AMINO ACID DIGESTIBILITY TO PIGS IN VARIOUS FIBER SOURCES
2. TRUE DIGESTIBILITY OF AMINO ACIDS IN ILEAL DIGESTA AND FECES

A. Nongyao[1], In K. Han[2] and Y. J. Choi

College of Agriculture
Seoul National University, Suweon 441-744, Korea

Summary

The effects of dietary fiber on true digestibility of amino acids by growing pigs were studied, using semi-purified diets formulated from alfalfa meal, cassava leaf meal, rubber seed meal and leucaena meal at 20% level. A protein-free diet including 5% cellulose was formulated for correcting the endogenous amino acid loss. Across all the diets, arginine was the most digestible while the least at ileal level was threonine; methionine and/or histidine at fecal level respectively. The true digestibility value of amino acids at ileal level were higher than at fecal level except control diet (cellulose). The true digestibility values at ileal level were similar for all diets but differed at fecal level in different magnitude. These results indicate that undigestible compound in individual feedstuff might confound. True digestibility should be studied together for accurate diet formulation as apparent digestibility decreased when their amino acid concentration in the diet was reduced.

(**Key Words**: Dietary Fiber, True Amino Acid Digestibility, Apparent Amino Acid Digestibility, Cannulated Pigs)

Introduction

Fibrous feedstuff varied in constituents and chemical composition depend on sources and species. The nutritive value of amino acids were depressed by high fiber had been reported. The magnitude of this negative influence depends on the type and origin of fiber source (Fernandez and Jorgensen, 1986). Values for amino acid digestibility have been determined at ileal and fecal level. However, apparent digestibility is influenced by the protein level in the diet (Taverner et al., 1981) and confounded with non-reabsorbed endogenous amino acids (Low, 1982).

Apparent digestibility values increased with increasing protein content especially at lower protein levels. Feedstuffs harvested from plant leaves always contain low protein and high fiber. It is expected that true, rather than apparent digestibility value will more accurate for diet formulation. The present study was carried out to determine the true digestibility to pigs of amino acids as affected by various fiber sources and also to provide a comparison with apparent digestibility values.

Materials and Methods

Design

The animals and the experimental diets used in present study were originated from previous experiment that have been published (Nongyao et al., 1990). For determining true digestibility value, a protein-free diet was formulated including 5% cellulose as shown in table 1. The collection procedure of ileal and fecal samples were carried out by Jorgensen et al. (1984) method. The proximate composition were analyzed for all diets and collected samples by AOAC (1984) procedure. The concentration of chromic oxide were determined using Fenton and Fenton (1979) method. Georing and Van Soest (1970) method was used to assay the content of fiber fractions. Amino acid contents in feed, feces and ileal digesta were analyzed by auto amino acid analyzer model KLB 4150 alpha.

Calculation of digestibility

Digestibility values were determined by refe-

[1]Present address: Faculty of Agriculture and Industrial Science, Chandrakasem Teachers' College, Bangkok 10900, Thailand.

[2]Address reprint requests to Dr. In K. Han, Animal Science Department, College of Agriculture, Seoul National University, Suweon 441-744, Korea.

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TABLE 1. FORMULATION OF PROTEIN-FREE DIET

Ingredients	%
Corn starch	68
Glucose	20
Corn oil	3
Cellulose	5
Vit. Min. premix[1]	0.25
Salt	0.25
Monosodium PO_4	0.50
Dicalcium-PO_4	2.50
Chromic oxide	0.50
Total	100.0

[1]Contributed the following nutrients per kg of diet: Zn, 100 mg; Cu, 10 mg; Mn, 20 mg; Fe, 150 mg; Se, 1 mg; Vitamin A, 4,000,000 IU; Vitamin D_3, 800,000 IU; Vitamin E, 6,000 IU; Vitamin K_3, 1,400 mg; thiamin, 800 mg; riboflavin, 2,000 mg; pyridoxine, 800 mg; cobalamine, 6,500 mg; Ca-pantothenate, 6,000 mg; niacin, 10,000 mg; folacin, 400 mg and ethoxyquine, 500 mg.

rence to relative concentrations of Cr in the diet and collected samples. True digestibility equation introduced by Dabrowski and Dabrowska (1981) were slightly changed and used in this study as follows.

True digestibility (%) =

$$100 \times \left\{ \frac{\frac{N\ feed}{Cr\ feed} - \frac{N\ feces}{Cr\ feces} - \frac{N\ metabolic}{Cr\ metabolic}}{\frac{N\ feed}{Cr\ feed}} \right\}$$

Where N = examined constituent (%)
Cr = chromium oxide (%)

Feces = it can mean the digesta from ileum

Metabolic = the level of examined constituents in ileum or total tract from pigs fed protein-free diet

Statistical analyses

Analysis of variance were used for identifying the true digestibility values as affected by different fiber sources. Duncan's New Multiple Range Test was used to compared the treatment mean. These data were analyzed using Proc Anova of SAS (1985) procedure.

Results and Discussion

Endogenous amino acid level

The endogenous values of amino acid output used to calculate the digestibility value of amino acids at ileum and fecal were presented in table 2. Using these values, it was calculated to correct the endogenous loss caused by dietary fiber for present study. It was shown that most of endogenous amino acid at ileal level were higher than fecal level. Glycine and proline were the prominent of endogenous amino acid, were well agreed with de Lange et al. (1989) and Furuya and Kaji (1987). Of the indispensable amino acids, histidine, phenylalanine and threonine were the prominent.

True ileal digestibility value

The true digestibility of arginine in cassava leaf meal diet was found to be the greatest among natural fibrous group whereas threonine was the least for all amino acids digestibility (table 3). Arginine, histidine, lysine and methionine

TABLE 2. ENDOGENOUS ILEAL AND FECAL LEVEL OF AMINO ACIDS IN PIGS (%)

Indispensable amino acids			Dispensable amino acids		
	Ileal	Fecal		Ileal	Fecal
Arg	0.25	0.18	Ala	0.33	0.32
His	0.57	0.32	Asp	0.40	0.41
Ile	0.23	0.23	Glu	0.69	0.61
Leu	0.33	0.41	Gly	1.31	0.28
Lys	0.13	0.22	Pro	2.24	0.25
Met	0.13	0.10	Ser	0.31	0.23
Phe	0.46	0.28	Tyr	0.23	0.16
Thr	0.37	0.25			
Val	0.30	0.28			

TABLE 3. EFFECT OF FIBER SOURCES ON TRUE AMINO ACIDS DIGESTIBILITIES MEASURED AT THE TERMINAL ILEUM OF GROWING PIGS (%)

Component	Dietary treatments[1]				
	C	AFM	CLM	RSM	LM
Dietary indispensable amino acids[2]					
Arginine	98.09^{a}	95.53^{a}	98.80^{a}	97.63^{a}	95.82^{a}
Histidine	99.15^{a}	95.40^{a}	97.57^{a}	92.14^{a}	94.51^{a}
Isoleucine	89.29^{bc}	94.01^{ab}	95.36^{a}	88.68^{bc}	86.27^{c}
Leucine	92.31^{bc}	95.09^{ab}	96.71^{a}	93.25^{abc}	89.74^{c}
Lysine	88.80^{a}	88.89^{a}	95.50^{a}	92.74^{a}	92.35^{a}
Methionine	92.72^{a}	93.94^{a}	95.79^{a}	94.26^{a}	94.79^{a}
Phenylalanine	91.34^{b}	98.08^{a}	97.20^{a}	95.13^{ab}	92.26^{b}
Threonine	92.91^{ab}	77.87^{b}	95.83^{a}	90.62^{ab}	88.78^{ab}
Valine	86.09^{b}	93.44^{ab}	94.89^{a}	88.94^{ab}	85.69^{b}
Dietary dispensable amino acids					
Alanine	92.65^{ab}	94.18^{ab}	96.07^{a}	91.64^{bc}	88.36^{c}
Aspartic acid	95.16^{ab}	94.34^{ab}	97.22^{a}	92.24^{b}	91.78^{b}
Glutamine	96.21^{ab}	94.56^{b}	97.76^{a}	95.86^{ab}	95.49^{ab}
Glycine	100.64^{b}	113.26^{a}	100.15^{a}	97.49^{b}	107.11^{ab}
Proline	126.07^{a}	136.62^{a}	98.97^{a}	108.03^{a}	110.64^{a}
Serine	97.71^{a}	96.99^{ab}	97.99^{a}	88.87^{b}	94.85^{ab}
Tyrosine	91.06^{ab}	97.69^{a}	95.63^{ab}	94.00^{ab}	90.68^{b}
Mean	95.79	97.20	96.93	93.77	93.54

[1]Abbreviated: C, control; AFM, alfalfa meal; CLM, cassava leaf meal; RSM, rubber seed meal; LM, leucaena meal.

[2]Means with the different superscripts within the same row were significant difference ($p < 0.05$).

in all diets were found no significant difference in digestibility values. All of amino acids found in cassava leaf meal diet were easily digestible. Among the treatments, the true digestibilities of isoleucine, leucine, threonine, alanine and serine were lower than the true digestibility of other amino acids. Proline and probably glycine recovered in ileal digesta could be overestimated when the protein-free diet was fed, resulting in calculated true amino acid digestibility exceeding 100 % (de Lange et al., 1989). These findings were well agreed with those documents. The lowest true ileal digestibility for threonine found in AFM-diet in this study agreed with prior reports for soya-bean meal (Furuya et al., 1986), rapeseed meal (Sauer et al., 1982), casein (Kies et al.,1986) and wheat (Furuya and Kaji, 1987) which might have resulted from the low absorption rate of threonine in the small intestine (Zebrowska, 1979).

True fecal digestibility value

Glutamine was also found to exceed 100% (table 4). This could be explained by the fact that the tissue of intestinal tract takes up large quantities of glutamine which can be mobilized to glutamate plus ammonia, citrulline and proline when fed a protein-free diet as described by Rodwell (1985). Alanine showed low true digestibility at fecal level found in alfalfa meal, rubber seed meal as well as leucaena meal diet and in leucaena meal diet at ileal level. These findings could be explained that more than 50% of the total α-amino acid was released from the muscle tissue of the pig fed protein-free diet (Rodwell, 1985), and supplied to the lumen of the gut. The lowered digestibility value of glycine found at fecal level in alfalfa meal, rubber seed meal and leucaena meal diet were similar to that reviewed by Corring (1982), in which the less well absorbed amino acid along the whole digestive tract were cystine and glycine, and that glycine was highly predominant in a digestive secretion such as a bile. Methionine and/or histidine were found to be the least digestible indispensable amino acid

TABLE 4. EFFECT OF FIBER SOURCES ON TRUE AMINO ACIDS DIGESTIBILITIES MEASURED BY THE TOTAL TRACT OF GROWING PIGS (%)

Component	Dietary treatments[1]				
	C	AFM	CLM	RSM	LM
Dietary indispensable amino acids[2]					
Arginine	99.74[a]	85.02[b]	92.69[b]	85.61[b]	90.62[b]
Histidine	97.06[a]	60.54[bc]	83.86[ab]	50.12[c]	78.01[ab]
Isoleucine	99.70[a]	63.64[c]	86.88[ab]	65.77[c]	80.90[bc]
Leucine	99.49[a]	69.76[c]	88.55[ab]	77.23[bc]	81.68[bc]
Lysine	98.12[a]	72.86[c]	90.92[ab]	82.19[bc]	85.29[b]
Methionine	97.83[a]	51.85[c]	84.06[ab]	67.58[bc]	76.51[b]
Phenylalanine	99.29[a]	68.54[c]	88.30[ab]	77.32[bc]	81.03[b]
Threonine	99.73[a]	62.25[c]	89.32[ab]	74.63[bc]	80.82[b]
Valine	99.88[a]	61.82[c]	85.17[ab]	65.47[bc]	76.30[bc]
Dietary dispensable amino acids					
Alanine	99.27[a]	56.73[c]	85.06[ab]	69.95[b]	76.11[a]
Aspartic acid	99.23[a]	78.52[b]	93.33[a]	84.62[ab]	88.64[a]
Glutamine	100.09[a]	81.65[d]	95.07[ab]	87.79[cd]	91.30[bc]
Glycine	99.51[a]	56.21[c]	86.60[ab]	68.93[bc]	77.60[b]
Proline	100.01[a]	73.13[b]	89.80[ab]	72.96[b]	83.02[ab]
Serine	99.69[a]	79.84[c]	92.02[ab]	81.30[c]	85.24[bc]
Tyrosine	97.76[a]	68.52[c]	85.97[ab]	77.39[bc]	82.56[bc]
Mean	99.14	68.07	88.55	74.15	82.17

[1]Abbreviated: C, control; AFM, alfalfa meal; CLM, cassava leaf meal; RSM, rubber seed meal; LM, leucaena meal.

[2]Means with the different superscripts within the same row were significant difference ($p < 0.05$).

among treatments. Alfalfa meal diet was the least digestible, lower than cassava leaf meal-diet, leucaena meal diet and rubber seed meal diet. The low digestible values of amino acids at fecal level for all diets, except control diet were found. Since these diets were already corrected for the endogenous loss caused by fiber but the digestibility values were still low. It indicated that these feedstuffs contained some undigestible complex which could not digest by animal enzymes.

Comparison on true ileal and fecal digestibility values

In general, the true digestibility of amino acids at ileal were higher than fecal level except control diet as shown in table 5. This was probably from the fact that cellulose in control diet differed from natural cellulosic materials in their physical and biological properties (Van Soest, 1978) and they might be more resistant than normal cellulose to microbial degradation in the large intestine of pigs (Stanogias and Pearce, 1985). Histidine, glycine and proline in control diet were found higher at ileal than fecal level. The magnitude of difference were ranged from 8.37, 10.84, 18.92, 8.37 to 28.69% for cassava leaf meal diet, leucaena meal diet, rubber seed meal diet and alfalfa meal diet, respectively.

Comparison on apparent and true digestibility

The apparent and true digestibility of each amino acid in various diets containing different fiber sources were compared in table 6. Among the indispensable amino acid group, the digestibility value of arginine in cassava leaf meal was found to be the highest. Methionine was found to be the lowest value in apparent digestibility from leucaena meal diet indicating that this kind of amino acid could be bound with lignin. However, the branched chain amino acids, such as valine, leucine and isoleucine tended to be low

TABLE 5. COMPARISON ON THE TRUE DIGESTIBILITY OF AMINO ACIDS MEASURED THE TERMINAL ILEUM AND THE TOTAL TRACT (%)

Components	Dietary treatments[1]				
	C	AFM	CLM	RSM	LM
Dietary indispensable amino acids					
Arginine	−1.72	10.51	6.11	12.02	5.20
Histidine	2.09	34.86	13.71	42.02	16.50
Isoleucine	−10.41	30.37	8.48	22.91	5.37
Leucine	−7.18	25.33	8.16	16.02	8.06
Lysine	- 9.32	16.03	4.58	0.55	7.06
Methionine	−5.11	42.09	11.73	26.68	18.28
Phenylalanine	−7.95	29.54	8.90	17.81	1.23
Threonine	- 6.82	15.62	6.51	15.99	7.96
Valine	−13.80	31.62	9.72	23.47	9.39
Dietary dispensable amino acids					
Alanine	−6.62	37.45	11.01	21.69	12.25
Aspartic acid	−4.07	15.82	3.89	7.62	3.14
Glutamine	−3.88	2.91	2.69	8.07	4.19
Glycine	7.13	57.05	13.55	28.56	29.51
Proline	26.06	63.49	9.17	35.07	27.62
Serine	−1.98	17.15	5.97	7.57	9.61
Tyrosine	- 6.70	29.17	9.66	16.61	8.12
Mean	−3.14	28.69	8.37	18.92	10.84

[1]Abbreviated: C, control; AFM, alfalfa meal; CLM, cassava leaf meal; RSM, rubber seed meal; LM, leucaena meal.

in digestibility values. Similar findings were made by Laplace et al. (1989). This could suggest that these amino acids were absorbed to a smaller extent than the other amino acids.

Of the dispensable amino acid group, true digestibility value of almost all amino acids in alfalfa meal diet except proline were low. Apparent digestibility value of proline in alfalfa meal diet was largely lower than true digestibility, and revealed that endogenous loss was greater than other diets. The true digestibility values of proline was not significantly different in all diets. This may be caused by high NDF value which the sloughing process of the mucus cell as well as enzymatic system interference concerned. True digestibility values of glutamine were higher than that of other amino acids across all diets. This revealed the fact that the tissue of the intestinal tract took up large quantities of glutamine which could be mobilized to glutamate as well as proline in compensate of meatabolic function when fed with high fiber diet. The notable depression of alanine apparent and true digestibility values resulted from the abundant of more than 50% of α-amino acid released from the muscle tissue, as decribed by Rodwell (1985).

The results of present data showed that adding fiber to the diet caused increased excretion of fecal protein and thus reduced both apparent and true protein digestibilities. The endogenous loss resulting from adding crude fiber had been found and responsible for the increased excretion of endogenous ileal and fecal nitrogen. These were supported with reports by many investigators. Increased losses of endogenous fecal nitrogen might be due to number of factors. Increased secretion of digestive enzymes (trypsin, chymotrypsin, lipase and amylase) had been shown to occur. Endogenous fecal nitrogen as well as amino acids included digestive glands secretion, desquamated cells from active replacement of gastro-intestinal mucosal lining and small amount of plasma protein (Matthews, 1975). The possibility that fiber might increased the sloughing of inte-

TABLE 6. COMPARISON OF APPARENT AND TRUE AMINO ACIDS DIGESTIBILITIES AS AFFECTED BY VARIOUS FIBER SOURCES REGARDLESS THE COLLECTION SITE (%)

Components		Dietary treatments[1]				
		C	AFM	CLM	RSM	LM
Indispensable amino acids						
Arginine	app	90.77^{ab}	88.24^{b}	95.20^{a}	86.43^{b}	88.24^{b}
	true	98.91^{a}	89.90^{b}	95.47^{ab}	93.22^{ab}	91.08^{b}
Histidine	app	73.89^{ab}	70.34^{ab}	87.91^{a}	65.37^{b}	66.33^{b}
	true	98.12^{a}	76.39^{bc}	90.09^{ab}	82.26^{abc}	69.21^{c}
Isoleucine	app	77.18^{ab}	79.92^{b}	89.73^{a}	68.85^{b}	75.08^{ab}
	true	94.49^{a}	77.44^{b}	90.73^{a}	83.59^{ab}	76.18^{b}
Leucine	app	81.32^{ab}	79.05^{b}	91.89^{a}	72.14^{b}	83.67^{ab}
	true	95.90^{a}	81.27^{c}	92.26^{ab}	85.71^{bc}	84.51^{bc}
Lysine	app	83.25^{bc}	78.60^{bc}	92.98^{a}	77.08^{c}	86.65^{ab}
	true	93.46^{a}	80.15^{b}	93.00^{a}	88.82^{a}	86.98^{ab}
Methionine	app	70.35^{b}	67.09^{b}	83.73^{a}	60.47^{b}	77.48^{ab}
	true	92.27^{a}	70.98^{b}	89.39^{a}	86.65^{ab}	79.91^{ab}
Phenylalanine	app	77.10^{ab}	65.36^{b}	91.00^{a}	68.31^{b}	79.86^{a}
	true	95.31^{a}	81.97^{b}	92.35^{ab}	87.03^{ab}	85.42^{ab}
Threonine	app	75.66^{a}	71.99^{b}	88.57^{a}	64.98^{b}	74.38^{ab}
	true	96.32^{a}	69.35^{c}	92.28^{ab}	84.80^{ab}	81.90^{b}
Valine	app	75.66^{a}	71.99^{b}	88.57^{a}	64.98^{b}	74.38^{ab}
	true	92.98^{a}	76.19^{b}	89.59^{ab}	80.99^{ab}	76.14^{b}
Dispensable amino acids						
Alanine	app	73.65^{ab}	70.59^{b}	89.57^{a}	62.77^{b}	78.15^{ab}
	true	95.96^{a}	73.75^{c}	90.07^{ab}	82.24^{bc}	79.65^{bc}
Aspartic acid	app	84.18^{b}	84.17^{b}	94.34^{a}	81.80^{b}	87.41^{b}
	true	93.69^{ab}	85.71^{c}	95.10^{a}	90.21^{abc}	88.09^{bc}
Glutamic acid	app	88.34^{a}	85.75^{a}	91.05^{a}	84.88^{a}	90.76^{a}
	true	98.15^{a}	87.52^{c}	96.29^{ab}	93.40^{ab}	91.46^{bc}
Glycine	app	75.53^{ab}	66.73^{b}	89.57^{a}	63.22^{b}	75.78^{a}
	true	103.76^{a}	84.14^{b}	92.76^{ab}	92.35^{ab}	81.91^{b}
Proline	app	84.13^{a}	46.55^{b}	91.73^{a}	76.12^{a}	80.47^{a}
	true	113.04^{a}	101.99^{a}	93.97^{a}	96.83^{a}	88.90^{a}
Serine	app	87.52^{a}	85.79^{ab}	93.87^{a}	80.43^{b}	86.39^{a}
	true	98.70^{a}	87.63^{bc}	94.74^{ab}	90.04^{bc}	84.79^{c}
Tyrosine	app	79.45^{ab}	78.93^{ab}	89.53^{a}	72.70^{b}	82.75^{ab}
	true	94.41^{a}	81.78^{b}	90.36^{ab}	86.62^{ab}	89.84^{ab}

[1]Abbreviated: C, control diet; AFM, alfalfa meal diet; CLM, cassava leaf meal diet; RSM, rubber seed meal diet; LM, leucaena meal diet; app, apparent.

[2]Means with the same superscripts within the same row were significant difference ($p < 0.05$).

stinal mucosal cell had been also suggested (Bergner and Zimmer, 1975; Sheard and Schneeman, 1980). A lowering of intestinal reabsorption of endogenous amino acids secreted into the gut had been observed with a fiber-supplemented diet (Bergner and Zimmer, 1975). Any reduction in intestinal transit time associated with fiber-containing diets differed in physical characteristics could also leave less time for digestion and absorption. In the present study, it should be noted that apparent digestibility of amino acids always decreased when their concentration in the diet was reduced. For this reason, for more accurate diet formulation, further studies on the digestible amino acid requirement should be performed together with the true ileal digestibility. In conclusion, the true ileal digestibility values of amino acids were higher at ileal than fecal level. The most digestible amino acid across all the diets was arginine whereas the least was threonine at ileal level and, methionine and/or histidine at fecal level. The true digestibilities at ileal level were similar for all diets. At fecal level the difference of digestibility was found among the diets with different magnitude. Cassava leaf meal diet was the most digestible diet among the natural fibrous feedstuff while the least at ileal level was leucaena meal diet and alfalfa meal diet at fecal level.

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