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Non Ruminant Nutrition

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Quantities of ash, Ca, and P in metacarpals, metatarsals, and tibia are better correlated with total body bone ash in growing pigs than ash, Ca, and P in other bones

Su A Lee,[†] L. Vanessa Lagos,[‡] Mike R. Bedford,[||] and Hans H. Stein[†,‡,1]

[†]Department of Animal Sciences, University of Illinois, Urbana 61801, USA, [‡]Division of Nutritional Sciences, University of Illinois, Urbana 61801, USA, [||]AB Vista, Marlborough, SN8 4AN, UK

[1]Corresponding author: hstein@illinois.edu

ORCiD number: 0000-0001-9351-7196 (S. A. Lee).

Abstract

The objective was to determine correlations between individual bones and total body bone ash to identify the bone that is most representative of total body bone ash in growing pigs. Twenty growing pigs were allotted to 1 of 2 diets that were formulated to contain 60% or 100% of the requirement for standardized total tract digestible (**STTD**) P. Both diets had an STTD Ca to STTD P ratio of 1.90:1. Growth performance and carcass weights were determined. Metacarpals, metatarsals, femur, tibia, fibula, 3rd and 4th ribs, and 10th and 11th ribs, and all other bones from the left half of the carcass were collected separately. Each bone was defatted and ashed. Pigs fed the diet containing 100% of required Ca and P had greater ($P < 0.05$) average daily gain, gain to feed, and ash concentration (%) in total and all individual bones except femur and fibula compared with pigs fed the diet containing 60% of required Ca and P. Calcium and P concentrations in bone ash were not affected by dietary treatments. Weights (g) of bone ash, bone Ca, and bone P were greater ($P < 0.05$) or tended to be greater ($P < 0.10$) for pigs fed the diet containing 100% of required Ca and P. Correlation coefficients between the weight of ashed metacarpals, metatarsals, and tibia and the weight of total bone ash were >0.95. In conclusion, metacarpals, metatarsals, and tibia were more representative of total body bone ash compared with other bones.

Key words: bone ash, calcium, phosphorus, pigs

Introduction

The most abundant minerals in the body are Ca and P and most Ca and P are deposited in bone tissue (Crenshaw, 2001; Létourneau-Montminy et al., 2015). Therefore, bone characteristics in pigs are affected by dietary Ca and P (Crenshaw et al., 1981; Lagos et al., 2019a; Vier et al., 2019). Using X-ray absorptiometry, it was determined that femur ash is a better indicator of total body mineral content in 25 kg pigs compared with fibula ash (Crenshaw et al., 2009). As a consequence, in recent work to determine effects of dietary Ca and P on body bone ash in growing pigs, femur ash has been used as a representative bone (González-Vega et al., 2016; Merriman et al., 2017; Lagos et al., 2019b) although metacarpal ash has also sometimes been measured (She et al., 2017; Vier et al., 2019). However, to our knowledge, no data to demonstrate, which bone most

Abbreviations

ADFI	average daily feed intake
ADG	average daily gain
BW	body weight
G:F	gain to feed ratio
STTD	standardized total tract digestible

closely predicts total body bone ash in pigs fed diets containing different levels of Ca and P have been reported.

Feeding diets to growing pigs that are deficient in dietary Ca and P results in reduced growth performance and bone mineralization (Gonzalez-Vega et al., 2016; Lagos et al., 2019a). In particular, a P deficiency results in reduced deposition of bone ash, and therefore, reduced deposition of Ca and P in bone ash as well. In addition, excess provision of Ca in diets with marginal dietary P may exacerbate the P deficiency because Ca reduces digestibility of P (Stein et al., 2011; Lee et al., 2020). Because the size of the bones is influenced by provisions of dietary Ca and P, feeding diets with Ca and P below the requirement usually results in a greater reduction in the quantity (g) of bone ash than in the percentage of bone ash or percentage of bone Ca and P, and the composition of bone ash is relatively constant (Blavi et al., 2019). There are, however, no data to demonstrate if this is the case for the total bone ash in the body of pigs, or if this is only observed in some bones. Therefore, the objective of this experiment was to test the hypothesis that feeding pigs a diet containing 60% of the requirement for Ca and P reduces growth performance, carcass weights, and bone ash, bone Ca, and bone P compared with pigs fed a diet containing 100% of the requirement for Ca and P. The second objective was to determine correlations between individual bones and total body bone ash to identify the bone that is most representative of total body bone ash in growing pigs.

Materials and Methods

The Institutional Animal Care and Use Committee at the University of Illinois reviewed and approved the protocol for the experiment before the animal work was initiated. Pigs were the offspring of Line 359 boars and Camborough females (Pig Improvement Company, Hendersonville, TN).

Animals, housing, feeding, and diets

Twenty growing pigs [initial body weight (**BW**): 40.78 ± 3.47 kg] were allotted to 2 diets using a randomized complete block design with sex and BW as blocking factors for a total of ten replicate pigs (5 gilts and 5 barrows) per diet. Pigs were housed individually in fully slatted pens (0.9 × 1.8 m). Room temperature was controlled and each pen had a feeder, a nipple drinker, and a fully slatted concrete floor. Pigs were allowed ad libitum access to feed and water was available at all times. Pigs were weighed at the start of the experiment, on day 14, and at the conclusion of the experiment (day 28). The amount of feed offered was recorded daily and the amount of feed left in the feeders was recorded on days 14 and 28.

The 2 experimental diets (Table 1) were based on corn and soybean meal and formulated to contain 60% or 100% of the requirement for standardized total tract digestible (**STTD**) P (NRC, 2012). Calcium was included in both diets to maintain an STTD Ca to STTD P ratio of 1.90:1, which is believed to maximize bone ash (Lagos et al., 2019a; Lee et al., 2019a). All nutrients except Ca and P were included in both diets at the requirement for growing pigs (NRC, 2012).

Table 1. Ingredient and nutrient composition of diets fed to growing pigs (as-fed basis)

	% of the STTD P requirement	
Item	60	100
Ingredient, %		
Ground corn	77.91	76.85
Soybean meal	19.00	19.00
Choice white grease	1.00	1.00
Calcium carbonate	0.81	1.11
Dicalcium phosphate	0.26	1.02
L-Lys·HCl	0.33	0.33
DL-Met	0.05	0.05
L-Thr	0.09	0.09
Sodium chloride	0.40	0.40
Vitamin–mineral premix[1]	0.15	0.15
Analyzed composition, %		
Dry matter	89.18	89.64
Gross energy	3,906	3,878
Crude protein	14.53	14.45
Ash	3.54	4.24
Calcium	0.49	0.74
Phosphorus	0.39	0.50
STTD Ca[2]	0.329	0.544
STTD P[2]	0.173	0.286
STTD Ca to STTD P ratio[2]	1.90	1.90

[1]The vitamin–mineral premix provided the following quantities of vitamins and microminerals per kilogram of complete diet: vitamin A as retinyl acetate, 11,150 IU; vitamin D_3 as cholecalciferol, 2,210 IU; vitamin E as selenium yeast, 66 IU; vitamin K as menadione nicotinamide bisulfate, 1.42 mg; thiamin as thiamine mononitrate, 1.10 mg; riboflavin, 6.59 mg; pyridoxine as pyridoxine hydrochloride, 1.00 mg; vitamin B_{12}, 0.03 mg; D-pantothenic acid as D-calcium pantothenate, 23.6 mg; niacin, 44.1 mg; folic acid, 1.59 mg; biotin, 0.44 mg; Cu, 20 mg as copper chloride; Fe, 125 mg as iron sulfate; I, 1.26 mg as ethylenediamine dihydriodide; Mn, 60.2 mg as manganese hydroxychloride; Se, 0.30 mg as sodium selenite and selenium yeast; and Zn, 125.1 mg as zinc hydroxychloride.
[2]Values for the STTD Ca and STTD P were calculated rather than analyzed (NRC, 2012; Lee et al., 2019b); the 1.90:1 ratio between STTD Ca and STTD P was expected to maximize bone ash of 40 kg pigs (Lagos et al., 2019a; Lee et al., 2019a).

Carcass weights and bone measurements

On the last day of the experiment, pigs were transported to the University of Illinois Meat Science Laboratory. Pigs were fasted for ~18 hr and weighed again to determine ending live weight. Following exsanguination, blood was weighed. Pigs were scalded, dehaired, and singed to remove all hair from the carcass, and toenails and tail were removed. The weight of the viscera (i.e., heart, kidneys, liver, gall bladder, spleen, lungs, trachea, reproductive tract, and emptied gastrointestinal tract) was recorded. Weights of bone with fat and muscle tissues on, feet, skin, and soft tissue from the left side of the carcass were recorded the day after pigs were killed. The weight of the left sides and of the heads was also recorded.

Third and 4th metacarpals, 3rd and 4th metatarsals, femur, tibia, fibula, 3rd and 4th ribs, and 10th and 11th ribs from the left half of the carcass were collected separately and stored at −20 °C. All remaining bones from the left half of the carcass except the bones in the head and tail were combined, the weight was recorded, and these bones were also stored at −20 °C. Each of the frozen bone samples was thawed and autoclaved at 125 °C for 55 min. Marrow and fat were removed and bones were soaked in petroleum ether under a chemical hood for 72 hr.

Defatted bone samples were dried for 2 hr at 135 °C and weighed and then ashed overnight at 600 °C. The weight of bone ash (g) in each sample was recorded and concentration of ash (%) was calculated as a percentage of bone dry weight. Concentrations of Ca and P (%) were calculated as a percentage of bone ash. Weight of defatted total body bone ash (g) was calculated as the sum of the weight of individual bones and all remaining bones. Because the entire half of the body was used to measure body bone ash, weight of total and individual bone ash was calculated by multiplying the bone ash weight from the left half by 2.

Chemical analysis

Diet samples were analyzed for dry matter (AOAC Int., 2019; method 930.15). Ash in diet and bone samples was also analyzed (AOAC Int., 2019; method 942.05). The gross energy in diet samples was measured using an isoperibol bomb calorimeter (Model 6400, Parr Instruments, Moline, IL). Crude protein in diet samples was calculated as N × 6.25 and N was measured by the combustion procedure (method 990.03; AOAC Int., 2019) using a LECO FP628 nitrogen analyzer (LECO Corp., Saint Joseph, MI). Calcium and P in diet and bone samples were analyzed (AOAC Int., 2019; method 985.01 A, B, and C) by inductively coupled plasma spectroscopy (ICP-OES; Avio 200, PerkinElmer, Waltham, MA). Sample preparation included dry ashing at 600 °C for 4 hr (AOAC Int., 2019; method 942.05) and wet digestion with nitric acids (method 3050 B; U.S. Environmental Protection Agency, 1996).

Statistical analysis

Normality of residuals was verified using the UNIVARIATE procedure (SAS Inst. Inc., Cary, NC) and homogeneity was also confirmed. Outliers were identified as values that deviated from 1st- and 3rd-quartile by more than 3 times the interquartile range within treatment (Tukey, 1977). However, no outliers were identified. The pig was the experimental unit. The statistical model included diet as fixed effect and sex and BW within sex as random effects. To compare Ca:P ratios in bone ash, the statistical model included the bone as fixed effect and sex, BW within sex, and diet as random effects. LSmeans were separated using the pdiff option with Tukey's adjustment. Data were analyzed using MIXED procedures of SAS (SAS Institute Inc., Cary, NC). Correlation coefficients (*r*) between total body bone and individual bones were determined using the CORR procedure of SAS. Statistical significance and tendency were considered at $P < 0.05$ and $0.05 \le P < 0.10$, respectively.

Results

All pigs remained healthy and had normal feed intake throughout the experiment. There were no effects of sex on any response criteria.

Growth performance and carcass weights

There was no effect of dietary Ca and P on BW at day 14, but pigs fed the diet containing 100% of the requirement for Ca and P tended to have greater ($P < 0.10$) BW on day 28 (Table 2). From days 1 to 14, average daily gain (**ADG**) and average daily feed intake (**ADFI**) of pigs were not affected by dietary Ca and P, but gain-to-feed ratio (**G:F**) of pigs fed the diet containing 100% of the requirement for Ca and P was greater ($P < 0.01$) compared with pigs fed the diet containing 60% of the requirement for Ca and P. There was no effect of dietary Ca and P on ADG, ADFI, or G:F of pigs from days 14 to 28. Overall, pigs fed the diet containing 100% of the requirement for Ca and P had greater ($P < 0.05$) ADG and G:F compared with pigs fed the diet containing 60% of the requirement for Ca and P, but there was no difference in ADFI between the 2 diets. There was no effect of dietary Ca and P on carcass weights of pigs, but the weight of head of pigs fed the diet containing 100% of the requirement for Ca and P was greater ($P < 0.05$) compared with pigs fed the diet containing 60% of the requirement (Table 3).

Bone ash

Weight (g) of dried, defatted bone in total and all individual bones was greater ($P < 0.05$) for pigs fed the diet containing 100% of the requirement for Ca and P compared with pigs fed the diet containing 60% of the requirement (Table 4). Concentration of ash (%) in total and in all individual bones

Table 2. Growth performance of pigs fed experimental diets ($n = 10$)[1]

	% of the STTD P requirement			
Item[1]	60	100	SEM	P-value
BW, kg				
Day 1	40.64	40.92	-	-
Day 14	52.22	54.14	2.29	0.116
Day 28	67.30	70.54	1.46	0.060
Days 1 to 14				
ADG, kg	0.83	0.94	0.06	0.102
ADFI, kg	2.23	2.16	0.07	0.510
G:F	0.37	0.44	0.03	0.001
Days 14 to 28				
ADG, kg	1.08	1.17	0.11	0.196
ADFI, kg	2.54	2.51	0.17	0.881
G:F	0.43	0.47	0.02	0.145
Overall				
ADG, kg	0.95	1.06	0.04	0.037
ADFI, kg	2.39	2.34	0.12	0.693
G:F	0.40	0.45	0.01	< 0.001

[1] Twenty growing pigs were housed individually and fed the 2 experimental diets with 10 pigs fed each diet.

Table 3. Carcass weights of pigs fed experimental diets ($n = 10$)

	% of the STTD P requirement			
Item	60	100	SEM	P-value
Ending live BW[1], kg	64.89	67.13	1.38	0.162
Hot carcass weight, kg	47.92	49.62	1.11	0.239
Left side				
Side weight, kg	23.43	24.18	0.56	0.235
Bone[2], kg	3.55	3.67	0.08	0.227
Feet, kg	0.74	0.75	0.02	0.741
Skin, kg	2.49	2.40	0.05	0.259
Soft tissue, kg	16.58	17.33	0.46	0.225
Blood, kg	2.86	2.96	0.14	0.570
Head, kg	5.13	5.32	0.08	0.004
Viscera[3], kg	6.99	7.09	0.18	0.483

[1]Ending live weight was determined ~18 hr after final BW had been recorded. No feed was provided during this period.
[2]Weights of bone were measured with tissues on and head, tail, and feet were not included.
[3]Visceral weight excluded the weight of digesta in the gastrointestinal tract.

Table 4. Bone ash concentration (%) and weight (g) in total body bone, metacarpals, metatarsals, femur, tibia, fibula, and ribs from pigs fed experimental diets[1] (*n* = 10)

	% of the STTD P requirement			
Item	60	100	SEM	P-value
Dried, defatted bone, g				
Total	665.81	827.18	22.73	<0.001
3rd and 4th metacarpals	10.02	11.26	0.33	0.007
3rd and 4th metatarsals	10.63	12.56	0.40	0.004
Femur	64.63	76.25	3.29	0.025
Tibia	30.63	37.44	1.12	0.001
Fibula	4.61	5.39	0.16	0.005
3rd and 4th ribs	7.88	9.84	0.26	< 0.001
10th and 11th ribs	8.05	10.49	0.60	0.002
Bone ash, % defatted and dried bones				
Total	54.61	57.17	0.39	< 0.001
3rd and 4th metacarpals	59.23	61.53	0.36	< 0.001
3rd and 4th metatarsals	59.24	61.32	0.37	< 0.001
Femur	56.89	58.17	0.53	0.118
Tibia	60.25	61.73	0.39	0.020
Fibula	59.81	61.18	0.65	0.089
3rd and 4th ribs	55.60	58.10	0.51	0.003
10th and 11th ribs	55.04	57.65	0.59	0.002
Bone ash, g				
Total	727.76	946.70	28.10	< 0.001
3rd and 4th metacarpals	11.87	13.84	0.38	0.001
3rd and 4th metatarsals	12.60	15.40	0.48	0.001
Femur	73.66	88.88	4.16	0.025
Tibia	36.91	46.29	1.51	0.001
Fibula	5.51	6.60	0.20	0.001
3rd and 4th ribs	8.76	11.44	0.32	< 0.001
10th and 11th ribs	8.87	12.09	0.75	0.001

[1]All bones were collected from the left half of the carcass. Therefore, weight of bone ash was calculated by multiplying by 2. Total body bones represent the sum of metacarpals, metatarsals, femur, tibia, fibula, ribs, and miscellaneous bones, but head and tail were not included.

Table 5. Concentrations of Ca and P and Ca to P ratio in bone ash from pigs fed experimental diets[1] (*n* = 10)

	% of the STTD P requirement			
Item	60	100	SEM	P-value
Ca in bone ash, %				
Total	36.29	36.93	0.47	0.262
3rd and 4th metacarpals	38.47	39.01	0.58	0.261
3rd and 4th metatarsals	36.89	36.80	0.39	0.829
Femur	41.72	41.87	1.85	0.824
Tibia	36.82	36.44	0.38	0.451
Fibula	36.56	37.03	0.29	0.129
3rd and 4th ribs	35.87	35.79	0.31	0.714
10th and 11th ribs	35.98	35.84	0.52	0.755
P in bone ash, %				
Total	18.67	18.66	0.18	0.965
3rd and 4th metacarpals	18.76	18.77	0.24	0.966
3rd and 4th metatarsals	19.81	19.87	0.22	0.749
Femur	18.16	18.16	0.72	0.995
Tibia	19.75	19.47	0.08	0.006
Fibula	19.99	20.05	0.13	0.622
3rd and 4th ribs	19.58	19.86	0.12	0.098
10th and 11th ribs	19.27	19.59	0.15	0.132
Ca:P ratio				
Total[c]	1.95	1.98	0.03	0.304
3rd and 4th metacarpals[b]	2.05	2.08	0.02	0.441
3rd and 4th metatarsals[d]	1.86	1.85	0.02	0.666
Femur[a]	2.30	2.31	0.03	0.901
Tibia[d]	1.86	1.87	0.02	0.793
Fibula[d]	1.83	1.85	0.02	0.312
3rd and 4th ribs[d]	1.83	1.80	0.01	0.136
10th and 11th ribs[d]	1.87	1.83	0.04	0.196

[a–d]Within a column, means with different superscripts differ ($P < 0.05$). The statistical model included different bone as fixed effect and sex, BW within sex, and diet as random effects.
[1]All bones were collected from the left half of the carcass. Total body bones represent the sum of metacarpals, metatarsals, femur, tibia, fibula, ribs, and miscellaneous bones, but head and tail were not included.

except femur and fibula was greater ($P < 0.05$) for pigs fed the diet containing 100% of the requirement for Ca and P compared with pigs fed the diet containing 60% of the requirement. Ash concentration (%) in the fibula tended to be greater ($P < 0.10$) if pigs were fed Ca and P at the requirement compared with pigs fed Ca and P below the requirement. Weights of total and individual bone ash (g) were greater ($P < 0.05$) for pigs fed the diet containing 100% of the requirement for Ca and P compared with pigs fed the diet containing 60% of the requirement.

Concentrations of Ca and P (%) in total and all individual bones except concentration of P in tibia were not affected by dietary Ca and P (Table 5). Concentration of P (%) in tibia was greater ($P < 0.05$) if pigs were fed the diet containing 60% of the requirement for Ca and P compared with pigs fed the diet containing 100% of the requirement. Calcium to P ratios did not differ between pigs fed the 2 diets. Regardless of diet, the Ca to P ratio in femur was greater ($P < 0.05$) than ratios in other bones, and the Ca to P ratio in metacarpals was greater ($P < 0.05$) than in total bone ash and in all other bones except femurs. The Ca to P ratio in total body bone ash was greater ($P < 0.05$) than in metatarsals, tibia, fibula, and ribs, but there was no difference in Ca:P ratio among metatarsals, tibia, fibula, 3rd and 4th ribs, and 10th and 11th ribs. For total bones and all individual bones, weights of bone Ca and bone P (g) were greater ($P < 0.05$) or tended to be greater ($P < 0.10$) for pigs fed the diet containing 100% of the requirement for Ca and P compared with pigs fed the diet containing 60% of the requirement (Table 6).

There were positive correlations ($P < 0.05$) between individual bones and total body bone for the concentration of ash (%), weight of bone ash (g), and weights of bone Ca and bone P (g; Table 7). Correlation coefficients between the weight (g) of ashed metacarpals, metatarsals, and tibia and the weight (g) of total body bone ash were >0.950, whereas correlation coefficients between the weights of ashed femur, fibula, 3rd and 4th ribs, and 10th and 11th ribs and total body ash were 0.846, 0.929, 0.876, and 0.817, respectively. Unlike bone ash weight, all correlation coefficients between individual bones and total body bone ash for percentage ash were <0.750 and all correlation coefficients for percentage Ca and P were close to 0.

Discussion

The analyzed Ca and P in both diets were in agreement with calculated values. It was expected that dietary ADFI would be reduced for pigs fed the diet with reduced Ca and P compared with the requirement because reduced dietary P often results in reduced ADFI. However, the reduction in ADFI as dietary P is reduced is primarily observed in diets where Ca is not reduced or where Ca is in excess of the requirement (Merriman et al., 2017). It is possible that the reason for the lack of a reduction in ADFI in the present experiment is that diets were formulated to have the same Ca:P ratio and Ca, therefore, was also reduced as dietary P was reduced.

Results demonstrated that growth performance and body bone ash of pigs were reduced if dietary Ca and P are below the requirement, but the negative effect was greater for bone ash than for growth performance. The growth difference was 12% between treatments whereas for bone ash weight it was 16% to 36% depending on the bone. This implies that compared with pigs fed 100% of the requirement for Ca and P, pigs that were fed the diet containing 60% of the requirement utilized a greater proportion of dietary Ca and P for the growth of soft tissues and a lower proportion for bone tissue synthesis. These data support the hypothesis that Ca and P requirements to maximize growth performance are less than requirements to maximize bone ash (NRC, 2012).

Bone mineralization is likely affected by the age of pigs and percentage of bone ash in finishing pigs is greater than in growing pigs (Crenshaw et al., 1981; Lagos et al., 2021). The observation that bone ash in pigs with a final BW of 71 kg was 59.6% indicates that this value was between previous values obtained from growing pigs (BW = 26 kg; bone ash = 50%) and finishing pigs (BW = 129 kg; bone ash = 61%; Lagos et al., 2021).

The observation that bone ash was reduced by lowering Ca and P in diets was in agreement with previous data (González-Vega et al., 2016; Lagos et al., 2019a; Vier et al., 2019). Likewise, the fact that concentrations of Ca and P (%) in ashed bone were not affected by dietary Ca and P was also in agreement with previous data (González-Vega et al., 2016). Whereas differences in percentage of ash in defatted bones between pigs fed diets with 60% and 100% of the requirement for Ca and P were small for most bones, the weights (g) of bone ash, bone Ca, and bone P were dramatically affected by dietary Ca and P. This was also observed in previous experiments (Crenshaw et al., 1981; González-Vega et al., 2016; Lagos et al., 2019a; Schlegel and Gutzwiller, 2020), and indicates that differences observed in bone ash weight are likely a result of differences in the size of bones rather than differences in the percentage of ash in the bone. This observation indicates that the composition of bone tissue does not change to a great extent, regardless of dietary provisions of Ca and P. However, mineral bone density is affected by diet Ca and P concentrations (Schlegel and Gutzwiller, 2020), which is the reason the weight of bone ash changed without changes in percentage of ash, Ca, and P. This observation may also be the reason for the lower correlation coefficients for percentage ash between ash in individual bone and total body bone ash and no correlation for concentrations of Ca and P. The observation that Ca:P ratios in bone ash differed among different bones was surprising because the majority of bone ash is deposited as hydroxyapatite, which has a Ca:P ratio of 2.16:1.00. It is, therefore, possible that some of the Ca and P in bones may be stored in different minerals than hydroxyapatite and that this proportion varies among bones in the body.

Most Ca and P in the body are present in skeletal tissue (Crenshaw, 2001). Therefore, analyzing bone ash has been a standard procedure for determining Ca and P availability or deposition (Cromwell, 1992; NRC, 1998). Different bones have been used, but most often bones from legs and feet have been used because these bones are believed to represent total body bone ash (Crenshaw et al., 1981; 2009). The reason legs or feet are frequently used may be that it is relatively easier to collect these bones compared with bones in other parts of the body. The observation that correlation

Table 6. Weights of bone Ca and P in pigs fed experimental diets[1] (*n* = 10)

Item	% of the STTD P requirement		SEM	*P*-value
	60	100		
Bone Ca, g				
Total	264.10	349.24	10.55	<0.001
3rd and 4th metacarpals	4.57	5.40	0.14	<0.001
3rd and 4th metatarsals	4.64	5.67	0.18	0.002
Femur	30.82	37.36	3.08	0.051
Tibia	13.59	16.89	0.63	0.002
Fibula	2.02	2.44	0.08	<0.001
3rd and 4th ribs	3.14	4.09	0.12	<0.001
10th and 11th ribs	3.19	4.33	0.32	0.002
Bone P, g				
Total	135.54	176.45	4.51	<0.001
3rd and 4th metacarpals	2.23	2.60	0.08	0.002
3rd and 4th metatarsals	2.49	3.06	0.10	0.002
Femur	13.39	16.13	1.20	0.026
Tibia	7.29	9.01	0.30	0.001
Fibula	1.10	1.32	0.04	0.001
3rd and 4th ribs	1.71	2.27	0.07	<0.001
10th and 11th ribs	1.71	2.37	0.14	0.001

[1]All bones were collected from the left half of the carcass. Therefore, weight of bone ash was calculated by multiplying by 2. Total body bones represent the sum of metacarpals, metatarsals, femur, tibia, fibula, ribs, and miscellaneous bones, but head and tail were not included.

Table 7. Correlation coefficients (*r*) between individual bones and total body bone concentration or weight of ash, Ca, and P[1] (*n* = 20)

Item	Total body bone					
	Bone ash, %	Bone Ca, %	Bone P, %	Bone ash, g	Bone Ca, g	Bone P, g
3rd and 4th metacarpals	0.741***	−0.324	−0.014	0.956***	0.938***	0.915***
3rd and 4th metatarsals	0.735***	0.053	0.131	0.971***	0.943***	0.939***
Femur	0.722***	−0.357	−0.056	0.846***	0.701**	0.758***
Tibia	0.644**	0.0002	−0.183	0.957***	0.909***	0.926***
Fibula	0.658**	−0.233	−0.019	0.929***	0.899***	0.891***
3rd and 4th ribs	0.539*	−0.021	−0.064	0.876***	0.881***	0.886***
10th and 11th ribs	0.704**	0.089	−0.218	0.817***	0.805***	0.831***

*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.
[1]All bones were collected from the left half of the carcass. Therefore, weight of bone ash was calculated by multiplying by 2. Total body bones represent the sum of metacarpals, metatarsals, femur, tibia, fibula, ribs, and miscellaneous bones, but head and tail were not included.

coefficients between bone ash weight of metacarpals, metatarsals, and tibia and total body bone ash weight were >0.950, whereas ribs had lower coefficients is in agreement with data, indicating that physical characteristics of metacarpals and metatarsals from pigs at 3 to 5 mo of age were more sensitive to different levels of dietary Ca and P than were other individual bones (Crenshaw et al., 1981). However, femur was a better indicator of total mineral content or physiological characteristics of bone in pigs at an early age compared with other individual bones (Crenshaw et al., 1981; 2009). It is possible that the most representative bone for total body bone ash depends on the age of pigs, but more research is needed to test this hypothesis by analyzing total body bone ash from pigs at different age groups.

Conclusion

Providing dietary Ca and P below the requirement negatively affected growth performance of pigs and reduced total body bone ash in growing pigs. As weight of total body bone ash increased, the weight of all ashed bones also increased. Weights (g) of bone ash, bone Ca, and bone P (g) better represent bone mineralization in growing pigs than concentrations (%) of bone ash, Ca, and P. Metacarpals, metatarsals, and tibia were more representative of total body bone ash in 41 to 69 kg growing pigs compared with femur, fibula, and ribs.

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Conflict of Interest Statement

The authors declare no real or perceived conflicts of interest.

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