Estimation of standardized mineral availabilities in feedstuffs for broilers¹

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ABSTRACT: Two experiments were conducted to estimate standardized mineral (Ca, P, Cu, Fe, Mn, and Zn) availabilities (SMA) in commonly used feedstuffs and verify the additivity of SMA for diet formulation of broilers. In Exp. 1, after 3 d of acclimation, a total of 96 22-d-old Arbor Acres male broilers were fasted for 24 h. Then the broilers were fed a mineral-free, corn, soybean meal (SBM), or corn-soybean meal (C-SBM) diet for 4 h and the excreta samples were collected for 48 h after feed withdrawal (totally 52 h). The results showed that the endogenous losses of Ca, P, Cu, Fe, Mn, and Zn for chickens fed the mineral-free diet were 13.7, 113, 0.064, 0.593, 0.094, and 0.132 mg/52 h per bird, respectively. The standardized availability values of Ca, P, Cu, Fe, Mn, and Zn were 20.8%, 39.6%, 37.8%, 47.4%, 10.7%, and 45.2% in corn as well as 54.3%, 51.1%, 39.7%, 27.6%, 26.1%, and 51.0% in SBM, respectively. The determined values of SMA in C-SBM diet were close

($P > 0.09$) to the predicted summations of SMA from corn and SBM (Ca, 48.6% vs. 48.1%; P, 44.9% vs. 46.0%; Cu, 41.7% vs. 39.1%; Fe, 35.3% vs. 33.2%; Mn, 22.7% vs. 22.2%; Zn, 46.2% vs. 48.4%). In Exp. 2, a total of 144 22-d-old Arbor Acres male broilers were used to estimate the SMA values in 6 feedstuffs by using the above procedure. The results showed that the standardized availability values in wheat, wheat bran, corn distillers dried grains with solubles, cottonseed meal, rapeseed meal, and corn gluten meal diets were 24.7% to 55.8% for Ca, 35.6% to 46.6% for P, 24.5% to 45.7% for Cu, 21.6% to 43.5% for Fe, 9.3% to 34.5% for Mn, and 22.9% to 52.9% for Zn, respectively. The results from the present study indicated that the mineral-free diet could be used for estimating the endogenous losses of the above minerals and SMA values of feedstuffs for broilers, and the estimates of SMA in feedstuffs for the C-SBM diet formulation were additive.

Key words: broiler, endogenous mineral loss, feedstuff, standardized mineral availability

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INTRODUCTION

Minerals are required for normal physiological functions of chickens (NRC, 1994). However, overfeeding of minerals in commercial broiler production will cause excessive mineral excretion, and this may cause severe environmental pollution (Bao et al., 2007; Lu et al., 2017). To accurately meet mineral requirements of broilers without overfeeding, the contribution of available minerals from individual feedstuff must be quantified. Although estimations of apparent mineral availabilities (AMA) in diets have been abundantly reported (Lan et al., 2002; Ravindran et al., 2006; Thomas and Ravindran, 2010), these estimates do not account for the contributions of endogenous mineral losses (EML). Nwokolo et al. (1976) reported a rapid and convenient procedure for determining standardized mineral availabilities (SMA) of feedstuffs by using a mineral-free diet to measure EML. Then many studies demonstrated that this procedure could be used for estimating SMA in feedstuffs for chickens (Nwokolo and Bragg, 1980; Aw-yong et al., 1983; Chan, 1983). However, it was also reported that SMA in feedstuffs was overestimated due to insufficient time for emptying chyme residues and excreta collection (Nwokolo, 1987; Zhong, 2006; Liu et al., 2012a). Under commercial conditions, chickens are fed a combination of feedstuffs in a formulated diet and it is currently not known if the SMA for individual feedstuff is additive in these formulated diets. Results from our laboratory have indicated that the standardized P availability (SPA) value for broilers from a corn–soybean meal (C-SBM) diet was close to its predicted value by using a P-free diet to measure EML (Liu et al., 2012b). But the P-free diet can only be used to calculate the SPA. Whether the mineral-free diet can be used for the estimation of the EML and SMA values, and whether the values of SMA for feed ingredients are additive in formulated diets for broilers need to be verified. Therefore, it was hypothesized that the mineral-free diet could be used for the estimation of the EML and SMA values of feedstuffs, and dietary SMA could be estimated by the addition of the SMA of dietary components in a formulated ration. The objectives of the current study were to determine the EML and the SMA of commonly used feedstuffs by using the mineral-free diet, and verify the additivity of SMA in feedstuffs for the C-SBM diet formulation of broilers.

MATERIALS AND METHODS

All experimental procedures were approved by the Animal Management Committee (in charge of

animal welfare) of the Institute of Animal Science, Chinese Academy of Agricultural Sciences (IAS-CAAS, Beijing, China) and performed in accordance with the guidelines. Ethical approval on animal survival was given by the animal ethics committee of IAS-CAAS.

Experimental Design and Treatments

A completely randomized design was used in the 2 experiments. There were a total of 4 dietary treatments, including the mineral-free (dextrose-gelatin), corn, soybean meal (SBM), and C-SBM diets in Exp. 1, and 6 dietary treatments, including wheat, wheat bran, corn distillers dried grains with solubles (DDGS), cottonseed meal, rapeseed meal, and corn gluten meal diets in Exp. 2.

Birds and Diets

From days 1 to 21 posthatching, all male Arbor Acres broiler chickens from a local hatchery (Huadu Broiler Breeding Corp., Beijing, China) were housed in electrically heated, thermostatically controlled, stainless cages coated with plastic (100 × 50 × 45 cm) and equipped with fiberglass feeders and waterers. Feed and tap water were available ad libitum.

Chickens were fed a standard broiler starter diet during days 1 to 21 posthatching and grower diet during recovery (Table 1) and maintained on a 24-h constant light schedule. The mineral-free diet was primarily formulated by using dextrose, gelatin, and synthetic amino acids to supply energy, protein, or amino acids. All experimental diets were formulated by replacing dextrose or gelatin with tested feed ingredients and contained equal contents of ME, CP, and main amino acids to those of the mineral-free diet. The amount of soybean oil was not more than 4%. These diets were formulated to meet or exceed the nutrient requirements of growing broilers from 3 to 6 wk of age recommended by NRC (1994) except minerals. The analyzed nutrient composition of 8 feedstuffs used in the 2 experiments is shown in Table 2. The composition and nutrient contents of the experimental diets in the 2 experiments are shown in Tables 3 and 4, respectively. During the experimental period, all birds were allowed ad libitum access to the deionized water.

Experiment 1. On day 22 at 0800 h, a total of 96 chickens with similar body weight were selected and randomly allotted to 1 of 4 dietary treatments with 6 replicate cages of 4 birds each. Chickens were sutured with a threaded hollow

Table 1. Composition of the starter and grower diets for broiler chickens (as-fed basis)

Item	Starter diet	Grower diet
Ingredient, %		
Corn	52.40	60.13
Soybean meal	39.50	33.00
Soybean oil	4.00	3.50
Limestone ¹	1.10	1.20
Dicalcium phosphate ¹	2.10	1.55
Salt ¹	0.30	0.30
DL-Met	0.23	0.09
Micro-nutrients ^{1,2}	0.37	0.23
Nutrient composition, %		
ME, MJ/kg	12.46	12.54
CP ³	21.39	19.86
Ca ³	0.98	0.91
Nonphytate P	0.45	0.35
Lys	1.10	0.96
Met	0.56	0.39
Met + Cys	0.91	0.72

¹Feed grade.

²For starter diet, provided per kilogram of diet: vitamin A (all-trans retinol acetate), 12,500 IU; cholecalciferol, 3,750 IU; vitamin E (all-rac- α -tocopherol acetate), 20 IU; vitamin K (menadione sodium bisulfate), 2.5 mg; thiamin (thiamin mononitrate), 2.5 mg; riboflavin, 8.0 mg; vitamin B₆, 2.5 mg; vitamin B₁₂, 0.015 mg; calcium pantothenate, 12.5 mg; niacin, 32.5 mg; folic acid, 1.25 mg; biotin, 0.125 mg; choline (choline chloride), 700 mg; Cu (CuSO₄·5H₂O), 8 mg; Mn (MnSO₄·H₂O), 100 mg; Fe (FeSO₄·7H₂O), 80 mg; Zn (ZnSO₄·7H₂O), 60 mg; I (KI), 0.35 mg; Se (NaSeO₃), 0.15 mg. For grower diet, provided per kilogram of diet: vitamin A (all-trans retinolacetate) 10,000 IU; cholecalciferol 3,000 IU; vitamin E (all-rac- α -tocopherol acetate) 16 IU; vitamin K (as menadione sodium bisulfate) 2.0 mg; thiamin (thiamin mononitrate) 2.0 mg; riboflavin 6.4 mg; vitamin B₆ 2.0 mg; vitamin B₁₂ 0.01 mg; calcium pantothenate 10 mg; niacin 26.0 mg; folic acid 1.0 mg; biotin 0.10 mg; choline (choline chloride) 500 mg; Cu (CuSO₄·5H₂O), 8 mg; Mn (MnSO₄·H₂O) 80 mg; Zn (ZnSO₄·7H₂O) 40 mg; Fe (FeSO₄·7H₂O) 80 mg; I (KI) 0.35 mg; Se (Na₂SeO₃) 0.15 mg.

³Determined by analysis and each value based on triplicate determinations.

plastic cap around the vent for screwing a plastic bag to collect excreta (Adeola et al., 1997). The cap and plastic bag could prevent contamination of excreta by feathers, skin, and other debris. Following attachment of the plastic caps, the birds recovered and acclimated for 62 h. On day 24 at 2200 h, feeds were withdrawn and all chickens were fasted overnight for 8 h. On day 25 at 0600 h, all chickens were fed a mineral-free diet for 4 h and then fasted for 24 h to clear residues in the gastrointestinal tract (Liu et al., 2012a). On day 26 at 1000 h, feeders (with weighed feeds) and fecal-collecting bags were installed from cages 1 to 24. The test diets (Table 3) were provided from cages 1 to 24 for 4 h, and then the remaining diets were also withdrawn and weighed from cages 1 to 24. Excreta collections started from feeding and lasted for a total of 52 h.

Experiment 2. On day 22 at 0800 h, a total of 144 chickens with similar body weight were selected and randomly allotted to 1 of 6 dietary treatments with 6 replicate cages of 4 birds each. The birds were prepared, fed, and sampled as described in Exp. 1.

Sample Collections and Preparations

In the 2 experiments, the excreta-collecting bag attached to the anus was fully filled with excreta in about 4 h, so excreta samples were collected at approximately 4-h intervals. Excreta samples of 4 chickens in each replicate cage were then pooled into 1 sample and dried in a forced-air oven at 65 °C. Ingredients and diets, as well as dried excreta samples, were ground through a 0.45-mm sieve by using a grinding mill (Yanshanzhengde Inc., Beijing, China) to facilitate analyses.

Determinations of Dry Matter, CP, Ca, P, Cu, Fe, Mn, and Zn Concentrations

Dry matter contents in feed ingredients, diets, and excreta samples were determined by using AOAC methods (2000), and CP contents in ingredients and diets were analyzed by Kjeldahl N (Thiex et al., 2002). The Ca, Cu, Fe, Mn, and Zn concentrations in ingredients, diets, and excreta samples were determined by inductively coupled plasma spectroscopy (Model IRIS Intrepid II; Thermal Jarrell Ash, Waltham, MA). Total P concentrations in ingredients, diets, and excreta samples were determined by using a molybdovanadate spectrophotometric method (AOAC, 2000), and phytate P determined by using the ferric precipitation method (Rutherford et al., 2004).

Calculations and Statistical Analyses

The determined values of AMA and SMA were calculated according to the following equations:

$$\text{AMA (\%)} = (M_I - M_O) / M_I \times 100, \quad (1)$$

$$\text{SMA (\%)} = [M_I - (M_O - \text{EML}_B)] / M_I \times 100, \quad (2)$$

where M_I is the total mineral intake (mg) of birds fed the tested diet, M_O is the total excreta mineral output (mg) of birds fed the test diet, and EML_B is the total excreta mineral output (mg) of chickens fed the mineral-free diet.

The predicted values of SMA for the C-SBM diet were based on the individually determined SMA for ingredients and their mineral intake according to the following equation:

Table 2. Nutritional composition of 8 ingredients for test (as-fed basis)

Item	Corn	SBM ¹	Wheat	Wheat bran	Corn DDGS ¹	Cottonseed meal	Rapeseed meal	Corn gluten meal
Proximate nutrient, %								
Dry matter ²	88.3	90.4	88.7	89.2	88.4	90.7	90.8	88.3
CP ²	8.3	47.5	14.4	18.6	26.1	45.0	38.5	57.0
Ether extract	3.5	1.5	1.7	3.9	10.1	0.5	1.4	6.0
Crude fiber	1.6	3.3	1.9	6.5	6.6	10.5	11.8	2.1
Crude ash	2.0	6.1	2.5	4.8	5.7	8.1	8.2	3.1
Nitrogen free extract	72.9	32.0	68.2	55.4	39.9	26.5	30.9	20.1
Amino acid, %								
Arg	0.32	3.40	0.54	1.23	0.98	4.30	1.48	1.84
His	0.23	1.29	0.34	0.50	0.72	1.46	0.94	1.37
Ile	0.28	2.04	0.47	0.39	0.85	1.33	1.30	2.27
Leu	0.97	3.84	0.88	1.02	3.20	2.32	2.72	10.71
Lys	0.28	3.01	0.38	0.75	0.75	1.74	1.00	1.04
Met	0.19	0.66	0.20	0.28	0.56	0.58	0.71	1.40
Phe	0.37	2.35	0.61	0.66	1.15	2.37	1.60	3.43
Thr	0.28	1.89	0.38	0.59	0.93	1.38	1.46	1.97
Trp	0.06	0.61	0.16	0.30	0.19	0.47	0.42	0.25
Val	0.39	2.19	0.59	0.76	1.24	1.94	1.78	2.68
Ala	0.66	2.16	0.50	0.76	1.93	1.72	1.67	5.48
Asp	0.55	5.21	0.63	0.93	1.52	4.02	2.24	3.48
Cys	0.18	0.63	0.29	0.38	0.44	0.62	0.67	0.92
Glu	1.64	7.72	4.09	3.54	3.96	9.20	5.83	11.50
Gly	0.33	1.94	0.55	0.82	0.94	1.81	1.73	1.61
Pro	0.76	2.46	1.37	1.22	2.19	1.53	2.35	5.65
Ser	0.35	2.30	0.60	0.72	1.17	1.72	1.42	2.88
Tyr	0.29	1.56	0.33	0.31	0.91	1.10	0.91	2.57
Mineral ² , %								
Ca	0.02	0.2	0.05	0.1	0.03	0.23	0.78	0.03
Total P	0.25	0.73	0.34	1.03	0.75	1.01	1.02	0.73
Nonphytate P ³	0.06	0.26	0.09	0.29	0.32	0.35	0.24	0.20
K	0.27	2.33	0.43	1.03	1.02	1.61	1.34	0.78
Mg	0.10	0.29	0.11	0.42	0.30	0.64	0.48	0.25
Cu, mg/kg	2.14	12.8	5.35	12.2	3.38	12.9	7.37	11.2
Fe, mg/kg	40.1	183	46.0	143	198	138	405	554
Mn, mg/kg	5.06	38.6	32.79	116	16.4	16.8	61.1	13.6
Zn, mg/kg	14.2	45.0	33.2	84.1	44.9	53.8	76.9	39.1

¹SBM = soybean meal; DDGS = distillers dried grains with solubles.

²Determined by analysis and each value based on triplicate determinations.

³Defined as the calculated difference between total P and phytate P contents.

$$\text{Predicted SMA (\%)} = \frac{\left[\begin{matrix} M_{II} \times \text{SMA}_{II} \\ + M_{III} \times \text{SMA}_{III} \end{matrix} \right]}{(M_{II} + M_{III}) \times 100}, \quad (3)$$

where M_{II} is the mineral intake (mg) from corn in the C-SBM diet fed to birds, M_{III} is the mineral intake (mg) from SBM in the C-SBM diet fed to birds, SMA_{II} is the SMA of corn, and SMA_{III} is the SMA of SBM.

Data from AMA and SMA values were carried out with one-way ANOVA by using the GLM procedures of SAS (release 9.4, SAS Institute Inc., Cary, NC). Data from predicted and determined SMA values in the C-SBM diet were carried out

with Student's *t* test by using TTEST procedure of SAS. Cage was the experimental unit, and $P < 0.05$ was considered to be statistically significant.

RESULTS

All the broilers were healthy throughout the 2 experiments and readily consumed their diets. The values of EML, AMA, and SMA for broiler chickens in Exp. 1 and 2 are presented in Tables 5 and 6, respectively. The determined values of AMA for corn were negative for all minerals except Zn. Apparent P availability values were all negative for all feedstuffs in both experiments. The AMA values were greater ($P < 0.03$) in SBM and C-SBM diet

Table 3. Composition and nutrient contents of mineral-free, corn, SBM, and C-SBM diets¹ in Exp. 1 (as-fed basis)

Item	Mineral-free diet	Corn diet	SBM ² diet	C-SBM ² diet
Ingredient, %				
Corn	—	85.05	—	72.00
SBM	—	—	40.22	27.75
Gelatin ³	19.00	12.26	—	—
Soybean oil	4.00	—	2.80	—
Dextrose	71.16	—	55.96	—
Cellulose ⁴	2.00	0.64	0.67	—
L-Lys HCl	0.28	0.32	—	—
DL-Met	0.57	0.30	0.20	0.10
L-Thr	0.43	0.30	—	—
L-Trp	0.17	0.12	—	—
L-Ile	0.52	0.36	—	—
L-Leu	0.62	—	—	—
L-Val	0.40	0.20	—	—
L-Phe	0.70	0.30	—	—
Vitamin premix ⁵	0.15	0.15	0.15	0.15
Nutrient level, %				
ME, MJ/kg	12.45	12.67	12.45	12.64
CP ⁶	19.64	20.04	19.84	19.44
Lys	1.01	1.00	1.21	1.04
Met+Cys	0.73	0.72	0.72	0.72
Ca ⁶ , mg/kg	19.30	147	805	650
Cu ⁶ , mg/kg	0.29	1.34	5.01	5.09
Fe ⁶ , mg/kg	9.12	19.88	63.71	58.95
Mn ⁶ , mg/kg	0.03	4.23	15.43	15.78
Zn ⁶ , mg/kg	0	11.78	16.39	22.73
Total P ⁶	0	0.19	0.30	0.40

¹Dextrose and cellulose were analytical grade, other ingredients except feedstuffs for test were food grade.

²SBM = soybean meal; C-SBM = corn-soybean meal.

³Obtained from Gelita Gelatine USA Inc., Sioux City, IA.

⁴Obtained from Yuanju Biotech Inc., Shanghai City, China.

⁵Provided per kilogram of diet: vitamin A (all-trans retinolacetate) 10,000 IU; cholecalciferol 3,000 IU; vitamin E (all-rac- α -tocopherol acetate) 16 IU; vitamin K (as menadione sodium bisulfate) 2.0 mg; thiamin (thiamin mononitrate) 2.0 mg; riboflavin 6.4 mg; vitamin B₆ 2.0 mg; vitamin B₁₂ 0.01 mg; calcium pantothenate 10 mg; niacin 26.0 mg; folic acid 1.0 mg; biotin 0.10 mg; choline (choline chloride) 500 mg.

⁶Determined by analysis and each value based on triplicate determinations.

than in corn. The standardized availability values of Ca, P, and Mn were greater ($P < 0.006$) in SBM than in corn and C-SBM diet. The standardized availability values of Fe were greater ($P < 0.0009$) in corn than in SBM and C-SBM diet. However, there were no differences ($P > 0.10$) in standardized availability values of Cu and Zn among corn, SBM, and C-SBM diets. The determined values of SMA for the C-SBM diet were close ($P > 0.09$) to the predicted summations of SMA from corn and SBM in Exp. 1 (Table 7). In Exp. 2, the apparent availability values of Ca and Cu were greater ($P < 0.03$) in cottonseed meal than in wheat bran, corn DDGS, and corn gluten meal. The apparent availability values of P were greater ($P < 0.0002$) in wheat bran, cottonseed meal, and corn DDGS than in wheat, rapeseed meal, and corn gluten meal. The apparent

availability values of Fe, Mn, and Zn were greater ($P < 0.006$) in corn DDGS than in wheat bran, rapeseed meal, cottonseed meal, and corn gluten meal. The standardized availability values of Ca, P, Fe, and Zn were greater ($P < 0.03$) in wheat than in rapeseed meal. The standardized availability values of Cu were greater ($P < 0.0001$) in rapeseed meal, corn DDGS, and corn gluten meal than in wheat, wheat bran, and cottonseed meal. The standardized availability values of Mn were greater ($P < 0.0001$) in corn DDGS than in wheat, wheat bran, cottonseed meal, rapeseed meal, and corn gluten meal.

DISCUSSION

The hypothesis that dietary SMA could be estimated by the summations of the SMA of dietary

Table 4. Composition and nutrient contents of wheat, wheat bran, cottonseed meal, rapeseed meal, corn gluten meal, and corn DDGS diets¹ in Exp. 2 (as-fed basis)

Item	Wheat diet	Wheat bran diet	Cottonseed meal diet	Rapeseed meal diet	Corn gluten meal diet	Corn DDGS ² diet
Ingredient, %						
Wheat	87.87	—	—	—	—	—
Wheat bran	—	54.55	—	—	—	—
Cottonseed meal	—	—	40.83	—	—	—
Rapeseed meal	—	—	—	48.47	—	—
Corn gluten meal	—	—	—	—	31.98	—
Corn DDGS	—	—	—	—	—	71.75
Gelatin ³	5.9	8.54	—	—	—	—
Soybean oil	1.0	4.0	4.0	4.0	—	4.0
Dextrose	2.68	30.47	53.94	46.53	64.97	23.4
Cellulose ⁴	0.33	—	—	—	1.33	—
L-Lys HCl	0.54	0.30	0.37	0.48	0.85	0.48
DL-Met	0.24	0.35	0.24	—	—	—
Vitamin premix ⁵	0.15	0.15	0.15	0.15	0.15	0.15
Nutrient level, %						
ME, MJ/kg	12.46	9.28	11.82	10.99	12.94	12.22
CP ⁶	20.24	19.66	19.28	19.78	19.97	19.69
Lys	1.0	1.0	1.0	1.0	1.0	1.0
Met+Cys	0.72	0.72	0.73	0.73	0.74	0.81
Ca ⁶ , mg/kg	431	569	956	3857	124	209
Cu ⁶ , mg/kg	4.21	7.24	5.47	3.96	3.93	2.43
Fe ⁶ , mg/kg	50.80	78.90	56.30	188	150	122
Mn ⁶ , mg/kg	25.74	62.42	6.97	30.00	4.40	10.49
Zn ⁶ , mg/kg	25.12	38.87	21.06	37.38	10.17	27.92
Total P ⁶	0.26	0.50	0.40	0.46	0.22	0.52

¹Dextrose and cellulose were analytical grade, other ingredients except feedstuffs for test were food grade.

²DDGS =distillers dried grains with solubles.

³Obtained from Gelita Gelatine USA Inc., Sioux City, IA.

⁴Obtained from Yuanju Biotech Inc., Shanghai City, China.

⁵Provided per kilogram of diet: vitamin A (all-trans retinolacetate) 10,000 IU; cholecalciferol 3,000 IU; vitamin E (all-rac- α -tocopherol acetate) 16 IU; vitamin K (as menadione sodium bisulfate) 2.0 mg; thiamin (thiamin mononitrate) 2.0 mg; riboflavin 6.4 mg; vitamin B₆ 2.0 mg; vitamin B₁₂ 0.01 mg; calcium pantothenate 10 mg; niacin 26.0 mg; folic acid 1.0 mg; biotin 0.10 mg; choline (choline chloride) 500 mg.

⁶Determined by analysis and each value based on triplicate determinations.

components in a formulated ration was supported by the results of this study. There was no significant difference between the SMA predicted by the SMA of each dietary component and the SMA measured from the mixed C-SBM diet. A previous study from our laboratory estimated the SPA values of corn, SBM, and C-SBM diet by using the P-free diet to determine endogenous P losses and found that the determined SPA value of the C-SBM diet was close to its predicted value (Liu et al., 2012b). Furthermore, the present study estimated the SMA values of corn, SBM, and C-SBM diet by using the mineral-free diet to determine EML and found that the SMA values predicted by the SMA of each dietary component were also close to the SMA values estimated from the mixed C-SBM diet. Moreover, the standardized availability values of Ca, P, Cu, Mn, and Zn for SBM in the present study were in agreement with the results reported by Liu et al.

(2012a) by using the nitrogen- and mineral-free diet. These above results indicated that the mineral-free diet could be used for the estimation of the EML and SMA values of feedstuffs for broilers and the SMA values for feed ingredients are additive in formulated C-SBM diet for broilers.

Total EML includes diet-dependent EML and basal EML. Dietary factors, such as fiber and antinutritional factors, could induce specific EML and thereby this part of endogenous losses is diet-dependent EML. The basal EML was measured by using the mineral-free diet in the present experiments and not affected by dietary factors (Petersen and Stein, 2006; Almeida and Stein, 2010). Liu et al. (2012a) found that the endogenous loss values of Ca, P, Cu, Mn, and Zn were 24.3, 155, 0.083, 0.118, and 0.159 mg/52 h per bird by using the nitrogen- and mineral-free diet (composed of 96.16% dextrose, 2.84% soybean oil, and

Table 5. The values of EML, AMA, and SMA in corn, SBM, and C-SBM diet for broilers in Exp. 1

Item		Corn	SBM	C-SBM diet	P value
Feed intake ¹ , g		58.9 (1.8)	51.3 (1.4)	52.9 (1.5)	
Excreta, g		11.8 (0.4)	11.0 (0.3)	15.2 (0.4)	
Ca	EML ² , mg per chicken	13.7 (0.4)	13.7 (0.4)	13.7 (0.4)	
	AMA, %	-139 (5) ^c	21.0 (1.7) ^a	8.6 (2.7) ^b	<0.0001
	SMA, %	20.8 (1.0) ^c	54.3 (1.0) ^a	48.6 (1.6) ^b	<0.0001
P	EML ² , mg per chicken	113 (7)	113 (7)	113 (7)	
	AMA, %	-62.2 (3.5) ^c	-21.9 (2.8) ^b	-9.6 (1.6) ^a	<0.0001
	SMA, %	39.6 (1.3) ^c	51.1 (1.5) ^a	44.9 (0.9) ^b	<0.0001
Cu	EML ² , µg per chicken	63.6 (13.7)	63.6 (13.7)	63.6 (13.7)	
	AMA, %	-43.2 (3.8) ^b	14.9 (2.0) ^a	18.0 (0.9) ^a	<0.0001
	SMA, %	37.8 (1.4)	39.7 (2.0)	41.7 (1.1)	0.21
Fe	EML ² , µg per chicken	593 (148)	593 (148)	593 (148)	
	AMA, %	-3.5 (3.0) ^c	9.4 (0.6) ^b	18.8 (1.9) ^a	<0.0001
	SMA, %	47.4 (2.0) ^a	27.6 (0.5) ^c	35.3 (1.7) ^b	<0.0001
Mn	EML ² , µg per chicken	94.2 (19.7)	94.2 (19.7)	94.2 (19.7)	
	AMA, %	-27.3 (1.5) ^b	14.2 (1.2) ^a	6.8 (0.9) ^a	<0.0001
	SMA, %	10.7 (0.8) ^c	26.1 (1.0) ^a	22.7 (0.7) ^b	<0.0001
Zn	EML ² , µg per chicken	132 (18)	132 (18)	132 (18)	
	AMA, %	26.1 (2.2) ^b	35.3 (3.0) ^a	35.2 (1.9) ^a	0.01
	SMA, %	45.2 (1.7)	51.0 (2.9)	46.2 (1.7)	0.10

^{a,b,c}Means with different superscripts within the same row differ ($P < 0.03$).

Each value represents the mean (SE) of 8 replicates ($n = 8$); the excreta samples were collected for 48 h after feed withdrawal (totally 52 h); AMA = apparent mineral availability; EML = endogenous mineral loss; SMA = standardized mineral availability; SBM = soybean meal; C-SBM = corn-soybean meal.

¹The average DM intake of birds in the mineral-free diet group was 35.9(1.6) g per chicken.

²Basal EML estimated by gelatin–dextrose mineral-free diet.

Table 6. The values of AMA and SMA in feedstuffs for broilers in Exp. 2

Item		Wheat	Wheat bran	Cottonseed meal	Rapeseed meal	Corn DDGS	Corn gluten meal	P value
Feed intake, g		63.1 (1.6)	46.7 (1.3)	58.7 (1.4)	39.5 (1.3)	42.6 (1.8)	43.0 (1.8)	
Excreta, g		18.2 (0.6)	16.5 (0.4)	15.5 (0.3)	16.6 (0.4)	17.2 (0.8)	8.2 (0.7)	
Ca	AMA, %	3.7 (1.6) ^b	-21.6 (5.1) ^c	16.6 (2.1) ^{ab}	29.9 (1.8) ^a	-127 (6) ^d	-235 (10) ^e	<0.0001
	SMA, %	55.8 (1.0) ^a	33.5 (1.6) ^c	41.1 (2.2) ^b	38.9 (1.5) ^b	28.6 (1.6) ^d	24.7 (1.6) ^d	<0.0001
P	AMA, %	-29.5 (4.6) ^b	-7.7 (1.4) ^a	-3.1 (3.0) ^a	-27.7 (2.5) ^b	-8.0 (4.6) ^a	-72.8 (6.0) ^c	<0.0001
	SMA, %	42.7 (1.6) ^{ab}	41.3 (1.7) ^b	45.9 (2.0) ^{ab}	35.6 (1.4) ^c	43.7 (2.4) ^{ab}	46.6 (2.0) ^a	0.0003
Cu	AMA, %	7.6 (1.2) ^{bc}	5.6 (1.0) ^{bc}	15.2 (1.7) ^a	4.2 (1.6) ^c	-16.3 (3.0) ^d	9.0 (0.8) ^b	<0.0001
	SMA, %	32.6 (1.2) ^b	24.5 (0.7) ^c	35.0 (1.8) ^b	45.1 (2.0) ^a	45.7 (1.0) ^a	43.7 (1.1) ^a	<0.0001
Fe	AMA, %	24.2 (1.6) ^b	17.1 (1.7) ^c	3.6 (0.9) ^d	15.3 (1.5) ^c	30.3 (1.1) ^a	14.9 (1.3) ^c	<0.0001
	SMA, %	43.5 (1.2) ^a	33.3 (1.6) ^b	21.6 (1.0) ^c	23.3 (1.5) ^c	41.8 (0.7) ^a	24.2 (1.1) ^c	<0.0001
Mn	AMA, %	14.9 (1.0) ^a	7.3 (0.7) ^b	-13.8 (0.9) ^c	4.0 (0.9) ^b	13.2 (1.7) ^a	-31.4 (2.8) ^d	<0.0001
	SMA, %	20.9 (0.9) ^b	10.5 (0.7) ^{cd}	9.3 (0.9) ^d	12.0 (0.9) ^c	34.5 (1.1) ^a	18.8 (1.2) ^b	<0.0001
Zn	AMA, %	45.8 (2.3) ^a	22.6 (0.8) ^c	27.4 (1.5) ^b	29.1 (2.0) ^b	41.6 (2.2) ^a	21.4 (1.7) ^c	<0.0001
	SMA, %	52.9 (2.0) ^a	29.0 (0.7) ^d	44.0 (1.6) ^b	38.0 (2.2) ^c	52.8 (1.8) ^a	51.7 (1.8) ^a	<0.0001

^{a,b,c,d,e}Means with different superscripts within the same row differ ($P < 0.05$).

Each value represents the mean (SE) of 8 replicates ($n = 8$); The excreta samples were collected for 48 h after feed withdrawal (totally 52 h); AMA = apparent mineral availability; SMA = standardized mineral availability; SBM = soybean meal; C-SBM = corn-soybean meal; DDGS = distillers dried grains with solubles.

1.0% cellulose), respectively. These above data were greater than those in the present study. However, the endogenous loss of P for SBM in Exp. 1 (113 mg) was in agreement with the results (123 and 109 mg) from Liu et al. (2012b) and Liu et al. (2013), who reported that chickens were fed the P-free diet for

4 h with a total of 52 h of excreta collection time. This indicated that the CP and other nutrients in the mineral-free diet used in the current study might reduce the EML of chickens. Cowieson et al. (2004) reported that the endogenous loss values of Ca, P, Cu, Fe, Mn, and Zn for 42-d-old female broilers

Table 7. Comparison of the determined and the predicted values of SMA in the C-SBM diet in Exp. 1

Item	Ca	P	Cu	Fe	Mn	Zn
Determined, %	48.6 (1.6)	44.9 (0.9)	41.7 (1.1)	35.3 (1.7)	22.7 (0.7)	46.2 (1.7)
Predicted, ¹ %	48.1 (0.6)	46.0 (0.7)	39.1 (1.0)	33.2 (0.7)	22.2 (0.7)	48.4 (1.3)
P value	0.73	0.42	0.10	0.42	0.63	0.31

Each value represents the mean (SE) of 8 replicates ($n = 8$); The excreta samples were collected for 48 h after feed withdrawal (totally 52 h); C-SBM = corn-soybean meal; SMA = standardized mineral availability.

¹Predicted values by determined SMA values of single feed ingredients.

were 72.5, 230, 0.30, 5.2, 0.5, and 12.0 mg/48 h per bird, respectively, by using a precision-fed assay. [Al-Masri \(1995\)](#) reported that endogenous P losses for 14- to 29-d-old male broilers ranged between 135 and 30 mg/d per bird with 4 different Ca:P ratios ranging between 1.0:1 and 2.5:1 by using the isotope dilution method. These diverse estimates might be explained by differences in age or genetic background of broilers or more likely by different approaches used to determine EML. Moreover, there is the likelihood that a significant portion of the EML may be coming from resident bacteria in the hind gut (large intestine and cecum), especially given the very low feed intake after fasting. This, to some extent, may have some effects not only on the EML but also on the SMA values. This needs to be further studied in the future experiments.

The AMA values have been shown to be very variable, and thus not reliable in diet formulation for pigs ([Fan and Sauer, 2002](#)) and ducks ([Chen, 2010](#)). If values of AMA are corrected for EML, more accurate estimate values can be generated ([Dilger and Adeola, 2006](#)). [Ravindran et al. \(2006\)](#) reported that the apparent ileal digestibility coefficients of Ca, P, Cu, Mn, and Zn in a low phytate diet for 28-d-old broiler chickens were 34.9%, 51.6%, 5.3%, -19.8%, and 25.8%, respectively. [Liu et al. \(2012a\)](#) reported that the availability values of Ca, P, Cu, Mn, and Zn were, respectively, 10.9%, -1.8%, -66.1%, -4.7%, and 11.7% for 36-d-old broiler chickens. The negative values are reflective of significant endogenous secretions into the gut and do not account for the confounding effects of EML. In the present study, the negative apparent P availability values were observed in the 2 experiments and the negative AMA values of corn except Zn were observed due to the EML. However, [Dilger and Adeola \(2006\)](#) used chromic oxide as an indigestible marker and fed chickens for 6 d and found that the average apparent P availability value in conventional SBM for 3-wk-old broilers was 49.0%. The above apparent difference might be due to the different experimental procedures used in different studies.

The SMA values of 6 commonly used feed ingredients derived from plants were evaluated in Exp. 2 following the same procedure in Exp. 1, so as to provide mineral availability data for diet formulation of broilers. The determined SMA values for wheat were lower than those reported by [Aw-Yong et al. \(1983\)](#) for all minerals except Zn. [Nwokolo and Bragg \(1980\)](#) reported that the standardized availability values in 7 rapeseed meals were 59.7% to 75.6% for Ca, 65.0% to 81.0% for P, 62.2% to 85.0% for Cu, 45.5% to 60.0% for Mn, and 23.0% to 57.6% for Zn, which were also greater than those in the current study. [Mutucumarana et al. \(2014\)](#) found that the true ileal P digestibility coefficients in wheat and corn DDGS were, respectively, 46.4% (close to the current study, 42.7%) and 72.7% (more than the current study, 43.7%) by using the regression method. The above differences might be related to the use of broiler chickens, feedstuffs from different batches, and sources or the different approaches used to determine mineral availabilities. Further studies are needed to determine the SMA values of other feed ingredients as well as standardized available mineral requirements of broilers so that diets can be formulated based on SMA values to more accurately meet their mineral requirements.

In summary, the results from the present study indicated that the mineral-free diet could be used for estimating the EML and SMA values of feedstuffs for broilers, and the estimates of SMA in feedstuffs for C-SBM diet formulation were additive. The information will allow more precision in diet formulation and result in less overfeeding and reduced mineral pollution.

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