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Manju

Assistant Professor, Department of Animal Nutrition, PGIVER, Jaipur, Rajasthan, India

CS Vaishnava

Retd. Professor, Department of Animal Nutrition, CVAS, Udaipur, Rajasthan, India

RK Khinchi

Assistant Professor, Department of Veterinary Medicine, CVAS, Udaipur, Rajasthan, India

Monika Karnani

Assistant Professor, Department of Animal Nutrition, PGIVER, Jaipur, Rajasthan, India

Padma Meel Senior Veterinary Officer,

Animal Husbandry, Udaipur, Rajasthan, India

Sheela Choudhary

Professor and Head, Department of Animal Nutrition, PGIVER, Jaipur, Rajasthan, India

Dharmendra Chharang

PhD Scholar, Department of Animal Nutrition, PGIVER, Jaipur, Rajasthan, India

Corresponding Author: Manju Assistant Professor Der

Assistant Professor, Department of Animal Nutrition, PGIVER, Jaipur, Rajasthan, India

Effect of utilization of *Moringa oleifera* seed meal with or without supplementation of acidifier on haematological parameters of broiler chicks

Manju, CS Vaishnava, RK Khinchi, Monika Karnani, Padma Meel, Sheela Choudhary and Dharmendra Chharang

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Abstract

The present study attempts to analyze the effect of utilization of Moringa oleifera seed meal (MOSM) with or without supplementation of acidifier on haematological parameters of broiler chicks. A feeding trial of 42 days was conducted for five iso-nitrogenous and iso-caloric rations having MOSM at 0, 1, 2, 3 and 4% level with or without supplementation of acidifier using 300 broiler chicks in 5X2 factorial design. Broiler chicks were assigned randomly to various experimental groups. The dietary treatments were basal diet as control without MOSM (T1), basal diet with MOSM at 1% (T2), 2% (T3), 3% (T4), 4% (T5), basal diet with Acidifier at 1g/kg feed (T6), basal diet with MOSM at 1, 2, 3 and 4% with Acidifier at 1g/kg feed for T7, T8, T9 and T10, respectively. Blood samples for haematological analysis were collected aseptically from randomly selected birds (two birds per replicate) at 21 and 42 day of experiment. Results showed that highly significant (p < 0.01) effect of MOSM in diets of broiler chicks on Hb, RBC, WBC and PCV at 21 day of age as well as 42 day of age in broilers were observed. The effect of supplementation of acidifier as feed additive in ration of broiler chicks were found to be highly significant (p < 0.01) on Hb, RBC, WBC and PCV at 21 day of age. Whereas, at 42 day of age, highly significant (p < 0.01) effect of acidifier in the ratio of broilers on Hb, RBC and PCV and significant (p < 0.05) effect on WBC were observed. It was concluded that MOSM with or without acidifier can be incorporated in the diet of the broiler chicks without any harmful effect on the health status of the broiler chicks.

Keywords: Acidifier, broiler, Moringa oleifera seed meal, haematological parameters

1. Introduction

In many tropical and subtropical countries, various parts of moringa (leaves, fruits, immature pods, flower and seeds) are incorporated into the traditional food of humans (Siddhuraju and Becke, 2003; Anhawange *et al.*, 2004) ^[18, 4]. Researches were conducted to study the effect of moringa leaf meal on the growth performance of layer chicks (Melesse *et al.*, 2011) ^[15], on the productive performance of laying hens (Kakengi *et al.*, 2007; and Abou-Elezz *et al.*, 2011) ^[13]. ^{2]}, on broiler performance (Olugbemi *et al.*, 2010) ^[17]. The effect of Moringa seed was also examined by researchers (Abbas and Ahmed, 2012; Hassan Kaleem *et al.*, 2017) ^[1, 11]. On other hand, several substances as feed additive have been investigated in recent years with the aim of finding alternatives to growth promoting antimicrobials that are able to support productive performance and prevent the incidence of some diseases in poultry (Huyghebaert *et al.*, 2011) ^[12]. Among such substances organic acids band their salts have received much attention because they have shown not only antimicrobial activity, but several additional effects that go beyond those of antibiotics and could improve productivity in poultry (Dibner and Buttin, 2002)^[9].

This study therefore consider the utilization of varying levels of *Moringa oleifera* seed meal and acidifier on the haematological parameters of broilers chickens.

2. Material and methods

The present investigation was conducted at Poultry farm and Department of Animal Nutrition of College of Veterinary and Animal Science, Navania, Vallabhnagar, Udaipur, Rajasthan University of Veterinary and Animal Sciences, Bikaner.

2.1 Experimental design

Three hundred, day old Vencobb broiler chicks of either sex individually weighed and randomly divided into ten groups of 30 chicks each having similar average body weight. Each group of 30 chicks was further subdivided randomly into 2 replicates of 15 chicks in a 5 x 2 factorial arrangement. Deep litter system of housing was adopted for the feeding trial in the present study with an objective to provide maximum comfort to the broilers. The pens were thoroughly cleaned and disinfected before starting of experiment. All the chicks were maintained under standard managemental regimen of brooding and lighting. Proper ventilation and biosecurity measures were ensured throughout the trial. Ad libitum clean and fresh water was provided throughout the trial. Fresh and dry wheat straw was used as bedding material.

The various feed ingredients for computing different experimental rations were purchased from the local market in one lot before starting the experiment and were analyzed for proximate composition according to AOAC (2005) ^[5]. *Moringa oliefera* seeds were collected from local market and sorted out manually before soaking in water for overnight and rinsed with water in next morning till free from the foamy water. Then Moringa seeds were dried and ground in a

domestic blender to obtain Moringa oliefera seed meal (MOSM) which was stored for subsequent inclusion into diet mixtures. Ingredient-wise compositions of experimental diets are presented in Table 1. Commercial acidifier AlvipH manufactured by Alivira Health Care a combination of Formic Acid, Propionic Acid, Benzoic Acid, Butyric Acid and Orthophosphoric Acid was included in the ration at 100 g per 100 kg of feed. The dietary treatments were basal diet as negative control (T1), basal diet mixed with graded level of MOSM @ 1% without acidifier (T2), 2% MOSM without acidifier (T3), 3% MOSM without acidifier (T4), 4% MOSM without acidifier (T5), basal diet with acidifier (T6), 1% MOSM with acidifier (T7), 2% MOSM with acidifier (T8), 3% MOSM with acidifier (T9) and 4% MOSM with acidifier (T10), respectively. The rations formulated for various treatment groups for starter and finisher chicks were made iso-caloric and iso-nitrogenous (Table 2 and 3). The broiler starter and broiler finisher rations were formulated as per the BIS (2007)^[6]. Experimental starter rations were offered up to 3 weeks of age and thereafter experimental finisher rations were offered up to 6 weeks of age with and without supplementation of acidifier to respective groups.

C No	Ingredients	Starter ration (0-3 weeks)				Finisher ration (4-6 weeks)					
5. 110.		0	1	2	3	4	0	1	2	3	4
1.	Moringa Seeds	0	1	2	3	4	0	1	2	3	4
2.	Maize	45	45	45	45	45	55	55	55	55	55
3.	Deoiled ricebran	15	15	15	15	15	11	11	11	11	11
4.	Soyabean meal	18	18	18	18	18	13	13	13	13	13
5.	Groundnut cake	12	11	10	9	8	12	11	10	9	8
6.	Fish meal	8	8	8	8	8	7	7	7	7	7
7.	Mineral- mixture.	2	2	2	2	2	2	2	2	2	2
8.	Groundnut oil (ml)	0.5	0.4	0.3	0.2	0	0.5	0.5	0.4	0.3	0.3

 Table 1: Ingredient composition of experimental rations (kg/100 kg feed)

 Table 2: Nutrient composition of experimental starter ration (on per cent DM basis)

Treatments	0	1	2	3	4
DM	89	89.01	89.03	89.04	89.06
OM	91.43	91.45	91.47	91.49	91.51
CP	22.81	22.83	22.85	22.87	22.89
EE	3.01	3.15	3.29	3.43	3.57
CF	6.11	6.16	6.22	6.27	6.32
TA	6.57	6.55	6.53	6.51	6.49
NFE	50.5	50.32	50.15	49.97	49.8
ME (kcal/kg)	2924.91	2921.79	2918.67	2915.56	2903.49

DM-dry matter, OM-organic matter, CP-crude protein, EEether extract, CF-crude fiber, TA-total ash, NFE-nitrogen free extract, ME-metabolizable energy.

 Table 3: Nutrient composition of experimental finisher ration (on per cent DM basis)

Treatments	0	1	2	3	4
DM	89.2	89.22	89.23	89.25	89.26
OM	92.23	92.25	92.27	92.29	92.31
СР	20.37	20.39	20.41	20.43	20.45
EE	3.26	3.4	3.54	3.68	3.82
CF	5.21	5.26	5.31	5.37	5.42
TA	5.77	5.75	5.73	5.71	5.69
NFE	54.6	54.42	54.25	54.07	53.9
ME (kcal/kg)	3000.42	3006.25	3003.13	3000.02	3005.85

DM-dry matter, OM-organic matter, CP-crude protein, EEether extract, CF-crude fiber, TA-total ash, NFE-nitrogen free extract, ME-metabolizable energy

At 21 day and 42 day, blood was collected from randomly selected chicks (four chicks from each treatment) for haematological parameters.

2.2 Haematological parameters

About 3 ml blood samples were collected aseptically from wing vein of four randomly selected birds from each treatment group at 21 day and 42 day of age for the estimation of different haematological parameters. The blood was transferred into Ethylene Diamine Tetra Acetic acid (EDTA) containing vacutainer tubes. These tubes were subjected to determine Haemoglobin (Hb) concentration, Red Blood Cell (RBC), White Blood Cell (WBC) and Packed Cell Volume (PCV). Heamoglobin was estimated by using Sahli's acid hematin method. Erythrocyte (RBC) and leukocytes (WBC) counts were determined using the haemocytometer. PCV was measured by a standard manual technique using Wintrobe tubes centrifuged at 3000 rpm for 30 minutes.

2.3 Statistical analysis

Data collected during the present investigation were subjected to statistical analysis by adopting appropriate methods of analysis of variance as described by Snedecor and Cochran, 2004. The significance of mean differences were tested by Duncan's New Multiple Range Test (Duncan's Range Test) as modified by Kramer (1957)^[14].

3. Results and discussion

The effect of utilization of MOSM with or without

supplementation of acidifier on hematological parameters of broiler chicks has been presented in Table 4 and 5.

 Table 4: The effect of graded level of MOSM with or without

 supplementation of acidifier on blood parameters at 21 day of age

Treatmonte	Sampling at 21 day of age								
Treatments	Hb(gm%)	RBCx10 ⁶ /µl	WBC x10 ³ /cumm	PCV %					
MOSM%									
0%	9.96 ^a	3.43 ^a	23.03ª	26.63 ^a					
1%	11.06 ^b	3.95 ^b	24.01 ^b	26.99 ^a					
2%	12.2 ^c	4.15 ^{bc}	24.33 ^{bc}	30.08 ^b					
3%	12.63 ^c	3.83 ^{ab}	24.81°	31.25 ^b					
4%	13.7 ^d	4.47 ^c	28.77 ^d	33.08 ^c					
	Acidifier								
Without Acidifier	11.11 ^a	3.7 ^a	24.48 ^a	28.38 ^a					
With Acidifier	12.72 ^b	4.24 ^b	25.52 ^b	30.84 ^b					
SEM	0.25	0.08	0.42	0.53					
Probabilities									
MOSM	S**	S**	S**	S**					
Acidifier	S**	S**	S**	S**					

A, b, c, d, e, f and g means in each column, within each item, bearing the same superscripts are not significantly different (P>0.05), S- significant at p<0.05; S**-significant at p<0.01; NS-non-significant, SEM= Standard error of mean

Table 5: The effect of graded level of MOSM with or without supplementation of acidifier on blood parameters at 42 day of age

	Sampling at 42 day of age						
	Hb(gm%)	RBCx10 ⁶ /µl	WBC x10 ³ /cumm	PCV %			
		MOSM%					
0%	10.5 ^a	3.76 ^a	25.30a	29.0 ^a			
1%	11.64 ^b	3.99 ^a	25.96a	30.28 ^b			
2%	12.40 ^c	4.34 ^b	26.73b	31.03 ^{bc}			
3%	13.02 ^d	4.33 ^b	27.41b	32.08 ^c			
4%	13.73 ^e	4.53 ^b	31.85c	34.8 ^d			
		Acidifier					
Without Acidifier	11.34 ^a	4.05 ^a	26.67 ^a	31.0 ^a			
With Acidifier	13.18 ^b	4.34 ^b	28.23 ^b	31.88 ^b			
SEM	0.25	0.08	0.24	0.07			
Probabilities							
MOSM	S**	S**	S**	S**			
Acidifier	S**	S**	S*	S**			

A, b, c, d, e, f and g means in each column, within each item, bearing the same superscripts are not significantly different (P>0.05), S- significant at p<0.05; S**-significant at p<0.01, SEM= Standard error of mean

Highly significant (p<0.01) effect of *Moringa oleifera* seed meal in diets of broiler chicks on Hb, RBC, WBC and PCV at 21 day of age as well as 42 day of age in broilers were observed. The effect of supplementation of acidifier as feed additive in ration of broiler chicks in respect to haematological parameters, were found to be highly significant (p<0.01) on Hb, RBC, WBC and PCV at 21 day of age. Whereas, at 42 day of age, highly significant (p<0.01) effect of acidifier in the ration of broilers on Hb, RBC and PCV and significant (p<0.05) effect on WBC were observed. Findings from this study have shown that there are significant differences in the sum of the means of the Hb, RBC, WBC and PCV across the groups (Figure 1(I), 1(II), 2(I), 2(II), 3(I), 3(II), 4(I) and 4(II)).

 Table 6: The effect of graded level of MOSM with or without

 supplementation of acidifier on blood parameters at 21 and 42 day of age

	Blood	parameters	Mean	SEM	t value	Level of significance
Dain 1	Hb	At 21 day	11.91	0.253	-3.37	.000
Pair I		At 42 day	12.25	0.241		
Pair 2	RBC	At 21 day	3.96	0.085	-4.22	.000
		At 42 day	4.19	0.065		
Pair 3	WBC	At 21 day	24.99	0.416	-5.62	.001
		At 42 day	27.45	0.456		
Pair 4	PCV	At 21 day	29.60	0.531	-6.28	000
		At 42 day	31.43	0.432		.000

SEM-Standard error of mean

Further, the results of analysis of data by paired t test showed that significant differences were found in all blood parameters in terms of Hb, RBC, WBC and PCV in treatment groups.



Fig 1(I): Effect of MOSM on Hb



Fig 1(II): Effect of Acidifier on Hb



Fig 2(I): Effect of MOSM on RBC



Fig 2(II): Effect of Acidifier on RBC



Fig 3(I): Effect of MOSM on WBC



Fig 3(II): Effect of Acidifier on WBC



Fig 4(I): Effect of MOSM on PCV



Fig 4(II): Effect of Acidifier on PCV

Red blood cells are responsible for the transportation of oxygen and carbon dioxide in the blood, hence higher values indicates a greater potential for these function and a better state of health (Nouala et al., 2006)^[16]. According to Brown (2010) [7] increased RBC values are associated with high quality dietary protein and with disease free animals. An increased PCV shows a better transportation of oxygen and nutrients. According to Chineke (2006)^[8], high PCV readings shows an increase in the number of RBC or reduction in circulating plasma volume. The increase in WBCs may be as a result of the ability of the plant to cause some degree of improvement in immunity. Inclusion of Moringa oleifera in the diet of broiler chicks resulted in increase in RBC, PCV and Hb values confirming the findings of Fuglie (2009) ^[10] who reported that Moringa oleifera has a blood boosting effect because of the high protein content. The comparable WBC of the birds suggests that the animals were healthy because a decrease in number of WBC below the normal range is an indication of allergic condition, anaphylactic shock and certain parasitism or presence of foreign body in circulating system Ahamefule et al. (2008)^[3].

4. Conclusion

The results obtained in the present study shows that the haematological parameters in term of Hb, RBC, WBC and PCV were improved in the broiler chicks fed MOSM supplementation with or without acidifier in the diet as compared to the control group. On the basis of the results, the conclusion was made that MOSM with or without supplementation of acidifier can be included in the diet of broiler chicks without any deleterious effects on health status of broiler chicks.

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