

# Effect of Calcium Carbonate Particle Size on the Available Phosphorus Requirement of Broiler Chicks

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**ABSTRACT** Sixteen hundred broiler strain cockerels housed in battery brooders 1 to 21 days of age were used to determine the effect of feeding different commercially available particle-sized calcium carbonate ( $\text{CaCO}_3$ ) on the phosphorus requirement. Five replicates of 8 chicks each were fed corn-soy type diets consisting of either 12 to 20, 20 to 60, or 100 to 200 (passed-retained basis) United States Bureau of Standards (USBS) sieve-sized  $\text{CaCO}_3$ . Each particle-sized  $\text{CaCO}_3$  was fed in diets containing either .20 to .30% (Experiment 1), .15 to 30% (Experiment 2), or .20 to .45% (Experiment 3) available phosphorus (AP) with .05% increments. Supplementary phosphorus was added as sodium phosphate monobasic.

Tibia ash and body weight were greater when the USBS 20 to 60 particle-sized  $\text{CaCO}_3$  was fed than when either the USBS 12 to 20 or 100 to 200 particle-sized  $\text{CaCO}_3$  was fed. When .90% dietary calcium was fed to broiler chicks, the phosphorus requirement was either .35% AP with the USBS 20 to 60 particle-sized  $\text{CaCO}_3$  or at least .45% AP with either the USBS 12 to 20 or 100 to 200 particle-sized  $\text{CaCO}_3$ .

Blood alkaline phosphatase, which indicates bone calcification, was inversely correlated ( $r = -.65$ ) with bone ash. Blood calcium and blood phosphorus increased, and blood alkaline phosphatase decreased with increasing dietary phosphorus.

These data show that the 21-day-old broiler chick's available phosphorus requirement is less in corn-soybean meal diets containing a USBS 20 to 60 particle-sized  $\text{CaCO}_3$  than in diets containing either the USBS 12 to 20 or 100 to 200 particle-sized  $\text{CaCO}_3$ .

(*Key words:* calcium, particle size, available phosphorus, broilers)

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## INTRODUCTION

Inorganic phosphorus is the most expensive mineral supplement in practical poultry diets. During periods of phosphorus shortages, nutritionists will normally decrease the dietary phosphorus level in an attempt to conserve existing phosphorus supplies. The available phosphorus requirement of broiler chicks has been reported by most researchers to be approximately .45% (Sieburth *et al.*, 1952; Fisher *et al.*, 1953; Almquist, 1954; Waldroup *et al.*, 1963; Lillie *et al.*, 1964; Peeler, 1965; and NRC, 1977). Singen *et al.* (1948) indicated that the chick's available phosphorus requirement was between .38% and .47%. Very low total phosphorus requirement values have been reported by Grau and Zweigart (1953), .45%; Gillis *et al.* (1949), .5%; and Hart *et al.* (1930), .3%. Scott *et al.* (1976) reported that the available phosphorus requirements of laying hens for maximum egg production and egg shell quality is no greater than .26%. Under field conditions, the available phosphorus level in practical broiler chick starter diets is approx-

imately .45% with .9 to 1.0% dietary calcium. However, during short phosphorus supplies, the level of dietary available phosphorus in broiler diets may fall well below .45% under practical field conditions.

The effects of calcium carbonate ( $\text{CaCO}_3$ ) particle size have been thoroughly investigated with both laying hens and broiler chicks. Scott *et al.* (1971) and Roland *et al.* (1972) concluded that calcium is "metered" from the gizzard into the blood stream when hens were fed calcium with large particle sizes. Roland *et al.* (1972) found that the majority of calcium (derived from the larger particle sizes of  $\text{CaCO}_3$ ) metered from the gizzard occurs during the summer. McNaughton *et al.* (1974) also found that calcium use by the broiler was dependent upon the particle size of the calcium supplement. A 40 to 50 United States Bureau of Standards (USBS) sieve particle-sized calcium supplement in chick diets appeared desirable to maximize tibia ash.

Hurwitz and Bar (1971) found that feeding low dietary calcium levels could strongly

depress phosphate absorption in broiler chicks; the inhibition effect diminished as the level of dietary calcium was increased. Vandepopuliere *et al.* (1961); Formica *et al.* (1961), and Hart *et al.* (1930) agree that the dietary level of either calcium or phosphorus may affect the requirement for the other element. Two facts appear to be evident in calcium and phosphorus metabolism: 1) phosphate absorption increases with increasing calcium levels until calcium requirement is reached (Hurwitz and Bar, 1971); and 2) calcium use is dependent upon the particle size of the calcium supplement (Scott *et al.*, 1971; Roland *et al.*, 1972; and McNaughton *et al.*, 1974). Therefore, the phosphorus requirement of broiler chicks may vary with different particle sizes of  $\text{CaCO}_3$ . A study was conducted to determine the phosphorus requirement of 1- to 21-day-old broiler chicks with three commercially-available  $\text{CaCO}_3$  particle sizes.

#### EXPERIMENTAL PROCEDURE

A total of 1600 broiler strain cockerels was used in three experiments to determine the effect of  $\text{CaCO}_3$  particle size on the broiler chick phosphorus requirement to 21 days of age. In each experiment, five lots of 8 chicks each were fed each experimental diet. At one day of age, chicks were wingbanded and randomly assigned to decks in electrically heated battery brooders with raised wire floors.

The basal diet reported by McNaughton *et al.* (1974) was used. The basal diet contained .11% calcium as determined by the atomic absorption spectrophotometer.<sup>1,2</sup> Each diet contained 1653 ICU vitamin  $\text{D}_3$  per kilogram of diet. All other nutrients except calcium and phosphorus sources were added to either meet or exceed recommendations of the National Research Council (NRC, 1977).

The  $\text{CaCO}_3$  of different particle sizes was purchased simultaneously from a commercial

source. All calcium sources were purchased from the same supplier and were of the same raw material. Three samples from each source were particle-sized in accordance with the USBS procedure with 50 g samples placed for 10 min in a Ro-Tap<sup>3</sup> sieve shaker. At least 80% of the  $\text{CaCO}_3$  particles fell within either a USBS 12 to 20, 20 to 60, or 100 to 200 (passed-retained basis) USBS sieve size range. Therefore, these sources are designated as USBS 12 to 20, 20 to 60, and 100 to 200 particle-sized  $\text{CaCO}_3$ .

Each particle-sized  $\text{CaCO}_3$  and either .20 to 30% (Experiment 1), .15 to 30% (Experiment 2), or .20 to .45% (Experiment 3) available phosphorus with .05% increments were fed to five lots of 8 broiler cockerels. Supplementary phosphorus was added in the form of sodium phosphate monobasic (certified grade containing 22.45% phosphorus). Supplementary calcium was added to furnish either .70% dietary calcium in Experiments 1 and 2 or .90% dietary calcium in Experiment 3.

In each experiment, all birds were individually weighed, feed consumption was determined, and tibia were removed for bone ash analysis; in Experiment 2, blood samples were collected at 21 days of age. All chicks were sacrificed at the end of each experiment. One tibia was taken from each surviving bird and frozen until the cleaning began. Each group of tibia was boiled for approximately 3 min in distilled water, cleaned with the use of a knife and cheesecloth, dried 12 hr at 105 C, ether-extracted, and dried 24 more hours at 105 C before bone ash determinations were made. All bones were extracted with ethyl ether at least 4 times to assure total fat removal. The fat-free, moisture-free tibia bones were then randomly pooled into lots of either one or two whole tibia bones and ashed at 600 C for 8 hr. For each experimental diet, there was a total of at least 12 ashings per diet.

Serum calcium<sup>4</sup> was determined by procedures described by Schwarzenbach (1955). Serum inorganic phosphorus<sup>4</sup> was determined by procedures described by Tausky and Schon (1953). Serum alkaline phosphatase<sup>4</sup> was determined by procedures described by Coleman (1966).

A factorially arranged randomized complete block design was used in each experiment. Data were examined statistically by the analysis of variance (Steel and Torrie, 1960). Duncan's new multiple range test (Duncan, 1955) was

<sup>1</sup>Model No. 485, Beckman Instrument, Inc., Fulton, CA.

<sup>2</sup>Mention of a trade name, proprietary product, or specific equipment does not constitute a guarantee or warranty by the US Department of Agriculture and does not imply its approval to the exclusion of other products that may be suitable.

<sup>3</sup>The W. S. Tyler Co., Menton, OH.

<sup>4</sup>Reagents obtained from Hycl, Inc., Houston, TX.

used to determine significant differences between means. All statements of significance refer to the 5% level of probability.

### RESULTS

The USBS screen analyses of the commercial calcium sources used in this study are shown in Table 1. Particle size separations indicate that at least 80% of the particles fell within either a 12 to 20, 20 to 60, or 100 to 200 (passed-retained basis) USBS sieve-size range.

*Experiment 1.* When the 20 to 60 particle-sized  $\text{CaCO}_3$  was fed to broiler chicks, both bone ash and body weight (Table 2) were significantly increased by feeding at least .25% available phosphorus (AP) with .70% dietary calcium. However, when either the 12 to 20 or 100 to 200 particle-sized  $\text{CaCO}_3$  was fed, both bone ash and body weight were maximized by feeding at least .30% AP.

Within each dietary AP level, calcium was used more efficiently with the 20 to 60 particle-sized  $\text{CaCO}_3$  than with either the 12 to 20 or 100 to 200 particle-sized  $\text{CaCO}_3$ , as indicated by bone ash and body weight, until the phosphorus requirement was met. Both bone ash and body weight appeared to be sensitive

criteria for determining phosphorus requirement in this experiment. The AP requirement appeared to be low because of the low dietary calcium fed to broiler chicks.

*Experiment 2.* Examination of the data in Table 3 shows that bone ash and body weight were maximum when the diet contained the USBS 20 to 60 particle-sized  $\text{CaCO}_3$  and at least .25% AP. Increasing the level of AP above .25% when the 20 to 60 particle-sized  $\text{CaCO}_3$  was fed did not significantly alter either bone ash or body weight. The presence of either the USBS 12 to 20 or 100 to 200 particle-sized  $\text{CaCO}_3$  resulted in almost equivalent bone ash and body weight when at least .30% dietary AP was fed.

By feeding the lowest level of AP (.15%), feed utilization (feed/gain) was significantly increased by furnishing the calcium in the form of a USBS 20 to 60 particle-sized  $\text{CaCO}_3$ , as compared to the feed utilization when the 100 to 200 particle-sized  $\text{CaCO}_3$  was furnished. By feeding more than .15% dietary AP, feed utilization was not significantly altered as a result of particle size of the calcium supplement.

Serum calcium and serum alkaline phos-

TABLE 1. USBS screen analysis of all commercial sources of calcium used in this study<sup>1</sup>

USBS screen	% Retained			Fine ground limestone
	Limestone Blend <sup>2</sup>			
	Coarse	Medium	Fine	
	(% Retained)			
10	.33	.....	.....	.....
12	4.23	.....	.....	.....
16	39.60	.43	.....	.....
20	43.84	13.71	.....	.22
30	8.53	38.36	.....	.67
40	1.00	28.64	.....	.....
60	.....	15.39	.54	.89
80	.....	1.63	2.13	.....
100	.....	.....	5.40	7.78
120	.....	.....	39.13	.....
170	.....	.....	25.53	.....
200	.....	.....	16.87	18.66
270	.....	.....	7.85	36.89
PAN <sup>3</sup>	2.47	1.84	2.55	34.89
Total	100	100	100	100

<sup>1</sup> Screen analysis of each commercial source of calcium was an average of three analyses.

<sup>2</sup> At least 80% of each limestone blend was retained in a range of either 16 to 20, 30 to 60, or 120 to 200 USBS screen sizes. Thus, limestone blends as either 12 to 20, 20 to 60, or 100 to 200 ranges in particle sizes on a passed-retained basis.

<sup>3</sup> Remaining particles.

TABLE 2. Effect of phosphorus level, limestone particle size and calcium source on broiler performance, Experiment 1

Range in particle size, USBS sieve sizes <sup>1</sup>	Calcium source	Available phosphorus, %		
		.20	.25	.30
21-day body weights, g				
	Ground limestone	.....	.....	405±15 <sup>b</sup>
12-20	Particle-sized limestone	332±4 <sup>f</sup>	351±12 <sup>c</sup>	390±7 <sup>c</sup>
20-60	Particle-sized limestone	390±7 <sup>c</sup>	427±13 <sup>a</sup>	428±12 <sup>a</sup>
100-200	Particle-sized limestone	350±4 <sup>e</sup>	365±4 <sup>d</sup>	396±5 <sup>bc</sup>
Bone ash, %				
	Ground limestone	.....	.....	42.15±.27 <sup>ab</sup>
12-20	Particle-sized limestone	32.79±.84 <sup>d</sup>	38.17±.52 <sup>c</sup>	41.04±.17 <sup>b</sup>
20-60	Particle-sized limestone	38.50±.47 <sup>c</sup>	42.68±.30 <sup>a</sup>	43.20±.70 <sup>a</sup>
100-200	Particle-sized limestone	33.32±.43 <sup>d</sup>	37.26±.46 <sup>c</sup>	41.22±.20 <sup>b</sup>

a,b,c,d,e,f Interaction means within each criterion grouping without a common superscript are significantly different (P<.05).

<sup>1</sup> Each value is shown on a passed-retained basis. At least 80% of material was retained on each sieve series.

phatase decreased, and serum inorganic phosphorus increased, with increasing dietary AP levels (Table 4). Serum calcium and serum inorganic phosphorus were significantly higher,

and serum alkaline phosphatase was significantly lower in birds consuming diets containing the 20 to 60 particle-sized CaCO<sub>3</sub> than in birds consuming diets containing either the 12 to 20

TABLE 3. Effect of phosphorus level and limestone particle size on broiler performance, Experiment 2

Range in particle size, USBS sieve sizes <sup>1</sup>	Available phosphorus, %				Mean
	.15	.20	.25	.30	
21-day body weight, g					
12-20	292±13 <sup>c</sup>	330±10 <sup>d</sup>	440±3 <sup>b</sup>	469±15 <sup>ab</sup>	383
20-60	346±8 <sup>cd</sup>	445±9 <sup>b</sup>	461±9 <sup>ab</sup>	488±11 <sup>a</sup>	435
100-200	218±9 <sup>f</sup>	368±12 <sup>c</sup>	448±5 <sup>b</sup>	488±10 <sup>a</sup>	380
Mean	285	381	450	482	
1-21 day feed/gain					
12-20	1.60±.03 <sup>d</sup>	1.46±.03 <sup>bc</sup>	1.38±.02 <sup>ab</sup>	1.37±.01 <sup>ab</sup>	1.45
20-60	1.52±.02 <sup>cd</sup>	1.37±.02 <sup>ab</sup>	1.35±.03 <sup>a</sup>	1.35±.01 <sup>a</sup>	1.40
100-200	1.86±.06 <sup>e</sup>	1.42±.02 <sup>ab</sup>	1.33±.01 <sup>a</sup>	1.36±.01 <sup>ab</sup>	1.49
Mean	1.66	1.42	1.35	1.36	
Bone ash, %					
12-20	27.55±.59 <sup>g</sup>	31.80±.59 <sup>e</sup>	36.46±.65 <sup>c</sup>	39.35±.63 <sup>a</sup>	33.79
20-60	29.30±.54 <sup>f</sup>	33.37±.58 <sup>d</sup>	39.08±.80 <sup>a</sup>	40.33±.91 <sup>a</sup>	35.52
100-200	27.64±.49 <sup>g</sup>	29.82±.54 <sup>f</sup>	35.08±.68 <sup>c</sup>	38.82±.42 <sup>b</sup>	32.84
Mean	28.16	31.66	36.87	39.50	

a,b,c,d,e,f Interaction means within each criterion grouping without a common superscript are significantly different (P<.05).

<sup>1</sup> Each value is shown on a passed-retained basis. At least 80% of material was retained on each sieve series.

TABLE 4. Effect of phosphorus level and limestone particle size on blood parameters, Experiment 2

Range in particle size, USBS sieve sizes <sup>1</sup>	Available phosphorus, %				Mean
	.15	.20	.25	.30	
	Serum Ca, mg/100 ml				
12-20	8.16±.34 <sup>c</sup>	7.84±.26 <sup>d</sup>	7.00±.22 <sup>e</sup>	6.51±.42 <sup>f</sup>	7.38
20-60	8.14±.59 <sup>c</sup>	7.72±.44 <sup>d</sup>	6.79±.26 <sup>ef</sup>	6.05±.44 <sup>g</sup>	7.18
100-200	8.88±.62 <sup>a</sup>	8.54±.34 <sup>b</sup>	7.99±.36 <sup>cd</sup>	6.98±.49 <sup>e</sup>	8.10
Mean	8.39	8.03	7.26	6.51	
	Serum inorganic phosphorus, mg/100 ml				
12-20	4.10±.02 <sup>f</sup>	4.43±.03 <sup>e</sup>	4.98±.07 <sup>d</sup>	5.46±.03 <sup>b</sup>	4.82
20-60	3.42±.05 <sup>i</sup>	3.87±.05 <sup>g</sup>	5.10±.02 <sup>c</sup>	5.76±.02 <sup>a</sup>	4.38
100-200	3.30±.02 <sup>j</sup>	3.34±.03 <sup>ij</sup>	3.75±.02 <sup>h</sup>	5.12±.03 <sup>c</sup>	3.96
Mean	3.61	3.88	4.61	5.45	
	Alkaline phosphatase, U/L				
12-20	85.69±1.50 <sup>c</sup>	77.04±.37 <sup>d</sup>	70.95±.60 <sup>e</sup>	66.11±.89 <sup>f</sup>	74.95
20-60	72.59±.37 <sup>e</sup>	68.94±1.24 <sup>ef</sup>	66.81±.25 <sup>f</sup>	60.09±1.37 <sup>g</sup>	67.11
100-200	119.45±1.47 <sup>a</sup>	94.63±1.36 <sup>b</sup>	89.03±1.28 <sup>c</sup>	78.89±1.10 <sup>d</sup>	95.50
Mean	92.58	80.20	75.60	68.36	

a,b,c,d,e,f,g,h,i,j Interaction means within each criterion grouping without a common superscript are significantly different ( $P < .05$ ).

<sup>1</sup>Each value is shown on a passed-retained basis. At least 80% of the material was retained on each sieve series.

or the 100 to 200 particle-sized limestone within any level of dietary AP fed.

*Experiment 3.* In Experiments 1 and 2, calcium supplements were added to furnish .70% dietary calcium. The NRC (1977) recommends .90% dietary calcium. Therefore, Experiment 3 was conducted to determine the effect of CaCO<sub>3</sub> particle size on the AP requirement when .90% dietary calcium was fed to broiler chicks to 21 days of age.

Feeding any AP level resulted in greater 21-day bone ash and body weight when the 20 to 60 particle-sized CaCO<sub>3</sub> was fed than when either the 12 to 20 or the 100 to 200 particle-sized CaCO<sub>3</sub> was fed (Table 5). When the 20 to 60 particle-sized CaCO<sub>3</sub> was fed, the AP requirement was found to be .35%; the AP requirement was at least .45% when either the 12 to 20 or the 100 to 200 particle-sized CaCO<sub>3</sub> was fed.

#### DISCUSSION

Evaluation of data presented in this study reveals that the USBS 20 to 60 particle-sized CaCO<sub>3</sub> when fed to 1- to 21-day-old broiler chicks produced greater body weights and bone

ash than either the USBS 12 to 20 or 100 to 200 particle-sized CaCO<sub>3</sub> sources, confirming results of McNaughton *et al.* (1974). The USBS 20 to 60 particle-sized CaCO<sub>3</sub> is apparently "metered" (as termed by Scott *et al.*, 1971) from the gizzard into the small intestine, and maximum calcium utilization results. The use of a USBS 20 to 60 particle-sized calcium supplement in 1- to 21-day-old chick diets appears desirable when maximum bone ash and body weight are used as criteria.

When the USBS 20 to 60 particle-sized CaCO<sub>3</sub> was fed, the AP requirement was either .25% AP with .7% dietary calcium or .35% AP with .9% dietary calcium. However, when either the USBS 12 to 20 or 100 to 200 particle-sized CaCO<sub>3</sub> was fed, the requirement was either .35% AP with .7% dietary calcium or at least .45% AP with .9% dietary calcium.

Hurwitz and Bar (1971) found that feeding low dietary calcium levels could strongly depress phosphate absorption in broiler chicks; the inhibition effect diminished as the amount of dietary calcium increased. This phenomenon appears to be correct until the phosphorus requirement is reached. Dietary phosphorus was apparently used more efficiently with the USBS

TABLE 5. Effect of phosphorus level and limestone particle size on broiler performance, Experiment 3

Range in particle size, USBS sieve series <sup>1</sup>	Available phosphorus, %					Mean
	.20	.25	.30	.35	.40	
	21-day body weight, g					
12-20	370±9 <sup>k</sup>	418±9 <sup>j</sup>	457±12 <sup>i</sup>	494±11 <sup>fgh</sup>	500±8 <sup>efg</sup>	522±10 <sup>cd</sup>
20-60	467±6 <sup>i</sup>	492±6 <sup>fgh</sup>	513±11 <sup>def</sup>	551±6 <sup>ab</sup>	558±7 <sup>a</sup>	547±4 <sup>ab</sup>
100-200	430±6 <sup>j</sup>	474±7 <sup>hi</sup>	488±8 <sup>gh</sup>	516±10 <sup>cde</sup>	536±4 <sup>bc</sup>	532±6 <sup>bcd</sup>
	422	461	456	520	531	534
	Bone ash, %					
12-20	30.22±.51 <sup>j</sup>	35.88±.95 <sup>h</sup>	37.26±.37 <sup>gh</sup>	39.51±.99 <sup>ef</sup>	40.44±.46 <sup>abc</sup>	42.68±.40 <sup>abc</sup>
20-60	33.54±.63 <sup>i</sup>	37.85±.56 <sup>fg</sup>	41.17±.65 <sup>cde</sup>	43.68±.24 <sup>ab</sup>	44.18±.26 <sup>a</sup>	44.19±.24 <sup>a</sup>
100-200	32.54±.49 <sup>i</sup>	36.86±.46 <sup>gh</sup>	38.33±1.16 <sup>fg</sup>	40.62±.72 <sup>dc</sup>	42.10±.21 <sup>bcd</sup>	42.67±.11 <sup>abc</sup>
	32.10	36.86	38.92	41.27	42.24	43.18

a, b, c, d, e, f, g, h, i, j, k Interaction means within each criterion grouping without a common superscript are significantly different (P < .05).

<sup>1</sup> Each value is shown on a passed-retained basis. At least 80% of the material was retained on each sieve series.

20 to 60 particle-sized  $\text{CaCO}_3$  because maximum bone ash and body weight occurred on lower dietary phosphorus levels than when either the 12 to 20 or 100 to 200 particle-sized  $\text{CaCO}_3$  was fed. Serum calcium, inorganic phosphorus, and alkaline phosphatase also indicated that phosphorus is being absorbed and used more efficiently for bone and body development with the 20 to 60 particle-sized  $\text{CaCO}_3$  calcium supplement than with other particle-sized  $\text{CaCO}_3$ .

These data indicate that the 1- to 21-day broiler chick's AP requirement is at least .05% lower in corn-soybean meal diets containing a USBS 20 to 60 particle-sized  $\text{CaCO}_3$  than in diets containing either the USBS 12 to 20 or 100 to 200 particle-sized  $\text{CaCO}_3$ . Results of this study indicate that the broiler chick's AP requirement is dependent upon calcium supplement availability. Therefore, the phosphorus requirement is lower when a USBS 20 to 60 particle-sized calcium supplement is used as compared to either a USBS 12 to 20 or 100 to 200 particle-sized calcium supplement.

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