Impact of Essential Amino Acid Balancing Postpartum on Lactation Performance by Dairy Cows

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Introduction

Precision feeding became an essential strategy to optimize income over feed cost in dairy herds, mainly through the increase in nutrient utilization and the corresponding benefits in milk and milk components production. Furthermore, environmental impact of dairy herds is lessened when nutrient utilization is improved. All these benefits may be achieved through the formulation of diets targeting for optimum amino acid (**AA**) balancing. For example, several research studies reported increases in milk and/or milk protein production with supplementation of rumen-protected amino acids (**RPAA**). In addition, RPAA allows dairy nutritionist to formulate diets with lower CP content while maintaining or sometimes improving performance; and these diets were often reported to reduce N excretion. Combined these results suggest that RPAA supplementation is desirable, especially in eras when or locations where protein feedstuffs are expensive. Thus, the objective of the present article is to discuss the relationship between dietary essential amino acids (**EAA**) concentration and lactation performance of early lactation dairy cows. Our focus will be to present some findings from a recent meta-analysis study from our group combined with recent literature.

Benefits of Balancing Essential AA on Performance of Dairy Cows

Meta-analysis description

Our meta-analysis study used an unconventional approach; instead of summarizing published literature data, a dataset comprised of 20 unpublished feeding trials was assembled and used (**Table 1**). This approach was selected due to the uniqueness of the dataset. All feeding trials were performed in collaboration between The William H. Miner Agricultural Research Institute (Chazy, NY) and Ajinomoto Heartland Inc. (Chicago, IL) in the 1990’s and were designed as continuous lactation trials to evaluate the effect of lysine or lysine/methionine supplementation on early-lactation performance by dairy cows. Diets from all 20 feeding trials were formulated by the same nutritionist using CPM/CNCPS (version 2) which provided a complete dietary AA profile (**Table 2**). Furthermore, feed and milk samples from all 20 feeding trials were analyzed at the same commercial laboratory.
Data analysis were performed to evaluate: 1) the relationship between individual dietary essential AA concentration (g of AA/Mcal of ME) and lactation performance of each of

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the initial 4 weeks of lactation (Ferraretto et al., 2016); and 2) to evaluate various strategies of RPAA supplementation on lactation performance (Ferraretto et al., unpublished). Although AA concentrations are often reported as percentage of metabolizable protein (**MP**) supply, our dietary AA concentrations were expressed in g of AA/Mcal of ME based on the recommendations of Higgs (2014). Higgs (2014) observed a relationship between AA supply to ME, but not MP, and underscored that perhaps its use could improve predictions of AA utilization.

Rumen Protected Methionine and Lysine

It is well documented that diets based on corn silage and soybean meal (typical North American diets) are limiting in lysine and methionine for milk production and milk protein synthesis (NRC, 2001). Therefore, balancing dairy cow diets with limiting and/or essential AA can be an effective strategy to increase milk and milk protein production. Several studies demonstrated that supplementation with rumen-protected lysine (**RPL**) and methionine (**RPM**) may improve feed intake, milk production, and content and production of milk protein. Those responses are more likely to be observed in high-producing dairy cows rather than in low-producing cows, when rumen-undegraded protein (**RUP**) supplies a greater portion of metabolizable protein (**MP)**, and for cows in early than mid- and late-lactation (NRC, 2001; Socha et al., 2005). In addition, increased dietary concentrations of lysine and methionine in MP have indicated that milk protein content is more sensitive than milk protein production; and that increases in milk protein content are independent of milk production (NRC, 2001).

Our meta-analysis (Ferraretto et al., unpublished) compared the effects of RPL supplementation in early to mid-lactation cows (**Table 3**). Treatments were control, RPL plus dietary sources of methionine, combination of RPL and RPM, and 2x RPL plus a dietary source of methionine. Overall, AA balancing enhanced intake and yield of milk and milk components. However, improvements were greater for RPL plus RPM than other AA treatments. Socha et al. (2005) evaluated the effect of supplementing corn-based diets to pre-partum and early post-partum cows with RPM alone or with a combination of RPM plus RPL on lactation performance. They reported that cows that were supplemented with RPM plus RPL increased production of energy-corrected milk (**ECM**), milk true protein, and milk fat. However, no effects on performance were detected when RPM was supplemented alone compared to basal diet and basal diet plus RPAA combination. Wang et al. (2010) evaluated the effect of supplementing corn-based diets of mid-lactation cows with RPM, RPL, and in combination (RPM + RPL) in diets slightly limiting in MP. The authors observed an increase in milk production of 1.5 kg/d by cows fed RPL or 2.0 kg/d by cows fed RPM alone, and an increase of 3.8 kg/d when RPM and RPL were supplemented together compared to the basal diet (without RPAA supplementation). Also, increased milk protein yield and fat content were observed when diets supplemented with RPM alone or in combination with RPM plus RPL were fed.

Relationships between dietary methionine or lysine concentrations and yields of milk and milk protein are in **Table 4** (Ferraretto et al., 2016). Our meta-analysis revealed positive relationships between dietary methionine and milk and milk protein yields during weeks 1 to 4. This is in agreement with a recent meta-analysis review which used CNCPS (version 6.5) to predict nutrient supplies from a large number of published experiments (Lean et al., 2018). These authors observed a positive relationship between metabolizable methionine (g/d) with milk protein yield and content. The supply of methionine estimated by Lean et al. (2018) was similar (57 vs. 54 g/d and 2.24 vs. 2.10% of MP) to that of Ferraretto et al. (2016).

Ferraretto et al. (2016) observed an increase in actual and ECM yields along with lysine concentration during weeks 1, 2 and 4, whereas milk protein yield increased during the 4 initial weeks of lactation (**Table 3**). Interestingly, no relationship was detected between lysine and milk protein or milk yield (Lean et al., 2018). However, the supply of lysine estimated by Lean et al. (2018) was lower (162 vs. 179 g/d and 6.38 vs. 6.88% of MP) than Ferraretto et al. (2016). According to CNCPS 6.5, the optimal lysine supply (% of MP) recommended to maximum protein yield is 6.68%. Perhaps the lack of a relationship in the review by Lean et al. (2018) is related to the supply of lysine being under the recommended concentration by CNCPS. Furthermore, there is a difference in lactation stage between the 2 meta-analyses (early- vs. mid-lactation) which may also have contributed to this difference as speculated in previous studies (Wang et al., 2010; Osorio et al., 2016). Overall, these results underscore the importance of balancing lysine and methionine when formulating diets for high-producing dairy cows; and this importance is increased during early lactation.

Reducing dietary CP with rumen-protected AA

The efficiency of N utilization for milk (milk N/N intake, %) in high-producing dairy cows can range from 25 to 35% with the remaining N excreted in feces and urine (Arriola Apelo et al., 2014b). Nitrogen can be a pollutant from animal operations, having a negative environmental impact (e.g., surface water eutrophication, ground water nitrate, and ammonia emissions; US EPA, 2011). Therefore, the US dairy industry is under pressure to reduce N excretion in dairy cow operations and, as a consequence, there is an increased interest to improve N utilization efficiency without compromising milk production and animal health (Lapierre et al., 2005; Wang et al., 2010). Previous studies highlighted that decreasing dietary CP concentration is an efficient way to reduce N excretion (Olmos Colmenero and Broderick, 2006; Agle et al., 2010). However, reducing intake of CP may result in a deficient supply of MP, and thereby reduce yields of milk and protein and milk protein content (Cabrita et al., 2011). A strategy to overcome this issue may be achieved by improving the balance of metabolizable AA supply (Lapierre et al., 2005; Lee et al., 2012a).

Ferraretto et al. (unpublished) evaluated effect of diets of low CP, RDP, or MP concentrations supplemented with and without RPL and RPM on performance of early-lactation dairy cows. Dry matter intake and yield of milk, milk protein, and ECM increased for cows fed diets supplemented with RPAA compared to the cows without supplementation (**Table 5**). Socha et al. (2005) evaluated the effect of supplementing corn-based diets of pre-partum and early post-partum cows with a combination of RPM plus RPL with two different concentrations of CP (18.5 vs. 16.0%, DM basis) on lactation performance. These authors observed that cows fed the diet containing 16% CP numerically increased DMI (+ 0.4 kg/d), ECM (+ 1.5 kg/d), and had greater gross efficiency of N utilization (35 vs. 29%) compared with cows fed a diet with 18.5% CP. Recently, Nursoy et al. (2017) evaluated the optimal dietary CP concentration for mid-lactation cows fed corn-based diets supplemented with soybean meal (**SBM**) plus RPM. The authors tested 4 CP concentrations (11, 13, 15, and 17%, DM basis), and all diets were formulated to maintain a methionine to lysine ratio of 3.1:1 (% of MP). The authors concluded that feeding a corn-based diet supplemented with SBM plus RPM with 15% CP was adequate for mid-lactation cows producing approximately 40 kg/d of milk. Compared to a 17% CP diet, cows fed the 15% CP diet improved N efficiency (29.5 vs. 32.7%, respectively). On the other hand, Cabrita et al. (2011) evaluated the effects of dietary CP concentration (16 vs. 14%, DM basis) and balance of MP by manipulating the main protein sources (SBM and corn byproducts), and by adding RPM plus RPL on lactation performance of dairy cows fed corn silage-based diets. Overall, authors did not observe benefits supplementing RPM and RPL in reducing dietary CP from 16 to 14%, since no significant differences were observed for all production traits evaluated.

Lee et al. (2012b) conducted 2 experiments evaluating the effects of supplementation of MP-deficient or MP adequate diets with RPM and RPL on milk production, milk components, and N utilization. In experiment 1 dietary treatments were: (1) MP-adequate diet without RPAA supplementation; (2) MP-deficient diet (approximately 12% of MP-adequate diet) plus supplementation with RPL; and (3) MP-deficient diet supplemented with RPL plus RPM. In experiment 2, dietary treatments were: (1) adequate MP supplemented with RPL; and (2) adequate MP supplemented with RPL plus RPM. Overall, authors did not observe significant differences in milk yield and components among the dietary treatments (albeit milk yield decreased by about 1 kg/d for both deficient-MP diets). When the MP-deficient diet was only supplemented with RPL, milk protein content decreased compared to adequate-MP diet.

Overall, these results indicate that supplementing RPL and RPM along with a reduction in dietary CP may be an important nutritional strategy to improve N utilization efficiency and minimize potential environmental pollution by dairy cow operations.

Other Essential AA for high producing dairy cows

Although lysine and methionine are the two most limiting AA when feeding corn-silage based diets, recent studies suggest that increasing the supply of other EAA may improve lactation performance of dairy cows. For example, recent studies suggested that histidine may be a limiting AA after lysine and methionine (Vanhatalo et al., 1999; Lee et al. 2012a,b; Giallongo et al. 2016). Furthermore, other studies indicate that other EAA may play an important role to improve yields of milk and protein (Haque et al., 2013; Arriola Apelo et al., 2014a).

Ferraretto et al. (2016) reported that dietary histidine had a positive relationship with milk yield, but a negative relationship to milk protein content on weeks 2 to 4 of lactation (**Table 6**). This is in agreement with previous research that suggested production benefits related to histidine (Lee et al., 2012a, b; Giallongo et al., 2016; Lean et al., 2018). For example, Giallongo et al. (2016) evaluated the effects of supplementation of RPAA (methionine, lysine, and histidine) in MP-deficient diets on performance of dairy cows. The diet deficient in MP supplemented exclusively with rumen-protected histidine increased milk protein content, and when supplemented with rumen-protected methionine, lysine, and histidine together, it further increased yields of milk fat, protein, and ECM and ECM feed efficiency compared to the diet deficient in MP without RPAA supplementation. Lean et al. (2018) observed that histidine elicited a positive response in milk yield despite the small difference between treatments; control and treatment groups supplied 2.57% and 2.61% of MP, respectively. These authors suggested that this response indicates that histidine plays a role as a co-limiting AA in dairy cow diets.

Dietary valine was positively related to ECM, but negatively to milk protein concentration on weeks 2 and 3 (Ferraretto et al., 2016). On weeks 3 and 4, a positive relationship between milk yield and valine was observed. Haque et al. (2013) evaluated milk protein responses to changes in post-ruminal infusion of EAA and depletion of arginine, isoleucine, and valine in dairy cows. The authors observed that when cows did not receive post-ruminal infusions of valine, milk protein synthesis was decreased compared to cows that did receive post-ruminal infusions of all EAA. They concluded that the lower level of valine (4.5% of MP) may explain the negative effect on milk protein. In our study the average concentration of valine was 5.7% of MP which is within the range reported by Doepel et al. (2004).

Other EAA were also related to lactation performance in our meta-analysis study (Ferraretto et al., 2016). Dietary concentration of arginine and threonine were negatively related to milk fat content and yield on weeks 3 and 4 of lactation. Moreover, quadratic relationships between milk or milk protein yields and dietary concentrations of leucine and phenylalanine were observed during weeks 1 to 4. Isoleucine concentration of the diet was negatively related to ECM and milk protein yields during weeks 3 and 4 and to milk and milk fat yields on week 3. Dietary concentration of tryptophan was negatively related to ECM, milk fat content, and milk fat yield. Leucine was associated with greater milk protein yield in the study by Lean et al. (2018). In addition, tryptophan and threonine affected milk yield positively (Lean et al., 2018).

Summary

Overall, benefits on lactation performance were observed with supplementation of RPAA to early lactation dairy cows, particularly when RP sources included both, methionine and lysine. In addition, RPAA supplementation is a viable tool to reduce concentration of CP, RDP or MP in dairy cow diets. Increased dietary concentrations of methionine, lysine, valine, and histidine enhanced lactation performance in lactating cows during the initial 4 weeks of lactation. In contrast, isoleucine and tryptophan were negatively related to lactation performance whereas arginine and threonine depressed milk fat. These results underscore the importance of amino acid balancing beyond the lysine to methionine ratio when formulating diets for early lactation dairy cows.

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Table 1. Cow numbers, supplementation period, length of collection of milk yield, and amino acid supplementation strategy of the 20 studies used for the meta-analysis.

Study	n[1]	Supplementation Period[2]	Lactation[3]	Lysine[4]	Methionine[5]
1	41	-21 to 140	140	+	+
2	7	-21 to 56	140	+	+
3	27	-14 to 42	112	+	-
4	17	-14 to 42	112	+	-
5	35	-21 to 42	112	+	-
6	20	1 to 28	56	+	+
7	11	1 to 28	112	+	+
8	16	1 to 28	112	+	-
9	40	-21 to 56	168	+	+
10	40	-21 to 42	70	+	-
11	13	-14 to 42	168	+	+
12	20	1 to 28	112	+	+
13	11	-21 to 70	308	+	+
14	45	-21 to 42	112	+	-
15	14	-21 to 42	168	+	+
16	14	1 to 42	168	+	-
17	30	-14 to 28	112	+	+
18	24	-21 to 42	168	+	-
19	14	-21 to 42	140	+	+
20	15	-21 to 301	308	+	+

[1] Number of lactating dairy cows used in trial per treatment.
[2] Period of rumen-protected amino acid supplementation, presented as days in milk.
[3] Days in milk at the end of lactation performance assessment.
[4] With or without rumen-protected lysine (+, -).
[5] With or without rumen-protected methionine (+, -).

Table 2. Selected nutrient composition of diets fed in studies used in the meta-analysis.

Item	Average	SD	Minimum	Maximum
Forage, % of DM	44.1	4.5	37.1	56.9
NDF, % of DM	30.6	2.4	26.8	37.0
NFC, % of DM	39.2	2.2	35.7	44.2
Fat, % of DM	5.5	1.3	2.6	7.2
Metabolizable energy, Mcal/d	61.8	3.0	55.3	68.1
NEL, Mcal/kg of DM	1.76	0.88	1.56	1.94
CP, % of DM	18.4	1.0	15.9	20.6
RDP, % of CP	60.1	4.7	50.1	69.9
RUP, % of CP	39.9	4.7	30.1	49.9
Metabolizable protein, g/d	2601	168	2381	2930
Methionine, g/d	54	7	44	67
Lysine, g/d	179	19	137	223
Arginine, g/d	157	10	134	176
Threonine, g/d	117	7	105	131
Leucine, g/d	220	25	180	287
Isoleucine, g/d	124	8	111	143
Valine, g/d	148	13	131	174
Histidine, g/d	69	6	56	82
Phenylalanine, g/d	132	10	115	158
Tryptophan, g/d	36	3	29	41
Essential amino acids, g/d	1236	80	1077	1396

Table 3. Effect of supplementation of rumen-protected amino acids on lactation performance by early lactation dairy cows.[1]

Item	CON	RPL	RPLM	RP2LM	SEM[2]	*P*-value
DM intake, kg/d	19.9[c]	21.6[a]	20.6[b]	21.6[a]	0.7	0.001
Milk, kg/d	41.3[d]	42.1[c]	45.0[a]	43.2[b]	1.8	0.001
ECM, kg/d	41.9[c]	42.8[b]	45.1[a]	44.2[a]	1.8	0.001
Milk fat, %	3.70	3.69	3.60	3.76	0.09	0.06
Milk fat, kg/d	1.51[c]	1.52[bc]	1.61[a]	1.58[ab]	0.07	0.001
Milk protein, %	2.85[b]	2.90[a]	2.81[c]	2.92[a]	0.03	0.001
Milk protein, kg/d	1.18[c]	1.23[b]	1.36[a]	1.28[b]	0.06	0.001
Milk lactose, %	4.74	4.77	4.76	4.76	0.03	0.13
Milk lactose, kg/d	1.80[b]	1.82[b]	1.98[a]	1.81[b]	0.13	0.001
MUN, mg/dL	17.5	16.0	17.8	18.2	1.5	0.28

[1] Treatments were control diet (CON), rumen protected lysine plus a dietary source of methionine (RPL), rumen protected lysine and methionine (RPLM), and 2x rumen protected lysine plus a dietary source of methionine (RP2LM).
[2] Standard error of the mean.

Table 4. Relationship between dietary intake of methionine or lysine (g/Mcal of ME) and yield of milk and milk protein by dairy cows during the initial four weeks of lactation.[1]

Item	Intercept	SE	Slope	SE	P-value	RMSE[2]
Methionine						
Week 1						
Milk yield, kg/d	25.28	3.59	6.494	3.714	0.10	1.75
Milk true protein, kg/d	0.97	0.14	0.245	0.126	0.07	0.11
Week 2						
Milk yield, kg/d	30.69	3.88	7.883	3.869	0.06	1.89
Milk true protein, kg/d	0.89	0.13	0.341	0.131	0.02	0.05
Week 3						
Milk yield, kg/d	33.87	4.05	8.116	4.428	0.08	2.19
Milk true protein, kg/d	0.97	0.10	0.225	0.087	0.02	0.07
Week 4						
Milk yield, kg/d	33.22	3.78	10.901	3.242	0.01	2.26
Milk true protein, kg/d	0.87	0.11	0.323	0.103	0.01	0.07
Lysine						
Week 1						
Milk yield, kg/d	23.93	3.55	2.423	1.073	0.04	1.85
Milk true protein, kg/d	0.85	0.16	0.119	0.052	0.03	0.10
Week 2						
Milk yield, kg/d	28.79	3.94	3.050	1.135	0.02	1.86
Milk true protein, kg/d	0.84	0.14	0.120	0.042	0.02	0.04
Week 3						
Milk true protein, kg/d	0.92	0.15	0.088	0.047	0.08	0.05
Week 4						
Milk yield, kg/d	32.77	4.48	3.498	1.289	0.02	1.93
Milk true protein, kg/d	0.88	0.14	0.108	0.046	0.03	0.03

[1] Adjusted for the random effect of study.
[2] Root mean square error.

Table 5. Effect of supplementation of rumen-protected methionine and lysine in diets formulated with low amounts of CP, RDP, or MP on lactation performance by early lactation dairy cows.[1]

Item	CON	RP-	SEM[2]	*P*-value
DM intake, kg/d	21.6	22.7	1.0	0.001
Milk, kg/d	31.6	33.9	3.0	0.001
ECM, kg/d	33.0	33.9	3.3	0.001
Milk fat, %	3.82	3.53	0.10	0.001
Milk fat, kg/d	1.21	1.18	0.14	0.10
Milk protein, %	2.89	2.92	0.08	0.52
Milk protein, kg/d	0.91	0.98	0.06	0.001
Milk lactose, %	4.71	4.73	0.02	0.36
Milk lactose, kg/d	1.50	1.60	0.13	0.001

[1] Treatments were control diet (CON) and low CP, RDP or MP diets with rumen protected methionine and lysine (RP-).
[2] Standard error of the mean.

Table 6. Effects of dietary histidine (g/Mcal of ME) on milk yield and milk protein content by dairy cows during the initial four weeks of lactation.[1]

Item	Intercept	SE	Slope	SE	P-value	RMSE[2]
Week 2						
Milk yield, kg/d	24.87	5.98	11.39	5.780	0.07	2.13
Milk true protein, %	3.65	0.16	-0.441	0.141	0.01	0.05
Week 3						
Milk yield, kg/d	22.31	7.16	16.629	6.781	0.03	2.34
Milk true protein, %	3.24	0.12	-0.334	0.103	0.01	0.06
Week 4						
Milk yield, kg/d	26.76	7.19	14.554	6.805	0.05	2.24
Milk true protein, %	3.07	0.16	-0.274	0.139	0.06	0.06

[1] Adjusted for the random effect of study.
[2] Root mean square error.

SESSION NOTES