Effects of Betaine on Performence, Carcass Characteristics and Hepatic Betaine-homocysteine Methyltransferase Activity in Finishing Barrows*

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ABSTRACT : This experiment was conducted to determine the effect of dietary betaine (0, 0.125%) on performance, carcass composition, pork quality and hepatic betaine-homocysteine methyltransferase (BHMT) activity of crossbred finishing barrows. Three replicates of ten pigs were used for each treatment. The results showed that average daily gain, feed intake and feed conversion were not affected by betaine. Compared with the control group, pigs treated with betaine had a 8.17% ($p<0.05$) decrease in carcass fat percentage, and a 8.84% ($p<0.05$) reduction in 10th-rib backfat thickness, but dressing percentage, percentage lean, longissimus muscle area, and average backfat thickness were not affected. There were also no significant differences in muscle color score, marbling score, pork pH value and water loss rate between the control and betaine-treated groups. Hepatic betaine-homocysteine methyltransferase (BHMT) activity was significantly increased by 13.97% ($p<0.05$) when pigs were offered 0.125% betaine. ***(Asian-Aust. J. Anim. Sci. 2006. Vol 19, No. 3 : 402-405)***

Key Words : Betaine, Barrow, Performace, Carcass Characteristics, Pork, BHMT Activity

INTRODUCTION

The use of betaine in swine diets has increased since Cadogan et al. (1993) reported a 14.8% decrease in backfat thickness in pigs offered diets supplemented with 1,250 mg/kg betaine. Following this report, many researchers evaluated the effects of betaine on pigs (Haydon et al., 1995; Emmert et al., 1996, 1998; Lawrence et al., 2002; Yu et al., 2004).

Studies on the dietary betaine effect of the performance of pigs show variable results (Haydon et al., 1995; Matthews et al., 1998, 2001a; Lawrence et al., 2002). In some experiments, betaine supplementation improved Average daily gain (ADG) and/or carcass quality parameters, whereas in others studies, betaine had no effect. The inconsistent results also can be seen in the effects of betaine on pork quality (Matthews et al., 1998; 2001b; Øverland et al., 1999). The variable responses to betaine supplementation across studies is likely due to the different mode of action of betaine tested and/or the difference in animal health and stress status between studies.

Betaine-homocysteine methyltransferase (BHMT), is produced primarily in the liver, and catalyzes the conversion of homocysteine to methionine by facilitating the transfer of a methyl group from betaine, thereby playing a critical role in maintaining normal physiological levels of homocysteine and methionine (Finkelstein et al., 1971; Finkelstein and Martin, 1984; Xue, 1986). The importance of BHMT in betaine metabolism is evident, but data regarding the influence of dietary betaine on BHMT activity in swine is limited. Although the effect of betaine on BHMT activity has been studied in young pigs (Emmert et al., 1998), it has not been investigated in finishing pigs yet. The objective of this study was to evaluate the effects of 1,250 mg/kg betaine (the most often studied dosage) on performance, carcass characteristics, pork quality and hepatic BHMT activity in pigs, when offered feed *ad libitum* from 62.5 to 92.5 kg.

MATERIALS AND METHODS

Animals and experimental design

Sixty crossbred barrows (Duroc×Landrace×Yorkshire) with an average initial weight of 62.5 kg were allotted to two dietary treatments on the basis of weight, and ancestry was equalized across treatments in a randomized complete block design. Three replications of ten pigs were used for each treatment. The treatment diets included a corn-soybean meal basal (Table 1) supplemented with 0 and 0.125% betaine at the expense of corn. The basal diet was supplemented with minerals and vitamins to meet or exceed the requirements for finishing pigs (NRC, 1998). All pigs were given *ad libitum* access to feed and water. Performance results as average daily gain (ADG), average daily feed intake (ADFI), and feed conversion (FCR) were collected.

Carcass measurements

At the end of the feeding trial, 12 pigs (six barrows

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Table 1. Ingredient and nutrients composition of experiment basal diet

Ingredient	%	Nutrient[3]	
Maize	69.25	DE (MJ/kg)	13.32
Soybean meal	18.00	Protein (%)	15.57
Rapeseed meal	3.00	Calcium (%)	0.67
Wheat middling	3.00	Phosphorus (%)	0.54
Wheat bran	4.00	Lysine (%)	0.79
Calcium phosphate	1.00	Methionine (%)	0.27
Limestone	1.00	Cystine (%)	0.30
Salt (NaCl)	0.40	Threonine (%)	0.60
Mineral premix[1]	0.18		
Vitamin premix[2]	0.02		
Lysine.HCl	0.15		

[1] Supplied the following in mg/kg of diet: Fe, 125; Cu, 10; Mn, 20; Zn, 120.
[2] Supplied the following per kilogram of diet: vitamin A, 5,000 IU; cholecalciferol, 700 IU; vitamin B_{12}, 27.2 μg; riboflavin, 5.5 mg; niacin, 30 mg; dl-α-tocopheryl acetate, 78 IU; D-pantothenic acid, 25 mg; biotin, 80 mg; thiamin, 2.4 mg; choline chloride, 450 mg.
[3] DE based on calculated values, others were analyzed value.

from each treatment) were selected to determine carcass composition and pork quality. All pigs were killed humanely, dehaired and eviscerated. The head was removed and the carcass was split longitudinally. One-half of the carcass was physically dissected into bone, muscle, fat and skin; with each of the dissected tissues being weighed to the nearest gram. Backfat thickness was measured at the carcass midline at the 10th rib. Average backfat was determined by averaging the backfat thickness at the first rib, last rib, and last lumbar vertebrae. Longissimus muscle area (LMA) was determined by tracing its surface area at the 10th rib. Marbling score and color score were evaluated on the cut surface of the longissimus muscle between the 10th and 11th rib according the methods of NPPC (1976); pH value and water loss rate were determined by the method of Xu et al. (1996).

Carcass dressing percentage was calculated by the following formula: hot carcass weight divided by final live weight ×100.

Laboratory analysis

Described as Emmert et al. (1998), liver samples were immediately removed, frozen in liquid nitrogen, and stored at -80°C for determination of BHMT activity.

Activity of BHMT was measured in crude extracts of liver by the method of Garrow (1996) and Emmert et al. (1998) described with several modifications. In brief, the assay contained 5 mM DL-Hcy, 2 mM betaine, and 50 mM Tris-HCl (pH 7.5). The final reaction volume was 0.5 ml. Reactions were initiated with the addition of tissue extract and carried out for 1.5 h at 37°C. Reactions were stopped by chilling the tubes in ice water, and reactants were applied to a Dowex 1-× 4 column. The unreacted betaine was washed from the column with water. Methylthioacetate and methionine were eluted into scintillation vials with 3 ml of 1.5 N HCl. Measurement of total protein in crude liver extracts was conducted using the method of Bradford (1976) with reagents obtained from Sigma company (*St.* Louis, USA). As measured, a unit of BHMT activity was defined as nanomoles of product formed per hour.

Table 2. The effects of betaine on growth performance of pigs offered feed adlibitum[1]

	Betaine (%)		SEM[2]
	0	0.125	
ADG (kg/d)	0.715	0.748	0.021
ADFI (kg)	2.34	2.39	0.05
FCR	3.27	3.22	0.04

[1] Values are presented as means; n = 3 for ADG, ADFI and FCR per treatment.
[2] Standard error of the mean.

Statistical analysis

The experiment was analyzed as randomized complete-block design, and analysis of variance was conducted on all data using the General Linear Models (GLM) procedure of SAS (1988). A significance level of 0.05 was used.

RESULTS AND DISCUSSION

Performance

The results showed that average daily gain, feed intake and feed gain ratio were not significantly affected in present study (Table 2). Previous investigations of the effects of betaine on finishing pig performance were variable. Betaine increased Average daily gain (ADG) in finishing pigs fed low protein-low energy diets and adequate protein-high energy diets (Matthews et al., 1998). The same trend was found in finishing pigs fed low-protein diets but not in those fed high-protein diets (Haydon et al., 1995). Smith et al. (1995) also indicated that betaine increased Average daily gain (ADG) in finishing pigs. The data presented here are consistent with the results of other studies, which indicated that there was no effect of betaine on performance of finishing pigs (Webel et al., 1995; Øverland et al., 1999). Matthews et al. (1998) suggested that an interaction between betaine, crude protein, and net energy on Average daily gain (ADG) might exist during the feeding periods. This could explain some of the conflicting results reported in the literature.

Carcass characteristics and pork quality

Carcass characteristics are shown in Table 3. As compared with the control, pigs fed 0.125% betaine had a 8.17% ($p<0.05$) decrease in percentage carcass fat, and a 8.84% ($p<0.05$) reduction in 10th-rib backfat thickness. Betaine has been reported to decrease backfat thickness (Cadogan et al., 1993; Casarin et al., 1997; Matthews et al. 2001b), and our data are in agreement. Betaine could also

Table 3. Effects of betaine on the carcass characteristics and some measures of pork quality of pigs[1]

	Betaine (%)		SEM
	0	0.125	
Dressing percentage	75.82	76.31	0.22
Percentage lean	63.92	65.26	0.54
Percentage fat	18.11b	16.63a	0.40
Percentage skin	5.58	5.63	0.11
Percentage bone	9.37	9.65	0.17
backfat thickness (10th rib) (cm)	2.49[a]	2.27[b]	0.07
Average backfat thickness (cm)	2.70	2.51	0.09
Longissimus muscle area (10[th] rib) (cm^2)	40.19	41.09	0.54
Leaf fat (kg)	1.27	1.15	0.05
Color score of muscle[2]	2.14	2.04	0.11
pH value	6.21	6.19	0.09
Water loss rate (%)	11.06	11.22	0.47
Marbling score of muscle 3	1.85	1.87	0.07

[1] Values are means for 6 pigs. Means within rows with different superscript letters differ significantly (p<0.05).
[2] 1-pale, 5-dark.
[3] 1-traces, 5-abundant.

increase carcass lean in pigs (Casarin et al., 1997; Cromwell et al., 1999). The Present study showed betaine increased percentage carcass lean and loin muscle area numerically. However, other reports have indicated that 0.125 or 1.0% betaine does not affect carcass traits (Webel, 1995; Matthews et al., 1998; Øverland et al., 1999), and Haydon et al. (1995) reported that backfat thickness and loin muscle area may be increased or decreased in pigs fed 0.10% betaine depending on the lysine:calorie ratio or energy level of the diet. There were no effects of betaine on dressing percentage, percentage carcass bone, or percentage carcass skin in our study.

It has been reported that betaine had no effect on subjective marbling and firmness-wetness of pigs fed 0.125% betaine or on sensory quality of pigs fed 1.0% betaine (Matthews et al., 1998; Øverland et al., 1999). This is agreement with this study. And we did not observe any effects on color of pigs fed betaine in the loin muscle in the current study. Previous research (Matthews et al., 1998) indicated that subjective color was paler in pigs fed betaine, More research is necessary on this point.

Betaine-homocysteine methyltransferase (BHMT) activity

The effect of betaine on hepatic betaine-homocysteine methyltransferase (BHMT) activity is presented in Figure 1. Pigs treated with 0.125% betaine had a significant improvement of 13.97% (p<0.05) in hepatic BHMT activity as compared with the control group (53.93±4.03 vs. 47.32±3.93). There is no information in the literature about the effect of betaine on hepatic BHMT activity in finishing swine. Previous work with rats and chicks has shown that

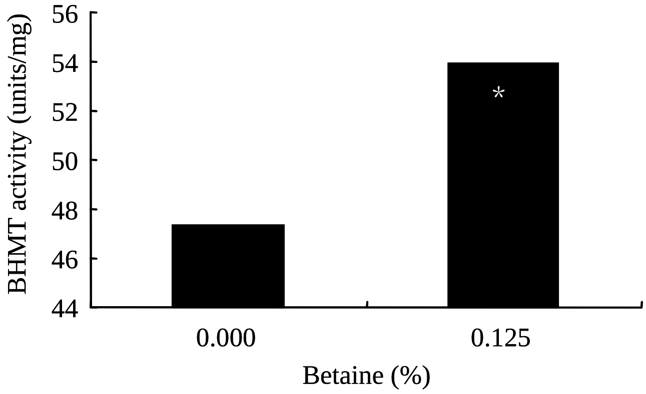

Figure 1. The effects of betaine on hepatic betaine-homocysteine S-methyltransferase activity of pigs (Values are means for 6 pigs. * Stands for value differ significantly compared with control.).

hepatic BHMT activity is substantially elevated under methionine-deficient conditions and is further elevated by supplementation with excess choline or betaine (Finkelstein et al., 1982a; Emmert et al., 1996; Park et al., 1997). Emmert et al. (1998) reported only moderate elevations in BHMT activity due to betaine in young pigs (8-20 kg). The present results with heavier pigs indicated that betaine increased hepatic BHMT activity. Betaine may serve as an important alternative methylating agent when normal methylating pathways are impaired (Barak and Tuma, 1983). A possible mode of action of methyl donors involves a methionine sparing effect in processes, such as the methylation of nucleic acids and other substrates (e.g., synthesis of carnitine, phosphatidylcholine) that requires methyl group transfer only from S-adenosyl-methionine, freeing additional methionine for other metabolic functions (Kidd et al., 1997). Campbell et al. (1995) suggested that betaine partially replaces methionine in sulfur amino acid-deficient diets fed to finishing pigs. The present diets were formulated to meet NRC (1998) recommendations for all nutrients including methionine and choline. The fact that hepatic BHMT activity was still elevated suggests that the diets maybe actually have been marginal in digestible methionine for the protein deposition capacity of the pigs used in the study. Methionine marginal deficiency condition increased hepatic BHMT activity to conserve methionine for the synthesis of the protein (Finkelstein et al., 1982a). This suggestion is further supported by the fact that in young pigs, rats and chicks that hepatic BHMT activity is elevated under methionine marginal deficient conditions and is further increased by supplementation with surfeit betaine (Finkelstein et al., 1982b, 1983; Emmert et al., 1996, 1998).

IMPLICATIONS

Addition of 0.125% betaine to the diet of finishing pigs may result in decreased carcass fat and backfat thickness, which are beneficial to the swine industry. And betaine could increase hepatic BHMT activity of finishing barrows. Further work is required to better define the conditions for which dietary betaine can improve performance and carcass characteristics.

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