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Full Length Research Paper

Study of possible reduction or withdrawal of vitamin premix during finisher period in floor and battery cage broiler raising systems

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The effect of dietary vitamin premix withdrawal or reduction between 29 and 35, 36 and 42, and 29 and 42 days of age on broiler chicken performance and immunocompetence was evaluated. The diets were formulated based on wheat and barley, and the experiment was conducted in floor pens (experiment 1) and battery cage (experiment 2) rearing systems in 7 treatments and 4 replicates for each treatment. The results of experiment 1 showed that vitamin premix reduction and withdrawal at 29 days of age did not impair performance during the final period of broiler chicken (29 to 42 days). The results of experiment 2 showed that there were no significant differences in performance with reduction or withdrawal of vitamin premix from diets in 29 to 35 days, but in 36 to 42 days of age, performance of birds fed with a diet that has no vitamin premix (T1) was significantly lower than other treatments ($P < 0.05$). The results of the two experiments demonstrated that immunocompetence response was not affected by treatments in the finisher period ($P > 0.05$). In conclusion, the results of this study indicated that in the battery cage system, it is possible to reduce dietary vitamin premix during finisher period, but withdrawal can negatively affect performance of broiler chickens, while in the floor system, it is possible to withdraw vitamin supplements in broilers' finisher diets.

Key words: Vitamin premix, reduction, broiler, wheat, immunocompetence.

INTRODUCTION

Vitamins are defined as a group of complex organic compounds present in small amounts in natural food-stuffs that are essential for normal metabolism, and its lack in the diet causes deficiency diseases (McDowell, 2000). They are organic compounds present in most feedstuffs in small amounts. Typically, vitamins represent only 0.05% of weight and 1.5% of complete feed cost. They also cause a specific deficiency disease if absent in the diet and cannot be synthesized by the host animal to meet vitamin requirements (Coelho and McNaughton, 1995). It has been reported that minerals and vitamins are being added to the diet when they may not be needed (Skinner et al., 1992). The withdrawal of vitamin or mineral supplements in finisher diets has been evaluated in the last few years as a way to reduce the costs of broiler chicken production. There are different reports about the influence of reduction and withdrawal of vitamin supplements in the diet of broiler chickens, based on corn and soya bean meal performance and immune system in the finisher period. Maiorka et al. (2002) indicated that vitamin and mineral mix withdrawal at 42 days of age did not impair feed intake or weight gain, but significantly affected the feed conversion ratio. Deyhim and Teeter (1993) detected the reduced performance for several growth and carcass traits, when the same withdrawal compound was examined. In contrast, Skinner et al. (1992) reported that removing vitamin (V) and trace mineral (TM) premixes from the diets of broilers that are 28 to 49 days of age had little impact on growth performance. Khajali et al. (2006) suggested that the vitamin and trace mineral contents of the finisher diet were sufficiently high in maintaining a humoral immune response. It seems that it is necessary to study the with-

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drawal or reduction of vitamin supplements in finisher diets based on wheat and barley in the battery cage system due to the following causes:

1. Instability of world corn prices and propensity of the producer to use wheat and barley, instead of corn in the diet of broiler chickens.
2. There are differences between wheat, barley and corn that contains vitamin.
3. Birds in cages require more dietary vitamins than those on floor housing because of more limited opportunity for coprophagy.

Therefore, this study was carried out to evaluate the effects of reduction or withdrawal of the vitamin premix (VP) from broiler diets during finisher period on performance and immunocompetence in two rearing systems (floor and battery cage).

MATERIALS AND METHODS

General procedure

Two experiments were conducted to evaluate the reduction or withdrawal of VP during the final phase of broiler chicken's growth (between 29 and 42 days of age). All birds were fed the same diet (wheat and barley based diet), from day 1 to day 29 (Table 1), and were kept in floor pens. Four hundred and sixteen (416) day-old male broiler chicks of commercial strain (Ross 308) were used and the average initial body weight of chicks in each pen was 42 g. Room temperature was kept at 34°C during the first 3 consecutive days of the trial, and then it reduced gradually according to their age until it reached 22°C on day 21. The light was continuous during the first 3 days, and then the lighting regimen was 23 h/day. The dietary treatments were: T1 - without VP during 29 to 42 days, T2 - 33% VP during 29 to 42 days, T3 - 33% VP during 29 to 35 days and without VP during 36 to 42 days, T4 - 66% VP during 29 to 42 days, T5 - 66% VP during 29 to 35 days and without VP during 36 to 42 days, T6 - 100% VP during 29 to 42 days as the control group, and T7 - 100% VP during 29 to 35 days and without VP during 36 to 42 days (Table 2). The ingredient composition of the experimental diets and the nutrient composition are shown in Table 1. Mash feed and water were available for *ad-libitum* consumption. Prior to formulation, all major dietary ingredients [crude protein (CP), crude fibre (CF), ether extract (EE) and ash] were analyzed as described by AOAC (2000) for AMEN and amino acid (AA) profiles [according to the prediction formula existing in NRC (1994)].

Experiment 1

Five hundred and four (504) male broiler chicks of commercial strain (Ross 308) were randomly allocated to 7 treatment groups, with 4 replicate pens of 18 birds each. The initial body weights were similar in all pens (1135±13.6g). A trial was used to assess the effect of the vitamin premix reduction and withdrawal from 29 and 42 days of age in the floor system.

Experiment 2

Two hundred and twenty-four (224) male broiler chicks weredistri-buted with similar average body weight into battery cages at 23 days of age and were fed similar grower diet up to 28 days of age. Treatments and diets of this experiment (29 to 42 days of age) were similar to trial 1 and the initial body weights were similar in all cages (1120±11.1 g).

Productive performance experiments

Mortality during 29 to 42 days was determined for each pen. The body weight (BW) and feed intake (FI) of chickens were determined at 35 and 42 days of age, then the feed conversion ratio was calculated from these data.

Lymphoid organs weight

At 35 and 42 days of ages, two birds of each replicate in experiments 1 and 2, with similar average weight of each replicate were selected and slaughtered. The relative weight of the lymphoid organs (bursa of Fabricius and spleen) relative weights were measured to the nearest 0.01 g.

Immunological studies

The sheep's red blood cells were used as T-dependent antigens to quantify the antibody response. In the trials, four birds were selected from each of the replicated groups (16/treatment) at 34 days of age and were inoculated intravenously with 0.1 ml of 1% suspension of SRBC (sheep red blood cell). Blood samples were collected at brachial vein 7 days after each injection. The serum from each sample was collected by heat inactivated at 56°C for 30 min and was then analyzed for total mercaptoethanol (Sigma Chemical Co., St. Louis, MO, USA) -sensitive (MES) IgM and mercaptoethanol-resistant IgG anti-SRBC antibodies as previously described (Delhanty and Solomon, 1966; Yamamoto and Glick, 1982; Qureshi and Havenstein, 1994). Briefly, 50 µL of serum was added in an equal amount of phosphate-buffered saline (PBS; pH 7.6) in the first column of a 96-well v-shaped bottom plate (Corning, Corning, NY, USA), and the solution was incubated for 30 min at 37°C. A serial dilution was then made (1:2), and 50 µl of 2% SRBC suspension was added to each well. Total antibody titers were then read after 30 min of incubation at 37°C. The well, immediately preceding a well with a distinct SRBC button, was considered as the endpoint titer for agglutination. For MES (IgM) response, 50 µl of 0.01 M mercaptoethanol in PBS was used instead of PBS alone, followed by the aforementioned procedure. The difference between the total response and the IgG response was considered to be equal to the IgM antibody level (Cheema et al., 2003).

Statistical analysis

The data were subjected to ANOVA as a completely randomized design using the GLM procedure of SAS software (SAS Institute, 2002). Anti-SRBC titers and lymphoid organ weights data were transformed to log2 and arc sine, respectively. Means were separated by Duncan's Multiple Range Test at a significance level of $P<0.05$.

RESULTS AND DISCUSSION

Performance

Experiment 1

Mortality for all groups was within the expected range and

Table 1. Composition of the starter and grower diets used in the pre-experimental.

Ingredient	Starter diet (g/kg)	Grower diet (g/kg)
Wheat	340	351.4
Barely	320	300
Soya bean meal (440 g/kg CP)	239.7	269.3
Gluten meal	56.2	25.1
Soya oil	10.3	18.0
Oyster shell	13.0	12.9
Dicalcium phosphate	10.5	10.5
Vitamin premix[1]	2.5	2.5
Trace mineral premix[2]	2.5	2.5
Sodium chloride	2.8	2.8
DL-Methionine	0. 6	2.1
L-Lysine-HCl	1.3	2.6
Enzyme	0. 5	0. 5
Calculated composition		
AME, kcal/kg	2850	2860
Analyses CP (g/kg)	208	200
Met(g/kg)	4.8	4.1
Met + Cys (g/kg)	1.0	8.6
Lys (g/kg)	1.3	11.2
Na	1.5	1.4
Ca	9.9	8.1
Available phosphorus (g/kg)	4.7	4.1

[1]2.5 kg of vitamin premix contain: retinol acetate (2700 mg), cholecalciferol (400 µg), DL-α-tocopherol acetate (18 mg), menadione (2000 mg), thiamine mononitrate (1800 mg), riboflavin (6600 mg), pyridoxol (3 g), cyanocobalamin (15 mg), D-biotin (100 mg), niacin (10 g), pantothenic acid (30 g), folic acid (1 g) and choline chloride (250 g). [2]2.5 kg of trace mineral premix contain: iron (50 g), copper (10 g), manganese (100 g), cobalt (5 g), zinc (100 g), iodine (1 g), selenium (200 mg) and molybdenum (0.5 mg).

Table 2. Compositions of the diets used during the experimental period (29 to 42 days of age) in the experiments.

Ingredient	Treatment (g/kg)*			
	T 1	T 2	T 4	T 6
Wheat	363.8	360.5	359.7	357.9
Barley	300	300	300	300
Soya bean meal (440 g/kg CP)	279.3	280.9	280.4	280.9
Soya oil	27.4	280	28.6	29
Oyster shell	12.4	125	124	12.4
Dicalcium phosphate	8.9	9	9	9
Vitamin premix	0.00	0.8	1.6	2.5
Trace mineral premix	2.5	2.5	2.5	2.5
Sodium chloride	2.8	2.8	2.8	2.8
DL-Methionine	1.7	1.8	1.8	1.8
L-Lysine-HCl	0.7	0.7	0.7	0.7
Enzyme	0.5	0.5	0.5	0.5
Total	1000	1000	1000	1000

Table 2. Continue.

Calculated composition				
AME, kcal/kg	2900	2900	2900	2900
Analyses CP (g/kg)	200	200	200	200
Met	3.7	3.7	3.7	3.7
Met + Cys (g/kg)	7.7	7.7	7.7	7.7
Lys (g/kg)	9.7	9.7	9.7	9.7
Na	1.6	1.6	1.6	1.6
Ca	7.6	7.6	7.6	7.6
Available phosphorus (g/kg)	3.7	3.7	3.7	3.7

*T1 - basal diet without VP; T2, T4 and T6 - basal diet with 33, 66 and 100% VP during 29 to 42 days, respectively; T3, T5 and T7 were similar to T2, T4 and T6 during 29 to 35 days and then without VP during 36 to 42 days.

Table 3. Vitamin premix reduction or withdrawal effects on weight gain, feed intake and feed conversion efficiency (Experiment 1).

Parameter	29 - 35 day			36 - 42 day			29 - 42 day			42 day
	FI (g)	Gain (g)	FCR[1]	FI (g)	Gain (g)	FCR	FI (g)	Gain (g)	FCR	BW
T 1	1017.5	595.8	1.71	1204.1	640.0	1.88	2222.2	1280.0	1.74	2365.6
T 2	1002.5	580.1	1.73	1183.5	620.6	1.91	2186.2	1241.2	1.76	2361.2
T 3	1012.3	607.5	1.67	1181.3	605.3	1.95	2193.7	1210.6	1.81	2390.3
T 4	1018.9	600.7	1.70	1185.8	672.3	1.76	2204.6	1344.6	1.64	2394.6
T 5	1025.9	602.6	1.70	1183.4	655.1	1.81	2209.3	1310.3	1.69	2393.3
T 6	1016.0	611.1	1.66	1203.2	654.7	1.84	2219.3	1309.4	1.69	2395.3
T 7	1021.02	608.5	1.68	1199.0	648.8	1.85	2220.0	1297.7	1.71	2391.4
SEM	21.8	17.52	0.05	20.06	35.06	0.07	14.56	21.02	0.04	25.36

[1] FCR = total feed intake/total body weight gain (g/g); FI, feed intakel BW, body weight.

there was no significant difference in the mortality of all treatments. The results obtained from Trial 1 for feed intake, body weight and feed conversion ratio are shown in Table 3. Vitamin premix reduction or withdrawal at different ages did not significantly affect FI, BW or FCR ($P > 0.05$). The findings of this study in Trial 1 were slightly similar to those reported by Skinner et al. (1992) and Maiorka et al. (2002), as they showed that vitamin and mineral premix withdrawal from the finisher diet of broiler chickens did not affect performance. Skinner et al. (1992) suggest that the lack of a withdrawal effect could be related to the availability in the body of vitamins and minerals for further growth, as the amount of these supplements usually exceeds two or three times the recommended broiler chicken requirement in poultry diets. In opposition, omitting vitamin from the finisher diet for the same removal period decreased weight gain in three different broiler strains (Deyhim and Teeter, 1993; Patel et al., 1997). These differences may be due to the type of rearing system (floor litter or cages) or differences in diet composition.

Experiment 2

FI, BW and FCR were affected by VP withdrawal. Broilers merely fed a diet without vitamin premix (T1) at 36 to 42 days of age had poorer performance as compared to those that received VP (Table 4).

The findings of this study differ slightly from those reported by Skinner et al. (1992) and Khajali et al. (2006) as they show that VP withdrawal from the finisher diet of broiler chickens did not affect performance, but the findings of this study are nearly similar to the reports of Deyhim and Teeter (1993). They also demonstrated that broiler chickens, reared in batteries under a cycling ambient temperature (24 to 35°C, creating heat stress) and fed diets without vitamin and mineral premix, had reduced weight gain and poorer feed conversion as compared to birds fed normal supplemented diets, whereas birds in cages required more dietary vitamins than those on floor housing because of more limited opportunity for coprophagy. Base on the results of Trials 1 and 2, it seems that we can reduce vitamin premix in

Table 4. Vitamin premix reduction or withdrawal effects on weight gain, feed intake and feed conversion efficiency (Experiment 2).

Parameter	29-35 day			36-42 day			29-42 day			42 day
	FI (g)	Gain (g)	FCR	FI (g)	Gain (g)	FCR	FI (g)	Gain (g)	FCR	BW
T 1	968.7	565.2	1.71	1076.8^{b}	445.1^{b}	2.42^{b}	2048.6^{b}	1010.4^{b}	2.03^{b}	2140.3^{b}
T 2	976.6	557.6	1.75	1223.4^{a}	654.2^{a}	1.87^{a}	2199.9^{a}	1212.0^{a}	1.82^{a}	2332.6^{a}
T 3	975.8	577.3	1.69	1229.5^{a}	637.6^{a}	1.93^{a}	2205.3^{a}	1075.0^{a}	2.05^{a}	2362.1^{a}
T 4	977.1	568.9	1.72	1204.3^{a}	663.0^{a}	1.82^{a}	2181.3^{a}	1231.9^{a}	1.77^{a}	2354.9^{a}
T 5	978.6	579.2	1.69	1239.9^{a}	678.4^{a}	1.83^{a}	2218.4^{a}	1253.3^{a}	1.77^{a}	2370.6^{a}
T 6	974.6	582.7	1.67	1257.5^{a}	684.8^{a}	1.84^{a}	2232.2^{a}	1267.4^{a}	1.76^{a}	2377.1^{a}
T 7	979.8	579.7	1.69	1253.0^{a}	679.2^{a}	1.84^{a}	2232.9^{a}	1259.2^{a}	1.77^{a}	2363.4^{a}
SEM	17.64	32.41	0.05	35.77	30.24	0.1	23.28	32.9	0.04	29.58

^{ab} Means in a column with different superscripts differ significantly ($P < 0.05$).

Table 5. Vitamin premix reduction or withdrawal effects on IgG and IgM anti-SRBC titers (Experiments 1 and 2).

Treatment	Experiment 1			Experiment 2		
	SRBC*	IgG	IgM	SRBC	IgG	IgM
T 1	7.75	2.50	5.25	6.00	2.50	3.50
T 2	5.75	2.25	3.50	5.75	2.25	3.50
T 3	7.25	2.50	4.75	6.25	2.50	3.75
T 4	7.00	2.75	4.25	6.50	2.50	4.00
T 5	6.50	2.50	4.00	6.00	2.50	3.50
T 6	6.75	2.75	4.00	7.00	2.75	4.25
T 7	6.00	2.25	3.75	6.25	2.75	3.50
SEM	1.07	0.25	0.80	0.70	0.30	0.40

*Data expressed as log2. SRBC, Sheep red blood cell.

the finisher broiler diets (until 33%) in 29 to 35 days of age and remove it during 36 to 42 days of age (T3), without a significant negative effect on the performance of broilers, while in the floor system, it can be possible to withdraw vitamin supplements in the finisher broiler diets.

Humoral immunocompetence response

Antibody response data against SRBC as measured by total IgM and IgG levels in experiments 1 and 2 are given in Table 5. In both experiments, humoral immune-competence responses (IgM, IgG and anti-SRBC titers) were not affected by different treatments ($P>0.05$). The effect of vitamin premix reduction or withdrawn on lymphoid organs weights in experiments 1 and 2 are shown in Tables 6 and 7. The bursa of Fabricius and spleen weights were not significantly different in chicks fed diets with various levels of vitamin premix in experiments 1 and 2 ($P>0.05$).

These results agreed with those reported by Deyhim and Teeter (1993) and Khajali et al. (2006). These findings suggested that the vitamin contents of the finisher diet are sufficiently high in maintaining a humoral immune response. These results suggested that the vitamin contents of wheat, barley and soy-bean meal diet were sufficiently high in maintaining a humoral immune response, when the grower diet was not fortified with VP during 29 to 42 days posthatching. It has been demonstrated that the immune system has a higher priority for circulating nutrients and it has the ability to compete favorably with other tissues when nutrient levels are low (Klasing, 1998).

Therefore, it seems that cage-reared broilers cannot access their feces to reach some vitamins which have departed from their intestine, but they can compensate the vitamin deficiency by using a small percentage of vitamin premix in the finisher diet. It seems that there are some reasons which may cause the removal or reduction of the usage of VP (an expensive essential nutrient in poultry's diets) in finisher period diets in the floor and battery cage systems, respectively, for instance: (1) The

Table 6. Vitamin premix reduction or withdrawal effects on lymphoid organs weight (Experiment 1).

Parameter	35 day		42 day	
	Bursa (g/kg)	Spleen (g/kg)	Bursa (g/kg)	Spleen (g/kg)
T 1	0.24	1.96	2.50	1.85
T 2	0.23	2.03	2.58	2.08
T 3	0.21	2.00	2.61	1.93
T 4	0.15	2.16	2.78	1.89
T 5	0.19	2.11	2.52	1.96
T 6	0.22	2.00	2.29	2.36
T 7	0.23	2.05	2.36	1.99
SEM	0.03	0.11	0.09	0.12

Table 7. Vitamin premix reduction or withdrawal effects on lymphoid organs weight (Trial 2).

Parameter	35 day		42 day	
	Bursa (g/kg)	Spleen (g/kg)	Bursa (g/kg)	Spleen (g/kg)
T 1	0.22	2.25	2.39	2.06
T 2	0.18	2.55	2.47	2.10
T 3	0.22	2.45	2.50	2.05
T 4	0.19	2.30	2.40	1.86
T 5	0.20	2.35	2.46	1.94
T 6	0.21	2.30	2.61	2.15
T 7	0.21	2.50	2.53	2.05
SEM	0.04	0.17	0.13	0.16

amount of VP usually exceeds two or three times the recommended broiler chicken requirement in poultry diets. (2) Fat-soluble vitamins may be stored by a bird in its liver and fatty tissue in sufficient quantities to meet requirements for up to 15 days or even longer. (3) There are some vitamins in diet ingredients, such as wheat, barley and soybean meal, that are not considered during diets formulation. (4) Floor-reared broilers can access their feces to reach some vitamins, which have departed from the intestine (this cannot be possible for the birds that are reared in the battery cage system). In conclusion, the results of this study indicate that we can reduce vitamin premix in 29 to 35 days of age and remove it during the second week of the finisher period, without a significant negative effect on performance of broilers; while in the floor system, it can be possible to withdraw vitamin supplements in the finisher broiler diets.

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