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The Effects of Ractopamine, Coenzyme Q_{10} and L-Carnitine Supplementation, Individual or in Combination, on the Hematological Parameters of Broiler Chickens

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Abstract: This study was conducted to evaluate the effect of ractopamine, coenzyme Q_{10} and L-carnitine, individually and in combination, on the hematological parameters of broiler chickens. A total of 512 one day-old male broiler chickens (Ross 308) were randomly divided to eight treatment groups. Each group had four replicates with 16 birds per each. Treatments were assigned as a 2 × 2 × 2 factorial arrangement based on completely randomized design, with two levels of ractopamine (0 and 10 mg/kg), coenzyme Q_{10} (0 and 40 mg/kg) and L-carnitine (0 and 200 mg/kg). Blood was collected at day 42 of age and hematological parameters were measured. Hemoglobin content, red blood cell counts, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration and white blood cell counts were not differ ($P>0.05$) among chicks fed different treatments. Dietary coenzyme Q_{10} + L-carnitine supplementation reduced ($P<0.05$) packed cell volume, which is a positive effect as it relates to ascite. Based upon these results, the addition of ractopamine, coenzyme Q_{10} and L-carnitine to broiler diet, individually or in combination, had no effect on hematological parameters of broiler chickens.

Key words: Ractopamine · Coenzyme Q10 · L- carnitine · Hematological parameters · Broiler chicken

INTRODUCTION

Ractopamine, a beta-adrenergic agonist, is used in cattle and swine diets [1]. The positive effects of beta adrenergic agonist on performance in meat producing animals, including poultry are well documented by several experiments [2-4]. Coenzyme Q_{10} is a vitamin- like substance which is the coenzyme for mitochondrial enzymes of oxidative phosphorylation pathway [5, 6]. L-carnitine is a non-essential amino acid that appears to be necessary for transport of long-chain fatty acids into the mitochondria for ß-oxidation [7, 8]. Producers have to use high amounts of lipids in broiler rations [9]; therefore the addition of these additives to diet is necessary. In the literature, the effects of these additives on hematological parameters have been evaluated alone. Akbariazad *et al*. [10] reported that addition of L-carnitine to diet of broilers at levels of 125 and 250 mg/kg had no significant effect on hemoglobin, red blood cell (RBC), mean corpuscular volume (MCV), mean corpuscular hemoglobin concentration (MCHC) and mean corpuscular hemoglobin (MCH). In contrast, Kardeniz *et al*. [11] showed that RBC, hemoglobin (Hb), packed cell volume (PCV) and MCHC of broilers significantly increased when L-carnitine was added to diet at level of 100 mg/kg. Hoshi *et al*. [12] reported that the use of ractopamine in pregnant sows had no significant effect on blood's hemoglobin. Geng *et al*. [13] reported that PCV was not influenced by coenzyme Q_{10} alone, but significantly decreased by L-carnitine + coenzyme Q_{10}.

In the literature, the effects of addition of ractopamine, coenzyme Q_{10}, L-carnitine to diet, in combination, on the hematological parameters has not been well defined in broiler chickens. The objective of the present study was to investigate the effects of dietary supplementation of ractopamine, coenzyme Q_{10} and L-carnitine, individually and in combination, on hematological parameters of broiler chickens.

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MATERIALS AND METHODS

Chickens and Diets: A total of 512 one-day- old male broiler chicken (Ross 308) was randomly allocated into eight treatments groups. Each group had four replicates with 16 birds per each. Corn and soybean meal based ration was formulated according to the requirements suggested by Ross 308 management guide. Diets were formulated as starter (days 1-10), grower (days 11-24) and finisher (days 25-42). Based on completely randomized design, a 2 × 2 × 2 factorial arrangement was employed with two levels (0 and 10 mg/kg) of ractopamine, two levels (0 and 40 mg/kg) of coenzyme Q_{10} and two levels of L-carnitine (0 and 200 mg/kg) supplementation during the experiment. Treatments included of T_1: control group, basal diet without no additives, T_2: basal diet containing ractopamine, T_3: basal diet containing coenzyme Q_{10}, T_4: basal diet containing L-carnitine, T_5: basal diet containing ractopamine + coenzyme Q_{10}, T_6: basal diet containing ractopamine + L-carnitine, T_7: basal diet containing coenzyme Q_{10}+L-carnitine, T_8: basal diet containing ractopamine + coenzyme Q_{10} + L-carnitine. The composition and nutrient levels of the starter, grower and finisher diets are presented in Table 1. Small amount of the diets was first mixed with the respective amount of ractopamine, coenzyme Q_{10} and L-carnitine as small as batch, then with a larger amount of the basal diets, so that the total amount of the respective diets was homogeneously mixed.

Management: The birds were housed in an environmentally controlled room. The room temperature was set at 32°C on day one which was lowered in a step wise manner to 23-24°C for the rest of experiment. Food and fresh water was offered to birds *ad libitum*. Humidity was held at 65%. The lighting Schedule was provided 23 hours light per day.

Hematological Data Collection: At day 42 of age, the blood samples were collected from wing vein of two birds in each replicate for measurement of hematological parameters. Measurements were included of: hemoglobin (Hb), packed cell volume (PCV), white blood cell count (WBC), red blood cell count (RBC) and the absolute numbers of each leukocyte type. A number of sterile test tubes containing anticoagulant (sodium citrate) were taken. PCV was determined by microhematocrit tube method. Hemoglobin concentration was measured using the cyanme themoglobin method. Values of mean corpuscular volume (MCV), mean corpuscular hemoglobin concentration (MCHC) and mean corpuscular hemoglobin (MCH) were calculated with the following formulas.

Table 1: Composition and nutrient levels of the starter, grower and finisher diets (%)

Ingredients	Starter (1-10)	Grower (11-24)	Finisher (25-42)
Corn	52.28	51.80	59.00
Soybean meal (44%)	36.37	34.90	28.20
Soybean oil	3.90	5.07	4.60
Fish meal	3.07	5.00	5.00
Dicalcium phosphate	2.09	1.19	1.10
Oyster shell	0.77	0.89	0.91
Vitamin/mineral premix	0.60	0.50	0.50
Salt	0.20	0.17	0.20
DL- methionine	0.33	0.24	0.22
L-Lysine	0.21	0.11	0.13
L- Threonine	0.07	0.01	0.04
$NaHCO_3$	0.11	0.12	0.10
Calculated nutrient content (%)			
Ingredients	Starter (1-10)	Grower (11-24)	Finisher (25-42)
ME (kcal/kg)	3025	3150	3198
Crude protein	22.4	22.4	20.0
Calcium	1.00	0.90	0.87
Available phosphorus	0.50	0.45	0.43
Methionine	0.68	0.57	0.52
Methionine+cystine	1.03	0.95	0.87
Lysine	1.41	1.34	1.20
Threonine	0.94	0.88	0.82

$$MCV(fl) = \frac{PCV(\%) \times 10}{RBC(10^6/ul)}$$

$$MCH(pg) = \frac{Hb(g/dl) \times 10}{RBC(10^6/ul)}$$

$$MCHC = \frac{Hb(g/dl) \times 10}{PCV(\%)}$$

Statistical Analysis: The statistical analysis was performed with SPSS 20 for windows. Anova GLM (general linear procedure) and Duncan's Multiple Range test were used. Values of $P<0.05$ were considered as significant. All values in Table have been reported as mean ± standard deviation (SD).

RESULTS AND DISCUSSION

The effect of ractopamine, coenzyme Q_{10} and L-carnitine on hematological parameters are presented in Table 2.

Treatments had no significant effect on hemoglobin, RBC, MCV, MCH and MCHC. This finding agreed with the report of Akbariazad *et al*. [10] who showed no

Table 2: Effects of supplemental dietary ractopamine, coenzymeQ_{10} and L-carnitine on hematological parameters of broiler chickens. (mean ± standard deviation)

Parameter			Hemoglobin (g/dl)	PCV[1](%)	RBC[2] (10^6/il)	MCV[3] (fl)	MCH[4] (pg)	MCHC[5] (%)
Ractopamine (mg/kg)								
0			10.9±1.1	26±3	2.6±0.49	101.8±14.5	43±8.2	42.5±9.6
10			11.2±1.3	28±4.4	2.8±0.6	100.4±14.8	41±9.6	41.2±6
Coenzyme Q_{10}(mg/kg)								
0			11±1.8	27.7±4.3	2.7±0.6	103.6±12	42±9.7	40.8±6.3
40			11.1±1.6	26.1±3.2	2.7±0.6	98.6±16.5	42±8.7	42.8±7.5
L-caraitine (mg/kg)								
0			10.8±1.1	28±4.3	2.8±0.6	102.8±13.3	40±8	39.5±5.3
200			11.3±1.6	26±3.1	2.66±0.48	99.4±15.7	43±9.6	44±7.7
Interactions								
R × Q_{10} × L- C								
0	0	0	10.4±0.5	26.9±4.3[ab]	2.57±0.6	106.2±9.3	42±9.6	39.11±5.5
10	0	0	11.25±1.3	31±5.8[a]	3.1±0.8	102.9±11.3	39±12.6	38.5±8.8
0	40	0	11±1.4	26±1.7[ab]	2.8±0.5	95.5±16.2	39±3.3	41.4±3.2
0	0	200	10.75±0.5	27±2.4[ab]	2.67±0.1	101.6±12.1	40±1.7	39.8±4.2
10	40	0	10.5±1.2	27±4.4[ab]	2.63±0.7	106.7 ±17.3	41±7	38.8±4
10	0	200	11.6±1/8	25.5±3.5[ab]	2.5±0.5	103.6±18.5	48±12.6	45.8±5.6
0	40	200	11.5±2.8	23.4±2.5[b]	2.3±0.5	104±21.7	50±11.2	49.5±12
10	40	200	11.4±0.95	27.5±3.1[ab]	3.1±0.36	88/3±7.5	36±1.4	41.5±3

Values with different superscripts in the same column are significantly different($p<0.05$).[1]PCV, Packed cell volume; [2]RBC, Red blood cell; [3]MCV, Mean corpuscular volume; [4] MCH,Mean corpuscular hemoglobin; [5]MCHC, Mean corpuscular hemoglobin concentration.

significant effect of L-carnitine at levels of 125 and 250 mg/kg on hemoglobin, RBC, MCV, MCH and MCHC. In contrast, Kardeniz *et al*. [11] showed significant increase in RBC, hemoglobin, PCV and MCHC of broilers, when L-carnitine was added to diet at level of 100 mg/kg. In agreement with our finding, Hoshi *et al*. [12] showed that the use of ractopamine in pregnant sows had no significant effect on blood's hemoglobin. Difference in broiler sex, level of L-carnitine and diet compositions justifies differences in results obtained in our experiment with others.

There was no significant interaction between feed additives used in this study, except for PCV. Packed cell volume of broilers fed diet containing 40 mg coenzyme Q_{10} per kg + 200 mg L-carnitine per kg (T7) were significantly lower than broiler fed with ractopamine (T2). This result agreed with Geng *et al*. [13, 14], who reported that PCV was not influenced by coenzyme Q_{10} alone, but significantly decreased by L-carnitine + coenzyme Q_{10}. A higher PCV indicates higher blood viscosity, which is one of the causes of ascite [15]. The decrease of PCV by coenzyme Q_{10} + L-carnitine may be associated with their benefits for the integrity of bio membranes.

CONCLUSION

The addition of ractopamine, coenzyme Q_{10} and L-carnitine, individually or in combination, had no effect on hematological parameters. Dietary coenzyme Q_{10} + L-carnitine supplementation reduced packed cell volume, which is a positive effect as it relates to ascite.

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