Integrating Energy and Amino Acids to Enhance Milk Component Yield

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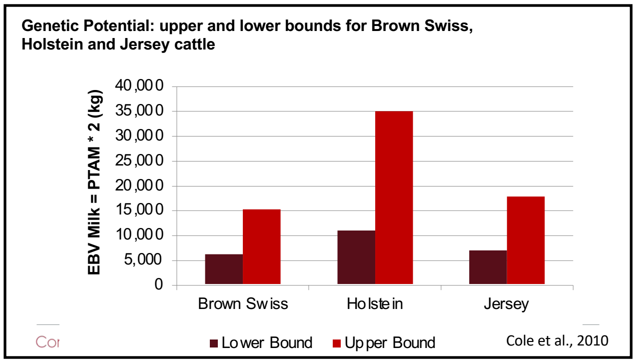
Introduction

- Feed efficiency and carbon intensity are directly correlated
  - Any increase in feed efficiency reduces carbon intensity (feed C neutral)
- Essential amino acids are required for protein synthesis, nutrient signaling, and conversion to other metabolites like non-essential amino acids, enzymes and hormones
- The system is constantly running, but it is not always using the energy efficiently – parallels energy spilling in bacteria
- There is an obligate requirement for amino acids in fatty acid synthesis and all of this is integrated in liver and mammary metabolism but is not well discussed

Introduction

- This talk will focus on milk components however it is important to appreciate the interaction between amino acids and fatty acids in enhancing milk components
- This aspect of nutrition is rarely, if ever, discussed, yet observations are available demonstrating the impact of the interactions
- And consideration of the right precursors for milk fat yield are also important



Genetic Potential: upper and lower bounds for Brown Swiss, Holstein and Jersey cattle

Cole et al., 2010

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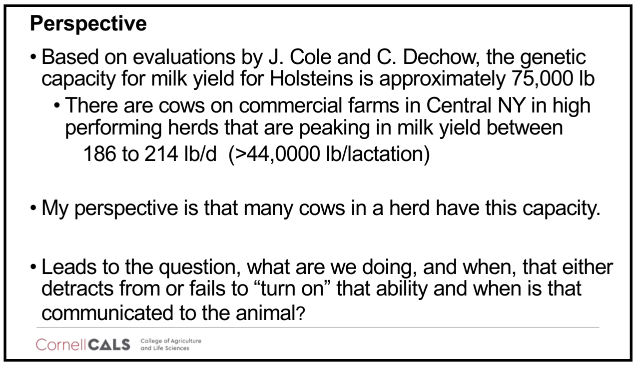

Perspective

- Based on evaluations by J. Cole and C. Dechow, the genetic capacity for milk yield for Holsteins is approximately 75,000 lb
  - There are cows on commercial farms in Central NY in high performing herds that are peaking in milk yield between 186 to 214 lb/d (>44,0000 lb/lactation)
- My perspective is that many cows in a herd have this capacity.
- Leads to the question, what are we doing, and when, that either detracts from or fails to “turn on” that ability and when is that communicated to the animal?

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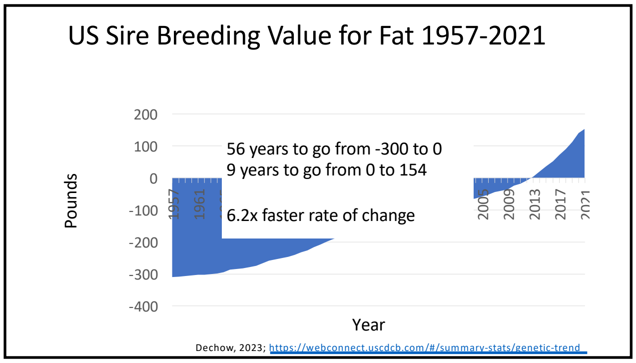

US Sire Breeding Value for Fat 1957-2021

56 years to go from -300 to 0
9 years to go from 0 to 154

6.2x faster rate of change

Dechow, 2023; https://webconnect.uscdcb.com/#/summary-stats/genetic-trend

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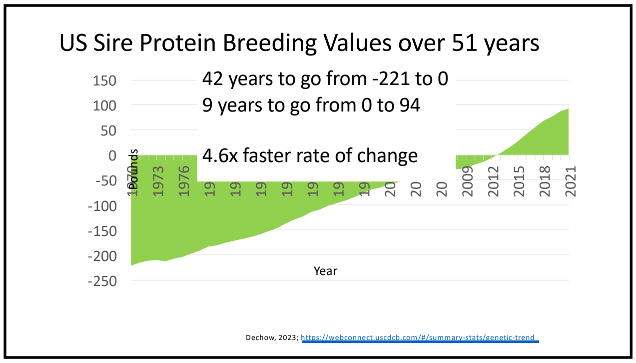
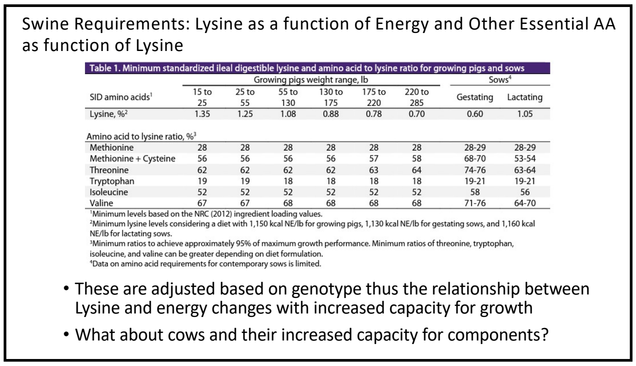


US Sire Protein Breeding Values over 51 years

42 years to go from -221 to 0
9 years to go from 0 to 94

4.6x faster rate of change

Dechow, 2023; https://webconnect.uscdcb.com/#/summary-stats/genetic-trend



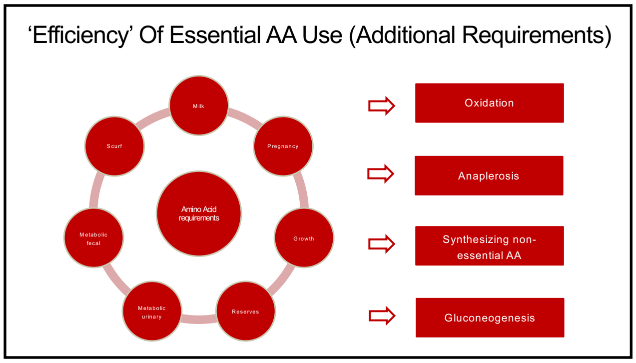
Swine Requirements: Lysine as a function of Energy and Other Essential AA as function of Lysine

Table 1. Minimum standardized ileal digestible lysine and amino acid to lysine ratio for growing pigs and sows

SID amino acids[1]	Growing pigs weight range, lb						Sows[4]	
	15 to 25	25 to 55	55 to 130	130 to 175	175 to 220	220 to 285	Gestating	Lactating
Lysine, %[2]	1.35	1.25	1.08	0.88	0.78	0.70	0.60	1.05
Amino acid to lysine ratio, %[3]								
Methionine	28	28	28	28	28	28	28-29	28-29
Methionine + Cysteine	56	56	56	56	57	58	68-70	53-54
Threonine	62	62	62	62	63	64	74-76	63-64
Tryptophan	19	19	18	18	18	18	19-21	19-21
Isoleucine	52	52	52	52	52	52	58	56
Valine	67	67	68	68	68	68	71-76	64-70

[1] Minimum levels based on the NRC (2012) ingredient loading values.
[2] Minimum lysine levels considering a diet with 1,150 kcal NE/lb for growing pigs, 1,130 kcal NE/lb for gestating sows, and 1,160 kcal NE/lb for lactating sows.
[3] Minimum ratios to achieve approximately 95% of maximum growth performance. Minimum ratios of threonine, tryptophan, isoleucine, and valine can be greater depending on diet formulation.
[4] Data on amino acid requirements for contemporary sows is limited.

- These are adjusted based on genotype thus the relationship between Lysine and energy changes with increased capacity for growth
- What about cows and their increased capacity for components?



‘Efficiency’ Of Essential AA Use (Additional Requirements)

⇨ Oxidation

⇨ Anaplerosis

⇨ Synthesizing non-essential AA

⇨ Gluconeogenesis

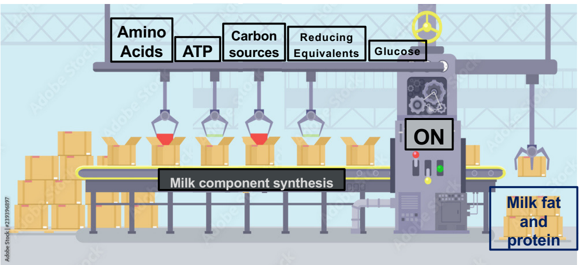
Protein-energy interactions

“Although it has been traditional to consider ‘protein’ and ‘energy’ metabolism as separate entities in mammalian metabolism, most scientists recognize this is an artificial divide. Indeed, they should be considered together as this reflects how nutrients are ingested and utilized as part of normal feeding patterns during evolution.”

Lobley, G. E. 2007. Protein-energy interactions: horizontal aspects. Pages 445-462 in Proc. Energy and protein metabolism and nutrition. Butterworths, Vichy, France.

The Conveyor Belt of Milk Component Production

- Meeting amino acid requirements improves overall nutrient and energy use efficiency for milk and component production

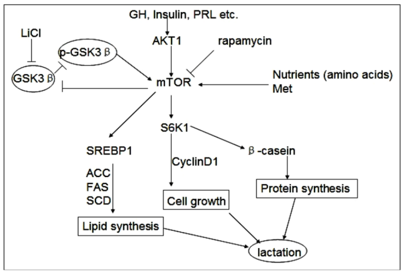


Nutrient signaling and metabolic flexibility in the mammary gland: Key to improved component yields?

Milk protein synthesis requires activation/repression of key metabolic pathways
- mTORC1 and AMPk pathways
  - Activated through hormone signaling (insulin, IGF-1), intracellular nutrients (AA supply; Leucine), and energy status (ATP:AMP ratio)
- Integrated stress response (ISR) pathway
  - Reduces cellular anabolic load in the presence of intracellular stress
  - Indirectly inhibited by insulin and IGF-1 and ATP status
- Unfolded protein response (UPR) pathway
  - Restores endoplasmic reticulum homeostasis through multiple cellular responses
  - Initiation causes direct phosphorylation of PERK → activation of ISR pathway

Optimal supply of AA with appropriate energy status → Maximized anabolic output

Pathways and Regulatory Signals for Regulation of Protein Synthesis in the Mammary Gland



https://doi.org/10.3390/molecules19079435

Insulin Effect on Milk Component Synthesis

- Proposed effect of insulin on milk fat synthesis:
  - Insulin-induced genes (INSIG2) responsive to insulin, effects translocation and activation of SREBP1 thus affecting the downstream FA genes and proteins (ACC, FAS, SCD, etc)
  - Insulin may regulate mTOR complex that affects downstream genes and proteins

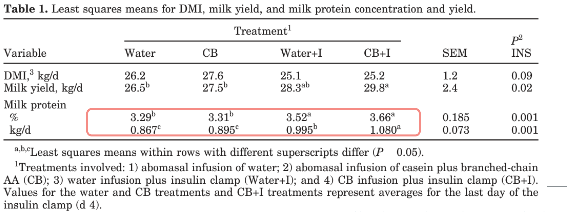


Table 1. Least squares means for DMI, milk yield, and milk protein concentration and yield.

	Treatment[1]					P^2
Variable	Water	CB	Water+I	CB+I	SEM	INS
DMI,[3] kg/d	26.2	27.6	25.1	25.2	1.2	0.09
Milk yield, kg/d	26.5^{b}	27.5^{b}	28.3^{ab}	29.8^{a}	2.4	0.02
Milk protein						
%	3.29^{b}	3.31^{b}	3.52^{a}	3.66^{a}	0.185	0.001
kg/d	0.867^{c}	0.895^{c}	0.995^{b}	1.080^{a}	0.073	0.001

[a,b,c]Least squares means within rows with different superscripts differ (P 0.05).

[1]Treatments involved: 1) abomasal infusion of water; 2) abomasal infusion of casein plus branched-chain AA (CB); 3) water infusion plus insulin clamp (Water+I); and 4) CB infusion plus insulin clamp (CB+I). Values for the water and CB treatments and CB+I treatments represent averages for the last day of the insulin clamp (d 4).

Mackle et al., 2000; Bionaz and Loor, 2008; Li et al., 2019

Mammary adaptability in varying nutrient supplies

Shifts in nutrient profile and supply → alterations in their efficient use according to mammary demand.

Extraction of BCAA changes across lactation

- Cellular maintenance and anabolic response (Mepham, 1982)

Lysine undergoes obligate catabolism in mammary (Lapierre, 2009)

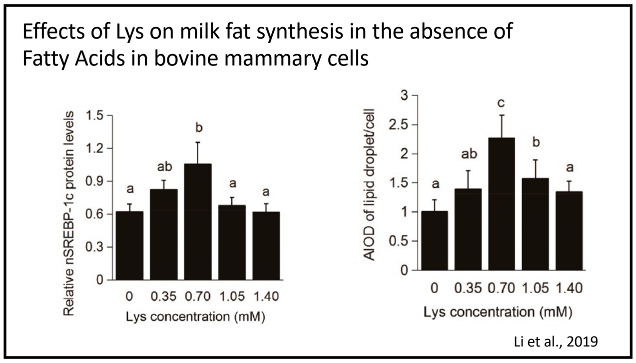
- Supplies N for NEAA synthesis
- Level of catabolism can shift in accordance with NEAA supply

Arginine is taken up in drastic excessive relative to milk protein output (~2.5x)

- Catabolism products include proline, ornithine, and urea (O'Quinn et al., 2002)
- Proline content in milk casein = 10.4% (2nd highest to glutamine)

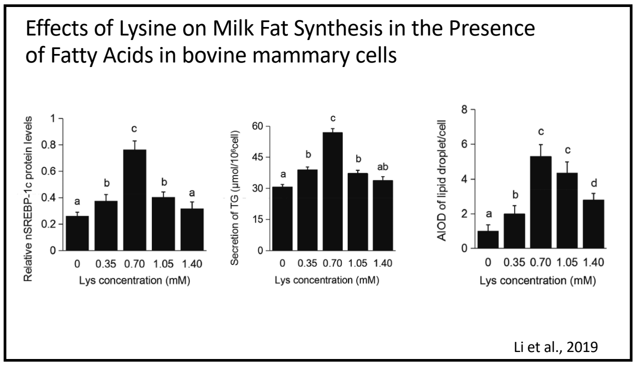
	AA Group (Mepham, 1982)		
	1	2	3
Amino Acid	**Histidine**	**Isoleucine**	**Alanine**
	Phenylalanine	**Leucine**	**Asparagine**
	Methionine	**Valine**	**Cysteine**
	Tyrosine	**Lysine**	**Glutamine**
	Tryptophan	**Arginine***	**Glycine**
		Threonine*	**Proline**
			Serine
Efficiency (AA –N uptake/AA-N Milk)	**1**	**> 1.15**	**< 1**

* Suggested group according to Lapierre et al. (2012)



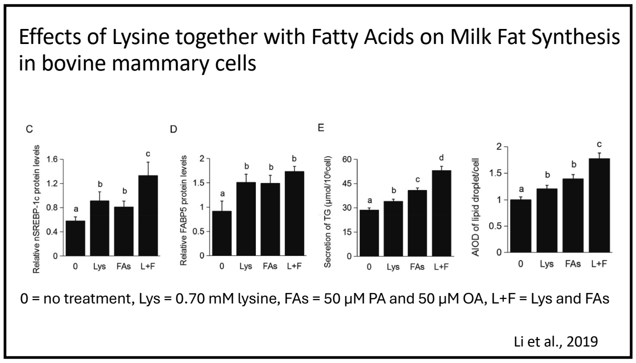
Effects of Lys on milk fat synthesis in the absence of Fatty Acids in bovine mammary cells

Li et al., 2019



Effects of Lysine on Milk Fat Synthesis in the Presence of Fatty Acids in bovine mammary cells

Li et al., 2019

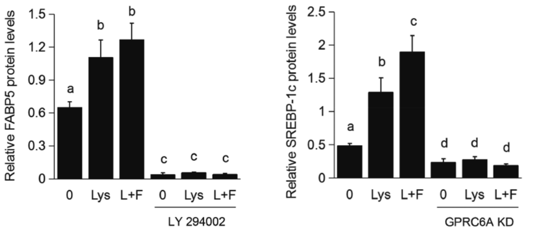


Effects of Lysine together with Fatty Acids on Milk Fat Synthesis in bovine mammary cells

0 = no treatment, Lys = 0.70 mM lysine, FAs = 50 μM PA and 50 μM OA, L+F = Lys and FAs

Li et al., 2019

Effects of PI3K Inhibition on Lysine Stimulated FABP5 Expression and SREBP-1c Expression and Maturation in bovine mammary cells



Li et al., 2019

Lysine and Milk Fat

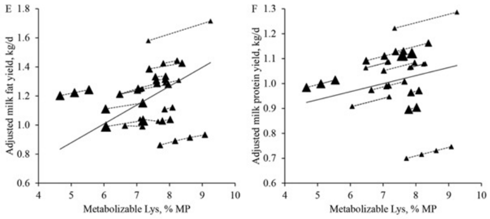
- In this study , using bovine mammary epithelial cells, Lysine-induced fatty acid-dependent SREBP-1c expression and maturation was used. SREBP-1c
- SREPB-1 is a key regulator of fatty acid synthesis in the mammary gland (Li et al., 2014) and is also sensitive to insulin
- This was done through regulation of theGPRC6A- the G protein-coupled receptor class 6A – which induces the PI3K/AkT (phosphatidy linositol 3-kinase) pathway
- FABP5 – Fatty acid binding protein 5 which regulates lipid metabolism

Effects of feeding rumen-protected lysine during the postpartum period on performance and amino acid profile in dairy cows: A meta-analysis

	Lysine % MP			
	6.5	**8.5**	**SEM**	**P**
Milk, kg	32.1	34.0	1.3	0.02
ECM, kg	33.4	35.8	1.6	0.03
Milk fat, %	3.68	3.73	0.12	0.07
Milk fat, kg	1.17	1.27	0.06	0.05
Milk protein, %	3.09	3.18	0.03	0.04
Milk protein, kg	0.99	1.06	0.05	0.07
Lactose, %	4.81	4.72	0.07	0.14

Arshad et al., 2024

Effects of feeding rumen-protected lysine during the postpartum period on performance and amino acid profile in dairy cows: A meta-analysis



Arshad et al., 2024

Amino Acids and De Novo FA Synthesis

- Lys increased enzymes related to de novo FA synthesis (ACS, ACC, FAS) through upregulation of FABP and SREBP1 (Li et al., 2019)
  - Further increased when supplemented with palmitic acid and oleic acid
- Additionally, Met and Leu increase expression of SREBP1– important regulator of enzymes for milk FA synthesis (Li et al., 2019).
- Arg increased de novo and mixed FA synthesis and expression of ACC, SCD, DGAT1 (Ding et al., 2022)

Fatty Acid Synthetase (FAS)

- FAS synthesizes de novo FA by elongating FA carbon chain
- Active sites with AA essential for function and transfer of intermediates during elongation of de novo FA
  - His, Lys, Ser, Cys (Smith et al., 2003; Wettstein-Knowles et al., 2005)
- FAS expression decreased in His- and Lys-deficient human liver cell medium (Dudek and Semenkovich, 1995)
  - This was reversible when His and Lys were reintroduced
- Expression of FAS increased by adding both NEAA and EAA compared each treatment individually (Fukuda and Iritani, 1986)
  - FAS complex likely has requirement for both types of AA

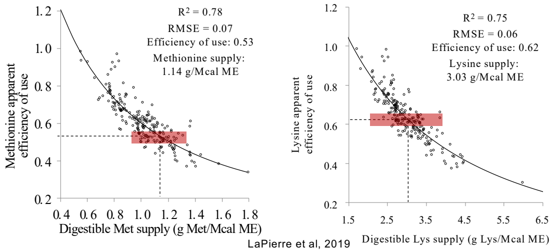
Optimum Supply Of Each EAA Relative To Metabolizable Energy – CNCPS v7.0

AA	R^2	Efficiency from our evaluation	Lapierre et al. (2007)	**g AA/ Mcal ME**	% EAA
Arg	0.81	0.61	0.58	**2.04**	10.2%
His	0.84	0.77	0.76	**0.91**	4.5%
Ile	0.74	0.67	0.67	**2.16**	10.8%
Leu	0.81	0.73	0.61	**3.42**	17.0%
Lys	0.75	0.67	0.69	**3.03**	15.1%
Met	0.79	0.57	0.66	**1.14**	5.7%
Phe	0.75	0.58	0.57	**2.15**	10.7%
Thr	0.75	0.59	0.66	**2.14**	10.7%
Trp	0.71	0.65	N/A	**0.59**	2.9%
Val	0.79	0.68	0.66	**2.48**	12.4%

Lys and Met requirements 14.9%, 5.1% - Schwab (1996) 2.9:1

Lys and Met requirements 14.7%, 5.3% - Rulquin et al. (1993) 2.77:1

Variation exists when contextualizing efficiency of use with amino acid and energy supply



LaPierre et al, 2019

Experimental design to test amino acid balancing

14-week longitudinal feeding trial

144 cows balanced in 9, **16-cow pens**

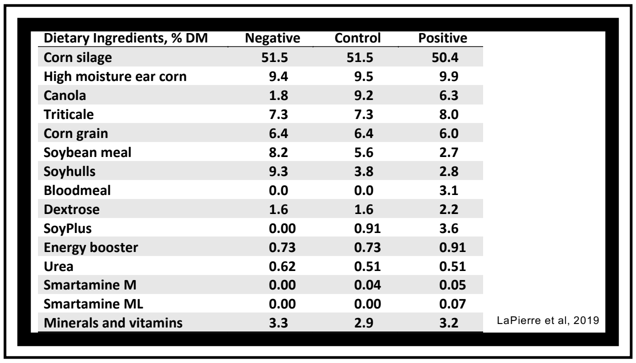
3 Diets formulated using CNCPS v.7:

1. Optimum g EAA/Mcal ME (14.8% CP) → **'Control'**
2. -1 Std Dev g EAA/Mcal ME (14.0% CP) → **'Negative'**
3. +1 Std Dev g EAA/Mcal ME (16.3% CP) → **'Positive'**

All diets formulated to be iso-caloric and in ME excess

- ☐ Nitrogen intestinal digestibility tested (Gutierrez-Botaro et al., 2022)
- ☐ Feed AA profile updated to refine supply (Van Amburgh et al., 2017)

LaPierre et al, 2019



Dietary Ingredients, % DM	Negative	Control	Positive
Corn silage	51.5	51.5	50.4
High moisture ear corn	9.4	9.5	9.9
Canola	1.8	9.2	6.3
Triticale	7.3	7.3	8.0
Corn grain	6.4	6.4	6.0
Soybean meal	8.2	5.6	2.7
Soyhulls	9.3	3.8	2.8
Bloodmeal	0.0	0.0	3.1
Dextrose	1.6	1.6	2.2
SoyPlus	0.00	0.91	3.6
Energy booster	0.73	0.73	0.91
Urea	0.62	0.51	0.51
Smartamine M	0.00	0.04	0.05
Smartamine ML	0.00	0.00	0.07
Minerals and vitamins	3.3	2.9	3.2

LaPierre et al, 2019

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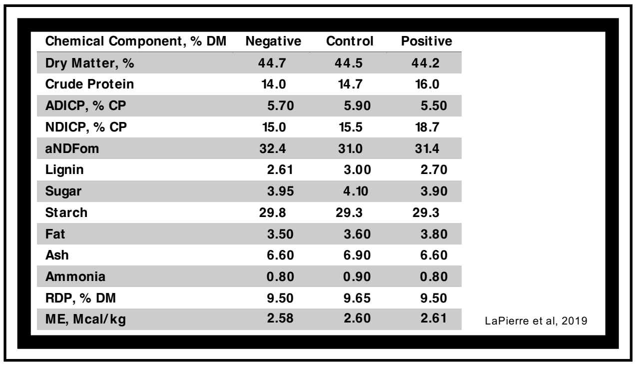

Chemical Component, % DM	Negative	Control	Positive
Dry Matter, %	44.7	44.5	44.2
Crude Protein	14.0	14.7	16.0
ADICP, % CP	5.70	5.90	5.50
NDICP, % CP	15.0	15.5	18.7
aNDFom	32.4	31.0	31.4
Lignin	2.61	3.00	2.70
Sugar	3.95	4.10	3.90
Starch	29.8	29.3	29.3
Fat	3.50	3.60	3.80
Ash	6.60	6.90	6.60
Ammonia	0.80	0.90	0.80
RDP, % DM	9.50	9.65	9.50
ME, Mcal/kg	2.58	2.60	2.61

LaPierre et al, 2019

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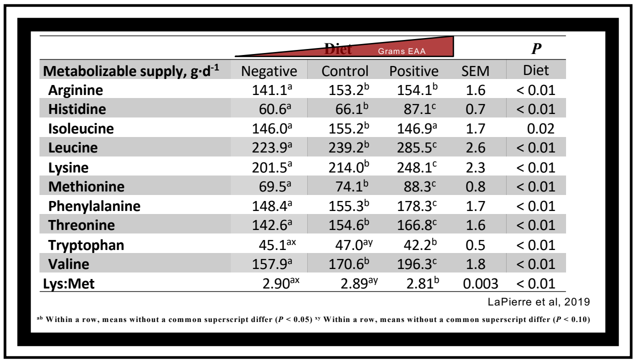

	Diet Grams EAA				*P*
Metabolizable supply, $g \cdot d^{-1}$	Negative	Control	Positive	SEM	Diet
Arginine	141.1^{a}	153.2^{b}	154.1^{b}	1.6	< 0.01
Histidine	60.6^{a}	66.1^{b}	87.1^{c}	0.7	< 0.01
Isoleucine	146.0^{a}	155.2^{b}	146.9^{a}	1.7	0.02
Leucine	223.9^{a}	239.2^{b}	285.5^{c}	2.6	< 0.01
Lysine	201.5^{a}	214.0^{b}	248.1^{c}	2.3	< 0.01
Methionine	69.5^{a}	74.1^{b}	88.3^{c}	0.8	< 0.01
Phenylalanine	148.4^{a}	155.3^{b}	178.3^{c}	1.7	< 0.01
Threonine	142.6^{a}	154.6^{b}	166.8^{c}	1.6	< 0.01
Tryptophan	45.1^{ax}	47.0^{ay}	42.2^{b}	0.5	< 0.01
Valine	157.9^{a}	170.6^{b}	196.3^{c}	1.8	< 0.01
Lys:Met	2.90^{ax}	2.89^{ay}	2.81^{b}	0.003	< 0.01

LaPierre et al, 2019

[ab] Within a row, means without a common superscript differ ($P < 0.05$) [xy] Within a row, means without a common superscript differ ($P < 0.10$)

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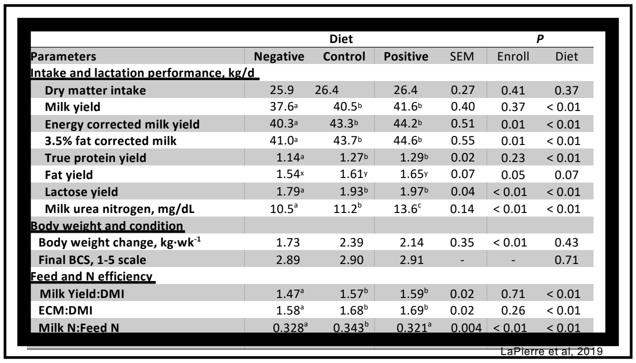
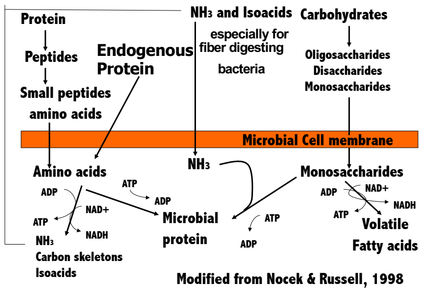


	Diet				P	
Parameters	**Negative**	**Control**	**Positive**	SEM	Enroll	Diet
Intake and lactation performance, kg/d						
Dry matter intake	25.9	26.4	26.4	0.27	0.41	0.37
Milk yield	37.6^a	40.5^b	41.6^b	0.40	0.37	< 0.01
Energy corrected milk yield	40.3^a	43.3^b	44.2^b	0.51	0.01	< 0.01
3.5% fat corrected milk	41.0^a	43.7^b	44.6^b	0.55	0.01	< 0.01
True protein yield	1.14^a	1.27^b	1.29^b	0.02	0.23	< 0.01
Fat yield	1.54^x	1.61^y	1.65^y	0.07	0.05	0.07
Lactose yield	1.79^a	1.93^b	1.97^b	0.04	< 0.01	< 0.01
Milk urea nitrogen, mg/dL	10.5^a	11.2^b	13.6^c	0.14	< 0.01	< 0.01
Body weight and condition						
Body weight change, kg·wk^{-1}	1.73	2.39	2.14	0.35	< 0.01	0.43
Final BCS, 1-5 scale	2.89	2.90	2.91	-	-	0.71
Feed and N efficiency						
Milk Yield:DMI	1.47^a	1.57^b	1.59^b	0.02	0.71	< 0.01
ECM:DMI	1.58^a	1.68^b	1.69^b	0.02	0.26	< 0.01
Milk N:Feed N	0.328^a	0.343^b	0.321^a	0.004	< 0.01	< 0.01

LaPierre et al, 2019

Protein and CHO To Optimize Microbial Yield



Modified from Nocek & Russell, 1998

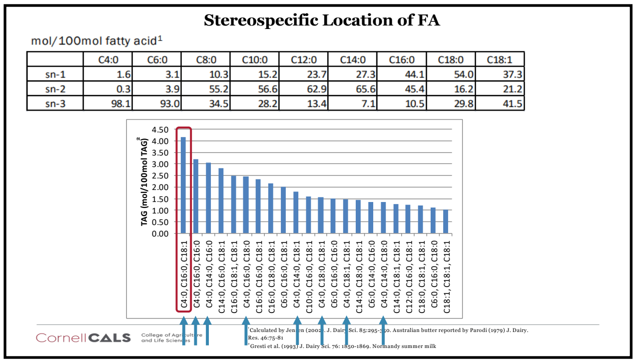
Fermentable Nonstructural Carbohydrates (NSC) to Optimize Microbial Yield and Milk Protein

Stage of lactation	Fermentable NSCHO, %DM	Fermentable starch, %DM	Fermentable sugar, %DM	Fermentable soluble fiber, %DM
Early	40-41	18.5 - 20	8	8
Peak	43	22 - 25	8	7
Mid	40	18.5 – 20.5	6	6

For high cows – 86% to 90% ruminal starch digestion

de Ondarza and Hoover: **Sugar in the 6% to 8% DM** range improved microbial yield and fiber digestion – likely due to protozoa. Higher sugar levels should be fed if starch levels are lower

Modified from Sniffen et al. and de Ondarza et al



Stereospecific Location of FA

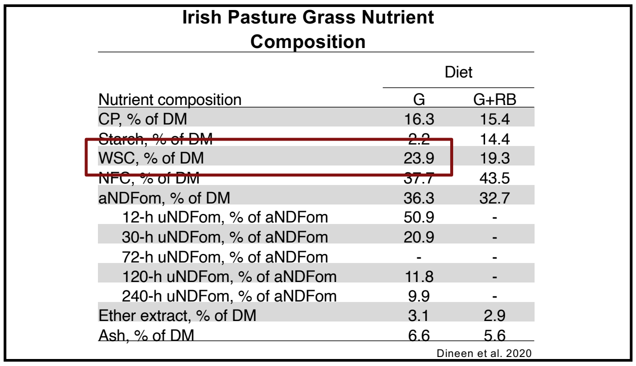
mol/100mol fatty acid[1]

	C4:0	C6:0	C8:0	C10:0	C12:0	C14:0	C16:0	C18:0	C18:1
sn-1	1.6	3.1	10.3	15.2	23.7	27.3	44.1	54.0	37.3
sn-2	0.3	3.9	55.2	56.6	62.9	65.6	45.4	16.2	21.2
sn-3	98.1	93.0	34.5	28.2	13.4	7.1	10.5	29.8	41.5

Cornell CALS College of Agriculture and Life Sciences

[1] Calculated by Jensen (2002). J. Dairy Sci. 85:295-350. Australian butter reported by Parodi (1979) J. Dairy. Res. 46:75-81

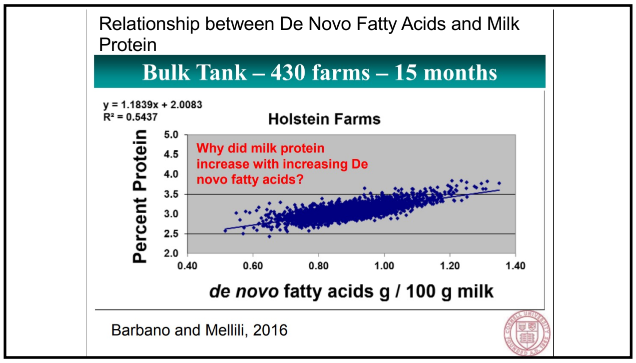
[2] Gresti et al. (1993) J. Dairy Sci. 76: 1850-1869. Normandy summer milk



Irish Pasture Grass Nutrient Composition

	Diet	
Nutrient composition	G	G+RB
CP, % of DM	16.3	15.4
Starch, % of DM	2.2	14.4
WSC, % of DM	23.9	19.3
NFC, % of DM	37.7	43.5
aNDFom, % of DM	36.3	32.7
12-h uNDFom, % of aNDFom	50.9	-
30-h uNDFom, % of aNDFom	20.9	-
72-h uNDFom, % of aNDFom	-	-
120-h uNDFom, % of aNDFom	11.8	-
240-h uNDFom, % of aNDFom	9.9	-
Ether extract, % of DM	3.1	2.9
Ash, % of DM	6.6	5.6

Dineen et al. 2020



Relationship between De Novo Fatty Acids and Milk Protein

Bulk Tank – 430 farms – 15 months

Why did milk protein increase with increasing De novo fatty acids?

Barbano and Mellili, 2016

Make Use of Fatty Acids

- Data emerging demonstrating that the profile of fatty acids at different stages of lactation impact insulin signaling
- Data from Lock et al and McFadden et al labs
- Implication is the cow has a FA requirement or a certain profile of FA improves energetic efficiency by altering partitioning of energy

Altering the ratio of dietary C16:0 (palmitic) and cis-9 C18:1 (oleic) impacts productivity

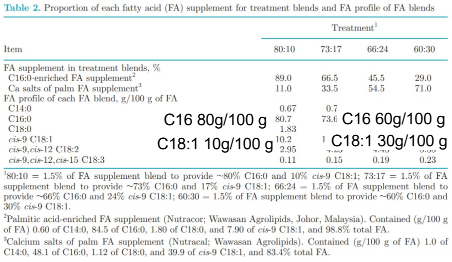


Table 2. Proportion of each fatty acid (FA) supplement for treatment blends and FA profile of FA blends

Item	Treatment[1]			
	80:10	73:17	66:24	60:30
FA supplement in treatment blends, %				
C16:0-enriched FA supplement[2]	89.0	66.5	45.5	29.0
Ca salts of palm FA supplement[3]	11.0	33.5	54.5	71.0
FA profile of each FA blend, g/100 g of FA				
C14:0	0.67	0.7	[illegible]	[illegible]
C16:0	80.7	73.6	[illegible]	[illegible]
C18:0	1.83	[illegible]	[illegible]	[illegible]
cis-9 C18:1	10.2	[illegible]	[illegible]	[illegible]
cis-9,*cis*-12 C18:2	2.95	[illegible]	[illegible]	[illegible]
cis-9,*cis*-12,*cis*-15 C18:3	0.11	0.15	0.19	0.23

C16 80g/100 g

C18:1 10g/100 g

C16 60g/100 g

C18:1 30g/100 g

[1] 80:10 = 1.5% of FA supplement blend to provide ~80% C16:0 and 10% *cis*-9 C18:1; 73:17 = 1.5% of FA supplement blend to provide ~73% C16:0 and 17% *cis*-9 C18:1; 66:24 = 1.5% of FA supplement blend to provide ~66% C16:0 and 24% *cis*-9 C18:1; 60:30 = 1.5% of FA supplement blend to provide ~60% C16:0 and 30% *cis*-9 C18:1.

[2] Palmitic acid-enriched FA supplement (Nutracor; Wawasan Agrolipids, Johor, Malaysia). Contained (g/100 g of FA) 0.60 of C14:0, 84.5 of C16:0, 1.80 of C18:0, and 7.90 of *cis*-9 C18:1, and 98.8% total FA.

[3] Calcium salts of palm FA supplement (Nutracal; Wawasan Agrolipids). Contained (g/100 g of FA) 1.0 of C14:0, 48.1 of C16:0, 1.12 of C18:0, and 39.9 of *cis*-9 C18:1, and 83.4% total FA.

De Souza et al., 2019 JDS

Diet and Ratio of Palmitic to Oleic Acids

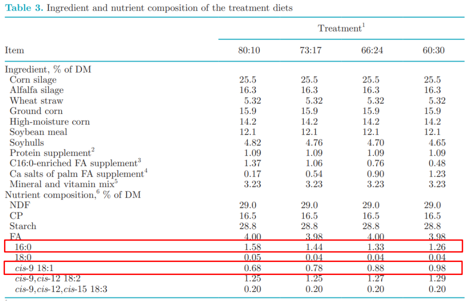


Table 3. Ingredient and nutrient composition of the treatment diets

Item	Treatment[1]			
	80:10	73:17	66:24	60:30
Ingredient, % of DM				
Corn silage	25.5	25.5	25.5	25.5
Alfalfa silage	16.3	16.3	16.3	16.3
Wheat straw	5.32	5.32	5.32	5.32
Ground corn	15.9	15.9	15.9	15.9
High-moisture corn	14.2	14.2	14.2	14.2
Soybean meal	12.1	12.1	12.1	12.1
Soyhulls	4.82	4.76	4.70	4.65
Protein supplement[2]	1.09	1.09	1.09	1.09
C16:0-enriched FA supplement[3]	1.37	1.06	0.76	0.48
Ca salts of palm FA supplement[4]	0.17	0.54	0.90	1.23
Mineral and vitamin mix[5]	3.23	3.23	3.23	3.23
Nutrient composition,[6] % of DM				
NDF	29.0	29.0	29.0	29.0
CP	16.5	16.5	16.5	16.5
Starch	28.8	28.8	28.8	28.8
FA	4.00	3.98	4.00	3.98
16:0	1.58	1.44	1.33	1.26
18:0	0.05	0.04	0.04	0.04
cis-9 18:1	0.68	0.78	0.88	0.98
cis-9,*cis*-12 18:2	1.25	1.25	1.27	1.29
cis-9,*cis*-12,*cis*-15 18:3	0.20	0.20	0.20	0.20

De Souza et al., 2019 JDS

Effect of Ratio of Palmitic to Oleic on Productivity

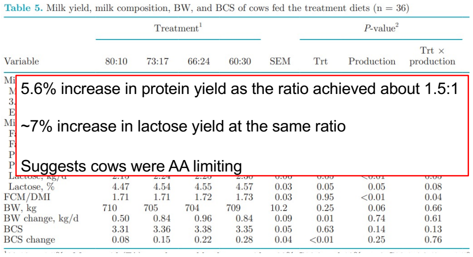


Table 5. Milk yield, milk composition, BW, and BCS of cows fed the treatment diets (n = 36)

	Treatment[1]					P-value[2]		
Variable	80:10	73:17	66:24	60:30	SEM	Trt	Production	Trt × production
Lactose, %	4.47	4.54	4.55	4.57	0.03	0.05	0.05	0.08
FCM/DMI	1.71	1.71	1.72	1.73	0.03	0.95	<0.01	0.04
BW, kg	710	705	704	709	10.2	0.25	0.06	0.66
BW change, kg/d	0.50	0.84	0.96	0.84	0.09	0.01	0.74	0.61
BCS	3.31	3.36	3.38	3.35	0.05	0.63	0.14	0.13
BCS change	0.08	0.15	0.22	0.28	0.04	<0.01	0.25	0.76

5.6% increase in protein yield as the ratio achieved about 1.5:1

~7% increase in lactose yield at the same ratio

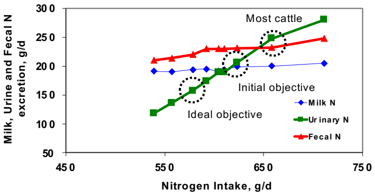
Suggests cows were AA limiting

De Souza et al., 2019 JDS

Take Home

- Cows have requirements for fatty acids like they do for amino acids – we just haven't figured it out yet
- It looks like when we feed a certain ratio of palmitic (16:0) to oleic (C18:1) the efficiency of use of absorbed nutrients increases
- 1.5:1 for Palmitic:Oleic and this is for intake
- For example, if you are supplying 280 g C16:0, you should formulate about 180 g of C18:1 to optimize the component response

Nitrogen excretion in diets varying in dietary nitrogen



Milk Nitrogen: ~200 g or 1.28 kg (2.81 lb) protein

Urinary N:

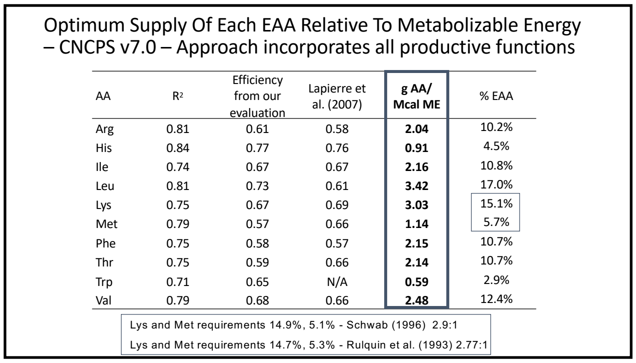
Most Cattle: ~250 g
Initial Objective: ~200 g
Ideal Objective : ~150 g

Milk N:Urinary N

Most Cattle: 0.8
Initial Objective: 1.0
Ideal Objective : 1.3

Metrics can be used as a proxy for improvements of Productive N:Urinary N

Van Amburgh et al. 2015 J. Dairy Sci



Optimum Supply Of Each EAA Relative To Metabolizable Energy – CNCPS v7.0 – Approach incorporates all productive functions

AA	R^2	Efficiency from our evaluation	Lapierre et al. (2007)	**g AA/ Mcal ME**	% EAA
Arg	0.81	0.61	0.58	**2.04**	10.2%
His	0.84	0.77	0.76	**0.91**	4.5%
Ile	0.74	0.67	0.67	**2.16**	10.8%
Leu	0.81	0.73	0.61	**3.42**	17.0%
Lys	0.75	0.67	0.69	**3.03**	15.1%
Met	0.79	0.57	0.66	**1.14**	5.7%
Phe	0.75	0.58	0.57	**2.15**	10.7%
Thr	0.75	0.59	0.66	**2.14**	10.7%
Trp	0.71	0.65	N/A	**0.59**	2.9%
Val	0.79	0.68	0.66	**2.48**	12.4%

Lys and Met requirements 14.9%, 5.1% - Schwab (1996) 2.9:1

Lys and Met requirements 14.7%, 5.3% - Rulquin et al. (1993) 2.77:1

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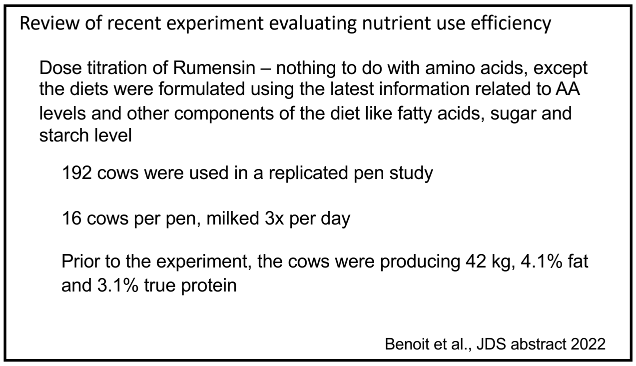

Review of recent experiment evaluating nutrient use efficiency

Dose titration of Rumensin – nothing to do with amino acids, except the diets were formulated using the latest information related to AA levels and other components of the diet like fatty acids, sugar and starch level

192 cows were used in a replicated pen study

16 cows per pen, milked 3x per day

Prior to the experiment, the cows were producing 42 kg, 4.1% fat and 3.1% true protein

Benoit et al., JDS abstract 2022

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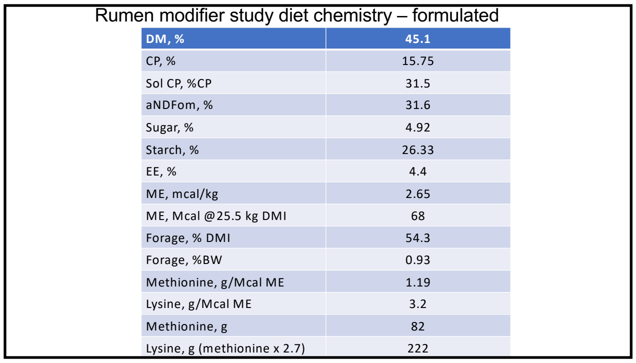

Rumen modifier study diet chemistry – formulated

DM, %	45.1
CP, %	15.75
Sol CP, %CP	31.5
aNDFom, %	31.6
Sugar, %	4.92
Starch, %	26.33
EE, %	4.4
ME, mcal/kg	2.65
ME, Mcal @25.5 kg DMI	68
Forage, % DMI	54.3
Forage, %BW	0.93
Methionine, g/Mcal ME	1.19
Lysine, g/Mcal ME	3.2
Methionine, g	82
Lysine, g (methionine x 2.7)	222

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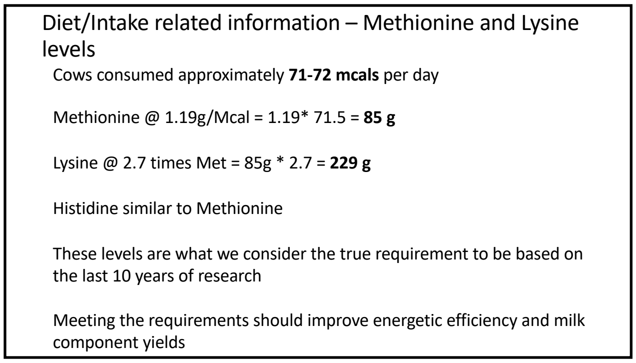


Diet/Intake related information – Methionine and Lysine levels

Cows consumed approximately **71-72 mcals** per day

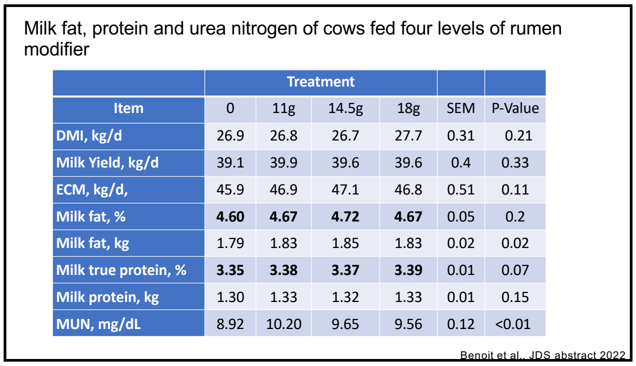
Methionine @ 1.19g/Mcal = 1.19* 71.5 = **85 g**

Lysine @ 2.7 times Met = 85g * 2.7 = **229 g**

Histidine similar to Methionine

These levels are what we consider the true requirement to be based on the last 10 years of research

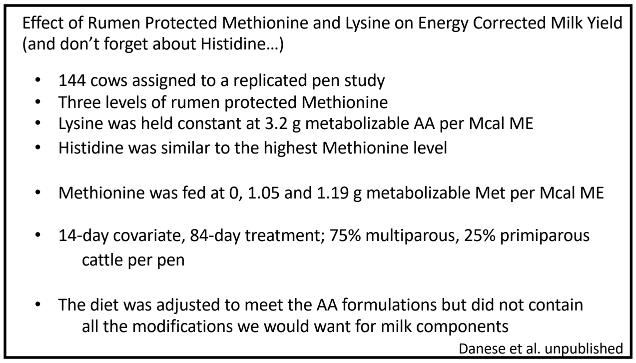
Meeting the requirements should improve energetic efficiency and milk component yields



Milk fat, protein and urea nitrogen of cows fed four levels of rumen modifier

	Treatment					
Item	0	11g	14.5g	18g	SEM	P-Value
DMI, kg/d	26.9	26.8	26.7	27.7	0.31	0.21
Milk Yield, kg/d	39.1	39.9	39.6	39.6	0.4	0.33
ECM, kg/d,	45.9	46.9	47.1	46.8	0.51	0.11
Milk fat, %	**4.60**	**4.67**	**4.72**	**4.67**	0.05	0.2
Milk fat, kg	1.79	1.83	1.85	1.83	0.02	0.02
Milk true protein, %	**3.35**	**3.38**	**3.37**	**3.39**	0.01	0.07
Milk protein, kg	1.30	1.33	1.32	1.33	0.01	0.15
MUN, mg/dL	8.92	10.20	9.65	9.56	0.12	<0.01

Benoit et al., JDS abstract 2022



Effect of Rumen Protected Methionine and Lysine on Energy Corrected Milk Yield (and don't forget about Histidine…)

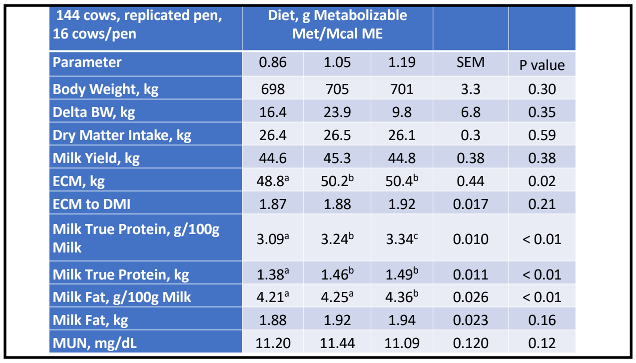
- 144 cows assigned to a replicated pen study
- Three levels of rumen protected Methionine
- Lysine was held constant at 3.2 g metabolizable AA per Mcal ME
- Histidine was similar to the highest Methionine level

- Methionine was fed at 0, 1.05 and 1.19 g metabolizable Met per Mcal ME

- 14-day covariate, 84-day treatment; 75% multiparous, 25% primiparous cattle per pen

- The diet was adjusted to meet the AA formulations but did not contain all the modifications we would want for milk components

Danese et al. unpublished



144 cows, replicated pen, 16 cows/pen	Diet, g Metabolizable Met/Mcal ME				
Parameter	0.86	1.05	1.19	SEM	P value
Body Weight, kg	698	705	701	3.3	0.30
Delta BW, kg	16.4	23.9	9.8	6.8	0.35
Dry Matter Intake, kg	26.4	26.5	26.1	0.3	0.59
Milk Yield, kg	44.6	45.3	44.8	0.38	0.38
ECM, kg	48.8^{a}	50.2^{b}	50.4^{b}	0.44	0.02
ECM to DMI	1.87	1.88	1.92	0.017	0.21
Milk True Protein, g/100g Milk	3.09^{a}	3.24^{b}	3.34^{c}	0.010	< 0.01
Milk True Protein, kg	1.38^{a}	1.46^{b}	1.49^{b}	0.011	< 0.01
Milk Fat, g/100g Milk	4.21^{a}	4.25^{a}	4.36^{b}	0.026	< 0.01
Milk Fat, kg	1.88	1.92	1.94	0.023	0.16
MUN, mg/dL	11.20	11.44	11.09	0.120	0.12

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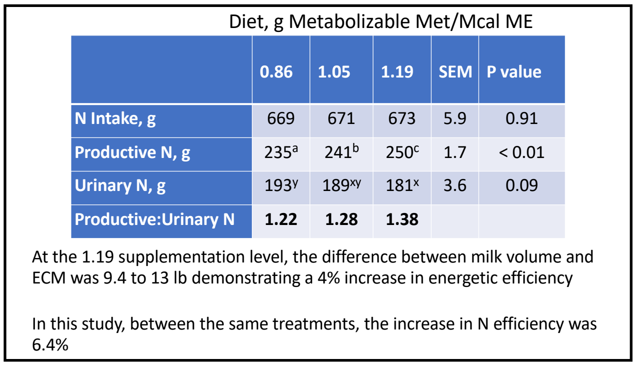

Diet, g Metabolizable Met/Mcal ME

	0.86	1.05	1.19	SEM	P value
N Intake, g	669	671	673	5.9	0.91
Productive N, g	235^{a}	241^{b}	250^{c}	1.7	< 0.01
Urinary N, g	193^{y}	189^{xy}	181^{x}	3.6	0.09
Productive:Urinary N	**1.22**	**1.28**	**1.38**		

At the 1.19 supplementation level, the difference between milk volume and ECM was 9.4 to 13 lb demonstrating a 4% increase in energetic efficiency

In this study, between the same treatments, the increase in N efficiency was 6.4%

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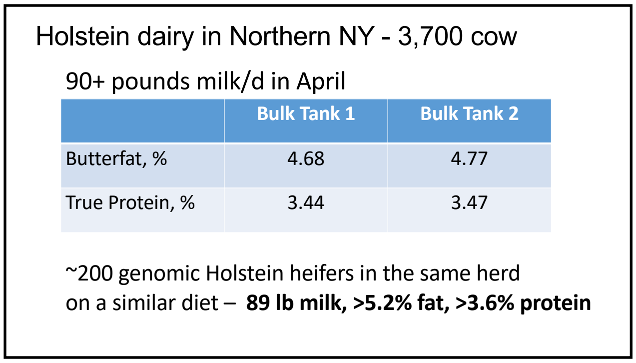

Holstein dairy in Northern NY - 3,700 cow

90+ pounds milk/d in April

	Bulk Tank 1	Bulk Tank 2
Butterfat, %	4.68	4.77
True Protein, %	3.44	3.47

~200 genomic Holstein heifers in the same herd on a similar diet – **89 lb milk, >5.2% fat, >3.6% protein**

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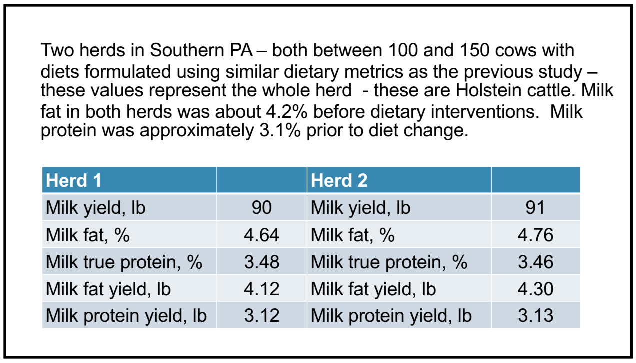
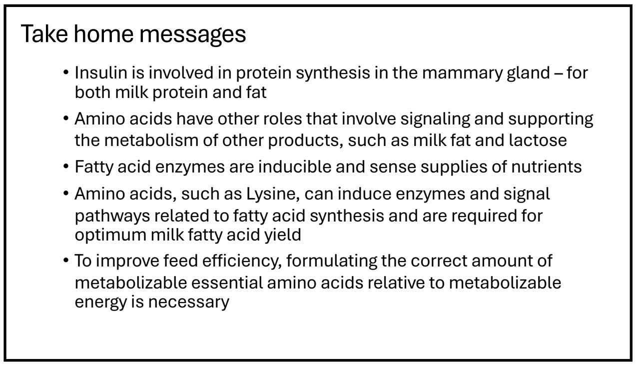


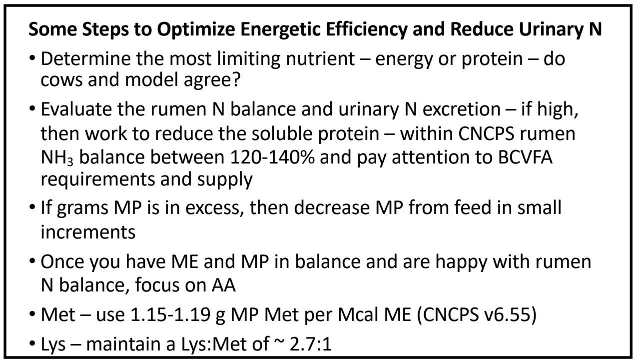
Two herds in Southern PA – both between 100 and 150 cows with diets formulated using similar dietary metrics as the previous study – these values represent the whole herd - these are Holstein cattle. Milk fat in both herds was about 4.2% before dietary interventions. Milk protein was approximately 3.1% prior to diet change.

Herd 1		Herd 2	
Milk yield, lb	90	Milk yield, lb	91
Milk fat, %	4.64	Milk fat, %	4.76
Milk true protein, %	3.48	Milk true protein, %	3.46
Milk fat yield, lb	4.12	Milk fat yield, lb	4.30
Milk protein yield, lb	3.12	Milk protein yield, lb	3.13



Take home messages

- Insulin is involved in protein synthesis in the mammary gland – for both milk protein and fat
- Amino acids have other roles that involve signaling and supporting the metabolism of other products, such as milk fat and lactose
- Fatty acid enzymes are inducible and sense supplies of nutrients
- Amino acids, such as Lysine, can induce enzymes and signal pathways related to fatty acid synthesis and are required for optimum milk fatty acid yield
- To improve feed efficiency, formulating the correct amount of metabolizable essential amino acids relative to metabolizable energy is necessary



Some Steps to Optimize Energetic Efficiency and Reduce Urinary N

- Determine the most limiting nutrient – energy or protein – do cows and model agree?
- Evaluate the rumen N balance and urinary N excretion – if high, then work to reduce the soluble protein – within CNCPS rumen NH_3 balance between 120-140% and pay attention to BCVFA requirements and supply
- If grams MP is in excess, then decrease MP from feed in small increments
- Once you have ME and MP in balance and are happy with rumen N balance, focus on AA
- Met – use 1.15-1.19 g MP Met per Mcal ME (CNCPS v6.55)
- Lys – maintain a Lys:Met of ~ 2.7:1

Some Steps to Optimize Energetic Efficiency and Reduce Urinary N

- Pay attention to aNDFom digestibility and allocate the highest digestibility forages to the fresh and high cows
- With forages you want the lowest uNDF pool as possible to maximize the digestible aNDFom
- Don't overfeed starch or fatty acids and add some sugar – need butyrate
- Formulate sugar at 5% to 8% DM
- Good rumen digestible starch sources in the 25-27% DM range
- Ether extract 4.4-4.7% and work towards a 1.5:1 relationship between palmitic and oleic

Formulation considerations for component yields

Pools	
Sugars	5 to 7% DM
Starch	26-27% and 90% ruminal digestibility
aNDFom	30-32% DM and >67% ruminal digestibility at 30 h uNDF as low as possible
Fatty acids	Less than 4.5%
Fatty acids	1.5:1 Palmitic:oleic
Amino acids	Met 1.19 g/Mcal ME Lys 3.21 g/Mcal ME or 2.7x Met

Thank you for your attention, for everyone who helped develop this work, and for the sponsors who keep it going.

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