

## Tryptophan Bioavailability in Soybean Meal for Young Pigs

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### Introduction

Several studies have been conducted to determine the bioavailability of amino acids for young pigs in the 22 to 45 lb live weight range (Sato et al., 1987; Kovar et al., 1993; Adeola et al., 1994). Bioavailability assays based on animal performance provide relative information on the capacity of a feed ingredient to provide a specific limiting amino acid for maintenance and growth. Commonly used dependent variables in a slope-ratio assay are weight gains (non-partitioned or partitioned) and gain:feed ratio. Although the independent variable used may be influenced by the choice of dependent variable, amino acid intake has been preferred by most investigators. There is evidence that bioavailability estimates vary depending on the choice of independent variable, but this has not been systematically investigated. An obvious question would be, "to what extent do the dependent and independent variables affect the estimates of bioavailability?" To evaluate this, the bioavailability of tryptophan in soybean meal was estimated by the slope-ratio assay using supplemental level, daily total intake, and daily supplemental intake of tryptophan as independent variables.

### Materials and Methods

#### *Pigs, Diets, and Housing*

Seventy-two 5-week-old crossbred pigs were used in the experiment. Thirty-six barrows and 36 gilts with an average initial weight of 22 lb were blocked by weight and randomly assigned to six treatments. The six treatments consisted of one basal diet, three reference diets, and two test diets. The composition of the basal diet is in Table 1. The content of all amino acids for the basal diet was at least 110% of NRC (1988) requirements for 22- to 45-lb pigs, except for tryptophan. The three reference diets were formulated by supplementing the basal diet with .2, .4, or .6 g of L-tryptophan/kg. The two test diets were formulated by supplementing the basal diet with tryptophan from soybean meal at .2 or .4 g/kg.

The pigs were randomly assigned to individual stainless steel pens equipped with nipple drinkers, stainless steel feeders, and plastic-coated, expanded metal floors. The pens were located in an environmentally regulated building maintained at approximately 75°F with a 12-hour light-dark cycle. Body weight and feed intake were recorded weekly, and ad-libitum access to feed and water was provided for 28 days.

#### *Statistical Methods*

The response criteria of average daily gain (ADG), average daily feed intake (ADFI), and gain:feed ratio were analyzed using a randomized complete block design. Common-intercept, multiple linear regression and slope-ratio techniques (Finney, 1978) were used to estimate availability of

tryptophan. Linear and quadratic contrasts were used to examine the relationship between growth performance response criteria and added tryptophan from L-tryptophan or soybean meal.

## Results

### *Ingredient and Basal Diet Composition*

The amino acid composition of corn and corn gluten meal, which together supplied 80% of the tryptophan in the basal diet, is given in Table 2. Corn used in the basal diet contained .6 g tryptophan/kg, which is considerably lower than the average tryptophan content of corn samples (.9 g/kg) published by NRC (1988). However, the .6 g tryptophan/kg is within the range of .5 to .8 g/kg reported for the 1994 corn crop in the U.S. (Blair, 1995). Tryptophan in corn gluten meal at 3.2 g/kg is comparable to the 3 g/kg reported by NRC (1988). The basal diet was formulated to contain 1.1 g tryptophan/kg on the basis of analyzed tryptophan in corn and corn gluten meal and 4.3 g tryptophan/kg of skim milk powder (NRC, 1988). Analyzed tryptophan content of soybean meal at 6.2 g/kg (Table 2) was used to formulate the test diets containing .2 or .4 g tryptophan/kg. The analyzed lysine content of the basal diet (10.4 g/kg) exceeded the NRC (1988) requirement of 9.5 g/kg for 22- to 45-lb pigs.

### *Growth Performance Response to Dietary Tryptophan*

Adding tryptophan to the diets increased ( $P<.05$ ) feed intake and consequently increased ( $P<.05$ ) both total and supplemental tryptophan intake (Table 3). Supplementing the basal diet with L-tryptophan produced a linear increase ( $P<.05$ ) in weight gain. Similarly, pigs responded linearly ( $P<.05$ ) to the increase in supplemental tryptophan from soybean meal. The linear response in daily weight gain to supplemental tryptophan from soybean meal was significant ( $P<.05$ ). Gain:feed ratio also increased linearly ( $P<.05$ ) to supplemental tryptophan from either L-tryptophan or soybean meal.

Increasing the dietary tryptophan level from 1.03 to 1.43 g/kg improved daily weight gain by 148% and gain:feed ratio by 62% with L-tryptophan supplementation. Daily feed consumption increased by 52 or 27% when dietary tryptophan was increased from 1.0 to 1.4 g/kg through L-tryptophan or soybean meal, respectively. However, the growth improvement was perhaps partially independent of feed intake, because only a portion of the growth improvement could be accounted for by feed intake differences. The dietary tryptophan-induced increase in feed intake observed in the present experiment may be mediated through the influence of tryptophan on the synthesis and release of serotonin, which would in turn influence feed intake through gastric emptying.

### *Tryptophan Bioavailability Based on Non-Partitioned and Partitioned Weight Gain*

Using daily weight gain as the dependent variable and supplemental tryptophan level in the diet as the independent variable, the estimate of tryptophan bioavailability in soybean meal relative to L-tryptophan for 22-lb pigs was 90% (Table 4). When daily total or supplemental tryptophan intake (g/day) was used as the independent variable, estimate of tryptophan bioavailability was 99 or 110%, respectively. Most growth assays conducted to determine bioavailability of amino acids in feed ingredients have used body weight gain as the dependent variable, and amino acid intake has been the

preferred independent variable by most investigators. In the present experiment, the regression of daily weight gain on supplemental tryptophan level, daily total tryptophan intake, or daily supplemental tryptophan intake produced different estimates of tryptophan bioavailability. It should be noted that the residual standard deviation was smallest, and the proportion of the total variation in the data set explained by the relationship between the dependent and independent variables ( $R^2$ ) was highest, when daily total tryptophan intake was used as the independent variable. The estimate of bioavailability of tryptophan in soybean meal relative to L-tryptophan was 106% when based on a common-intercept, multiple linear regression of partitioned daily weight gain (kg/day) on supplemental tryptophan intake (Table 4).

#### *Tryptophan Bioavailability Based on Gain:Feed Ratio*

Common-intercept, multiple linear regression of dependent variable gain:feed ratio on the independent variable supplemental tryptophan level gave 90% as the estimate of bioavailability of tryptophan in soybean meal relative to L-tryptophan (Table 4). The use of daily total or supplemental tryptophan intake as the independent variables gave estimates of 95 or 104%, respectively.

#### **Discussion**

Bioavailability assays based on animal performance include all of the processes of digestion, absorption, and utilization of the specific limiting amino acid. The purpose of choosing several combinations of dependent and independent variables to analyze the data statistically was to demonstrate that the estimates of bioavailability generated depend on the method used for their derivation. It is clear from the present experiment that bioavailability estimates for a given amino acid in a feed ingredient vary with the response criterion, and with the independent variable for a given response criterion. With daily supplemental tryptophan intake as the independent variable, estimates of tryptophan bioavailability were 106 and 110% for partitioned and non-partitioned weight gains, respectively, and 104% for gain:feed ratio. None of these estimates of bioavailability is different from 100%, as reflected in the standard errors presented in Table 4. For a given response criterion such as weight gain, estimates of tryptophan bioavailability in soybean meal range from 90 to 110%, depending on the independent variable used in the common intercept, multiple linear regression model.

As a result of the increase in voluntary feed intake with increasing tryptophan level from either the L-tryptophan or soybean meal diets, tryptophan intake from ingredients in the basal diet increased. This implies that part of the increase in weight gain of pigs fed the supplemented diets was due to consumption of more tryptophan from ingredients in the basal diet. As discussed by Netke and Scott (1970), the partitioning of growth to reflect weight gain solely attributable to supplemental tryptophan represents an attempt to adjust for differences in feed intake. The bioavailability estimate from partitioned weight gain (106%) was lower than that from non-partitioned weight gain (110%).

Bioavailability estimates greater than 100% imply that the dependent/independent variable combination is influenced by a variety of nutritional factors. One factor is a more rapid absorption of crystalline tryptophan relative to amino acids in proteins when diets containing free tryptophan are fed. The likely consequence of a more rapid absorption of crystalline tryptophan is a reduction in efficiency

of utilization for growth, because it reaches utilization sites when other amino acids are not available in proper amounts for use in protein synthesis. Other factors might include peptides in soybean meal that could be stimulatory to digestion/absorption, and the profile of amino acids in diets containing soybean meal.

### **Implications**

The purpose of the experiment was to present trends in estimates of bioavailability with each dependent/independent variable combination. The experiment indicates that a nutritionist should not be dogmatic about the correctness of an estimate of bioavailability generated by a particular dependent/independent variable combination. Each combination introduces its own bias and may not produce a more definitive answer. Estimates of tryptophan bioavailability in soybean meal for young pigs vary with dependent and independent variable combination used in the regression analysis and range from 90 to 117%.

### **References**

- Adeola, O., B.V. Lawrence, and T.R. Cline. 1994. Availability of amino acids for 10- to 20-kilogram pigs: Lysine and threonine in soybean meal. *J. Anim. Sci.* 72:2061.
- Blair, M. 1995. An update on the amino acid values of feed ingredients: The 1994 corn crop. Proceedings of the 1995 Degussa Technical Symposium. pp 51-56.
- Finney, D.J. 1978. *Statistical Methods in Biological Assay* (3rd Ed.). Charles Griffin & Co., London.
- Kovar, J.L., A.J. Lewis, T.R. Radke, and P.S. Miller. 1993. Bioavailability of threonine in soybean meal for young pigs. *J. Anim. Sci.* 71:2133.
- Netke, S.P., and H.M. Scott. 1970. Estimates on the availability of amino acids in soybean oil meal as determined by chick growth assay: Methodology as applied to lysine. *J. Nutr.* 100:281.
- NRC. 1988. *Nutrient Requirements of Swine* (9th Ed.). National Academy Press, Washington, DC.
- Sato, H., T. Kobayashi, R.W. Jones, and R.A. Easter. 1987. Tryptophan availability of some feedstuffs determined by pig growth assay. *J. Anim. Sci.* 64:191.

Table 1. Composition (g/kg) of basal diet on as-fed basis<sup>a</sup>.

Ingredient	
Corn (7.5% CP)	280
Cornstarch <sup>b</sup>	275.9
Corn gluten meal (60% CP)	220
Sucrose	50
Skim milk powder (34% CP)	50
Corn oil	40
Solka Floc	30
L-Lysine•HCl	7.5
Mineral, vitamin and antibiotic mix <sup>c</sup>	46.6
Total	1000.0

<sup>a</sup> Calculated tryptophan content of basal diet was 1.1 g/kg.

<sup>b</sup> L-tryptophan or soybean meal replaced cornstarch in the reference or test diets, respectively. Test diets contained 32 or 64 g of soybean meal/kg of diet.

<sup>c</sup> Provided per kilogram of diet: 6.3 g Ca; 4.8 g P; .9 g K; .55 g Mg; 4.9 g NaCl; 179 mg Fe (as FeSO<sub>4</sub>·H<sub>2</sub>O); 60 mg Mn (as MnO); 150 mg Zn (as ZnO); 17.5 mg Cu (as CuO); 3 mg I (as Ca(IO<sub>3</sub>)<sub>2</sub>); 300 µg Se; 6,108 IU vitamin A; 600 IU vitamin D<sub>3</sub>; 23 IU vitamin E; 1.2 mg menadione sodium bisulfite; 31 µg vitamin B<sub>12</sub>; 6 mg riboflavin; 22.5 mg d-pantothenic acid; 35 mg niacin; 520 mg choline; 1 mg thiamin; .3 mg folic acid; 1.5 mg pyridoxine; 10.4 mg tylosin phosphate.

Table 2. Determined composition of corn, corn gluten meal, soybean meal, and basal diet on an as-fed basis.

Nutrient	Corn	Corn gluten meal	Soybean meal	Basal diet
Dry matter, g/kg	881.5	901.9	889.9	893.2
Ether extract, g/kg	33.3	22.7	21.3	54.9
CP, g/kg	71.9	598.1	467.4	163.7
Indispensable amino acids, g/kg				
Arginine	3.3	19.1	34.1	5.4
Histidine	2.1	12.4	12.6	3.7
Isoleucine	2.5	23.1	20.7	6.8
Leucine	8.2	95.9	35.9	24.4
Lysine	2.5	10.9	29.5	10.4
Methionine	1.7	15.4	6.6	4.1
Phenylalanine	3.5	38.0	23.8	9.8
Threonine	2.6	20.1	18.2	5.6
Tryptophan	.6	3.2	6.2	1.03
Valine	3.3	26.2	21.6	7.7
Dispensable amino acids, g/kg				
Alanine	5.1	51.8	20.2	13.1
Aspartate	4.9	36.4	51.9	10.4
Cystine	1.9	12.1	7	3.3
Glutamate	12.3	119.2	79.2	32.7
Glycine	3.1	16.9	19.5	4.7
Proline	6.2	54.5	23.6	15.1
Serine	3.2	27.7	20.6	7.1
Tyrosine	2.1	31.7	16.7	7.3

Table 3. Final weight, ADG, ADFI, and gain:feed ratio of pigs fed diets containing graded levels of supplemental tryptophan from L-tryptophan or soybean meal.

Tryptophan source:	Basal	L-tryptophan			Soybean meal		
Total tryptophan, g/kg:	1.03	1.23	1.43	1.63	1.23	1.43	
Supplemental Tryptophan, g/kg:	0	.2	.4	.6	.2	.4	SE
<u>Item</u>							
Tryptophan intake, g/d <sup>abc</sup>	.582	.865	1.242	1.381	.891	1.094	.028
Supplemental Tryptophan intake, g/d <sup>abc</sup>	0	.144	.354	.517	.149	.312	.019
Initial weight, lb	21.52	21.60	21.60	21.23	21.36	21.52	.20
Final weight, lb <sup>ab</sup>	29.35	34.91	41.03	46.11	33.97	39.71	1.50
ADG, lb <sup>ab</sup>	.279	.475	.695	.887	.449	.649	.053
ADFI, lb <sup>abc</sup>	1.280	1.586	1.949	1.896	1.635	1.718	.090
Gain:feed, lb/lb <sup>ab</sup>	.216	.288	.352	.488	.271	.361	.028
N	12	12	12	12	12	12	

<sup>a</sup> Linear response ( $P < .05$ ) to added tryptophan from L-tryptophan.

<sup>b</sup> Linear response ( $P < .05$ ) to added tryptophan from soybean meal.

<sup>c</sup> Quadratic response ( $P < .05$ ) to added tryptophan from L-tryptophan.

Table 4. Estimates of bioavailability of tryptophan in soybean meal from daily weight gain (kg/day) and gain:feed ratio (kg/kg) as response criteria, using supplemental dietary tryptophan level (g/kg), daily total tryptophan intake (g/day), or daily supplemental tryptophan intake (g/day) as an independent variable.

Dependent variable	Independent variable	Common-intercept multiple regression equation	R <sup>2</sup>	SE	Relative bioavailability, %	Approx. SE, rel. bioavailability
ADG	Supplemental tryptophan level	$Y = .125 + .467X_1 + .42X_2$	.451	.028	90	11.7
ADG	Total tryptophan intake	$Y = -.059 + .318X_1 + .315X_2$	.726	.021	99	3.8
ADG	Supplemental tryptophan intake	$Y = .114 + .577X_1 + .636X_2$	.646	.023	110	9.6
Partitioned ADG	Supplemental tryptophan intake	$Y = .34X_1 + .359X_2$	.457	.020	106	14.0
Gain:feed	Supplemental tryptophan level	$Y = .201 + .441X_1 + .397X_2$	.446	.027	90	11.8
Gain:feed	Total tryptophan intake	$Y = .145 + .186X_1 + .176X_2$	.275	.032	95	9.9
Gain:feed	Supplemental tryptophan intake	$Y = .224 + .421X_1 + .439X_2$	.375	.029	104	16.4