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Effects of Dietary Energy Concentration and Lysine on the Digestible Energy Ratio for Apparent Amino Acid Digestibility in Finishing Barrows

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ABSTRACT : This experiment was performed to investigate the effects of two energy levels and four lysine:digestible energy (DE) ratios on the apparent digestibility of nutrients in finishing pigs. The experiment was conducted using a 2×4 randomized complete block (RCB) design with three replicates. Twenty-four cross-bred finishing barrows ((Landrace×Yorkshire)×Duroc) with an average body weight of 64.2±0.69 kg were assigned to one of eight treatments. Each barrow was placed in an individual metabolism crate and dietary treatment and water was provided *ad libitum*. Diets were designed to contain lysine:ME ratios of 1.5, 1.8, 2.1 and 2.4 g/Mcal at 3.35 and 3.6 Mcal/kg of diet in a 4×2 factorial arrangement. Dry matter (DM), ash, Ca and P digestibility were not affected by energy density or lysine:DE ratios. Crude fat digestibility increased as the energy density increased from 3.35 to 3.6 Mcal of DE/kg. Increasing the lysine:DE ratio also increased crude protein digestibility. There were no interactions between energy density and lysine:DE ratio in terms of nutrient digestibility. Nitrogen excretion via feces was not affected by energy density and lysine:DE ratio, while nitrogen excretion via urine was significantly affected by energy density and lysine:DE ratio. The apparent digestibility of all amino acids except for isoluecine, arginine and aspartic acid as well as average values of essential amino (EAA), non-essential amino acids (NEAA) and total amino acid digestibility ($p>0.05$) were not affected by energy density. The apparent digestibility of all amino acids except for leucine, proline, alanine and tyrosine, NEAA and total amino acid digestibility were significantly affected by lysine: DE ratio ($p<0.05$). Interactive effects of energy and lysine:DE ratio also significantly affected amino acid digestibility except for isoleucine, alanine, cystine, leucine, phenylalanine, glutamine and proline ($p<0.05$). In conclusion, these results suggest that maintaining the appropriate lysine:DE ratio becomes more important as the energy density of the diet increases. Consequently, increasing the lysine:DE ratio can result in increased crude protein digestibility and urinary nitrogen excretion, although apparent protein digestibility and nitrogen excretion were not affected by energy density Furthermore, increasing the lysine:DE ratio also increased the apparent digestibility of essential amino acids, except for leucine, regardless of energy density. The optimum lysine:DE ratio for maximum essential amino acid digestibility of the 64.2±0.69 kg pig is approximately 2.4 g of lysine/Mcal of DE. (**Key Words :** Barrows, Lysine:DE Ratio, Amino Acid Digestibility, Urinary Nitrogen)

INTRODUCTION

Recently the tendency of pork industry has been to maximize lean growth in pigs which is sufficient the lean growth demand of the consumers through of genetic selection and the nutrition system. Consequently, a lot of studies were conducted to figure out the specific ratio of amino acids and the energy level for maximum lean pork growth becoming accomplished regardless of animal's genetic potential.

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Feed intake and amino acids intake have been reported to decrease with increasing the DE content in diet when pigs had fed *ad libitum* (Campbell and Dunkin, 1983). We hypothesize that the amino acid levels of a diet should be related to its DE concentration. Several studies have been conducted on a wide range of ratios of total lysine:energy (Campbell et al., 1988; Nam and Ahere, 1994; Urynek et al., 2003; Tang et al., 2007). In a trial using barley-based diets, Campbell et al. (1988) observed maximal growth in gilts, increasing in weight by 20 to 60 kg when the diet contained 2.97 g lysine/Mcal DE. Castell et al. (1994) and Blanchard et al. (1999) reported that the consumption of diets with low amino acid:energy ratios resulted in a higher carcass and intramuscular fat levels. This is in contrast to findings by Gahl et al. (1991), Heger and Frydrich (1985) and

Table 1. Formula and chemical composition of the experimental diets

DE level	3.35 Mcal of DE/kg				3.60 Mcal of DE/kg			
g of lysine/Mcal of DE	1.5	1.8	2.1	2.4	1.5	1.8	2.1	2.4
Ingredients :								
Corn	85.60	81.26	75.92	70.19	86.16	80.35	73.89	69.90
Corn gluten feed	7.99	8.37	8.50	8.75	3.95	4.25	4.66	4.65
Soybean meal (44%)	0.48	4.54	9.82	15.39	3.58	9.19	15.37	19.40
Molasses	3.50	3.50	3.50	3.50	-	-	-	-
Tallow	-	-	-	-	4.00	4.00	4.00	4.00
Limestone	0.51	0.78	0.70	0.67	0.60	0.66	0.72	0.76
Tricalcium phosphate	0.93	0.58	0.63	0.61	0.75	0.63	0.50	0.42
NaCl	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
L-Lysine·HCl	0.22	0.21	0.17	0.12	0.19	0.15	0.09	0.10
Mineral mixture[1]	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Vitamin mixture[2]	0.27	0.27	0.27	0.27	0.27	0.27	0.27	0.27
Total	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00
Chemical composition[3]								
DE (kcal/kg)	3,350	3,350	3,350	3,350	3,600	3,600	3,600	3,600
Crude protein (%)	9.4	10.9	12.8	14.8	9.8	11.8	14.1	15.5
Lysine (%)	0.50	0.60	0.70	0.80	0.54	0.65	0.76	0.87
Methionine (%)	0.18	0.19	0.21	0.23	0.18	0.20	0.22	0.24
Ca (%)	0.55	0.55	0.55	0.55	0.50	0.50	0.50	0.50
Total P (%)	0.50	0.45	0.48	0.50	0.4	0.45	0.45	0.45

[1] Provided the following per kilogram of diet: choline chloride 700 mg; selenium 0.15 mg; manganese 0.03 g; zinc 0.1 g; iron 0.1 g; iodine 0.5 mg; magnesium 0.1 g.

[2] Provided the following per kilogram of diet: vitamin A 5,500 IU; vitamin D_3 550 IU; vitamin E 27 IU; menadione sodium bisulfate 2.5 mg; pantothenic acid 27 mg; niacin 33 mg; riboflavin 5.5 mg; vitamin B_{12} 0.04 mg; thiamin 5 mg; pyridoxine 3 mg; biotin 0.24 mg; folic acid 1.5 mg.

[3] Calculated values.

Batterham et al. (1990) showing a decline in the marginal efficiency of lysine utilization as the lysine intake approached the amount required for maximum growth performance. These discrepancies in the optimum lysine:energy ratio may be ascribed to differences in the sources of the ingredients, and the genotypes and ages of the pigs used.

The apparent amino acid digestibility depended on the amino acid contents in the experimental diet. Eggum (1973) demonstrated in rats that the apparent fecal CP digestibility in soybean meal (SBM) increased curvilinearly with increasing dietary CP content. Therefore, values for apparent fecal amino acid digestibility were only meaningful under standardized conditions, at least with respect to the amino acid content in the diet. The determinations of apparent fecal and ileal digestibilities were carried out at various dietary amino acid levels (Holmes et al., 1974; JØrgensen et al., 1984; Knabe et al., 1989). However, the results from these studies did not allow for the establishment of a detailed relationship between the lysine:energy ratio and apparent amino digestibility.

Therefore, the experiment was performed to evaluate the effect of four lysine:DE ratios on apparent and fecal amino acid digestibility of nutrients in finishing pigs at two dietary energy concentrations, and to estimate the optimal lysine:energy ratio in their feed.

MATERIALS AND METHODS

This experiment utilized a 2×4 randomized complete block (RCB) design conducted in three replicates. Twenty-four cross-bred finishing barrows ((Landrace× Yorkshire)× Duroc) with an average body weight of 64.2±0.69 kg were assigned to one of eight treatments. Each barrow was placed in an individual metabolism crate and provided with dietary treatment twice daily at 08:00 and 20:00, throughout the experimental period, along with water on an *ad libitum* basis. Feed intake was adjusted to a constant rate at approximately 90% of intake for a 4-day adjustment period. Pigs were fed 2.8 times the energy required for maintenance, which was assumed to be 106 kcal per ME/kg of $BW^{0.75}$ (NRC, 1998). After a 14-day total adaptation period, feces were collected twice daily from the pigs for six consecutive days, placed in plastic bags and frozen. At the end of the collection period, the daily fecal samples from each pig were pooled and frozen. Two digestible energy (DE) levels (3.35 and 3.60 Mcal/kg) and four lysine:DE ratios (1.5, 1.8, 2.1 and 2.4 g lysine/Mcal DE) were evaluated. The formulas and calculated nutritional values of the experimental diets are presented in Table 1.

The collected samples were mixed and dried in an air forced drying oven at 55°C for 72 h and ground in a 1-mm Wiley Mill for chemical analysis. Proximate analysis of experimental diets and excreta were analyzed according to

Table 2. Effects of dietary energy level and lysine:DE ratio on the apparent digestibility of finishing barrows

DE level	3.35 Mcal of DE/kg				3.60 Mcal of DE/kg				MSE[1]
g of lysine/Mcal of DE	1.5	1.8	2.1	2.4	1.5	1.8	2.1	2.4	
DM	87.62	88.79	85.98	86.12	87.22	87.71	87.50	89.03	0.55
CP	78.98	80.46	80.88	81.76	75.81	81.21	82.93	87.41	1.07
C. fat	76.90	72.73	66.17	56.67	80.08	77.18	81.89	86.13	2.24
C. fiber	70.24	71.93	65.70	60.95	59.88	66.43	69.77	75.44	1.89
Ash	51.31	51.79	52.93	61.08	56.91	66.34	57.31	60.46	2.18
Ca	53.98	53.37	42.51	44.08	54.49	63.87	50.85	64.30	3.28
P	42.86	47.83	35.14	38.56	35.68	42.76	36.51	42.97	2.88
GE	83.94	84.74	81.27	81.25	81.91	82.57	83.53	85.15	0.79

Probability (%)	DM	CP	C. fat	C. fiber	Ash	Ca	P	GE
DE level	0.31	0.28	<0.01	0.77	0.07	0.06	0.64	0.57
Lysine:DE ratio	0.52	<0.01	0.45	0.64	0.41	0.42	0.31	0.76
DE×lysine	0.21	0.11	0.03	0.01	0.41	0.58	0.61	0.05

[1] Mean standard error.

Table 3. Effects of dietary energy level and lysine:DE ratio on the nutrient excretion of finishing pigs

DE level	3.35 Mcal of DE/kg				3.60 Mcal of DE/kg				MSE[1]
g of lysine/Mcal of DE	1.5	1.8	2.1	2.4	1.5	1.8	2.1	2.4	
Excretion (g/d)									
Fecal N	20.90	24.20	27.93	30.66	26.00	26.56	25.86	22.90	1.32
Urinary N	6.75	5.40	7.35	8.20	7.95	6.30	9.00	8.40	0.28
Total N	27.66	29.60	35.26	38.86	33.93	32.86	34.86	31.30	1.36

Probability (%)	Fecal N	Urinary N	Total N
DE level	0.73	<0.01	0.82
Lysine:DE ratio	0.47	<0.01	0.17
DE×lysine	0.08	0.44	0.06

[1] Mean standard error.

AOAC methods (1995) and the gross energy content measured on an Adiabatic Bomb Calorimeter (Model 1241, Parr Instrument Co., USA). Amino acid availability in the experimental diets was determined by acid hydrolysis with 6 N HCl at 110°C for 24 h prior to using an amino acid analyzer (Biochrom 20, Pharmacia Biotech, England). Sulfur-containing amino acids were analyzed after overnight treatment in cold performic acid oxidation and subsequent hydrolysis.

Statistical analysis was carried out by comparing means with the Duncan's multiple range test, using the General Linear Model (GLM) protocol in SAS (1985). In this study, two-way ANOVA was used to test the interactive effects of lysine:DE ratios and dietary DE levels on apparent amino digestibility.

RESULTS AND DISCUSSION

The effects of digestible energy (DE) level and four dietary lysine:DE ratios on nutrient digestibility in finishing pigs are given in Table 2. Dry matter (DM), ash, Ca and P digestibility were not affected by energy density or lysine:DE ratios. Crude fat digestibility was affected by DE level or DE×lysine:ME ratio. Increasing the DE from 3.35 to 3.6 Mcal of DE/kg increased fat digestibility. Increasing the lysine:DE ratio also increased crude protein digestibility. In pigs fed low-energy density diets, increasing the lysine: DE ratio from 1.5 to 2.1 g/Mcal increased crude protein digestibility (%) from 79.0 to 81.8. In pigs fed high-energy diets, increasing lysine:DE ratio increased crude protein digestibility up to a maximum 87.4%. However, there were no interactions between energy density and lysine:DE ratios in terms of nutrient digestibility.

The results of this study indicated that although the crude protein digestibility was unaffected by dietary DE density, increasing the DE level of the diet from 3.35 to 3.6 Mcal of DE/kg could resulted in increase crude fat digestibility. Furthermore, when sufficient digestible energy is available, increasing the lysine:DE ratio to 1.5 to 2.4 g of lysine/Mcal of DE may increase crude protein digestibility. However, we did not detect any interactions between energy density and the lysine:DE ratio. Lawrence et al. (1994) detected that energy, DM and nitrogen digestibility increased as the energy density increased from 3.5 to 3.75 Mcal of DE/kg in 20 kg pigs. This response was caused more by pigs reaching the optimum DE level than by lysine:DE ratio.

The effects of dietary energy (DE) level and four dietary lysine:DE ratios on nitrogen excretion are shown in Table 3. Fecal nitrogen excretion was unaffected by the energy density or lysine:DE ratio of the diet, while urinary nitrogen excretion increased in response to increasing the energy

Table 4. Effects of dietary energy level and lysine : DE ratio on the apparent digestibility of amino acids in finishing barrows

DE level	3.35 Mcal of DE/kg				3.60 Mcal of DE/kg				MSE[1]	
g of lysine/Mcal of DE	1.5	1.8	2.1	2.4	1.5	1.8	2.1	2.4		
Threonine**	74.15	75.80	72.33	78.60	68.36	77.55	78.59	83.35	1.48	
Valine	78.55	78.97	76.65	80.49	73.43	79.62	80.71	85.99	1.36	
Methionine	80.57	79.76	78.96	80.06	74.11	79.05	80.32	84.91	1.18	
Isoleucine	71.63	74.09	69.82	78.21	72.39	77.74	77.68	86.50	1.74	
Leucine	83.82	83.16	81.56	84.20	79.63	83.15	84.20	87.76	1.00	
Phenylalanine	81.27	81.60	77.01	82.30	76.31	81.89	80.49	84.31	1.21	
Lysine	81.66	83.50	80.69	81.98	77.52	81.52	82.48	86.65	1.14	
Histidine	84.02	87.83	75.85	86.57	78.19	88.32	83.34	88.72	1.41	
Arginine	84.84	89.35	83.10	89.00	81.68	87.89	91.08	92.63	1.02	
EAA avg.	80.05	81.56	77.33	82.38	75.74	81.86	82.10	86.76	1.23	
Aspartic acid	75.56	79.60	75.66	83.50	72.15	81.96	83.53	88.18	1.45	
Cystine	86.74	88.88	85.96	87.39	85.20	87.75	87.90	89.71	0.59	
Serine	81.49	82.79	80.22	84.63	77.40	83.50	84.99	88.36	1.09	
Glutamine	86.49	87.13	86.17	88.82	83.48	88.36	89.29	92.09	0.86	
Proline	88.50	88.77	88.15	89.49	85.74	88.84	89.19	91.59	0.65	
Glycine	76.82	77.62	73.97	79.35	69.70	78.82	79.39	84.00	1.41	
Alanine	80.97	79.52	78.04	79.88	74.70	79.37	80.58	84.34	1.20	
Tyrosine	81.08	79.35	76.55	78.64	74.92	79.88	79.96	84.58	1.19	
NEAA avg.	82.20	82.96	80.59	83.96	77.91	83.56	84.35	87.86	1.03	
TAA avg.	81.07	82.22	78.86	83.12	76.76	82.66	83.16	87.27	1.13	
Probability (%)	THR	VAL	CYS	MET	ILE	LEU	PHE	LYS	HIS	ARG
DE level	0.24	0.35	0.55	0.83	0.01	0.68	0.87	0.93	0.49	0.07
Lysine level	<0.01	0.01	0.05	0.04	<0.01	0.13	0.04	0.04	<0.01	<0.01
DE×lysine	0.04	0.05	0.12	0.02	0.43	0.15	0.12	0.04	0.05	<0.01
Probability (%)	EAA	ASP	SER	GLU	PRO	GLY	ALA	TYR	NEAA	TAA
DE level	0.29	0.04	0.27	0.25	0.90	0.42	0.92	0.54	0.37	0.32
Lysine:DE ratio	<0.01	<0.01	<0.01	0.01	0.09	<0.01	0.27	0.33	0.01	0.01
DE×lysine	0.02	0.04	0.06	0.12	0.27	0.01	0.10	0.06	0.06	0.05

density and lysine:DE ratios. There were no interactions between energy density and lysine:DE ratios related to fecal and urine nitrogen excretion

In this study, the numerical increase in urinary nitrogen excretion at the 3.6 Mcal of DE level indicates a increment in amino acid deamination and suggests that, at this digestible energy levels there may be insufficient DE available for minimal N excretion. In contrast, Lawrence et al. (1994) showed that high-energy diets (3.75 Mcal of DE) resulted in lower urinary nitrogen excretion than low-energy diets (3.5 Mcal of DE). Han et al. (1995) showed that pigs fed low CP diets without amino acid supplementation excreted lower amounts of urinary nitrogen than pigs fed high CP diets. With environmental considerations becoming increasingly important in animal production, reduction of nitrogen excretion through feed re-formulations or proper feeding regimens may become necessary in the livestock industry. Our results will be helpful for this goal, as we showed that feeding pigs diets with the proper lysine levels in the finishing period could reduce nitrogen excretion without retarding growth performance (The growth performance data weren't showed in this article).

The response of amino acid digestibility to two energy levels and four lysine:DE ratios are summarized in Tables 4 and 5. The apparent digestibility of all amino acids except for isoluecine, arginine and aspartic acid as well as average values of essential amino (EAA), non-essential amino acids (NEAA) and total amino acids digestibilities ($p>0.05$) were not affected by energy density. The apparent digestibility of all amino acids except for leucine, proline, alanine and tyrosine, NEAA and total amino acids digestibility were significantly affected by lysine: DE ratio ($p<0.05$).

Interactive effects of energy and lysine:DE ratio also significantly affected amino acid digestibility except for isoleucine, alanine, cystine, leucine, phenylalanine, glutamine and proline ($p<0.05$). This is consistent with the findings Chiba et al. (1991), who did not detect any interactions between energy density and the lysine:calorie ratio in heavier pigs.

Based on these results, we observed that increasing lysine:DE ratios increased the average values of essential amino acids digestibility. This result is supported by Jung et al. (1999), who observed that pigs fed high lysine:DE ratio diets showed higher than average values of essential amino acid digestibility. Holmes et al. (1974) demonstrated that the apparent ileal digestibility of lysine in soybean meal (48%) was 90.7% in studies where the dietary lysine content was 1.46%. However, Rudolph et al. (1983) suggested a value of 83.5% in studies where the dietary lysine content was 0.70. These results indicated that a well-balanced diet with an optimized lysine:DE ratio should be fed to finishing pig to improve nutrient utilization and reduce their environmental impact.

When energy density was converted to ME, it assumed that ME is 96% of DE .The ME level effects observed in Campbell et al. (1983) and Urynek et al. (2003) where high-energy diets resulted in higher N deposition and faster growth of young pigs than low-energy diets. However, the lysine:ME ratio effects were observed by Urynek et al. (2003) wherein although daily N retention, ADG, and were affected by ME concentration, increasing the lysine:ME ratio may increase daily N retention and growth criteria if sufficient ME available. However, Lawrence et al. (1994) failed to observe a response to increasing the lysine:DE ratio because energy intake may have been limiting N retention.

According to the present result, increasing the lysine:DE ratio can result in increases in crude protein digestibility and urinary nitrogen excretion, although apparent protein digestibility and nitrogen excretion were not affected by energy density Further, increasing the lysine:DE ratio also increased the apparent digestibility of essential amino acids except for leucine regardless of energy density. The data suggest that maintaining the appropriate lysine:DE ratio becomes more important as DE concentration of the diet increases.

In conclusion, our results suggest that maintaining the optimum lysine:DE ratio that would maximize essential amino acid digestibility in finishing pigs (64.2±0.69 kg) is approximately 2.4 g of lysine/Mcal of DE.

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