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Effect of Particle Size, Ash Content, and Processing Pressure on the Bioavailablity of Phosphorus in Meat and Bone Meal for Swine.

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The attached reprint of the American Society of Animal Science (ASAS) article issubmitted as the final report of FPRF project conducted by Dr. Gary Cromwell and associates. Previous studies conducted by Dr. Carl Parsons, University of Illinois, have validated a significant reduction in the bioavailability of amino acids as the result of pressure processing meat and bone. Dr. Parsons's experiments were conducted using broiler chickens and dogs as experimental animals. Processing pressures of 0, 30, 45, and 60 pounds per square inch (psi) resulted in a 10 to 15% reduction in the bioavailability of many of the essential amino acids beginning at the 30 psi range. Bioavailability reductions were greatest for those amino acids with sulfur containing structures. Reductions in bioavailability were enhanced with increasing pressure treatment.

Dr. Cromwell's work did not demonstrate any influence in the relative availability of phosphorus in this test. This study validates the relatively high bioavailability of the phosphorus supplied by meat and bone meal. Values of at least 90%, relative to that of monosodium phosphate, is used by most practicing nutritionists when formulating meat and bone meal in most of the meat producing species diets. This study lends support to the higher values currently used when compared to dated databases. Similarly little value is acquired from pressure processing protein meals excepting feather and hair. It is a negative influence on protein and amino acid digestibility.

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# Effects of particle size, ash content, and processing pressure on the bioavailability of phosphorus in meat and bone meal for swine<sup>1,2</sup>

# S. L. Traylor<sup>3</sup>, G. L. Cromwell<sup>4</sup>, and M. D. Lindemann

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**ABSTRACT:** Meat and bone meal (MBM), when supplemented with tryptophan, is an excellent protein source for pigs. It is also a rich source of Ca and P. but some research has suggested that the bioavailability of P is variable. Three experiments were conducted to determine whether particle size, ash content, or processing pressure of MBM influences the bioavailability of P. Each experiment involved six replications of six treatments with individually penned pigs initially averaging 13 to 17 kg of BW. A low-P basal diet was fed with or without 0.1 or 0.2% added P (as-fed basis) from monosodium phosphate (MSP) or with three types of MBM added at levels that supplied 0.2% P (as-fed basis). The Ca level was 0.70%, and the lysine level was 0.95% in all diets. Pigs were allowed to consume their diets (meal form) on an ad libitum basis. At the end of the study, pigs were killed, and femurs and third and fourth metacarpals and metatarsals were removed for determination of breaking strength and ash content. Bone traits were regressed on added P intake for each

P source, and slope-ratio procedures were used to estimate the bioavailability of P in MBM relative to that in MSP. In Exp. 1, a blended source of MBM ground to three particle sizes (amount that passed through 6-, 8-, or 12-mesh screens) was evaluated. In Exp. 2, low-ash MBM of porcine origin, high-ash MBM of bovine origin, and a 1:1 blend of the two sources were assessed. In Exp. 3, normally processed MBM was subjected to an additional 2.1 and 4.2 kg/cm<sup>2</sup> of pressure for 20 min to determine whether excessive heat treatment would influence the bioavailability of P. Fineness of grind of MBM or processing pressure did not influence the relative bioavailability of P in this study; however, ash content of MBM affected P bioavailability. The relative availability of P in low-ash MBM of porcine origin (with composition typical of meat meal) was approximately 15 percentage units less than that in high-ash MBM of bovine origin. Overall, the bioavailability of P in MBM. relative to that in MSP, averaged 85%.

Key Words: Meat and Bone Meal, Phosphorus, Pigs

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## Introduction

Meat and bone meal (MBM) contains relatively high levels of protein, Ca, and P, and it is commonly included in swine diets. Studies have shown that growth performance in pigs fed relatively high levels of MBM is not decreased when diets are supplemented with 0.03% tryp-

tophan for every 10% addition of MBM to the diet (Cromwell et al., 1991).

Conflicting information exists on the bioavailability of Ca and P in MBM. Huang and Allee (1981) indicated that the relative bioavailability of P in MBM was 93%, but later studies reported P bioavailabilities that were considerably lower, ranging from 64 to 72% (Burnell et al., 1988, 1989; Coffey and Cromwell, 1993). A recent study by our group indicated that the bioavailability of P in MBM, relative to that in monosodium phosphate (MSP), was approximately 91% (Traylor et al., 2005), which agrees more closely with the value of 93% reported by Huang and Allee (1981).

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<sup>&</sup>lt;sup>2</sup>Appreciation is extended to G. G. Pearl, President and Director of Technical Services, Fats and Proteins Research Foundation, Bloomington, IL, for partial support of this research with a grantin-aid; to S. D. Thomas, Griffin Industries, Cold Spring, KY, for providing the meat and bone meal used in the first two studies; to K. C. Behnke, Grain Sci. Dept., Kansas State Univ., Manhattan, for grinding the meat and bone meal for the particle size study; and to C. R. Hamilton, Darling International, Inc., Irving, TX, for providing and processing the meat and bone meal for the pressure/temperature study. Appreciation also is given to Akey, Lewisburg, OH, for providing the vitamin premix and to Ajinomoto Heartland LLC, Chicago, IL, and Archer Daniels Midland, Decatur, IL, for providing the Llysine HCl and L-tryptophan used in the studies.

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Table 1. Composition of meat and bone meal (%, air-dry basis)

		Exp	p. 2ª	Exp. 3 <sup>h</sup>			
Item	Exp. 1	Low ash	High ash	0 kg/cm <sup>2</sup>	2.1 kg/cm <sup>2</sup>	4.2 kg/cm <sup>2</sup>	
CP	44.8	59.7	40.0	49.8	49.5	50.0	
Crude fat	9.0	9.8	9.6	7.7	7.4	7.2	
Ash	32.3	23.1	42.7	35.1	34.0	33.4	
Ca	10.9	7.4	14.3	12.3	12,0	10.9	
P	5.5	3.7	7.1	5.8	5.7	5.2	
AA							
Arginine	3.13	4.01	2.91	3.53	3.37	3.42	
Histidine	0.75	1.19	0.60	0.79	0.80	0.78	
Isoleucine	1.10	1.70	0.94	1,26	1.29	1.27	
Leucine	2.48	3.66	2.03	2.73	2.78	2.77	
Lysine	2.15	3.07	1.86	2.30	2.28	2.21	
Methionine	0.58	0.84	0.47	0.54	0.54	0.52	
Cysteine	0.35	0.61	0.27	0.54	0.44	0.38	
Phenylalanine	1.41	2.03	1.17	1.54	1.54	1.55	
Tyrosine	0.92	1.44	0.67	1.02	0.97	1.02	
Threonine	1.33	1,99	1.04	1.44	1.46	1.44	
Tryptophan	0.26	0.37	0.22	0.31	0.29	0.29	
Valine	1.70	2.52	1.57	1.88	1.92	1.93	

"The low-ash meat and bone meal (MBM) was of porcine origin, and the high-ash MBM was of bovine origin.

<sup>13</sup>Normally processed MBM was further processed at a high temperature for 20 min at 2.1 and 4.2 kg/cm<sup>2</sup> (30 and 60 pounds/square inch, respectively).

Phosphorus digestibility studies also have produced variable results. Poulsen (1995) reported that the apparent digestibility of P in MBM for pigs ranged from 54 to 80%; however, in two studies in our laboratory, the true digestibility of P in MBM averaged 81.9 compared with a true digestibility value of 86.7% for MSP (Traylor et al., 1999a,b), suggesting that the P in MBM was 94% as digestible as the P in MSP.

The reason for the wide range in estimates of P bioavailability is not clear. Source of raw material and processing have been suggested as factors that may contribute to variation in quality of meat byproducts (Knabe, 1995). Large particles of bone in MBM have been suggested to decrease P availability (Burnell et al., 1988), in that the P in large bone particles may not be as efficiently released for absorption as the P in more finely ground bone.

This study was conducted to determine whether factors such as particle size, ash content (reflecting species origin), or processing pressure of MBM affect the bioavailability of P in MBM for growing pigs.

## Materials and Methods

Three experiments were conducted to determine the bioavailability of P in MBM of different particle sizes and ash contents and in MBM subjected to different processing pressures. The experiments were 35 d in length and were conducted at the University of Kentucky Swine Research Unit, Lexington. The studies were conducted under protocols approved by the University of Kentucky Institutional Animal Care and Use Committee.

Pigs originated from Hampshire  $\times$  Yorkshire-Landrace crosses and initially averaged 16.5, 12.7, and 15.6 kg of BW in Exp. 1, 2, and 3, respectively. In each study, the pigs were allotted randomly to six dietary treatments with six replications per treatment from outcome groups of weight, gender, and ancestry. They were individually penned in elevated, wire mesh-floored pens (0.6 m  $\times$  1.2 m) in a temperature-controlled building and allowed to consume their diets (meal form) on an ad libitum basis from stainless steel self-feeders. Water was provided from nipple waterers. Pig weights and feed intake were determined weekly.

## Meat and Bone Meals

The MBM sources used in the first two studies (Table 1) were provided by Griffin Industries (Cold Spring, KY). In Exp. 1, MBM cracklings of porcine and bovine origin were obtained from a plant and sent to the Grain Science Department at Kansas State University, Manhattan. The cracklings were equally divided into three lots; then, each batch was ground in a hammer mill equipped with three screen sizes to produce ground MBM that passed through 6-, 8-, or 12-mesh screens (representing coarse, medium, and fine, respectively). The mean geometric particle size (ASAE, 2003) of the coarse, medium, and fine MBM was 635, 535, and 471  $\mu$ , respectively. The processing was performed in collaboration with K. C. Behnke (Grain Sci. Dept., Kansas State Univ.).

In Exp. 2, two sources of MBM, one that was low in ash content (23.1%) and a second one that was high in ash content (42.7%), were evaluated. The low-ash MBM was from a packing plant that processed swine, and the high-ash MBM was from a plant that processed dairy

Table 2. Composition of diets in Exp. 1 (%, as-fed basis)

	Diet							
	1	2	3	4, 5, and 6				
	Basal	MS	SP <sup>a</sup>	MBMª				
Item	(0.00% added P)	0.10% added P	0.20% added P	(0.20% added P)				
Ground corn	71.00	71.00	71.00	71.00				
Soybean meal, dehulled	21.72	21.72	21.72	21.72				
Cornstarch	3.64	2.79	1.95	0.39				
Corn oil	1.00	1.40	1.80	1.57				
MSP	_	0.45	0.89					
MBM	_	_	_	3.64				
Ground limestone	0.60	0.60	0.60	0.60				
Calcium carbonate <sup>b</sup>	0.98	0.98	0.98					
Iodized salt	0.50	0.50	0.50	0.50				
Vitamin-trace mineral premix <sup>c</sup>	0.18	0.18	0.18	0.18				
L-Lysine·HCl	0.13	0.13	0.13	0.13				
L-Tryptophan	-	_		0.02				
Antibiotic <sup>d</sup>	0.25	0.25	0.25	0.25				
Calculated analysis <sup>e</sup>			% <del></del>					
CP	16.4	16.4	16.4	18.0				
Lysine	0.95	0.95	0.95	1.03				
Ca	0.70	0.70	0.70	0.70				
P	0.34	0.44	0.54	0.54				
ME, Meal/kg	3.365	3.365	3.365	3.365				

 $^{n}MSP$  = monosodium phosphate; MBM = meat and bone meal. Included three particle sizes (coarse, medium, and fine) of the same MBM that passed through a 6-, 8-, or 12-mesh screen. Mean geometric particle sizes were 635, 535, and 471  $\mu$ , respectively.

<sup>b</sup>Technical grade.

<sup>e</sup>Provided the following per kilogram of diet: 6,600 IU of vitamin A (2,270 μg of retinyl acetate), 880 IU of vitamin  $D_3$  (22 μg of cholecalciferol), 22 IU of vitamin E (22 mg of  $D_4$ -α-tocopherol acetate), 6.4 mg of vitamin K (as menadione sodium bisulfite complex), 8.8 mg of riboflavin, 22 mg of pantothenic acid, 44 mg of niacin, 0.022 mg of vitamin  $B_{12}$ , 0.22 mg of p-biotin, 1.1 mg of folic acid, 135 mg of Zn (ZnO), 135 mg of Fe (FeSO<sub>4</sub>·H<sub>2</sub>O), 45 mg of Mn (MnO), 13 mg of Cu (CuSO<sub>4</sub>·5H<sub>2</sub>O), 1.5 mg of I (CaI<sub>2</sub>O<sub>6</sub>), and 0.3 mg of Se (NaSeO<sub>3</sub>).

dMecadox (Phibro Animal Health, Fairfield, NJ) provided 55 mg of carbadox/kg of diet.

Based on calculated analysis of corn and soybean meal (NRC, 1998) and determined analysis of MBM,

cows. Particle size was similar (10-mesh screen) for the two sources.

In Exp. 3, MBM was subjected to excessively high processing pressure and temperature to determine whether the additional processing would have a negative effect on the bioavailability of Ca and P, as it does with AA (Batterham et al., 1986; Johns et al., 1987; Wang and Parsons, 1998b). A blended MBM from Darling Industries (Wahoo, NE) that had been processed under typical industry conditions was used in this study. The MBM was processed in a continuous system at atmospheric pressure for 40 to 90 min; time and temperature were monitored to provide an exit temperature of 135 to 143°C. (This process meets or exceeds the European Union requirement of 133°C for 20 min.) The MBM was divided into three batches. One batch was not further processed and served as the control. A second batch was placed in a laboratory-scale model cooker, and steam was injected to bring the pressure to 2.1 kg/cm<sup>2</sup> (30 pounds/ square inch [psi]) for 20 min. A third batch was processed similarly, except that it was subjected to 4.2 kg/ cm<sup>2</sup> (60 psi) of pressure for 20 min. The further processing was performed in collaboration with C. R. Hamilton (Darling Industries, Irvine, TX).

#### Diets

The experimental diets in Exp. 1 are shown in Table 2. Diet 1 was a low-P (0.34%, as-fed basis) basal diet consisting mainly of ground corn and dehulled soybean meal with a small amount of cornstarch. All of the P in this diet was supplied by the corn and soybean meal; thus, the P was largely in the form of phytate, from which P is poorly available (Cromwell and Coffey, 1993). Diets 2 and 3 consisted of the basal diet with two graded levels of supplemental P (0.10 and 0.20%) supplied by MSP, a highly available source of P. Diets 4, 5, and 6 consisted of the basal diet with MBM of the three particle sizes described previously, at levels that provided 0.20% P. The MSP and MBM were substituted for cornstarch. Technical-grade calcium carbonate was included in Diets 1 to 3 to provide the same amount of Ca as supplied by the MBM. A constant amount of ground calcitic limestone was included in all diets so that the dietary Ca was maintained at 0.70%. In addition, L-lysine HCl was added to all diets, and L-tryptophan was added to the MBM diets to provide 3 g of tryptophan for every kilogram of MBM (Cromwell et al., 1991). Corn oil was adjusted to maintain a constant level of ME in all diets.

Table 3. Composition of diets in Exp. 2 (%, as-fed basis)

	Diet							
	1	2	3	4	5	6		
		MS	SP"		MBM <sup>a</sup>			
Item	Basal (0.00% added P)	0.10% added P	0.20% added P	0.20% added P (low-ash MBM)	0.20% added P (blended MBM)	0.20% added P (high-ash MBM)		
Ground corn	69.35	69.35	69.35	69.35	69.35	69.35		
Soybean meal, dehulled	21.85	21.85	21.85	21.85	21.85	21.85		
Cornstarch	5.65	4.80	3.97	0.00	1.77	3.53		
Corn oil	0.50	0.90	1.29	1.75	1.25	0.75		
MSP	_	0.45	0.89	_		_		
MBM		_	_	5.36	4.09	2.83		
Ground limestone	0.50	0.50	0.50	0.50	0.50	0.50		
Calcium carbonate <sup>b</sup>	0.98	0.98	0.98	_	*****	****		
Iodized salt	0.50	0.50	0.50	0.50	0.50	0.50		
Vitamin-trace mineral premix <sup>c</sup>	0.18	0.18	0.18	0.18	0.18	0.18		
L-Lysine·HCl	0.14	0.14	0.14	0.14	0.14	0.14		
L-Tryptophan		_	_	0.02	0.02	0.02		
Antibiotic <sup>d</sup>	0.25	0.25	0.25	0.25	0.25	0.25		
Calculated analysis				%				
CP	16.2	16.2	16.2	19.5	18.4	17.4		
Lysine	0.95	0.95	0.95	1.12	1.06	1.00		
Ca	0.70	0.70	0.70	0.70	0.70	0.70		
P	0.34	0.44	0.54	0.54	0.54	0.54		
ME, Mcal/kg	3.352	3.352	3.352	3.352	3.352	3.352		

"MSP = monosodium phosphate; MBM = meat and bone meal. The low-ash MBM (23.1% ash) was of porcine origin, and the high-ash MBM (42.7% ash) was of bovine origin. The blend contained equal quantities of the two sources on a P basis (32.9% ash).

<sup>b</sup>Technical grade.

<sup>e</sup>Provided the following per kilogram of diet: 6,600 IU of vitamin A (2,270 μg of retinyl acetate), 880 IU of vitamin D<sub>3</sub> (22 μg of cholecalciferol), 22 IU of vitamin E (22 mg of DL-α-tocopherol acetate), 6.4 mg of vitamin K (as menadione sodium bisulfite complex), 8.8 mg of riboflavin, 22 mg of pantothenic acid, 44 mg of niacin, 0.022 mg of vitamin B<sub>12</sub>, 0.22 mg of D-biotin, 1.1 mg of folic acid, 135 mg of Zn (ZnO), 135 mg of Fe (FeSO<sub>4</sub>·H<sub>2</sub>O), 45 mg of Mn (MnO), 13 mg of Cu (CuSO<sub>4</sub>·5H<sub>2</sub>O), 1.5 mg of I (CaI<sub>2</sub>O<sub>6</sub>), and 0.3 mg of Se (NaSeO<sub>3</sub>).

dMecadox (Phibro Animal Health, Fairfield, NJ) provided 55 mg of carbadox/kg of diet.

<sup>e</sup>Based on calculated analysis of corn and soybean meal (NRC, 1998) and determined analysis of MBM.

All diets were fortified with salt, trace minerals, and vitamins to meet or exceed NRC (1998) requirements, except for P. An antimicrobial agent also was included in the diets.

In Exp. 2, a low-P (0.34% P, as-fed basis) basal diet of similar composition as in Exp. 1 was fed (Table 3). Two levels of added P (0.10 and 0.20%) from MSP were added to the basal diet as in Exp. 1. Low-ash, intermediate-ash, and high-ash MBM were the three additional treatments. The low- and high-ash MBM contained 23.1 and 42.7% ash, respectively, and they were blended 1:1 to give an intermediate-ash MBM of 32.9% ash. The MBM was added at levels of 5.36, 4.09, and 2.83%, which supplied the same level of added P (0.20%) as the highest inclusion of MSP. All other dietary components and adjustments were similar to those of Exp. 1.

The composition of the diets in Exp. 3 is given in Table 4. A low-P basal diet (0.34% P, as-fed basis) and two diets with 0.10 and 0.20% added P from MSP, similar in composition to those in Exp. 1 and 2, were used in this study. Three additional treatments included MBM that had either not been further processed or that been subjected to 2.1 or 4.2 kg/cm² (30 or 60 psi) of pressure for 20 min, as described previously. When added at 3.50, 3.58, and 3.78% to the diet, each of the three MBM

supplied 0.20% added P, the same level of added P as provided by the highest inclusion of MSP. All other dietary components and adjustments were similar to those of Exp. 1 and 2.

## Bone Extraction and Analyses

At the end of the studies, all pigs were transported to the University of Kentucky Meats Laboratory and humanely killed (electrically stunned followed by exsanguination). The two femurs were collected, and both front and rear feet were removed at the knee and hock joint, respectively. The femurs and feet were sealed in plastic bags and frozen (–20°C). Later, the feet were thawed and placed in an autoclave at 121°C for 8 min to aid in removal of the third and fourth metacarpals and metatarsals. Following extraction, the bones were again frozen.

Before bone processing, the femurs, metatarsals, and metacarpals were allowed to thaw for 5 to 6 h and then were subjected to breaking strength determinations with an Instron machine (Instron Model TM 1123; Instron Corp., Canton, MA). All bones were broken in a thawed state. Breaking strength is defined as the amount of force, before fracture, applied by a wedge mounted on a

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Table 4. Composition of diets in Exp. 3 (%, as-fed basis)

				Diet		
	1	2	3	4	5	6
		MS	5P <sup>a</sup>		MBM <sup>b</sup>	
Item	Basal (0.00% added P)	0.10% added P	0.20% added P	0.20% added P (0 kg/cm <sup>2</sup> )	0.20% added P (2.1 kg/cm <sup>2</sup> )	0.20% added P (4.2 kg/cm <sup>2</sup> )
Ground corn	71.29	71.29	71.29	71.29	71.29	71.29
Soybean meal, dehulled	21,66	21.66	21.66	21.66	21.66	21.66
Cornstarch	3.31	2.47	1.63	0.38	0.28	
Corn oil	1.10	1.49	1.89	1.56	1.58	1.67
MSP	_	0.45	0.89	_	_	_
MBM	_	_	_	3.50	3.58	3.78
Ground limestone	0.53	0.53	0.53	0.53	0.53	0.53
Calcium carbonate <sup>c</sup>	1.05	1.05	1.05	_	_	_
Iodized salt	0.50	0.50	0.50	0.50	0.50	0.50
Vitamin-trace mineral premix <sup>d</sup>	0.18	0.18	0.18	0.18	0.18	0.18
L-Lysine·HCl	0.14	0.14	0.14	0.14	0.14	0.14
L-Tryptophan	_	_	_	0.02	0.02	0.02
Antibiotic <sup>e</sup>	0.25	0.25	0.25	0.25	0.25	0.25
Calculated analysis <sup>f</sup>				%		
CP	16.4	16.4	16.4	18.1	18.1	18.2
Lysine	0.95	0.95	0.95	1.03	1.03	1.03
Ca	0.70	0.70	0.70	0.70	0.70	0.70
P	0.34	0.44	0.54	0.54	0.54	0.54
ME, Mcal/kg	3.368	3.368	3.368	3.368	3.368	3.368

"MSP = monosodium phosphate.

Technical grade.

"Mecadox (Phibro Animal Health, Fairfield, NJ) provided 55 mg of carbadox/kg of diet.

pressure-sensitive compression cell that was positioned at the center of the bone when placed horizontally on two supports spaced 7.0 cm (femurs) or 3.2 cm (metacarpals and metatarsals) apart. Peak force was recorded on graph paper, and the height of the peak was determined. The metacarpals were then cut into halves, so that the marrow could be removed, and then were dried, wrapped in cheesecloth, and extracted with fresh petroleum ether three times at 24-h intervals. They were then dried at room temperature in a chemical hood for 24 h, dried in an oven overnight, then ashed in a muffle furnace at 600°C for at least 6 h. Ash weight was recorded and also was calculated as a percentage of dry, fat-free bone.

Bone strength and ash weight were regressed on the daily quantity of added P consumed by the pigs fed the MSP diets, with the basal diet included in determining the regression slopes. A similar procedure was used for the MBM treatments, except that it was based on single-point regression techniques (the basal and one level of added P). Single-point regression is justified based on the linear response of bone traits to level of added P up to 0.20% of added P. The linear response was clearly demonstrated in our previous study involving similar diets and pigs of approximately the same BW (Traylor

et al., 2005) and has been demonstrated in numerous other studies at our station. A comparison of the two slopes gives the bioavailability of the P in the specific MBM relative to the bioavailability of P in MSP (given a value of 100). The P bioavailabilities based on femur strength, mean of metacarpal-metatarsal strengths, and metacarpal ash weight were then averaged to give an overall estimate of the relative bioavailability of P in the MBM sources. The bioavailabilities were calculated with an unforced y-intercept.

## Chemical Analyses

Representative samples of MBM were analyzed in duplicate or triplicate for CP by a N analyzer (N  $\times$  6.25), for crude fat based on ether extraction, and for ash in a muffle furnace. After wet ashing, Ca was determined by atomic absorption chromatography, and P was determined by a gravimetric procedure. All methods were based on standard procedures (AOAC, 1995). Amino acids were analyzed with ion-exchange chromatography after acid hydrolysis. Methionine and cysteine were oxidized to methionine sulfone and cysteic acid by treatment with performic acid before hydrolysis. Tryptophan

<sup>&</sup>lt;sup>b</sup>MBM = meat and bone meal. The normally processed MBM (0 kg/cm<sup>2</sup>) was further processed at a high temperature for 20 min at 2.1 or 4.2 kg/cm<sup>2</sup> (30 or 60 pounds/square inch, respectively).

<sup>&</sup>lt;sup>d</sup>Provided the following per kilogram of diet: 6,600 IU of vitamin A (2,270 μg of retinyl acetate), 880 IU of vitamin D<sub>3</sub> (22 μg of cholecalciferol), 22 IU of vitamin E (22 mg of DL-α-tocopherol acetate), 6.4 mg of vitamin K (as menadione sodium bisulfite complex), 8.8 mg of riboflavin, 22 mg of pantothenic acid, 44 mg of niacin, 0.022 mg of vitamin B<sub>12</sub>, 0.22 mg of p-biotin, 1.1 mg of folic acid, 135 mg of Zn (ZnO), 135 mg of Fe (FeSO<sub>4</sub>·H<sub>2</sub>O), 45 mg of Mn (MnO), 13 mg of Cu (CuSO<sub>4</sub>·5H<sub>2</sub>O), 1.5 mg of I (CaI<sub>2</sub>O<sub>6</sub>), and 0.3 mg of Se (NaSeO<sub>4</sub>).

<sup>&</sup>quot;Based on calculated analysis of corn and soybean meal (NRC, 1998) and determined analysis of MBM.

Table 5. Effects of particle size of meat and bone meal (MBM) on performance and bone traits of growing pigs, Exp. 1<sup>a</sup>

				Diet			
	1	2	3	4	5	6	
		MS	5P <sup>b</sup>		$\mathrm{MBM^c}$		
Item	Basal (0.00% added P)	0.10% added P	0.20% added P	0.20% added P (6 mesh)	0.20% added P (8 mesh)	0.20% added P (12 mesh)	SE
ADG, kg <sup>ri</sup>	0,577	0.696	0.795	0.756	0.743	0.744	0.033
ADFI (as-fed basis), kg <sup>d</sup>	1.31	1.48	1.53	1.52	1.46	1.47	0.07
Feed:gain <sup>d</sup>	2.26	2.12	1.91	2.01	1.98	1.98	0.06
Added P intake, g/d <sup>d</sup>	0.00	1.51	3.05	3.03	2.92	2.95	0.12
Bone strength, kg							
Femur <sup>d</sup>	85.2	165.4	231,2	206.8	227.3	212.8	12.6
MT and MC, avg <sup>de</sup>	19.8	33.6	41.4	38.5	39.0	37.8	1.9
MC ash, g <sup>d</sup>	1.75	2.51	2.81	2.65	2.63	2.68	0.09
MC ash, % <sup>df</sup>	47.9	53.0	53.9	53.4	53.3	54.0	0.4

<sup>&</sup>quot;Each diet was fed to six individually penned pigs per treatment for 35 d. Initial and final BW averaged 16.5 and 41.7 kg, respectively.  $^{\rm b}$ MSP = monosodium phosphate.

was analyzed after alkaline hydrolysis. Calcium and AA assays were conducted at the University of Missouri Experiment Station Chemical Laboratories (Columbia, MO), and the other assays were conducted at the University of Kentucky.

## Statistical Analyses

The data were analyzed as a randomized complete block design (Steel and Torrie, 1980) using the GLM procedure of SAS (Version 8.2; SAS Inst., Inc., Cary, NC). The statistical model included the effects of replication, diet, and replication × diet (error). Preplanned treatment comparisons in Exp. 1 were basal vs. mean of other five treatments; linear and quadratic effects of P level within MSP, with the basal diet included in both regressions; linear and quadratic effects of particle size of MBM; and the highest level of MSP vs. the mean of the three MBM. In Exp. 2 and 3, the contrasts were the same as in Exp. 1, except that, within MBM, linear and quadratic effects of ash content (Exp. 2) and processing pressure (Exp. 3) were assessed. In all instances, pen was the experimental unit. Unless stated otherwise, an  $\alpha$ level of P < 0.05 was considered statistically significant.

#### Results

# Composition of Meat and Bone Meals

The composition of the MBM used in the experiments is shown in Table 1. In general, the blended MBM in Exp. 1 and 3 were slightly higher in fat, Ca, and P and lower in protein than values listed by NRC (1998) for MBM. As a result of the lower protein content, all AA were slightly lower in our MBM than NRC (1998) values. For example, NRC (1998) lists 2.51% lysine for MBM,

whereas the MBM in Exp. 1 and 3 ranged from 2.15 to 2.30%. The differences in composition between MBM were greatest in Exp. 2, where the low- and high-ash MBM was tested. In that study, the low-ash MBM contained considerably less ash, about one-half as much Ca and P, and in some instances, twice as much of the AA as the high-ash MBM. These differences are typical of a low-ash MBM derived from pork processing plants compared with high-ash MBM derived from beef processing plants. Actually, the low-ash MBM is more typical in composition to meat meal as listed by NRC (1998). High-pressure processing of MBM in Exp. 3 had essentially no effect on the composition of the MBM.

## Experiment 1

As expected, performance and bone traits were decreased (P < 0.01) in pigs fed the low-P basal diet compared with those fed diets with added P (Table 5). Body weight gain, feed intake, efficiency of feed utilization, femur and metacarpal-metatarsal strength, and metacarpal ash weight improved linearly (P < 0.01) when MSP was added to the basal diet (Table 5). Bone ash, expressed as a percentage of fat-free bone, also responded to increased P but in a quadratic fashion (P < 0.01). The growth performance and bone responses were similar among pigs fed the three particle sizes of MBM, indicating that particle size, within the range tested, had little effect on response patterns. Bone strength and ash were slightly less in pigs fed 0.20% added P from MBM compared with pigs fed the same level of P from MSP, but the differences were not of sufficient magnitude to be statistically significant.

## Experiment 2

As in Exp. 1, growth performance and bone mineralization were considerably less (P < 0.01) in pigs fed the

The same MBM was ground to three particle sizes (coarse, medium, and fine), which represented the MBM that passed through a 6-, 8-, or 12-mesh screen.

<sup>&</sup>lt;sup>d</sup>Basal vs. others, P < 0.01; MSP linear within Diets 1 to 3, P < 0.01.

eThird and fourth metatarsal (MT) and metacarpal (MC).

<sup>&</sup>lt;sup>f</sup>Quadratic response within Diets 1 to 3, P < 0.01.

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Table 6. Effects of ash content of meat and bone meal (MBM) on performance and bone traits of growing pigs, Exp. 2<sup>a</sup>

				Diet			
	1	2	3	4	5	6	
		MS	SP <sup>b</sup>		MBM <sup>c</sup>		
Item	Basal (0.00% added P)	0.10% added P	0.20% added P	0.20% added P (low-ash MBM)	0.20% added P (blended MBM)	0.20% added P (high-ash MBM)	SE
ADG, kg <sup>d</sup>	0.422	0.626	0.681	0.678	0.682	0.670	0.023
ADFI (as-fed basis), kg <sup>d</sup>	0.99	1.21	1.24	1.29	1.20	1.28	0.05
Feed:gain <sup>d</sup>	2.35	1.94	1.82	1.91	1.76	1.91	0.08
Added P intake, g/d <sup>d</sup> Bone strength, kg	0.00	1.21	2.48	2.58	2.41	2.57	0.09
Femur <sup>d</sup>	70.8	141.3	196.2	164.2	177.0	183.5	10.2
MT and MC, avg <sup>def</sup>	12.96	25.03	33.54	28.01	28.53	31.73	1.46
MC ash, g <sup>d</sup>	1.26	1.82	2.19	2.00	1.99	2.14	0.06
MC ash, % <sup>dg</sup>	47.1	51.4	53.5	53.2	53.5	53.8	0.69

<sup>&</sup>lt;sup>a</sup>Each diet was fed to six individually penned pigs per treatment for 35 d. Initial and final BW averaged 12.7 and 34.7 kg, respectively. <sup>b</sup>MSP = monosodium phosphate.

low-P basal diet compared with those fed diets with added P (Table 6). All of these traits improved linearly (P < 0.01) with the addition of P from MSP, except for bone ash percentage, which increased quadratically (P < 0.01) with MSP addition. In general, bone strength and ash weight tended to increase as ash content of the MBM increased, but these differences were not significant. Metacarpal-metatarsal strength was less (P < 0.05) in pigs fed the MBM diets compared with those fed the highest level of P addition from MSP. A similar trend was observed for the femur, but the difference was not significant.

## Experiment 3

As in Exp. 1 and 2, growth performance and bone mineralization were markedly decreased (P < 0.01) in pigs fed the low-P basal diet compared with those fed diets supplemented with P from MSP or MBM (Table 7). Linear (P < 0.01) improvements in all traits occurred with the addition of P from MSP, although the responses in ADG and ADFI also tended to be quadratic (P < 0.10) in this study. Processing pressure linearly affected (P < 0.05) femur strength, with greater strength exhibited when the MBM was treated with excess pressure for 20 min; however, this response pattern did not occur for metacarpal-metatarsal strength or ash. Bone strength was less (P < 0.05) when 0.20% P was added as MBM compared with MSP. A similar trend occurred for metacarpal ash (P < 0.10).

## Slope Ratio of Bone Traits

The slope-ratio data based on femur strength, the mean of metatarsal and metacarpal strength, and meta-

carpal ash weight for pigs in the three experiments are shown in Table 8. Regression of treatment means for the basal and the two levels of MSP on daily supplemental P intake resulted in good fits with  $r^2$  values for femur strength of 0.996, 0.992, and 0.999 in Exp. 1, 2, and 3, respectively. The  $r^2$  values for metatarsal-metacarpal strength were 0.971, 0.988, and 0.983, and for metacarpal ash weight, they were 0.936, 0.982, and 0.996 in Exp. 1, 2, and 3, respectively. The strong linear trends and excellent fits of the data to the regression line in the MSP treatments give justification for the use of single point regression slopes in the determination of the relative bioavailability of P in the MBM sources.

Based on the mean slope ratios of the three bone traits in Exp. 1 (Table 8), the bioavailability of P ranged from 94% in the 8-mesh MBM to 86% in the 6-mesh MBM. The finer particle-sized MBM was intermediate at 90% bioavailability. None of these differences were significant at P = 0.50.

In Exp. 2, the slope ratio data indicated that the ash content of the MBM affected (P < 0.02) the bioavailability of P (Table 8). The availability of P seemed to increase as the ash content of the MBM increased, with a difference of 17 percentage points from the low-ash to the high-ash MBM. This finding suggests that the P in MBM (actually more typically meat meal) of porcine origin is less available than that of bovine origin.

In Exp. 3, the slope ratio results indicated that the P bioavailabilities in the three MBM ranged from 80 to 91% (Table 8). It is evident from the data that the extremely high processing pressure to which MBM was subjected did not negatively affect the bioavailability of P in MBM. In fact, if anything, it might have improved the availability of P.

<sup>&</sup>quot;The low-ash MBM was of porcine origin, and the high-ash MBM was of bovine origin. The blend contained equal quantities of the two sources on a P basis.

<sup>&</sup>lt;sup>d</sup>Basal vs. others, P < 0.01; MSP linear within Diets 1 to 3, P < 0.01. <sup>e</sup>Third and fourth metatarsal (MT) and metacarpal (MC).

<sup>&</sup>lt;sup>f</sup>Diet 3 vs. mean of Diets 4 to 6, P < 0.05.

<sup>&</sup>lt;sup>g</sup>Quadratic response trend within Diets 1 to 3, P < 0.10.

Table 7. Effects of processing pressure of meat and bone meal (MBM) on performance and bone traits of growing pigs, Exp. 3<sup>a</sup>

				Diet			-
	1	2	3	4	5	6	
		MS	5P <sup>b</sup>		MBM <sup>e</sup>		
Item	Basal (0.00% added P)	0.10% added P	0.20% added P	0.20% added P (0 kg/cm <sup>2</sup> )	0.20% added P (2.1 kg/cm <sup>2</sup> )	0.20% added P (4.2 kg/cm <sup>2</sup> )	SE
ADG, kg <sup>de</sup>	0.565	0.737	0.791	0.817	0.803	0.804	0,023
ADFI (as-fed basis), kgde	1.18	1.46	1.50	1.47	1.48	1.48	0.037
Feed:gain <sup>d</sup>	2.10	1.99	1.90	1.81	1.85	1.84	0.04
Added P intake, g/dd	0.00	1.46	3.00	2,94	2.97	2.96	0.07
Bone strength, kg							
Femur <sup>dfg</sup>	79.5	167.3	264.7	204.8	217.7	244.2	10.4
MT and MC, avg <sup>dfh</sup>	20.67	41.27	54.96	49.44	48.92	50.54	2.19
MC ash, g <sup>di</sup>	1.45	2.14	2.73	2.51	2.51	2.63	0.08
MC ash, % <sup>di</sup>	47.8	52.8	56.3	54.3	55.4	55.3	0.64

Each diet was fed to six individually penned pigs per treatment for 35 d. Initial and final BW averaged 15.6 and 41.9 kg, respectively. <sup>h</sup>MSP = monosodium phosphate.

## Discussion

According to the Official Publication of AAFCO (2000), MBM is described as the rendered product from mammal tissues, including bone, exclusive of any added blood, hair, hoof, horn, hide trimmings, manure, stomach, and ruminal contents, except such amounts as may occur unavoidably in good processing practices. It should contain a minimum of 4.0% P, and the Ca level should not exceed 2.2 times the Plevel. Except for the low-ash MBM

**Table 8.** Relative bioavailability of P for meat and bone meal (MBM) with varying particle size, ash content, and processing pressure in Exp. 1, 2, and 3

	Bor	ne strength		
Item	Femur	MC-MT <sup>n</sup>	MC ash, g	Average
		— Relative bioavailal	bility of P in MBM, %b -	
Particle size, Exp. 1 <sup>e</sup>				
6-mesh (coarse)	84	88	86	86
8-mesh (medium)	102	94	87	94
12-mesh (fine)	91	87	91	90
SE	6	6	4	4
Ash content, Exp. 2 <sup>d</sup>				
Low (porcine origin)	73	69	75	72
Medium (1:1 blend)	88	78	81	82
High-ash (bovine origin)	87	89	92	89
SE	5	6	5	4
Processing pressure, Exp. 3e				
0 kg/cm <sup>2</sup>	69	86	84	80
2.1 kg/cm <sup>2 f</sup>	76	84	84	81
4.2 kg/cm <sup>2 f</sup>	90	88	94	91
SE	6	7	7	5
Overall MBM average				85

<sup>&</sup>lt;sup>a</sup>MC = metacarpal; MT = metatarsal.

The normally processed MBM (0 kg/cm<sup>2</sup>) was further processed at a high temperature for 20 min at 2.1 or 4.2 kg/cm<sup>2</sup> (30 or 60 pounds/ square inch, respectively). 

dBasal vs. others, P < 0.01; MSP linear within Diets 1 to 3, P < 0.01. 
MSP quadratic within Diets 1 to 3, P < 0.10.

Diet 3 vs. mean of Diets 4 to 6, P < 0.05. <sup>g</sup>Linear effect within Diets 4 to 6, P < 0.05.

<sup>&</sup>lt;sup>h</sup>Third and fourth metatarsal (MT) and metacarpal (MC).

<sup>&</sup>lt;sup>i</sup>Diet 3 vs. mean of Diets 4 to 6, P < 0.10.

<sup>&</sup>lt;sup>b</sup>Relative to monosodium phosphate, which was given a value of 100%.

<sup>&</sup>quot;No effect of particle size of MBM, P = 0.50.

<sup>&</sup>lt;sup>d</sup>Linear effect of ash content of MBM, P < 0.02.

 $<sup>^{\</sup>rm e}$ No effect of processing pressure of MBM, P=0.15.  $^{\rm f}$ 2.1 and 4.2 kg/cm<sup>2</sup> = 30 and 60 pounds/square inch, respectively.

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in Exp. 2, all of the MBM sources in this study were within these limits. The low-ash product of porcine origin evaluated in Exp. 2 contained 3.7% P, which falls into the category of meat meal according to the AAFCO (2000) definition. The higher CP content (59.7%) of the low-ash MBM in Exp. 2 compared with the CP content of the other MBM (40.0 to 50.0%) is more typical of meat meal than MBM.

The bioavailability of P in MBM in our three studies, based on slope-ratio procedures, averaged 85% across particle sizes, ash contents, and processing pressures. This value is in reasonably close agreement with an estimated 91% bioavailability of P in MBM previously reported by our group (Traylor et al., 2005) and with an earlier estimate of 93% by Huang and Allee (1981), in which they used similar procedures. These estimates also agree with results of recent balance studies conducted at our station in which the P in MBM was 94% as digestible as the P in MSP. Jongbloed and Kemme (1990) reported slightly lower bioavailabilities of P in MBM (74 to 85%). Because much of the P in MBM comes from bone, it is not surprising that the bioavailability estimates for P in MBM are in line with the estimated P bioavailability of 82% for steamed bone meal (Cromwell and Coffey, 1993).

Conversely, some studies have produced lower estimates of P bioavailability in MBM than ours. For example, bioavailability estimates of 63 to 67% (Burnell et al., 1988), 69% (Coffey and Cromwell, 1993), and 72% (Burnell et al., 1989) have been reported. Poulsen (1995) also reported a low estimate of apparent digestibility of P in MBM for pigs (54%) compared with a review of other cited research, in which the apparent digestibility ranged from 69 to 80%. The reason for this discrepancy in our recent estimates and the earlier ones is unknown. In the study of Burnell et al. (1989), the possibility that the low estimate might have been due to the large particle size in the MBM was suggested. In that study, approximately 30% of the P in MBM was in particles ≥4 mm in diameter.

Very few studies have investigated the effects of particle size of MBM on the utilization of minerals, and those have been conducted with poultry. Sell and Jeffrey (1996) found that particle size did not affect the utilization of P in MBM by turkey poults. In their study, the P in MBM was as available as the P in dicalcium phosphate. In a study in which steamed bone meal of different particle sizes was evaluated, Orban and Roland (1992) concluded that large bone fragments decreased the availability of P in bone meal for chicks.

Until the present study, we are not aware of other studies with pigs in which the effects of particle size of MBM on utilization of P have been assessed. Our study indicates that particle size of MBM over the range tested has very little, if any, affect on the bioavailability of P. Most processors grind MBM with a 10-mesh screen, and our study involved MBM that had been ground through 8- to 12-mesh screens, which covers the range of the majority of MBM products on the market (personal com-

munication, G. G. Pearl, Bloomington, IL). It is possible that the MBM used by Burnell et al. (1989) was coarser than the MBM that we tested in the present study.

Another possible factor that might explain the wide range in estimates of P bioavailability in MBM is the percentage of ash in the MBM tested. Meat and bone meals with high ash contents, and thereby high Ca and P contents, have a greater proportion of Ca and P supplied by bone and a lower proportion supplied by soft tissues. Most of the high-ash MBM sources are produced from packing plants that kill and process cattle (often dairy cattle that have a higher bone:soft tissue than finished cattle), whereas low-ash MBM are produced in packing plants that kill and process swine. Our second experiment clearly showed that the P in high-ash MBM of bovine origin was more highly available to pigs than the P from low-ash MBM of porcine origin. We are unaware of any other studies that would support or refute this finding in pigs. This result could be interpreted to mean that the P in bone is more highly available to pigs than the P in the soft tissues (muscle, cartilage, etc.).

Finally, the third experiment was conducted to determine whether MBM that is subjected to excessive processing pressure and temperature contributes to the utilization of P from MBM. The additional processing pressure of 2.1 and 4.2 kg/cm² (30 and 60 psi) was chosen in our study because it brackets the additional processing pressure of at least 3.15 kg/cm² (45 psi) for 20 min that was required by the European Union for processing MBM to protect against bovine spongiform encephalopathy. The regulation states that animal byproducts must be heated to a core temperature of >133°C for at least 20 min without interruption at a pressure (absolute) of at least 3 bars (equivalent to 3.15 kg/cm²) produced by saturated steam (Regulation No. 1774/2002 of the European Parliament and of the Council of October 3, 2002).

The current study clearly showed that excessive processing pressure did not negatively influence the bioavailability of P in MBM. In fact, there was a tendency for the P to be more available in MBM that was subjected to the additional processing; however, excessive processing of MBM might have dramatic negative effects on the availability of AA as indicated in other studies conducted with pigs (Batterham et al., 1986) and poultry (Johns et al., 1987; Wang and Parsons, 1998a,b).

In summary, the results of the present study, along with the results of a previous study at our station (Traylor et al., 2005), indicate that the bioavailability of P in MBM is relatively high for pigs, ranging from 85 to 90% of that of MSP and other commonly used inorganic phosphate sources. The source of the raw material from which MBM is derived seems to have an effect on the availability of P; high-ash MBM of bovine origin has greater bioavailability of P than low-ash MBM of porcine origin. In contrast, MBM particle sizes of 470 to 635  $\mu$  (ground to pass 12- and 6-mesh screens) did not affect P bioavailability, nor did excessive heat treatment represented by up to 4.2 kg/cm² (60 psi) of additional processing pressure for 20 min.

## **Implications**

The results of this study indicate that the bioavailability of phosphorus in meat and bone meal relative to that in monosodium phosphate is not affected by particle size or processing temperature, but ash content of the meat and bone meal seems to affect phosphorus bioavailability. The phosphorus in high-protein, low-ash meat meal of porcine origin seems to be less available than the phosphorus in low-protein, high-ash meal of bovine origin. Overall, the bioavailability of phosphorus in meat and bone meal in this study was relatively high, averaging 85% of the bioavailability of phosphorus in monosodium phosphate.

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