A vision on sustainability in the formulation of AA and proteins for dogs and cats

Anna Kate Shoveller, PhD, PAS
Department of Animal Biosciences, University of Guelph, Canada

Protein quality

Protein quality is dependent upon:

1. Protein content
2. Amino acid composition
3. Protein digestibility and amino acid availability

compared to the requirements of the species of interest.

Study	Animals	Diet approach	Outcome	> 14 weeks NRC (MR)	Adult maintenance AAFCO (min)
Methionine and cysteine	Puppies and/or growing dogs = **OVER**	Titration with synthetic or semi-synthetic diets = **UNDER**	Weight gain or nitrogen retention = **UNDER**	2.1 g/kg for both Met and Cys	3.3 g/kg Met and 3.2 g/kg Cys
Lysine				5.6 g/kg	6.3 g/kg
Isoleucine				4.0 g/kg	3.8 g/kg
Leucine				6.5 g/kg	6.8 g/kg

Human indispensable amino acid requirements

- Current protein recommendation: EAR= 0.66 g/kg/day; RDA= 0.8 g/kg/day
  - Data based on nitrogen balance method (since 1985)
- Indispensable amino acid recommendation increased from 1985 to 2005/2007 guidelines

	1985	2005/2007	
Amino Acids	Adults	EAR (mg/kg/d)	RDA (mg/kg/d)
Histidine	8	11	14
Isoleucine	10	15	19
Leucine	14	34	42
Lysine	12	31	38
Methionine+ Cysteine	13	15	19
Phenylalanine+ Tyrosine	14	27	33
Threonine	7	16	20
Tryptophan	3.5	4	5
Valine	10	19	24

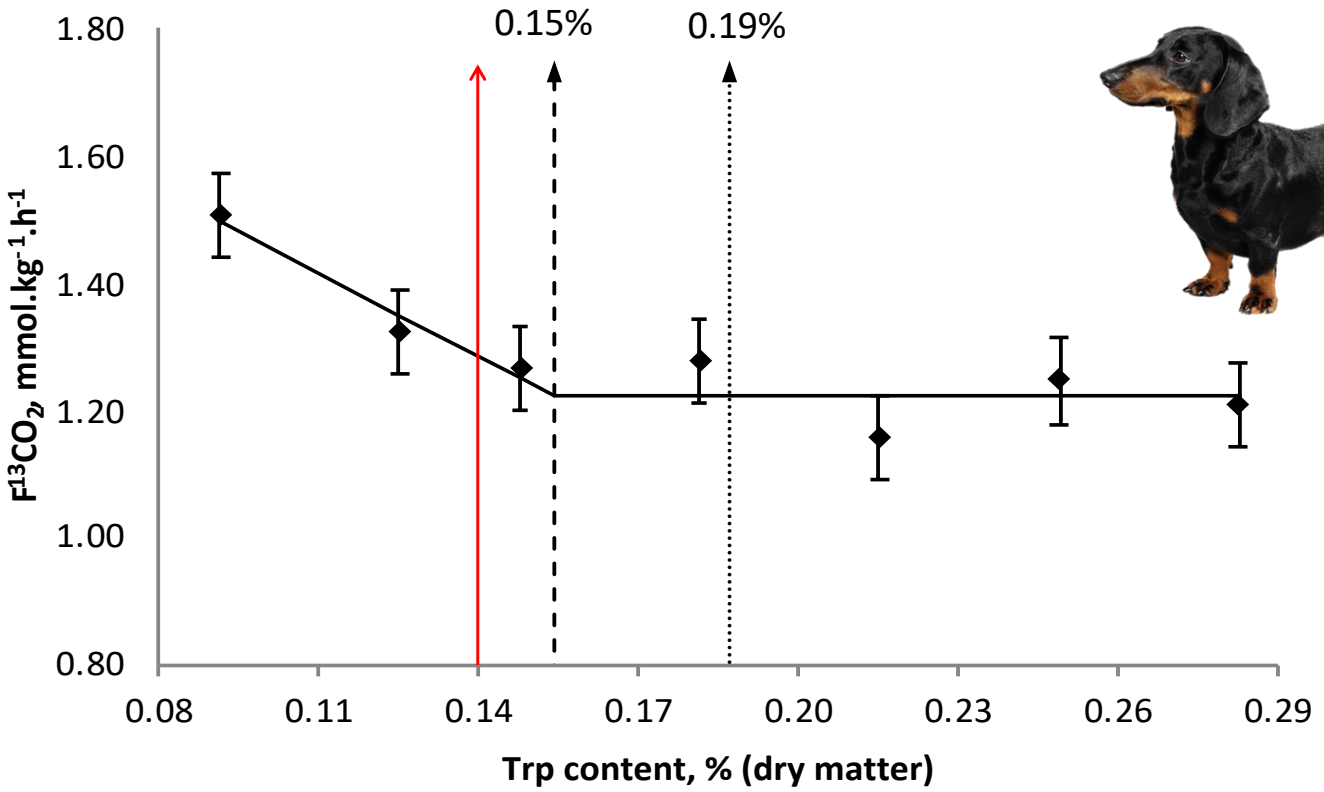
(Institute of Medicine, 2005), FAO/WHO, 2007), (National Academies Press, 1985)

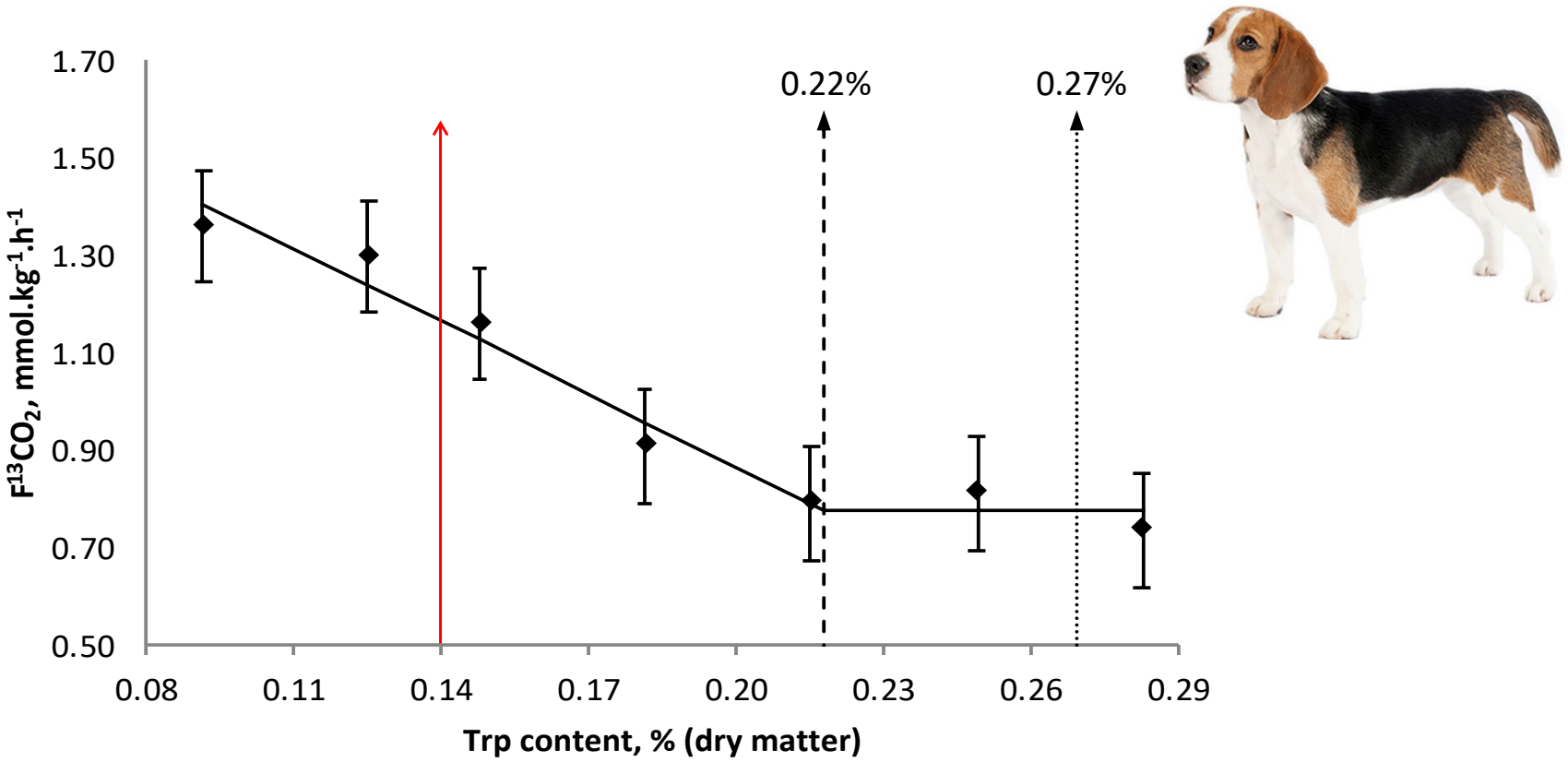
Which life stage? Which life style? Which breed?

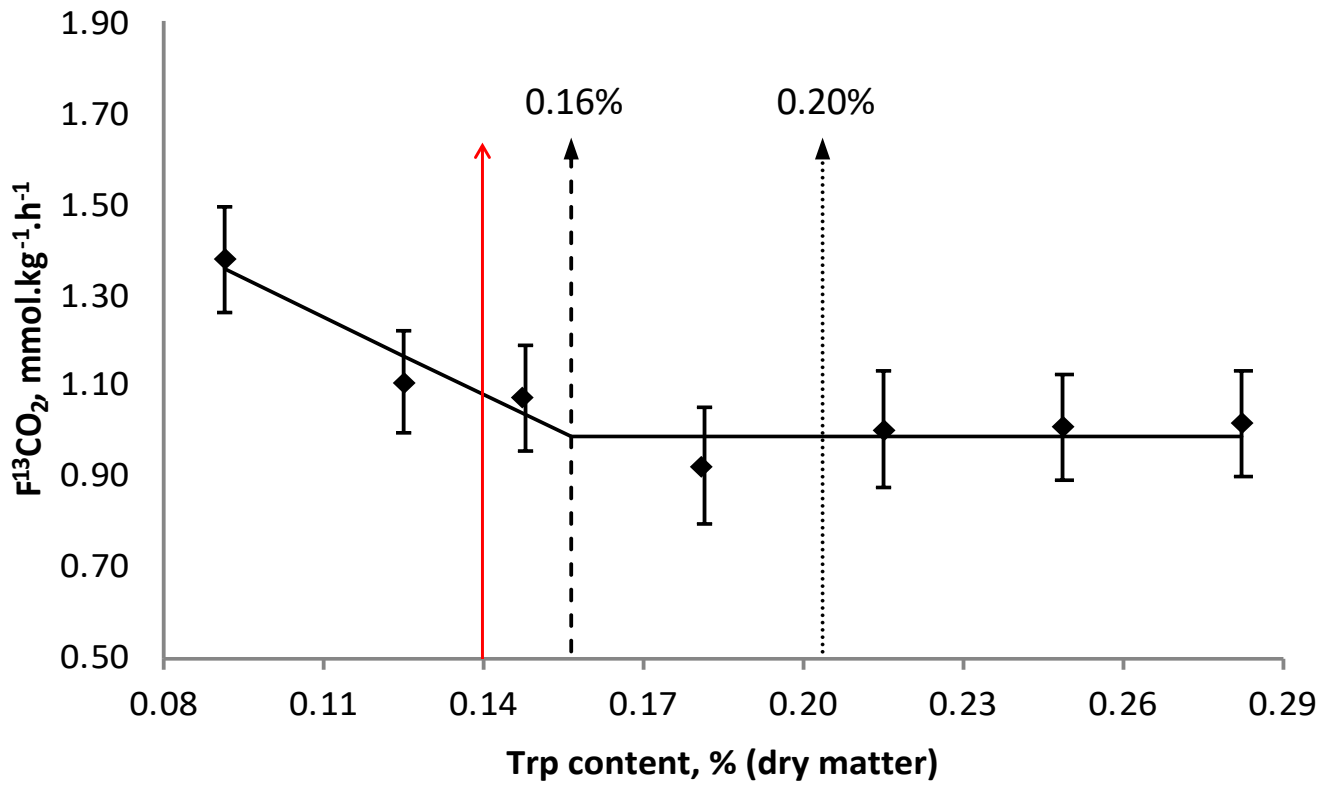
Begging the question: Who do we target? What do we need to know to make good decisions?

For these reasons I would like to echo the concluding remarks of Tang et al. (2014) regarding the IAAO technique in that “the limitations of this short-term, noninvasive method underscore the need for new research that uses alternative experimental designs and measuring physiologic, morphologic, and health-related outcomes.” *Juan Marini, 2015, Commentary in Journal of Nutrition*

Tryptophan


0.15%
0.19%
1.80
1.60
1.40
1.20
1.00
0.80
$F^{13}CO_2$, mmol.kg^{-1}.h^{-1}
0.08
0.11
0.14
0.17
0.20
0.23
0.26
0.29
Trp content, % (dry matter)


1.70
1.50
1.30
1.10
0.90
0.70
0.50
$F^{13}CO_2$, mmol.kg^{-1}.h^{-1}
0.22%
0.27%
0.08
0.11
0.14
0.17
0.20
0.23
0.26
0.29
Trp content, % (dry matter)


1.90
1.70
1.50
1.30
1.10
0.90
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0.50
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Trp content, % (dry matter)

Amino acid requirements (g/100g DM)

	AAFCO/NRC RA	Small	Medium	Large
Phenylalanine (Shoveller et al, 2018 JAS)	0.44/ 0.45	0.39	0.40	0.50
Tryptophan (Templeman et al, 2019 JAS)	0.16/0.14	0.18	0.26	0.20
Threonine (Mansilla et al, 2020 JAS)	0.48/0.43	--	0.60	0.57
Lysine (Sutherland et al, 2020, TAS)	0.63/0.35	--	0.58	
Methionine (Mansilla et al, 2020, JAS)	0.33/0.33	0.34	0.50	0.57

Tryptophan ratios

For 11 weeks, dogs were fed either:

Control: fed a basal diet (Trp:LNAA ratio of 0.047:1)

Treatment: fed the basal diet with a dietary Trp supplementation (Trp:LNAA ratio of 0.075:1)

JOURNAL ARTICLE

Effects of incremental exercise and dietary tryptophan supplementation on the amino acid metabolism, serotonin status, stool quality, fecal metabolites, and body composition of mid-distance training sled dogs Get access

James R Templeman, Emma Thornton, Cara Cargo-Froom, Eli J Squires, Kelly S Swanson, Anna K Shoveller

Journal of Animal Science, Volume 98, Issue 5, May 2020, skaa128, https://doi.org/10.1093/jas/skaa128

PLOS ONE

OPEN ACCESS PEER-REVIEWED

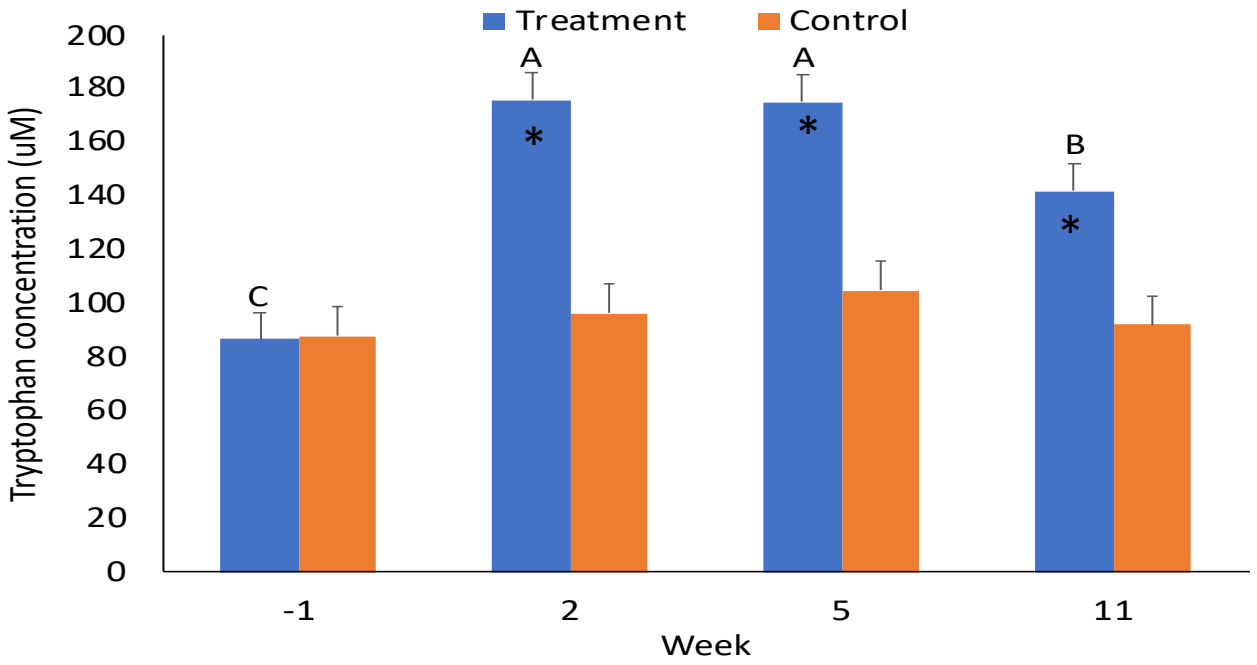
RESEARCH ARTICLE

Investigating the effects of incremental conditioning and supplemental dietary tryptophan on the voluntary activity and behaviour of mid-distance training sled dogs

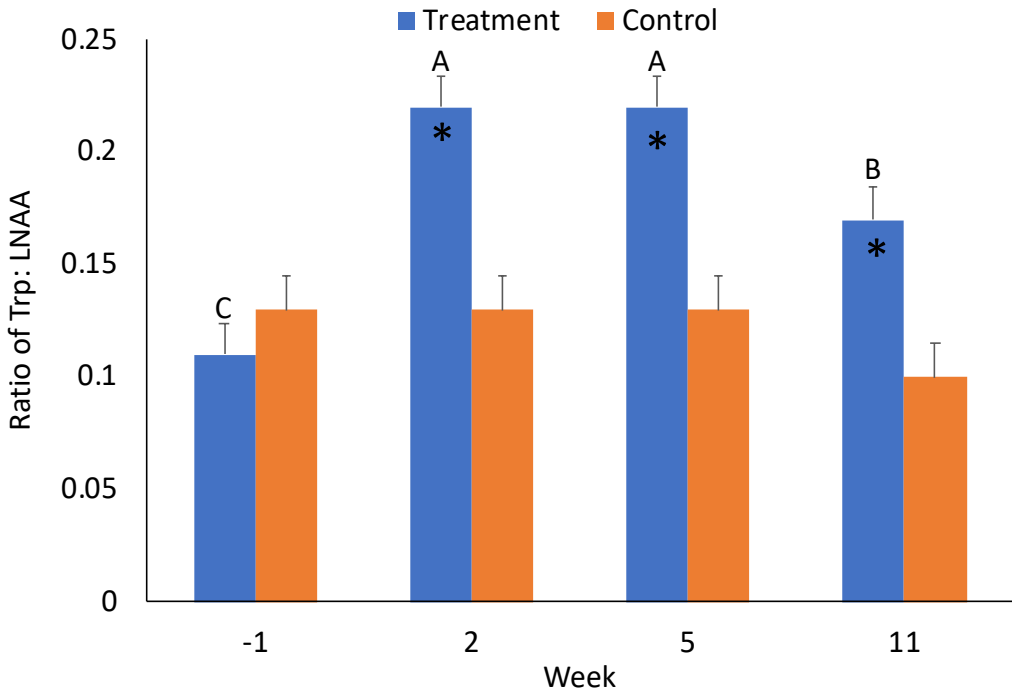
Eve Robinson, James R. Templeman, Emma Thornton, Candace C. Croney, Lee Niel, Anna Kate Shoveller

Published: August 13, 2020 • https://doi.org/10.1371/journal.pone.0232643

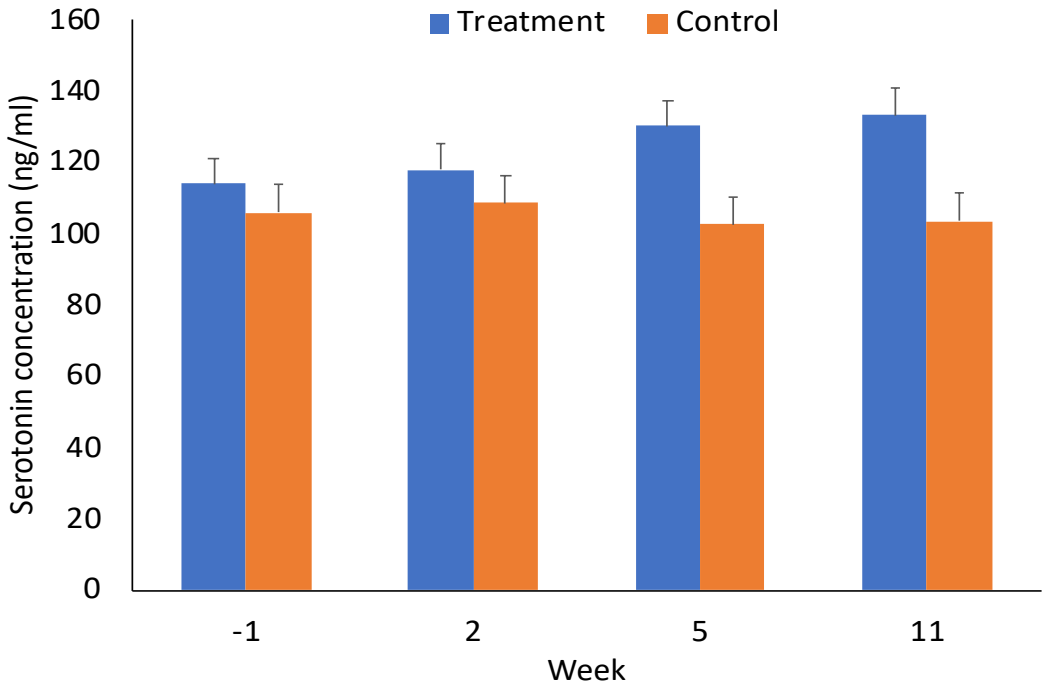
Serum tryptophan concentrations at 1 hour after feeding



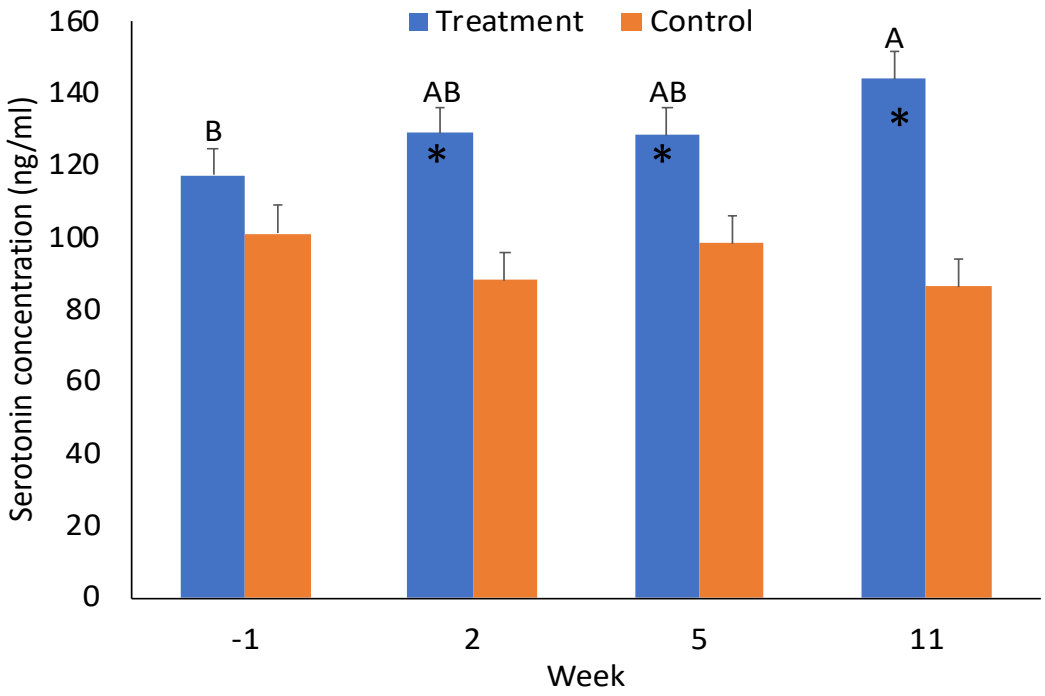
Ratio of tryptophan to LNAA in serum at 1 hour after feeding



Serum serotonin concentrations at 1 hour after feeding



Serum serotonin concentrations at 4 hours after feeding

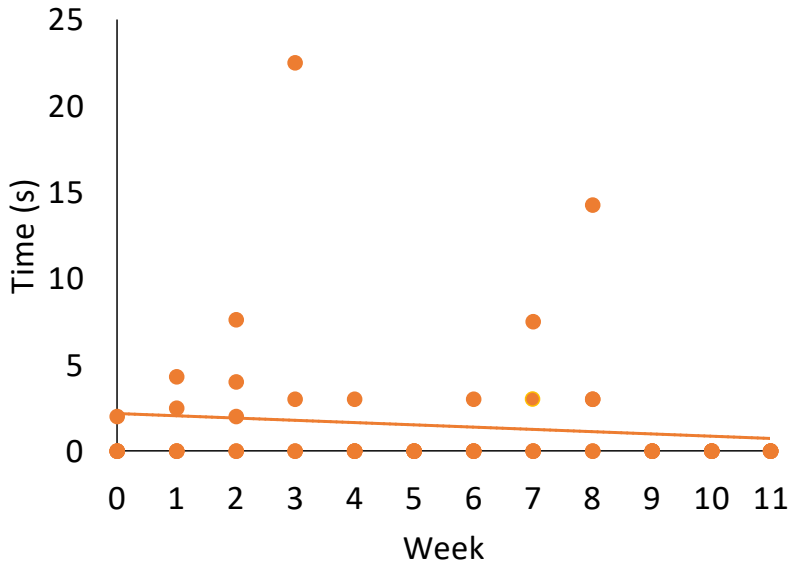


Behavioural Evaluation

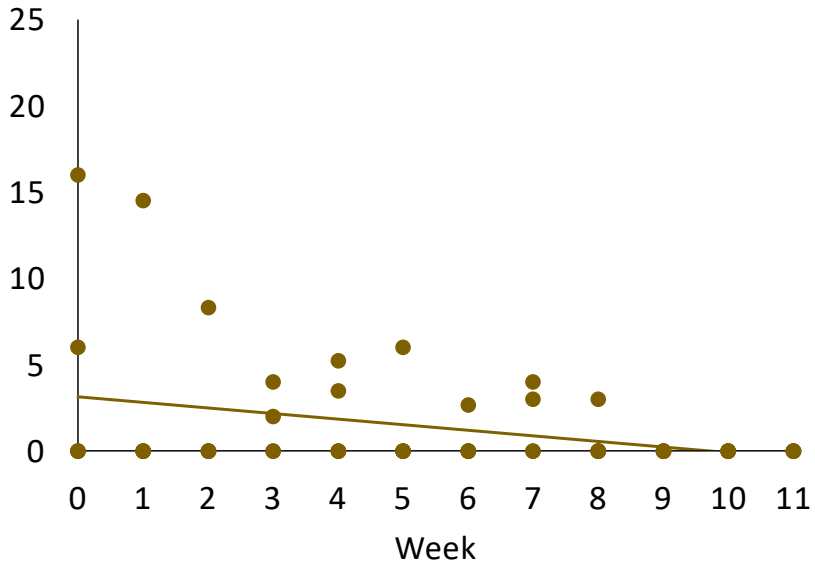
Step-up exercise protocol of dogs, km ran

- Dogs were recorded for 5 minutes prior to exercise, and for 5 minutes immediately post exercise
- Quantified the time each dog spent performing:
  - **Agonistic behaviours**, chewing on the gangline, digging, jumping, lunging, changing posture, sitting, standing and lying down

Tryptophan decreased pre-run agonistic behaviours



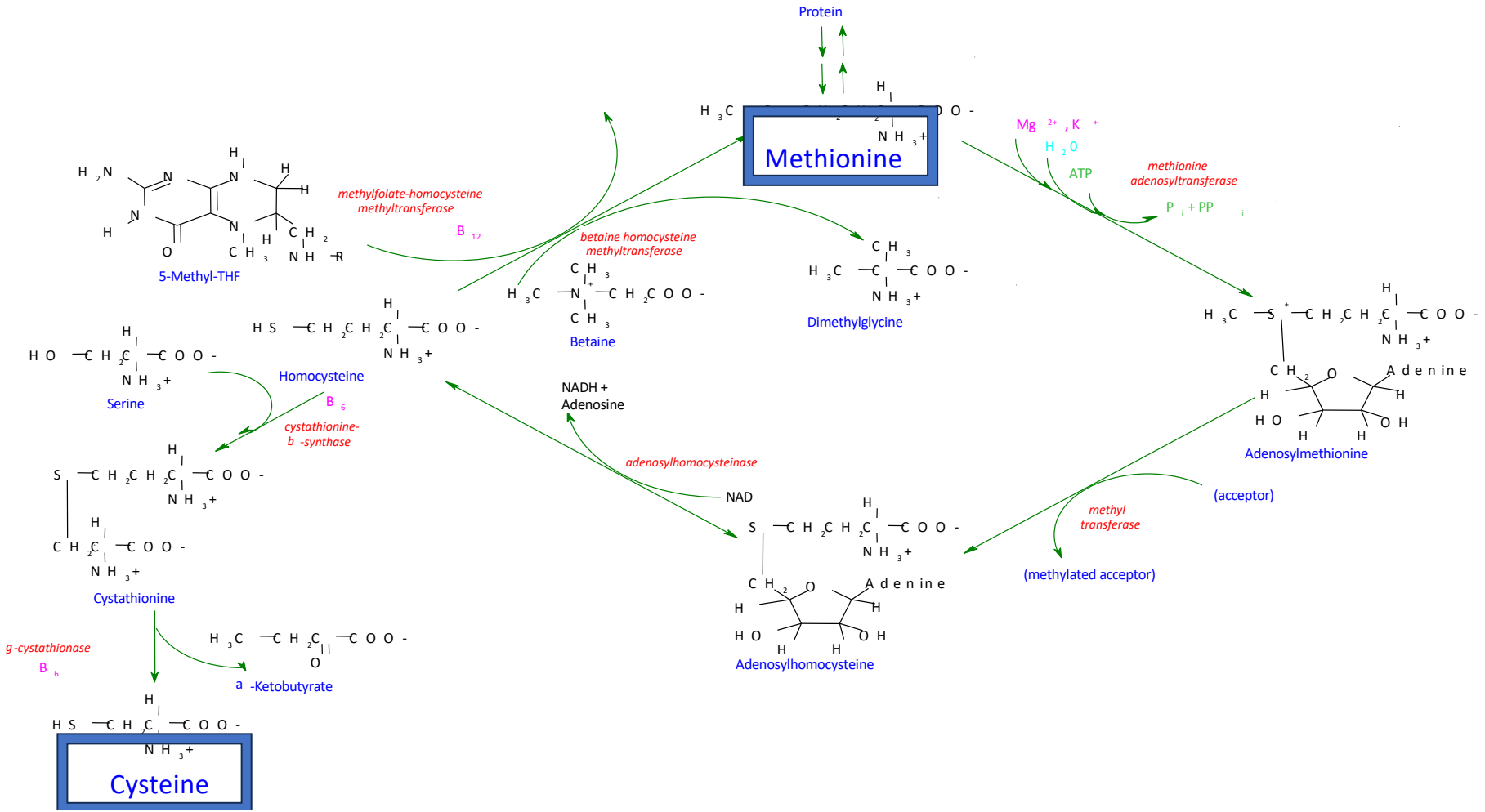
Control: β = -0.13, 95% CI [-0.41, 0.15], **P > 0.05**



Treatment: β = -0.32, 95% CI [-0.55, -0.09], **P < 0.05**

Tryptophan, gut health, and behaviour

- TRP requirements are greater than regulatory guidance
- TRP participates in secondary metabolism that may take much longer to measure than changes in protein, amino acids, or nitrogen metabolism
- Some secondary metabolites, like serotonin, may have implications for the health and well-being of dogs


Protein
Methionine
Mg 2+ , K +
H 2 0
ATP
methionine adenosyltransferase
P i + PP i
methylfolate-homocysteine methyltransferase
B 12
5-Methyl-THF
betaine homocysteine methyltransferase
Betaine
Dimethylglycine
Homocysteine
B 6
cystathionine- b -synthase
Serine
Cystathionine
g -cystathionase
B 6
a -Ketobutyrate
Cysteine
NADH + Adenosine
adenosylhomocysteinase
NAD
Adenosylhomocysteine
Adenosylmethionine
(acceptor)
methyl transferase
(methylated acceptor)
Adenine

Journal of Animal Science, 2020, Vol. 98, No. 11, 1–10

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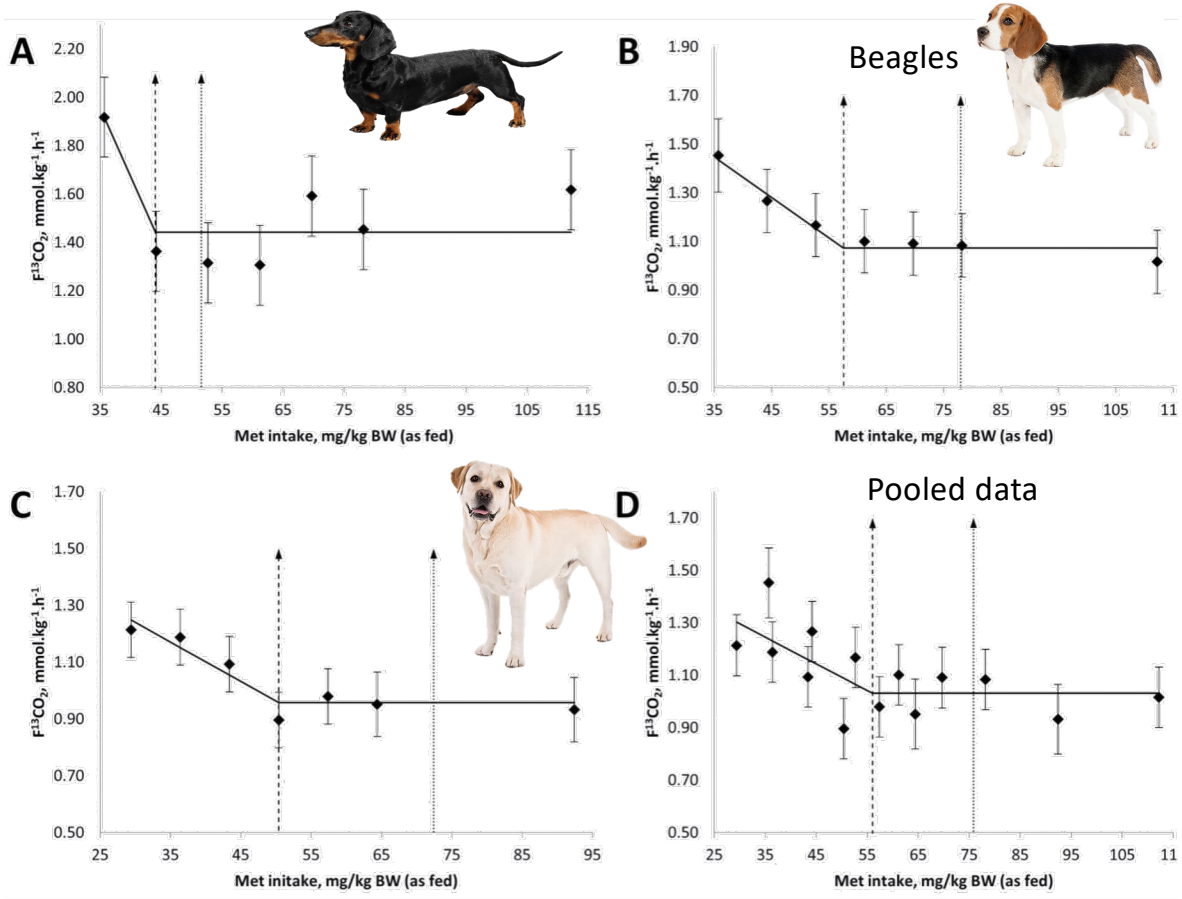
Received: 23 July 2020 and **Accepted:** 29 September 2020
Companion Animal Nutrition

COMPANION ANIMAL NUTRITION

Minimum dietary methionine requirements in Miniature Dachshund, Beagle, and Labrador Retriever adult dogs using the indicator amino acid oxidation technique

Wilfredo D. Mansilla,[†,1] James R. Templeman,[†] Lisa Fortener,[‡,2] and Anna K. Shoveller[†,‡,3]



Minimum methionine requirements (Mansilla et al. 2020)

	AAFCO	FEDIAF (110 kcal/kg$^{0.75}$)	NRC		Miniature Dachshunds		Beagles		Labrador Retrievers		Beagles and Labradors (pooled data)	
			MR	RA	MR	CL	MR	CL	MR	CL	MR	CL
g/100 g DM	0.33	0.40	0.26	0.33	[0.21-0.26]	0.304	0.338	0.458	0.360	0.517	0.360	0.482
g/Mcal ME	0.83	1.00	0.65	0.83	[0.57-0.70]	0.822	0.914	1.238	0.973	**1.397**	0.973	1.303
mg/kg BW					[35.7-45.0]	51.6	57.5	77.9	50.4	72.4	56.0	75.8
mg/kg BW$^{\wedge 0.75}$			85	110	--	--	107.7	147.8	121.8	159.6	118.4	150.5

Amino acid requirements (g/100g DM)

	AAFCO/NRC RA	Small	Medium	Large
Phenylalanine (Shoveller et al, 2018 JAS)	0.44/ 0.45	0.39	0.40	0.50
Tryptophan (Templeman et al, 2019 JAS)	0.16/0.14	0.18	0.26	0.20
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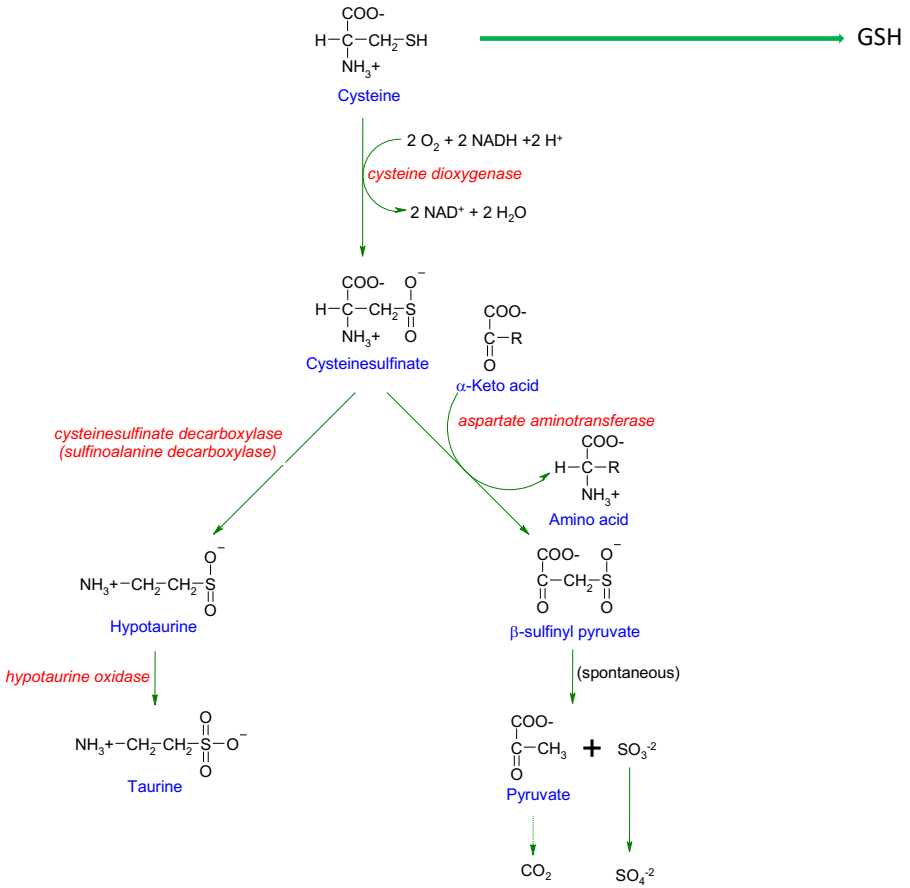
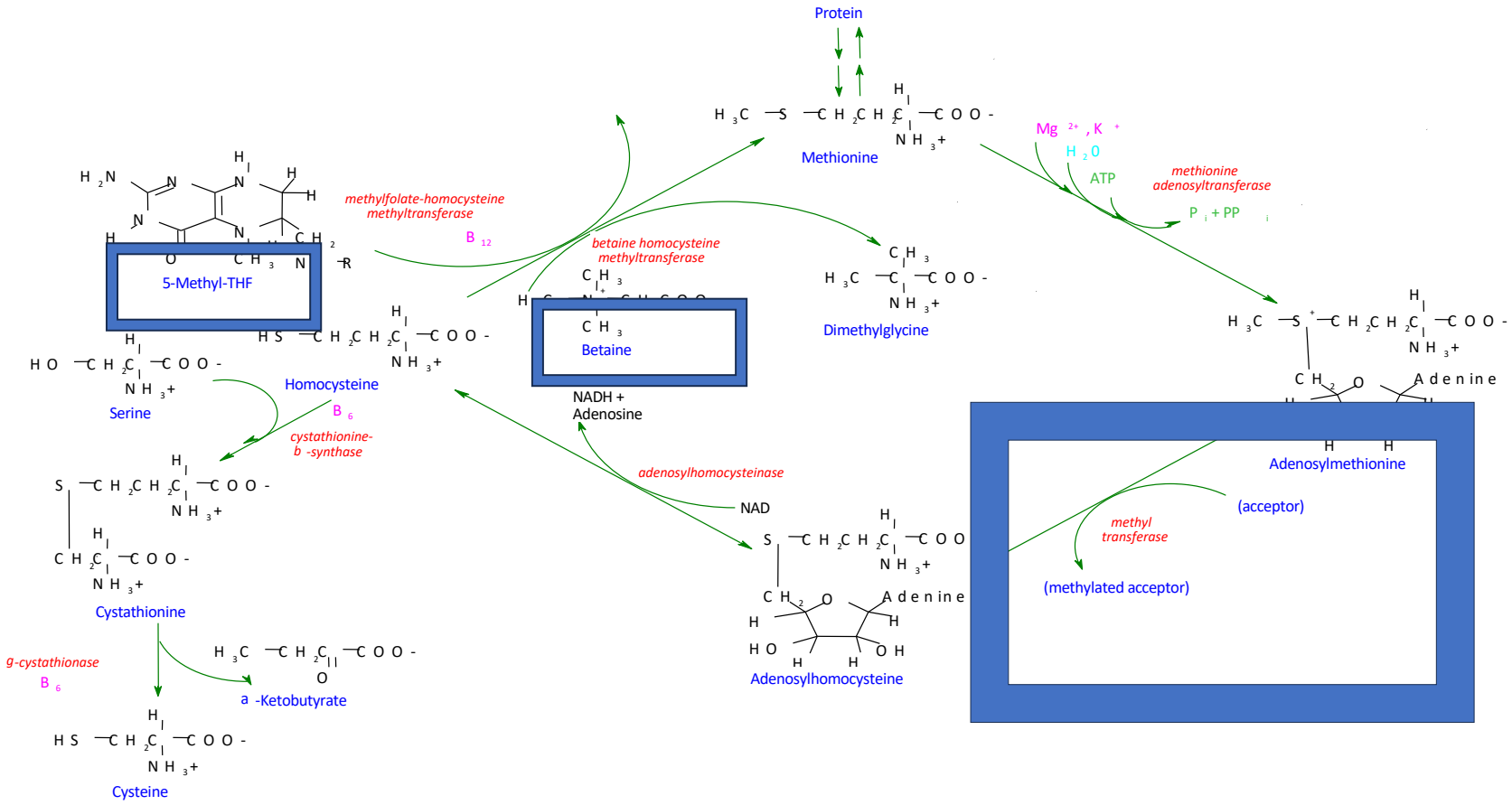
Methionine (Mansilla et al, 2020, JAS)	0.33/0.33	0.34	0.50	0.57

Cysteine is used for glutathione, taurine and sulfate synthesis.

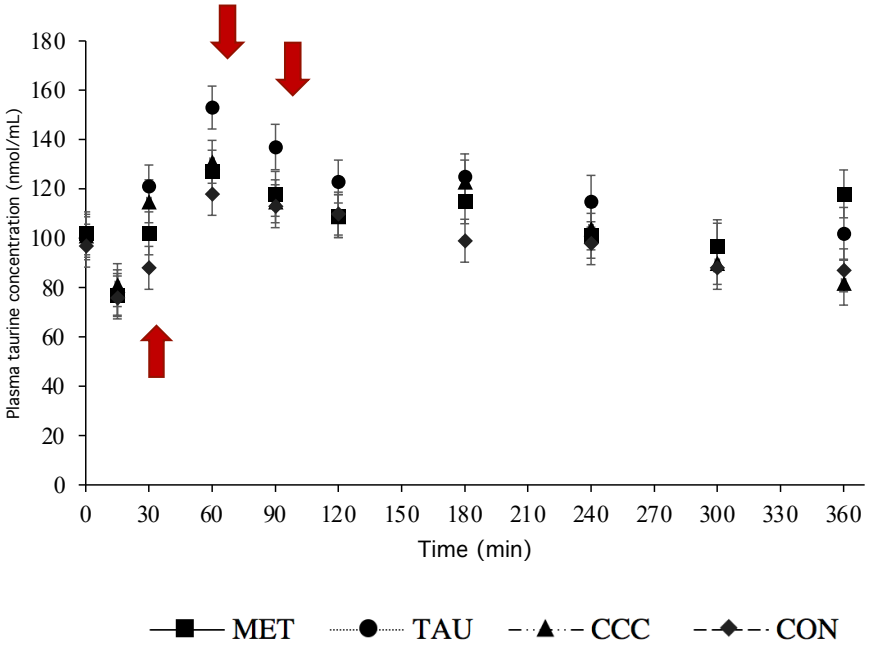


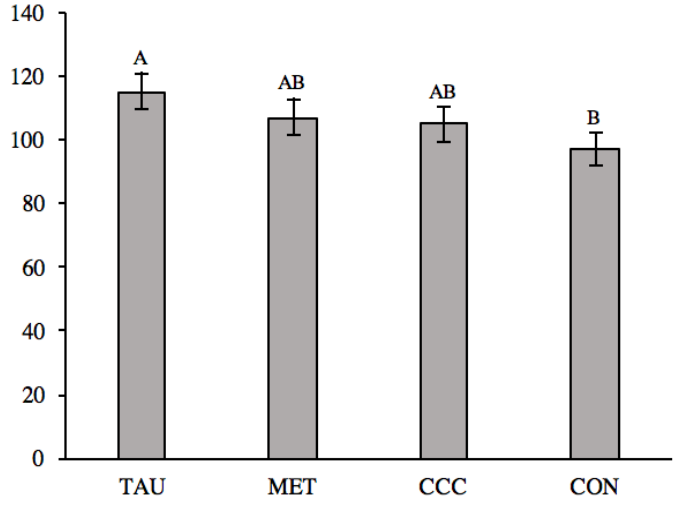
Plasma SAA concentrations (Mansilla et al. 2020)

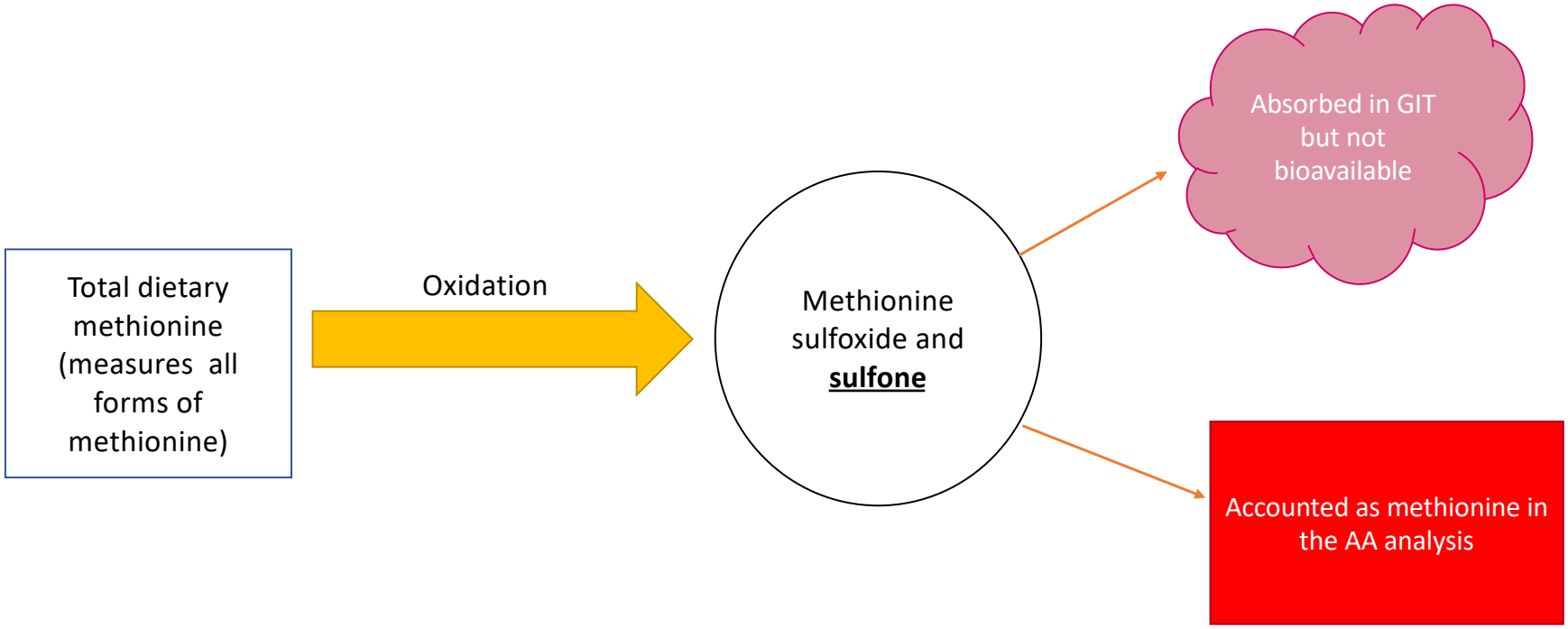
AA, µM	Breed	Dietary Met, % (n=4)										
		0.21	0.26	0.31	0.36	0.41	0.46	0.66	SEM[1]	Breed	Met	Interaction
Methionine	Dachshunds	26.1	23.1	34.7	43.9	52.4*	63.4*	63.2*	7.0	<0.001	0.016	0.125
	Beagles	141.0	220.6	199.3	238.5	304.3	222.8	344.9*	51.0			
	Labradors	280.6	285.0	147.6	224.8	194.1	252.2	347.6	41.0			
Taurine	Dachshunds	266.1	227.0	195.6	238.5	226.0	217.6	208.7	25.5	<0.001	0.243	0.882
	Beagles	175.9	159.4	141.4	176.6	184.5	192.2	177.1	24.0			
	Labradors	133.8	131.3	110.1	126.9	119.3	116.2	127.4	12.8			

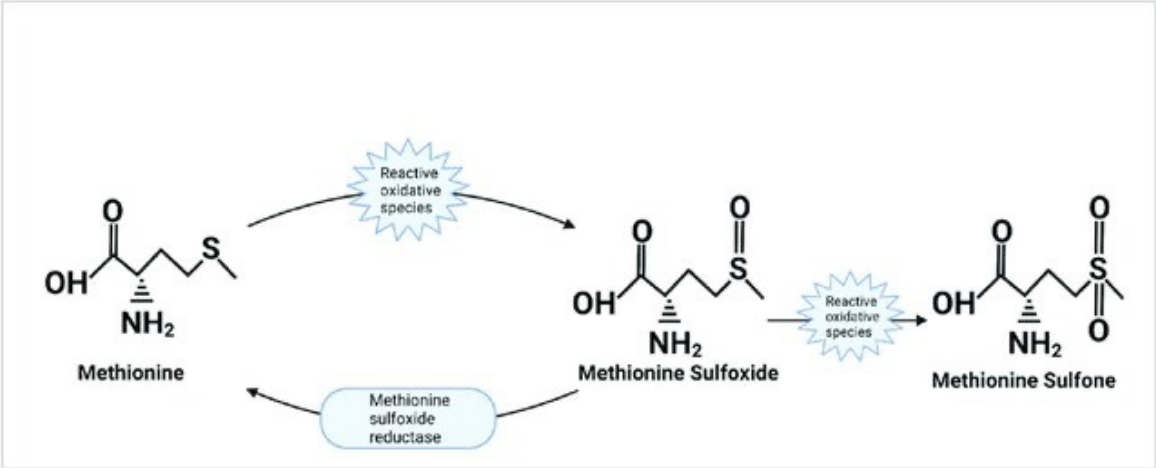

Protein
Methionine
Mg 2+ , K +
H 2 0
ATP
methionine adenosyltransferase
P i + PP i
methylfolate-homocysteine methyltransferase
B 12
5-Methyl-THF
betaine homocysteine methyltransferase
Betaine
Dimethylglycine
Homocysteine
B 6
cystathionine-b -synthase
Serine
NADH + Adenosine
adenosylhomocysteinase
NAD
Adenosylmethionine
(acceptor)
methyl transferase
(methylated acceptor)
Adenosylhomocysteine
Cystathionine
g-cystathionase
B 6
a -Ketobutyrate
Cysteine

Plasma taurine is greater when either methionine or choline, carnitine and creatine are provided in contrast to control (Banton et al. 2021)






Total dietary methionine (measures all forms of methionine)
Oxidation
Methionine sulfoxide and sulfone
Absorbed in GIT but not bioavailable
Accounted as methionine in the AA analysis



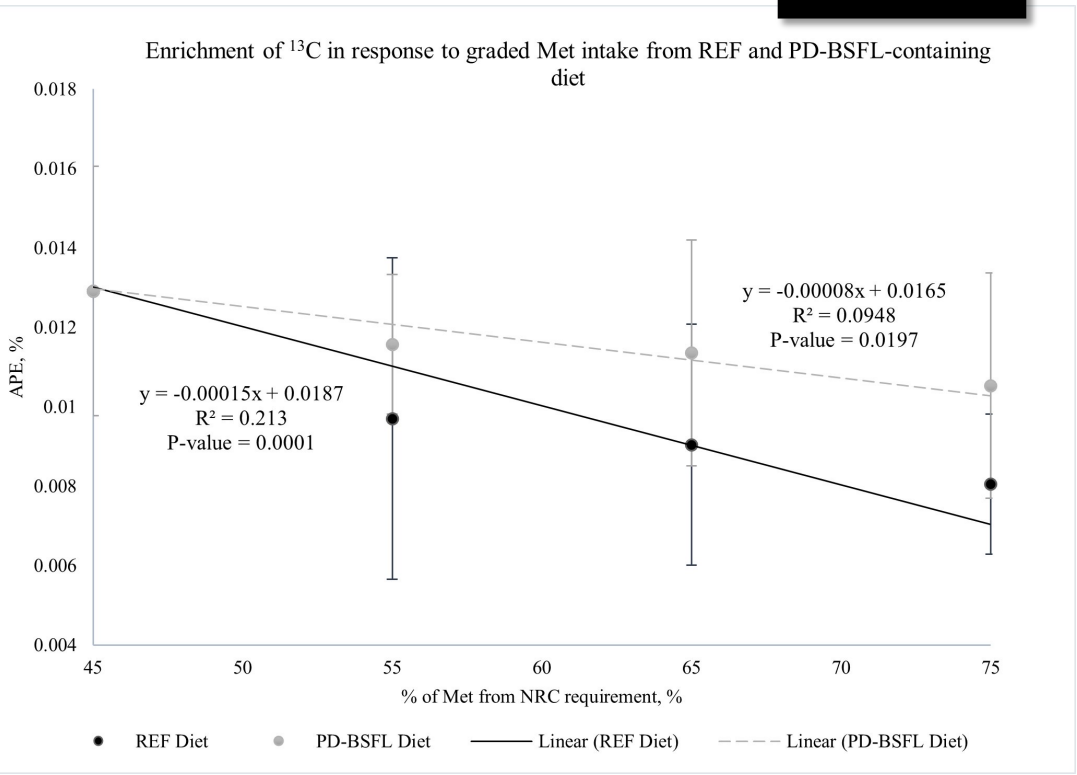
DL-methionine 100% **Met-sulfoxide 60%** **Met-sulfone 0%**

Anderson H. et al. Journal of Nutrition, 106(8): 1108-1114.

Journal of Animal Science, 2023, **101**, 1–12
https://doi.org/10.1093/jas/skac420
Advance access publication 25 December 2022
Non ruminant nutrition

Evaluation of standardized ileal digestibility of amino acids and metabolic availability of methionine, using the indicator amino acid oxidation method, in black soldier fly larvae (*Hermetia illucens*) meal fed to growing pigs

Fiona Tansil,[1] Júlia G. Pezzali,[1] Cara Cargo-Froom,[1] Lee-Anne Huber,[1] Elijah G. Kiarie,[1] Glenda Courtney-Martin,[2,3], Crystal L. Levesque,[4] and Anna K. Shoveller[2,3],



Item	SID, %	SEM	SID content, %
Crude protein	76.1	3.08	37.5
Indispensable AA, %			
Arg	93.0	0.99	2.2
His	83.4	1.91	1.2
Ile	86.0	1.08	1.6
Leu	88.8	1.36	2.8
Lys	87.6	1.52	2.3
Met	90.4	1.60	0.4
Phe	89.3	1.48	1.6
Thr	83.0	2.40	1.1
Val	86.2	1.29	2.3

The sulfur amino acids

- MET requirements are greater than regulatory guidance
- Sulfur amino acid metabolism affects and is affected by the cofactors needed for transmethylation, remethylation and transsulfuration
- MET bioavailability currently is not measured in canine nutrition and physiology and is needed to understand the protein quality of the foods we feed

Amino acid requirements (g/100g DM)

	AAFCO/NRC RA	Small	Medium	Large
Phenylalanine (Shoveller et al, 2018 JAS)	0.44/ 0.45	0.39	0.40	0.50
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(Institute of Medicine, 2005), FAO/WHO, 2007), (National Academies Press, 1985)

Enabling calculations of protein quality …

J Anim Sci. 2022 Nov 1;100(11):skac279. doi: 10.1093/jas/skac279.

Digestible indispensable amino acid scores of animal and plant ingredients potentially used in dog diet formulation: how this protein quality metric is affected by ingredient characteristics and reference amino acid profile

James R Templeman [1], Anna K Shoveller [1]

Affiliations + expand

PMID: 36029066 PMCID: PMC9624197 DOI: 10.1093/jas/skac279

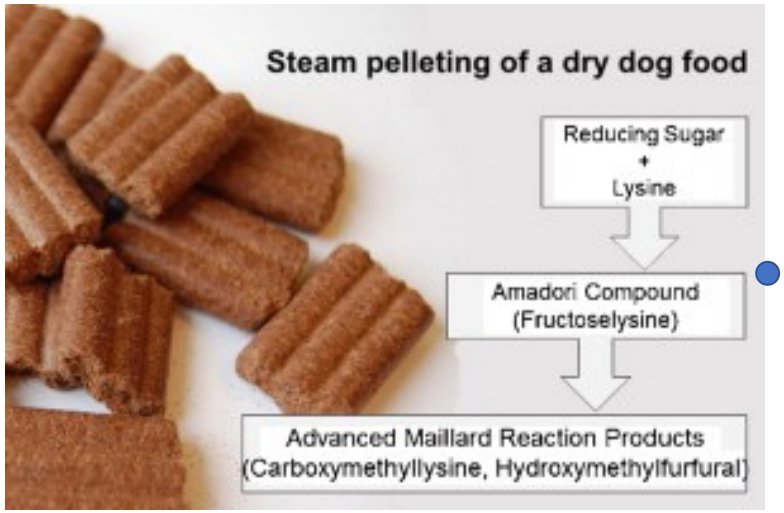
Free PMC article

MiNACS

Protein quality can be quantified and conveyed to the veterinarian and consumer

$$\text{DIAAS} = \frac{(\text{mg digestible IAA / 1 g test ingredient crude protein})}{(\text{mg of same IAA / 1 g reference protein})} \times 100$$

Reference patterns? What happens when those are not accurate?



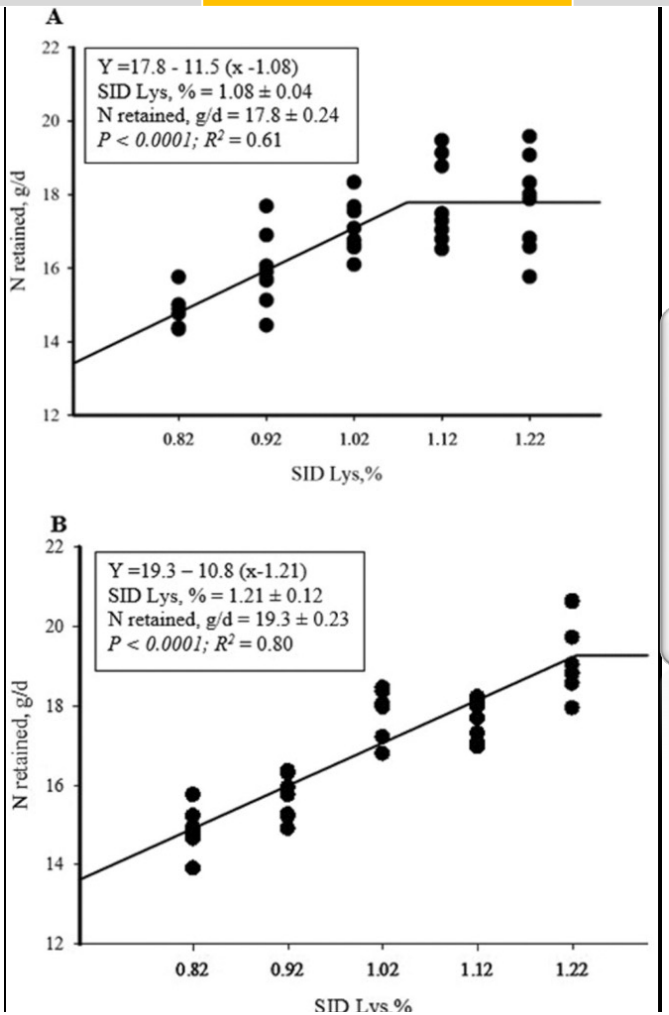
Van Rooijen et al. 2014

Analytical accuracy of measurements of AA in forms that mammals can utilize or compounds that may metabolically, but not digestively alter the use of AA for protein synthesis

The SID Lysine requirement is greater (1.21%) when pigs are fed low ratio of indispensable nitrogen: total nitrogen ratios (0.33) in comparison to pigs fed a high ratio (0.36).

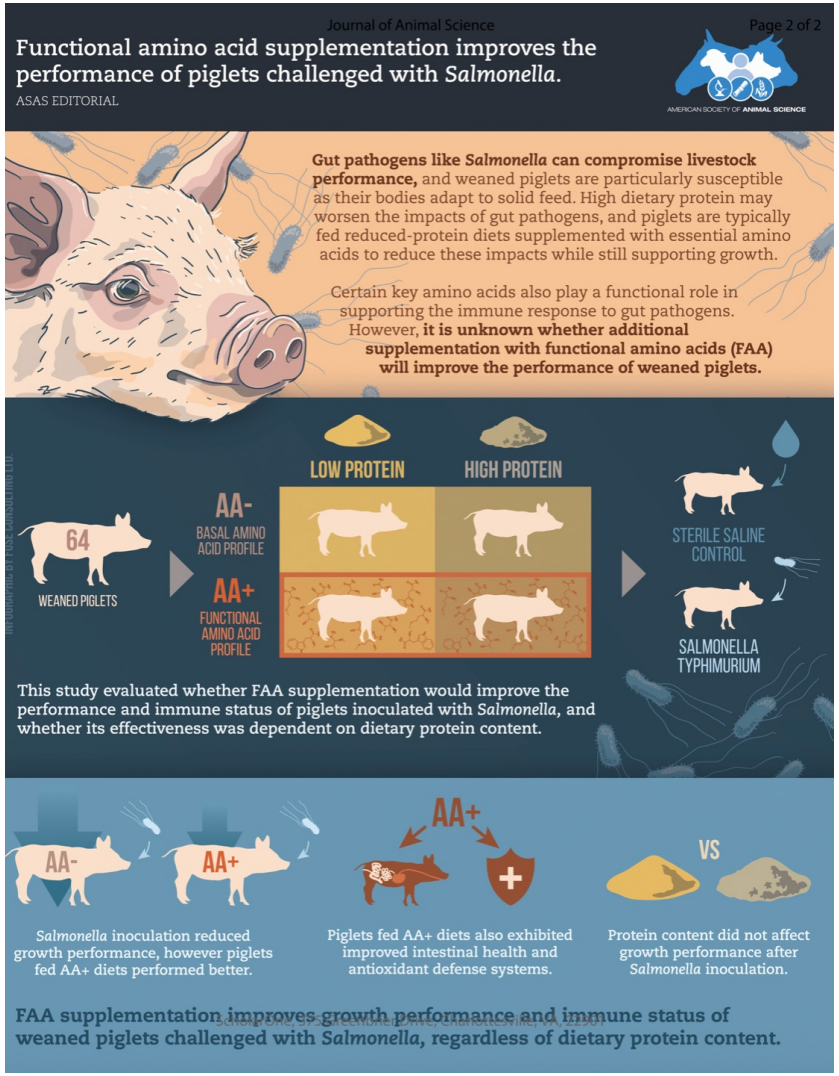
What does this mean for the variability in amino acid and protein content of commercial dog and cat foods?

Camiré et al. 2023



How does the ratio of indispensable AA nitrogen to total nitrogen affect AA requirements in dogs and cats?

Is there a role or requirements for dispensable AA in dogs and cats? Could this help ameliorate sarcopenia in aging dogs and cats?



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doi:10.1093/jas/skaa365
Advance Access publication February 2, 2021
Received: 2 October 2020 and **Accepted:** 9 November 2020
Non Ruminant Nutrition

NON RUMINANT NUTRITION

Functional amino acid supplementation, regardless of dietary protein content, improves growth performance and immune status of weaned pigs challenged with *Salmonella* Typhimurium

Lucas A. Rodrigues,[†,‡] Michael O. Wellington,[‡] J. Caroline González-Vega,[‖] John K. Htoo,[‖] Andrew G. Van Kessel,[‡] and Daniel A. Columbus[†,‡,1]

Journal of Animal Science, 2022, **100**, 1–15
https://doi.org/10.1093/jas/skac267
Advance access publication 17 August 2022
Non Ruminant Nutrition

Functional amino acid supplementation attenuates the negative effects of plant-based nursery diets on the response of pigs to a subsequent *Salmonella* Typhimurium challenge

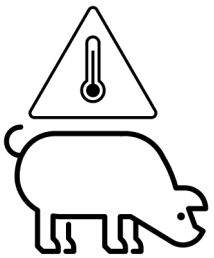
Lucas A. Rodrigues,[†,‡] Josiane C. Panisson,[†,‡] Andrew G. Van Kessel,[‡] and Daniel A. Columbus[†,‡,1]

Journal of Animal Science, 2024, **102**, skae120
https://doi.org/10.1093/jas/skae120
Advance access publication 30 May 2024
Gastrointestinal Biology

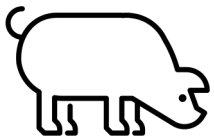
Effects of supplemental citrulline on thermal and intestinal morphology parameters during heat stress and feed restriction in growing pigs

Sara K. Kvidera, Edith J. Mayorga, Carrie S. McCarthy, Erin A. Horst, Megan A. Abeyta, and Lance H. Baumgard[1]

Department of Animal Science, Iowa State University, Ames, Iowa 50011, USA
[1]Corresponding author: baumgard@iastate.edu

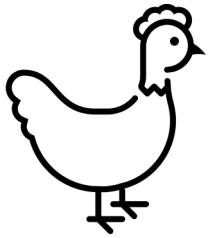


Dietary glycine supplementation enhances postweaning growth and meat quality of pigs with intrauterine growth restriction



Wenliang He, Erin A. Posey, Chandler C. Steele, Jeffrey W. Savell, Fuller W. Bazer, and Guoyao Wu[1]

Dietary protein reduction with stepwise addition of crystalline amino acids and the effect of considering a minimum glycine-serine content in broiler diets



Wilfredo D. Mansilla,[*,1] Saritha Saraswathy,[†] and Ana I. García-Ruiz[*]

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Dietary glycine supplementation prevents heat stress-induced impairment of antioxidant status and intestinal barrier function in broilers

Chenxi Deng,[*,†,1] Jun Zheng,[*,†,1] Hua Zhou,[*,†] Jinming You,[*,†] and Guanhong Li[*,†,2]

American Society for Nutrition

The American Journal of CLINICAL NUTRITION

journal homepage: https://ajcn.nutrition.org/

Original Research Article

Reduced plasma glycine concentration in healthy and chronically diseased older adults: a marker of visceral adiposity?

Lars NJ Deutz[1], Raven A Wierzchowska-McNew[1], Nicolaas EP Deutz[1,2], Mariëlle PKJ Engelen[1,2,*]

Summary

- AA requirements may be too low when dogs are fed to weight maintenance and food restricted in cases to avoid weight gain
- The ratio of amino acids should be considered in research and subsequently in commercial formulation, ie. Trp: LNAA and the as a ratio to nitrogen, such as employed in DIAAS measures of protein quality
- Protein and AA metabolism is affected by other nutrients, ie. micronutrients, and need to be considered together in formulation
- Protein quality also considers the bioavailability of the AA and you should understand what you are measuring, such as in the case of MET
- The physiological state of animals will predict the efficacy of individual formulation approaches and need to be considered in formulation


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