestibility of rumen undegradable protein (RUP) may vary and this may have effects on the supply of amino acids. For example, in the report of (Paz et al. (2014) the average digestibility of RUP (dRUP) of DDGS was observed to be 83.9 ± 10.5 %. This digestibility coefficient is similar to the NRC (2001) assumption of 80% but the reported estimates were also highly variable ranging from 59.2 - 95.0%. Similar variation in ruminal and intestinal digestibility has been reported in German canola meal (Steingass et al., 2013). Analytically speaking measures of rumen and intestinal digestibility is laborious and have traditionally been estimated by the mobile bag technique (Hvelplund et al., 1992) and in vitro techniques (Calsamiglia and Stern, 1995; Gargallo et al., 2006; Ross et al., 2013)have been developed to not only estimate digestibility but serve as a tool to be used in routine analysis to track variability.

Reducing Nitrogen Excretion

To this point we have discussed the value of protein and amino acids in dairy production, but it should also be noted that excessive feeding of dietary protein results in negative environmental impacts because the practice contributes to both air and water nitrogen pollution. Recently, considerable research has been conducted to identify nutritional methods to reduce the amount of protein fed so nitrogen utilization by the cow is maximized and excess nitrogen is not lost to the environment. Replacment of nutrients from rumen protected technologies may allow a nutritionist to formulate a diet with less "safety factors" and with less protein and consequently reduce nitrogen excretion by the animals. As producers in consultation with their nutritionists consider sources of protein

and the utilization, the overall goal should be to match the animals needs with the overall protein supply. This can be done by using ration balancing software to estimate the supply of protein and amino acids, and in some cases, the inclusion of commercial available rumen protected amino acids.

Conclusions

The use of byproducts by the dairy industry is a practice that will continue. Such a practice increases the environmental sustainability of the industry because byproducts are resources that are unfit for human consumption. Feeding studies indicate that these feeds also supply valuable nutrients but subtle differences in availability of nutrients require good understanding of chemical composition and in some cases, in vitro testing provides information that can be contribute to our need for knowledge on whole animal nutrient supply and utilization.

References

Bradford, B.J., and C.R. Mullins. 2012. Invited review: Strategies for promoting productivity and health of dairy cattle by feeding nonforage fiber sources. J. Dairy Sci. 95:4735–4746. doi:10.3168/jds.2012-5393.

Calsamiglia, S., and M.D. Stern. 1995. A three-step in vitro procedure for estimating intestinal digestion of protein in ruminants.. J. Anim. Sci. 73:1459–1465.

Capper, J.L., R.A. Cady, and D.E. Bauman. 2009. The environmental impact of dairy production: 1944 compared with 2007. J. Anim. Sci. 87:2160–2167. doi:10.2527/jas.2009-1781.

DeGroot, M.A., J.R. Miller, M.J. Arana, and E.J. DePeters. 2007. Variability in chemical composition and digestibility of twelve by-product feedstuffs utilized in the california dairy industry1. Prof. Anim. Sci. 23:148–163.

Gargallo, S., S. Calsamiglia, and A. Ferret. 2006. Technical note: A modified three-step in vitro procedure to determine intestinal digestion of proteins. J. Anim. Sci. 84:2163–2167.

Hvelplund, T., M.R. Weisbjerg, and L.S. Andersen. 1992. Estimation of the true digestibility of rumen undegraded dietary protein in the small intestine of ruminants by the mobile bag technique. Acta Agric. Scand. -Anim. Sci. 42:34–39.

Karlsson, J., R. Spörndly, M. Lindberg, and K. Holtenius. 2018. Replacing human-edible feed ingredients with by-products increases net food production efficiency in dairy cows. J. Dairy Sci. 101:7146–7155. doi:10.3168/jds.2017-14209.

Paz, H.A., T.J. Klopfenstein, D. Hostetler, S.C. Fernando, E. Castillo-Lopez, and P.J. Kononoff. 2014. Ruminal degradation and intestinal digestibility of protein and amino acids in high-protein feedstuffs commonly used in dairy diets. J. Dairy Sci. 97:6485–6498. doi:10.3168/jds.2014-8108.

Ross, D.A., M. Gutierrez-Botero, and M.E. Van Amburgh. 2013. Development of an in vitro intestinal digestibility assay for ruminant feeds. Pages 190–202 Proceedings of the Cornell Nutrition Conference for Feed Manufacturers. Cornell Univ., Ithaca, NY, East Syracuse, NY.

Steingass, H., G. Kneer, G. Wischer, and M. Rodehutscord. 2013. Variation of in situ rumen degradation of crude protein and amino acids and in vitro digestibility of undegraded feed protein in rapeseed meals. animal 7:1119–1127. doi:10.1017/S175173111300030X.

St-Pierre, N.R., and W.P. Weiss. 2015. Partitioning variation in nutrient composition data of common feeds and mixed diets on commercial dairy farms. J. Dairy Sci. 98:5004–5015. doi:10.3168/jds.2015-9431.

VandeHaar, M.J., and N. St-Pierre. 2006. Major advances in nutrition: Relevance to the sustainability of the dairy industry. J. Dairy Sci. 89:1280–1291.

Table 1. Summary of general sampling recommendations based on on-farm observations of variation in feed composition (St-Pierre and Weiss, 2015).

Farm variation	**Feed**	**Recommendation**
Farm WAS a significant source of variation	Corn silage Haycrop silages Wet corn gluten feed Wet brewers grains Wet DDGS High moisture corn	Multiple samples to be taken at the farm level and summarized by farm
Farm WAS NOT a significant source of variation	Dry corn Soybean meal Dry corn gluten feed Cottonseed	Use data from feed tables
Farm was OFTEN a significant source of variation	DDGS	If you don't know the source test

Modeling and Integrating Metabolizable Energy and Protein Supply and Requirements in Dry and Lactating Dairy Cattle to Optimize Nitrogen Utilization

Mike Van Amburgh, Debbie Ross, Sam Fessenden, Ryan Higgs,
Marcelo Gutierrez-Botero, Andres Ortega and Andrew LaPierre

Dept. of Animal Science, Cornell University
Ithaca, NY

Introduction

Improving the prediction of supply and use of metabolizable energy (ME) and protein (MP) is dependent on several factors that can be measured routinely or predicted with reasonable precision. The prediction of ME is dependent on factors such as total feed intake, the chemical composition of the feed consumed, and ruminal and post-ruminal digestibility. The prediction of MP is dependent on the same factors, although MP is more complex as it is highly dependent on the quantity, profile and digestibility of amino acids that escapes the rumen, whereas substrates for ME can be absorbed anywhere along the GIT, recognizing how those substrates are partitioned are different as they are absorbed farther down the GIT. Feed protein is one of the most expensive macronutrients in dairy cattle diets, and overfeeding degradable protein relative to rumen requirements results in excessive N losses to the environment (Huhtanen and Hristov, 2009). Efficient use of feed N can be achieved by first meeting the requirements of the rumen microbial population, followed by balancing diets to meet the amino acid requirements of the cow. To decrease competition for quality protein that could otherwise be fed to humans, dairy cattle are fed byproducts of human food production, thereby converting waste products into highly valuable milk protein and other nutrients.

To frame the thesis of this paper, the modeling approach used in Cornell Net Carbohydrate and Protein System (CNCPS) will be utilized to describe the relationships and accounting necessary to integrate ME, MP and amino acid supply and requirements (Tylutki et al., 2008; Higgs, 2014; Van Amburgh et al., 2015). There are at least five major steps necessary to improve the prediction of MP and AA supply and requirements in a lactating and dry cow. Most of this discussion will involve basic structural changes in thinking relative to predictions and requirements. Those five areas are the use, characterization and application of crude protein, recycled urea and endogenous protein, intestinal digestibility and determining first limiting nutrients through integration of protein requirements with energy supply.

Crude protein

Crude protein is an antiquated metric and one that is difficult to apply to ruminants in general. A previous review and meta-analysis by Ipharaguarra and Clark (2005) clearly pointed out the inadequacy of using CP to describe milk yield. The first concern with CP is that it is a ratio of (N x 6.25)/DM which makes it difficult to partition into the amount of

"CP" required to meet the ruminal requirements (rumen degradable protein) for fermentation, microbial digestion and yield, and the amount of CP that escapes fermentation (rumen undegradable protein), especially if you are attempting to predict the AA supply to the cow. Further, as pointed out by Jones (1941) not all protein uses the same correction factor and this becomes more problematic as you attempt to describe the degradation of plant material through the gastrointestinal tract as the N content and thus the protein content changes (Van Soest, 1994). This, coupled with the ubiquitous use of 6.25 for all feeds analyzed and used in feed libraries creates variation in predicting individual MPAA supplies to the cow. Overall, to better predict MP, there has to be consideration for the AA within the MP from feed, endogenous protein and microbes as that is what the cow requires for milk yield and productivity. Some of the current approaches commonly used might confound this approach.

Using soybean meal as an example, starting with 3,000 g of soybean meal which is 51.5% CP or 8.24% N and 1.34% methionine on a CP basis or 0.64% methionine on a DM basis the grams of content can be calculated either way. In Table 1, the left column uses a typical approach of methionine as a percent CP which results in 20.7 g of methionine in the soybean meal whereas on the right side the calculation was made on a DM and N basis resulting in 19.2 g methionine, a difference of 1.5 g. Although this difference looks modest, when arrayed across all of the feed and other sources of AA leads to significant differences, especially when attempting to reconcile diets to a few grams during formulation.

Table 1. Grams of methionine in soybean meal calculated as a percent of crude protein or on a nitrogen basis.

Grams CP	3,000 x 0.515 = 1,545 g	Grams N	3000 x 0.0824=247.2
Met., %CP	1.34	Met. N, %DM	0.64/0.0824 =7.77%
Met., g	(1,545 g x 0.0134) = 20.7 g	Met., g	247.2 x 0.0777= 19.2
		Or g x Met DM%	3000 x 0.0064 = 19.2

Rumen ammonia, recycled urea and endogenous protein

To improve the ability to predict AA supply and requirements, separating the ruminal N requirements for microbial growth from the post-ruminal AA requirements and supply is an important step in moving in a better direction for predicting AA supply for the cow and helping reconcile the difference in N partitioning throughout the gastrointestinal tract. One important factor that strongly influences rumen NH_3-N is feed intake pattern, which in turn, impacts microbial growth and N demands and supply. For example, a comparison of predicted NH_3-N using continuous intake, 4 meals/d and 8 meals/d is in Figure 1. For this evaluation, microbial growth in the model becomes limited when rumen NH_3-N falls below 5.0 mg/dl (Satter and Roffler, 1975). The variation in feed intake causes the oscillating behavior observed in Figure 1, where NH_3-N falls below 5.0 mg/dl when the meal pattern is 4 meal/d. The effect of N recycling within the model is evident as rumen NH_3-N slowly increases until the next meal is consumed although the rate of change and the nadir will be very dependent on the total amount of N being fed in the diet. A similar

pattern in rumen ammonia supply was presented by Schwab et al. (2005) using in-vivo data from cattle fed various diets with ranges in DM and N intake. When modeling continuous feed intake with 8 meal/d, the rumen NH_3-N concentration remains above 5.0 mg/dl, demonstrating the importance of feed intake pattern on rumen N supply. This suggests that improper time budgets and inadequate feed availability will negatively affect rumen N balance even though full urea-N recycling is being calculated in the example below. In response to intake restrictions, diet N content might need to be increased to keep rumen ammonia above the desired level for adequate microbial function depending on the level of N in the diet.

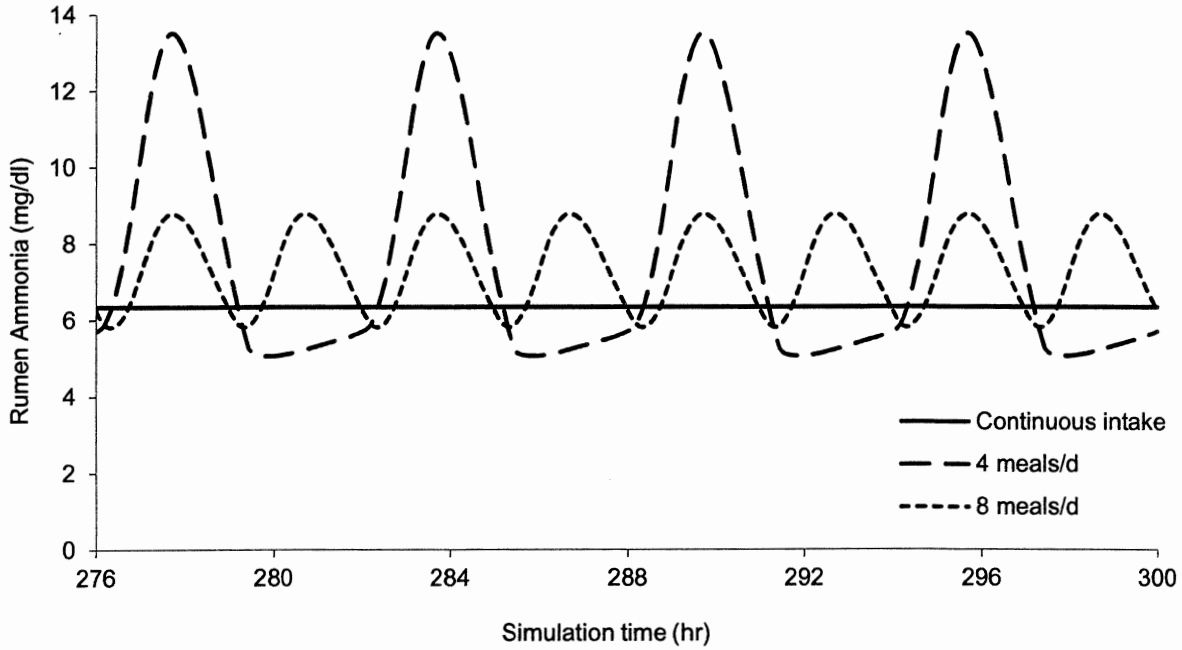

Figure 1. Variation in rumen NH_3-N (mg/dl) among three different meal intake distributions represented by continuous intake, four meals per day and eight meals per day. The ammonia concentration does not decrease below 5 mg/dl in this example because below that value, microbial yield is decreased thus offsetting the demand and reducing feed digestibility accordingly. The model becomes stable after 280 h of simulation time.

Ruminants have a remarkable ability to recycle N back to the GIT in order to sustain favorable conditions for microbial protein function. The recycling of N appears to be an obligate function with a low energy requirement (Reynolds, 1992). While previous versions of the CNCPS model have accounted for N recycling (Fox et al., 2004), the dynamics are difficult to capture in a static model. A great deal of work has been published that attempts to describe the mechanisms and quantitative aspects of N recycling, and while the exact mechanisms remain evasive, quantitative aspects of N fluxes are reasonably well understood and described (Lapierre and Lobley, 2001; Marini et al., 2008; Marini and Van Amburgh, 2003; Recktenwald et al., 2014).

The proportion of urea returned to the GIT relative to urea production is remarkably uniform among experiments when animals are fed diets at, or in moderate excess of MP

requirements (Lapierre et al., 2004; Ouellet et al., 2004; Recktenwald, 2007; Valkeners et al., 2007). However, recycling increases when N supply is limited (Reynolds and Kristensen, 2008; Valkeners et al., 2007) and decreases when N supply is greatly in excess (Lapierre et al., 2004; Reynolds and Kristensen, 2008). To estimate the proportion of urea returned to the GIT, the equations presented in Recktenwald et al. (2014) and Reynolds and Kristensen (2008) were used in combination in v7 of CNCPS. Recktenwald et al. (2014) showed a linear relationship between urea production and urea recycling in high producing cows fed diets ranging from 15% - 17% CP, while, Reynolds and Kristensen (2008) showed an increase in the proportion of urea recycled at very low N intakes and a plateau at higher N intakes. Therefore, using equations from Recktenwald et al. and Reynolds and Kristensen in combination allowed for a wider range in dietary conditions to be represented. Recycled urea is distributed to the rumen, large intestine or small intestine and continues to cycle through the system at steady state based primarily on the size of the overall N pool in the animal. Overall, between 50 and 70% of intake N is converted to urea N and approximately 50% of the urea production is recycled into the GIT when N intake is within the linear portion of the relationship. The total pool size of N in the animal dictates how much of the hepatic urea production is captured in the rumen or excreted in the urine. As N intake increases, the probability of recycled urea N being captured by the microbes is reduced; thus, the objective during formulation is to find the balance between the ruminal requirements for ammonia N, N intake and the capacity for N recycling to ensure the optimum efficiency of use.

Urea N is not the only recycled N source in cattle. The endogenous nitrogen (EN) secretions occur at various places along the gastrointestinal tract (GIT). Important sources include saliva, gastric juices, bile, pancreatic secretions, sloughed epithelial cells and mucin (Tamminga et al., 1995). Gross EN to the forestomach and intestines were calculated in the CNCPS v7 based on the data from Ouellet et al. (2002) and Ouellet et al. (2010). The data were subsequently partitioned into individual components (Table 2) using estimates reported in Egan et al. (1984). Endogenous contributions are reasonably consistent among diets when expressed relative to DMI or OMI (Marini et al., 2008; Ouellet et al., 2010; Ouellet et al., 2002; Tamminga et al., 1995). Thus, the model expresses each component as g EN per kg DMI. Again, this information is necessary to fully characterize the AA supply available at the small intestine. Taken together, the endogenous N supply can contribute up to 20% of the AA arriving at the small intestine. Further, data generated by Ouellet and Lapierre have demonstrated at least half of the endogenous N is utilized by the rumen microbes, thus half of the endogenous N arrives in free form and the rest is incorporated in to rumen microbes.

Intestinal digestibility

Current cattle diet formulation models rely on library estimates of intestinal digestibility of proteins and carbohydrates to predict metabolizable energy (ME) and protein (MP) supply (NRC, 2001; Fox et al., 2004; Tylutki et al., 2008). As models become more accurate and precise in the prediction of nutrient supply and nutrient balance, there is a greater need to evaluate and be able to adapt the inputs currently used as static library values. As diets are formulated closer to the MP requirements of cattle and subsequently

lower in N content, accurate estimates of intestinal digestibility (ID) or indigestibility of protein and amino acids are increasingly important to ensure an adequate supply of those nutrients. Use of outdated feed library values for all feeding conditions can lead to under- and over-estimations of MP and amino acid supply, resulting in variation from expected production.

Table 2. Endogenous contributions used to predict endogenous AA requirements and supply in v7 of the CNCPS.

Endogenous component	Secretion (g N/kg DMI)
Saliva	0.9
Rumen sloughed cells	4.3
Omasum/abomasum sloughed cells	0.3
Omasum/abomasum secretions	0.2
Pancreatic secretions	0.4
Bile	0.1
Small intestine sloughed cells[1]	0.7
Small intestine secretions[1]	0.7
Large intestine sloughed cells	0.3

[1] Includes secretions past the pancreatic and bile duct and prior to the terminal ileum

Several approaches have been developed to predict the intestinal digestibility of protein in feeds and are a departure from the detergent system of feed chemical composition (Calsamiglia and Stern, 1995; Ross et al., 2013). An in vitro assay was developed by Ross et al., (2013) that predicts intestinal N indigestibility in cattle using a multi-step approach. To evaluate the capability of the assay, a study was conducted by formulating two different diets in high producing cattle using two different blood meals with different predicted intestinal protein indigestibility to test the accuracy and precision of both the assay and our ability to apply those values in the CNCPS for diet formulation.

Ninety-six multiparous cows (726 ± 14.2 kg BW; 147 ± 64 DIM) and thirty-two primiparous cows (607 kg ± 29.5 kg BW; 97 ± 20 DIM) were distributed by DIM and BW into 8 pens of 16 cows (12 multiparous and 4 primiparous). Pens were stratified into four levels of milk production, and each stratum randomly allocated to treatments. Diets were formulated using CNCPS, (Van Amburgh et al., 2015) using the chemical composition of the ingredients used in the experimental diets (Table 3). The lactation trial consisted of a two week adaptation period, one week covariate period and 9 week experimental period at Cornell University Ruminant Center (Harford, NY). All cows were fed the LOW uN diet during adaptation and covariate periods. Cows were housed in pens in a four row barn design with one bed and more than one headlock per cow and free access to water. All cows received rBST (Posilac, Elanco Animal Health, Indianapolis, IN) on a 14 day schedule throughout the length of the trial.

Overall DMI and N intake for the treatments were similar and milk yield was significantly different for cattle fed the two treatments (Table 4). Milk yield was 1.6 kg/d lower for cattle fed the HIGH uN diet and energy corrected milk (ECM) was 1.9 kg/d lower

on the same diet. Further, cattle fed the HIGH uN diet had significantly lower MUN levels that cattle fed the LOW uN diet (Table 4). From this information, it is apparent that the cattle fed the different blood meals had significantly different MP supply, consistent with the predicted values from the uN assay. The predicted difference described earlier (38.5 g N) is equal to approximately 240 g MP, about the amount required to produce up to 5 kg of milk under the conditions of this study assuming all MP was partitioned to milk production.

Table 3. The ingredient content and chemical composition of two diets containing blood meals with Low and High indigestible intestinal N digestibility.

	Treatment	
Ingredient, % DM	LOW uN	HIGH uN
Alfalfa haylage	11.5	11.5
BMR corn silage	49.3	49.3
Bakery byproduct	1.8	1.8
Blood meal High	3.7	---
Blood meal Low	---	4.0
Canola meal	3.0	3.0
Corn grain	16.1	16.1
Energy Booster 100	1.8	1.8
Molasses	1.8	1.8
Rumen protected met.	0.1	0.1
Sodium bicarbonate	0.6	0.5
Soybean hulls	4.6	4.5
Urea	0.2	0.2
Wheat midds	4.6	4.5
Min/vit mix	1.0	1.0
Chemical composition		
DM, % as fed	50.00	50.50
CP, % DM	15.20	15.20
NDF, % DM	31.90	32.30
ADF, % DM	21.30	20.50
Ether extract, % DM	4.30	3.90
Starch, % DM	30.40	31.20
Sugar, % DM	3.60	3.30
Ca, % DM	0.65	0.60
P, % DM	0.43	0.43
ME[1], Mcal/kg DM	1.80	1.70
Lys:Met[1], % MP	3.21	3.19

LOW: low uN diet, HIGH: high uN diet. [1]CNCPS predicted

However, the observed difference on an ECM basis was 1.9 kg, thus the difference between the absolute levels measured in the assay and the observed ECM yield are either due to differences in digestibility within the cow, the amount of the blood meal

arriving at the small intestine, the amount of nutrients partitioned to body reserves, or a combination of all of those factors. Although the change in BW and BCS were not significant, the changes are still biologically relevant given the partitioning of nutrients to reserves and away from milk.

Table 4. Effect of N availability on intake, milk production, milk composition and body weight gain of dairy cows fed diets with low and high unavailable N

	Treatment			
Item[1]	LOW uN	HIGH uN	SEM	P-value
DMI, kg	27.40	27.10	0.61	0.75
N Intake, kg DM	671.10	664.40	14.80	0.77
Milk production				
Milk, kg	42.00	40.40	0.31	<0.01
ECM, kg	41.90	40.00	0.32	<0.01
Fat, kg	1.51	1.42	0.02	<0.01
Protein, kg	1.26	1.23	0.01	0.03
Milk composition				
Fat, %	3.60	3.50	0.03	<0.03
Protein, %	3.03	3.06	0.02	0.20
Lactose, %	4.90	4.86	0.02	0.18
MUN, mg/dl	9.40	8.00	0.18	<0.01
SCC (log1000/ml)	3.90	4.00	0.05	0.13
BW and BCS				
BW initial, kg	684.10	692.10	10.10	0.58
BW change, kg	34.70	29.70	2.25	0.12
BCS change, (1-5)	0.20	0.16	0.03	0.29
Efficiency				
Feed efficiency[2]	1.56	1.50	0.03	0.34
Milk N efficiency[3]	30.00	29.70	0.70	0.76

[1] DMI: dry matter intake, ECM: energy corrected milk yield (Tyrrell and Reid, 1965)
[2] calculated as kg milk / kg DMI
[3] calculated as milk N/N intake*100

To evaluate the outcome of the study, CNCPS v6.5 with the updated feed library rates and pool sizes was used to evaluate the predictions. The chemical composition of the feeds used in the study was inputted into the model. To evaluate the assay within the structure of the model and against the study data, the blood meal values for the uN and ADIN were the only values changed. For the two blood meals, the uN values were inputted in place of the ADIN value, and intestinal digestibility left at zero. Further, the intestinal digestibility of the NDIN value were set to 100% although after being analyzed

for aNDFom, the blood meals do not contain any ND residue, so that pool is zero. With this approach, all of the protein in blood meals is in the A2, B1 and C fractions.

The current intestinal digestibility of the NDIN fraction for all feeds is 80% and it appears that the assay of Ross et al. (2013) captures that portion of the indigestible protein, therefore by difference; the remaining fractions should be set at 100% digestibility. Thus, with continued testing and implementation of the uN assay for all feeds, the NDIN fraction ID will be set to 100% because it appears that in NDF containing feeds, the uN assay spans both the ADIN and NDIN fractions.

For the cattle inputs, the expected BW change based on the target growth approach was used and the BCS change was also inputted over the period of the study (9 wks), which accounted for the distribution of nutrients to other productive uses and not just milk output. With all of the inputs accounted for, the prediction of ME and MP allowable milk with the uN assay information is in Table 5.

In the CNCPS evaluation in Table 5, it is apparent that the feed chemistry described through by the detergent system is not appropriate to allow the model to predict the most limiting nutrient in this comparison using blood meal as the treatment. When the uN data are used to describe the chemistry of the blood meals, the model provides an acceptable and realistic prediction of the most limiting nutrient. It is also important to recognize that an accurate and complete description of the animal characteristics was crucial to make this evaluation and in the absence of that information, the model would predict over 4 kg of MP allowable milk difference. The sensitivity of the model predictions to complete and accurate animal characterization cannot be overstated and helps explain why literature data to evaluate the model rarely allows for robust predictions of most limiting nutrients due the lack of complete information.

Table 5. The actual and energy corrected milk and the metabolizable energy (ME) and protein (MP) allowable milk for both treatments predicted by the CNCPS using the assay data of Ross et al., (2013) to estimate intestinal digestibility of blood meal, or using the original fractionation approach using acid detergent insoluble nitrogen as the unavailable fraction

	Treatment	
Item	LOW uN	HIGH uN
Actual milk, kg	42.0	40.4
Energy corrected milk, kg	41.9	40.0
Using uN assay inputs		
ME allowable milk, kg	45.0	46.0
MP allowable milk, kg	42.6	39.3
Using NDIN and ADIN		
MP allowable milk, kg	44.9	44.6

In summary, the uN assay appears to provide protein indigestibility predictions that are consistent with cattle responses and serves as a platform for modifying the approach to predict protein digestibility within the CNCPS and will improve the model's ability to

identify the most limiting nutrient. The data also demonstrate that we are ready to move beyond the detergent system of fractionation for protein, to a system that fractionates proteins based on solubility and indigestibility.

Accurate and complete amino acid measure and values

Most systems use a factorial approach to calculate AA supply, so accurate profiles of AA in undegraded feed, bacteria, protozoa, and endogenous portions of post-rumen protein flows are important. The AA content of protein has historically been determined by single time point hydrolysis, as this represents a compromise between maximal release of AA from the matrix while minimizing the loss of acid labile AA (Rutherfurd, 2009). Determination at multiple time points followed by least-squares non-linear regression provides more accurate estimates of the true amino acid profile (Darragh and Moughan, 2005).

Microbial samples obtained from the omasum were used to determine the AA content after multiple time point hydrolysis. The AA content of all samples was determined by HPLC following hydrolysis at 110°C in a block heater (Gehrke et al., 1985) for 2, 4, 6, 12, 18, 21, 24, 30, 48, 72, 120 and 168 h. All AA except Trp were determined using 6N HCl hydrolysis, with Met and Cys undergoing an additional pre-oxidation step. Tryptophan was determined using florescence detection after hydrolysis in barium hydroxide at the same time points as the acid hydrolysis. The entire time course was performed twice for each sample, and the reported values are the mean of the two determinations.

The comparison of the multiple time point vs. single time point indicates that the AA profile is affected by the rate at which AA are hydrolyzed in the assay (Table 6). This means that when using a single time point hydrolysis at 21 or 24 h, the acid stable and slower releasing AA will be underestimated, while the faster releasing and acid labile AA might be overestimated. In a quantitative sense, this might not account for much of the rumen-undegraded portion individual feed ingredient AA. However, when assigning a profile of AA to the microbial flows, error in the analysis will have a large effect on predicted AA flows when using the factorial approach, as the microbial portion is usually responsible for 40-60 % of the total AA supply.

Similar measurements have been made for feeds and similar differences have been observed, especially in the branched chain amino acids (Table 7). This data suggests that the 21 hr hydrolysis of AA (Gehrke et al., 1985) does not completely recover all of the AA. Further, a multiple time point process should be evaluated to determine if the release of more slowly hydrolyzed AA are uniformly recovered after the 168 hr hydrolysis or if more feeds need to analyzed to determine the true AA content.

Recently, Lapierre et al. (2019) published correction factors for the AA content of rumen microbes and feeds to adjust for specific AA that would allow data from 24 hr hydrolysis to be adjusted to a "true" AA content based on long-term hydrolysis data. This approach allows us to make the appropriate adjustments to the AA content of the supply side of the

predictions, but we need additional data to evaluate milk and tissue to ensure the requirement side does not behave in a similar manner.

Table 6. Comparison of measured AA composition after single hydrolysis time point vs. estimated AA composition determined using least-squares non-linear regression after multiple hydrolysis times for omasal bacteria and protozoa isolates from trial B.

	Bacteria			Protozoa		
Item	24 h[1]	Mult[2]	% Δ	24 h[1]	Mult[2]	% Δ
Essential AA, % of AA						
ARG	4.96	4.88	1.6	5.37	5.41	-0.7
HIS	2.24	2.17	3.0	2.50	2.59	-3.6
ILE	4.25	4.77	-12.4	4.03	4.51	-12.0
LEU	5.48	5.47	0.3	6.83	6.43	5.8
LYS	7.52	7.40	1.6	8.90	8.79	1.2
MET	4.71	4.81	-2.0	3.44	3.87	-12.6
PHE	6.15	5.94	3.4	6.79	6.76	0.4
TRP	5.51	5.93	-7.7	4.26	5.49	-29.1
THR	5.67	5.70	-0.5	4.84	5.09	-5.1
VAL	6.58	7.14	-8.4	4.67	4.88	-4.6
Total EAA	53.07	51.73	2.5	51.61	51.01	1.2
Non-essential AA, % of AA						
ALA	6.68	7.15	-7.0	5.36	5.17	3.6
ASP	10.46	11.13	-6.3	9.65	10.42	-7.9
CYS	1.43	1.45	-1.4	2.37	2.22	6.5
GLU	11.25	11.39	-1.3	12.94	13.40	-3.5
GLY	5.01	4.98	0.6	4.67	4.53	2.9
PRO	2.00	1.97	1.2	2.99	2.97	0.7
SER	4.48	5.03	-12.2	5.14	5.43	-5.8
TYR	5.61	5.82	-3.6	5.27	4.83	8.3
Total NEAA	46.93	48.90	-4.2	48.39	49.22	-1.7
Total AA, % of DM	346.60	339.00	2.2	295.00	290.70	1.4

[1]AA composition after 24 h hydrolysis time

[2]AA composition determined from least-squares non-linear regression from multiple hydrolysis times.

Concept of first limiting nutrient and integration of ME and AA

Using the information presented so far, the AA supply can be more accurately described which then allows for calculations of requirements on a more refined basis. Requirements for each individual EAA in the CNCPS v7 are predicted for processes that are quantified by the model (maintenance, lactation, pregnancy, growth) and subsequently divided by the efficiency of transfer to that process to give the total AA requirement (O'Connor et al., 1993; Fox et al., 2004). The efficiency of transfer could also be thought of as the additional requirement for each AA relative to the requirements

Table 7. The amino acid composition (mg/g DM) of three feeds analyzed at 24 and 168 hr of hydrolysis and content calculated by logistic regression of the content of the residues.

	Canola 1			Soybean meal 1			Blood meal		
AA[1]	24h	168h	SEM[2]	24h	168h	SEM[2]	24h	168h	SEM[2]
Arg	22.17	20.84	0.67	30.99	34.74	1.88	36.26	39.20	1.47
His	12.50	11.04	0.73	12.40	13.95	0.78	50.40[a]	61.58[b]	5.59
Ile	12.40[a]	14.27[b]	0.93	18.99	21.07	1.04	6.06	6.19	0.06
Leu	22.48	25.53	1.53	30.26	39.19	4.47	96.90[a]	119.97[b]	11.54
Lys	17.76[a]	19.54[b]	0.89	25.39	31.70	3.16	71.77[a]	87.10[b]	7.67
Phe	18.51	17.81	0.35	25.88	29.09	1.61	67.93	71.90	1.99
Thr	15.08[a]	12.31[b]	1.39	17.30	14.92	1.19	37.75	37.64	0.05
Val	17.22	18.40	0.59	20.65	21.91	0.63	59.52[a]	83.09[b]	11.79
Met	10.81	11.55	0.37	9.10[a]	9.78[b]	0.34	17.56	17.50	0.03
Trp	8.82	9.42	0.30	10.57	10.48	0.05	25.14[a]	56.32[b]	15.59

[a, b] Different superscripts for a given feed at 24h vs 168h signifies $p < 0.05$

quantified by the model. Such processes include oxidation across the gut or in other tissues, anaplerotic requirements, synthesis of non-essential AA, gluconeogenesis etc. (Lapierre et al., 2005; Lapierre et al., 2006; Lemosquet et al., 2010; Lobley, 2007). The apparent efficiency of AA use for any given diet can be calculated by dividing model predicted amino acid requirement (AAR) by amino acid supply (AAS), which can be variable, and typically decreases as AAS increases relative to either AAR or metabolizable energy (Hanigan et al., 1998). This decrease in apparent efficiency of AA use represents AA being increasingly used for purposes other than those quantified or described by the model. If the utilization of each AA for every process in metabolism could be adequately quantified, the term 'efficiency of use' would become obsolete as it would be 100% (there would be no additional requirement above model predictions). The ability of cows to direct AA to other uses demonstrates the interactions among different nutrients and is an example of the metabolic flexibility that allows productivity to be maintained across a wide range of nutrient inputs and supply (Lobley, 2007). The pertinent question for ration balancing is: what level of additional AA supply is required above the predicted requirements for milk protein synthesis and body protein requirements to maximize productivity and minimize AA wastage? The answer to this question is going to differ among models as supply and requirements are calculated in different ways.

The optimum supply of EAA in v7 was estimated similarly to Doepel et al. (2004) using a dataset of studies that infused AA into the abomasum, duodenum, or intravenously and

fitted a logistic curve (Higgs, 2014). The optimum supply of each EAA was defined as the point in which a logistic curve was approaching plateau most rapidly (Lysine example; Figure 2). This point is similar to the break-point in the segmented linear model used in the NRC (2001) but is further integrated with the ME supply to describe the relationship based on the energy driven demand for AA and not just as a percent of protein.

The optimum ratio of model predicted AAR to AAS (efficiency of use) for each AA in v7 are in Table 8. The impact of energy supply on the utilization of AA was also investigated by regressing the ratio of AAR and AAS against AA supply relative to total ME (Lysine example; Figure 3). Interestingly, the optimum supply of Met and Lys estimated using this approach was 15.1% and 5.7% of EAA, respectively, which is similar to results found in other studies that used different approaches (Rulquin et al., 1993; Schwab, 1996; Schwab et al., 1992). However, under these circumstances, no relationship was observed between the 'efficiency' of AA use when AA supply was expressed relative to MP supply but a strong relationship was observed when AA were expressed relative to ME supply which is in agreement the findings of Van Straalen et al. (1994). These data suggest when balancing rations it might be more appropriate to consider AA supply relative to ME which is the approach used in swine (NRC, 2012). Establishing requirements for monogastrics is less complicated than in ruminants as the true AA supply is more easily determined (Lapierre et al., 2006). That being said, with the available AA infusion study data and the updated techniques described previously in this paper, AA requirements in the ruminant animal are becoming both more accurate and precise. To extend the comparison of non-ruminant to ruminant, the predicted Lys requirement for a lactating sow in the NRC (2012) model is 2.72 g Lys/Mcal ME which is similar to the 3.03 g Lys/Mcal ME calculated in this study for dairy cows. Likewise, the recommended ratios for each EAA and Lys are similar in the dairy cow and sow with the exception of Met and His (Table 8). These data suggest, as improvements are made to the predictions of true AA supply in dairy cows, consideration of the approach used to balance AA in other species where AA supply is more easily determined could provide opportunities to improve productivity and the efficiency of nutrient use.

Summary

To better describe AA supply and requirements and develop approaches to formulate closer to meet the requirements, several steps have been taken to improve the predictions. These approaches provide solutions to offset bias in calculations, improve chemistry to provide information about improved recoveries and digestibilities and provide new insights into how to evaluate AA requirements on an energy allowable basis consistent with monogastric species. It is anticipated that actualizing all of these approaches will allow for lower N feeding and more efficient diets that result in lower cost and less environmental impact of dairy cattle.

Table 8. Efficiency of use and optimum supply of each EAA relative to total EAA, ME and Lys.

AA	Efficiency of use	% EAA	g AA/ Mcal ME	Lys:AA Dairy[1]	Lys:AA Swine[2]
Arg	0.55	10.2%	2.04	1.49	1.85
His	0.70	4.5%	0.91	3.33	2.50
Ile	0.61	10.8%	2.16	1.40	1.78
Leu	0.67	17.1%	3.42	0.89	0.89
Lys	0.62	15.1%	3.03	1.00	1.00
Met	0.53	5.7%	1.14	2.66	3.71
Phe	0.53	10.7%	2.15	1.40	1.82
Thr	0.53	10.7%	2.14	1.41	1.49
Trp	0.58	2.9%	0.59	5.16	5.33
Val	0.62	12.4%	2.48	1.22	1.15

[1] Optimum Lys:EAA ratio for the data set used
[2] Optimum Lys:EAA ratio for a lactating sow (NRC, 2012)

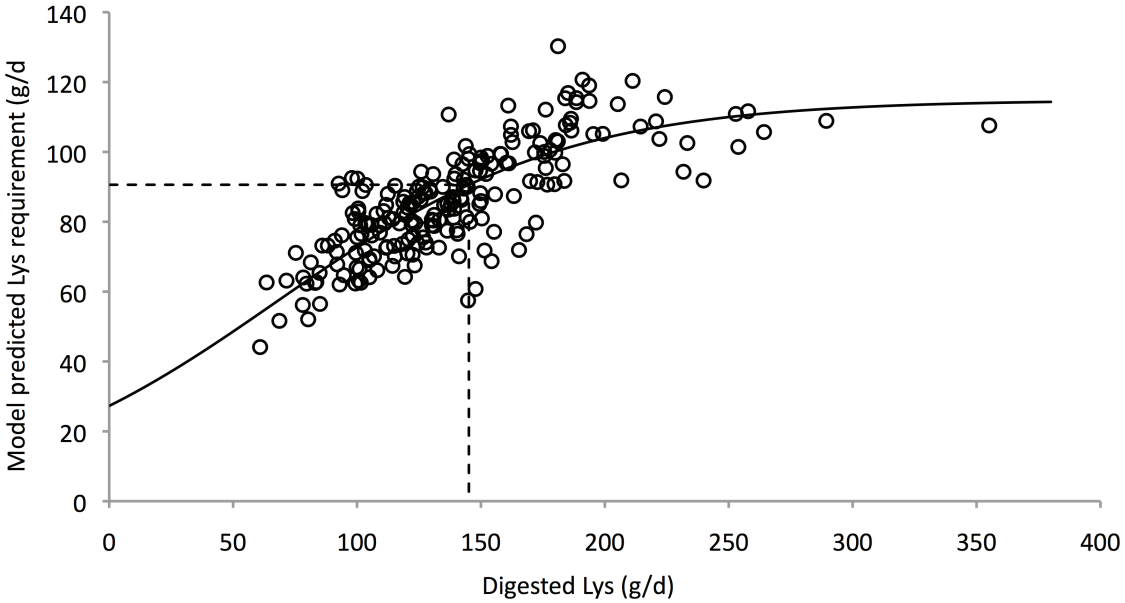

Figure 2. Logistic fit of model predicted Lys requirement and Lys supply. The dashed line represents the optimum ratio of Lys requirement and Lys supply

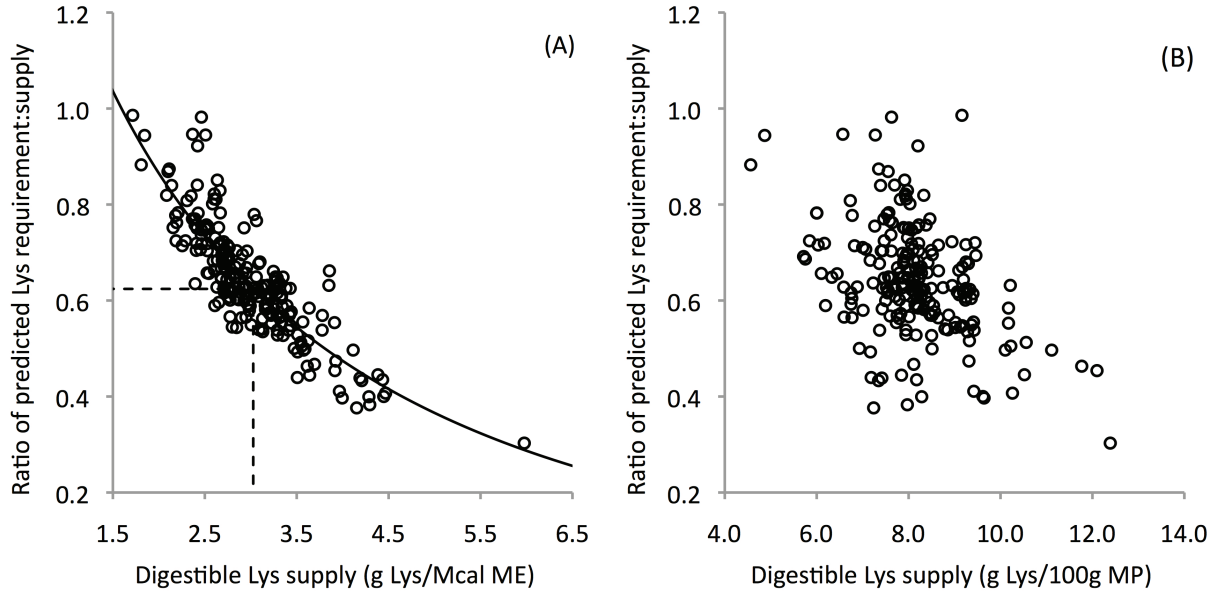

Figure 3. Relationship between model predicted Lys requirement:supply and Lys supply relative to ME (A) or MP (B). The dashed line in (A) represents the Lys supply at the optimum ratio of model predicted Lys requirement and supply. No significant relationship was determined in (B).

A version of this paper was published in the Proceedings of the Canadian Nutrition Conference, 2018.

References

Broderick, G. A., P. Huhtanen, S. Ahvenjärvi, S. M. Reynal, and K. J. Shingfield. 2010. Quantifying ruminal nitrogen metabolism using the omasal sampling technique in cattle - A meta-analysis. J. Dairy Sci. 93: 3216-3230.

Calsamiglia, S. and Stern, M. D. 1995. A three-step *in vitro* procedure for estimating intestinal digestion of protein in ruminants. J Anim. Sci. 73:1459-1465.

Darragh, A. J., and P. J. Moughan. 2005. The effect of hydrolysis time on amino acid analysis. J. AOAC Int. 88:888–893.

Doepel, L. et al. 2004. Milk protein synthesis as a function of amino acid supply. J. Dairy Sci. 87: 1279-1297.

Egan, A. R., K. Boda, and J. Varady. 1984. Regulation of nitrogen metabolism and recycling. In: International Symposium on Ruminant Physiology, Banff, Canada. p 386-402.

Fox, D. G. et al. 2004. The Cornell Net Carbohydrate and Protein System model for evaluating herd nutrition and nutrient excretion. Anim. Feed Sci. Technol. 112: 29-78.

Gehrke, C.W., L. Wall Sr, J. Absheer, F. Kaiser, and R. Zumwalt. 1985. Sample preparation forchromatography of amino acids: acid hydrolysis of proteins. Journal of AOAC International. 68:811-821.

Hanigan, M., J. Cant, D. Weakley, and J. Beckett. 1998. An evaluation of postabsorptive protein and amino acid metabolism in the lactating dairy cow. J. Dairy Sci. 81: 3385-3401.

Higgs, R. J. 2014. Development of a dynamic rumen and gastro-intestinal model in the Cornell Net Carbohydrate and Protein System to predict the nutrient supply and requirements of dairy cattle. PhD Dissertation, Cornell Univeristy, Ithaca, NY.

Higgs, R. J., L. E. Chase, D. A. Ross, and M. E. Van Amburgh. 2015. Updating the Cornell Net Carbohydrate and Protein System feed library and analyzing model sensitivity to feed inputs. J. Dairy Sci. 98: 6340-6360.

Hristov, A. N., and J.-P. Jouany. 2005. Nitrogen requirements of cattle. In: A. Pfeffer and A. N. Hristov (eds.) Factors Affecting the Efficiency of Nitrogen Utilization in the Rumen. p 117-166. CABI, Wallingford, UK.

Huhtanen, P., and A. N. Hristov. 2009. A meta-analysis of the effects of dietary protein concentration and degradability on milk protein yield and milk N efficiency in dairycows. J. Dairy Sci. 92:3222-3232.

Isaacson, H. R., F. C. Hinds, M. P. Bryant, and F. N. Owens. 1975. Efficiency of Energy Utilization by Mixed Rumen Bacteria in Continuous Culture. J. Dairy Sci. 58: 1645-1659.

Ipharraguerre I.R., J.H. Clark. 2005. Impacts of the source and amount of crude protein on the intestinal supply of nitrogen fractions and performance of dairy cows. J. Dairy Sci. 2005 88:E22-37 (Suppl 1).

Jones, D. B. 1941. Factors for converting percentages of nitrogen in foods and feeds into percentages of proteins. USDA Circular No. 183. Washington, D.C.

Lapierre, H. et al. 2005. The route of absorbed nitrogen into milk protein. Animal Science 80: 10-22.

Lapierre, H., and G. E. Lobley. 2001. Nitrogen recycling in the ruminant: A review. J. Dairy Sci. 84: 233-236.

Lapierre, H., Ouellet, D.R., Berthiaume, R., Girard, C., Dubreuil, P., Babkine, M., Lobley, G.E. 2004. Effect of urea supplementation on urea kinetics and splanchnic flux of amino acids in dairy cows. J. Anim. Feed Sci.13:319–322.

Lapierre, H., G. E. Lobley, D. R. Quellet, L. Doepel, and D. Pacheco. 2007. Amino acid requirements for lactating dairy cows: Reconciling predictive models and biology. In: Cornell Nutrition Conference, Syracuse, NY. p 39-59.

Lapierre, H. et al. 2006. What is the True Supply of Amino Acids for a Dairy Cow? J. Dairy Sci. 89: E1-14.

Lapierre, H., S. Binggeli, M. Sok, D. Pellerin, and D. R. Ouellet. 2019. Estimation of correction factors to determine the true amino acid contentration of protein after a 24-hour hydrolysis. J. Dairy Sci. 102:1205-1212.

Lobley, G. E. 2007. Protein-energy interactions: horizontal aspects. In: Energy and protein metabolism and nutrition, Vichy, France. p 445-462.

Marini, J. C., D. G. Fox, and M. R. Murphy. 2008. Nitrogen transactions along the gastrointestinal tract of cattle: A meta-analytical approach. J. Anim. Sci. 86: 660-679.

Marini, J. C., and M. E. Van Amburgh. 2003. Nitrogen metabolism and recycling in Holstein heifers. J. Anim. Sci. 81: 545-552.

NRC. 2001. Nutrient requirements of dairy cattle. 7th revised ed. National Academy Press, Washington, DC.

NRC. 2012. Nutrient requirements of swine. National Academy Press, Washington, DC.

O'Connor, J. D., C. J. Sniffen, D. G. Fox, and W. Chalupa. 1993. A net carbohydrate and protein system for evaluating cattle diets: IV. Predicting amino acid adequacy. J. Anim. Sci. 71: 1298-1311.

Ouellet, D. R. et al. 2010. Effect of method of conservation of timothy on endogenous nitrogen flows in lactating dairy cows. J. Dairy Sci. 93: 4252-4261.

Ouellet, D. R., R. Berthiaume, G. E. Lobley, R. Martineau, and H. Lapierre. 2004. Effects of sun-curing, formic acid-treatment or microbial inoculation of timothy on urea metabolism in lactating dairy cows. Journal of Animal and Feed Sciences 13: 323-326.

Ouellet, D. R. et al. 2002. Effect of dietary fiber on endogenous nitrogen flows in lactating dairy cows. J. Dairy Sci. 85: 3013-3025.

Recktenwald, E. B., D. A. Ross, S. W. Fessenden, C. J. Wall, and M. E. Van Amburgh. 2014. Urea-N recycling in lactating dairy cows fed diets with 2 different levels of dietary crude protein and starch with or without monensin. J. Dairy Sci. 97: 1611-1622.

Reynolds, C. K., and N. B. Kristensen. 2008. Nitrogen recycling through the gut and th nitrogen economy of ruminants: An asynchronous symbiosis. J. Anim. Sci. 86 (E Suppl.):E293–E305.

Reynolds, C. K. 1992. Metabolism of nitrogenous compounds by ruminant liver. J. Nutr. 122: 850-854.

Ross, D. A. 2013. Methods to analyze feeds for nitrogen fractions and digestibility for ruminants with application for the CNCPS. PhD Dissertation, Cornell University, Ithaca, NY.

Ross, D. A., M. Gutierrez-Botero, and M. E. Van Amburgh. 2013. Development of an in-vitro intestinal digestibility assay for ruminant feeds. Pp. 190-202. Proc. Cornell Nutr. Conf. Dept. of Animal Sci. Syracuse, NY

Rulquin, H., P. Pisulewski, R. Vérité, and J. Guinard. 1993. Milk production and composition as a function of postruminal lysine and methionine supply: a nutrient-response approach. Livestock Production Science 37: 69-90.

Russell, J. B., J. D. O'Connor, D. G. Fox, P. J. Van Soest, and C. J. Sniffen. 1992. A net carbohydrate and protein system for evaluating cattle diets: I. Ruminal fermentation. J. Anim. Sci. 70: 3551-3561.

Rutherfurd, S. M. 2009. Accurate determination of the amino acid content of selected feedstuffs. International Journal of Food Sciences and Nutrition. 60:53-62.

Satter, L., and R. Roffler. 1975. Nitrogen requirement and utilization in dairy cattle. J. Dairy Sci. 58: 1219-1237.

Schwab, C. G. 1996. Rumen-protected amino acids for dairy cattle: Progress towards determining lysine and methionine requirements. Anim. Feed Sci. Technol. 59: 87-101.

Schwab, C. G., C. K. Bozak, N. L. Whitehouse, and V. M. Olson. 1992. Amino acid limitation and flow to the duodenum at four stages of lactation. 2. Extent of lysine limitation. J. Dairy Sci. 75: 3503-3518.

Schwab, C. G., P. Huhtanen, C. W. Hunt, and T. Hvelplund. 2005. Nitrogen requirements of cattle. In: A. Pfeffer and A. N. Hristov (eds.) Nitrogen and Phosphorus Nutrition of Cattle and the Environment. p 13-70. CABI, Wallingford, UK.

Sniffen, C. J., J. D. O'Connor, P. J. Van Soest, D. G. Fox, and J. B. Russell. 1992. A net carbohydrate and protein system for evaluating cattle diets: II. Carbohydrate and protein availability. J. Anim. Sci. 70: 3562-3577.

Tamminga, S., H. Schulze, J. Vanbruchem, and J. Huisman. 1995. Nutritional significance of endogenous N-losses along the gastrointestinal-tract of farm-animals. Archives of Animal Nutrition 48: 9-22.

Tylutki, T. P. et al. 2008. Cornell Net Carbohydrate and Protein System: A model for precision feeding of dairy cattle. Anim. Feed Sci. Technol. 143: 174-202.

Valkeners, D., H. Lapierre, J. C. Marini, and D. Ouellet. 2007. Effect of metabolizable protein supply on nitrogen metabolism and recycling in lactating dairy cows. Pages 417–418 in Proc. 2nd Int. Symp. Energy and Protein, Vichy, France. EAAP publication 124. European Federation of Animal Science (EAAP), Rome, Italy.

Van Amburgh, M. E. et al. 2015. The Cornell Net Carbohydrate and Protein System: Updates to the model and evaluation of version 6.5. J. Dairy Sci. 98: 6361-6380.

Van Amburgh, M. E., E. B. Recktenwald, D. A. Ross, T. R. Overton, and L. E. Chase. 2007. Achieving better nitrogen efficiency in lactating dairy cattle: Updating field usable tools to improve nitrogen efficiency. In: Cornell Nutrition Conference, Syracuse, NY. p 25-38.

Van Soest, P. J. 1994. Nutritional ecology of the ruminant. 2nd ed. Cornell University Press, Ithaca, NY.

Van Soest, P. J. et al. 1982. A net protein system for cattle: The rumen submodel for nitrogen. In: Protein requirements for cattle: Oklahoma Symposium. p 265.

Van Straalen, W. et al. 1994. Validation of protein evaluation systems by means of milk production experiments with dairy cows. Netherlands Journal of Agricultural Science 42: 89-104.

Vensim. 2010. Vensim DSS for Windows Version 5.10e. Ventana Systems, Harvard MA.

Waldo, D. R., L. W. Smith, and E. L. Cox. 1972. Model of cellulose disappearance from the rumen. J. Dairy Sci. 55: 125-129.

Walker, N. D., C. J. Newbold, and R. J. Wallace. 2005. Nitrogen requirements of cattle. In: A. Pfeffer and A. N. Hristov (eds.) Nitrogen Metabolism in the Rumen. p 71-115. CABI, Wallingford, UK.

WINNER OF THE 2019 CANC ANIMAL NUTRITION SCHOLARSHIP

CARLYN PETERSON

Effects of the supplemental feeding of a phytogenic feed additive on enteric greenhouse gas emissions and performance of lactating dairy cows.

C. B. Peterson, E. J. DePeters, E. G. Ross, A.V. Carrazco, S. J. Werth, Y. J. Zhao, Y. Pan, and F. M. Mitloehner - Department of Animal Science, University of California, Davis

With new legislation from the State of California to reduce greenhouse gas emissions, research into mitigation strategies of dairy cattle emissions is of great importance. The objective of the study was to mitigate greenhouse gas (GHG) emissions from enteric fermentation of dairy cattle, while maintaining milk production, by feeding diets supplemented with a phytogenic feed additive (PH). Twenty lactating Holstein cows, 10 primiparous and 10 multiparous, were blocked by parity and days-in-milk and were assigned to one of two dietary treatments, with 10 cows per treatment, organized in a randomized complete-block design. The treatments included a control diet with no PH and a diet supplemented with PH (1g/head/day). Treatment was delivered as a top-dress at a rate of 0.5g/head/feeding mixed with ground corn (49.5g corn grain, total 50.0 grams of top-dress/feeding). The control animals received 50g/head/feeding of corn grain only. The diets were primarily composed of corn silage, along with other commodities commonly fed across California dairies. Data on milk yield were collected twice daily. Milk samples were analyzed for components, including fat and protein. Enteric GHG emissions from each cow were sampled for a 12 h period at 14 d intervals (d 0, 14, 28, 42, and 56 of the study) using a head-chamber system that was connected to a mobile air emissions laboratory. Gas emissions measured included the GHGs methane (CH_4), nitrous oxide (N_2O), and carbon dioxide (CO_2), as well as the criteria pollutant ammonia (NH_3). Data were analyzed using the LSMeans package in R (R Core Team, Vienna, Austria). Gases were analyzed alone as averages, and as a percentage of both dry matter intake (DMI) and energy corrected milk (ECM). Gas averages of PH vs control fed cows differed in the interaction of treatment by day for CH_4, CO_2, and NH_3 ($P \geq 0.05$), but were similar for N_2O. When the gases were analyzed as a percentage of DMI, treatment vs control fed cows were found to be similar for all gases. Analysis on gas production expressed as a percentage of ECM did reflect differences for the interaction of treatment by day for CH_4 ($P = 0.012$) and for CO_2 ($P = 0.002$). Performance data of PH treated vs control cows for ECM and DMI were similar between treatments. Milk fat tended to be higher in PH vs control animals for the main effect of treatment ($P = 0.07$) and milk protein tended to be higher in PH treated vs control animals for the interaction of treatment by day ($P = 0.066$). Supplementation with a phytogenic additive to the diet of lactating cows showed minimal differences for the main effect of treatment, for enteric gases and performance, but showed interactions of treatment by time. Changes in the diet during the first three weeks of the trial may have influenced the results.

Integrating concepts of pre-pubertal mammary development and rates of body growth to describe differences in first lactation milk yield

M. E. Van Amburgh, F. Soberon†, M. J. Meyer and R. A. Molano
Department of Animal Science, Cornell University
Ithaca, NY 14850
†Nutreco USA, 115 Executive Drive, Highland, IL 62249

INTRODUCTION

The growth of dairy replacement animals has several objectives: low cost and a low number of days of non-productive life, adequate body size, appropriate body composition, and capacity for optimum milk yield over their lifetimes associated with a long productive life. The actual growth objectives are a function of several factors such as mature body weight (MBW), the body weight (BW) at calving and calving age that optimizes raising cost and nutrients partitioned to growth during lactation thus allowing for optimum milk yield and a prospective decision concerning the age at first calving (Hoffman, 1996; Fox et al., 1999). The integration of those factors provides the target growth rate necessary to meet the BW goal for first parturition within the stated amount of time and this then determines the nutrient supply required daily to meet the objectives.

Overall, changes in body chemical composition are effected by growth, or an altered level of nutrient intake above maintenance energy requirements, or both, which results in dilution of empty body ash, water, and protein (CP) by fat (Reid, 1972; Garrett, 1987). This is because the chemical composition of the fat-free matter is largely unresponsive to BW or level of nutrient intake (Reid, 1972; Fortin et al., 1980; Garrett, 1987; Waldo et al., 1997). Furthermore, it is the retention rate of body CP relative to body fat, which decreases with increasing BW and increasing level of nutrient intake, which limits CP composition of the empty body. Data suggest that the rate of protein synthesis becomes limiting in cattle beyond a certain energy intake and growth rate and therefore energy not deposited as protein is deposited as fat and this breakpoint appears to be at approximately 1 to 1.1 kg/d gain and is most likely MBW dependent (Byers, 1982).

The current Dairy NRC (2001) recommendations for nutrient requirements of growing Holstein heifers are based largely upon a data set collected from beef breeds (Garrett, 1980). With the exception of data collected at Cornell in the 1950's and 1960's (Fortin et al., 1980), little serial harvest data existed that described the composition of retained tissue in the growing Holstein heifer. Since the NRC's publication, the chemical growth of both pre-weaned and pre-pubertal dairy breed heifers has received considerable research interest in the last 20 years. Further, research has been conducted to evaluate the effects of diet, nutrient intake, or both, on the composition of retained tissue between approximately 100 kg BW (Moallem et al., 2004) or 180 kg BW (Waldo et al., 1997) and puberty in Holstein heifers. The data of Waldo et al. (1997) suggested that the composition of the Holstein heifers in that study contained less body fat than previously described and that this observation might be due to changes in mature size associated

with selection for milk yield.

Further, data published over 40 years suggested that milk yield was reduced as pre-pubertal ADG increased, especially if the associated energy intake allowed for increased adipose deposition in the animal. Swanson (1960) was the first to publish data demonstrating that as energy intake increased, fat deposition increased in the heifer, milk yield was reduced and "fatty infiltration" of the mammary gland was likely the causative factor. This observation lead to much more work investigating mammary development and growth rate with the objective of understanding why high growth rate reduced mammary parenchymal development (Sejrsen et al., 1982; Peticlerc et al., 1984; Mantysaari et al., 1995). The data from Waldo et al. (1997, 1998) and Capuco et al., (1995) were the first to quantitatively integrate body growth, mammary development and milk yield and made observations that were not consistent with the previously held perspective that decreased pre-pubertal mammary development negatively impacted milk yield. However this information was largely ignored.

The goals for raising replacement heifers go beyond achieving a specific weight gain. Given that they are future dairy cows, the final goal of heifer rearing should be to optimize their future milk production potential. Body composition is directly related to growth rate, diet composition and stage of maturity at the time the growth occurred. With this in mind, it is vital to remember the effects of body condition or body composition at calving on milk yield. The effect of greater body condition on performance of dairy cattle was reported as a linear decrease in milk yield (Garnsworthy and Topps, 1982). More contemporary data has refined this observation and associated it with reduced dry matter intake and further, this is the focus of much research into transition cow metabolism, insulin resistance and the interaction between obesity and milk yield (Ingvartsen and Andersen, 2000; Douglas et al. 2006; Allen et al., 2009; Overton, 2011). Thus, when evaluating the data integrating pre-pubertal growth rates, mammary development and milk yield, the composition of growth, and therefore the final body composition of the heifer at calving are essential when comparing studies related to milk production.

This paper will integrate concepts of body growth, composition, and nutrient requirements along with mammary development and first lactation milk yield to provide a systems based approach to describe the effects of incorrect body growth on first lactation milk production that has been associated primarily with mammary development. The purpose of this review is to describe nutrient requirements and body composition and discuss how the stage of maturity and the rate of gain at each stage of physiological development can result in changes in body composition that help explain the milk yield observed in previously published studies. This information can be used to reduce total replacement raising cost and improve lactation performance by promoting growth at each stage of maturity while considering the final body composition of the animals. For a more thorough evaluation of body composition and energy and protein content of tissues over time, please see Van Amburgh et al. (2019).

Mammary development and milk yield

Traditionally, body composition has been overlooked when analyzing the effects of pre-

pubertal growth rates on first lactation performance. However, just as body composition and obesity influence the performance of mature dairy cattle, those factors are also a crucial determinant of first lactation heifer performance. As reported by Swanson (1960), when heifers were fattened and bred to calve at the same age as their non-fattened twins, the fattened heifers produced considerably less milk during their first lactation. Although the goals of that study were to compare fattened vs. non-fattened heifers and their corresponding lactation performance, the data was associated with the concept that something other than body composition was impacting the lactation performance.

Subsequently, the seminal work by Sejrsen et al. (1982; 1983) describing the effect of high energy intake on mammary development and the relationship with circulating growth hormone linked the relationship between pre-pubertal growth, mammary development and future milk yield. The primary outcome of this work was to provide an intuitive mechanism to explain why rapid growth during the pre-pubertal phase resulted in reduced milk production in the first lactation. The observation of reduced mammary development could be repeated in almost every experiment (Pritchard et al., 1972; Petitclerc et al., 1984; Mäntysaari et al., 1995; Capuco et al., 1995; Meyer et al., 2006ab). These repeatable observations lead to the conclusion that high energy intakes reduced mammary development through altered hormone status or signaling processes. However, Meyer et al. (2006ab) were the first to recognize that mammary development was not reduced by high energy intake, and instead was the time to reach puberty and the associated signals to change allometric mammary growth that were altered. The mammary gland, like all other reproductive organs, grows in proportion to the size of the body and not in proportion to nutrient intake during the post-weaning, pre-pubertal phase.

To evaluate whether the time effect associated with the mammary development observed in Meyer et al. (2006ab) was similar to previous studies, the amount of mammary development (measured in milligrams of DNA accumulation per day) was determined. Meyer (2005) hypothesized that if the observation was consistent among studies, mammary development should be predictable based on days on treatment. The daily DNA accumulation from Meyer et al. (2006b) was compared to five other studies with adequate descriptions of the experimental design (Figure 1). In that comparison, a majority (R^2=0.83) of the difference in mammary development could be explained by time on study, suggesting that in all of these studies, energy intake hastened the time to puberty, and earlier puberty and the hormonal changes associated with puberty were responsible for the decreased mammary development.

Tissue harvest was the endpoint in most of these studies of mammary development which precludes evaluation of milk yield. There are a few studies where tissue harvest and pregnancy and milk yield data were collected under similar feeding conditions to be able to measure heifers in a "pair-fed" experimental design. The studies with direct comparisons are those of Capuco et al. (1995), Waldo et al. (1998) and Smith (2002). Other studies with similar but sequential study data are from Radcliff et al. (1997; 2000). In each of these studies, the authors observed significant changes in mammary development, without significant changes in first lactation milk yield.

Capuco et al. (1995) observed a 52% decrease in mammary development at puberty in heifers fed for higher rates of pre-pubertal gain, but in the pair fed animals, there was

no significant difference in milk yield (Waldo et al., 1998). Smith (2002) fed a calcium salt of conjugated linoleic acid (Ca-CLA) and measured differences in body composition and pre-pubertal mammary development and in pair-fed animals, measured milk yield. In this study, mammary development was reduced by approximately 60% in heifers fed Ca-CLA, however there was no significant difference in milk yield of the pair-fed heifers.

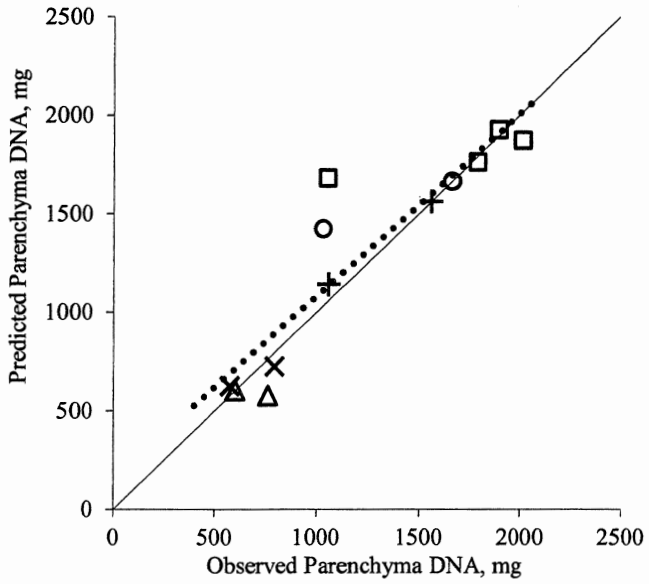

Figure 1. Evaluation of the prediction of "normal" and "diet impaired" pre-pubertal parenchyma development in Holstein heifers. The data points are predicted versus observed. Observed data are from previously published papers [Pritchard et al., 1972, (Δ); Sejrsen et al., 1982, (+); Petitclerc et al., 1984, (X); Capuco et al., 1995, (□); and Mäntysaari et al., (1995), (○)]. Predicted values were generated using the mean daily DNA accretion rate determined in the current study and the average age at harvest as published in the respective papers. Slope of predicted verses observed (dotted line) is 0.95, r^2 = 82% ($P < 0.01$). The solid line represents unity (X = Y). Meyer, 2005.

In the studies by Radcliff et al. (1997; 2000), bST was administered from 125 to 336 kg (276 to 740 lbs) of body weight to enhance pre- pubertal mammary development. In the tissue harvest study, mammary development was enhanced approximately 48% by the use of growth hormone (Radcliff et al. 1997). Milk yield from the heifers treated pre-pubertally with growth hormone did increase by approximately 5.9%, but that was not significant and not highly correlated with the increase in mammary parenchyma development (Radcliff et al. 2000). Thus, mammary development, measured as DNA content of the parenchyma at puberty, varied by about 100% (+48 to -60%) with no significant difference in milk yield. This strongly suggests that mammary development when measured as DNA content at puberty is not a good indicator of future milk yield. This is not to dismiss the concept that mammary development is important, but rather to provide opportunity to consider specific cell types instead of gross measurements using DNA as a proxy for cell number (Sinha and Tucker, 1969; Ballagh et al., 2008).

Body composition and milk yield

One aspect that is harder to quantify is the difference in body composition among heifers at calving in studies investigating the effect of age at first calving on milk yield. Again, for example, Swanson (1960) compared the milk yield of fat versus moderately conditioned heifers and observed that the fatter heifers did not perform as well. Based on data describing the productivity of dairy cattle calving at higher than desired body condition scores, dry matter intake, milk yield and post-partum health are usually at greater risk of being compromised (Grummer et al. 2004; Allen et al 2005; Douglas et al., 2006; Ospina et al. 2010). Thus, body composition at calving as it relates to energy balance is as important for first lactation cattle as multiparous cattle. Further, any difference in body composition of heifers at puberty or pregnancy will most likely be maintained or enhanced since under most conditions the animals remain in positive energy balance from puberty to calving. Thus, experiments evaluating rapid growth prior to puberty are potentially measuring the long-term effect of altered body composition at calving.

There are currently data to make accurate predictions of the maintenance and growth requirements of dairy heifers, as well as to model growth and body composition while taking into consideration stage of maturity of the heifer. In this paper, we used published equations describing energy and protein requirements and body composition to predict body composition at various stages of growth up to calving. Predictions were made using the current Nutrient Requirements of Dairy Cattle 7th edition (NRC, 2001) publication, as well as optimizations of requirements calculated from data generated after the publication of the NRC (Van Amburgh and Drackley, 2005). In addition, equations from Fox, et al. (1999) and the Nutrient Requirements of Beef Cattle (NRC, 1996) were also used to predict nutrient requirements or body composition.

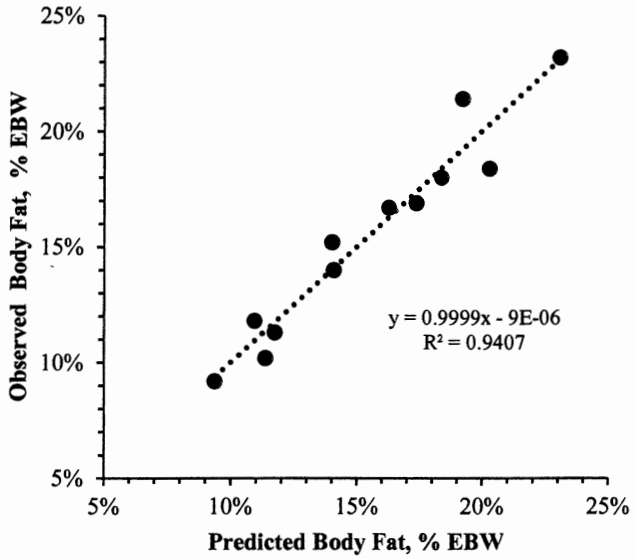

Figure 2. Regression of predicted and observed empty body fat content of dairy calves and heifers at different weights for calves grown at two different rates of gain. Measured body compositions taken from Meyer et al., 2005.

The predictions for body fat percent were evaluated using data from Meyer (2005) and resulted in a R^2 of 0.94 (Figure 2). Protein and lean tissue composition were also considered and the body composition of the protein content was also predicted by the model.

The model was evaluated with data from Gibb et al. (1992), where post-calving body composition was available for cows fed to grow at three different ADG pre-calving. A distinctive characteristic of this study was that the mature body weight of the cattle used could be described as approximately 700 kg since the study utilized cattle with 3 and greater lactations. The reported body fat content of cattle grown at the 3 different pre-calving body growth rates were 18.6%, 19.4% and 21.2%. When accounting for themature weight of the population, the estimates from our model for post-calving body fat content were 18.5%, 19.5% and 20.8% for each of the respective ADG.

To better understand the relationship between rate of gain, composition of gain and age at first calving (AFC), the model was used to develop estimates of the body composition at calving of heifers who were grown at different rates of gain, bred when they had achieved 55% of their mature body weight and had calved at 82% of their mature body weight as per current recommendations (Fox et al. 1999; NRC, 2001). In the base scenario, all animals were assumed to double their birth weight by 60 days and have an ADG of 0.6 kg (1.3 lbs) during pregnancy, excluding the weight of the gravid uterus. Three different pre-pubertal growth rates were used: 0.75 kg/d (1.7 lbs/d), 0.64 kg/d (1.4 lbs/d) and 0.56 kg/d (1.2 lbs/d) that allowed for AFC of 22, 24 and 26 mo., respectively. Given these growth rates, the three groups of animals were estimated to calve at 25% body fat and 15% protein, and would not be expected to have differences in milk production, although the animals that calve at 22 months would be producing milk 4 months sooner than those set to calve at 26 months.

Subsequently, several different scenarios were created based on published data to represent studies and potential on-farm conditions that describe various management approaches to decision making for AFC and BW at or post-calving. When pre-pubertal growth rates were adjusted but heifers were bred by age, the predicted body composition of heifers in each group changed significantly. Using similar assumptions, if calves double their birth weight by 60 d and grow at 0.7 kg/d (1.5 lbs/d) during pregnancy (without the weight of pregnancy), and all heifers are bred at 16 months for an expected AFC of 25 months, but during pre-puberty had ADG of 1 kg (2.2 lbs), 0.8 kg (1.8 lbs) or 0.6 kg (1.3 lbs) they would calve at 30%, 27% or 23% body fat and 14%, 15% and 16% protein, respectively. Data are not available to fully characterize the body composition at calving that provides the most optimum energy balance for first lactation cattle, however the difference in body fat from 23 to 30% would be enough to increase the BCS by at least 1 score, equivalent to 40 kg (88 lb) body fat in a 560 kg (1,250 lb) Holstein heifer. These calculations are consistent with data where heifers were bred to calve at the same age but at different body weights; consequentially, heavier (fatter) heifers produced less milk during first lactation (Swanson, 1978).

To better understand the effects of pre-pubertal ADG on future milk production, we estimated the body composition at calving for heifers from published studies where milk yield was evaluated.

Valentine et al. (1987) reported growth rates from 0.18 kg (0.40 lbs) to 0.94 kg (2.07 lbs), where AFC ranged from 26.9 mo for the slowest treatment to 22.4 mo for the treatment gaining over 0.9 kg/d. After calving, the estimated body fat percent for all groups was 22% and researchers reported no difference in milk production among any of the groups. This data suggests that if little difference exists in body composition at calving, and BW are reasonably similar, dry matter intake, energy balance and milk yield will not be negatively impacted.

Hohenboken et al. (1995) compared three different growth rates from 6 wk of life to 300 kg (661 lb). In this study, all heifers were bred at the same age generating AFC of 29, 26 and 23 mo for heifers raised at ADG of 0.6 kg (1.3 lbs), 0.7 kg (1.6 lbs) or 0.9 kg (1.9 lbs) respectively. These treatments resulted in a predicted body composition of 17% and 25% body fat and 18% and 16% protein for calves raised at 0.6 kg and 0.9 kg respectively. The treatment heifers with 17% predicted body fat produced 500 kg (1,103 lbs) more milk than the group with higher body fat percent. This is consistent with the data describing the potential impact of greater body condition score on dry matter intake, energy balance and milk yield (Garnsworthy and Jones, 1987; Allen et al., 2005; Janovick and Drackley, 2010)

In agreement with these calculations are the results from Hoffman et al. (1996), who reported the effects of different growth rates post-puberty (~45% of mature body weight) and during pregnancy on first lactation milk yield. Heifers on this study were fed to achieve an ADG of 0.97 or 0.79 kg (2.14 or 1.74 lbs) from 10 mo of age until calving. The group fed for higher gain was bred at 10 mo while the control group was bred at 14 mo. At calving, both groups had similar body weights but researchers reported that the group with higher gains had lower wither height and pelvic area. The interpretation of these results suggest that calves with higher ADG during pregnancy had a higher fat composition given the fact that they were smaller framed animals but had similar weights than control. Furthermore, milk production of the calves with higher ADG during pregnancy was 2 kg/d (4.4 lbs) lower than control calves but their milk fat yield was higher during the first 2 months of lactation. These observations are consistent with the lactation performance of over-conditioned cattle.

One of the most crucial and overlooked variables in the effects of growth rate on future performance is mature size. As previously mentioned, the composition of the gain is dependent on the stage of maturity, therefore, when evaluating growth rates pre- puberty, it is important to characterize the growth rates within the stage of physiological maturity. This concept was described for dairy cattle by Fox et al. (1999), where they described the percent of mature BW at pregnancy (55%) and post-calving BW (minimum 82%) necessary to optimize first lactation milk yield. The key factor in this approach is utilizing the mature BW of the herd to adjust for stage of maturity for nutrient requirements instead of using a population value. In all of the studies conducted on heifers prior to the publication of the Dairy NRC (NRC, 2001), no consideration was given to the mature size of the cattle, thus most data were not adjusted for stage of growth and under those conditions, energy intake is almost always greater than required for dairy replacements (Van Amburgh and Meyer, 2005).

Foldager and Sejrsen (1987) concluded that the optimal growth rate of dairy calves

between 90 and 350 kg (200 and 770 lbs) live weight should be 0.6 kg/d (1.3 lbs/d). However, representative animals from that data set are shown in Figure 3. From this picture, over- conditioning of the fastest growing heifers was not included in the analysis and was probably a confounding factor in milk production. To better describe this, the growth data from Foldager and Sejrsen (1987) were used to predict body composition at calving, however, we had to make assumptions about the mature body weight of the animals represented and chose a range of mature weights for comparison. Predicted body composition at calving for cattle with mature body weights from 500 to 700 kg (1,103 to 1,544 lbs) are presented in Table 1. As mature weight increased, body fat decreased at similar calving weights.

Table 1. Calculated body composition at calving of heifers grown at different pre-pubertal rates with different mature body weights.

Mature body weight, kg	ADG from 90 to 350 kg, g/d	Calculated body fat, %	Calculated body protein, %
700	400	18.5%	18.0%
700	600	19.5%	17.7%
700	800	20.8%	17.4%
600	400	20.8%	17.3%
600	600	22.0%	17.0%
600	800	23.6%	16.6%
550	400	22.2%	16.9%
550	600	23.6%	16.5%
550	800	25.3%	16.1%
500	400	23.9%	16.4%
500	600	25.4%	16.0%
500	800	27.2%	15.5%

The cattle represented in the study appear to be small framed cattle with mature body weights between 500 and 550 kg (1,103 to 1,213 lb). If this study had been performed with larger framed cattle, conclusions on the effects of growth rate on milk performance might have been different due to the composition of the gain of the animals. Again, depending on the mature size of the cattle, the differences in fat percent translate into differences in BCS of at least 1 unit and this would have a significant effect on post-partum DMI, and milk yield. Milk production on this study differed by 500 kg (1,102 lb) in the first 250 d of lactation where heifers grown at 0.6 kg/d (1.3 lb/d) produced 5,100 kg (11,245 lb) of milk compared with 4,600 kg (10,143 lb) produced by heifers grown at 0.8 kg/d (1.76 lb/d) during the pre-pubertal period.

The overall goal of heifer rearing is to provide the management and nutrition that allows for optimum milk yield in the first and subsequent lactations. Research has evaluated many aspects of heifer rearing, however, most of the focus has been on pre-pubertal growth rate and its effects on mammary development. Little to no attention has been placed on the effects of such growth rates on body composition or maturity at calving as it relates to energy balance or growth during the lactation. Transition cow research has

unequivocally shown the negative effects of over conditioned cattle at the time of calving on DMI, metabolic problems and milk yield. These findings also apply to first lactation heifers. When accounting for predicted body composition at calving, we are able to explain most of the variation in milk production observed in different studies. Body composition explains both the lack of differences in production observed in some studies (Valentine et al., 1987; Waldo et al., 1998) as well as the differences in milk production observed in others (Swanson, 1978; Foldager and Sejrsen, 1987; Hohenboken et al., 1995). Thus in many studies evaluating mammary development and milk yield, directly or indirectly, the outcome was most likely better predicted by body composition at calving and not mammary development. Moreover, body composition during different growth stages is greatly influenced by mature size. When mature size is not accounted for in diet formulation, energy is often over-fed, resulting in greater fat deposition in growing heifers in subtle but significant outcomes.

Figure 3. Three 18 months old heifers grown at ADG of 400, 600 and 800 g (0.88, 1.32 and 1.76 lb). Live weights were 250, 402 and 540 kg (551, 886, 1,190 lb) respectively (Foldager and Sejrsen, 1987).

An additional scenario commonly seen in practical conditions is reducing AFC without taking into consideration the physiological maturity of heifers (MBW), necessary to adjust growth rates and achieving target weights. This practice might lead to heifers with a significant growth requirement during lactation and growth will always be a priority for nutrient use as the heifer partition energy away from lactation. This is most likely one of the primary factors effecting milk yield in herds were AFC is identified as an important metric with no discussion of body weight, pre- or post-pubertal growth rates or mature size of the herds were considered. Under sized heifers will not only be unable to consume and compete for feed as much as their more mature counterparts, but also more of the consumed nutrients will be partitioned towards growth than for lactation functions (Van Amburgh et al., 1998; Fox et al., 1999). The CNCPS has a growth function based on current BW, MBW and time to achieve the expected BW for the next

lactation established to predict the amount of nutrients required to meet the target growth to achieve MBW. In a scenario for which herd MBW was 750 kg, a standard diet was balanced using the Cornell Net Protein and Carbohydrate System (Van Ambugh et al., 2015) for meeting 100% of ME, MP and Met requirements of a first calf heifer at 82% of MBW (615 kg) and producing 38 kg of ECM/d with an expected ADG in the first lactation of 0.19 kg/d and at this stage of growth, the ME requirement is 2 Mcal/d and 74 g/d of MP. A contemporary heifer that calved at 72% MBW (540 kg) was evaluated similarly and expected ADG to meet the BW target for the following lactation was 0.39 kg/d, more than twice the animal calving at 615 kg and this is an ME requirement of 3.8 Mcal/d and 133 g/d for MP. This difference in ME and MP required for growth would come at the expense of milk yield and is equal to approximately 1 kg of milk/d, not accounting for the DMI difference that would exist between the heifers of at least 1.5 to 2 kg due to the BW difference. Thus, for every 0.1 kg of SBW gain required per day, daily ME and MP requirements were increased by 1 Mcal and 30 g, respectively.

Summary

Data presented in this paper support the current growth benchmarks for heifer rearing (Fox et al., 1999; NRC, 2001) to achieve a body composition by calving that does not compromise post-partum energy balance or milk yield and allows for earlier age at first calving. Heifers should be bred between 55 and 60% of their mature body weight to achieve a post-calving weight of 82 to 85% of the mature body weight of the herd. When these targets are attained, heifers can successfully calve earlier without a negative impact on milk production, with the added benefit of having reduced the length of the non-productive stage.

References

Allen, M. S., B. J. Bradford, and K. J. Harvatine. 2005. The cow as a model to study food intake regulation. Ann. Rev. Nutr. 25:523-547.

Allen, M. S., B. J. Bradford, and M. Oba. 2009. The hepatic oxidation theory of the control of feed intake and its application to ruminants. J. Anim. Sci. 87:3317-3334.

Ballagh K., N. Korn, L. Riggs, S. L. Pratt, F. Dessauge, R. M. Akers, and S. Ellis. 2008. *Hot Topic:* Prepubertal ovariectomy alters the development of myoepithelial cells in the bovine mammary gland. J. Dairy Sci. 91:2992–2995.

Byers, F. M. 1982. Protein growth and turnover in cattle: Systems for measurements and biological limits. In: Protein Requirements for Cattle: Symposium. Oklahoma State University Division of Agriculture Bulletin MP-109: Pp. 141-165.

Capuco A. V., J. J. Smith, D. R. Waldo, and C. E. Rexroad. 1995. Influence of prepubertal dietary regimen on mammary growth of Holstein heifers. J. Dairy Sci. 78:2709-2725

Douglas, G. N., T. R. Overton, H. G. Bateman II, H. M. Dann, and J. K. Drackley. 2006. Prepartal plane of nutrition, regardless of dietary energy source, affects periparturient metabolism and dry matter intake in Holstein cows. J. Dairy Sci. 89:2141-2157.

Foldager J., and Sejrsen K. 1987. Mammary gland development and milk production in dairy cows in relation to feeding and hormone manipulation during rearing. Research in cattle production Danish status and perspectives. 8:102-116.

Fortin, A., S. Simpfendorfer, J. Reid, H. Ayala, R. Anrique, and A. Kertz. 1980. Effect of level of energy intake and influence of breed and sex on the chemical composition of cattle. J. Anim, Sci. 51:604-614.

Fox D.G., Van Amburgh M.E., Tylutki T.P. 1999. Predicting requirements for growth, maturity, and body reserves in dairy cattle. J Dairy Sci. 82:1968-77.

Garnsworthy P. C., G. P. Jones. 1987. The influence of body condition at calving and dietary protein supply on voluntary food intake and performance in dairy cows. Anim. Prod. 44:347- 353.

Garnsworthy P. C., J. H. Topps. 1982. The effect of body condition of dairy cows at calving on their food intake and performance when given complete diets. Anim. Prod. 35:113-119.

Garrett, W. 1980. Energy utilization by growing cattle as determined in 72 comparative slaughter experiments. Energy Metab. Proc. Symp. 26: 3-7.

Garrett, W. 1987. Relationship between energy metabolism and the amounts of protein and fat deposited in growing cattle. Energy Metab. Proc. Symp. 32:528-547.

Gibb M. J., W. E. Ivingst, M. S. Dhanoa, J. D. Sutton. 1992. Changes in body components of autumn-calving Holstein-Friesian cows over the first 29 weeks of lactation. Anim. Prod. 55:339-360.

Grummer, R. R., D. G. Mashek, and A. Hayirli. 2004. Dry matter intake and energy balance in the transition period. Vet. Clin. Food Anim. 20:447-470.

Hoffman P. C., N. M. Brehm, S. G. Price, A. Prill-Adams. 1996. Effect of accelerated postpubertal growth and early calving on lactation performance of primiparous Holstein heifers. J. Dairy Sci. 79:2024-2031.

Hohenboken W. D., J. Foldager, J. Jensen, P. Madsen, B. B. Andersen. 1995. Breed and nutritional effects and interactions on energy intake, production and efficiency of nutrient utilization in young bulls heifers and lactating cows. Acta Agric. Scand. Sect. A Anim. Sci. 45:92.

Ingvartsen, K. L., J. B. Andersen. 2000. Integration of metabolism and intake regulation: A review focusing on periparturient animals. J. Dairy Sci. 83:1573-1597.

Janovick N. A., Drackley J. K. 2010. Prepartum dietary management of energy intake affects postpartum intake and lactation performance by primiparous and multiparous Holstein cows. J Dairy Sci. 93:3086-102

Mäntysaari P., K. L. Ingvartsen, V. Toivonen, K. Sejrsen. 1995. The effect of feeding level and nitrogen source of the diet on mammary development and plasma hormone concentrations of pre-pubertal heifers. Acta. Agric. Scand. Sect. A 45:236-244.

Meyer M. J. 2005. Developmental, nutritional, and hormonal regulation of mammary growth, steroid receptor gene expression and chemical composition of retained tissues in prepubertal bovine. Ph.D. Diss. Cornell University, Ithaca, NY.

Meyer M. J., A. V. Capuco, D. A. Ross, L. M. Lintault, and M. E. Van Amburgh. 2006a. Developmental and nutritional regulation of the prepubertal heifer mammary gland: I. Parenchyma and fat pad mass and composition. J. Dairy Sci. 89:4289-4297

Meyer M. J., A. V. Capuco, D. A. Ross, L. M. Lintault, and M. E. Van Amburgh. 2006b. Developmental and nutritional regulation of the prepubertal bovine mammary gland: II. Epithelial cell proliferation, parenchymal accretion rate, and allometric growth. J. Dairy Sci. 89:4298-4304.

Moallem, U., G. Dahl, E. Duffey, A. Capuco, D. Wood, K. McLeod, R. Baldwin, and R. Erdman. 2004. Bovine somatotropin and rumen-undegradable protein effects in prepubertal dairy heifers: Effects on body composition and organ and tissue weights. J. Dairy Sci. 87:3869-3880.

National Research Council. 1996. Nutrient Requirements of Beef Cattle. Seventh Revised Edition. National Academy Press, Washington, DC.

National Research Council. 2001. Nutrient requirements of Dairy Cattle. Seventh Revised Edition. National Academy Press, Washington, DC.

Ospina, P. A., D.V. Nydam, T. Stokol, and T.R. Overton. 2010. Evaluation of NEFA and β hydroxybutyrate (BHB) in transition dairy cattle in the northeast USA. Critical thresholds

for prediction of clinical diseases. J. Dairy Sci. 93:546-54
Overton T. R. 2011. Managing the dynamics of feed intake and body condition score during the transition period and early lactation. Proc. Cornell Nutr. Conf.
Petitclerc D., Chapin L. T., Tucker H. A. 1984. Carcass composition and mammary development responses to photoperiod and plane of nutrition in Holstein heifers. J Anim Sci. 58:913-9.
Pritchard D. E., Hafs H. D., Tucker H. A., Boyd L. J., Purchas R. W., Huber J. T. 1972. Growth, mammary, reproductive, and pituitary hormone characteristics of Holstein heifers fed extra grain and melengestrol acetate. J Dairy Sci. 55:995-1004.
Radcliff R. P., M. J. VandeHaar, A. L. Skidmore, L. T. Chapin, B. R. Radke, J. W. Lloyd, E. P. Stanisiewski, H. A. Tucker. 1997. Effect of diet and bovine somatotropin on heifer growth and mammary development. J. Dairy Sci. 80:1996-2003.
Radcliff R. P., M. J. Vandehaar, L. T. Chapin, T. E. Pilbeam, D. K. Beede, E. P. Stanisiewski, H. A.Tucker. 2000. Effects of diet and injection of bovine somatotropin on pre-pubertal growth and first-lactation milk yield of Holstein cows. J. Dairy Sci. 83:23-29.
Reid, J. T. 1972. Body composition in animals: Interspecific, sex, and age peculiarities, and the influence of nutrition. Pages 217-237 in Festkrift til. K. Breirem and Maricudals Boktrikkeri A.s Gjovik, eds., Oslow, Norway.
Sejrsen K., Huber J. T., Tucker H. A. 1983. Influence of amount fed on hormone concentrationsand their relationship to mammary growth in heifers. J Dairy Sci. 66:845-55.
Sejrsen K., Huber J. T., Tucker H. A., Akers R. M. 1982. Influence of nutrition of mammary development in pre- and postpubertal heifers. J Dairy Sci. 65:793-800.
Sinha Y. N., Tucker H. A. 1969. Mammary development and pituitary prolactin level of heifers from birth through puberty and during the estrous cycle. J Dairy Sci. 52:507-12.
Smith J. M. 2002. Nutritional modulation of growth and development of Holstein heifers. Ph.D. Diss. Cornell University, Ithaca, NY.
Stelwagen K., D. G. Grieve. 1992. Effect of plane of nutrition between 6 and 16 months of age on body composition, plasma hormone concentrations and first-lactation milk production in Holstein heifers. Can. J. Anim. Sci. 72:337-346.
Swanson E. W. 1960. Effect of rapid growth with fattening of dairy heifers on their lactational ability. J. Dairy Sci. 43:377-387.
Swanson, E. W. 1978. Heifer performance standards: relation of rearing systems to lactation. Chapter 21 in "Large dairy herd management". University Presses of Florida, Gainesville. Editors: C. J. Wilcox, H. H. Von Horn, B. Harris, Jr., H. H. Head, S. P. Marshall, W. W. Thatcher, D. W. Webb, J. M. Wing. Pp.494-511.
Valentine, S. C., R. C. Dobos, P. A. Lewis, B. D. Bartsch, R. B. Wickes. 1987. Effect of live weight gain before or during pregnancy on mammary gland development and subsequent milk production of Australian Holstein-Friesian heifers. Aust. J. Exp. Agric. 27:195-204.
Van Amburgh, M.E., F. Soberon, M.J. Meyer, R.A. Molano. 2019. *Symposium review:* Integration of post-weaning nutrient requirements and supply with composition of growth and mammary development in modern dairy heifers. J. Dairy Sci. 102:3692 3705.
Van Amburgh, M., E. Collao-Saenz, R. Higgs, D. Ross, E. Recktenwald, E. Raffrenato, L. Chase, T. Overton, J. Mills, and A. Foskolos. 2015. The Cornell Net Carbohydrate and Protein System: Updates to the model and evaluation of version 6.5 J. Dairy Sci. 98:6361-6380.
Van Amburgh, M. E., and M. J. Meyer. 2005. Target growth and nutrient requirements of post weaned dairy heifers. Dairy calves and heifers integrating biology and management. NRAES 175:54-65
Van Amburgh, M. E. and J. K. Drackley. 2005 Current perspectives on the energy and protein requirements of the pre-weaned calf. Chap. 5 in "Calf and heifer rearing: Principles of

rearing the modern dairy heifer from calf to calving". Nottingham Univ. Press. P.C. Garnsworthy, ed. Pp.67-82.

Waldo, D., H. Tyrrell, A. Capuco, and C. Rexroad Jr. 1997. Components of growth in Holstein heifers fed either alfalfa or corn silage diets to produce two daily gains. J. Dairy Sci. 80:1674-1684.

Waldo D. R., Capuco A. V., Rexroad C. E. Jr. 1998. Milk production of Holstein heifers fed either alfalfa or corn silage diets at two rates of daily gain. J Dairy Sci. 81:756-64.

Role of the Rumen Microbiome in Feed Digestion and Beef Cattle Health

Tim A. McAllister[1], Rodrigo Passetti[2] and Ludmila Passetti[3]
1Agriculture and Agri-Food Canada, Lethbridge Research Centre, Lethbridge, AB. tim.mcallister@canada.ca
[2]Department of Animal Science, State University of Maringá, Maringá, PR, Brazil. racpassetti@gmail.com
[3]Institute of Agricultural Sciences, Federal University of the Valleys of Jequitinhonha and Mucuri, Unaí *Campus*, Unaí, MG, Brazil. ludmilacoutogomes@gmail.com

I. Fundamental rumen microbiology

1. Rumen ecosystem

Ruminants are herbivores that primarily use forages and other by-products unsuitable for human consumption as sources of energy, protein and other nutrients. Even in intensive productions systems that utilize grains to finish beef cattle, forages account for ≈ 80% of the feed consumed by the entire beef herd. Despite their reliance on forages as their primary feed, ruminants do not produce any of the enzymes required to degrade plant cell walls. Rather, they rely on a symbiotic relationship with microorganisms (bacteria, fungi, methanogens, and protozoa) that degrade plant cell walls to end products that can be used by the host to produce milk and meat.

To accomplish this task, ruminants have a biologically unique structure within their digestive system known as the reticulo-rumen, which is an ideal anaerobic environment for the growth of these fermentative microorganisms. The rumen is essentially a continuous fermenter where substrate in the form of feed is regularly provided and the pH and osmolality are controlled by the inflow of saliva, the absorption of fermentation acids and the passage of microbial protein and digesta to the lower digestive tract However, the rumen is also a dynamic system, and microorganisms have to be able to adapt to changes in the composition, physical structure as well as the quantity and frequency of feed availability. Consequently, changes in the forage to concentrate ratio, feed processing methods and the frequency and quantity of feed delivered to the host can have a significant impact on the microbial population and the health of the host. It is important that cattle producers have an understanding of how feeding practices can alter the rumen environment and its microbial inhabitants.

2. Bacteria

Rumen bacteria are the most abundant and diverse group of microorganisms in the rumen ecosystem. Ruminal contents can contain 10^{10} to 10^{11} bacteria/mL, which are responsible for the majority of feed digestion in the rumen. As a whole they possess a multitude of enzymatic activities including, amylases, cellulases, hemicellulases, proteases and lipases which digest starch, plant cell walls, proteins and lipids, respectively. Most of bacteria (70 - 80%) are attached to feed, as this is a perquisite for the digestion of insoluble feeds in the rumen (**Fig. 1**). Bacteria are also present in the liquid fraction (20-30%) of ruminal contents, where they utilize soluble substrates or are in transition from one feed particle to the next. A small fraction of bacteria (< 1%) are also attached to the rumen epithelium. These bacteria utilize the oxygen that diffuses from the blood and is toxic to most other microbial residents within the rumen. They also hydrolyze the urea that diffuses from the blood into the rumen, producing the ammonia that rumen microorganisms can

combine with carbon skeletons to synthesize amino acids. So although this small group of bacteria does not contribute significantly to feed digestion, they still make a significant contribution to the microbial community (Cheng et al., 1979).

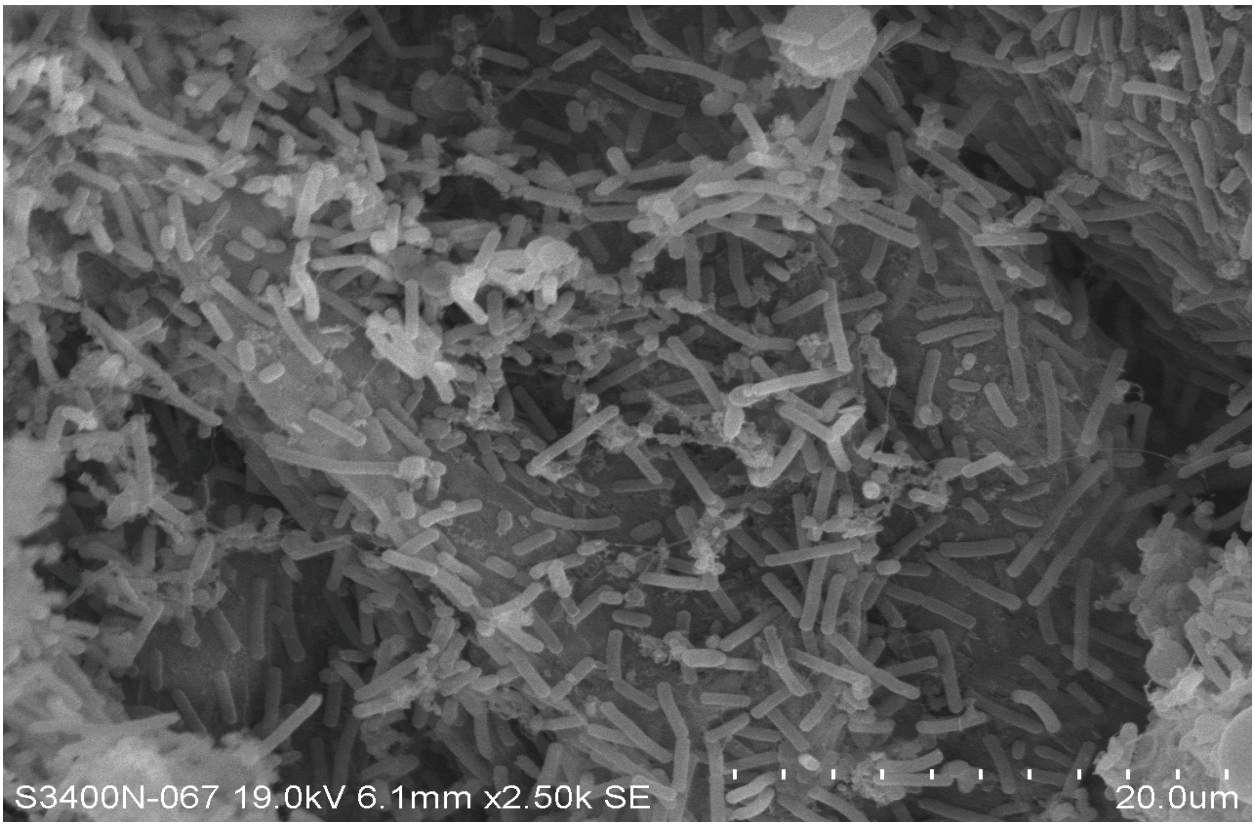

Fig. 1. Adherence of mixed of rumen bacteria to plant material

Diet is the major factor that determines the composition and diversity of the rumen bacterial population.(Fernando et al., 2010; Henderson et al., 2015). When cattle are fed high forage diets, cellulolytic bacteria proliferate (e.g., *Ruminococcus flavefaciens*, *Ruminococcus albus*, *Bacteroides succinogenes* and *Butyrivibrio fibrisolvens*), in the rumen. In contrast, if cattle are fed high grain diets, amylolytic bacteria (e.g., *Ruminobacter amylophilus, Prevotella ruminocola, Streptococus bovis*,) that digest starch and those that utilize the end products of starch digestion (e.g., *Megasphaera elsdeni; Selenomonas ruminantium)* such as lactic acid proliferate. This change in the microbiome leads to a decrease in acetate and increase in propionate concentration in the rumen. The production of more fermentation end products leads to a decline in pH and if it reaches 5.0 or lower, lactate concentration in the rumen also usually increases [(Nagaraja and Titgemeyer, 2007); **Fig. 2**]. At a pH below 5.8, many of the cellulolytic bacteria are inhibited and as a result ruminal fiber digestion also decreases.

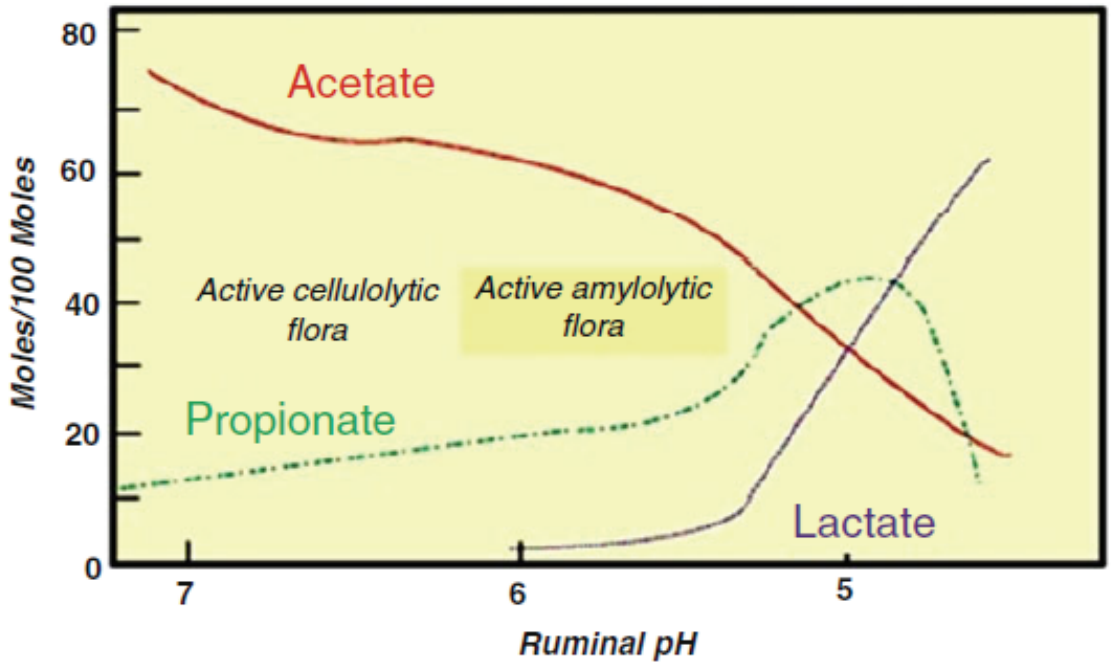

Fig. 2. Relationship of ruminal pH with proportions of acetate, proprionate and lactate. Adapted from (Kaufmann et al., 1980)

3. Protozoa

Protozoa are present in rumen fluid at 10^3 to 10^6 cells per ml of rumen fluid and may account for up to 50% of the total rumen microbial biomass. Protozoa are thought to be responsible for ¼ to ⅓ of the fiber digestion in the rumen. Although protozoa are often associated with rumen fluid, large numbers may also attach to the surface of feed particles (**Fig. 3**) or to the rumen epithelium. Protozoa are predators of rumen bacteria, thus the number of protozoa in the rumen fluctuates inversely with the number of bacteria. As a result of bacterial predation, protozoa are also responsible for the turnover of a large portion of the microbial protein within the rumen. They also contribute to the degradation of feed protein and are associated with higher ruminal concentrations of ammonia. Ruminants can survive without any protozoa in the rumen, and as protozoa and methanogens have a close symbiotic relationship, removal of protozoa often results in a transient decline in methane emissions.

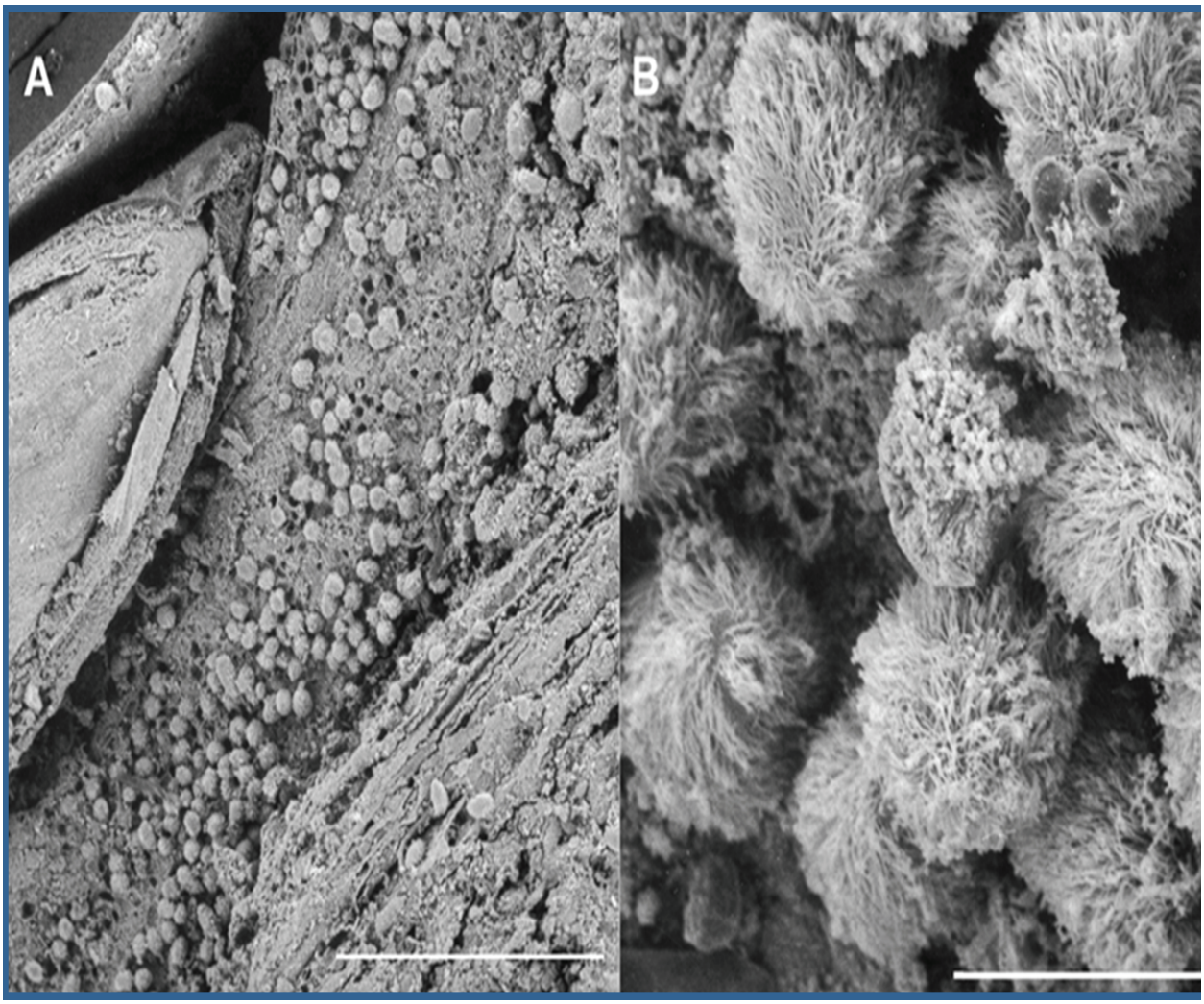

Fig. 3. Protozoa colonizing barley grain after 2h of ruminal incubation

Ruminants that lack protozoa are said to be defaunated, (Newbold et al., 2015) and although a reduction in methane emissions and ruminal protein breakdown could be of benefit, it is often at the expense of a decrease in organic matter and fiber digestibility (Newbold et al., 2015). The diversity of protozoa in the rumen declines with increasing concentrate in the diet, but those protozoa that remain are capable of engulfing starch granules, modulating starch digestion and reducing the risk of ruminal acidosis **(Fig. 4)**. Thus, the benefits of eliminating rumen protozoa is a matter of debate and as protozoa are readily transmitted among individuals within the herd, it is very difficult to ensure that ruminants remain defaunated under practical production conditions.

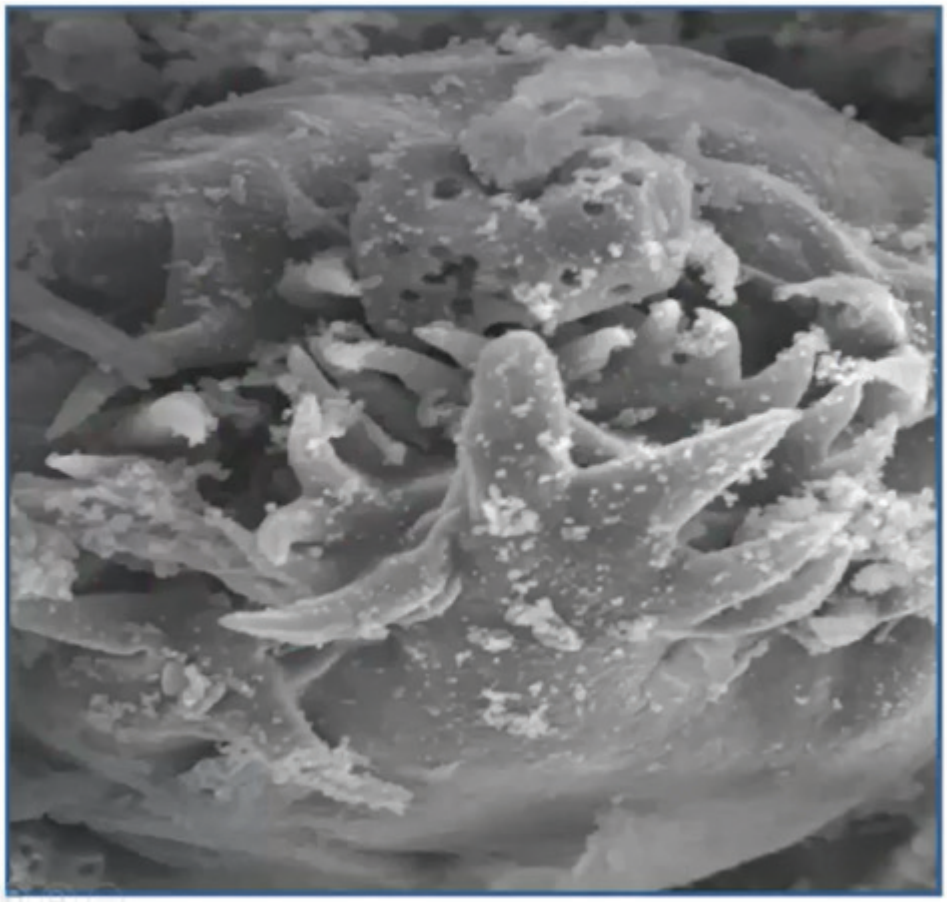

Fig. 4. Protozoa engulfing starch

4. Fungi

Fungi are the least populous of the rumen microorganisms (10^3 to10^6 zoospore/mL), and represent < 20% of rumen microbial biomass (Rezaeian et al., 2004). However, rumen fungi are among the most important microorganisms involved in the digestion of low quality forages such as cereal straw. Anaerobic fungi, also produce a wide array of carbohydrate degrading enzymes, but are most well-known for the highly active fibrolytic enzymes. Furthermore, they produce filamentous structures known as rhizoids that have the capacity to exert a physical force and penetrate highly lignified plant cell walls (Krause et al., 2003). The enzymes that they produce are concentrated at the tip of these structures and the openings that are created in the plant cell wall provide bacteria with access to the interior of the plant cell (**Fig. 5**).

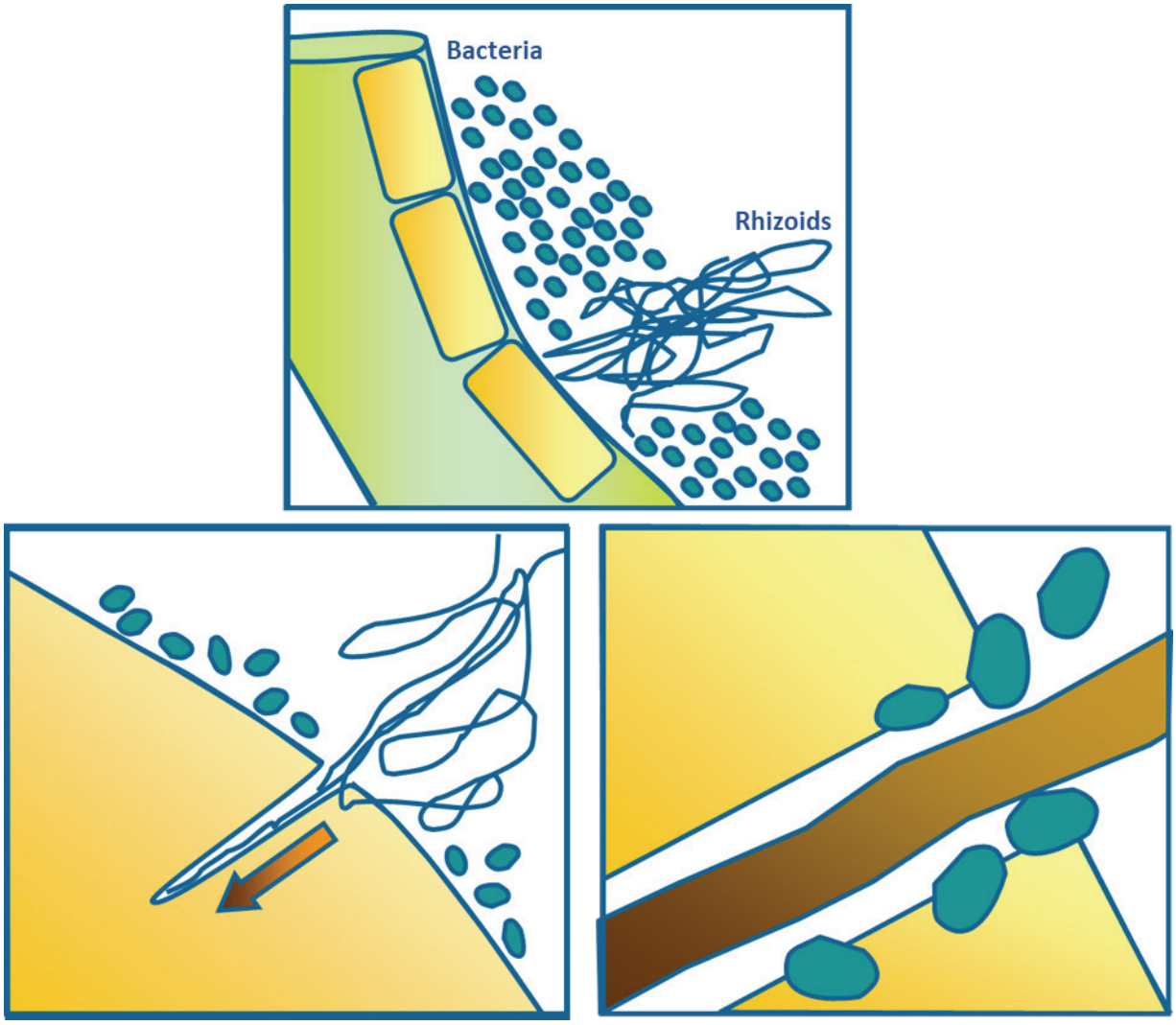

Fig. 5. Synergism between fungi and bacteria on colonizing plant material

5. Methanogens

Methanogens are members of a unique group of microorganism known as the Archaea, and are responsible for methane production in the rumen. Methanogens are not prominent members of the microbial community (10^6 cell/mL), but their ability to reduce methylamines, formate and carbon dioxide to methane is an integral component of fermentation. Eructation, (not flatulence) is responsible for the majority of methane produced in the rumen. Ruminal methane emission is not desirable for either the host or the environment, since it represents a significant energetic loss and has 25 times the global warming potential of carbon dioxide.

Because of that, considerable research effort has been expended into methods of methane mitigation. The most effective mitigation strategy is to reduce the amount of feed necessary to produce one unit of product (e.g., kg carcass; L of milk). Other strategies consist of the usage of feed additives (e.g., ionophores, probiotics or natural extracts) that are either toxic to methanogens or redirect H_2 to other electron acceptors (Leng, 2014). More recently, enzymes produced by bacteriophage (viruses that predate bacteria) have been also being researched (Altermann et al., 2018). However, due to the large variety of species and the capacity of methanogens to adapt,

several of these developed technologies only result in a short term reduction in methane emissions and some have had undesirable side effect on feed digestibility and host productivity.

6. Bacteriophage

Bacteriophages are "bacterial viruses" that reside in the rumen in concentrations of 10^{10} cells/mL of ruminal fluid. These viral particles need a host (bacteria) for replication which may culminate in rupture of the bacterial cell (Lytic cycle) or in the integration of the bacteriophage genome (Lysogenic cycle) into the genome of the host (**Fig. 6**)

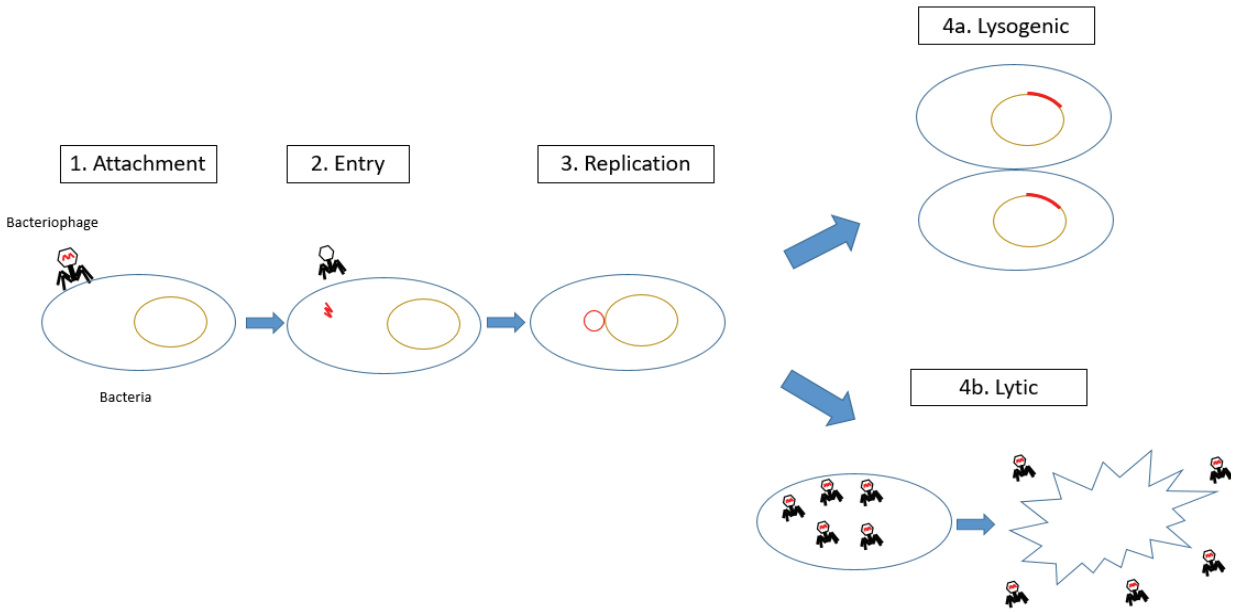

Fig. 6. Lysogenic and Lytic cycle of bacteriophage infecting rumen bacteria.

Bacteriophages are extremely host specific, often being able to infect only a few strains of a given bacterial species. To date, more than 125 different morphological types of bacteriophage have been identified, however there is still many more types that remain unidentified and uncharacterized. Although largely unstudied, it is well known that bacteriophage play an important role in density and types of bacteria that reside in the rumen. Recently, bacteriophage have been gaining increasing attention by rumen microbiologists. Although not presently practical on farm, it is believed that bacteriophage could be used to target undesirable bacterial species (e.g., *Streptococcus bovis*) in the rumen that contribute to digestive upsets such as acidosis and bloat. This approach may have the potential to replace the use of antibiotics in the future.

II. Rumen contents

1. Dimension

In ruminants, the digestion of fibrous material requires a fermentative chamber of a great volumetric capacity (reticulum and rumen), that can retain feed for the long period of time required to digest recalcitrant cell walls. Ruminants actually have a 4 chamber stomach (**Fig. 7**) which 80 % is accounted for by the rumen, 5 % by the reticulum, 8 % by the omasum, and 7 % by the

abomasum (Dyce et al. (2004). The rumen accounts for about 6 % of the live weight and in adult ruminants can have a volumetric capacity of up to 200 L.

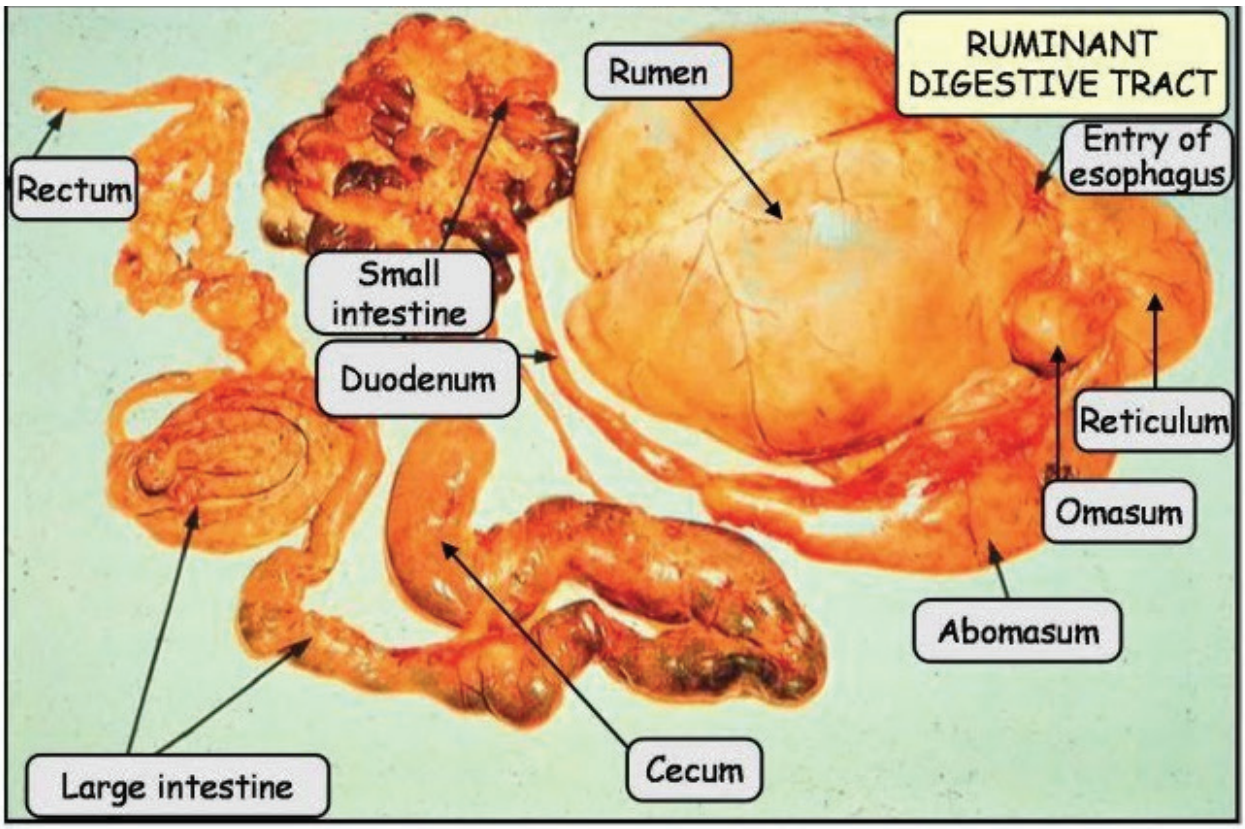

Fig. 7. The ruminant digestive tract

2. Osmolarity, pH and redox potential

The temperature in the rumen ranges between 38 °C and 41 °C, conditions that are optimal for growth of the rumen microorganisms. Ruminal pH is far more variable, ranging from 7.2 to as low as 4.6. Ruminal pH below 5.0 are undesirable as they are associated with subclinical or clinical acidosis. Ruminal pHs below 5.8 inhibit the activity of fibrolytic microorganisms and account for the inevitable decline in fiber digestion as a result of increasing levels of concentrate in the diet. Ruminal pH typically exhibits a diurnal patter where it declines shortly after feed consumption and recovers to pre-feeding levels prior to the next meal. Individual animals can vary substantially in their sensitivity to low pH, with some individuals going of feed when the pH declines below 5.4 and other showing no adverse effects even when the ruminal pH reaches 5.2

Diets with a high-fiber content (NDF > 40 %) and moderate amount of starch (<20 %), usually result in ruminal pH being no lower than 6.0- 6.2; as acid production occurs at a moderate rate and the presence of forage stimulates rumination and saliva production which contains sodium bicarbonate that buffers the rumen (Van Soest et al., 1991). However, with high-concentrate diets (over 65 % concentrate and/or more than 45 % of non-fibrous carbohydrates) are fed the rapid fermentation of starch accelerates the production of organic acids and the lack of fiber reduces rumination and saliva production. As a consequence, ruminal pH values frequently decline to levels below 6.0 (Agle et al., 2010; Kljak et al., 2017).

The rumen is an anaerobic and reduced environment with a redox potential (Eh) ranging between -250 e -450 mV. The strictly anaerobic microorganisms in the rumen require rumen digesta to be kept at a very low redox potential for their growth. In fact, if the redox potential becomes too high it can create conditions that benefit bacteria such as *Streptococcus bovis* that can grow in the presence or absence of oxygen. Growth of *Streptococcus bovis*, is considered undesirable because it produces high quantities of lactic acid.

Normally the osmolality (number of dissolved particles per unit of solvent) of rumen digesta ranges from 260 to 340 mosmol/kg. The degree of osmolality of rumen digesta depends on the extent to which end products accumulate in the rumen. For example, with a high grain diet rumen osmolality can reach 350 to 400 mosmol, conditions that can promote the lyses of bacterial cells and be indicative of the development of acidosis. These conditions can lead to the accumulation of water in the digestive tract and the development of diarrhea. High osmolality can also impair the ruminal digestion of fiber (240–265 mosmol) and starch (280–300 mosmol) (Garza et al., 1989).

3. The Short Chain Fatty Acids (SCFA)

Short Chain Fatty Acids (**SCFA**) are the major end products formed as a result of microbial fermentation and represent the principal energy source (50-70%) for the ruminant host. Acetic, propionic and butyric acid are the main SCFA produced in the rumen. In the rumen these acids are rapidly ionized to acetate, propionate and butyrate, respectively. The concentration of SCFA generally rank as acetate > propionate > butyrate, but with a high concentrate diet the amount of propionate produced increases and the ratio of acetate : propionate comes closer to 1:1. Supplying more concentrate in the diet also tends to increase the total concentration of SCFA in the rumen.

SCFA produced in the rumen are rapidly absorbed across the ruminal wall and while the majority of butyrate is metabolized in the ruminal epithelium, acetate and propionate enter into the bloodstream. Acetate is the main VFA metabolized in peripheral tissues, while propionate is utilized by the liver to synthesize glucose in a process known as gluconeogenesis. Glucose is required for proper function of the nervous system, with concentrations in the bloodstream maintained between 35–55 mg/dL in cattle and 35–60 mg/dL in sheep.

4. Saliva

Saliva is the main secretion of the digestive system, with adult cattle producing 170–180 L of saliva/day. The daily volume of saliva produced depends on the amount of time that the ruminant spends eating and ruminating. Although ruminants are well equipped to chew fibrous feeds, chewing is not efficient during the consumption of feed, thus feed needs to be regurgitated from the rumen, through the esophagus to the mouth where it is, re-chewed, re-salivated and re-swallowed.

Bovine saliva contains 126 mEq/L of sodium, 126 mEq/L of bicarbonate, 26 mEq/L of phosphate, 7 mEq/L of chloride, and 6 mEq/L of potassium. The bicarbonate ions (HCO_3), in saliva act as a buffer of ruminal pH. Phosphate in saliva ensures that this mineral is frequently introduced into the rumen to support microbial growth (Cunningham, 2011). Finally, saliva is also a rich source of N, with 65–70 % of the total nitrogen being in the form of urea. This non-protein N source can be used by rumen microorganism to synthesize amino acids which are intern assembled into microbial protein.

5. Protein absorption

The microbial protein produced in the rumen is the principal protein source of the ruminant host. The rumen is anatomically positioned before the abomasum and duodenum. Rumen microorganisms that flow from the rumen either with feed or fluid are subject to digestion in the abomasum and small intestine where released amino acids can be absorbed.

Often, protein levels in the diet may not be sufficient to meet the animals' requirements. Under these circumstances non-protein nitrogen and fermentable carbohydrates can be added to the diet in an effort to optimize the efficiency of microbial protein synthesis. Depending on the nature of the protein, as much as 60–90 % of the feed protein consumed can be degraded and the nitrogen released as ammonia (NH_3) in the rumen. The excess ammonia in rumen is absorbed across the rumen wall and converted into urea in the liver. If nitrogen is in excess, the urea will be excreted in the urine. However, if there is a deficiency of ruminal N, the urea can be recycled via the saliva or across the rumen wall where it can once again be used be the rumen microorganisms to synthesize amino acids (**Fig. 8**).

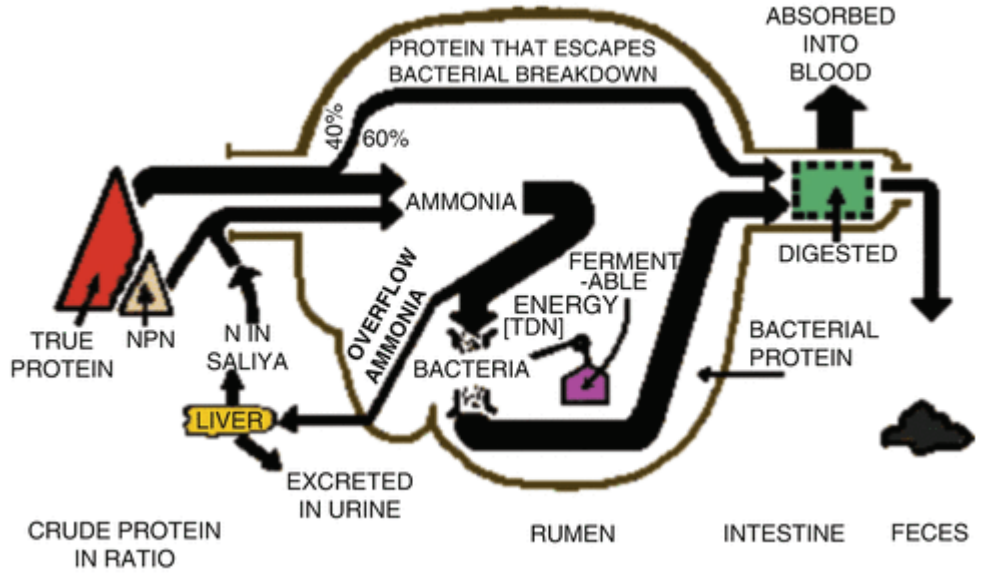

Fig. 8. Digestion and absorption of protein in ruminants (Satter, 1978).

6. Methane

As previous discussed, the fermentation activity of the rumen produces from 500–1000 L of gas per day in the rumen of an adult bovine. In general, this gas consists of 0.2 % hydrogen, 0.5 % oxygen, 7 % nitrogen, 26.8 % methane and 65.5 % carbon dioxide (Cunningham, 2011).It is the methanogenic *archea* that are responsible for the production of methane. Eructation is a vital and essential physiological mechanism for the removal of this gas from the rumen. Adult cattle produce 30–50 L of gas/h, whereas sheep and goats produce approximately 5 L/h (Cunningham, 2011). In cattle, CO_2 and methane account for 60–70 % and 30–40 % of the gas produced, respectively.

7. Minerals and vitamins

Rumen microorganisms require trace minerals to sustain the biochemical pathways that they employ in the fermentation of feed. Some in vitro studies suggest that deficiencies in Cu, Mn and Zn can negatively impact fiber digestion. However, most research has shown that compared to the

host, rumen microorganism require only trace amounts of Cu, Mn and Zn (Genther and Hansen, 2013).

In the rumen, microorganisms are capable of synthetizing all the B and K vitamin complexes that are required to meet the maintenance and growth requirements of beef cattle. As a result, ruminants do not require supplementation of these vitamins. Supplementation with B and K vitamins may be beneficial for calves and lambs which have yet to develop a fully functional rumen microbiome.

References

Agle, M., A. Hristov, S. Zaman, C. Schneider, P. Ndegwa, and V. Vaddella. 2010. Effect of dietary concentrate on rumen fermentation, digestibility, and nitrogen losses in dairy cows. Journal of Dairy Science 93(9):4211-4222.

Altermann, E., L. R. Schofield, R. S. Ronimus, A. K. Beatty, and K. Reilly. 2018. Inhibition of rumen methanogens by a novel archaeal lytic enzyme displayed on tailored bionanoparticles. Frontiers in microbiology 9:2378.

Cheng, K., R. McCowan, and J. Costerton. 1979. Adherent epithelial bacteria in ruminants and their roles in digestive tract function. The American journal of clinical nutrition 32(1):139-148.

Cunningham, J. 2011. Tratado de fisiologia veterinária. Elsevier Health Sciences.

Dyce, K. M., C. J. Wensing, and W. O. Sack. 2004. Tratado de anatomia veterinária. Elsevier Brasil.

Fernando, S. C., H. Purvis, F. Najar, L. Sukharnikov, C. Krehbiel, T. Nagaraja, B. Roe, and U. DeSilva. 2010. Rumen microbial population dynamics during adaptation to a high-grain diet. Applied and Environmental Microbiology 76(22):7482-7490.

Garza, F., F. Owens, and J. Breazile. 1989. Effects of diet on ruminal liquid and on blood serum osmolality and hematocrit in feedlot heifers. Miscellaneous publication-Agricultural Experiment Station, Oklahoma State University (USA)

Genther, O. N., and S. L. Hansen. 2013. The Effect of Trace Mineral Source and Concentration on Mineral Solubility in the Rumen and Diet Digestibility. Animal Industry Report 659(1):29.

Henderson, G., F. Cox, S. Ganesh, A. Jonker, W. Young, G. R. C. Collaborators, L. Abecia, E. Angarita, P. Aravena, and G. N. Arenas. 2015. Rumen microbial community composition varies with diet and host, but a core microbiome is found across a wide geographical range. Scientific reports 5:14567.

Kaufmann, W., H. Hagemeister, and G. Dirksen. 1980. Adaptation to changes in dietary composition, level and frequency of feeding, Digestive physiology and metabolism in ruminants. Springer. p. 587-602.

Kljak, K., F. Pino, and A. Heinrichs. 2017. Effect of forage to concentrate ratio with sorghum silage as a source of forage on rumen fermentation, N balance, and purine derivative excretion in limit-fed dairy heifers. Journal of dairy science 100(1):213-223.

Krause, D. O., S. E. Denman, R. I. Mackie, M. Morrison, A. L. Rae, G. T. Attwood, and C. S. McSweeney. 2003. Opportunities to improve fiber degradation in the rumen: microbiology, ecology, and genomics. FEMS microbiology reviews 27(5):663-693.

Leng, R. 2014. Interactions between microbial consortia in biofilms: a paradigm shift in rumen microbial ecology and enteric methane mitigation. Animal Production Science 54(5):519-543.

Nagaraja, T., and E. Titgemeyer. 2007. Ruminal acidosis in beef cattle: the current microbiological and nutritional outlook. Journal of Dairy Science 90:E17-E38.

Newbold, C. J., G. de la Fuente, A. Belanche, E. Ramos-Morales, and N. R. McEwan. 2015. The role of ciliate protozoa in the rumen. Frontiers in microbiology 6:1313.

Rezaeian, M., G. W. Beakes, and D. S. Parker. 2004. Distribution and estimation of anaerobic zoosporic fungi along the digestive tracts of sheep. Mycological research 108(10):1227-1233.

Satter. 1978. Use of nitrogen by the ruminant, Minnesota Nutrition Conference Proceedings.

Van Soest, P., J. Robertson, and B. Lewis. 1991. Symposium: carbohydrate methodology, metabolism, and nutritional implications in dairy cattle. Journal of Dairy Science 74(10):3583-3597.

Successful Blending of Nutrition and Animal Behavior on Robotic Milking Facilities

Dr. Mike Brouk
Professor and Extension Dairy Specialist
Kansas State University
mbrouk@ksu.edu
785-565-3434

Take Home Messages

- Nutrition and feeding must encourage cow movement
- Focus on cows the first 120 days of lactation
- Every extra pound of dry matter intake could equal an additional 3 pounds of milk
- Feed tables generally underestimate early lactation cattlel
- Provide opportunities to increase dry matter intake
- Forage quality is generally the most limiting nutritional factor
- Intake in early lactation is related to the quantity and digestibility of the fiber consumed
- Keep it simple

Introduction

Robotic milking systems continue to be installed at a fast pace in North America. In the last couple of years, larger dairy farms have begun exploring the possibility of adopting robotic milking and this interest is intensifying. Currently about 2% of the cows in the US are milked with robotic systems and this number is increasing quickly. Currently, box stalls that milk 1 cow at a time are the most popular. Use of these stalls requires that the dairy farm rethink and rearrange nearly all of the dairy chores of a conventional dairy farm. While there are advantages to these milking systems, successful adoption requires an open mind to new concepts and practices. The emphasis of this paper is to introduce some ideas and concepts of blending nutrition and animal behavior into a successful robotic milking system.

Facilities and Cow Flow

Facility design is critical in creating an environment that will allow cows to move freely toward the robotic milking stalls. Avoiding 90 degree turns, providing adequate width for 2-way cow traffic, avoiding slick surfaces, avoiding steps, avoiding dead ends and eliminating corners in the waiting area are key issues in encouraging cows to move freely around the pens. In addition, adequate space must be provided in front of the robotic milking unit to allow cows to freely enter the milking stalls. Designs which hinder cow movement will reduce cow flow and productivity. Timid cows need to be provided with adequate space to move by other animals in order to access the milking stall. Cow interactions between the resting area and milking robot are greatly increased when alleys are too narrow or if adequate space is not provided around the robot. Addressing these requirements in the design phase is a critical step toward achieving the desired cow flow in a robotic milking facility.

Functions of the Milking Stall

The milking stall is more than just a device that harvests milk. It collects various data and serves as a feeding stall. The goal of the system is to create a stable milking environment which exposes the cow to similar experiences at every milking. The system preps the teats for milking, harvests the milk and then applies a postdip to the teats. The system also checks for the presence of blood in the milk and discards non-salable milk. While the cow is being milked, a tremendous amount of data is collected about the milking visit, cow activity and cow health. In addition to data collection, the cow is offered an amount of feed, usually a pellet. The feed serves to attract the cow to the milking stall, focuses the cow's attention away from milking and provides additional nutrition. Cows on pasture will generally be grazing while the calf is nursing. Feeding may be important to reduce stress during the milking process, resulting in a reduction in incomplete milkings. Following milking, the system can also sort the animal for additional treatment or individual management procedures. The system is integral to the milking, feeding and management of the cow herd. It performs many of the routine dairy farm tasks without human assistance.

Robot Factors Involved in the Feeding Experience

The goal is to provide a consistent feeding experience at every milking. There are several settings that combine to determine the amount of feed offered at an individual milking. First, there is a maximum allowed per day. This may be based on milk production, days in milk or simply a fixed amount for each animal. This amount is generally divided by the expected number of milkings per day or allotted by the time between each milking based on a 24-hour period. Next, there is a setting which limits the amount which an animal can receive at each milking. The final setting is the rate at which the robot dispenses feed. Ideally, there will be feed dispensed over most of the milking. Near the end of milking, dispensing may stop to allow the cow to finish the meal before the end of milking. This encourages the cow to exit the stall immediately after milking rather than lingering to finish the meal or leaving feed for the next animal entering the stall. Lastly, the total amount of feed an animal can receive from the robot is directly related to the number of milkings each day. The number of milkings or permission to be milked is based either on the time since the previous milking or the amount of expected milk in the udder based on the individual cow's production and the length of time since the previous milking.

For example, if one allows milking permission of early-lactation cows every 4 hours, there would be 6 possible milkings each day. If the nutritionist and the dairy producer agreed that a maximum meal size at each milking was 3 pounds, then the total maximum allowed per day would be 18 pounds (6 milkings x 3 pounds each milking). However, very few cows will actually milk more than 4 times each day. Most animals visit 3 or fewer times each day. In practice, animals visiting for milking 5 or 6 times each day would be producing more than 150 pounds of milk daily. Remember, the system will allow milking permission based on time between the previous milking or expected milk yield. Now, if the meals size is 3 pounds, the dispensing rate is related to milking time. In general, most animals will milk in 5 to 6 minutes with a total box

time of 7 to 8 minutes. Dispensing 3 pounds over 6 minutes would suggest a dispense rate of 0.5 pounds per minute (3 pounds / 6 minutes). By following this logic, the machine should be set to provide a consistent feeding experience at every milking during the day.

Changes in the amount of feed offered or the quality of the feed at different milking times each day can result in decreased visits and incomplete milking. Feeding systems should provide for a similar feeding experience at every milking of the day. There are many settings that control the amount and rate of the feed that is dispensed during an individual milking. While there has been considerable focus on developing a consistent milking experience, there has been less attention given to the development of standards for the feeding system that allow for a consistent feeding experience. Depending on the various settings, cows may be offered different amounts of feed at each milking.

Type of Feed Offered in the Robot

There is considerable debate in the industry concerning the type of feed offered in the robot. Most farms with robotic milking systems will feed a pelleted feed in the robot. This provides a consistent density and moisture feed which is easily consumed by the cattle. Intake of pelleted feed by dairy cattle will be about 1.0 pound per minute as compared to about 0.6 pound for a meal-type feed. Fines generally encourage cattle to lick the feed manger and may result in cattle lingering in the milking stall after milking is complete.

Common feeds included in a pellet include corn, wheat, barley, soybean meal, canola meal, beet pulp, soybean hulls, wheat mids, corn gluten feed, salt and molasses. A commercial pellet binder may also be included to increase the pellet integrity. Addition of minerals, fats and feedstuffs that do not produce a hard pellet will increase the fines due to the number of times pellets are handled before reaching the cow. Inconsistent pellet quality or poor pellet quality will generally result in decreased milking visits and increased fetching effort. It is critical to have consistent pellet quality that entices intake when training animals.

Based on my experience with many different farms across the US and in Canada, feeding a high-quality pellet will increase visits and reduce fetching effort. Many farms have switched from pellets to a meal or other feed. Initially, cows seem to accept the new feed; however, after a period of only a few weeks, visits and milk production decrease and fetch effort increases. Most farms then switch back to the pelleted feed.

Increased Nutrition Cost with Robots

Feed cost often accounts for more than 50% of the total cost of milk production. With the recent decrease in feed prices, feed cost may be slightly less than 50% of total production cost, but it still represents the single greatest cost of production. When switching from conventional milking to robotic milking, daily feed cost per cow will generally increase. However, if there is also an increase in milk production, the feed cost per cwt of milk may actually decrease. One

of the critical factors in determining the cost of the feed fed in the robot is the additional cost associated with feeding a pelleted feed. The feed nutrients would need to be purchased, so the concern is the extra cost of feeding a pelleted feed. In general, one can assume that the cost of the pelleting will increase the cost of feed by $40 per ton. If feeding an average of 10 pounds of feed per day in the robot, this would amount to about $0.20 per cow daily. If milk is $0.15 per pound, this requires just over 1 pound of increased milk production to cover this expense. When calculating feed cost and making comparisons between conventional milking and robotic milking, it is important to convert feed cost to reflect changes in milk production rather than cost per cow. While cost per cow may increase, feed cost per unit of milk decreases. One should also consider the differences between shrink in the commodity shed vs shrink with an enclosed feeding system. Shrink in commodity sheds often increases actual cost by 5-15% depending on the feed and length of storage.

Consumption of Robot Feed vs Bunk Ration

Several studies have looked at the impact of feeding more feed through the robot and the resulting impact on intake at the feed bunk. These studies have used post-peak animals and have consistently shown that increased intake in the robot reduces feed intake at the bunk. However, it is important to note that the study animals were post-peak. Post-peak animals will rarely show an increase in milk production and intake is largely controlled by metabolic and hormonal factors. In early lactation, total intake is related to the extent of rumen fill and how often the rumen can be refilled. Cows in early lactation (pre-peak) are driven to consume additional feed with increased milk production. Thus, giving the opportunity to consume at the robot as well as at the bunk can result in an increase of feed at the bunk and at the robot. Since early lactation feed intake is on the increase, balanced intake at the bunk and robot is desirable.

Why Do Cows Flow

One of the critical factors in robotic milking systems is independent cow movement. In a conventional milking system, we create animal flow by moving cows from the housing area to the milking parlor 2 or 3 times daily. This creates movement to the bunk following milking and can stimulate feed intake. In a robotic system, we want the cattle to move independently to the milking center. Thus, understanding cow movement and drive is important. First, the animals must be healthy and standing on a sound set of feet and legs. Animals suffering from health issues or lameness tend to spend more time resting and will likely become fetch animals. Movement of early lactation animals is generally driven by hunger, which is related to the level of milk production. Cows with a higher level of milk production consume more feed and have a higher rate of passage than lower-producing cows. Thus, feeding to encourage increased early lactation milk production will generally result in increased visits to the milking center and increased cow movement. Feedstuffs differ in the rate of fermentation and forages have a much longer residence time than other feeds. Thus, forage digestibility can be a limiting factor in the rate at which the rumen can be refilled and can limit overall feed intake and thus milk production of the dairy herd.

Forage Digestibility and Cow Flow

The rate and extent of forage fiber digestibility can be measured in terms of uNDF at various time points. Most commonly, we look at 30, 120 and 240 hours of exposure. By looking at 3 different time points, we get a picture of the rate and extent of forage fiber digestion. A newer method is TTNDFD, which combines the rate and extent of fiber digestion into a single term. As forage quality decreases, there is an increase in NDF content and the digestibility of the NDF may also decrease. This results in a decrease in total intake and may result in an increase in the rumen residence time. For the cow, this is a double edged sword: intake decreases and each mouthful of feed contains less available nutrients. Loss of milk production and decreased intake coupled with increased rumen residence time will result in less cow movement. For robotic dairies dependent on spontaneous cow movement, this is a critical issue. When trying to achieve higher levels of milk production, forage quality is often the most limiting factor. Thus, attention to the details of forage quality for a successful robotic facility is more important than on a conventional facility due to the impact of forage quality on cow behavior and movement.

Focusing on the Right Cows

Successful robotic dairies focus on dry, pre-fresh and animals in the first 120 days of lactation. Animals with a successful transition into lactation will start faster, move more often and achieve higher levels of milk production than those that transition poorly. Again, one of the goals of robotic milking is to remove the barriers and reduce the need for human intervention. Fixing transition issues is a must. Providing appropriate nutrition and cow comfort, including cow cooling, is required.

Once cattle calve, get out to the way and let them go. Heifers and cows being introduced to robotic milking need to have appropriate training in the first 5 days of lactation. They should be flowing on their own by day 7. Flowing is defined as 3 or more independent visits daily. This may require 4-5 training visits per day in the first few days. One of the keys to attracting cows to the robot is a consistent feeding experience as described earlier. Animals do have expectations and we need to allow animals to access increased amounts of feed in early lactation relative to visits and milk production. Correct use of the robot feeding system can be an effective method to lead feed cows as they increase in production. As production rises, intake of both robot feed and the bunk should rise together. Due to the high level of nutrient demand in early lactation, giving cattle the opportunity to consume additional total feed can be a key factor in obtaining higher levels of milk production. Again, the robot limits the amount of feed at each visit, so increased total intake at the robot is tied to appropriate visits and milkings.

Tying It All Together

As one considers the adoption of robotic milking centers, it is important to understand the impact and interactions of cow behavior and nutrition. Removing the fetching to the milking parlor 2-3 times each day significantly impacts cow behavior. Offering a consistent feeding experience in the robot coupled with excellent quality forages will

enable a cow to exhibit more of its genetic capability and increase the success of robotic milking facilities. In many cases, producers have limited access to robot feed in early lactation, which results in decreased total intake and milk production as well as increasing fetching. Allowing the cow access to additional feed in response to increased milk production and visits to the robot will allow for a more successful lactation. The amount of feed offered in the robot needs to be consistent at each visit and offered in a safe amount to prevent rumen upset. Offering excessive amounts at each visit can be detrimental to cow health and milk component production. Focusing on early lactation cows to allow adequate intake of both the robot feed and the bunk is critical in achieving higher levels of milk production. Healthy cows in a higher state of milk production will move more in the barn and exhibit a greater number of milkings each day. This will reduce the need for fetching and improve the overall efficiency of the farm.

References

Bach, A. and V. Cabrera. 2016. Robotic milking: Feeding strategies and economic returns. J. Dairy Sci. 100:7720-7728.

Bach. A., C. Iglesias, S. Calsamiglia, and M. Devant. 2007. Effect of amount of concentrate offered in automatic milking systems on milking frequency, feeding behavior, and milk production of dairy cattle consuming high amounts of corn silage. J. Dairy Sci. 90:5049-5055.

Hare, K., T.J. DeVries, K.S. Schwartkopf-Genswein, and G.B. Penner. 2018. Does the location of the concentrate provision affect voluntary visits, and milk and milk component yield for cows in an automated milking system? Can. J. Anim. Sci. 98:399-404

John, A.J., S.C. Garcia, K.L. Kerrish, M.J. Freeman, M.R. Islam and C.E.F. Clark. 2017. Short communication: The diurnal intake and behavior of dairy cows when access to a feed of consistent nutritive value is restricted. J. Dairy Sci. 100:9279-9284.

Menajovsky, S.B., C.E. Walpole, T.J. DeVries, K.S. Schwartzkopg-Genswein, M.E. Walpole. 2018. The effect of the forage-to-concentrate ratio of a particular missed ration and the quantity of concentrate in an automatic milking system for lactating Holstein cows. J. Dairy Sci. 101:9941-9953.

Paddick, K.S., T.J. DeVries, K. Schwartzkoph-Genswein, M.A. Steele, M.E. Walpole, and G.B. Penner. 2018. Effect of the amount of concentrate offered in an automated milking system on dry matter intake, milk yield, milk composition, ruminal digestion, and behavior of primiparous Holstein cows fed isocaloric diets. J. Dairy Sci. 102:2173-2187.

Rodenburg, Jack. 2017. Robotic milking: Technology, farm design, and effects on work flow. J. Dairy Sci. 100:7729-7738.

Siewert, J.M., J.A. Salfer, and M.I. Endres. 2018. Factors associated with productivity on automatic milking system dairy farms in the Upper Midwest United States. J. Dairy Sci. 101:8327-8334.

Feeding strategies associated with robotic dairies

Gregory Penner[1], Jennifer Haisan[1,2], Keshia Paddick[3], and Trevor DeVries[4]

[1]University of Saskatchewan, Saskatoon, SK; [2]University of Alberta, Edmonton, AB; [3]Lakeland College, Vermillion, AB; [4]University of Guelph, Guelph, ON
Email: greg.penner@usask.ca

Introduction

Diversity in nutritional management among Canadian dairy farms is evident and dictated by factors such as producer goals, milking system, amounts and types of feed (silage, hay, and cereal grains) produced on farm and those purchased off farm. With automated (robotic) milking systems (AMS), the diversity is increased relative to that in total mixed ration (TMR) fed herds, because the division of the TMR into a partial mixed ration (PMR) and the AMS pellet imposes a fundamental shift in nutritional management. In addition, the nature of the PMR, allocation of the PMR, type of pellet, and feeding strategy of the pellet delivered in the AMS differ. The large diversity coupled with relatively recent adoption of AMS and limited controlled research regarding feeding management have led to recommendations being largely based on survey studies or based on anecdotal data from single-farm case studies. However, research on feeding management strategies for cows managed in AMS has increased, particularly in Canada, and this paper will describe the current state of knowledge along with areas where research is needed.

Does Cow-traffic Design Influence the Feeding Management Approach?

There are two main goals when considering the nutritional program for cows milked with AMS. The first, as with all planned nutritional programs, is to provide a diet that meets nutrient requirements for maintenance and production. However, with AMS, there is a perception that this goal can be shifted from the pen level to the cow level. Thus, producers could be providing a different diet for each cow within the same pen by adjusting the amount of pellet provided in the AMS. The second goal, which is unique to AMS, is to stimulate cows to voluntarily enter the AMS by dispensing pellet in the AMS. A disproportionately large focus has been placed on the AMS pellet, considering that the PMR provides the majority of the dry matter and nutrients consumed. For example, assuming a static dry matter intake (DMI) of 28 kg, the PMR could be estimated to contribute between 89 and 71% of the total dietary dry matter for cows offered three and eight kg of pellet in the AMS (dry matter basis), respectively.

Current survey data suggest that producers with free-flow traffic barns program greater AMS pellet allocations than those with guided-flow traffic barns (Salfer and Endres, 2018). Feeding greater quantities of pellet in the AMS, by default, also indicates the PMR will be less nutrient dense. While this may not be considered to be a problem, recent research has demonstrated that feeding a PMR with a greater proportion of forage increases the ability of cattle to sort that PMR (Menajovsky et al., 2018; Paddick et al., 2019). Providing more pellet in the AMS with free-flow barns is typically done because cows can choose when, and if, they voluntarily enter the AMS, whereas with guided flow barns, cows are ultimately directed to the commitment pen and the AMS using automated sorting gates. While the survey data indicate that producers with free-flow barns provide more pellet in the AMS, it is not known whether those cows consume more AMS pellet because the amount actually delivered and the amount consumed are not reported. The difference between the computer programmed value, amount delivered, and amount consumed for the AMS pellet is of major importance and will be discussed subsequently in this paper. Moreover, survey-based studies have neglected to evaluate PMR composition and do not have the ability to evaluate PMR intake at a cow level (Bently et al., 2013; Tremblay et al., 2016; Salfer and Endres, 2018). With single-farm case-studies, many confounding factors may promote a specific response, but the study cannot delineate which one or ones are contributing factors to that response. These confounding factors may or may not be related to the nutritional strategies and as such, data is not robust to apply more broadly. Thus, caution should be applied when considering survey-based data or single-farm case-study data as a means to evaluate potential recommended feeding strategies.

Salfer and Endres (2018) reported that the upper limit for pellet allocation in AMS (computer programmed value) in their survey was 11.3 kg /cow/day. Assuming cows could consume 11.3 kg/day, each cow would need to consume over 2.8 kg/milking (assuming 4 milkings/day) equal to 350 to 400 g/minute if milking duration was between seven and eight minutes. This high rate of pellet feeding may outpace the ability of cows to consume pellet while milking, and likely would result in a significant quantity of pellet that is either not delivered to the cow (Penner et al., 2017) or delivered in the AMS but not consumed by the cow (Bach and Cabrera, 2017).

Unfortunately, there is a lack of data evaluating whether traffic flow truly affects the amount of pellet required to be offered in the AMS. A study conducted in a feed-first, guided-flow barn reported no effect on voluntary attendance or milk yield when the amount of pellet delivered varied from 0.5 to 5.0 kg of DM/day (Paddick et al., 2019), whereas similar treatments in a free-flow barn resulted in more frequent voluntary milkings (DM basis; DeVries, personal communication). It would be nice to conclude that these data provide support for allocating greater quantities of AMS pellet under free-flow systems; however, the AMS pellet composition, PMR composition, total DMI, and days in milk also differed between the two studies thereby preventing a direct comparison. Moreover, Bach et al. (2007) reported that the amount of pellet provided in a free-flow system did not affect

voluntary attendance or milk yield. As a result, studies should not be interpreted to indicate the absolute amount of pellet provided because the amount likely differs on a farm-to-farm basis.

Does Increasing the AMS Pellet Allocation Increase Voluntary Attendance and Milk Yield?

One of the most common claims with AMS feeding strategies is that increasing the amount of pellet delivered in the AMS will stimulate voluntary attendance and milk yield. The approaches used to increase the AMS pellet allocation should be considered because there are two very different nutritional strategies. First, producers need to decide how much pellet is required from a basal level and this basal amount must consider the formulation of the PMR. Previous studies have been conducted to evaluate how the amount of pellet offered in AMS affects production responses when the total dietary nutrient supply is equivalent. In other words, with this strategy, increasing the amount of pellet provided in the AMS requires an equal reduction in the amount of pellet in the PMR such that the total diet (PMR + AMS) does not differ. The first study published using this nutritional strategy compared treatments with computer programmed values of three or eight kg of pellet in the AMS in a free-flow barn design (Bach et al., 2007). In that study, despite having programmed values of 3 and 8 kg/day, pellet delivery was 2.6 and 6.8 kg/day (dry matter basis) and the amount of pellet delivered did not affect milk production or milk component production. In two recent studies conducted in a guided-flow barn at the University of Saskatchewan, AMS pellet delivery ranged between 0.5 and 5.0 kg of dry matter/cow/day (Hare et al., 2018; Paddick et al., 2019). Altering the amount of AMS pellet while maintaining equal dietary nutrient composition did not affect voluntary visits, milk yield or milk component yield. In contrast, a recent study conducted at the University of Guelph in a free-flow barn reported that with total diets (PMR + AMS pellet) that were the same in nutrient composition, increasing the AMS pellet from 3 to 6 kg/day (and correspondingly reducing the same pellet in the PMR), stimulated DMI, increased voluntary visits by 0.5 milkings/day, and numerically (but not significantly) increased milk yield by 1.5 kg/day (DeVries, personal communication).

It might seem counter-intuitive that increasing the AMS pellet allocation does not necessarily stimulate voluntary visits or milk yield. However, simply providing more pellet in the AMS does not necessarily translate to greater DMI. For example, Hare et al. (2018) reported that for every 1 kg increase in AMS pellet delivered, there was a corresponding decrease in PMR DMI of 1.58 kg. Bach et al. (2007) reported a 1.14 kg reduction in PMR DMI and Paddick et al. (2019) reported that PMR DMI decreased by 0.97 kg for every one kg increase in AMS pellet delivered. The large or at least equal reduction in PMR DMI with increasing AMS pellet intake demonstrates that nutrient intake may not be positively affected. In contrast, DeVries (personal communication) reported that for every 1 kg increase in AMS pellet intake there was only a 0.63 kg reduction in PMR DMI (Table 1). In that case, providing more pellet in the AMS resulted in greater total DMI and likely explains the numerical improvement in milk yield observed in that study. The variable and currently unpredictable substitution rate may challenge the ability to formulate diets for individual cows in the same pen given that only the amount or types of pellet in the AMS can differ. It should be noted that the inability to predict the substitution rate (and hence PMR intake) does not preclude imposing such precision feeding programs; we simply cannot evaluate the individual response or adequately predict the outcome. Clearly, this remains a challenge for nutritionists and producers alike.

Table 1. Effect of increasing pellet in the automated milking system (AMS) on the reduction in PMR intake (DM basis).

Study	DIM (mean ± SD)	Cows, parity, and study design	Traffic and diet, dietary scenario	Substitution ratio, kg PMR/kg AMS concentrate
Bach et al., 2007	191 ± 2.13	69 primiparous Holstein, 46 multiparous Holstein Completely randomized design	Free Isocaloric	1.14
Hare et al., 2018	227 ± 25 123 ± 71	5 multiparous Holstein 3 primiparous Holstein	Guided Isocaloric	1.58
Henriksen et al., 2018	32-320 14-330	22 primiparous Holstein, 19 multiparous Holstein 11-week study	Free Static PMR with 2 concentrate	0.58 – 0.92
Henriksen et al., 2018	29-218 17-267	14 primiparous Jersey 28 multiparous Jersey 11-week study	Free Static PMR with 2 concentrate allocations	0.69-0.50
Menajovsky et al., 2018	141 ± 13.6	8 multiparous Holstein Replicated 4x4 Latin square	Guided Low energy PMR High energy PMR	0.89 0.78
Henriksen et al., 2019	Early (5 to 14) Mid (15 to 240) Late (241 to 305)	128 cows (68 Holstein + 60 Jersey) Continuous lactation study	Free Static PMR with 2 differing concentrate allocations	5 1.1 2.9
Paddick et al., 2019	90.6 ± 9.8	8 primiparous Holstein Replicated 4x4 Latin square	Guided Isocaloric	0.97

As a second strategy, the energy density of the diet for an individual cow can be changed by increasing or decreasing the AMS pellet allocation without changing the composition of the PMR. This approach is one strategy to apply precision feeding management. There has been limited research with this strategy; however, in a recent study where cows received two or six kg of AMS pellet (dry matter basis), there were only subtle differences in milking frequency and only numerical improvements for milk and milk protein yield (Menajovsky et al., 2018). At a farm level, Tremblay et al. (2016) reported a negative relationship between the amount of pellet offered in the AMS and milk yield. Their rationale was that poor forage quality requires more pellet; however, there was no information provided on PMR characteristics. To our knowledge, there is still a lack of research focusing on the use of precision feeding strategies, particularly with high-yielding and early lactation cows.

A challenge with adopting precision feeding strategies is that predictions are needed for the amount of PMR and AMS pellet that the cow will consume on a daily basis. The data are clear that increasing the quantity of AMS pellet offered in the AMS increases the day-to-day variability in the consumption of the AMS pellet and hence creates more dietary variability (Hare et al., 2018; Menajovsky et al., 2018; Paddick et al., 2019). Based on the available data, the coefficient of variation (CV) in AMS pellet delivered averages 13.5%. Using this CV, we can calculate the standard deviation for AMS pellet delivery by multiplying the amount delivered by the CV (Figure 1). Using this approach, it is clear that as the amount of AMS pellet delivered increases, the day-to-day variation in the amount delivered also increases. In fact, we would expect that the day-to-day variation in the amount of pellet delivered for 96% of the cows would increase from 0.54 kg/day to 2.7 kg/day as the AMS pellet delivered increases from 2 to 10 kg/day. Using a 10 kg/day value and a fixed DMI of 28 kg/day, we would expect that AMS pellet would range between 8.7 and 11.4 kg/day. If we assume that total DMI (AMS pellet + PMR) is relatively constant, the variability in AMS pellet delivery could imply that PMR intake could also vary from 19.4 to 16.7 kg/d. However, the amount of pellet offered in the AMS did not affect PMR intake or variability in PMR intake in previous studies (Menajovsky et al., 2018; Paddick et al., 2019).

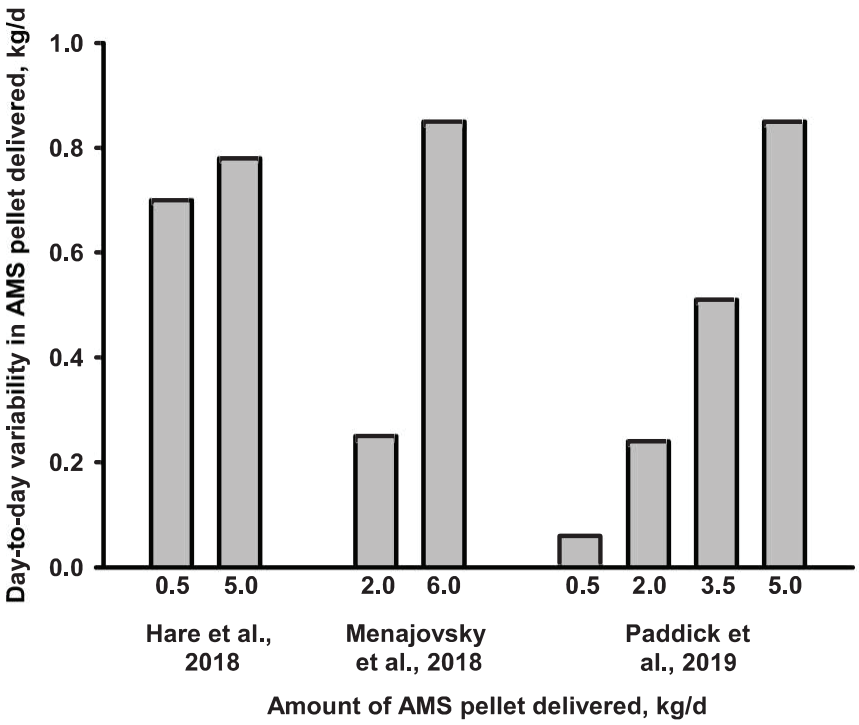

Figure 1. Variability in day-to-day pellet delivered in the AMS based on the amount of pellet offered in the AMS.

In most studies, a fundamental assumption is that as AMS pellet delivered, and presumably consumed, increased, PMR intake would decrease with an equal magnitude. We know this assumption is not true as substitution rates (amount of decrease in PMR intake for every 1 kg increase in AMS pellet intake) range from 0.62 to 1.58 kg (Table 1). Obviously, the reduction in PMR intake with increasing AMS pellet allocation will change the nature of the total diet and depending on the direction and magnitude of the PMR substitution, the proportions of forage neutral detergent fibre (NDF) or physically effective NDF may become marginal coupled with increases in ruminally degradable starch.

In AMS systems, there are three values that are relevant when considering AMS pellet delivery. The first value is the computer programmed target value. This value is the maximum amount that can be offered to cows in the AMS, assuming that carry-over of pellet is not included in the equation. The second value is the amount that is delivered to the cows in the AMS. The third value is the amount consumed in the AMS. The amount of pellet programmed in the computer does not correspond with the

amount delivered (Figure 2). For example, Bach et al. (2007) allocated either 3 or 8 kg/day in the AMS but only 2.6 and 6.8 kg/day were delivered, respectively.

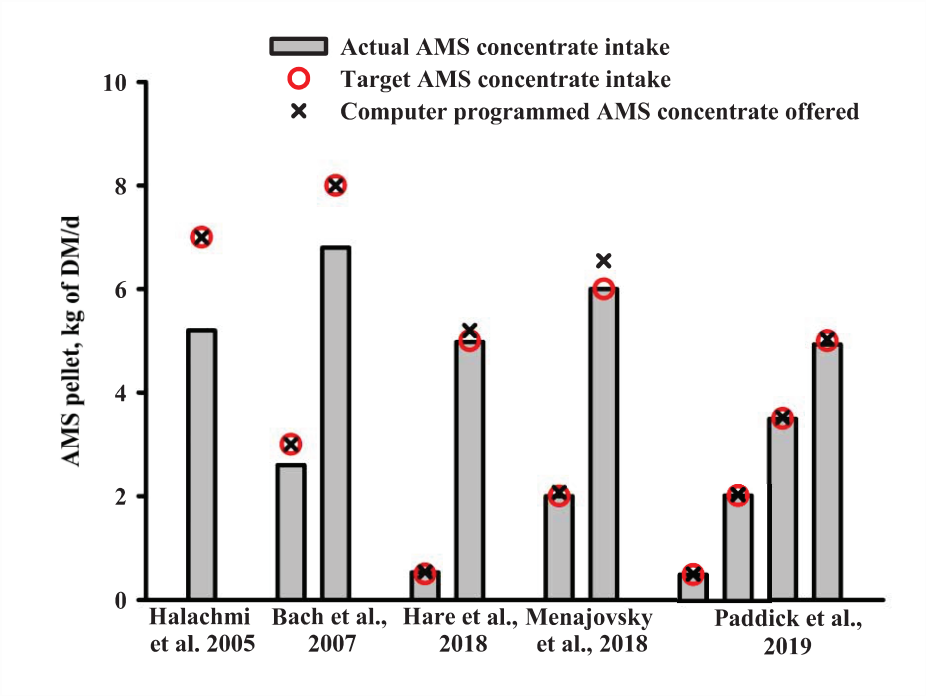

Figure 2. Comparison of computer programmed target AMS pellet allocation and AMS pellet consumption. The circles indicate the target quantity of AMS pellet desired, the 'x' indicate the computer programmed quantity, and the grey vertical bars indicate the average quantity that cows are delivered (adapted from Paddick and Penner, 2018).

Halachmi et al. (2005) offered either 7 kg/day or 1.2 kg/visit to cows and reported that cows offered 7 kg/day were only delivered 5.2 kg/day while those offered 1.2 kg/visit received 3.85 kg/day. Pellet delivery and pellet consumption below that of the formulated diet are major concerns. Evaluating the deviation between the amount programmed and the amount offered is an important management tool because it demonstrates the ability to deliver the formulated diet to the cows. The deviation between the amount programmed and the amount delivered increases as the amount programmed increases (Figure 2). This can also be viewed under commercial settings (Figure 3). The data in Figure 3 were obtained from a commercial free-flow barn in Alberta and demonstrate some important findings.

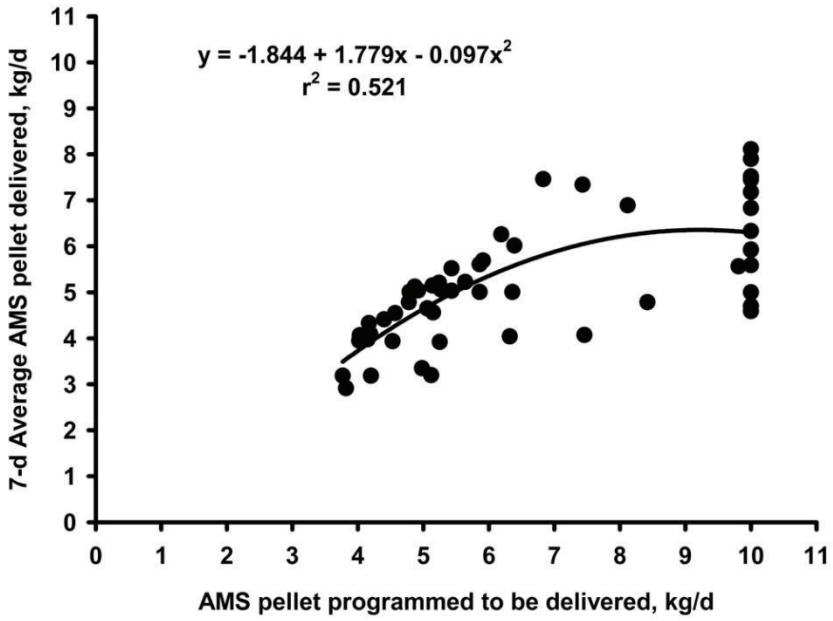

Figure 3. Relationship between the programmed quantity of pellet to be provided in the AMS and the actual amount delivered in a commercial herd in Alberta.

Firstly, in this barn, the average maximum pellet delivered was approximately 6.3 kg in the AMS, although some individual cows were delivered as much as 8.1 kg. It is important to note these maxima as the amount of pellet programmed to be available was 10 kg. Data from this farm also show a similar response as reported by Menajovsky et al. (2018) and Paddick et al. (2019). Specifically, as the quantity of AMS pellet programmed increases, the deviation between the computer programmed quantity and the amount that is delivered increases (Figure 4). Based on a linear regression, we would expect that cows programmed to receive 2.7 kg did in fact receive that quantity, while as the quantity of AMS pellet programmed increased by 1 kg, cows only were delivered an additional 0.62 kg. However, the variability in the difference between the programmed and delivered quantities was very large, particularly at the higher target pellet allowances. While it cannot be evaluated on farm easily, residual pellet left in the AMS feeder also increases with increasing pellet allocation in the AMS (Bach and Cabrera, 2017). Differences among the amount of pellet programmed, amount delivered in the AMS, and amount consumed by cows in the AMS can pose a challenge to dairy producers and their nutritionists, and diminish the ability to formulate diets that reasonably predict production outcomes.

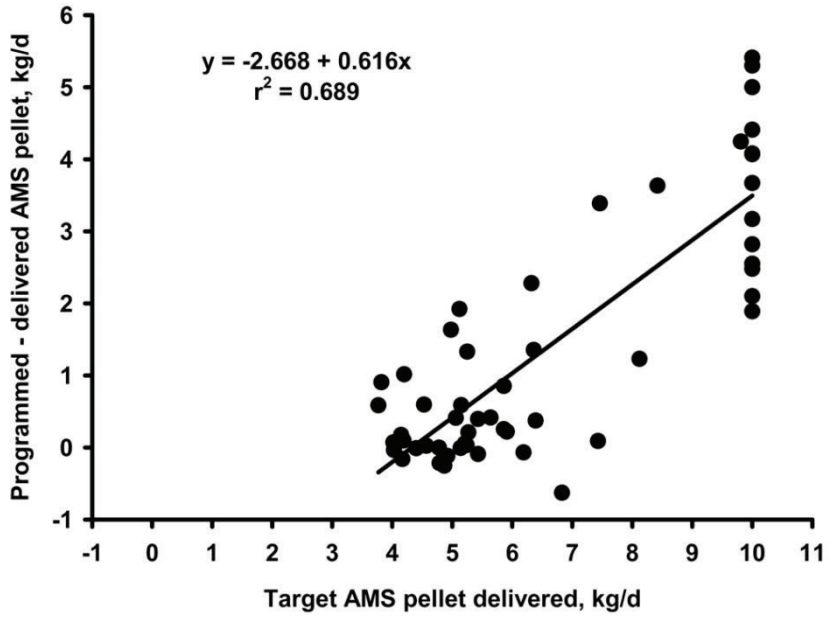

Figure 4. Influence of the amount of AMS pellet programmed for delivery and the difference between the programmed and delivered AMS pellet on a commercial farm in Alberta.

From a nutritional standpoint, minimizing the range in the amount of pellet allocated in the AMS among cows within a group can help to ensure the diet is adequately balanced despite still allowing for differences in the amount of pellet allocated through the AMS. Maintaining a moderate quantity of pellet provided in the AMS can also reduce the bias between the computer programmed amount, amount delivered to the cow in the AMS, and amount consumed by the cow in the AMS. Moderate pellet allocations in the AMS should also minimize day-to-day variability in AMS pellet delivered (Figure 1).

Automated milking systems also enable producers to impose adaptation programs for cows in early lactation. While increasing the energy density of the diet by increasing pellet allocation may seem like a plausible option, recent results suggest that such an approach may actually decrease DMI and milk yield (Deiho et al., 2016). We are not aware of any studies that have evaluated precision feeding strategies to determine whether such approaches improve milk and milk component yield and profitability. Studies that have evaluated fermentability of the diets following parturition have shown that increasing the rate of grain inclusion (Deiho et al., 2016), or increasing fermentability by including more rapidly fermentable grain sources may not be optimal (Albornoz and Allen, 2016). Deiho et al. (2016) reported that cows adapted to a diet that consisted of a concentrate supplement (45% DM basis) with the remainder of the diet consisting of grass silage, corn silage, and soybean meal. Their results showed that increasing concentrate gradually (0.25 kg/day) resulted in greater milk production than increasing concentrate rapidly (1 kg/day increase). Despite the improvement in milk yield, fermentable organic matter intake was less for gradually adapted cows than rapidly adapted cows, suggesting that feeding strategies designed to more closely meet nutrient demand may overwhelm the ability of cows to consume such diets and may not improve performance. Increased concentrate feeding is expected to increase feed costs without the corresponding increase in revenue. Although Albornoz and Allen (2016) used a completely different model, they found that replacement of dry rolled corn with high moisture corn reduced DMI and milk yield. Collectively, these studies highlight the need for future research under AMS conditions.

Is the AMS Pellet Likely to Induce Ruminal Acidosis?

There is often concern about risk for ruminal acidosis with AMS because a component feeding system is imposed and large quantities of pelleted feed may be programmed to be offered through the AMS. We have recently reported that the PMR formulation, rather than the quantity of pellet in the AMS, has a greater impact on ruminal pH (Menajovsky et al., 2018). It is logical that the PMR had greater impact than the AMS pellet considering it accounted for over 80% of the DMI in that study. Additionally, AMS pellet meal size in that study was constrained to a maximum of 2.5 kg and the amount delivered in the AMS was managed to not exceed 6 kg/cow/day on a DM basis. Based on recent information, cows in commercial operations may be provided up to 11.2 kg (as fed basis) of pellet in the AMS (Salfer and Endres, 2018). With this strategy, large swings in dietary composition can occur based on the expected reduction in PMR intake and increased pellet intake in the AMS. Under such scenarios, we could expect that the dietary physically effective NDF content would be dramatically reduced (and potentially

deficient) and that ruminally degradable carbohydrate content would increase thereby creating a diet (PMR + AMS pellet) that could be perceived to be high risk for ruminal acidosis. Currently, there are no data to support or dispute the previous claim.

How Important is the Type of Supplement Provided in the AMS?

In addition to general feeding management, palatability of the pellet provided in the AMS is also important. Madsen et al. (2010) evaluated pellets containing barley, wheat, a barley-oat mix, maize, artificially dried grass, or pellets with added lipid with all cows fed a common PMR. They observed that AMS pellet intake and voluntary visits were greatest when the pellets contained the wheat or the barley-oat mix. However, pelleted barley and wheat are expected to have a rapid rate of fermentation in the rumen and feeding substantial quantities would be expected to increase the risk for low ruminal pH. To reduce fermentability, pellets could be prepared with low-starch alternatives (Miron et al., 2004; Halamachi et al., 2006 and 2009). Substituting starch sources with soyhulls did not negatively affect voluntary attendance at the AMS or milk yield (Halamachi et al., 2006, 2009), and may slightly improve milk fat and reduce milk protein concentrations (Miron et al., 2004).

Producers may also choose to use home-grown feeds in the AMS. In a recent study, we tested whether feeding a pellet was required or if we could deliver steam-flaked barley as an alternative (Gardner et al., unpublished) in a feed-first guided-traffic flow barn. In that study, the pellet comprised only barley grain and the same source of barley grain was used for the steam-flaked treatment. In all cases, cows were programmed to have 2.0 kg of the concentrate in the AMS delivered. While PMR (27.0 kg/d DM basis) and AMS concentrate intake (1.99 kg/d DM basis) did not differ among treatments, cows fed the steam-flaked barley tended to have fewer visits (2.99 vs. 2.83; $P = 0.07$) to the AMS, tended to have a longer interval between milking events (488 vs. 542 minutes; $P = 0.10$), and spent 28 minutes more in the commitment pen prior to entering the AMS ($P = 0.01$) than those fed pelleted barley. While this did not translate into differences in milk yield (average of 44.9 L/d), it may be expected that with a longer-term study, production impacts would be observed. In contrast, Henriksen et al. (2018) reported greater voluntary visits when a texturized feed (combination of pellet and steam-rolled barley) was provided in comparison to a pellet alone. Regardless, utilization of a pellet as the sole ingredient or part of the mix may limit the ability of producers to use home-grown feeds in the AMS.

Partial Mixed Ration: the major, but forgotten component of the diet

As mentioned previously, all surveys that have been published to date focus on AMS feeding with little or no information collected to describe PMR composition or intake. The lack of focus on the PMR is likely because only group intakes can be determined and many of the studies have been conducted using retrospective analysis. However, drawing conclusions or making recommendations for feeding management without considering the PMR could lead to erroneous decisions. We recently completed a study where we varied the formulation of the PMR such that we increased the energy density of the PMR by a similar magnitude to that commonly used when increasing the amount of pellet in the AMS (Menajovsky et al., 2018). Feeding the PMR with a greater energy density tended to increase milk yield (39.2 vs. 37.9 kg/d; $P = 0.10$) likely because of greater energy supply. In several studies we have also noted that formulation of the PMR impacts sorting characteristics of the PMR (Menajovsky et al., 2018; Paddick et al., 2019). In both cases, reducing the energy density of the PMR (greater forage content as a percentage of DM) increased the sorting potential of the PMR. This may lead to cows selecting for dietary components in an undesirable manner (Miller-Cushon and DeVries, 2017). Future research is needed to understand how PMR composition can affect the ability to stimulate voluntary visits and to meet nutrient requirements for cows milked with AMS.

Conclusions

The use of AMS systems is increasing in Canada and sound feeding management practices are needed to support efficient and cost-effective milk production. The data that are available do not support the recommendation that feeding greater quantities of pellet in the AMS will result in greater milk production, likely because of the overall shift in the diet as cows substitute PMR for pellet. Moreover, feeding to meet milk production by increasing AMS pellet provision may not result in the expected benefits, again potentially due to a reduction in PMR intake and the potential shifts in dietary forage-to-concentrate ratio when both the PMR and AMS pellet are considered. Our data suggest that low-to-moderate AMS pellet provision will help to minimize variability in AMS pellet intake and, therefore, allow cows to consume diets more similar to that formulated. Low-to-moderate AMS pellet provision may also allow for greater flexibility for the pellet composition provided in the AMS. Regardless of the strategy employed, producers must not only consider the AMS feeding strategy, but also the interaction between how AMS feeding approaches may alter PMR consumption.

References

Bach, A., and V. Cabrera. 2017. Robotic milking: Feeding strategies and economic returns. J. Dairy Sci. 100:7720-7728.

Bach, A., C. Iglesias, S. Calsamiglia, and M. Devant. 2007. Effect of amount of concentrate offered in automatic milking systems on milking frequency, feeding behavior, and milk production of dairy cattle consuming high amounts of corn silage. J. Dairy Sci. 90:5049-5055.

Bently, J.A., L.F. Tranel, L. Timms, and K. Schulte. 2013. Automated milking systems (AMS) – Producer surveys. Animal Industry report (AS 659). http://lib.dr.iastate.edu/cgi/viewcontent.cgi?article=1826&context=ans_air. Accessed April 10, 2016.

Dieho, K., A. Bannink, A.L. Guerts, J.T. Schonewille, G. Gort, and J. Dijkstra. 2016. Morphological adaptation of rumen papillae during the dry period and early lactation as affected by rate of increase of concentrate allowance. J. Dairy Sci. 99:2339-2352.

Halachmi, I., S. Ofir, and J. Miron. 2005. Comparing two concentrate allowances in an automatic milking system. Anim. Sci. 80:339-343.

Halamachi, I., E. Shoshani, R. Solomon, E. Maltz, and J. Miron. 2006. Feeding of pellets rich in digestible neutral detergent fiber to lactating cows in an automatic milking system. J. Dairy Sci. 89: 3241-3249.

Halamachi, I., E. Shoshani, R. Solomon, E. Maltz, and J. Miron. 2009. Feeding soyhulls to high-yielding dairy cows increased milk production but not milking frequency, in an automatic milking system. J. Dairy Sci. 92:2317-2325.

Hare, K., T. DeVries, K. Schwartzkopf-Genswein, and G.B. Penner. 2018. Does the location of concentrate provision affect voluntary visits, and milk and milk component yield for cows in an automated milking system? Can. J. Anim. Sci. 98:399-404.

Henriksen, J.C.S., L. Munksgaard, and M.R. Weisbjerg. 2018. Short-term responses in production and behavior during periods of change in concentrate allowance for dairy cows. J. Dairy Sci. 101:7942-7953.

Henriksen, J.C.S., M.R. Weisbjerg, P. Løvendahl, T. Kristensen, and L. Munksgaard. 2019. Effects of an individual cow concentrate strategy on production and behavior. J. Dairy Sci. 102:2155-2172.

Madsen, J., M.R. Weisbjerg, and T. Hvelplund. 2010. Concentrate composition for automatic milking systems – Effect on milking frequency. Livest. Sci. 127:45-50.

Menajovsky S.B., C.E. Walpole, T.J. DeVries, K.S. Schwartzkopf-Genswein, M.E. Walpole, and G.B. Penner. 2018. The effect of the forage-to-concentrate ratio of the partial mixed ration (PMR) and the quantity of concentrate in an automatic milking system (AMS) for lactating Holstein cows. J. Dairy Sci. 101:9941-9953.

Migliorati L., M. Speroni, S. Calogero, and G. Pirlo. 2009. Influence of feeding flavouring-appetizing substances on activity of cows in an automatic milking system. Ital. J. Anim. Sci. 8:417-419.

Migliorati, L., M. Speroni, S. Lolli, and F. Calza. 2005. Effect of concentrate feeding on milking frequency and milk yield in an automatic milking system. Ital. J. Anim. Sci. 4:221-223.

Miller-Cushon, E.K., and T.J. DeVries. 2017. Feed sorting in dairy cattle: Causes, consequences, and management. J. Dairy Sci. 100:4172-4183. doi:10.3168/JDS.2016-11983.

Miron, J., E. Yosef, M. Nikbachat, A. Zenou, E. Maltz, I. Halachmi, and D. Ben-Ghedalia. 2004. Feeding behavior and performance of dairy cows fed pelleted nonroughage fiber byproducts. J. Dairy Sci. 87:1372-1379.

Miron, J., M. Nikbachat, A. Zenou, D. Ben-Ghedalia, R. Solomon, E. Shoshani, I. Halamachi, N. Livshin, A. Antler, and E. Maltz. 2004. Lactation performance and feeding behavior of dairy cows supplemented via automatic feeders with soy hulls or barley-based pellets. J. Dairy Sci. 87:3808-3815.

Paddick, K.S., T.J. DeVries, K. Schwartzkopf-Genswein, M.A. Steele, M.E. Walpole, and G.B. Penner. 2019. Effect of the amount of concentrate offered in an automated milking system on milk yield, milk composition, ruminal digestion, and behavior of primiparous Holstein cows fed iso-caloric diets. J. Dairy Sci. 102: 2173–2187.

Salfer, J., and M. Endres. 2018. Feeding and management practices for robotic milking success. Proc. 4-State Dairy Nutrition Conference. pp. 77-80. June 13 and 14. Debuque, Iowa, USA.

Tremblay, M., J.P. Hess, B.M. Christenson, K.K. McIntyre, B. Smink, A.J. van der Kamp, L.G. de Jong, and D. Döpfer. 2016. Factors associated with increased milk production for automatic milking systems. J. Dairy Sci. 99:3824–3837. doi:10.3168/jds.2015-10152.

TECHNICAL SYMPOSIUM SPEAKERS

Phil Cardoso, Ph.D. is an associate professor at the University of Illinois at Urbana-Champaign. He received his D.V.M. and M.S. degrees from the Universidade Federal Do Rio Grande do Sul in Brazil, and his Ph.D. from the University of Illinois. Since 2012, Cardoso has established a unique program that seamlessly blends his teaching, extension, and research efforts using a business model to give students opportunities to evaluate dairy farms. Phil's Dairy Science program impact by placing students in applied positions and academia. Phil and his students have published over 51 peer-reviewed manuscripts (original research and invited reviews) and 3 invited book chapters between 2011 and 2018. Of those, 22 papers have been published in J. Dairy Sci., and over 40 deal with research on nutritional physiology and reproduction of dairy cows. His research builds from questions asked by dairy producers and focuses on the impact of nutrition on metabolism, reproduction and health in dairy cows, as well as mechanisms of metabolic adaptation and forage quality.

Sebastian Arriola Apelo, Ph.D. is an Assistant Professor at the University of Wisconsin-Madison. The overall objective of Dr. Arriola's research program is to improve dietary nitrogen utilization and reduce nitrogen excretion in ruminants. To achieve that objective, his research focuses in understanding post-absorptive utilization of amino acids by splanchnic and peripheral tissues, including mammary gland. Dr. Arriola is particularly interested in cellular mechanisms that regulate tissue extraction of amino acids and synthesis of proteins.

Paul Kononoff, Ph.D. is a native of Saskatoon, Saskatchewan, Canada. Paul holds a B.S.A and a M.Sc. in Animal Science from the University of Saskatchewan (Saskatoon, SK) and a Ph.D. in Dairy Nutrition from The Pennsylvania State University (University Park, PA). A dairy nutritionist, he has industry experience as a Technical Support Specialist for Renaissance Nutrition (Roaring Spring, PA) and as a Project Director of the Ruminant Feed Analysis Consortium, formally located at the University of New Hampshire (Durham, NH). Since 2005 Paul has been employed at the University of Nebraska-Lincoln with an appointment of 68% research, 30 % extension, and 2% service. As a Dairy Nutrition Extension Specialist, he is involved with the education of dairy producers and allied industry personnel on nutritional methods to optimize profitability in a sustainable manner. Additionally, and in collaboration with others, he develops programs that support dairy management strategies and systems to facilitate profitable and sustainable decisions by dairy farm managers. Paul has several major areas of dairy nutrition research namely, 1) forage particle size and effective fiber, 2) feed characterization, 3) byproducts, 4) and energy metabolism. Most recently, he has built a program and research capabilities which employs indirect calorimetry to explore the effects of diet on methane production and

whole animal energy utilization in lactating dairy cattle. His research has impacted the dairy industry by improving the nutritional knowledge of dairy producers and nutritionists. Paul currently serves as a Senior Editor for the Journal of Dairy Science and serves on the NRC Committee of Nutrient Requirements of Dairy Cattle (8th Revised Edition).

Mike Van Amburgh, Ph.D. is a Professor in the Department of Animal Science and a Stephen H. Weiss Presidential Fellow at Cornell University where he has a dual appointment in teaching and research. His undergraduate degree is from The Ohio State University and his Ph.D. is from Cornell University. He teaches multiple courses and leads the Cornell Dairy Fellows Program, advises approximately 50 undergraduate students and is the advisor for the Cornell University Dairy Science Club. Mike currently leads the development of the Cornell Net Carbohydrate and Protein System, a nutrition evaluation and formulation model used worldwide and through that effort is focused on enhancing the efficiency of nutrient use by ruminants to improve the environmental impact of animal food production. A significant focus of his current work is to understand whole animal and ruminal nitrogen metabolism and amino acid supply and requirements to enhance the development of the Cornell Net Carbohydrate and Protein System. Further, his group is active in developing methods to better describe the interaction between forage and feed chemistry, rumen function and nutrient supply to compliment the model. He has authored and co-authored over 70 journal articles and many conference proceedings and is the recipient of several awards including the American Dairy Science Foundation Scholar Award, the Land O'Lakes Teaching and Mentoring Award from ADSA, the American Feed Ingredient Association Award for Research, the CALS Professor of Merit Award and the CALS Distinguished Advisor Award and in 2016, was named a Stephen H. Weiss Presidential Fellow, the highest teaching award given by Cornell University.

CANC SPEAKERS

Ryan David Leaf – Keynote Speaker - Ryan David is an advocate for those struggling with mental, and behavioral health issues and encourages audiences to transform the way we think about mental health issues and addiction. Ryan works to eliminate the mental health stigma and says, "asking for help is a sign of strength, not weakness." The strong presence Ryan holds in a room is not just due to his 6-foot-6 frame, but the impassioned commitment he has made to serve those who are struggling.

Football was Leaf's life. However, the pressure to perform on such an elite level was more than he was prepared to deal with, ultimately leading to the demise of his professional football career. After his official retirement from the NFL in 2003, Ryan dealt with severe depression and substance abuse. He would serve 32 months in prison for crimes related to that substance abuse. While in prison he found a guardian angel in the form of his roommate and upon his release in Dec 2014 he began to travel down a new path revolving around accountability, spirituality, community, and service to others. Ryan is currently the CEO and President of RAM Consultant, Inc. as well as program ambassador for Transcend Recovery Community. He also works significantly within the sports world as a college football analyst for the PAC 12 network, Sirius XM Radio, & FOX Sports. Ryan also works aggressively for his non-profit, the Focused Intensity Foundation, in which he is the chairman. The foundation raises money for mental health and substance abuse treatment for those who can't afford it.

Mike Van Amburgh, Ph.D. is a Professor in the Department of Animal Science and a Stephen H. Weiss Presidential Fellow at Cornell University where he has a dual appointment in teaching and research. His undergraduate degree is from The Ohio State University and his Ph.D. is from Cornell University. He teaches multiple courses and leads the Cornell Dairy Fellows Program, advises approximately 50 undergraduate students and is the advisor for the Cornell University Dairy Science Club. Mike currently leads the development of the Cornell Net Carbohydrate and Protein System, a nutrition evaluation and formulation model used worldwide and through that effort is focused on enhancing the efficiency of nutrient use by ruminants to improve the environmental impact of animal food production. A significant focus of his current work is to understand whole animal and ruminal nitrogen metabolism and amino acid supply and requirements to enhance the development of the Cornell Net Carbohydrate and Protein System. Further, his group is active in developing methods to better describe the interaction between forage and feed chemistry, rumen function and nutrient supply to compliment the model. He has authored and co-authored over 70 journal articles and many conference proceedings and

is the recipient of several awards including the American Dairy Science Foundation Scholar Award, the Land O'Lakes Teaching and Mentoring Award from ADSA, the American Feed Ingredient Association Award for Research, the CALS Professor of Merit Award and the CALS Distinguished Advisor Award and in 2016, was named a Stephen H. Weiss Presidential Fellow, the highest teaching award given by Cornell University.

Tim A. McAllister, Ph.D. grew up on his parents' cow/calf farm in Innisfail, AB. He obtained a B.Sc.(Agr) and M.Sc. from the University of Alberta in Edmonton, and a Ph.D. (with distinction) in ruminant nutrition and microbiology from the University of Guelph, ON. He accepted an NSERC post-doctoral fellowship at the University of Calgary in 1991, and joined Agriculture and Agri-Food Canada in Lethbridge, AB in 1992. Dr. McAllister has been a research scientist in Rumen Microbiology, Feed and Nutrition since 1997. His research focuses on microbiology, nutrition and beef production and on food and environmental safety issues related to livestock production, strategies for mitigation of *Escherichia coli* 0157:H7, prion inactivation within the environment, and more recently, studies of antimicrobial resistance in bacteria in feedlots. He also has extensive research experience in GHG emissions within animals from manure and the impact of manure handling procedures, such as composting, on emissions. He is the author or co-author of over 550 peer-reviewed scientific papers and 60 reviews, as well as 800 abstracts and conference proceedings, and over 100 final reports for collaborative research projects. Dr. McAllister has been recognized internationally for his leadership role and significant contributions to agricultural research and innovation in the areas of ruminant nutrition/microbiology and molecular biology as they apply to animal health, environmental health and food quality for the benefit of the agricultural industry in Alberta, Canada, and beyond.

Micheal Brouk, Ph.D., professor in the Department of Animal Sciences and Industry at Kansas State University (KSU) has enjoyed career experiences in academia and allied industry. He was raised on a crop farm in central Missouri and obtained BS degrees in Agronomy and Dairy Science at the University of Missouri-Columbia (UMC). He then worked as a research specialist at UMC studying the effects of dairy processing plant solids on forage crops. He then completed a MS degree at UMC in Dairy Science. He was then employed by Land O'Lakes as a livestock production specialist in southwest Minnesota. Later, he completed a PhD program at South Dakota State University (SDSU). He was then hired by SDSU in a teaching and research position. He worked at SDSU for about 2.5 years and then accepted a Commercial Agriculture Extension position at UMC. Following a couple of years at UMC, Dr. Brouk has been employed at KSU for the last 20 years in an extension and teaching position. His area of interests include dairy cattle nutrition, heat abatement and facility design. Dr. Brouk recently completed a sabbatical leave with DeLaval North America examining the application of robotics on large scale dairy operations. In addition to his responsibilities at KSU, he has also served as the co-chairman of the Western Dairy Management Conference for the past 20 years. Mike and his

wife, Michelle, reside in Manhattan, Kansas, where they and their 5 children are active in their church and local community.

Gregory Penner, Ph.D. is an Associate Professor and Centennial Enhancement Chair in Nutritional Physiology. He joined the Department of Animal and Poultry Science at the University of Saskatchewan in 2009 after obtaining his BSA (2004) and M.Sc. (2004) degrees from the same University, and his Ph.D. from the University of Alberta (2009). His research covers forage utilization, ruminant nutrition, and regulation gut function in cattle. He has developed 2 indwelling pH measurement systems that have been adopted by the research community worldwide. Dr. Penner has a well-funded research program supporting and has trained more than 10 graduate students and postdoctoral fellows. He has published over 60 peer-reviewed papers and provided over 40 invited presentations. Greg also has an active extension program helping to communicate research results to end users and serves as the co-chair for the Saskatchewan Beef and Forage Symposium.

Anja Raudabaugh is the Chief Executive Officer of Western United Dairymen. Anja came to WUD from the Madera County Farm Bureau where she served as the Executive Director for four years. Prior to her role at the Farm Bureau, she worked as a Project Manager for a Fortune 500 Company, and as the Senior Legislative Assistant for Congressman Doug Ose, representing California's 3rd District in Washington D.C. Previously the Marketing Director for the California Asparagus Commission and District Representative for State Senator Dick Monteith; her extensive experience in the political environment adds tremendous value to the organization. Anja's time in Sacramento working as a project manager and technical adviser handling CEQA and NEPA requirements as they applied to state and federal agencies, has lent her a skill set adept with navigating complex issues.

Marit Arana, Ph.D. is the head of the Nutrition Department at A. L. Gilbert Company in Oakdale and Keyes, California. Marit received her B.S. degree from California State Polytechnic University, San Luis Obispo, her M.S. degree from California State University, Fresno and her Ph.D. degree from the University of Arizona. Prior to joining A. L. Gilbert in 2003, Marit spent 9 years as the Area Dairy Advisor for the University of California Cooperative Extension Service in the central valley and delta area. Marit served on the CANC steering committee from 2000-2008 and was the Chair in 2007. She has also been active in ARPAS, she served as President of the California Chapter in 2004 after moving through all the other chapter governing council positions. She has also served National ARPAS, as President in 2008-2009 and as the Chair of the National ARPAS examining committee, 2002-2007. Marit has also served as President of the American College of Animal Sciences, 2014-2015, and Nutrition Discipline Chair 2009-13. Marit is also serving as the Vice-Chair on the Feed Inspection Advisory Board of the California Department of Food and Agriculture. Marit is also a member of the American Dairy Science Association; Equine Science Society;

American Society of Animal Science; National Mastitis Council; California Women for Agriculture; and American Agri Women.

Corwin Holtz is President of and an active consultant in Holtz Nelson Dairy Consultants, LLC. – a group of nine independent dairy nutrition and management consultants working with dairy producers in New York, Pennsylvania, Ohio and the New England states. Corwin formed this group in early 2004. The groups focus is on maximizing cow health and productivity through the use of client grown forages and grains and supplying management consultation over a wide variety of daily management topics that impact farm profitability, land resource use and environmental stewardship. Corwin grew up in the California dairy industry and obtained his degree in Dairy Science from Cal Poly, San Luis Obispo in 1978. This was followed by two years in the A.I. industry and then working as a farm manager for a 350 cow dairy in California. He then left for academia – obtaining a Masters degree in Ruminant Nutrition and Reproduction at Cornell University. Corwin was then a faculty member at Cal Poly for 1.5 years and then for 5 years in the Dairy Management teaching and extension program at Cornell University. In 1994 he entered the commercial feed business and held a variety of technical support and research positions with several cooperative and privately held commercial and specialty feed ingredient companies. From 2000-2002 he was a co-project and farm manager for the building and development of a 1,000 cow commercial/research dairy facility in central New York for DeLaval. In addition to his work activities Corwin is a member of ADSA, PAS and is a very active board member of the Northeast Agribusiness and Feed Alliance – a 300+ member organization involved in agricultural education and lobbying activities at the state, regional and national levels. He also serves on the scientific advisory boards of three national/international feed ingredient companies. Corwin resides in the central New York town of Dryden with his wife Debby and has the good fortune of having their daughter, son-in-law and two grandchildren living in the same town.

Rick Lundquist, Ph.D. is the owner of Lundquist & Associates, LLC and a partner in Nutrition Professionals Inc. Rick received his Ph.D. in Dairy Nutrition from the University of Minnesota in 1984 and began his career as a private consultant to the dairy industry in Arizona that same year. Rick now consults throughout the United States and internationally and has extensive experience in sub-tropical dairy feeding and management. Rick and his seven partners work with over 400 dairy producers and over 300,000 milking cows. In addition, Rick was a regular contributor to *Farm Journal's Dairy Today* magazine for over 20 years and currently writes a nutrition column for *Dairy Herd Management Magazine.* Rick is author of several scientific publications in *The Journal of Dairy Science.* Rick is a professional member of the American Dairy Science Association and is a Board Certified Diplomat in the American College of Animal Nutrition, American Registry of Professional Animal Scientists. Rick is also a member of Gamma Sigma Delta, The Honor Society of Agriculture.

California Animal Nutrition Conference
2019 Steering Committee

Chairperson: David Ledgerwood, M.S., P.A.S. graduated in 2004 with a BS degree in Animal Science focusing on livestock and dairy cattle from the University of California, Davis. Upon graduating he worked in the university ruminant nutrition lab with Dr. Ed DePeters as a lab technician performing feed nutrient and milk analysis while assisting graduate students run various ruminant nutrition focused research trials. In 2007 he graduated with a MS degree in Animal Biology focusing on ruminant nutrition working with Dr. DePeters. After graduation in 2007 he worked as a lab and research program manager in the field of animal behavior/welfare on the UC Davis campus performing various research trials focused on improving cow comfort. He accepted a job with the Veterinary Medicine Teaching and Research Center in Tulare as a research program manager for the clinical department performing various research projects covering cow behavior, calf health, and nutrition. In April of 2011 he was offered a position with Western Milling LLC working as a dairy nutritionist. In 2018 David joined Novita Nutrition as a Technical Service Manager. In the Spring of 2019 David began working with Chr-Hansen.

Vice Chairperson: Jennifer Heguy, M.S., P.A.S. – UCCE Farm Advisor
Jennifer Heguy is a native of California's San Joaquin Valley. She received her B.S. in Animal Science, with an emphasis in Livestock and Dairy, at the University of California, Davis. In 2006, she received her M.S. degree at UC Davis, focusing on dairy cattle nutrition. Jennifer currently serves as the University of California Dairy Farm Advisor in Merced, Stanislaus and San Joaquin Counties where milk is a major agricultural commodity. Jennifer's major program focus is improving silage and feeding management practices on California dairies.

Ex Officio: Jason Brixey, M.S., P.A.S. - Consulting Animal Nutritionist
Jason D. Brixey was born and raised in Crescent City, CA. He graduated in 2001 from Cal Poly San Luis Obispo with a Dairy Science and Ag Business Undergraduate Degree. He attended the University of Idaho in Moscow under Dr. Mark McGuire, graduated with a Masters of Science in 2003. He started his animal nutrition career with Westside Milling LLC in Phoenix, AZ as the Nutrition Technical Representative. In April of 2005, Jason was hired by Pine Creek Nutrition Service Inc. out of Denair, CA to work on dairy farms as a Consulting Animal Nutritionist. He became partner at Pine Creek Nutrition Service in January of 2008. Married to my lovely wife Jodie that I share in the duties of raising three wonderful children; Jack (10), Kaydee (8), and Will (5).

Committee Members:

Ruben Almada, B.A.Sc., Kemin Animal Nutrition & Health, was born and raised in the Hilmar, California area. Growing up on a dairy steered him toward a life in the dairy world. He graduated from California Polytechnic State University – San Luis Obispo in 2006 with a Dairy Science Degree. Upon completing his degree Ruben joined Cargill Animal Nutrition in the fall of 2006 where he was a Dairy Management Consultant for 3 years. He then joined Kemin Animal Health and Nutrition in July of 2010 as Key Account Manager covering California and Arizona for the Dairy Segment. He is married to his wonderful wife, Jennifer, and they have two children, Kinley (5) and Jaxson (3).

Tyler Harris, Ph.D., Kemin Animal Nutrition & Health, grew up in South Texas on a small farm and beef operation. He graduated from Texas Tech University with a Bachelor's degree in Animal Science in 2010, and obtained his Masters of Science in Muscle Biology in 2013. It was during his doctorate work that he was introduced to the dairy industry, and he completed his Ph.D. in 2016 with a degree in nutritional immunology. Since finishing his graduate work, Tyler has lived in California. He currently works as a Technical Service Manager for Kemin Industries.

Juliana M. Huzzey, Ph.D. is an Assistant Professor, Behavior and Welfare Specialist in the Animal Science Department at Cal Poly, San Luis Obispo. She received her Ph.D. Animal Science from Cornell University Ithaca, NY and M.S. Animal Science and B.S. Agroecology from University of British Columbia, Vancouver, BC, Canada. The overall aim of Julie Huzzey's program is to enhance the welfare of animals in managed systems by teaching classes and conducting research focused in applied animal behavior and welfare. She is particularly interested in cattle feeding and social behavior and the role these behaviors play in biological functioning and performance. By understanding these processes better, Dr. Huzzey aims to identify optimal housing and management practices that can be directly applied on farms to facilitate improved animal health and wellbeing. Dr. Huzzey is a member of the American Association of Dairy Science, the American Society of Animal Science and the International Society of Applied Ethology.

Jamie Jarrett, Ph.D., grew up in Bakersfield, California on a farm surrounded by pigs, chickens, horses, cattle and basically any animal she could convince her dad to

let her have. That passion for animals lead her to Cal Poly, where she edarned a degree in Animal Science with a pre-veterinary concentration. While there, she developed a deep interest in research through her senior project, which led her to graduate school at Oklahoma State where she worked in a lob that concentrated on low-nutrient excretion experiments and antibiotic alternatives. She would eventually complete her Ph.D. in 2011 at Virginia Polytechnic Institute and State University, where she completed her thesis: *"Effects of exogenous phytase and forage particle length on site and extent of phytate digestion in lactating cows."* Jamie joined Alpha Dairy Consulting in 2017 as a nutrition consultant.

Zachery Meyer, was raised in Ixonia, Wisconsin. He grew up immersed in his family's business, Rock River Laboratory. Meyer spent many hours helping in various jobs around the laboratory, seeing first-hand the dedication and commitment his father and the late Twilah Kulow had to the business and their customers. Meyer gathered business experience at Clear Channel and GE Medical while working toward his degree from the University of Wisconsin-Milwaukee. In 2007, Meyer resumed his involvement in Rock River Laboratory, starting as a soil sampler, moving to outside sales and eventually taking on his current role of director of operations.
Meyer still gathers inspiration from the Rock River Laboratory employees and mentors who cultivated his drive for customer satisfaction and service, while continuing to learn and deepen his understanding of animal nutrition and agronomy. When he isn't building relationships with customers or overseeing laboratory operations, Zac spends his time playing or watching sports and sharing in family time with his wife and two young daughters.

Honorary Members:

Amanda Gipe McKeith, Ph.D. is an Assistant Professor in the Department of Animal Sciences & Agricultural Education at Fresno State. She serves as the graduate program coordinator and co-advisor for both the Meat Science Club and Young Cattlemen's Association. Her research focus areas are how nutrition affects meat quality, processed meat ingredients, and food safety in food products. Dr. Amanda Gipe McKeith is originally from Merced, CA where she grew up on a 700-acre farm/ranch. Her family raises purebred registered Shorthorn cattle and grows corn silage, alfalfa, oats, and pasture hay. Amanda grew up showing livestock with her sister, Amy, and parents, Alpha and Sherri. Her family still runs the operation that now includes her brother-in-law, Tim and nephew, TJ as well as her husband Russell McKeith. Amanda serves as President for the National Shorthorn Lassie Association, and Treasurer for the California Shorthorn Breeders Association. Amanda received her A.A. in General Agriculture from Merced Community College, B.S. in Animal Science and Food Science at Kansas State University, M.S. in Animal Science with an emphasis in Meat Science from Kansas State University, and Ph.D. in Animal Science with an emphasis in Meat Science from The Pennsylvania State University. Amanda judged on both the Meats and Livestock Judging teams at Kansas State University.

She coached meats judging teams at Kansas State and Western Kentucky University and livestock judging teams at Penn State and Western Kentucky.

Kyle Thompson, Ph.D. received his B.S. degree in animal science from Fresno State (2006) and his master's and Ph.D. degrees in animal science from Oklahoma State (2011/2015). He joined the Fresno State staff in the fall of 2016 after taking classes and teaching at Oklahoma State from January 2007-June 2016 and serving as the graduate student assistant manager of the campus dairy cattle center. His research included dairy nutrition research trials and lactating cow probiotics. He also assisted in research for bovine respiratory disease, rumen temperature bolus, milk production by weigh-suckle-weigh and swine antimicrobial replacements. He also assisted in 4-H and FFA Field Day dairy judging competitions. While in Stillwater, OK, he owned and operated Wild Acre Farms and Exotics, which raised ewes, game birds, free range hens and other fowl/animals, and produced grasses and winter wheat for grazing and hay production. As a Fresno State student, he worked in the sheep unit three years, served as a campus farm tour guide, and dairy unit herdsman and feed/hospital technician. He also worked as an exotic animal nutrition intern (2009) and global nutrition fellow at the San Diego Zoo (2013).

CALIFORNIA ANIMAL NUTRITION CONFERENCE HISTORY

YEAR	CHAIRPERSON	COMPANY AFFILIATION
2018	Jason Brixey, M.S., P.A.S.	Pine Creek Nutrition Service
2017	Dr. Phillip Jardon, DVM, MPVM	Elanco Animal Health
2016	Dr. Phillip Jardon, DVM, MPVM	Elanco Animal Health
2015	Mr. Ben Tarr	Adisseo USA Inc.
2014	Dr. Jeffrey M. DeFrain	Zinpro Performance Minerals
2013	Mr. Doug DeGroff	Diversified Dairy Solutions, LLC
2012	Mr. Eduardo Galo	Novus International, Inc.
2011	Dr. Michael A. DeGroot	DeGroot Dairy Consulting
2010	Dr. Jim Tully	Pine Creek Nutrition Service, Inc.
2009	Mr. Michael Braun	Phibro Animal Health
2008	Dr. Luis Rodriguez	Zinpro Corporation
2007	Dr. Marit Arana	A.L. Gilbert Company
2006	Mr. Dennis Ervin P.A.S.	Prince Agri Products, Inc.
2005	Dr. Lawson Spicer	Nutri Management Inc.
2004	Dr. Luis Solorzano	Purina Mills, Inc.
2003	Dr. Alfonso Mireles, Jr.	Foster Farms
2002	Mr. Edmund Vieira	Pine Creek Nutrition Service, Inc.
2001	Dr. Melinda Burrill	California State Polytechnic University - Pomona
2000	Mr. Dave Fischer	Foster Farms
1999	Dr. M. Steven Daugherty	California State Polytechnic University - SLO
1998	Dr. Doug Dildey	Alltech, Inc.
1997	Ms. Carla Price	Nutritionist
1996	Dr. H.John Kuhl, Jr.	Nest Egg Nutrition
1995	Mr. Dennis Ralston	M. Rinus Boer Co., Inc.
1994	Dr. Doug Dildey	Alltech, Inc.
1993	Dr. Mark Aseltine	Consulting Animal Nutritionist
1992	Dr. Carl Old	MacGowan-Smith Ltd.
1991	Mr. Nick Ohanesian	Ohanesian & Associates
1990	Mr. Rod Johnson	M. Rinus Boer Co., Inc.
1989	Mr. Timothy Riordan	Nutri-Systems, Inc.
1988	Dr. Russ W. Van Hellen	Great West Analytical
1987	Dr. John E. Trei	California State Polytechnic University, Pomona
1986	Dr. A.A. Jimenez	Ancon, Inc.
1985	Dr. Wm. A. Dudley-Cash	Foster Farms
1984	Dr. Joel Kemper	Penny-Newman Co.
1983	Dr. Alex J. Kutches	O.H. Kruse Grain & Milling Co.
1982	Dr. Howard Waterhouse	Bell Grain & Milling
1981	Mr. Don Ulrich	Diamond Shamrock Chemical Co.
1980	Mr. Tom Geary	PMS-West, Inc.
1979	Dr. Frank Parks	Kemlin Industries
1978	Mr. Fred Pfaff	Zacky Farms
1977	Mr. Rene Lastreto	Diamond Shamrock Chemical Co.
1976	Mr. Rene Lastreto	Diamond Shamrock Chemical Co.
1975	Dr. R.D. Hendershott	Nulaid Foods
1974	Dr. R.D. Hendershott	Nulaid Foods

CALIFORNIA ANIMAL NUTRITION CONFERENCE HISTORY- Continued

YEAR	CHAIRPERSON	COMPANY AFFILIATION
1973	Dr. Leland Larsen	Nutri-Systems, Inc.
1972	Dr. Leland Larsen	Nutri-Systems, Inc.
1971	Mr. Rene Lastreto	Diamond Shamrock Chemical Co.
1970	Mr. Fred Pfaff	Balfour Guthrie
1969	Mr. Fred Pfaff	Balfour Guthrie
1968	Mr. Fred Pfaff	Balfour Guthrie
1967*	Mr. Gary L. Frame	J.G. Boswell Co.
1966*	Mr. Gary L. Frame	J.G. Boswell Co.
1965*	Mr. Arne Jalonen	Topper Feed Mills
1964*	Mr. Arne Jalonen	Topper Feed Mills
1963*	Dr. W.P. Lehrer	Albers Milling Co.
1962*	Dr. H.J. Almquist	The Grange Co.
1961*	Dr. H.S. Wilgus	The Ray Ewing Co.
1960*	Mr. Bert Maxwell	Nulaid Foods
1959*	Mr. Bert Maxwell	Nulaid Foods
1958*	Mr. Robert Caldwell	Anderson Smith Milling Co.
1957*	Mr. Emery Johnson	P.C.A., Los Angeles
1956*	Mr. Emery Johnson	P.C.A., Los Angeles
1955*	Dr. H.J. Almquist	The Grange Co.
1954*	Dr. H.J. Almquist	The Grange Co.
1953*	Mr. Clifford Capps	California Milling Co.
1951*	Mr. Dolph Hill	Golden Eagle Milling Co.
1950*	Dr. H.J. Almquist	The Grange Co.
1949*	Dr. H.J. Almquist	The Grange Co.
1948*	Dr. H.J. Almquist	The Grange Co.

* California Animal Industry Conference

History of the California Animal Nutrition Conference

The California Animal Nutrition Conference (CANC) originated in the 1940's as the California Animal Industry Conference, sponsored by the California Grain & Feed Association (CGFA). CGFA wanted to expand the continuing education program into a forum encompassing animal health, nutrition and management. The expectations were that communications between (nutritionists) industry, educational institutions and regulatory agencies would be improved. In 1972, CGFA discontinued sponsoring the Animal Industry Conference.

After the conference was discontinued, a small group of nutritionists began meeting annually in Fresno. Two or three invited speakers from industry or the universities presented information on nutrition, especially poultry.

In 1975 a set of organizational bylaws were developed by the steering committee. CANC was established and was provided support by CGFA. The CGFA Board of Directors appointed a chairperson annually and approved the steering committee. In 1978, Dr. Frank Parks, the Chairperson, requested that CANC be granted independent status and be established as a self-governing committee of CGFA. This request was granted.

For a few years, meetings were held in Fresno and Corona, California. For a couple of years starting in 1978, CANC published "Nutri-Facts", a "newsletter" consisting of articles in animal production.

In 1979, donations were requested from industry companies to help keep registration fees low. During the 1980's and through the 1990's the attendance at CANC continued to grow as the quality of the conference improved and the conference became known nationwide. In the 1990's a pre-symposium was added. The pre-symposium is sponsored by a company selected by the CANC Steering Committee. This process allows the selected company to showcase its research and products. In the year 2000, posters on research by students were included.

Attendance at the conference has grown from 50 in the 1970's to over 300 attendees. To encourage attendance, different activities have been tried such as keynote speakers, skiing expeditions and a very successful barbeque dinner put on by the Animal Science Department at Fresno State University.

The California Grain & Feed Association has supported and allowed CANC to work and grow. The premise of the CGFA and CANC relationship is to work together to educate the feed industry with information for problem solving and to disseminate valuable research information. CANC is not an industry, university, or government entity, but a committee collectively working together for the good of agriculture in California.

STUDENT ABSTRACTS FOR

POSTER PRESENTATION

AT THE

CALIFORNIA ANIMAL

NUTRITION CONFERENCE

MAY 8 & 9, 2019

Determining the effects of commercially available direct fed microbial product on overall growth, rumen development and health parameters in neonatal dairy calves

B. Britton,* M. Fringer,* A. McKeith,* S. Williamson,* and K. Thompson*

*Department of Animal Sciences and Agricultural Education, California State University, Fresno, Fresno, CA 93740

I. Abstract

As the concern for antibiotic resistant bacteria continues to grow, the animal industry is continually searching for new methods or products that will improve animal health while being a supplement or replacement for antibiotics. The addition of direct-fed microbial (DFM) supplements aid in establishing a beneficial microbial population in the rumen and gastrointestinal tract of the calf and therefore increase feed intake and feed efficiency of the animal. However, very little research has been done on colostrum-deprived calves, an area within the industry that has vast room for improvement. With up to fifty percent morbidity and mortality rate, it remains unclear if the health benefits of DFM's will overcome the calves insufficient immune system. A *lactobacillus* fermented product was tested on two different subpopulations; sixty colostrum-deprived Jersey calves and ten Holstein calves that received colostrum. Each treatment calf received 2.5 mL of *lactobacillus* fermented product for calves and were randomly assigned to be submitted for necropsy starting at 30 days of age. Individual weights were recorded for kidneys, liver, lungs, spleen and heart. Two sections from the duodenum, ilium and jejunum were collected and stored in 10% formaldehyde for histological examination of brush-border health. Additionally, individual fecal scores and incidence of diarrhea as a percent of overall life-time percentage and antibiotic treatments were recorded daily thus inferring general health of the calf. Biometric parameters such as average daily gain and feed conversion rate were also calculated. The addition of the DFM to the colostrum-deprived population did not reduce the average morbidity and mortality rate thus indicating that an increased population of beneficial bacteria cannot overcome the lack of immunoglobulins the calf would have received from colostrum or colostrum replacer. Treatment did not have a significant effect on body weight (treatment 57.95 ± 14.15 kg versus control 52.31 ± 14.23 kg), average daily gain (treatment 0.280 ± 0.297 kg versus control 0.271 ± 0.347 kg) or fecal scores (treatment 0.327% ± 0.347 versus control 0.452% ± 0.265

life with diarrhea). There was very little difference of ruminal development between treatment and control calves (treatment 0.0101% ± 0.00268 BW versus control 0.0226% ± 0.0402 BW). Intestinal health showed no apparent difference between treatment and control (treatment 2.491 ± 0.984 versus control 2.293 ± 1.121). There was a one-hundred percent survivability rate of the treatment group within the Holstein population thus indicating a positive enhancement on healthy calves. Weight, average daily gain, feed efficiency and fecal scores seem to be unaffected. There was seemingly no difference in intestinal parameters.

Due to the high morbidity and mortality rate of colostrum deprived calves, more research is needed to determine the specific mode of action in order to improve efficiency of its use in dairy calves and cattle.

Impact of replacing canola meal with solvent extracted distillers grain with solubles as a protein source on milk production

R. J. Edwards*[1], D.N. Ledgerwood[2], D.N. Waldner[3], H.A. Rossow[1]; [1]University of California, Davis, Tulare CA USA, [2]Novita, Brookings SD USA, [3]Valley Nutrition, Visalia, CA USA

Due to the high unsaturated fatty acid content of dried distillers grains with solubles (DDGS), usage of DDGS as a protein supplement for dairy cows is limited. However, new extraction processes can remove corn oil from distillers grains and produce solvent extracted distillers grain with solubles (NM; NovaMeal, Brookings, SD). The objective of this study was to evaluate the impact of replacing canola meal (CM) with NM as a protein source in lactating dairy cattle TMR on milk yield and components. A total of 2,817 Holstein cows averaging 63 ± 46 DIM were enrolled to one of four dietary treatment groups: 1) primiparous control group (PC; 12.98% CM DM basis); 2) primiparous treatment group (PT; 8.8% NM, 4.2% CM DM basis); 3) multiparous control group (MC; 12.76% CM DM basis); and 4) multiparous treatment group (MT; 8.6% NM, 4.1% CM DM basis). Primiparous control TMR contained 3.1 kg (DM basis) of CM and PT TMR replaced CM with 2.08 kg (DM basis) of NM. Multiparous control TMR contained 3.47 kg (DM basis) of CM and treatment TMR replaced CM with 2.35 kg (DM basis) of NM. The TMR were formulated to be isonitrogenous and isoenergetic with similar fatty acid profiles. All TMR averaged 17% CP, 5.5% fat and contained 45% forage and 55% concentrate. The TMR were fed for 6 wk and then switched between control and treated pens. After a 2 wk adjustment period, data collection resumed. Primiparous and multiparous cows DMI were different ($P < 0.05$; 23.2 and 29.7 kg/d, respectively), however DMI did not differ by treatment. Primiparous and multiparous cows were different for least square mean (LSM) milk yield ($P < 0.05$, 34 kg, 49 kg, respectively), LSM milk protein percentage ($P < 0.05$, 3.92 %, 2.64 %, respectively), LSM milk fat yield ($P < 0.05$, 1.99 kg, 2.13 kg, respectively) and LSM milk fat percentage ($P < 0.05$, 5.85 %, 4.74 %, respectively) but not different by treatment. The LSM milk protein yield was not different between primiparous and multiparous cows or by treatment groups. These results show that NM with supplemented urea can be used to replace CM as a protein supplement in a lactating dairy cattle TMR.

KEYWORDS
Solvent extracted distillers grain with solubles
Canola Meal
Bypass protein

Effects of *Bacillus subtilis* probiotics on growth performance, diarrhea, and intestinal health of weaned pigs experimentally infected with an enterotoxigenic *Escherichia coli*

Yijie He,[1] Kwangwook Kim,[1] Lauren Kovanda,[1] Cynthia Jinno,[1] Minho Song,[2] Xunde Li,[3] and Yanhong Liu[1]

[1]Department of Animal Science, University of California, Davis, CA, [2]Department of Animal Science and Biotechnology, Chungnam National University, Daejeon, South Korea, [3]School of Veterinary Medicine, University of California, Davis, CA

The objective of this experiment was to investigate the effects of dietary supplementation of *Bacillus subtilis* probiotics on growth performance, diarrhea, and intestinal health of weaned pigs experimentally infected with an enterotoxigenic *Escherichia coli* (***E. coli***). Weaned pigs (n = 48, 6.17 ± 0.36 kg BW) were individually housed in disease containment rooms and randomly allotted to one of four dietary treatments: negative control (**NC**, a corn-soybean meal control diet without *E. coli* challenge), positive control (**PC**, control diet with *E. coli* challenge), and two *E.coli* challenged treatment groups with either 50 mg/kg supplementation of carbadox (**AGP**) or 500 mg/kg supplementation of *Bacillus subtilis* probiotics (**PRO**). The experiment was conducted over 28 days with 7 days before and 21 days after the first *E. coli* inoculation. After 7 d of adaptation, pigs in *E. coli* challenged groups were orally inoculated with an F18 *E. coli* inoculum. The F18 *E. coli* expressed LT, STb, and SLT-2 toxins and were provided at 10^{10} CFU/3 mL dose for three consecutive days. Average daily gain (**ADG**), average daily feed intake (**ADFI**) and Gain:Feed ratio were calculated for d -7 to 0 before *E. coli* inoculation, d 0 to 7, d 7 to 14, and d 14 to 21 post-inoculation (**PI**). Diarrhea score (1, normal, to 5, watery diarrhea) was recorded daily for each pig. Fecal samples were collected on d 0, 3, 7, 14, and 21 PI to analyze for β-hemolytic coliforms. At the end the study at d 21 PI, pigs were euthanized and jejunal and ileal mucosa were collected to analyze tight junction protein mRNA expression and inflammatory mediator mRNA expression. Data were analyzed using the Mixed Procedure of SAS with repeated measure. Pigs in PRO group had greater ($P < 0.05$) body weight on d 21 PI, compared with the pigs in PC group. Pigs in AGP group had greater ($P < 0.05$) body weight on d 7, 14, and 21 PI than pigs in the PC and PRO group. Pigs in both AGP and PRO group had improved ($P < 0.05$) ADG and feed efficiency from d 0 to 21 PI. Pigs in AGP and PRO groups had reduced

($P < 0.05$) frequency of diarrhea (score ≥ 4) throughout the experiment and had decreased ($P < 0.05$) fecal β-hemolytic coliforms on d 7 PI compared with pigs in the PC. Pigs in PRO had increased ($P < 0.05$) claudin mRNA expression in jejunal mucosa and decreased ($P < 0.05$) COX2 and IL6 mRNA expression in ileal mucosa, compared with pigs in PC. In conclusion, results of the current experiment indicate that supplementation of *Bacillus subtilis* probiotics enhanced disease resistance and reduced intestinal inflammation of weaned pigs infected with F18 *E. coli*. Both *Bacillus subtilis* and antibiotics improved growth performance of *E. coli* challenged pigs compared with pigs in the positive control.

Key words: *Bacillus subtilis* probiotics, diarrhea, growth performance, intestine health, weaned pigs

Enzymatically digested food waste altered the fecal microbiota of growing-finishing pigs

Cynthia N. Jinno, Perot Saelao, Elizabeth Maga, and Yanhong Liu
Department of Animal Science, University of California, Davis, 95616

Enzymatic digestion converted food waste (bakery, fruits, vegetables, and meats) from supermarkets into pasteurized liquid pig feed. This study was conducted to observe the fecal microbiota of growing-finishing pigs fed with enzymatically digested food waste. Fifty-six crossbred pigs (approximately 32.99 kg BW) were randomly assigned to one of the 2 dietary treatments with 7 replication pens and 4 pigs per pen. A 3-phase feeding program was used with d 0 to 28 as Phase 1, d 28 to 53 as Phase 2, and d 53 to 79 as Phase 3. The 2 dietary treatments were: a corn-soybean meal control diet, and a liquid diet produced from enzymatically digested food waste. All diets met the estimates for nutrient requirements of growing-finishing pigs based on the NRC (2012). Pigs were fed control or liquid diet in phases 1 and 2, while all pigs were fed with control diet in phase 3. Fecal samples were collected on d 0, 28, 53, and 79 from the same pig per pen and fecal microbiota was analyzed using 16S rRNA gene sequencing at the V4 hypervariable region and compositional data was analyzed using QIIME2 (2018.6). Alpha diversity and relative abundance were analyzed with PROC MIXED of SAS with pen as experimental unit and the statistical model included diet as fixed effect and block as random effect. Beta diversity were analyzed using packages in the R program (ver. 3.5.2). Observed species and Shannon diversity indexes were similar in both treatment groups on d 0, but pigs fed with food waste had greater ($P < 0.05$) Shannon diversity than control pigs on d 28, 53, and 79. Feeding food waste tended ($P < 0.10$) to decrease the relative abundance of *Firmicutes* compared with pigs fed control diet. Within this phylum, the abundance of

Lachnospiraceae and *Ruminococcaceae* was increased ($P < 0.05$) and the abundance of *Streptococcaceae* and *Clostridiaceae* was decreased ($P < 0.05$). The analysis of Bray Curtis PCoA displayed a separate cluster between food waste and control groups on d 28 and 53. Results of this experiment indicate that feeding enzymatically digested food waste alters microbial diversity within the gastrointestinal tract of growing-finishing pigs. The likely reason for this change was the different nutrient components in food waste diet versus corn and soybean meal diet.

Effects of antibiotics on growth performance, diarrhea, bacterial translocation, and blood profiles in weanling pigs experimentally infected with a pathogenic *E. coli*

Kwangwook Kim[1], Yijie He[1], Cynthia Jinno[1], Seijoo Yang[1], Minho Song[2], Peng Ji[1], Yanhong Liu[1]

[1]University of California, Davis, CA, [2]Chungnam National University, Daejeon, Republic of Korea

The experiment was conducted to investigate the effects of antibiotics on growth performance, diarrhea, fecal β-hemolytic coliforms, bacterial translocation, and blood parameters of weanling pigs experimentally infected with F18 *Escherichia coli* (***E. coli***). Thirty-four pigs (6.88 ± 1.03 kg BW) were individually housed in disease containment rooms and randomly allotted to one of three treatments with 9-13 replicate pigs per treatment. The three dietary treatments were control diet and 2 additional diets supplemented with 0.5 or 50 mg/kg carbadox, respectively. The experiment lasted 18 d [7 d before and 11 d after first inoculation (d 0)]. The F18 *E. coli* inoculum was orally provided to all pigs with the dose of 10^{10} cfu/3 mL for 3 consecutive days. Growth performance was measured on d -7 to 0 before inoculation, and d 0 to 5 and 5 to 11 post-inoculation (**PI**). Diarrhea scores were daily checked throughout the experiment. Fecal samples were collected on d 0 before inoculation, and d 2, 5, 8, and 11 PI to test the percentage of β-hemolytic coliforms in total coliforms. Blood samples were collected on d 0 before *E. coli* inoculation and on d 2, 5, 8, and 11 PI. Total and differential blood cell count were analyzed by CBC test. 17 pigs were euthanized on d 5 PI, whereas the remained pigs were euthanized at the end of the experiment to collect mesenteric lymph node to analyze bacterial translocation. All data were analyzed by ANOVA using PROC MIXED of SAS. Pigs supplemented with high-dose of antibiotics had greater ($P < 0.05$) final BW and lower ($P < 0.05$) overall frequency of diarrhea, compared with pigs in control and low-dose antibiotics groups. Pigs supplemented with low-dose antibiotics had lowest ($P < 0.05$) ADG and feed efficiency from d 0 to 5 PI, and had highest ($P < 0.05$) percentage of β-hemolytic coliforms in fecal samples on d 2 and 5 PI, and greatest ($P < 0.05$) bacterial colonies in mesenteric lymph nodes on d 11 PI, compared with pigs in the other two groups. Supplementation of low-dose antibiotics had greatest ($P < 0.05$) neutrophils but lowest ($P < 0.05$) monocytes on d 2 PI, compared with control and high-dose antibiotics groups. Pigs in the low-dose antibiotics group still had higher ($P < 0.05$) white blood cell counts and lymphocytes than

pigs in the other groups on d 11 PI. In consistent with CBC results, pigs supplemented with low-dose antibiotics had greatest ($P < 0.05$) serum C-reactive protein on d 2 and 5 PI and serum TNF-α on d 5 PI, compared with pigs in the control and high-dose antibiotics groups. No differences were observed in the red blood cell profiles between pigs in control and low dose antibiotics groups, whereas supplementation of high-dose antibiotics had lowest ($P < 0.05$) packed cell volume but highest ($P < 0.05$) mean corpuscular hemoglobin concentration among three treatments. In conclusion, low-dose antibiotic supplementation may exacerbate the detrimental effects of *E. coli* infection on pig performance, and increase diarrhea and systemic inflammation of weanling pigs.

Key words: blood profile, carbadox, diarrhea, growth performance, pathogenic *E. coli*, weanling pigs

Dietary spray dried plasma on systemic immune responses of lactating sows and their litters

Sheena Kim1, Byeonghyeon Kim1, Junsu Kim1, Kwangwook Kim2, Jeong Jae Lee1, Joowon Kang1, Daye Mun1, Jangryeol Baek1, Soyun Kim1, Yanhong Liu2, Jeehwan Choe1, and Minho Song1
1Division of Animal and Dairy Science, Chungnam National University, Daejeon, Republic of Korea, 2Department of Animal Science, University of California, Davis, CA, USA

The objective of this experiment was to investigate the effects of spray dried plasma (**SDP**) in last gestating and lactating diets on systemic immune responses of lactating sows and their litters. A total of 12 last gestating sows (227.75 ± 7.50 kg BW; parity = 2.0 ± 0.7) were randomly assigned to 2 dietary treatments in a completely randomized design. The dietary treatments were a typical last gestating and lactating diet based on corn and soybean meal (**CON**) and CON supplemented with 1% of SDP (**SDP**). Sows were fed experimental diets from d 30 before expected farrowing to weaning. Weaned pigs from each sow were transferred to a nursery barn and group-housed by dietary treatments of sows and fed a typical nursery diet for 6 wk. Blood Samples were collected from sows on d 0, 3, and 7 after farrowing and randomly selected 2 piglets in each sow on d 3 and 7 after birth, weaning day, d 3, and 7 postweaning. Serum tumor necrosis factor-α (**TNF-α**), transforming growth factor-β (**TGF-β**), C-reactive protein (**CRP**), cortisol, and immunoglobulin (**Ig**)G, M, and A from their litters were analyzed by the enzymelinked immunosorbent assay. Data were analyzed using the PROC GLM of SAS. Sows fed SDP tended ($P < 0.10$) to have lower serum TNF-α on d 3 (264.94 vs. 281.96 pg/ml) and d 7 (249.35 vs. 272.15 pg/ml) than sows fed CON. Moreover, SDP tended ($P < 0.10$) to decrease of serum TGF-β (311.37 vs. 448.07 pg/ml) and cortisol (0.47 vs. 0.55 ng/ml) on d 3 compared with CON. The litters from sows fed SDP tended ($P < 0.10$) to reduce serum TNF-α (349.87 vs. 423.57 pg/ml), TGF-β (853.49 vs. 980.41 pg/ml), and cortisol (0.62 vs. 1.05 ng/ml) on d 7 than litters from sows fed CON. Supplementation of SDP tended ($P < 0.10$) to decrease serum TNF-α on d 3 (344.11 vs. 449.80 pg/ml) and CRP on d 7 (78.41 vs. 112.28 ng/ml) compared with the piglets from lactating sows fed CON. The addition of SDP also reduced ($P < 0.05$) serum cortisol on d 3 (1.40 vs. 1.88 ng/ml) and TGF-β on d 7 (718.33 vs. 836.48 pg/ml) compared with CON. However, there were no differences on CRP, IgG, IgM, and IgA of sows and their offspring between CON and SDP. In conclusion, supplementation of dietary spray dried plasma in last gestating and lactating diets may modulate systemic immune responses of sows and their litters.

Key words: Immune responses, Lactating sows, Spray dried plasma, Weaned pigs

Susceptibility of several species of gram-negative and gram-positive bacteria to organic acids and their derivatives

Lauren Kovanda[1], Xunde Li[1], Wen Zhang[1], Xiaohong Wei[1], Jia Luo[1], Xixi Wu[1], Edward Robert Atwill[1], Richard Sygall[2], Jacob Lum[2], and Yanhong Liu[1]
[1]University of California, Davis, 95616, and [2]Perstorp Feed and Food, Sweden

The objective of this experiment was to determine the *in vitro* antimicrobial activity of several organic acids and their derivatives against gram-positive (G+) and gram-negative (G-) bacteria. Monopropionin, monovalerin, monolaurin, and sodium formate were tested at 10 to 12 concentrations from 0.001 to 2.50 mg/L; monobutyrin was tested at 16 concentrations from 0.001 to 5.00 mg/L; butyric acid and valeric acid were tested at 8 concentrations from 0.001 to 0.35 mg/L; while formic acid was tested at 8 concentrations from 0.001 to 1.00 mg/L. The tested bacteria included G- bacteria (*Escherichia coli, Salmonella* Typhimurium, *and Campylobacter jejuni*) and G+ bacteria (*Enterococcus faecalis, Clostridia perfringens, Streptococcus pneumonia*, and *Streptococcus suis*). Antimicrobial activity of tested compounds was expressed as minimum inhibitory concentration (MIC) that prevented growth of tested bacteria in treated culture broth. Butyric acid, valeric acid, and formic acid at highest tested concentrations inhibited the growth of all bacterial strains. The MICs of the three acids varied among bacterial strains with lowest MIC of 0.05-0.10 mg/L on two strains of *Campylobacter*. Sodium formate at highest tested concentrations did not inhibit the growth of *E. coli*, *S.* Typhimurium, and *E. faecalis* but the compound inhibited the growth of other tested bacteria with MIC values from 0.20 to 1.88 mg/L. The MIC values of monovalerin, monolaurin, and monobutyrin ranged between 0.001 and 1.50 mg/L on all bacterial strains with the exception that monobutyrin did not inhibit *C. jejuni* (ATCC 33560) growth at the highest concentration tested (5.0 mg/L). Monopropionin at tested concentrations did not inhibit the growth of all tested bacteria with the exception of 1.13 mg/L as MIC on *C. perfringens* (ATCC 12915). The MIC tests indicate that all organic acids and their derivatives, except monopropionin, tested in this experiment exhibited strong antimicrobial effects *in vitro* against tested bacterial strains.

Key words: antimicrobial effects, minimum inhibitory concentration, organic acids

Influences of fiber length on nutrient digestibility in leopard tortoises (*Stigmochelys pardalis*)

A.G. Mullin, B.S., M.S. Edwards, Ph.D
Animal Science Department, California Polytechnic State University, San Luis Obispo, CA

Vertebrate animals lack endogenous enzymes to convert insoluble plant fiber into absorbable nutrients, and therefore are dependent on symbiotic gut microbes to utilize this abundant energy source. Physically effective fiber (peNDF), a concept developed for dairy cattle, considers both physical and chemical characteristics of fiber that influence digestibility, hindgut health and benefit both host and microorganisms (Bjorndal et al., 1990; Mertens, 1997). The need to describe physical and chemical effects of fiber in non-ruminant herbivores has been identified (Bjorndal, 1989; Edwards, 1991; Barboza, 1995; Hatt, 2005; NRC, 2007). Some commercial tortoise feeds include high fiber products physically reduced in size during processing, but the efficacy of these high fiber ingredients has not been investigated. Hindgut fermenting tortoises provide unique insights into digestive processes on fiber because, unlike mammalian herbivores, they do not reduce food particle size via mastication. *Miscanthus* grass, an economical ingredient used to increase insoluble fiber in some complete pet foods, was used as a supplemental source of longer stem fiber. Our objective was to evaluate the influence of *Miscanthus* grass particle length on digestibility and digesta passage in leopard tortoises (*Stigmochelys pardalis*). We hypothesized increased particle length of plant fiber would increase digesta retention, resulting in increased digestibility. Fifteen, individually housed leopard tortoises were offered one of two diets supplemented with *Miscanthus* grass (M-Fiber, Aurora, MO) differing in physical length. Individuals were allocated to treatment based on body mass such that mean body mass between treatments did not differ (P = 0.6883). Chopped, dried *Miscanthus* grasses as supplied by the manufacturer were sieved using a Penn State particle separator (Nasco, Fort Atkinson, WI) to produce forage remaining on a 1.18 – 4 mm (SHORT) and 8 – 19 mm (LONG) sieve. Diets offered comprised of a nutritionally complete tortoise diet (78.1%, mass as-is basis; Mazuri® 5M21, PMI Feeds, St. Louis, MO) and *Miscanthus* grass (21.9%, mass as-is basis). Test diets contained 91% dry matter (DM), 82-84% organic matter (OM, DMB) and 41% amylase neutral detergent fiber (aNDF, DMB). All diets were hand-prepared daily and dried (50°C, > 8h) prior to feeding. Total food intake and fecal output were quantified. Diet quantities fed satisfied 50% of herbivorous reptile field metabolic rate (kJ ME/d) based on body mass on d 1 of diet transition (Nagy et al., 1999). Individuals were transitioned to the respective test diet over 14 d. On d 1 of the 42 d acclimation period each animal was fed 100 indigestible, 2.0 mm acetate beads. Over the next 72 d, fecal collection occurred daily at 0900h, 1400h, and 1900h. Markers were recovered from fecal samples. Upon collection, fecals were immediately refrigerated, frozen within 48 h, dried (50°C) to constant mass and ground through a 1 mm sieve (AOAC, 2016). Acid insoluble ash (AIA) of diet and feces was measured and used to calculate DM and nutrient digestibility (n = 14) (Van Keulen, 1977). Transit time (TT_1) differed with treatment (LONG, 14.14 ± 5.05 d; SHORT, 7.25 ± 2.27 d, $P < 0.05$); however, mean retention time (R_{GIT}) did not differ (LONG, 41.14 ± 12.72 d; SHORT, 47.25 ± 19.44 d, P = 0.2604). No difference in apparent digestibility (aDig, %) of DM (LONG, 79.16 ± 9.32 %; SHORT, 75.50 ± 5.92 %, P = 0.3940) and OM (LONG, 86.18 ± 15.00 %; SHORT, 89.49 ± 12.30 %, P = 0.6610) was detected. Previous research with the same animals found no difference in aDig% of DM (88 – 90%) or OM (89 – 90%) across different lengths of purified cellulose (0.2 mm and 2 mm, 32 – 39 %aNDF, DMB) supplemented diets (Modica, 2016). When consuming only the nutritionally complete tortoise diet offered as a portion of the intake in this study, aDigDM was 88.5%. Because apparent DM and OM digestibility did not differ across these treatments, there may be preferential digestion of certain nutrients. *Xerobates agassizii* fed a mixed diet of high fiber pellets and hay resulted in shorter retention time (R_{GIT} of 14.2 – 14.8 d) (Barboza 1995). Retention of *Miscanthus* grass particles may be due to chemically higher aNDF% and subsequently promote further microbial fermentation of the insoluble fiber. Physical disruption of plant cells is minor because of poor mastication in herbivorous reptiles (Throckmorton, 1976). Therefore, due to limited mastication, the simple hindgut anatomy of tortoises may demonstrate greater flexibility in utilizing higher fiber, longer stemmed roughages. These results are consistent with extent of digestibility of natural forage containing diets consumed by other non-ruminant herbivores. *Miscanthus* grass demonstrates potential as an insoluble dietary fiber source for herbivorous reptiles, including tortoises.

A comparison of peripheral blood mononuclear cell mitochondrial enzyme activity to genetic markers of lactation performance in high and low producing Holstein cows

A.M. Niesen, H.A. Rossow
Department of Population Health and Reproduction, SVM VMTRC
University of California, Davis, CA

Mitochondria are central to metabolism and are the primary energy producers for all biosynthesis. The objective of this study was to compare mitochondrial enzyme activities of high and low producing dairy cows in early lactation to genetic markers of lactation performance. Blood samples were collected from 56 Holstein cows (70 ± 11 DIM) and mitochondria isolated from peripheral blood mononuclear cells (PBMC). Mitochondrial function was assessed by measuring the activity rates of citrate synthase (CS), complex I (CI), complex IV (CIV), and complex V (CV). Milk samples were collected 9 times within a week of blood collection and analyzed for major components using a MilkoScan FT2 by FOSS. Data were analyzed using GLM and the Mixed procedure of SAS (Version 9.4, SAS Inst. Inc., Cary, NC) for high or low groups for each production parameter with cow as the experimental unit of interest and dependent variables parity, and DIM as covariates. There were no interactions between milk yield level (high or low) and parity. Complex I and CV activities were lower in low producing cows than high producing cows for both the multiparous and primiparous groups ($P \leq 0.04$) and CV was not influenced by parity for both multiparous and primiparous cows ($P > 0.1$) across production parameters (ECM, milk fat, and total solids) suggesting that it may serve as a marker of a cow's ability to produce regardless of parity. Genetic indices of fluid merit, milk and protein were not correlated to CS, CI, CIV and CV activities ($P > 0.1$). The genetic index for fat was positively associated with CI activity for primiparous cows ($P \leq 0.02$, $R^2 = 0.27$). Comparisons of genetic predictors for milk fat to actual fat yield had positive correlations and were observed only in primiparous cows ($P \leq 0.01$, $R^2 = 0.40$) while multiparous cows showed no associations ($P > 0.1$, $R^2 = 0.05$) suggesting that genetic markers alone may not accurately predict fat production beyond the first lactation and that the use of mitochondrial enzyme activities may provide a better index of cow production potential as it considers both the animal's ability to process nutrients and parity related changes.

A SYSTEMATIC REVIEW OF RELATIONSHIP BETWEEN RUMEN ACIDOSIS AND LAMINITIS

Lorena Teixeira Passos*[1,2], Luciano Antônio Ritt[2], Vivian Fischer[2], Maria Eugênia Andrighetto Canozzi[3], Noelia Silva Del-Río[1]; [1]Veterinary Medicine Teaching and Research Center, University of California Davis, Tulare, CA, USA, [2]Federal University of Rio Grande do Sul,Postgraduate Animal Science Program, Porto Alegre, RS, Brazil, [3]INIA La Estanzuela - Colonia, Uruguai,

It is widely accepted that laminitis is associated with rumen acidosis; however, the scientific evidence supporting this premise might be limited. The objective of our study was to systematically review the current litterature demonstrating a causal relationship of induced rumen acidosis (ingestion of highly fermentable carbohydrates) and the occurrence of laminitis in cattle. A systematic review was conducted from january to august of 2017 using four electronic databases to identify research articles: PubMed (n = 252), Science Direct (n = 322), Scopus (n = 84) and Web of Science (n = 109). After elimination of duplicates the total number of research articles included was 664. Additional records identified through the Brazilian thesis bank entered the database (n = 2). Abstracts were screened by two reviewers to identify studies that met the inclusion criteria based on three questions: 1) Is this study a primary research?; 2) Does this study investigate rumen acidosis in dairy or beef cattle? and 3) Does this study investigates the association of rumen acidosis and locomotion problems in cattle? After the first screening, 21 studies were selected to be read as full paper. Relevant information was tabulated using Microsoft Excel. The final database was composed of 4 peer-review papers and 2 PhD Thesis. Two papers and 2 thesis induced rumen acidosis with the objective of observing occurrence of acute laminitis, 2 papers with intention of study occurrence of subclinical laminitis. Rumen acidosis was induced by two ways: dietary supply of NFC and oral administration of oligofructose (Table 1). The relationship between acidosis and acute laminitis was partially demonstrated in this systematic review based on the fact, that the authors (2 peer-review papers and 2 PhD Thesis) included in this study presented indicators of acidosis (rumen pH, feces pH, clinical signs) in the animals evaluated and were able to demonstrate clinical changes compatible with the expected ones in cases of acute laminitis. The variation of the diagnostic methods used to diagnose laminitis made it impossible to use the meta-analysis.

Table 1: General characteristics of the relevant experiments

Author(s)	Language	Study design	Country	Animals	Breed	Intervention	Laminitis occurrence
Momcilovic et al., 2000	English	Controlled trial	EUA	16 steers	Holstein	Induced by feeding NFC?	Subclinical
Donovan et al., 2004	English	Controlled trial	EUA	98 cows	Holstein	Induced by feeding NFC?	Subclinical
Thoefner et al., 2004	English	Controlled trial	Not reported	12 heifers	3 Jersey; 6 Holstein-friesian;2 Ayrshire; 1 Guernsey	Induced by oligofructose	Acute
Danscher et al., 2009	English	Quasi-experiment	Denmark	8 heifers	Holstein	Induced by oligofructose	Acute
Noronha, 2017	Portuguese	Controlled trial	Brazil	6 steers	Zebuine Cross-breed	Induced by oligofructose	Acute
Sousa, 2017	Portuguese	Controlled trial	Brazil	29 cows	Nelore	Induced by oligofructose	Acute

KEYWORDS:

Laminitis, bovine, acidosis

Broccoli Stems and Leaves Meal (BSLM) as a Feed Ingredient for Laying Hens

G. Peña[1], A.J. King[1]

[1] Department of Animal Science, University of California, Davis

Animal feed costs for diets account for 65 – 70% of total input. Poultry producers can reduce their cost of feed by utilizing byproducts generated from the human food industry. California is the top producer of broccoli (*Brassica oleracea*), a cruciferous vegetable containing anti-aging, anti-inflammatory, and anti-carcinogenic properties. Consequently, a substantial amount of nutritious agricultural waste (>50%) is generated from the harvest and processing of broccoli plants. Nutritious waste, such as broccoli stems and leaves meal (BSLM), could be used as a feed ingredient in poultry diets. However, broccoli also contains glucosinolates, a secondary plant metabolite, and feeding high glucosinolate concentrations to poultry can lead to reduced feed intake, reduced growth, and organ abnormalities. The objectives of this project were to investigate the effect of 15% BSLM on production and overall health of laying hens as well as nutrient content of eggs and their quality. A total of 110 H&N Nick Chick layers were used for this study. Two diets, one corn/soy-based basal diet as the control and one basal + 15% BSLM as the experimental treatment, were fed to a group of 55 layers for 6 weeks. Feed consumption per bird was determined for weeks 1 to 2 and weeks 4 to 6. Egg weight, Haugh units, egg yolk color, and egg shell break force were analyzed using an EggAnalyzer®. Every two weeks, 14 hens from each diet were culled to remove and weigh liver and thyroid glands. Haugh units were not statistically different between diets for the duration of the study ($p>0.05$). However, egg yolk color values from eggs of hens fed 15%BSLM were statistically higher ($p<0.05$) than that from yolk of hens fed the basal diet by the first week and remained higher throughout the study. Egg production parameters, detrimental effects on layers, deposition rates of glucosinolates in egg yolk and nutrient content of eggs under various heating conditions are yet to be evaluated. Herein reported are the effects of 15% BSLM on egg quality.

Key words: broccoli byproducts, poultry, feed

Factors that contribute to ketosis in early lactation Holstein dairy cattle

C.M. Sousa, H.A. Rossow

Department of Population Health and Reproduction, SVM VMTRC

University of California, Davis, CA

Ketosis in early lactation dairy cattle can negatively impact health events, milk yield and reproductive performance causing a loss in profit for dairy producers. The objective of this retrospective observational study was to identify factors such as management, metabolic profile and milk production that contribute to ketosis in early lactation. One hundred and thirty - two multiparous dry and lactating Holstein cows from a California commercial dairy herd were bled weekly from three weeks prior to calving to the dry period via coccygeal venipuncture into evacuated sterile tubes containing sodium fluoride and potassium oxalate. Milk yield, fat and protein percentage were measured monthly by Tulare County Dairy Herd Improvement Association using a Bentley Instrument ChemSpec 150 (Chaska, MN). Blood metabolite data including non-esterified fatty acids (NEFA) using a commercial kit from Wako Chemicals Inc. (Richmond, VA), glucose and BHBA using a Precision Xtra hand held meter (Abbott Diabetes Care Inc., Alameda, CA), and metabolic data using an Abaxis Vet Scan analyzer Large animal profile rotor (VetScan®, Abaxis, Inc., Union City, CA) were measured weekly. Management variables and milk production from Dairy Comp 305 (Valley Agricultural Software, Tulare, CA) were collected monthly in the previous and current lactation. Using the General Linear Model Procedure of SAS (SAS Institute v.9.4, 2018), BHBA were regressed on blood metabolites, management data and milk production. Variables were eliminated from the regression if $P > 0.05$. The significant management variables that remained in the regression were total days in milk in previous lactation and days dry in previous lactation ($P < 0.05$, $R^2 = 0.70$). When metabolic variables such as aspartate amino transferase, globulin, blood urea nitrogen, NEFA, and glucose from -7 to 14 days relative to calving were added, and milk production variables such as total solids and total fat in the previous lactation were added to the regression, $P < 0.05$ and $R^2 = 0.92$. Residuals were normally distributed and unbiased. These equations indicate that cows at risk for ketosis could be identified prior to calving to help dairy producers manage ketosis before it negatively impacts health and production performance.

Effect of Activo Liquid on early egg production and quality during pullet to layer transition

S.M. Temple[a], T. O'Lear Reid[a], R. Cabrera[b], D.C. Bennett[a]
[a]Animal Science Department, California Polytechnic State University, San Luis Obispo, USA
[b]EW Nutrition USA, Des Moines, Iowa, USA

The transition from the pullet rearing stage to full production is a stressful and demanding period in the life of a laying hen. Birds are exposed to many novelties, including: vaccination, new facilities, social stress, and are expected to reach 90% lay by 25 wk. Previous studies demonstrate plant extracts and essential oils have a positive effect on production performance and health of laying hens; however, these studies focus on mature, in-lay hens, and not during the pullet to layer transition period. Our objective was to evaluate the use of Activo Liquid (EW Nutrition) on the livability of pullets during the pullet to layer transition, and on the development of subsequent egg production. Lohman LSL-Lite pullets (n = 1,380) were reared in conventional cages (72 cages; average density: 19 birds/cage) and transferred at 18 wk to enriched cages (64 cages; average density: 21.5 ± 0.1 birds/cage). Birds received Activo Liquid in their drinking water (0.366 mL Activo/L) for 0, 3, 5 or 7 days, pre- and/or post-transfer to the laying facility in a 4x4 factorial design, with four replicate cages per cell. Mortality was monitored throughout the experiment. Egg production was recorded daily between 19 and 26 wk, and again at 35 wk. At 25 and 35 wk, all eggs were collected, weighed, and assessed for eggshell cracks (checks). Two eggs per cage were randomly selected to assess egg shell quality (weight, thickness, and breaking strength). Data were analyzed with repeated measures ANOVA in Systat 9 (SPSS Science, Chicago, IL), with the length of time birds were provided with Activo Pre- and Post-transfer as main effects, and week of measure (age) as the repeated effect. Statistical significance for detecting differences among the treatment groups was accepted when $P < 0.05$. Post-hoc LSD means separation were performed to separate treatment effects. No mortality was recorded for hens between 18 to 26 wk. Pre-transfer dosing of Activo significantly affected egg production between 19 and 24 wk, due to lower egg production in the 7-day treatment group. Pre-transfer dosing of Activo did not significantly affect egg production or eggshell quality at 25, 26, and 35 wk. Post-transfer dosing of Activo did not significantly affected egg production between 19 and 24 wk; however, egg production was significantly affect at 25 and 26 wk, mainly due to higher production in the 3-day treatment group, but this affect was not observed at 35 wk. Post-transfer dosing of Activo did not significantly affect eggshell quality. These results support that early commercial layer production was improved by 3-d post-transfer dosing of Activo.

Enzymatically digested food waste as a feed supplement in chicken diets

L. Truong[1], D. Morash[2], Y. Liu[1], A.J. King[1]

[1] Department of Animal Science, University of California, Davis, CA
[2] California Safe Soil, LLC, McClellan Park, CA

ABSTRACT

Due to concerns of feeding 9 billion humans by 2050 and reducing greenhouse gas emissions, methods of diverting current food waste were explored. This study evaluated the performance and lipid parameters of Cobb broiler chicks fed non-pathogenic food waste from local Californian grocery stores mixed with bakery waste (FW, 1:1, wt/wt, as-fed basis) as a feed ingredient in broiler starter diets. A total of 432 straight-run chicks (1-day-old) were randomly assigned to a corn/soy basal diet (Control); 75% Control + 25% FW (75:25, wt/wt, dry matter basis); or 50% Control + 50% FW (50:50, wt/wt, dry matter basis). The experiment was conducted for 14 days. On days 6, 10, and 14, individual bird weights (g), feed intake (g), serum, jejunum, and thigh meat were collected. Feed conversion ratio was calculated based on weight and feed intake. Jejunum samples were measured for villi height (VH) and crypt depth (CD) on days 6 and 14. The FW and diets were analyzed for fatty acid (FA) composition. Thigh meat from each treatment were also analyzed for FA and lipid oxidation in the raw or cooked state. Significant differences were determined at $p<0.05$. Results indicated that on days 6, 10, and 14, birds fed 75:25 had significantly greater body weights when compared to that of Control and 50:50. On day 14, feed intake was significantly greater in 75:25 and feed conversion ratio was not significantly different than Control. There were no significant differences in VH, CD, VH:CD, or serum parameters among diets. There were no significant differences for FA among separate batches of FW, indicating that batches could be replicated. Numerically, there was (1) a lower n-6:n-3 polyunsaturated fatty acids (PUFA) and (2) a higher saturated fatty acids (SFA):PUFA in FW, the 50:50 diet, and the subsequent thigh meat of broilers fed 50:50 when compared to Control and 75:25. The Control diet and thigh meat had the highest concentration of PUFA in comparison to 50:50 which could lead to increased oxidative processes and decreased shelf life. The 75:25 diet and thigh meat had intermediate levels of n-6:n-3 as well as SFA:PUFA. There was no difference in lipid oxidation among treatments; however, Control thigh meat, with higher concentrations of PUFA, had numerically higher lipid oxidation values than that of meat from other diets. Therefore, results indicated that 25% FW included in a broiler starter diet for 2 weeks could increase production rate of Cobb broiler chicken when compared to the Control and can be a substitute for corn and soybean meal in Cobb broiler starter diets.

Effect of postpartum milking strategy on plasma calcium concentration and risk of subclinical hypocalcemia in dairy cows

A Valldecabres*[1], R Lopes[1], A Lago[2], C Blanc[3], N Silva-del-Río[1];

Veterinary Medicine Teaching and Research Center, University of California Davis, Tulare, CA, USA[1].
DairyExperts Inc., Tulare, CA, USA[2].
GTV Dairies, Tipton, CA, USA[3].

The aim of the present study was to evaluate the effect of different postpartum milking strategies on plasma Ca concentration in multiparous dairy cows. A total of 83 Jersey and Jersey × Holstein crossbreed cows of 2nd to 8th parity, were enrolled in the study before 1st postpartum milking. Milking strategies implemented during the first 2 d postpartum were: once-a-day milking (1M; cows were milked every 24 h; n = 24), twice-a-day milking (2M; cows were milked every 12 h; n = 21), delayed milking (DM; cows were not milked for the first 2 postpartum milkings and were milked every 12 h afterwards; n = 19), and restricted milking (RM; cows were milked 3 L every 12 h; n = 19). Blood samples for total plasma Ca analysis were collected from the coccygeal vessels into heparinized vacuum tubes starting before 1st postpartum milking, every 4 h up to 48 h and at 72 h postpartum. Plasma Ca concentration changes during the study period and risk of subclinical hypocalcemia (SCH; Ca ≤2.12 mmol/L) at 48 and 72 h postpartum were evaluated using MIXED and GENMOD procedures of SAS, respectively. Prevalence of SCH before 1st postpartum milking was 48%. There were effects of treatment ($P = 0.03$), parity ($P = 0.003$), time ($P < 0.001$), initial calcemic status ($P < 0.001$; normocalcemic or subclinically hypocalcemic) and time by initial calcemic status ($P < 0.001$) on plasma Ca concentration. Overall, lower plasma Ca concentration was observed for 2M cows (2.04 mmol/l) compared to DM (2.17 mmol/L; $P = 0.04$) and RM cows (2.17 mmol/L; $P = 0.04$), but no differences were observed with 1M (2.12 mmol/l). At 48 h postpartum the risk of SCH was lower for 1M (Risk ratio; RR = 0.27; $P < 0.001$), DM (RR = 0.55; $P = 0.02$) and RM cows (RR = 0.41; $P < 0.001$) than for 2M cows. At 72 h postpartum the risk of SCH was lower for 1M (RR = 0.26; $P < 0.001$) and DM cows (RR = 0.41; $P < 0.001$) than for 2M cows. Our results suggest that postpartum plasma Ca concentration and risk of SCH may be influenced by postpartum milking strategies, warranting its study as a prophylactic strategy for hypocalcemia.
Project funded by USDA-NIFA (1013457 CFAH).

KEYWORDS

Dairy cow, hypocalcemia, transition cow

Comparison of nutrition composition, quality, and sensory differences among Dorper, domestic commercial crossbred and Australian lamb meat.

K.M. Villatoro[1], Z. Bauer[2], T. Lew[2], X. Yang[1], D. Woerner[3] and M. Chao[2].

[1]Department of Animal Science, University of California, Davis, [2]College of Agriculture, California State University, Chico, [3]Department of Animal Sciences, Colorado State University.

The objective of this study was to compare the nutritional, quality, and sensory attributes among Dorper sheep breed, domestic commercial crossbred (DCC) and Australian crossbred lamb meat. **Methods:** A total of 60 untrimmed lamb saddles (NAMP #231) from three sources (Dorper, n=20; DCC, n=20; and Australian, n=20) were purchased from commercial packing plants and warehouses. Lamb saddles from lamb sources were aged in a cooler (2°C) according to their production dates to achieve an aging time between 29-32 days. All aged saddles were frozen (-20°C) until sample preparation. On the sample preparation day, each saddle was cut on a bandsaw to 2.54 cm chops, deboned and trimmed to 0.30 cm subcutaneous fat. The chops were used to measure pH, objective color (L^*, a^*, and b^*), objective tenderness [Warner-Bratzler Shear Force (WBSF)], cook loss (differences in weight between raw and cooked samples) and nutrient analysis (moisture, protein, fat, ash, carbohydrate, and calories). A consumer panel of 120 untrained participants was used to evaluate tenderness, flavor acceptability, juiciness, and overall acceptance using a 9-point hedonic scale (1=Dislike extremely and 9=Like extremely). A trained panel with 6 trained panelists was used to evaluate flavor intensity, tenderness, and off-flavor intensity using an unstructured line scale anchored at both ends (0= absence or low intensity of specified attribute, 100= extreme intensity of specified flavor attribute). Data were analyzed as a completely randomized design with three treatments. One-way ANOVA was performed to compare the means of the attributes between the lamb sources using SAS 9.4, and the Least Significant Difference test (LSD) was calculated at the 5% significance level to compare the treatment means.
Results: The DCC lamb meat had lower ($P < 0.05$) pH, carbohydrate content and off-flavor intensity compared to Australian and Dorper lamb meat. The DCC lamb meat was also rated with more flavor acceptability ($P < 0.05$) compared to Dorper lamb meat by untrained panelists, while Australian lamb meat was rated similar ($P > 0.05$) in flavor acceptability compared to DCC and Dorper lamb meat. Untrained panelists preferred ($P < 0.05$) the tenderness of Australian lamb, which was also rated with greater ($P < 0.05$) tenderness by trained panelists compared to Dorper lamb. Finally, Dorper lamb meat had greater ($P < 0.05$) WBSF value and was rated with the lowest ($P < 0.05$) rating in overall acceptance by the untrained panelists compared to Australian and DCC lamb meat. No differences ($P > 0.05$) were found for L*, a*, b*, cook loss, moisture, fat, ash, calories, juiciness and flavor intensity among the treatments. **Conclusions:** These results indicated that there are apparent meat quality differences among the three lamb meat sources. Overall, consumers preferred DCC lamb meat compared to Dorper and Australian lamb meat. However, factors such as specific genetic makeups, age and diets were not accounted for in this research. Additional research with a more controlled environment is needed to shed light on the true palatability traits of Dorper lamb meat.

Keywords: Lamb, meat quality, Dorper, Australian lamb, Domestic cross-bred

Incidence of subclinical and clinical ketosis in the California central valley: similarities among commercial herds

M Wukadinovich, H.A. Rossow
Department of Population Health and Reproduction, SVM VMTRC
University of California, Davis

Ketosis is a common metabolic disease in postpartum dairy cows due to negative energy balance at the onset of lactation resulting from insufficient dry matter intake and disorders of energy metabolism. Ketosis results in hyperketonemia, hypoglycemia, decreased milk yield, and increased risk of being culled from the herd. Often, it is associated with other primary health disorders. To estimate the incidence of subclinical (SCK) and clinical ketosis (CK) in the California Central Valley, blood samples and milk production data from 172 Holstein multiparous cows across 10 commercial dairy herds in Tulare, Kings, and Kern counties were collected. In February 2018, an average of 17 cows from each herd were bled during week 1 and week 2 postpartum and whole blood was analyzed for glucose and acetoacetate (AcAc) levels using NovaMax® meters (Nova Diabetes Care, Inc., Billerica, MA). Subclinical and clinical ketosis were defined as 1.0-1.4 mmol/L and >1.4 mmol/L AcAc in whole blood, respectively. Previous lactation milk fat yield and 305-day mature equivalent milk yield were collected from DairyComp305 (Valley Ag Software, Tulare, CA). All data were analyzed using the Mixed Procedure of SAS with repeated measures by cow or a Generalized Linear Model Procedure (v. 9.4, SAS Institute 2015). Across all herds, average SCK was 12% (repeat cases 5%) and CK was 11% (repeat 61%). All repeat CK cases progressed from SCK in week 1. Across herds, the lowest incidence of hyperketonemia (> 1.0 mmol/L AcAc) was 0% and the highest incidence was 44%. Glucose and AcAc were inversely related ($P < 0.001$; $R^2 = 0.12$). Hyperketonemic cows produced 393 kg more milk ($P = 0.1$) and 44 kg more milk fat ($P = 0.02$) in their previous lactation compared to non-ketotic cows. Five dairies housed fresh cows in free stall corrals and five dairies housed fresh cows in free stall corrals with an attached exercise corral or in an open lot style corral. Cows that had access to an exercise corral or were housed in an open lot had lower blood AcAc levels compared to cows housed in free stalls (0.55 vs. 0.64 mmol/L; $P = 0.02$, $R^2 = 0.17$). Additionally, cows that had access to an exercise corral had a decreased recurrence of a ketotic event in the same lactation (5% vs. 26%). Therefore, factors impacting blood AcAc levels on these commercial herds were glucose, previous lactation milk and fat yield, and access to an exercise corral. Greater producing cows and cows without access to an exercise corral were more predisposed to developing ketosis in the postpartum period.