Inorganic feed phosphate type determines mineral digestibility, broiler performance, and bone mineralization

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Primary Audience: Feed Manufacturers, Nutritionists, Researchers

SUMMARY

An experiment was conducted to evaluate early broiler performance, tibia mineralization, and mineral digestibility of broilers fed with diets that differed in inorganic feed phosphates (IFP) but were formulated to be similar in dietary nonphytate phosphorus (nPP) content and calcium-to-phosphorus ratio (Ca:P), total mineral content, or calculated precedal digestible phosphorus (CPDP). Dicalcium phosphate (DCP), monodicalcium phosphate (MDP), monocalcium phosphate (MCP), and defluorinated phosphate were used in formulations with similar nPP content and Ca:P. In addition to these 4 treatments, a mixture of MCP, monosodium phosphate, and DCP was used to assimilate total mineral content of MDP (DMM). Moreover, a mixture of MCP and silicon dioxide was used to produce a similar CPDP content as the DCP diet (MCP + SiO_2). The MCP + SiO_2 diet had the lowest IFP inclusion. The study used a randomized complete block design with 10 replications of 10 male broilers fed with each of the 6 dietary treatments for 21 D. Linear contrasts were used to compare treatments of interest. Birds fed with MCP demonstrated increased live weight gain, tibia ash (mg/chick), and mineral digestibility compared with birds fed with DCP when diets were formulated to similar nPP content and Ca:P (P < 0.05). When diets were formulated to similar total mineral content, MDP-fed birds showed an increase in tibia ash (%) and mineral digestibility compared with birds fed with DMM (P < 0.05). Birds fed with MCP + SiO₂ had increased mineral digestibility compared with birds fed with DCP when diets were formulated to similar CPDP content (P < 0.05). Dietary inclusion and efficacy data suggest that diets formulated using CPDP may optimize feed P incorporation and utilization.

Key words: inorganic feed phosphate, monocalcium phosphate, dicalcium phosphate, monodicalcium phosphate, phosphorus digestibility

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DESCRIPTION OF PROBLEM

P and Ca are essential minerals for proper growth performance and bone mineralization in

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poultry. In addition, Ca is significant for eggshell formation, blood clotting, enzyme activation, and muscle contraction, while P plays an important role in cellular and membrane function, metabolism of fats and carbohydrates, and acid-base balance (Leeson and

Summers, 2001; Applegate and Angel, 2008; Veum, 2010; Kleyn, 2013). Although Ca and P are present in plant origin feedstuffs (Nelson et al., 1968; Huyghebaert et al., 1980; Viljoen, 2001; Adedokun and Adeola, 2013), inorganic feed phosphates (IFP) are still needed to cover mineral requirements in diets for poultry (Gordon and Roland, 1999; Applegate and Angel, 2008; Shastak et al., 2012). Some of the IFP used commercially include dicalcium phosphate (DCP), monocalcium phosphate (MCP), defluorinated phosphate (DFP), and monosodium phosphate.

The production of these IFP varies between products. When phosphoric acid reacts with lime and sulfuric acid, MCP and hydrated DCP are produced (Kleyn, 2013). Defluorinated phosphate is produced by the reaction of phosphate rock concentrate and phosphoric acid in the presence of sodium carbonate. With further processing, F is removed; the final product is tricalcium phosphate containing low F levels and a relatively high Na concentration (Lima et al., 1999). Monocalcium phosphate, DCP, and MDP contain various amounts of MCP and DCP. For a product to be considered DCP, it must contain more than or equal to 51% DCP. When an IFP contains less than 80% MCP but more than 51% MCP, it is labeled MDP. For an IFP to be labeled MCP, it must contain more than 80% MCP (Yara International, Oslo, Norway).

Phosphate rock is a nonrenewable resource used to produce IFP. Cordell et al. (2009) estimated that global reserves may last for 50 to 100 yr more. Inorganic feed phosphates vary in mineral content and P and Ca availability to the animal. This variability is due to many factors such as differences in the chemical structure, particle size, pH, crystallinity, the production process, source of ingredients used, and concentration of contaminating elements (Huyghebaert et al., 1980; Lima et al., 1999; Kleyn, 2013; WPSA, 2013). The sustainability of the poultry industry relies on the responsible use of IFP in diets. The exact knowledge about P availability in each type of IFP is needed to optimize nutrient use efficiency and avoid extra costs and excessive excretion (Viljoen, 2001; Applegate and Angel, 2008).

Historically, poultry diets were formulated based on the calcium-to-phosphorus ratio (Ca:P), which is defined as total Ca and total P in the diet. Today, in the United States, diets are typically formulated based on total Ca and available P (nonphytate P [nPP]) for better utilization of P by the bird. P requirements for broilers in the NRC (1994) are expressed in terms of total P and nPP. However, this does not take into account that phytate P may be partially available and that nPP may not be completely available to the bird (Leske and Coon, 2002).

Different assays have been classically used to determine the P and Ca availability of an ingredient (Shastak et al., 2012; Shastak and Rodehutscord, 2015). Bone ash, blood inorganic P concentration, and body weight gain assays provide relative values of P and Ca availability, which makes the comparison of data from different studies impossible. Therefore, these methods have limited value for feed formulation (Coon et al., 2002). Quantitative values of P and Ca digestibility of IFP can be obtained using retention measurements and prececal mineral digestibility. These data make it possible to formulate diets that closely match P and Ca requirements while minimizing excess and Ca supplementation (Shastak and Rodehutscord, 2015). The experimental effort to obtain quantitative measurements of P retention is high, and results can be influenced by excess dietary P intake. The Working Group 2 Nutrition of the European Federation of Branches of the World's Poultry Science Association recommends using prececal mineral digestibility to determine available P (WPSA, 2013). The group has developed a standard protocol for establishing a unique commonly accepted system to assess this parameter.

The objective of this study was to understand the impact of using different systems to evaluate the potential of an IFP. For this purpose, diets that differed in IFP source but were similar in either 1) dietary nPP content and Ca:P, 2) total mineral content or, 3) calculated preceal digestible P (CPDP) content (according to Centraal Veevoeder Bureau [CVB], 2016) were formulated, and their impact on early broiler performance, tibia mineralization, and mineral digestibility was assessed.

Table 1. Diet formulations of diets provided to broilers from day 1 to 21.

Ingredients	DCP	MDP	MCP	DFP	DMM	$MCP + SiO_2$
Inclusion (%)						
Corn	59.22	59.22	59.22	59.46	59.22	59.18
Soybean meal (48%)	29.17	29.17	29.17	29.17	29.17	29.18
Corn DDGS	6.00	6.00	6.00	6.00	6.00	6.00
Soybean oil	2.27	2.27	2.27	2.19	2.27	2.28
DL-methionine	0.34	0.34	0.34	0.34	0.34	0.34
Vitamin mineral premix ¹	0.25	0.25	0.25	0.25	0.25	0.25
Sodium bicarbonate	0.10	0.10	0.10	0.010	0.10	0.10
Lysine	0.36	0.36	0.36	0.36	0.36	0.36
Threonine	0.21	0.21	0.21	0.21	0.21	0.21
Feed phosphate	0.50	0.49	0.41	0.51	0.45	0.30 MCP
						0.15 SiO_2
Limestone	1.34	1.39	1.42	1.22	1.42	1.40
Salt	0.26	0.20	0.26	0.19	0.21	0.26

Abbreviations: DCP, dicalcium phosphate; DDGS, distillers dried grains with solubles; DFP, defluorinated phosphate; DMM, 1/5 MCP, 1/5 monosodium phosphate, 3/5 DCP; MCP, monocalcium phosphate; MCP + SiO₂, MCP + SiO₂, (67:33); MDP, monodicalcium phosphate.

¹Vitamin mineral premix (NB3000; Nutra Blend, Neosho, MO) supplied the following per kg of diet: manganese, 0.02 mg; zinc, 0.02 mg; iron, 0.01 mg; copper, 0.0025 mg; iodine, 0.0003 mg; selenium, 0.00003 mg; folic acid, 0.69 mg; choline, 386 mg; riboflavin, 6.61 mg; biotin, 0.03 mg; vitamin B6, 1.38 mg; niacin, 27.56 mg; pantothenic acid, 6.61 mg; thiamine, 2.20 mg; menadione, 0.83 mg; vitamin B12, 0.01 mg; vitamin E, 16.53 IU; vitamin D3, 2,133 ICU; vitamin A, 7,716 IU.

Table 2. Nutrient specifications 1,2 and pH change of diets provided to broilers from day 1 to 21.

Treatments	DCP	MDP	MCP	DFP	DMM	$MCP + SiO_2$
Calculated nutrients (%)						
ME (kcal/kg)	3,000	3,000	3,000	3,000	3,000	3,000
Crude protein	20.00	20.00	20.00	20.00	20.00	20.00
Digestible lysine	1.20	1.20	1.20	1.20	1.20	1.20
Digestible methionine	0.63	0.63	0.63	0.63	0.63	0.63
Digestible methionine + cysteine	0.90	0.90	0.90	0.90	0.90	0.90
Digestible threonine	0.84	0.84	0.84	0.84	0.84	0.84
Digestible tryptophan	0.20	0.20	0.20	0.20	0.20	0.20
Calcium	0.70	0.69	0.68	0.70	0.70	0.66
Phosphorus	0.49	0.49	0.49	0.49	0.49	0.46
Ca:P	1.44:1	1.42:1	1.41:1	1.44:1	1.43:1	1.43:1
nPP	0.22	0.22	0.22	0.22	0.22	0.20
Sodium	0.17	0.17	0.17	0.17	0.17	0.17
$CPDP^{3}$ (g)	0.509	0.735	0.791	0.413	0.748	0.579
Analyzed nutrients (%)						
Crude protein ⁴	18.7	18.6	18.5	18.2	17.9	17.5
Crude fat ⁵	4.96	4.93	4.86	4.60	5.14	5.11
Calcium ⁶	0.63	0.74	0.74	0.75	0.74	0.69
Phosphorus ⁷	0.47	0.51	0.50	0.47	0.49	0.47
nPP ⁸	0.24	0.28	0.27	0.25	0.24	0.23
Phytase analysis (FTU/kg) ⁹	1,300	930	650	970	920	530
pH change						
0-h incubation	7.3	7.2	7.2	7.3	7.2	7.2
24-h incubation	5.9	5.9	6.1	6.0	5.9	6.2
48-h incubation	5.4	5.7	6.2	5.2	5.5	5.7

All diets were top dressed with 1,000 FTU/kg of phytase.

Abbreviations: AOAC, Association of Official Analytical Chemists; CVB, Centraal Veevoeder Bureau; DCP, dicalcium phosphate; DFP, defluorinated phosphate; DMM, 1/5 MCP, 1/5 monosodium phosphate, 3/5 DCP; IFP, inorganic feed phosphates; MCP, monocalcium phosphate; MCP + SiO₂, MCP + SiO₂ (67:33); MDP, monodicalcium phosphate; nPP, nonphytate phosphorus.

MATERIALS AND METHODS

Diet Formulation and Dietary Treatments

Diets were corn and soybean meal based containing 6% distillers dried grains with solubles and were formulated to contain similar calculated nutrient values that were based on commercial (Donohue, 2013) and research guidelines (Tillman and Dozier, 2013) (Tables 1 and 2). However, nPP and Ca levels were lower than the requirements even with phytase inclusion to better demonstrate potential treatment differences on bird performance and mineral digestibility. A total of 6 dietary treatments were

¹Metabolizable energy and available phosphorus were based on Agristat values, as suggested by Donohue (2013).

²Digestible amino acids were based on values suggested by Tillman and Dozier (2013).

 $^{^3}$ CPDP: calculated prececal digestible P; values were calculated using values from the CVB (2016) data and IFP calculated values. Considering the DCP diet, DCP is included into the diet at 0.5% concentration, which is 5 g of IFP. The P content of DCP is 18.5%; therefore, 0.93 g of P is added to the diet with the addition of DCP. The standard digestibility coefficient obtained from the CVB (2016) data for DCP is 55%, meaning that the digestible P supplied through DCP corresponds to 0.5 g (0.93 g \times 55%).

⁴AOAC Method 988.05, 920.87, 991.20; Kjeldahl.

⁵AOAC Method 920.39, 934.0; Ether Extraction.

⁶AOAC Method 968.08, 927.02, 935.13, 985.35; Atomic Absorption Spectrophotometry.

⁷AOAC Method 965.17; Spectrophotometry.

⁸Nonphytate phosphorus = total phosphorus (AOAC Method 965.17) – $[0.282 \text{ X phytic acid (AOAC Method 986.11)}] \times 100.$

⁹AOAC method 2000.12; Phytase.

Table 3. Descriptive characteristics of the inorganic feed phosphates and mixtures used in experimental diets.

Treatments	DCP	MDP	MCP	DFP	DMM	MCP + SiO ₂
Chemical formula	CaHPO ₄	Ca(H ₂ PO ₄) ₂ H ₂ O-CaHPO ₄ 2H ₂ O	Ca(H ₂ PO ₄) ₂ H ₂ O	Ca ₃ (PO ₄) ₂	MCP: CaHPO ₄ DCP: Ca(H ₂ PO ₄) ₂ H ₂ O MSP: H ₂ NaO ₄ P	Ca(H ₂ PO ₄) ₂ H ₂ O SiO ₂
Calculated values (%)						
Phosphorus	18.5	19.0	22.7	18.0	20.4	15.2
Calcium	23.0	16.5	16.0	30.0	17.1	10.7
Sodium	_	4.5	_	5.3	4.0	_
Analyzed values (%)						
Phosphorus ¹	19.2	19.6	22.5	19.0	20.6	13.7
Calcium ²	20.3	17.8	18.8	34.7	15.2	11.6
Sodium ³	_	2.13	_	2.55	1.78	_

Abbreviations: AOAC, Association of Official Analytical Chemists; DCP, dicalcium phosphate; DFP, defluorinated phosphate; DMM, 1/5 MCP, 1/5 monosodium phosphate, 3/5 DCP; MCP, monocalcium phosphate; MCP + SiO₂, MCP + SiO₂, (67:33); MDP, monodicalcium phosphate; MSP, monosodium phosphate.

¹AOAC Method 965.17; Spectrophotometry.

²AOAC Method 968.08, 927.02, 935.13, 985.35; Atomic Absorption Spectrophotometry.

³AOAC Method 968.08, 935.13, 985.35; Atomic Absorption Spectrophotometry.

used in this experiment. Each treatment contained a different IFP or IFP mixture:

- DCP; CaHPO₄ (Feed Products & Service Company, St. Louis, MO),
- 2. MDP; Ca(H₂PO₄)₂ H₂O-CaHPO₄ 2H₂O (The Mosaic Company, Plymouth, MN),
- 3. MCP; Ca(H₂PO₄)₂ H₂O (Yara International),
- DFP; Ca₃(PO₄)₂ (EuroChem, Moscow, Russia),
- A mixture of DCP (60%), MCP (20%), and monosodium phosphate (H₂NaO₄P; 20%) formulated to assimilate total mineral content of MDP (DMM),
- A mixture of MCP (67%) and silicon dioxide (SiO₂) (33%) formulated to contain similar CPDP content as the DCP (MCP + SiO₂).

Five of the diets (DCP, MDP, MCP, DFP, and DMM) were formulated to contain 0.22% nPP and 0.70% Ca, with a 1.4:1 Ca:P. The MCP + SiO₂ diet had the lowest IFP content and lower nPP (0.20%) and Ca (0.66%) values than the other 5 treatments (Tables 1 and 2). The MCP + SiO₂ diet was formulated this way to be similar in CPDP content as determined for the DCP diet: DCP is included into the diet at 0.5% concentration, which is 5 g of IFP in a 1-kg batch of feed. The P content of DCP is 18.5%; therefore, 0.93 g of P is added to the diet with the addition of DCP. The standard digestibility coefficient obtained from the CVB (2016) data for DCP is 55%, meaning that the digestible P supplied through DCP corresponds to 0.5 g/kg $(0.93 \text{ g} \times 55\%)$ (Table 2). In addition, the pH of the diets was measured following the procedures described by Cox et al. (2013). Five grams of each treatment were added to 45 mL of buffered peptone water, stirred, and incubated at 37°C for 24 and 48 h. At 0, 24, and 48 h, pH was measured using a pH meter. The samples were analyzed in duplicate, and the averages of these values can be found in Table 2.

Titanium (**Ti**) dioxide was used as an indigestible marker for mineral digestibility determination, and all diets were top dressed with 1,000 FTU/kg of phytase (Quantum Blue; AB Vista, Marlborough, Wiltshire, United Kingdom). Silicon dioxide was used as a non-nutritive additive and potential pellet die scouring agent in the MCP + SiO₂ diet.

Feed Manufacture and IFP

All feed was manufactured at the West Virginia University pilot feed mill. All diets were mixed for 10 min in dry form and 10 min after soybean oil addition. Multiple samples of each treatment were collected, pooled, and sent to commercial laboratories for nutrient, Ti (NP Analytical Laboratories, St. Louis, MO; AOAC International, 2000; Method 968.08, 984.27, 935.13, 985.35, 975.03, 990.08, and 993.14), and phytase analysis (Eurofins Nutrition Analysis Center, Des Moines, IA) (Table 2). All feed was provided to birds in mash form.

The DCP product used in this study contained more than 85% calcium bis(dihydrogen orthophosphate) (Feed Products & Service Company). The MDP product was composed of 40 to 60% MCP monohydrate and 25 to 40% DCP dihydrate (The Mosaic Company). The MCP product contained more than or equal to 80% but less than 90% calcium bis(dihydrogen orthophosphate) monohydrate (Yara International). The inclusion rates of MCP and DCP in each product are based on manufacturers' statements. Individual IFP were analyzed for P, Ca, and Na (NP Analytical Laboratories) (Table 3).

Live Bird Performance

A total of six hundred 1-day-old male Ross × Ross 308 (Aviagen, Inc., Huntsville, AL) chicks were obtained from a commercial hatchery (Longnecker's Hatchery, Elizabethtown, PA). On day 1, chicks were individually weighed and allotted into 10 weight groups. One bird from each weight group was placed in one of the 60 raised wire cages to create the experimental unit. Each of the 6 dietary treatments was randomly assigned to a pen within a block. A block consisted of 6 adjacent cages, and there were 10 blocks or replications per treatment. Housing, lighting, and temperature conditions were similar to methodologies described by Lamp et al. (2015). Feed and water were provided for ad libitum consumption. On day 21, birds were exposed to 6 h of darkness and then allowed to consume feed for 6 h to ensure that digesta were present in the digestive tract to perform ileal collection and mineral

digestibility measurements. The study period was a total of 21 D, and performance measurements were taken on day 7, 14, and 21. Measured variables associated with performance included day 21 ending bird weight, pen feed intake (FI), mortality corrected feed conversion ratio (FCR), bird live weight gain (LWG), and pen percent mortality. All animals were reared according to protocols approved by the West Virginia University Animal Care and Use Committee.

Bone Mineralization and Mineral Digestibility

On day 21, all birds were euthanized via cervical dislocation. The left tibia was excised and used to determine dry defatted tibia ash (%) and tibia ash (mg/chick). Excised tibiae were placed in a freezer until tibia ash analysis began. Tibiae were placed in a drying oven at 105°C for 48 h. Once dried, tibiae were wrapped in a filter paper and placed in a Soxhlet apparatus and refluxed with petroleum ether for 16 h. After fat extraction, the tibiae were allowed to dry. Tibiae were then removed from the filter paper, weighed, and placed in an ashing oven at 600°C for 18 h. The remaining inorganic matter was weighed, and ash content was determined (Boney and Moritz, 2017).

vitamin D₃, whereas passive transport of Ca takes place throughout the small intestine (Walling, 1977; Bukley and Bronner, 1980; Pansu et al., 1983; Bronner et al., 1986; Auchère et al., 1998; Adedokun and Adeola, 2013; Kleyn, 2013; Proszkowiec-Weglarz and Angel, 2013). Furthermore, Rodehutscord (2012) concluded that P absorption is not complete at or before Meckel's diverticulum; his study reported a greater disappearance of P in the distal ileum. Therefore, in the present study, the ileum (defined as the portion of the small intestine extending from Meckel's diverticulum to the ileal-cecal junction) was excised from each bird (Ravindran et al., 1999). This section was cut into half, and the lower section was used for collection. The lower ileum was gently flushed with distilled water, and the digesta washing was collected. The collected digesta were pooled per pen and lyophilized at -40°C until dry (Evans et al., 2015). Digesta samples were analyzed for Ti, Ca, and contents (NP Analytical Laboratories).

Apparent ileal nutrient (Ca and P) digestibility coefficients were calculated according to Adedokun et al (2015):

AID Ca/AID P coefficient (%) =
$$\left[1 - \left(\frac{Ti_{Diet}}{Ti_{Digesta}}\right) \times \left(\frac{Nutrient_{Digesta}}{Nutrient_{Diet}}\right)\right] \times 100$$

Five birds from each pen were randomly selected to determine the apparent ileal Ca digestibility (AID Ca) coefficient and apparent ileal P digestibility (AID P) coefficient. Cal-

The digestible Ca and P concentrations of the diet were calculated using the following equation:

Digestible P/digestible Ca concentration (%) =
$$\left(\frac{AID\ nutrient(\%)}{100}\right) \times Nutrient_{Diet}(\%)$$

cium is absorbed across the intestinal wall through active (saturable; transcellular) or passive (unsaturable; paracellular) transport. Active transport occurs primarily in the duodenum and upper jejunum and involves where $Ti_{Digesta}$ and Ti_{Diet} are the analyzed concentrations of Ti (%) in the digesta and diets, respectively, and Nutrient_{Digesta} and Nutrient_{Diet} are the analyzed concentrations of Ca or P in the digesta and diets, respectively.

Table 4. Growth	performance of ross \times ross	308 male broilers and contrast	probability values recorde	d on day 7, 14, and 21.

Day	Treatments	DCP	MDP	MCP	DFP	DMM	MCP + SiO ₂	Treatment P-value	Tukey's HSD ⁴	Treatment SEM	Linear contrasts ⁵
7	Bird weight (kg)	0.117	0.121	0.124	0.118	0.119	0.116	0.0689	_	0.0019	a
	Pen FI ¹ (kg)	0.982	1.03	1.07	1.01	0.981	0.989	0.1141	_	0.0248	a
	Bird LWG ² (kg)	0.075	0.080	0.082	0.077	0.077	0.075	0.0635	_	0.0019	a
	FCR ³	1.32	1.29	1.30	1.31	1.27	1.34	0.1234	_	0.0185	_
14	Bird weight (kg)	0.341	0.353	0.356	0.342	0.348	0.335	0.0460	0.0215	0.0051	a
	Pen FI (kg)	4.37	4.49	4.52	4.38	4.40	4.37	0.0944	_	0.0463	a
	Bird LWG (kg)	0.299	0.312	0.314	0.291	0.307	0.293	0.1151	_	0.0072	a
	FCR	1.49	1.45	1.46	1.47	1.47	1.53	0.1497	_	0.0224	_
21	Bird weight (kg)	0.751	0.767	0.779	0.750	0.752	0.740	0.2149	_	0.0115	a
	Pen FI (kg)	10.10	10.35	10.39	10.19	10.15	10.24	0.4318	_	0.1144	_
	Bird LWG (kg)	0.710	0.726	0.738	0.708	0.711	0.699	0.2135	_	0.0115	a
	FCR	1.48	1.45	1.44	1.48	1.48	1.51	0.4397	_	0.0240	a

Abbreviations: DCP, dicalcium phosphate; DFP, defluorinated phosphate; DMM, 1/5 MCP, 1/5 monosodium phosphate, 3/5 DCP; MCP, monocalcium phosphate; MCP + SiO₂, MCP + SiO₂ (67:33); MDP, monodicalcium phosphate.

¹FI: feed intake.

²LWG: live weight gain.

³FCR: feed conversion ratio (feed:gain) was calculated using mortality weight.

⁴HSD: honestly significant difference.

⁵Significant (P < 0.05) linear contrasts were expressed by letters as follows: a = DCP vs. MCP; b = MDP vs. DMM; and c = DCP vs. MCP + SiO₂.

Table 5. Tibia ash and phosphorus and calcium digestibility analysis recorded on day 21.

Treatments	DCP	MDP	MCP	DFP	DMM	MCP + SiO ₂	Treatment P-value	Tukey's HSD ¹	Treatment SEM	Linear contrasts ²
Tibia ash (%)	47.2 ^b	48.0°	47.8 ^{a,b}	47.2 ^b	47.5 ^{a,b}	47.4 ^{a,b}	0.0078	0.6833	0.1624	b
Tibia ash (mg/chick)	675.0^{b}	723.1 ^{a,b}	769.0^{a}	712.8 ^{a,b}	753.6 ^a	$708.3^{a,b}$	0.0026	68.065	16.1727	a
Tibia weight (mg)	1430.7 ^b	1506.5 ^{a,b}	1610.4 ^a	1509.3 ^{a,b}	1585.7 ^a	1495.7 ^{a,b}	0.0078	144.38	34.3050	a
Ashed tibia weight (mg)	675.0^{b}	723.1 ^{a,b}	768.9 ^a	712.8 ^{a,b}	753.6 ^a	$708.3^{a,b}$	0.0026	68.065	16.1727	a
AID Ca coefficient (%)	44.78°	75.02 ^a	62.50 ^b	69.82 ^{a,b}	68.62 ^{a,b}	68.92 ^{a,b}	< 0.0001	10.468	2.4872	a, b, c
AID P coefficient (%)	29.05 ^b	74.65 ^a	68.19 ^a	66.41 ^a	74.21 ^a	74.87 ^a	< 0.0001	9.1215	2.1673	a, c
Digestible Ca concentration (%)	0.280°	0.557 ^a	0.463 ^b	0.523 ^{a,b}	0.506 ^{a,b}	0.476 ^b	< 0.0001	0.0694	0.0165	a, b, c
Digestible P concentration (%)	0.136 ^c	0.378 ^a	0.343 ^{a,b}	0.315 ^b	0.350 ^{a,b}	0.350 ^{a,b}	< 0.0001	0.0436	0.0103	a, b, c

 $[\]overline{a-c}$ Means within a column not sharing a common superscript differ significantly (P < 0.05).

Abbreviations: AID Ca coefficient, apparent ileal Ca digestibility coefficient; AID P coefficient, apparent ileal P digestibility coefficient; DCP, dicalcium phosphate; DFP, defluorinated phosphate; DMM, 1/5 MCP, 1/5 monosodium phosphate, 3/5 DCP; MCP, monocalcium phosphate; MCP + SiO₂, MCP + SiO₂ (67:33); MDP, monodicalcium phosphate.

¹HSD: honestly significant difference.

²Significant (P < 0.05) linear contrasts were expressed by letters as follows: a = DCP vs. MCP; b = MDP vs. DMM; and c = DCP vs. MCP + SiO₂.

Statistical Analysis

A randomized complete block design was used with one pen of 10 broilers as the experimental unit. Data were analyzed using the GLM procedure of SAS (SAS Institute, 2013), considering cage as the experimental unit for all variables studied, and pen location within the room was used as the blocking criterion. Significance was set at P < 0.05, and tendency was set at P < 0.10. Multiple comparisons of means were made using Tukey's honestly significant difference test. Linear contrasts were used to compare diets of interest that were formulated to be either similar in 1) dietary nPP content and Ca:P (DCP vs. MCP), 2) total mineral content (MDP vs. DMM), or 3) CPDP content (DCP vs. $MCP + SiO_2$).

RESULTS AND DISCUSSION

Realized Diet Composition and IFP

Overall Comparison. The analyzed nutrients of the experimental diets are shown in Table 2. Five of the diets (DCP, MDP, MCP, DFP, and DMM) were formulated to 0.22% nPP, 0.70% Ca, 0.49% P, and 20% CP. Analyzed values of the 5 diets were nPP (0.24-0.28%),Ca (0.63-0.75%),(0.47–0.51%), and CP (17.9–18.7%). The $MCP + SiO_2$ diet was formulated to 0.20% nPP, 0.66% Ca, 0.46% P, and 20% CP. Analyzed values of the MCP + SiO₂ diet were 0.23% nPP, 0.69% Ca, 0.47% P, and 17.5% CP. Phytase activity analysis performed on complete diets ranged from 530 to 1,300 FTU/kg (Table 2). The target phytase activity for each diet was 1,000 FTU/kg. All diets showed decrease in pH as incubation time increased. Similar results were observed by Cox et al. (2013); finished poultry feeds in buffered peptone water showed decrease in pH as incubation time increased. The analyzed P and Ca values of the IFP products were similar to the calculated values (Table 3). The MCP product contained the highest P value compared with the other products. The DFP product had the highest Ca and Na values among all products. The MCP + SiO₂ product had the lowest P and Ca values compared with all other products.

Similar Dietary nPP content and Ca:P (DCP vs. MCP). The MCP diet contained higher nPP, P, and Ca levels but lower CP levels than the DCP diet. The MCP product contained lower Ca and higher P levels than the DCP product. Owing to the higher P content and the higher AID P coefficient of MCP than DCP, the needed inclusion level of DCP was 22% higher than the MCP product when diets were formulated to similar nPP content and Ca:P (Table 1).

Similar Total Mineral Content (MDP vs. DMM). The MDP diet had a slightly higher CP, P, and nPP content than the DMM diet. The MDP product had lower P content and higher analyzed Ca content than the DMM IFP mixture.

Similar CPDP content (DCP vs. $MCP + SiO_2$). The DCP diet had higher CP and nPP content but lower Ca content than the MCP + SiO_2 diet. The DCP product contained higher P and Ca content than the MCP + SiO_2 product. When diets were formulated to similar CPDP content, the DCP product had a 67% higher inclusion in the diet than the MCP product in the MCP + SiO_2 diet (Table 1).

Overall Comparison

Live Bird Performance. The overall comparison among treatments indicated that birds fed with MCP diet had the numerically highest ending bird weight on day 14, followed MDP-, DMM-, DFP-, DCP-, $MCP + SiO_2$ -fed birds (P = 0.0460) (Table 4). In addition, no significant differences were found for bird LWG or FCR with the overall comparison. Bikker et al. (2016) similarly reported that day 27 ending bird weight was numerically highest when birds were fed an MCP diet, followed by ending bird weight of birds fed with MDP, DCP, and DFP diets being intermediate, and birds fed with the basal diet had the lowest ending bird weight (P < 0.001). Bikker et al. (2016) sought to determine the prececal P digestibility of various IFP at marginal levels of P supply. The study of Bikker et al. (2016) used a basal diet and 4 additional diets containing MCP (0.88% inclusion), MDP (0.92%), DCP (1.11%), or DFP (1.07%). The 5 diets had a Ca:P of 1.4:1 and an nPP content of 0.28%. However, in the present study, no weight

differences were apparent in the overall comparison at day 21. Pen percent mortality was not significant at any time period (P > 0.05) and did not exceed 5% for any treatment.

Bone Mineralization. The overall comparison among treatments indicated that MDP fed birds had the highest tibia ash (%); MCP, DMM, and MCP + SiO₂ fed birds were intermediate, and birds provided DCP and DFP had the lowest tibia ash (%) (P = 0.0078). Birds fed MCP and DMM had the highest tibia ash (mg/chick); MDP, DFP, and MCP + SiO₂ fed birds were intermediate, and birds provided DCP had the lowest tibia ash (mg/chick) (P = 0.0026).

Mineral Digestibility. The overall comparison among treatments indicated that MDPfed birds had the highest AID Ca coefficient; DFP-, MCP + SiO₂-, DMM-, and MCP-fed birds had intermediate AID Ca coefficient; and DCP-fed birds had the lowest AID Ca coefficient (P < 0.0001) (Table 5). Birds fed with DCP had the lowest AID P coefficient compared with all other treatments (P < 0.0001). Results reported by Bikker et al. (2016) do not agree with our results, stating that birds fed with MCP and MDP had the highest precedal P digestibility; birds fed with the basal diet were comparable with birds provided MDP, and birds fed with DCP and DFP had the lowest precedal P digestibility (P < 0.001). Birds fed with the basal diet, MCP, and MDP had the highest prececal Ca digestibility; DCP-fed birds were comparable with MDP-fed birds, and birds provided DFP had the lowest precedil Ca digestibility (P < 0.001) (Bikker et al., 2016). Their study used 4 suppliers of monohydrated MCP, 3 suppliers of MDP, and 3 suppliers of DCP; this could explain the difference in mineral digestibility results between the 2 studies. In the present study, the MDP diet had the highest digestible Ca and P concentrations, followed by the DFP, DMM, MCP + SiO_2 , and MCP diets having intermediate digestible Ca and P concentrations, and the DCP diet had the lowest digestible Ca and P concentrations (P < 0.0001).

Similar Dietary nPP Content and Ca:P (DCP vs. MCP)

Live Bird Performance. Ending bird weight, LWG, and FI on day 7 and 14 were high

for birds fed with MCP compared with birds fed with DCP (P < 0.05) (Table 4). On day 21, MCP-fed birds had an increased ending bird weight (P = 0.0022) and LWG (P = 0.0021), as well as a decreased FCR (P = 0.0461) compared with DCP-fed birds. Bikker et al, (2016) also reported that birds had increased day 27 ending bird weight, average daily gain, and average daily FI when provided a diet with MCP compared with birds fed with a diet formulated to similar nPP content and Ca:P with DCP. According to the CVB (2016) data and results of the present study, MCP has a higher P digestibility and digestible P content than DCP, which potentially caused birds provided diets with MCP to have increased skeletal growth and greater ending bird weight and LWG. Nonetheless, the DCP diet had the highest phytase activity and still was not able to compensate.

Bone Mineralization. Birds fed with MCP had increased tibia ash (mg/chick) (P = 0.0046) but not tibia ash (%) (P > 0.05) compared with birds fed with DCP (Table 5). The authors speculate that MCP-fed birds had increased tibia ash (mg/chick) compared with DCP-fed birds owing to the fact that MCP has a higher P digestibility and digestible P content than DCP (CVB, 2016). Similarly, Nelson et al., 1990 reported no differences for tibia ash (%) when Cobb 500 male broilers were fed with diets containing either MCP or DCP formulated to 0.16% nPP. Furthermore, the tibia ash values in the study of Nelson et al., 1990 were lower than the ones found in the present study, likely owing to a lower nPP content in diets used in the study of Nelson et al., 1990.

Mineral Digestibility. When diets were formulated to similar nPP content and Ca:P, birds fed with MCP had an increased AID Ca coefficient (P = 0.0045), AID P coefficient (P < 0.0001), digestible Ca concentration (P = 0.0002), and digestible P concentration (P < 0.0001) compared with birds fed with DCP (Table 5). Bikker et al. (2016) also reported that birds had increased prececal Ca and P digestibility when they were provided a diet with a 0.88% inclusion of MCP compared with birds fed a diet with a 1.11% inclusion of DCP. However, results by Anwar et al., 2018 disagree with the current findings. They reported that 24day-old male Ross 308 broilers fed with

semipurified diets had similar AID Ca when fed with a diet with either 4.6% DCP or 5.77% MCP (P > 0.05). The basal diet that contained DCP had a Ca:P of 1.11:1 and 0.81% nPP, while the diet containing MCP had a Ca:P of 0.79:1 and 1.29% nPP (Anwar et al., 2018).

Similar Total Mineral Content (MDP vs. DMM)

Live Bird Performance. Diets formulated to have similar total mineral content were similar in all performance measurements (P > 0.05) (Table 4). The MDP and DMM diets had similar CPDP content (0.7 g); therefore, birds provided with these diets were expected to have similar performance metrics.

Bone Mineralization. Tibia ash (%) was high for birds fed with MDP compared with birds fed with DMM when diets were formulated to similar total mineral content (P = 0.0093) (Table 5). The results mentioned later in this article show that the MDP diet had a higher digestible Ca and P concentration (P < 0.05) than the DMM diet, which could have led to the increase in tibia ash.

Mineral Digestibility. Birds fed with MDP had increased AID Ca coefficient (P = 0.0074), digestible Ca concentration (P = 0.0054), and digestible P concentration (P = 0.0321) compared with birds fed with DMM when diets were formulated to similar total mineral content. When an IFP contains less than 80% MCP but more than 51% MCP, it is considered MDP (Yara International). The MDP product was composed of 40 to 60% MCP monohydrate and 25 to 40% DCP dihydrate (The Mosaic Company). The DMM product contained 20% MCP and 60% DCP. Therefore, the MDP product potentially contained more MCP than the DMM product, causing the MDP product to have increased mineral digestibility compared with the DMM product. As mentioned earlier, MCP has an increased P digestibility and digestible P content compared with DCP (CVB, 2016).

CPDP (DCP vs. $MCP + SiO_2$)

Live Bird Performance. Diets formulated to have similar CPDP content were similar in all performance measurements (P > 0.05)

(Table 4). The DCP and MCP + SiO₂ products had similar CPDP content (0.5 g); therefore, birds provided with these diets were expected to have similar performance metrics.

Bone Mineralization. No significant differences in bone mineralization were found when diets were formulated to similar CPDP content (P > 0.05) (Table 5). However, results of the digestible Ca and P concentration of each diet do not agree with the results found for bone mineralization; the MCP + SiO₂ diet had increased digestible Ca and P concentrations compared with the DCP diet (P < 0.05). The phytase activities of the 2 diets could lead to an explanation of no differences observed in bone mineralization. The DCP diet had a phytase activity of 1,300 FTU/kg, whereas the MCP + SiO₂ diet had a phytase activity of 530 FTU/kg.

Mineral Digestibility. Birds fed with MCP + SiO₂ had increased AID Ca coefficient (P = 0.0019) and AID P coefficient (P < 0.0001) compared with birds fed with DCP when diets were formulated to similar CPDP content (Table 5). The authors speculate that the increased AID P coefficient of birds fed with MCP + SiO₂ compared with birds fed with DCP could be due to the CVB (2016) data underestimating the current studies' P digestibility of the MCP product or overestimating the P digestibility of the DCP product. However, despite these digestibility differences, broiler performance and tibia ash were not affected. As previously stated, IFP vary in P and Ca digestibility; this variability is due to many factors (Huyghebaert et al., 1980; Lima et al., 1999; Kleyn, 2013; WPSA, 2013). In addition, measurable differences in mineral digestibility may not translate to differences in performance or tibia ash measures. The authors do not imply that similar results would be obtained using different IFP products. In addition, pellet manufacture efficiency and pellet quality influences of IFP products should be considered in IFP selection.

CONCLUSIONS AND APPLICATIONS

 Birds fed with MCP had increased ending bird weight and LWG at each measurement period, tibia ash (mg/chick), and mineral digestibility compared with birds fed with DCP when diets were formulated to similar nPP content and Ca:P, despite the DCP product having a 22% higher dietary inclusion compared with the MCP product. This is explained by the greater digestible P content in the MCP diet.

- 2. When diets were formulated to similar CPDP content, birds fed with MCP + SiO₂ had increased mineral digestibility compared with birds fed with DCP, despite the greater DCP inclusion (67%, compared with MCP in the MCP + SiO₂ diet). As expected, similar CPDP content translated to similar broiler performances and tibia mineralization status, showing the importance of a proper evaluation system for P digestibility.
- 3. Different IFP may affect early broiler performance, tibia mineralization, and Ca and P digestibility differently, despite diets being formulated to similar i) nPP content and Ca:P and ii) total mineral content. These differences are likely associated with variation in digestible minerals of specific IFP.
- 4. Diets formulated using CPDP may lower total P inclusion and optimize feed P utilization.

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