

# Kansas Agricultural Experiment Station Research Reports

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Volume 0  
Issue 10 *Swine Day (1968-2014)*

Article 100

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1973

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T E. Eckert

G L. Allee

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### Recommended Citation

Eckert, T E. and Allee, G L. (1973) "Limiting amino acids in milo for the growing pig," *Kansas Agricultural Experiment Station Research Reports*: Vol. 0: Iss. 10. <https://doi.org/10.4148/2378-5977.3520>

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## Limiting Amino Acids in Milo for the Growing Pig

Thomas E. Eckert and Gary L. Allee

### Summary

Three growth trials and two nitrogen retention trials were conducted with 118 growing pigs to determine the limiting amino acids in milo. Lysine supplementation of the basal milo diet markedly improved daily gain, feed efficiency, and nitrogen retention. Adding threonine to diets containing supplemental lysine or lysine and methionine increased daily gain and improved feed efficiency. Similarly nitrogen retention was increased by adding threonine to the basal milo diet supplemented with lysine. Supplementing the basal milo diet with methionine in the presence of lysine gave no beneficial response in two trials and markedly depressed growth in a third. Supplementing with nonessential amino acids (glycine and glutamic acid) in the presence of all other amino acids studied gave no beneficial response. These data demonstrate that lysine is the first-limiting, and threonine the second-limiting amino acid in milo protein for the growing pig and that methionine is not third-limiting. The data suggest that isoleucine is the third-limiting amino acid in milo protein.

### Introduction

Milo has become a major feed grain in the U. S., and especially in Kansas. Without adequate supplementation, however, it has serious amino acid limitations. Milo is deficient in several amino acids for the growing pig, including lysine, methionine, threonine, isoleucine, and tryptophan. The increased occurrence of amino acid supplementation of swine diets makes it important to learn the qualitative and quantitative amino acid limitations of milo protein.

### Procedures

General. In the growth trials, pigs were housed in groups of three in elevated pens with expanded metal floors in a building where temperature was maintained at approximately 72°F. Feed and water were available at all times. Initial and final weights were recorded and daily gain and feed efficiency were determined at the end of the trials. Trials varied from 21 to 28 days.

In the nitrogen retention studies, pigs were housed individually in metal metabolism cages allowing for separate collection of feces and urine. Daily feed intake was constant, fed in two equal portions at approximately 8:00 a.m. and 5:00 p.m. Water was available at all times. A five day pre-test period preceded a five day collection period.

Composition of the fortified milo basal diet is shown in Table 1.1, with its amino acid composition in Table 1.2. Crystalline amino acids were added to the basal diet to equal the amino acid content of a 16%-protein, milo-soybean meal diet in trials 1 and 2. In trials 3, 4, and 5 amino acids were supplemented to meet N. R. C. (1968) requirements. Amino acids were added in the order calculated to be limiting, then rearranged in subsequent trials. Only the L-isomer of amino acids was added except in the case of DL-methionine. Lysine was added as L-lysine·HCl.

Trial 1. Thirty Hampshire and Yorkshire pigs weighing 37 pounds each were allotted based on weight, breed and sex to the following five treatments: (1) basal milo diet, (2) 1 plus 0.56% lysine, (3) 2 plus 0.22% DL-methionine, (4) 3 plus 0.33% threonine, and (5) a 16%-protein, milo-soybean meal as a control. The trial consisted of six pigs per treatment and lasted 28 days.

Trial 2. A nitrogen metabolism study was conducted using four Littermate Yorkshire gilts weighing an average of 40 pounds. The gilts were randomly assigned to treatments 1, 2, 3 and 4 used in trial 1. A Latin Square design was used and daily intake was held constant at 2.5 pounds throughout the trial.

Trial 3. Forty-two Duroc, Hampshire, and Yorkshire pigs weighing an average of 48 pounds were allotted based on weight, breed, and sex to the following seven treatments: (1) basal milo, (2) 1 plus 0.47% lysine, (3) 2 plus 0.13% threonine, (4) 3 plus 0.27% methionine, (5) 4 plus 0.12% isoleucine, (6) 5 plus 1.70% glycine and 1.70% glutamic acid, and (7) 16%-protein, milo-soybean meal control. Diets six and seven were equal in nitrogen. The trial was replicated with a total of six pigs per treatment and lasted 21 days.

Trial 4. Thirty Duroc and Yorkshire pigs weighing an average of 37 pounds were allotted by weight and breed to five treatments: (1) basal plus 0.40% lysine, (2) 1 plus 0.11% threonine, (3) 1 plus 0.26% methionine, (4) 1 plus 0.11% threonine plus 0.26% methionine, and (5) 16%-protein, milo-soybean meal control. The trial consisted of six pigs per treatment and lasted 22 days.

Trial 5. A nitrogen retention study was conducted using twelve Duroc and Yorkshire barrows weighing an average of 35 pounds. Three groups of four littermates were used in a randomized complete block design. In each group, treatments 1, 2, 3 and 4 employed in trial 4, were randomly assigned for one period (10 days), then the pigs were re-allotted for another period to provide a replicate. Daily intake was 2.0 pounds for the first period and 2.2 pounds for the second.

Table 1.1. Composition of Basal Milo Diet<sup>a</sup>

Ingredient	%
Milo <sup>b</sup>	96.11
Dicalcium phosphate	1.92
Ground limestone	0.42
Salt	0.50
Vitamin and antibiotic premix	1.00
Trace mineral premix	0.05

<sup>a</sup>Diet was ground.

<sup>b</sup>Crude protein in trials 1, 2 and 3 = 8.7%; in trials 4 and 5 = 9.1%.

Table 1.2. Amino Acid Composition of Fortified Milo Basal Diet<sup>a</sup>

Amino Acids	%	
	<u>Trials</u> 1, 2 & 3	<u>Trials</u> 4 & 5
Lysine	0.230	0.299
Methionine	0.093	0.096
-Cystine	0.140	0.144
Threonine	0.315	0.343
Isoleucine	0.380	0.366
Leucine	1.217	1.271
Tryptophan	0.163	0.115
Arginine	0.378	0.453
Valine	0.458	0.455
Histidine	0.214	0.262
Phenylalanine	0.470	0.490
-Tyrosine	0.345	0.388

<sup>a</sup>Per cent of air-dry diet.

## Results

Trial 1. Results of this growth trial are presented in Table 1.3. Supplementing the basal diet with lysine (0.56%) significantly ( $P<.05$ ) improved daily gain and feed efficiency. Adding methionine (0.22%) in the presence of lysine slightly improved daily gain and feed efficiency. Threonine (0.33%) in the presence of lysine and methionine significantly ( $P<.05$ ) improved both daily gain and feed efficiency. As expected, the milo-soybean meal diet produced the fastest ( $P<.05$ ) and most efficient gains.

Table 1.3. Performance of Growing Pigs Fed Fortified Milo Diets Supplemented with L-lysine, DL-methionine, and L-Threonine<sup>a</sup> (Trial 1)

Diets	Daily gain (lbs.)	Feed/gain
1. Basal	0.23 <sup>b</sup>	7.52 <sup>b</sup>
2. as 1 + 0.56% lysine	0.40 <sup>c</sup>	5.07 <sup>c</sup>
3. as 2 + 0.22% methionine	0.54 <sup>c</sup>	4.34 <sup>cd</sup>
4. as 3 + 0.33% threonine	0.75 <sup>d</sup>	3.35 <sup>cd</sup>
5. Milo + Soybean meal	1.38 <sup>e</sup>	2.26 <sup>f</sup>

<sup>a</sup>Six pigs per diet, avg. initial wt., 37 lbs., 28 day trial.

bcdef Means with different superscripts in the same column differ significantly ( $P<.05$ ).

Trial 2. Data from the nitrogen metabolism study are in Table 1.4. Adding lysine (0.56%) to the basal diet significantly ( $P<.05$ ) increased nitrogen retention. Methionine (.22%) in the presence of lysine increased nitrogen retention by 0.78 g per day over that observed for lysine alone. However, this difference was not statistically significant. Adding threonine (0.33%) in the presence of lysine, and methionine, significantly ( $P<.05$ ) increased nitrogen retention over that observed on the basal diet supplemented with lysine or with lysine and methionine.

Trial 3. Average daily gain (ADG) and feed/gain (F/G) data are presented in Table 1.5. Adding lysine (0.47%) to the basal milo diet tripled ADG and reduced F/G by 50%. Adding threonine (0.13%) in the presence of lysine increased ADG by 0.11 lbs. over that observed from lysine alone and decreased F/G. Adding 0.27% methionine in the presence of lysine and threonine reduced ADG and increased F/G over that observed from the basal diet with added lysine and threonine. Isoleucine (0.12%), in the presence of lysine, threonine, and methionine increased ADG ( $P<.05$ ) and lowered F/G compared with results from the other amino acid diets.

Table 1.4. Nitrogen Retention of Pigs Fed Fortified Milo Diets Supplemented with L-Lysine, DL-Methionine, and L-Threonine<sup>a</sup> (Trial 2)

Diets	Intake	Daily N (g)		Retained
		Urine	Fecal	
1. Basal	16.62	6.29	4.08	6.25 <sup>b</sup>
2. as 1 + 0.56% lysine	18.18	5.19	4.39	8.60 <sup>c</sup>
3. as 2 + 0.22% methionine	18.52	4.70	4.44	9.38 <sup>c</sup>
4. as 3 + 0.33% threonine	19.05	3.71	4.02	11.32 <sup>d</sup>

<sup>a</sup>Four Yorkshire gilts weighing 40 lbs., 4 X 4 Latin Square design.

<sup>bcd</sup>Means with different superscripts differ ( $P < .05$ ) significantly.

Table 1.5. Effects of Adding Indicated Amino Acids to a Fortified Milo Diet<sup>a</sup> (Trial 3)

Diets	Daily gain (lbs.)	Feed/gain
1. Basal	0.15 <sup>b</sup>	11.46 <sup>b</sup>
2. as 1 + 0.47% lysine	0.45 <sup>c</sup>	5.33 <sup>cd</sup>
3. as 2 + 0.13% threonine	0.56 <sup>c</sup>	4.33 <sup>de</sup>
4. as 3 + 0.27% methionine	0.47 <sup>c</sup>	4.56 <sup>de</sup>
5. as 4 + 0.12% isoleucine	0.85 <sup>d</sup>	3.43 <sup>ef</sup>
6. as 5 + 1.70% glutamic acid + 1.70% glycine	0.25 <sup>b</sup>	5.92 <sup>c</sup>
7. Milo + Soybean meal	1.40 <sup>e</sup>	2.43 <sup>f</sup>

<sup>a</sup>Six pigs per diet, avg. initial wt., 48 lbs., 21-day trial.

<sup>bcd</sup>Means with different superscripts in the same column differ ( $P < .05$ ) significantly.

Including nonessential amino acids in the presence of lysine, threonine, methionine, and isoleucine sharply reduced gain and feed efficiency. The 16%-protein, milo-soybean diet produced the highest gains and the lowest F/G of all diets studied.

Trial 4. Results of this growth trial are presented in Table 1.6. Supplementing the basal diet with 0.40% lysine produced a daily gain of only 0.73 lbs. with a 3.84 F/G. Adding threonine (0.11%) to the basal + lysine diet increased ( $P<.10$ ) daily gain and reduced ( $P<.05$ ) F/G. Adding methionine in the presence of lysine significantly ( $P<.05$ ) reduced daily gain. Adding both threonine and methionine in the presence of lysine resulted in almost identical responses in both ADG and F/G as that observed when only threonine was added in the presence of lysine. ADG for the milo-soybean meal diet was greater ( $P<.10$ ) than other treatments although the F/G was only slightly better (lower) than from added threonine.

Table 1.6. Performance of Growing Pigs Fed Fortified Milo Diets Supplemented with Indicated Amino Acids<sup>a</sup> (Trial 4)

Diets	Daily gain (lbs.)	Feed/gain
1. Basal + 0.40% lysine	0.73 <sup>b</sup>	3.84 <sup>f</sup>
2. as 1 + 0.11% threonine	0.89 <sup>c</sup>	2.92 <sup>g</sup>
3. as 1 + 0.26% methionine	0.49 <sup>d</sup>	4.71 <sup>f</sup>
4. as 1 + 0.11% threonine + 0.26% methionine	0.88 <sup>c</sup>	2.94 <sup>g</sup>
5. Milo + Soybean meal	1.46 <sup>e</sup>	2.19 <sup>g</sup>

<sup>a</sup>Six pigs per diet, avg. initial wt., 37 lbs., 22-day trial.

<sup>bcde</sup>Means with different superscripts differ ( $P<.10$ ) significantly.

<sup>fg</sup>Means with different superscripts differ ( $P<.05$ ) significantly.

Trial 5. Results from the nitrogen retention study are in Table 1.7. Adding 0.40% lysine to the basal diet resulted in 6.11 g of nitrogen retained per day. Adding 0.11% threonine to the basal + lysine diet significantly ( $P<.10$ ) increased nitrogen retention to 6.95 g. Adding methionine (0.26%) to the basal + lysine diet did not improve nitrogen retention. Methionine in the presence of lysine and threonine did not increase nitrogen retention over that from the basal diet supplemented with lysine and threonine.

Table 1.7. Nitrogen Retention of Barrows Fed Basal Milo Diet Supplemented with L-lysine, L-Threonine and DL-Methionine<sup>a</sup> (Trial 5)

Diets	Intake	Daily N (g)		Retained
		Urine	Fecal	
1. Basal + 0.40% lysine	13.45	3.35	3.99	6.11 <sup>b</sup>
2. as 1 + 0.11% threonine	13.56	2.44	4.17	6.95 <sup>c</sup>
3. as 1 + 0.26% methionine	13.68	3.31	4.14	6.23 <sup>b</sup>
4. as 1 + 0.11% threonine + 0.26% methionine	13.81	3.37	3.81	6.63 <sup>bc</sup>

<sup>a</sup>Six pigs per diet, initial wt., 35 lbs.

<sup>bc</sup>Means with different superscripts differ ( $P < .10$ ) significantly.

### Discussion

Studies reported here confirm and extend previous findings by demonstrating, using growth and nitrogen retention studies, that lysine is the first-limiting amino acid in milo for the growing pig and threonine is the second-limiting amino acid in milo protein. Adding lysine to the fortified milo basal diet markedly improved daily gain, feed efficiency, and nitrogen retention.

These experiments demonstrate that threonine is the second-limiting amino acid, although it had been calculated to be third-limiting. Adding threonine in the presence of lysine or lysine and methionine improved both daily gain and feed efficiency. Similarly, threonine increased nitrogen retention when added to diets containing either lysine or lysine and methionine.

Although methionine is calculated to be the second-limiting amino acid in milo protein, adding methionine to lysine fortified diets failed to increase daily gain or nitrogen retention.

Adding isoleucine in the presence of lysine, threonine and methionine improved ADG and improved F/G over diets with no supplemental isoleucine. This suggests that isoleucine is the third-limiting amino acid in milo.

Adding nonessential amino acids (glycine and glutamic acid) in the presence of lysine, threonine, methionine and isoleucine depressed both daily gain and feed efficiency. Apparently, nonessential amino acids were sufficient in the diets and additional quantities contributed to an imbalance.



The superiority of performance by pigs fed the 16%-protein, milo-soybean meal diet indicates that additional amino acids other than those studied are limiting in milo protein for growing pigs.