Original Research

Poultry

Growth performance and bone characteristics of broiler chickens fed corn-soy diet supplemented with different levels of vitamin premix and sources of mineral premix

T. Ao, M.A. Paul, A.J. Pescatore, L.M. Macalintal, M.J. Ford and K.A. Dawson

Alltech-University of Kentucky Nutrition Research Alliance

Summary

Inorganic trace mineral salts in the premix have a detrimental effect on the stability of vitamins due to redox reactions. A study was conducted to investigate the effect of dietary supplementation of different levels of vitamin premix with different mineral premixes on the performance and bone characteristics of broilers. A 2 x 2 factorial dietary treatment was used with two levels of vitamins and two types of minerals in the premix. A total of 1056, one-day old chicks were randomly assigned to four dietary treatments with 12 replicate pens of 22 chicks for 28 d. An interactive effect between vitamin levels and mineral sources on weight gain and feed intake of chickens was detected. Chickens fed the diet containing 100% vitamin premix with either source of mineral premix had higher ($P < 0.01$) weight gain and feed intake than those fed the diet containing 30% vitamin premix with either source of mineral premix. However, the chickens fed the diet containing the 30% vitamin premix with the organic minerals had higher ($P < 0.01$) weight gain and feed intake than those fed the diet containing 30% vitamin premix with inorganic minerals. Chickens fed the diet containing organic mineral premix had lower ($P < 0.05$) mortality and feed to gain ratio and higher ($P < 0.01$) bone breaking strength and ash content of tibia than those fed the inorganic mineral premix treatment. Chickens fed the diet containing 100% vitamin premix had higher ($P < 0.01$) breaking strength of femur and tibia ash than those fed the diet containing 30% vitamin premix. The results from this trial indicated that total replacement of inorganic trace minerals with organic minerals can increase the storage stability of vitamins in feed premixes containing both vitamins and trace minerals, which is reflected in better growth performance in poultry.

Keywords: broiler, performance, vitamin mineral premix, bone characteristics, vitamin activity

(Received 21 February 2019 – Accepted 31 March 2019)

Introduction

Vitamins are essential in diets and form the most expensive feed ingredients in animal feed. However, most vitamins are not stable and can undergo significant deterioration during feed processing and storage (Scott, 1972; Killeit, 1988; Coelho, 1991). Besides many other factors, trace minerals such as Zn, Cu and Fe have a detrimental effect on the stability of vitamins due to redox reactions, which is an important consideration in feed premixes containing both vitamins and trace minerals

(Wornick, 1968; Shurson *et al.*, 2010). Research showed that 55% of vitamin B6 activity was lost when included in a multi-vitamin premix containing inorganic trace minerals which was stored at 36.7°C for three months, compared to a 24% loss in a similar premix containing no trace minerals (Adams, 1972). It was reported that free metal ions are the most reactive and destructive to vitamins in premixes, followed by sulphates, carbonates and oxides (Creek *et al.*, 1960; BASF, 1999; Coelho 2002). Some *in vitro* observations have indicated that

chelated minerals have a less destructive effect on the vitamin stability than inorganic forms (Luo *et al.*, 2005; Lu *et al.*, 2010). This is possibly due to the structure of chelation that protects active free metal ions from reactions with other compounds, such as vitamins. There is little information about vitamin stability in premixes with organic mineral chelates in current literature. In order to investigate the effect of different mineral premixes (inorganic or organic) on the activity of vitamins in the premix, a marginal level of vitamins (30% commercial level as reported by Chadwick *et al.*, 2018), was used in the current trial. Besides the analysis of vitamin activity in the premix after storage, growth performance and bone characteristics of broiler chickens were observed.

Materials and Methods

Chickens and diet

One-day-old male broiler chicks from a local hatchery (Cobb-Vantress, Monticello, KY) were used. Chickens were housed in floor pens (122 × 183 cm) with room temperature following the current management guidance for Cobb 500. Light was provided at 20L: 4D daily. Chickens were given *ad libitum* access to feed and water. All procedures were approved by the University of Kentucky Institutional Animal Care and Use Committee.

A corn-soybean meal basal diet (Table 1), fed in mash form, was used and formulated to be adequate in all nutrients (National Research Council, 1994). Minerals were added to the diet as either inorganic mineral salts or organic mineral proteinates (Bioplex[®] mineral, Alltech Inc. Nicholasville, KY) for Zn, Cu, Mn and Fe (Table 1). Organic Se (Sel-Plex[®], Alltech Inc., Nicholasville, KY) at 0.3 mg/kg of diet was used to replace sodium selenite at 0.3 mg/kg of diet in control diet.

Experimental design and sampling

A randomised, complete block design was used in this study, with blocks based on physical location of the pens within the room. Treatments were a 2 × 2 factorial arrangement with two levels of vitamins (100 or 30% commercial level) and two forms of minerals (inorganic mineral premix at a commercial level or organic mineral premix at recommended level by the company). The vitamin premix was provided by DSM Nutritional Products, Inc. (Parsippany, NJ). The low level of vitamins (30% commercial level) was used to represent a marginal

Table 1. Ingredients and nutrient composition (as-fed basis) of the basal diet

Ingredient	Starter diet (1–21d) % of diet	Grower diet (22–28d) % of diet
Corn	56.71	62.34
Soybean meal (48% CP)	36.00	30.90
Soybean oil	2.95	2.80
Salt	0.45	0.45
Limestone	1.26	1.10
Dicalcium phosphate	1.75	1.58
L-lysine	0.12	0.10
DL-Methionine	0.26	0.23
Vitamin premix ¹	0.25	0.25
Mineral premix ²	0.25	0.25
Nutrient (calculated values)		
AME _n , kcal/kg	3.05	3.15
Protein, %	22.00	20.00
Ca, %	1.00	0.90
Nonphytate P, %	0.45	0.41
TSAA, %	0.90	0.72
Lysine, %	1.24	1.11

¹Supplied per kg diet for all diets: 9900 IU of vitamin A (retinyl acetate), 2750 IU of vitamin D₃, 1.98 mg of vitamin K₃, 1.98 mg of thiamine, 6.6 mg of riboflavin, 44 mg of niacin, 11 mg of Ca pantothenate, 3.96 mg of vitamin B₆ (pyridoxines), 0.11 mg of biotin, 1.32 mg of folic acid, 495 mg of choline, 19.8 µg of vitamin B₁₂ (cyanocobalamin). Vitamin E (DL-α-tocopheryl acetate): 33 IU/kg.

²Supplied per kg diet for the diets containing inorganic salts: 45 mg Fe as FeSO₄·H₂O, 100 mg Mn as MnSO₄·H₂O, 10 mg Cu as CuSO₄·5H₂O, 100 mg Zn as ZnO, 0.35 mg I as KIO₃, and 0.30 mg Se as sodium selenite. Supplied per kg diet for the diets containing organic minerals: 11 mg Fe as Bioplex Fe, 40 mg Mn as Bioplex Mn, 4 mg Cu as Bioplex Cu, 40 mg Zn as Bioplex Zn, 0.35 mg of iodine as KIO₃, and 0.30 mg Se as Sel-Plex.

level under research conditions based on findings reported by Chadwick *et al.* (2018). The four treatments were: (1) corn-soy basal diet containing 100% vitamin levels and inorganic minerals; (2) corn-soy basal diet with 100% vitamin levels and organic minerals; (3) corn-soy basal diet with 30% vitamin levels and inorganic minerals; (4) corn-soy basal diet with 30% vitamin levels and organic minerals. The premixes containing 30% vitamin level with an inorganic minerals (MV1) and 30% vitamin level with organic minerals (MV2) were stored in room temperature for 90 days before feeding.

A total of 1056, one-day old chicks were randomly assigned to one of four dietary treatments to give 12 replicate pens of 22 chicks. The experiment lasted for 28 days. Chickens and feed were weighed at the beginning of the trial and then weekly. Feed conversion ratio was calculated and corrected for mortality. On d 21, one chicken from each pen (12 chicks per treatment) was randomly selected and euthanised using argon gas followed by cervical dislocation. The tibia and femur were taken for the analysis of breaking strength and ash content. Right side tibias and femurs were cleaned and used for the assay of breaking strength by using an Instron Testing Instrument (Instron Ltd, Norwood, MA, USA). Left tibias were boiled in deionized water for 15 min, cleaned

Table 2. Effects of dietary treatments on the performance of broiler chickens

Diet	Factor		Performance (d 1-28)			
	Mineral Source	Vitamin Level	Weight gain, g/bird	Feed intake, g/bird	Feed to gain	Mortality, %
1	Inorganic	100%	1608 ^a	2317 ^a	1.44	2.27
2	Organic	100%	1627 ^a	2313 ^a	1.42	0.38
3	Inorganic	30%	1123 ^c	1615 ^c	1.44	5.68
4	Organic	30%	1527 ^b	2135 ^b	1.40	0.76
SEM			20.9	28.9	0.01	0.97
Main Effect of Mineral Source						
	Inorganic		1365	1966	1.44 ^a	3.98 ^a
	Organic		1577	2224	1.41 ^b	0.56 ^b
Main Effect of Vitamin Level						
	100%		1618	2315	1.43	1.33
	30%		1325	1875	1.42	3.22
Source of Variation						
	Mineral source		0.001	0.001	0.03	0.001
	Vitamin level		0.001	0.001	0.34	0.056
	Mineral source x Vitamin level		0.001	0.001	0.45	0.124

Data represent means of 12 replicate groups of 22 chickens
a, b, c: $P < 0.05$

of all soft tissue and dried at 60°C for 72 h. Following this, tibias were extracted in petroleum ether for 72 h and dried for 12 h at 105°C before ashing at 600°C overnight in a muffle furnace. The defatted dry weight and ash weight of the tibia was recorded and used for the calculation of ash percentage.

Statistical methods

Data were subjected to ANOVA as a 2×2 factorial design using the linear model of Statistix V. 9 (Analytical Software, 2008). Mean differences were determined using Fisher's LSD test. The group averages of all birds (22) per pen were used for analysis of growth performance data while the averages of 12 randomly selected birds per treatment were used for analysis of bone breaking strength and ash content.

Results and discussion

The effects of dietary treatments on the growth performance of broiler chickens are shown in Table 2.

An interactive effect between vitamin level and mineral source on weight gain and feed intake of chickens was detected. Chickens fed diets containing 100% vitamin levels with either source of minerals had higher ($P < 0.01$) weight gain and feed intake than those fed the diet containing 30% vitamin levels with either source of minerals. However, the chickens fed the diet containing 30% vitamin levels with organic minerals had higher ($P < 0.01$) weight gain and feed intake than those fed the diet with inorganic minerals. Studies have indicated that inorganic trace mineral

supplements, such as sulphate and oxide with copper, zinc and iron, were the most reactive and had a significant impact on vitamin stability in premixes (Creek *et al.* 1960; Wornick, 1968; Christian 1983; Shurson *et al.*, 2010). Chelated minerals, such as proteinates, are more stable and have a lower propensity for initiating formation of free radicals and, therefore, were the least reactive (Luo *et al.*, 2005; Lu *et al.*, 2010). Chadwick *et al.* (2018) reported that the growth performance and feed intake of chickens during a 42 d growing period were not affected by feeding 30% of normal commercial levels of vitamins under research conditions. The current results were different from previously reported findings, probably because the

Table 3. Effects of dietary treatments on the bone breaking strength of broiler chickens

Diet	Factor		Breaking strength, kg force	
	Mineral Source	Vitamin Level	Tibia	Femur
1	Inorganic	100%	27.8	24.1
2	Organic	100%	29.0	25.1
3	Inorganic	30%	25.7	21.8
4	Organic	30%	30.3	23.0
SEM			1.14	0.82
Main effect of mineral source				
	Inorganic		26.7 ^b	23.0
	Organic		29.7 ^a	24.1
Main effect of vitamin level				
	100%		28.4	24.6 ^a
	30%		28.0	22.4 ^b
Source of Variation				
	Mineral source		0.01	0.19
	Vitamin level		0.71	0.01
	Mineral source x Vitamin level		0.16	0.92

Data represent means of 12 replicates (1 bird/replicate)
a, b: $P < 0.05$

Table 4. Effects of dietary treatments on the tibia ash content of broiler chickens*

Diet	Factor		Tibia ash	
	Mineral Source	Vitamin Level	Ash content, g/tibia	Ash%
1	Inorganic	100%	1.15	53.4
2	Organic	100%	1.17	53.1
3	Inorganic	30%	1.10	51.9
4	Organic	30%	1.22	52.8
SEM			0.03	0.42
Main effect of mineral source				
	Inorganic		1.13 ^b	52.6
	Organic		1.20 ^a	52.9
Main effect of vitamin level				
	100%		1.16	53.2 ^a
	30%		1.16	52.3 ^b
Source of Variation				
	Mineral source		0.03	0.44
	Vitamin level		0.96	0.03
	Mineral source x Vitamin level		0.12	0.15

Data represent means of 12 replicates (1 bird/replicate)

* Tibia was fat free

a, b: $P < 0.05$

vitamin mineral premixes were stored at room temperature for 90 days before being mixed in the diets. Chickens fed the diet containing organic minerals had lower ($P < 0.05$) bird mortality and feed to gain ratio than those fed the diets containing inorganic minerals. Chickens fed the diet containing 100% vitamin levels tended to have lower ($P = 0.056$) mortality than those fed the diet containing 30% vitamins.

Table 3 lists the effect of dietary treatments on the bone breaking strength of chickens.

Chickens fed the diet containing organic minerals had higher ($P < 0.01$) breaking strength of tibia than those fed inorganic minerals, and those fed the diet containing 100% vitamin levels had higher ($P < 0.01$) breaking

strength of femur than those fed the diet containing 30% vitamin levels. The effect of dietary treatments on the ash content of tibia is shown in Table 4.

Chickens fed the diet containing organic minerals had higher ($P < 0.01$) ash content than those fed inorganic minerals. Chickens fed the diet containing 100% vitamin levels had higher ($P < 0.01$) ash percentage than those fed the diet containing 30% vitamin levels. Vitamins such as vitamin D and vitamin K are important to bone development and health. Current results indicated different ash content of tibias from chickens fed different forms of mineral premixes. This is possibly due to the different impact of mineral premixes on the stability of vitamin premix.

Table 5 shows the effect of different mineral premixes on the stability of vitamins in broiler diet premixes after 150 days of storage.

Compared with the calculated activity of vitamins at day 0, all vitamins except pantothenic acid, niacin and biotin in MV1 and thiamin, riboflavin, pantothenic acid, niacin and biotin in MV2 lost a significant portion ($>25\%$) of their activity at the end of the storage period. Overall, the vitamins in MV1 lost more activity than those in MV2; for example, the activity of vitamin A and vitamin K in MV1 was only 10.36 and 6.11% of the activity in MV2 respectively. The activity of thiamin and folic acid in MV1 was only 23.77% and 33.27% of the activity recorded in MV2. The activity of vitamin D3 in MV1 was reduced by almost 50% compared with that in MV2. The activity of riboflavin and pyridoxine in MV1 lost 35% compared with that in MV2. These results were consistent with and reflected the performance and bone characteristic results from the broilers,

Table 5. Effects of different mineral premixes on the storage stability of vitamins in broiler diet premixes¹

Vitamin	Unit/kg	Calculated activity at day 0	Analysed activity of MV1 after storage ²	% MV1 Activity of day 0	Analysed activity of MV2 after storage ³	% MV2 Activity of day 0	% MV1 activity of MV2 after storage
A	IU	594000	23300	3.92	225000	37.88	10.36
D3	IU	165000	43800	26.55	86900	52.67	50.40
E	IU	1980	8.91	0.45	10.1	0.51	88.22
K	mg	118.8	3.21	2.70	52.5	44.19	6.11
Thiamin	mg	118.8	27.1	22.81	114	95.96	23.77
Riboflavin	mg	396	283	71.46	436	110.10	64.91
Pantothenic Acid	mg	660	659	99.85	675	102.27	97.63
Niacin	mg	2640	2490	94.32	2720	103.03	91.54
Pyridoxine	mg	237.6	112	47.14	176	74.07	63.64
Biotin	mg	6.6	6.42	97.27	7.99	121.06	80.35
Folic Acid	mg	79.2	18.7	23.61	56.2	70.96	33.27

¹the vitamin mineral premix was analysed after animal trial. The total storage time is 150 days²MV1: vitamins with inorganic mineral premix³MV2: vitamins with organic mineral premix

whereby chickens fed MV1 which lost more vitamin activity had significant lower growth rate and feed intake and weaker bones than those fed diets with MV2.

Conclusions

The growth performance and bone characteristics of broiler chickens combined with the analytical results of vitamin activity indicated that total replacement of inorganic trace minerals with organic minerals can increase the storage stability of vitamins in feed premixes containing both vitamins and trace minerals.

References

- Adams C.R.** (1972) Effect of environmental conditions on the stability of vitamins in feeds. In: *Effect of Processing on the Nutritional Value of Feeds*. National Academy of Sciences, Washington, D.C, pp. 142.
- Analytical Software** (2008) Statistix 9 User's Manual. Tallahassee, U.S.A., Analytical Software.
- BASF** (1999) Vitamin stability in premixes and feeds: a practical approach. BASF Keeping Current KC 9138, 6TH ED., BASF Corporation, Mount Oliver, New Jersey.
- Chadwick E., Malheiros R., Ferket P., North R.B.** (2018) Microbiota, growth performance, and processing characteristics of broilers fed a standard vs low vitamin and trace mineral diet. *Poultry Science*, **97** (E-suppl 1) M71
- Christian L.D.** (1983) Vitamin stability in mineral mixes formulated with different calcium phosphates stored at two temperatures and relative humidities. In: *Proceedings AFIA Nutrition Council*, pp 22.
- Coelho M.** (1991) Fate of vitamins in premixes and feeds: vitamin stability. *Feed Management* **42** (October): pp 24.
- Coelho M.** (2002) Vitamin stability in premixes and feeds: a practical approach in ruminant diets. In: *Proceedings 13th Annual Florida Ruminant Nutrition Symposium*, pp 127–145.
- Creek R.D., Carrick C.W., Hauge S.M. and Parker H.E.** (1960) The Effect of Technical Grade Manganese Sulfate on Vitamin Stability. *Poultry Science*, **39**:109–111.
- Dove C.R. and Evan R.C.** (1991) Effect of trace minerals on the stability of vitamin E in swine grower diets. *Journal of Animal Science*, **69**:1994–2000.
- Huyghebaert A.** (1991) Stability of vitamin K3 in a mineral premix. *World Poultry*, **7**:71.
- Killeit V.** (1988) The Stability of Vitamins – A Selection of Current Literature. No. 2124, F. Hoffmann-LaRoche Co. Ltd., Basel, Switzerland.
- Lu L., Wang R.L., Zhang Z.J., Steward F.A., Luo X., Liu B.** (2010) Effect of dietary supplementation with copper sulfate or tribasic copper chloride on the growth performance, liver copper concentrations of broilers fed in floor pens and stabilities of vitamin E and phytase in feeds. *Biological Trace Element Research* **138**(1-3):181–189.
- Luo X.G., Ji F., Lin Y.X., Steward F.A., Lu L., Liu B., Yu S.X.** (2005) Effects of dietary supplementation with copper sulfate or tribasic copper chloride on broiler performance, relative copper bioavailability and oxidation stability of vitamin E in feed. *Poultry Science* **84**(6): 888–893.
- National Research Council** (1994) *Nutrient Requirements of Poultry*. 9th ed. National Academy Press, Washington, D.C.
- Scott M.L.** (1972) Effect of processing on the availability and nutritional value of vitamins. In: *Effect of Processing on the Nutritional Value of Feeds*. National Academy of Sciences, Washington, D.C, pp 119.
- Shurson G.C., Salzer T.M., Koehler D.D. and Whitney M.H.** (2010) Effect of metal specific amino acid complexes and inorganic trace minerals on vitamin stability in premixes. *Animal Feed Science Technology* **163**: 200–206.
- Wornick R.C.** (1968) The stability of micro-ingredients in animal feed products. *Feedstuffs* **40**: 25–27.