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Evaluation of Bioplex Zn[®] as an Organic Zinc Source for Chicks

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Abstract: A study was conducted to evaluate Bioplex Zn[®] (a chelated Zn proteinate) as an organic zinc source for broiler chicks. Corn-soybean meal diet without Zn supplementation, containing 23 mg/kg Zn, was used as a basal diet. Day-old male broiler chicks were raised for three weeks. Feed and water with no detectable Zn (<0.001 ppm) were supplied on an ad libitum basis. Treatments consisted of feeding the basal diet alone or with four supplemental levels of Zn (5, 10, 20 and 40 mg/kg) either from Bioplex Zn[®] or from analytical ZnSO₄·7H₂O. Eight replicate cages of six chicks were randomly assigned to each of nine dietary treatments. Zinc supplementation from both sources linearly increased (P<0.01) tibia Zn. Weight gain and feed intake increased quadratically (P<0.05) with the increasing levels of Zn from Bioplex Zn[®] and increased linearly with the increasing levels of Zn from ZnSO₄·7H₂O. Slope-ratio analysis regressing weight gain and tibia zinc content on supplemental Zn level below the inflection points indicated the relative bioavailability value of Bioplex Zn[®] compared with zinc sulfate were 183% and 157% for weight gain and tibia Zn content respectively. Broken-line analysis of the weight gain data determined the required supplemental level of Zn as Bioplex Zn[®] was 9.8 mg/kg of diet for optimal weight gain.

Key words: Bioavailability, chick, zinc proteinate, zinc requirement, zinc sulfate

Introduction

Zinc has been known to be an essential nutrient for normal growth and feathering of chicks for half a century (O'dell and Savage, 1957). The Zn requirement of chicks was first defined to be 30 ppm by Roberson and Schaible (1958). Subsequent research provided evidence for the 1994 Nutrient Requirements for Poultry (NRC, 1994) to set the requirements of broilers at 40 ppm. However, most of these research data are more than 10 years old and do not represent the needs of modern strains of commercial poultry (Leeson, 2005). Purified or semi-purified diet was traditionally used as basal diet in trace mineral requirement studies. The data from these trails do not reflect the real requirement due to lack of soluble fiber and phytate in basal diet that have negative influence to the bioavailability of trace minerals (O'dell and Savage, 1960). On the other hand, organic mineral sources, such as proteinate and amino acid chelate, have been used increasingly in recent years due to their higher bioavailability (Wedekind and Baker, 1989; Wedekind *et al.*, 1992; Cao *et al.*, 2000) and lower manure loading (Manon *et al.*, 2005; Pierce *et al.*, 2005). In general, inconsistency exists among research results regarding bioavailability of organic Zn sources. Some studies indicated small or no differences in bioavailability of Zn between inorganic and organic Zn sources (Hill *et al.*, 1986; Pimentel *et al.*, 1991; Ammerman *et al.*, 1995). The objective of this study was to compare the relative bioavailability value of Bioplex Zn[®] (a chelated Zn proteinate) with reagent grade ZnSO₄·7H₂O in chicks and to re-evaluate the

requirement of Zn for optimal growth of broiler chicks during 1 to 21d period when different sources of Zn were supplemented in practical corn soybean meal diet.

Materials and Methods

Chicks and diet: A total of 432 one-day-old male broiler chicks from a local hatchery (Avian Division, Cobb-Vantress, Monticello, KY, USA) were used. Chicks were housed in mesh wire-floored pullet starter cages (61 cm x 51 cm x 36 cm) with temperature of 31°C for the first week and 27°C for the remainder of the study. Each cage had one feeder that was covered by plastic and two nipple adjustable drinkers. Chicks were provided *ad libitum* access to feed and tap water that contains no detectable zinc (<0.001 ppm). Continuous light (22L:2D) was provided daily. All procedures were approved by the University of Kentucky Institutional Animal Care and Use Committee.

A corn-soybean meal basal diet (Table 1) was used and formulated to be adequate in all nutrients except Zn (NRC, 1994). The analyzed Zn concentration in the basal diet was 23 mg/kg. Dietary additions of Zn included 5, 10, 20 and 40 mg/kg diet and was provided as reagent grade Zn sulfate (ZnSO₄·7H₂O) or Bioplex Zn[®], a chelated Zn proteinate, that contained 10% Zn and was supplied by Alltech Inc. Nicholasville, KY, USA. Zn was added to the diet at the expense of corn.

A randomized complete block design was used with blocks based on physical location of the cages within the room. Chicks were randomly distributed to each of nine dietary treatments within each block with eight

Table 1: Ingredient and composition (as fed basis) of the basal diet¹

Ingredient	% of Diet	Nutrient ¹	Calculated value
Corn	54.91	AMEn, kcal/kg	3150
Soybean meal (48%)	36.5	Protein, %	22
Corn oil	4.6	Calcium, %	1
Salt	0.35	Available P, %	0.45
Calcium carbonate (UPS grade)	1.45	TSAA, %	0.91
Dicalcium phosphate (UPS grade)	1.08	Lysine, %	1.25
Sodium phosphate	0.4	Sodium, %	0.2
Vitamin premix ²	0.25		
Zn free mineral premix ³	0.25		
DL-Methionine	0.21		

¹Contained 23 mg Zn/kg by analysis. ²Supplied per kg diet: 11,025 I.U. vitamin A, 3,528 I.U. vitamin D3, 33 I.U. vitamin E, 0.91 mg vitamin K, 2 mg thiamin, 8 mg riboflavin, 55 mg niacin, 18 mg Ca pantothenate, 5 mg vitamin B-6, 0.221 mg biotin, 1 mg folic acid, 478 mg choline, 28 µg vitamin B-12. ³Supplied per kg diet: 80 mg iron, 60 mg manganese, 13 mg copper and 0.15 mg selenium.

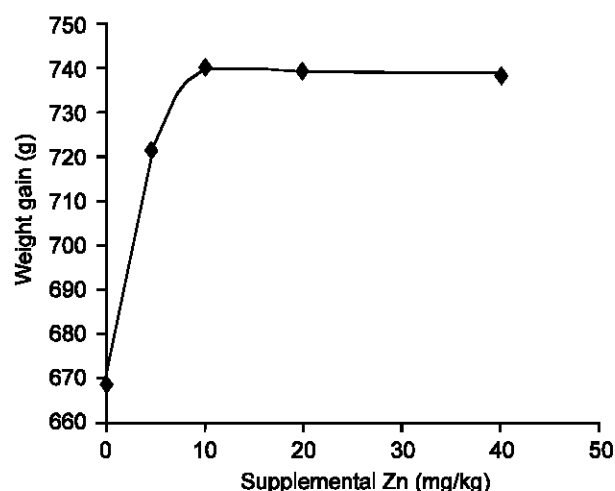


Fig. 1: Broken-line analysis plot of weight gain as a function of supplemental Zn as Bioplex Zn[®]. Breaking point occurred at 9.8 mg/kg diet

replicate cages of six chicks per cage. The study lasted 21 days. Chicks and feed were weighed at the beginning and every week period. On d 21, three chicks from each cage (24 chicks per treatment) were randomly selected and euthanized with argon gas followed by cervical dislocation. The liver and right tibia were removed and pooled by cage. The tissues were stored in -20°C until analysis.

Chemical analysis: Zn in diets, tap water and tissues were determined by an Inductively Coupled Plasma Optical Emission Spectrometer (ICP-OES) (Varian Analytical Instruments, Walnut Creek, CA). Livers were homogenized and dried at 60°C for 72 h. The dry matter content of liver was determined (AOAC, 1995). Bones were boiled in deionized water for 15 min, cleaned of all soft tissue and dried at 60°C for 72 h. Then the bones were extracted in petroleum ether for 72 h and dried for 12 h in 105°C before they were ashed at 600°C overnight in a muffle furnace. The ash weight of the bone was

recorded and used for the calculation of total tibia Zn content. The feed, liver and bone ash were then microwave digested with HNO₃ (AOAC, 1995) before going to the ICP-OES analysis.

Statistical analysis: Data were analyzed by ANOVA for a randomized complete block design using the linear model of Statistix V. 8. (2003) (Analytical Software, Tallahassee, FL). Mean differences were determined using Fisher's LSD test. Linear and quadratic effects were tested using polynomial contrasts. Multiple linear regression and slope-ratio methodology (Finney, 1978) were used to determine the relative bioavailability of Bioplex Zn[®] comparing with zinc sulfate. A SAS (2003) NLIN procedure was used to determine the breakpoint wherein the weight gain or total tibia zinc content was regressed on dietary supplemental zinc concentration. Two different broken-line regression models including a simple two straight-line, one-breakpoint model and a quadratic broke-line model (Robbins *et al.*, 2006) were used.

Results and Discussion

The Zn content of all treatment diets was analyzed and was consistent with the theoretical value (data not shown). The results of growth performance and tissue mineral content are listed in Table 2. Dietary addition of Zn from both sources increased (P<0.01) liver Zn content. The feed intake and weight gain were increased differently when different sources of Zn were supplemented in the diet. The feed intake and weight gain were increased quadratically with increasing levels of dietary Zn as Bioplex Zn[®]. Both feed intake and weight gain were linearly increased by dietary supplementing Bioplex Zn[®] until 10 ppm, after which no further increase occurred. The feed intake and weight gain were linearly (P<0.05) increased with increasing levels of dietary Zn as zinc sulfate until 40 ppm. However, the increasing rate is bigger when the supplemental level of Zn is less than 20 ppm. One-slope, quadratic broken-line analysis of weight gain to dietary supplemented levels of Zn from

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Table 2: Performance and tissue mineral content of chicks fed Bioplex Zn[®] and zinc sulfate in a corn-soybean meal diet¹

Zinc source	Added Zn (mg/kg)	Feed intake (g)	Weight gain (g)	Total Tibia Zn (µg)	Liver Zn (µg/g dry weight)
Basal	0	886 ^e	669 ^d	286 ^a	66.4 ^e
Bioplex Zn	5	948 ^{ab}	722 ^{bc}	423 ^e	71.1 ^{cd}
	10	973 ^a	750 ^{ab}	548 ^d	74.8 ^{bc}
	20	956 ^{ab}	732 ^{abc}	710 ^b	74.6 ^{bcd}
	40	948 ^{ab}	734 ^{abc}	783 ^a	76.4 ^{ab}
Zinc sulfate	5	933 ^b	713 ^c	353 ^f	70.5 ^d
	10	951 ^{ab}	730 ^{abc}	469 ^e	71.5 ^{cd}
	20	963 ^{ab}	751 ^{ab}	649 ^e	76.3 ^{ab}
	40	973 ^a	760 ^a	795 ^a	79.4 ^a
Pooled SEM	-	13	12	20	1.5

¹The performance data represent means of eight replicate groups of six chicks during the period 1 to 21d. The tissue data represent means of eight replicate groups of three chicks. a, b, c, d, e, f, g, P<0.01.

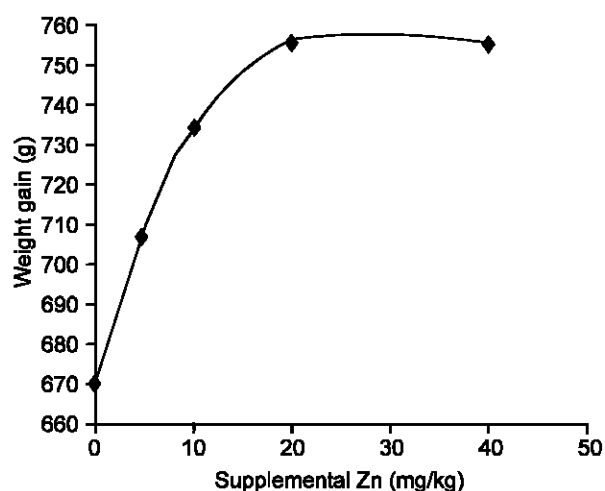


Fig. 2: Broken-line analysis plot of weight gain as a function of supplemental Zn as zinc sulfate. Breaking point occurred at 20.1 mg/kg diet

Bioplex Zn[®] or zinc sulfate resulted in the following two models. The model: $y = 739 - 0.727 * (9.8-x) * (9.8-x)$, where $(9.8-x)$ is zero at value of $x > 9.8$, fitted to the data from Bioplex Zn[®] groups ($P < 0.05$, $r^2 = 0.95$) (Fig. 1). This model indicated that the break point occurred at 9.8 ppm, which means that the required dietary supplemental level of Zn as Bioplex Zn[®] in the corn soybean meal diet is 9.8 mg/kg diet for optimal growth rate. The model: $y = 755 - 0.208 * (20.1-x) * (20.1-x)$, where $(20.1-x)$ is zero at value of $x > 20.1$, fitted to the data from zinc sulfate group. This model indicated that the break point occurred at 20.1 ppm ($P < 0.05$, $r^2 = 0.98$) (Fig. 2), which means that the required dietary supplemental level of Zn as zinc sulfate in the corn soybean meal diet is 20.1 ppm for optimal growth rate. This value is close to the NRC (1994) Zn requirement of 40 ppm when the Zn content of 23 ppm in the basal diet is taken into consideration as a contributor to the total requirement. This result is also consistent with the data reported by Batal *et al.* (2001). Multiple linear regression of weight gain on supplemental Zn level from Bioplex

Zn[®] and zinc sulfate below breaking point resulted in a following equation: $Y = 683.6 + 6.84 (\pm 1.59) X_1 + 3.73 (\pm 0.83) X_2$ ($r^2 = 0.89$), in which Y represents weight gain, X_1 represents supplemental level of Zn as Bioplex Zn[®] and X_2 represents supplemental level of Zn as zinc sulfate. The slope ratio from this equation indicated that the relative bioavailability value of Bioplex Zn[®] was 183% compared with zinc sulfate.

Dietary supplementation of Bioplex Zn[®] and zinc sulfate linearly ($P < 0.01$) increased total tibia Zn content. At each supplemental level of Zn except the maximal level of 40 ppm, the total tibia zinc content of chicks from Bioplex Zn[®] supplemental group was significantly ($P < 0.01$) higher than that of chicks from zinc sulfate supplemental group. For the Bioplex Zn[®] supplemental group, the increase rate of total tibia Zn content with increasing level of dietary Zn was higher when the Zn supplemental level was below 20 ppm. One-slope, straight broken-line analysis of total tibia Zn content to dietary supplementing levels of Zn as Bioplex Zn[®] resulted in the model: $Y = 746.5 - 26.2 * (17.5 - x)$, where $(17.5 - x)$ is defined as zero if $x > 17.5$. This model indicated that the break point occurred at 17.5 ppm ($P < 0.01$, $r^2 = 0.98$). Multiple linear regression of total tibia Zn content on supplemental Zn level from Bioplex Zn[®] and zinc sulfate below breaking point resulted in a following equation: $Y = 321.8 + 20.1 (\pm 2.61) X_1 + 12.8 (\pm 1.34) X_2$ ($r^2 = 0.95$), in which Y represents total tibia Zn content, X_1 represents supplemental level of Zn as Bioplex Zn[®] and X_2 represents supplemental level of Zn as zinc sulfate. The slope ratio from this equation indicated that the relative bioavailability value of Bioplex Zn[®] was 157% compared with zinc sulfate. Cao *et al.* (2000) reported that the bioavailability of Zn from proteinate was 139% that of zinc sulfate based on multiple linear regression slope ratio of bone Zn.

In summary, these results from this study indicate that the relative bioavailability value of Bioplex Zn[®] is 183% that of zinc sulfate based on the weight gain data, and 157% that of zinc sulfate based on the total tibia Zn content. The supplemental level of Zn as Bioplex Zn[®] and zinc sulfate in corn soybean meal diet required for

optimal growth rate of broiler chicks during starting phase (1-21d) were 9.8 mg/kg diet and 20.1 mg/kg diet respectively.

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