



*Research article*

## **Effect of *Moringa oleifera* seeds powder on performance and immunity of broiler chicks**

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**Abstract:** This study aims to investigate the effect of feeding different dietary levels of *Moringa oleifera* seeds powder (MOSP) and phytase (500 FTY/kg diet) on some productive performance, immune response, and blood constituents of broiler chicks. A total of 240, 1-day old unsexed Cobb 500 chicks were individually weighed and randomly were distributed into 6 equal groups. Each group had 40 birds in 5 replicates of 8 chicks. Groups were allocated to different six experimental diets; 1- A control basal, 2- A control plus phytase enzyme, 3- A control plus 0.75% MOSP, 4- A control plus 0.75% MOSP and phytase, 5- A control plus 1% MOSP and 6- A control plus 1% MOSP with phytase). The growth trial lasted for 38 days. The results show that birds fed MOSP at 0.75% with phytase had significantly the highest values of live body weight gain, lowest feed intake and best feed conversion ratio. Blood serum of total cholesterol and LDL were lower in chicken fed 0.75% MOSP (with and without) enzyme compared to the control and the other dietary levels of MOSP. Values of AST were lower in broilers fed MOSP at 0.75% with enzyme and those at 1% without enzyme, compared to control and other dietary treatments. However, no significant differences were observed in the ALT values between dietary treatments. A better antibody titer against Newcastle disease and immune response was observed in broilers received MOSP at 0.75% with phytase supplementation. It could be concluded that using MOSP at 0.75% gave better values of growth, immune organs, blood constituents, and the general health of broilers.

**Keywords:** *Moringa oleifera* seeds; immunity; newcastle disease; growth performance; broiler; blood

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**Abbreviations:** MOSP: *moringa oleifera* seed powder; LDL: low-density lipoprotein; AST: aspartate transaminase; ALT: alanine aminotransferase

## 1. Introduction

*Moringa oleifera* can play an important role in the economy of the poultry industry as a good source of vitamins and amino acids. Makkar *et al.*, Compaore *et al.*, and Abbas stated also that *Moringa oleifera* seeds are a good source of fats, proteins, and minerals [1–3]. It is an excellent source of proteins for monogastric animals [4]. Anwar and Rashid published that *Moringa oleifera* seeds contained 31.65% crude protein and 7.54% fiber on basis of dry matter [5]. Abbas and Ahmed reported an improvement in growth performance of broiler chicks fed diets fortified with *Moringa oleifera* seed powder (MOSP) at 0.37% of the diet [6]. Ochi *et al.*, found that MOSP at levels of 0.5, 1 and 2% in the diet of the broilers enhanced their live body weights and weight gains [7]. Ferreira *et al.*, [4] reported that growth performance of broiler chicks fed a diet supplemented with 50 g/kg of MOSP was improved compared to their counterparts of the control group. Furthermore, dietary supplementation with *Moringa oleifera* was found to enhance the oxidative stability of chicken meat [8]. Furthermore, seed extracts of *Moringa oleifera* have been reported to possess antimicrobial properties [9,10]. Gaia added that *Moringa oleifera* is effectual in reducing the activity of pathogenic bacteria and molds and improving the digestibility of nutrients, thus helping the chickens to express their natural genetic potential ties [11]. Walter *et al.*, reported that extract of *Moringa oleifera* seeds created inhibitory effects on *Salmonellatyphii* and *Escherichia coli*, as the extract may contain antibiotic metabolites, such as carboxylic acid, 2,4-diacetyl phloroglucinol and, cell wall-degrading enzymes and chitinases that improve the health of broilers [12]. Yang *et al.*, suggested that *Moringa oleifera* is a potential plant that could be used to enhance immune response and to improve the intestinal health of broiler chicken [13]. Chollom *et al.*, demonstrated that *M. oleifera* seed extract has nutritional value as well as strong antiviral activity against Newcastle disease virus (NDV) [14]. This study aimed at studying the performance and immune response of broilers as affected by providing MOSP and phytase enzyme in diets.

## 2. Materials and methods

### 2.1. Processing of *Moringa oleifera* seeds

*Moringa oleifera* seeds were purchased from the local market and cleaned before grinding to powder by using a grinding machine. Then, *Moringa oleifera* seeds powder (MOSP) was added to the basal diet.

### 2.2. Experimental house management

The broiler chicken house was thoroughly cleaned with water and then disinfected with formalin (NTK, Polokwane). All drinkers and feeders were thoroughly cleaned and disinfected before use. The house was left empty for two weeks after cleaning to break the life cycle of any disease-causing organisms that were not killed by the disinfectant. Heating equipment turned on 24 hours before the arrival of the chicks. The chicks housed in galvanized metal battery cages during the

experimental period from 0 to 38 days of age. The temperature was 34 °C in the first week and decreased gradually up to 24 °C in the third week and then all chicks kept under the same managerial conditions.

### 2.3. *Experimental chicks and design*

The total number of used chicks was 240 unsexed of Cobb 500 broiler chicks' day-old. Chicks were distributed randomly into six treatment groups, each treatment group containing 40 birds in five replicates with 8 birds, each. Feed and water were provided freely throughout the experiment and light was provided continuously.

### 2.4. *Experimental diets*

Birds were fed-on 23% crude protein (CP) and 3000 kcal ME/kg. Table 1 shows the composition and calculated analysis of experimental diets. Dietary treatments were assigned to contain (0 or control, 0.75%, and 1% MOSP with and without phytase enzyme), the composition of diets done according to the guide-book for Cobb 500 strain Broiler Performance and Nutrition Supplement [15].

### 2.5. *Chemical analysis of Moringa seeds*

Chemical composition of the *M. oleifera* seeds including moisture and ash contents of MOSP were determined by the Codex-adopted AOAC method [16], crude protein was determined by micro Kjeldahl method AOAC [17], crude lipids were determined according to the method described by AOAC [18], crude fiber was determined by AOAC [17] and ANKOM Technology and total carbohydrate were determined by operating instruction manual for isoperibol bomb calorimeter (Parr 1261) 1997. Amino acids determination was done according to AOAC [17].

### 2.6. *Performance parameters*

#### 2.6.1. Body weight

The average group body weights were recorded at weekly intervals during the experimental period.

#### 2.6.2. Feed intake (g)

The feed was weighed every week to determine the average feed intake per chick for the different treatment groups. Feed intake was calculated by the remained feed and divided by the number of birds in each group per day and totalized to be per week.

### 2.6.3. Weight gain (g)

The birds were weighed every week to determine the average weight gain per chick for the different treatment groups. Weight gain was calculated as the difference between two successive weekly body weights as follows:

$$\text{Weight Gain} = \text{Final weight} - \text{Initial weight} \quad (1)$$

### 2.6.4. Feed conversion ratio

The feed conversion ratio was calculated as the total amount of feed consumed divided by the weight gain of the birds in the pen.

**Table 1.** Composition and calculated chemical analysis of experimental diets.

Ingredients %	Control	0.75%	1%
Yellow corn (7.5%)	55.000	54.258	54.008
Soybean meal (43%)	30.300	30.300	30.300
Corn gluten meal (60%)	8.602	8.600	8.600
Vegetable oil	1.566	1.566	1.566
Di-calcium phosphate	1.892	1.872	1.866
Limestone	1.189	1.200	1.204
D-L. methionine	0.172	0.173	0.174
L-lysine (HCl)	0.396	0.398	0.399
Salt	0.400	0.400	0.400
Vit. & min. Mixture <sup>1</sup>	0.400	0.400	0.400
Choline chloride	0.083	0.083	0.083
MOSP	0.000	0.750	1.000
Calculated analysis			
CP (%)	22.890	22.837	22.819
ME (kcal/kg)	3000.517	2975.717	2967.464
L-lysine (%)	1.360	1.360	1.360
D-L Methionine (%)	0.585	0.585	0.585
TSAA (%)	0.980	0.980	0.980
Calcium (%)	0.960	0.960	0.960
Available P (%)	0.450	0.450	0.445

<sup>1</sup> Vitamins-minerals mixture provided per kg of diet: vitamin A, 12000 I.U; vitamin D3, 2010 I.U; vitamin E, 12 mg; vitamin K3, 2.5 mg; vitamin B2, 5 mg; vitamin B6, 1.7 mg; vitamin B1, 1.2 mg; vitamin B12, 11 µg; Biotine, 51 µg; B complexe, 10mg; Niacine, 30 mg; Manganèse, 60 mg; Zinc, 50 mg; Iron, 30 mg; Folié Acid, 1 mg; Copper, 10.2 mg; Idoine, 1.1 mg; Sélénium, 0.11 mg and Cobalt, 0.1 mg.

<sup>2</sup> Composition of diets was done according to guide-book for Cobb 500 strain Broiler Performance and Nutrition Supplement [15].

## 2.7. Slaughter test and blood constituents

At the end of the experiment (38 days), ten birds were slaughtered from each treatment to obtain the carcass, giblets (gizzard, liver, and heart), and the lymphoid organs were separately weighed.

Blood samples were taken from 5 birds per replicate to determine WBC's count and their fractions. The hemagglutination inhibition test performed [19] to examine the effects of different MOSP levels on antibody titers against Newcastle disease. The rest of the birds 5 per replicate selected for blood collection. Five ml of blood was collected with an anticoagulant and then centrifuged at 1500 rpm for 15 minutes to get the serum, which determines serum content of total protein, albumin, globulin, cholesterol, and liver enzymatic activity (AST and ALT) using commercial kits.

### 2.8. Carcass characteristics

The birds slaughtered on 38<sup>th</sup> day after the withdrawal of feed 12 hours before slaughter. Birds live body weights (LBW) recorded. The birds slaughtered by cutting the jugular vein, and after complete bleeding, the weights were recorded. The birds were then de-feathered and eviscerated. The respective bird's weight and organs were measured and expressed as a percentage of live body weight.

### 2.9. Immune organs

Evaluated the effect of dietary MOSP supplementation on some immune organs of broiler chicks was by measuring the weights of immune organs, including thymus and bursa as the primary immune organs, and the secondary immune organs being the spleen and liver.

### 2.10. Statistical analyses

Obtained data were statistically analyzed as a two-way analysis of variance, using the General Linear Model procedure described in SAS users guide (SAS, 1990) [20], according to the following model:

$$Y_{ijk} = \mu + D_i + E_j + DE_{ij} + \varepsilon_{ijk}$$

Where:

$Y_{ijk}$  = is the value of the respective variable

$\mu$  = is the overall mean of the respective variable

$D_i$  = is the effect due to the  $i^{\text{th}}$  diets,  $i = 1, 2, 3$  (1= basal diet, 2= 0.75% MOSP and 3= 1% MOSP)

$E_j$  = is the effect of the  $j^{\text{th}}$  enzyme ( $j = 0, 500$  FTU)

$DE_{ij}$  = is the effect of interaction.

$\varepsilon_{ijk}$  = is a random error associated with the  $ijk^{\text{th}}$  observation.

Differences between treatment means were tested using Duncan's multiple range test (Duncan's, 1955) [21]. Differences were considered statistically significant if  $P < 0.05$ .

## 3. Results

### 3.1. Chemical composition of MOSP

MOSP was found to contain crude protein 27.3%, crude fiber 17.2%, moisture 6.4%, ash 3.2%, NDF 29.6%, ADF 26.3%, ADL 14.9%, crude lipid 25.66% and gross energy 5718 cal/g (Table 2).

The amino acid content was determined (Table 2). The data illustrated that MOSP contains reasonable values of Arginine (3.36%), leucine (1.36%) and, valine (1.11%) and lower levels of lysine (0.49%) and threonine (0.65%).

**Table 2.** Proximate chemical analysis and amino acid content of *Moringa oleifera* seed powder (MOSP).

<sup>1</sup> Amino Acid				<sup>2</sup> Proximate analysis	
EAA	%	NEAA	%	Item	%
Isoleucine	0.75	Aspartic	1.21	Crude protein	27.3
Leucine	1.36	Serine	0.75	Crude lipid	25.66
Phenylalanine	1.06	Glutamic	5.26	Crude fiber	17.2
Histidine	0.57	Proline	1.43	Moisture	6.4
Threonine	0.65	Glycine	1.08	Ash	3.2
Lysine	0.49	Alanine	1.05	Gross energy	5718
Valine	1.11	Cysteine	1.03	NDF	29.6
Argenine	3.36	Tyrosine	0.60	ADF	26.3
Methionine	0.53			ADL	14.9
				Phosphorus	0.51

<sup>1</sup> EAA, essential amino acid; NEAA, nonessential amino acid.

<sup>2</sup> NDF, nitrogen detergent fibre; ADF, acid detergent fibre; ADL, acid detergent lignin.

### 3.2. Growth performance

Body weight (BW), body weight gain (BWG), and feed conversion ratio (FCR) are shown in (Table 3, 4, and 5). Birds fed MOSP at 0.75% with enzyme had significantly higher body weight (BW) than control with enzyme and other dietary treatments either after 14 or 28 days of age; however, at 38 days of age, it was superior to other treatments except for the control without enzyme supplementation. The positive results of 0.75% MOSP with phytase enzyme indicate the positive effect of phytase that worked on the breakdown of phytates.

**Table 3.** Effect of dietary *Moringa oleifera* seed Powder (MOSP), enzyme supplementation and their interaction on growth performance (starter period).

Starter (1–14 d)				
Item	BW <sup>1</sup>	BWG	FI	FCR
MOSP effect (%)				
0	359.50 <sup>c</sup>	314.50 <sup>c</sup>	377.60 <sup>a</sup>	1.27 <sup>a</sup>
0.75	383.28 <sup>a</sup>	338.28 <sup>a</sup>	365.75 <sup>b</sup>	1.09 <sup>b</sup>
1%	362.50 <sup>b</sup>	317.50 <sup>b</sup>	361.43 <sup>b</sup>	1.13 <sup>b</sup>
SEM	3.03	3.03	4.20	0.06
P-value	≤0.01	≤0.01	0.02	≤0.01
Enzyme effect (FTU)				
0	372.66 <sup>a</sup>	327.66 <sup>a</sup>	373.16 <sup>a</sup>	1.140 <sup>a</sup>
500	378.52 <sup>a</sup>	333.52 <sup>a</sup>	363.36 <sup>b</sup>	1.08 <sup>b</sup>

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Starter (1–14 d)				
Item	BW <sup>1</sup>	BWG	FI	FCR
SEM	2.48	2.48	3.43	0.05
P-value	0.09	0.09	0.04	≤ 0.01
Interaction effect (MOSP X Enzyme)				
0 X 0	359.50 <sup>c</sup>	314.50 <sup>c</sup>	399.50 <sup>a</sup>	1.27 <sup>a</sup>
0 X 500	373.07 <sup>b</sup>	328.07 <sup>b</sup>	355.71 <sup>b</sup>	1.08 <sup>b</sup>
0.75 X 0	369.00 <sup>b</sup>	324.00 <sup>b</sup>	365.00 <sup>b</sup>	1.13 <sup>b</sup>
0.75 X 500	393.00 <sup>a</sup>	348.60 <sup>a</sup>	366.51 <sup>b</sup>	1.05 <sup>c</sup>
1 X 0	355.50 <sup>c</sup>	310.50 <sup>c</sup>	355.00 <sup>b</sup>	1.15 <sup>b</sup>
1 X 500	369.50 <sup>b</sup>	324.50 <sup>b</sup>	367.87 <sup>b</sup>	1.13 <sup>b</sup>
SEM	4.08	4.08	5.85	0.06
P-value	≤ 0.01	≤ 0.01	≤ 0.01	≤ 0.01

SEM = standard error of the mean. <sup>a,b,c</sup> Means within a row with different subscripts differ when  $P < 0.05$ .

<sup>1</sup> BW, body weight in g; BWG, body weight gain in g; FI, feed intake in g; FCR, feed conversion ratio (g feed/g weight gain).

**Table 4.** Effect of dietary *Moringa oleifera* seed Powder (MOSP), enzyme supplementation and their interaction on growth performance (grower period).

Grower (15–28 d)				
Item	BW	BWG	FI	FCR
MOSP effect (%)				
0	1315.00 <sup>c</sup>	955.50 <sup>b</sup>	1188.85 <sup>a</sup>	1.26 <sup>a</sup>
0.75	1387.42 <sup>a</sup>	1004.14 <sup>a</sup>	1171.50 <sup>a</sup>	1.18 <sup>c</sup>
1%	1263.75 <sup>c</sup>	901.25 <sup>c</sup>	1115.42 <sup>b</sup>	1.25 <sup>a</sup>
SEM	11.45	9.26	7.94	0.06
P-value	≤ 0.01	≤ 0.01	≤ 0.01	≤ 0.01
Enzyme effect (FTU)				
0	1318.50 <sup>b</sup>	945.83 <sup>b</sup>	1150.66 <sup>a</sup>	1.22 <sup>a</sup>
500	1345.78 <sup>a</sup>	967.26 <sup>a</sup>	1166.52 <sup>a</sup>	1.20 <sup>c</sup>
SEM	9.35	7.56	6.48	0.05
P-value	0.04	0.04	0.08	≤ 0.01
Interaction effect (MOSP X Enzyme)				
0 X 0	1315.00 <sup>c</sup>	955.50 <sup>b</sup>	1203.00 <sup>a</sup>	1.26 <sup>a</sup>
0 X 500	1359.86 <sup>b</sup>	986.79 <sup>a</sup>	1174.71 <sup>b</sup>	1.19 <sup>c</sup>
0.75 X 0	1356.00 <sup>c</sup>	937.00 <sup>b</sup>	1151.50 <sup>bc</sup>	1.23 <sup>b</sup>
0.75 X 500	1380.50 <sup>ab</sup>	991.50 <sup>a</sup>	1191.50 <sup>a</sup>	1.20 <sup>c</sup>
1 X 0	1234.50 <sup>d</sup>	879.00 <sup>c</sup>	1097.50 <sup>d</sup>	1.27 <sup>a</sup>
1 X 500	1293.00 <sup>c</sup>	923.50 <sup>b</sup>	1133.36 <sup>c</sup>	1.23 <sup>b</sup>
SEM	14.09	11.41	4.93	0.05
P-value	≤ 0.01	≤ 0.01	≤ 0.01	≤ 0.01

SEM=standard error of the mean.

<sup>a,b,c,d</sup> Means within a row with different subscripts differ when  $P < 0.05$ .

<sup>1</sup> BW, body weight in g; BWG, body weight gain in g; FI, feed intake in g; FCR, feed conversion ratio (g feed/g weight gain).

**Table 5.** Effect of dietary *Moringa oleifera* seed Powder (MOSP), enzyme supplementation and their interaction on growth performance (finisher period).

Finisher (29–38 d)				
Item	BW	BWG	FI	FCR
MOSP effect (%)				
0	2214.80 <sup>c</sup>	899.80 <sup>b</sup>	1570.45 <sup>a</sup>	1.78 <sup>a</sup>
0.75	2279.21 <sup>a</sup>	891.78 <sup>a</sup>	1549.40 <sup>a</sup>	1.74 <sup>b</sup>
1%	2191.56 <sup>b</sup>	884.44 <sup>a</sup>	1566.76 <sup>a</sup>	1.77 <sup>b</sup>
SEM	10.94	6.17	8.44	0.07
P-value	≤0.01	0.6	0.1	≤0.01
Enzyme effect (FTU)				
0	2206.10 <sup>b</sup>	860.31 <sup>b</sup>	1584.54 <sup>a</sup>	1.79 <sup>a</sup>
500	2277.26 <sup>a</sup>	916.63 <sup>a</sup>	1539.86 <sup>b</sup>	1.67 <sup>c</sup>
SEM	8.93	5.03	6.89	0.06
P-value	≤0.01	≤0.01	≤0.01	≤0.01
Interaction effect (MOSP X Enzyme)				
0 X 0	2214.79 <sup>c</sup>	899.79 <sup>b</sup>	1601.40 <sup>a</sup>	1.78 <sup>a</sup>
0 X 500	2216.64 <sup>c</sup>	856.78 <sup>c</sup>	1539.51 <sup>b</sup> <sup>c</sup>	1.80 <sup>a</sup>
0.75 X 0	2236.22 <sup>b</sup> <sup>c</sup>	880.22 <sup>b</sup>	1576.71 <sup>b</sup>	1.70 <sup>c</sup>
0.75 X 500	2292.32 <sup>b</sup>	911.82 <sup>a</sup>	1522.10 <sup>d</sup>	1.67 <sup>c</sup>
1 X 0	2217.68 <sup>c</sup>	896.43 <sup>b</sup>	1575.54 <sup>ab</sup>	1.76 <sup>b</sup>
1 X 500	2165.45 <sup>d</sup>	872.45 <sup>bc</sup>	1557.99 <sup>c</sup>	1.79 <sup>ab</sup>
SEM	14.67	8.11	11.02	0.02
P-value	≤0.01	≤0.05	≤0.01	≤0.01

SEM = standard error of the mean.

<sup>a,b,c,d</sup> Means within a row with different subscripts differ when  $P < 0.05$ .

<sup>1</sup> BW, body weight in g; BWG, body weight gain in g; FI, feed intake in g; FCR, feed conversion ratio (g feed/g weight gain).

Data in Table 3 indicate that the feed intake of broilers of all dietary treatments was significantly lower at 14 days of age compared to the control. At 28 days of age, both the control and birds fed 0.75% MOSP were significantly equal and consumed higher feed than other dietary treatments Table 4. At 38 days of age, broilers fed 0.75% MOSP with enzyme consumed less feed compared to control and other dietary treatments Table 5. With regard to the effect of MOSP on feed conversion ratio (FCR), it was observed that chicks fed MOSP at 0.75% plus phytase exhibited the best corresponding values either at 14, 28, or at 38 days of age (Tables 3, 4, and 5).

### 3.3. Blood constituents

The results of blood parameters at 38 days old as affected by dietary *Moringa oleifera* seed Powder (MOSP) are presented in Table 6. Blood examination has its tangible values in the poultry rearing business e.g., it provides information on the assessment of poultry health and nutritional deficiency, as well as physiological changes in the growth of broilers. It is well known that the hematological value has a significant impact on physiological changes and the health status of poultry. The results showed that the White Blood Cells (WBC) ranged from  $23.00 \pm 0.36$  to  $18.33 \pm 0.76$  with



statistical differences among the different dietary treatments. Higher WBC count was elevated in birds received 1% MOSP without enzyme and those of the control diets compared with the other dietary treatments.

**Table 6.** Effect of dietary *Moringa oleifera* seed powder (MOSP) on blood constituents of Cobb broilers at 38 days old.

Treatment parameter	0% MOSP without enzyme	0% MOSP with enzyme	0.75% MOSP without enzyme	0.75% MOSP with enzyme	1% MOSP without enzyme	1% MOSP with enzyme	SEM <sup>4</sup>
WBC <sup>1</sup> ( $\times 10^3$ /mL)	22.66 <sup>a</sup>	22.00 <sup>a</sup>	18.33 <sup>c</sup>	20.00 <sup>b</sup>	23.00 <sup>a</sup>	19.00 <sup>b</sup>	1.19
H %	57.33 <sup>a</sup>	56.00 <sup>a</sup>	46.00 <sup>d</sup>	49.33 <sup>c</sup>	51.00 <sup>b</sup>	54.00 <sup>ab</sup>	3.69
L%	44.00	46.00	45.66	47.00	45.00	43.33	4.37
H/L ratio	1.30	1.22	1.01	1.05	1.13	1.25	
T.Col (mg/dL)	103.76 <sup>b</sup>	98.70 <sup>bc</sup>	96.60 <sup>bc</sup>	91.83 <sup>c</sup>	110.63 <sup>a</sup>	109.90 <sup>a</sup>	5.81
HDL (mg/dL)	63.77 <sup>c</sup>	72.48 <sup>b</sup>	68.01 <sup>c</sup>	67.00 <sup>c</sup>	79.43 <sup>a</sup>	79.96 <sup>a</sup>	3.52
LDL (mg/dL)	18.29 <sup>b</sup>	19.12 <sup>b</sup>	18.54 <sup>b</sup>	13.61 <sup>c</sup>	15.36 <sup>bc</sup>	21.32 <sup>a</sup>	3.23
AST (u/L)	133.00 <sup>b</sup>	130.33 <sup>b</sup>	163.00 <sup>a</sup>	116.66 <sup>c</sup>	105.00 <sup>d</sup>	137.33 <sup>b</sup>	12.68
ALT (u/L)	17.26	16.33	15.86	18.20	16.33	16.80	1.86
ND <sup>2</sup>	4.00 <sup>c</sup>	4.00 <sup>c</sup>	5.30 <sup>b</sup>	9.00 <sup>a</sup>	5.00 <sup>b</sup>	5.00 <sup>b</sup>	0.63

SEM = standard error of the mean.

<sup>a,b,c</sup> Means within a row with different subscripts differ when  $P < 0.05$ .

<sup>1</sup> WBC, white blood cells; H, heterophils; L, lymphocytes; T.Col, total cholesterol; HDL, high-density lipoprotein.

<sup>2</sup> ND, newcastle disease.

The results indicated that heterophils were lower in number in broilers fed MOSP at 0.75% level either with or without enzyme, compared to control; this shows the health of birds fed on Moringa seeds at 0.75%, which is reflected on their immunity response (Table 6). There were no significant differences in the numbers of lymphocytes of birds fed on MOSP diets compared to the control (Table 6). Lower total cholesterol content was observed in chicken fed on 0.75% MOSP (with and without) enzyme when compared to the control and the other dietary levels of MOSP. On the other hand, the result showed that HDL content at all levels of MOSP supplementation was higher than the control (Table 6). Results in Table 6 indicated that values of AST varied significantly and the lower values were found in broilers fed MOSP at 0.75% with enzyme and those at 1% without enzymes, compared to control and other dietary treatments. However, no significant differences were observed in the ALT values between dietary treatments.

### 3.4. Immunity

Regarding the antibody titers against Newcastle disease, Significant differences were observed ( $P < 0.05$ ) among MOSP levels and control (Table 6). The results showed that 0.75% MOSP with enzyme recorded higher antibody titers against Newcastle disease, as well as other MOSP levels also have good rates and were higher than control.

### 3.5. Carcass characteristics

The effects of feeding different levels of *Moringa oleifera* seed powder on dressing percentage and the weights of edible and lymphoid organs are shown in Table 7. Results showed that MOSP at both levels of 0.75% and 1% with enzyme significantly improved the breast weight (%) compared to the control and other treatments. It is worthy to note that the percentage of gizzard weight in all dietary treatments fed on MOSP achieved significantly ( $P < 0.05$ ) the best values compared with the control. The development of the gizzard ensures complete grinding, a well-regulated digester flow, and secretion of digestive juices which helps broilers to grow well.

**Table 7.** Effect of dietary *Moringa oleifera* seed Powder (MOSP) on carcass characteristics and lymphoid organs of Cobb broiler at 38 days old.

Treatment parameter	0% MOSP without enzyme	0% MOSP with enzyme	0.75% MOSP without enzyme	0.75% MOSP with enzyme	1% MOSP without enzyme	1% MOSP with enzyme	SEM
Carcass characteristics							
Pre-slaughter (g)	2324.7	2330.3	2347.8	2410.9	2328.4	2283.1	1.76
Dressing %	73.97 <sup>bc</sup>	74.39 <sup>bc</sup>	75.40 <sup>a</sup>	73.96 <sup>bc</sup>	73.88 <sup>c</sup>	74.52 <sup>b</sup>	1.77
Breast %	21.38 <sup>c</sup>	21.68 <sup>c</sup>	23.73 <sup>a</sup>	22.46 <sup>b</sup>	21.46 <sup>c</sup>	23.91 <sup>a</sup>	1.75
Thigh %	28.56 <sup>ab</sup>	28.89 <sup>a</sup>	27.88 <sup>c</sup>	28.23 <sup>bc</sup>	28.60 <sup>b</sup>	28.06 <sup>c</sup>	1.20
Liver %	1.84 <sup>a</sup>	1.85 <sup>a</sup>	1.79 <sup>b</sup>	1.73 <sup>c</sup>	1.80 <sup>ab</sup>	1.84 <sup>ab</sup>	0.16
Gizzard %	1.87 <sup>d</sup>	1.99 <sup>c</sup>	2.08 <sup>b</sup>	2.05 <sup>bc</sup>	2.20 <sup>a</sup>	2.11 <sup>b</sup>	0.22
Heart %	0.53 <sup>d</sup>	0.58 <sup>bc</sup>	0.59 <sup>abc</sup>	0.57 <sup>c</sup>	0.60 <sup>ab</sup>	0.61 <sup>a</sup>	0.08
Fat %	0.95 <sup>a</sup>	0.93 <sup>a</sup>	0.80 <sup>b</sup>	0.62 <sup>c</sup>	0.94 <sup>a</sup>	0.73 <sup>bc</sup>	0.34
Lymphoid organs							
Bursa %	0.28 <sup>ab</sup>	0.27 <sup>ab</sup>	0.18 <sup>c</sup>	0.26 <sup>b</sup>	0.26 <sup>b</sup>	0.30 <sup>a</sup>	0.08
Spleen %	0.09 <sup>d</sup>	0.11 <sup>b</sup>	0.09 <sup>d</sup>	0.10 <sup>c</sup>	0.10 <sup>c</sup>	0.13 <sup>a</sup>	0.02
Thymus %	0.23 <sup>d</sup>	0.31 <sup>c</sup>	0.29 <sup>c</sup>	0.39 <sup>b</sup>	0.44 <sup>a</sup>	0.43 <sup>a</sup>	0.10

SEM = standard error of the mean.

<sup>a,b,c</sup> Means within a row with different subscripts differ when  $P < 0.05$ .

### 3.6. Mortality rate

The effect of dietary treatments on the mortality rate of chickens observed throughout the entire experiment. The absence of death cases among the birds received MOSP might be due to the antimicrobial and availability of vitamins, proteins, and minerals in *Moringa* that may increase immunity.

## 4. Discussion

### 4.1. Chemical composition of MOSP

These results for the chemical composition of MOSP are in agreement with Francis *et al.*, [22]. The major nonessential amino acids were found to be glutamic acid (5.26%), glycine (1.08%), and Alanine (1.05%), respectively. These results correspond to El-Massry *et al.*, [23] found that both arginine and glutamic acids are present in high percentages in *Moringa* different parts if compared to other amino acids. In this respect, Ferreira *et al.*, [4] recommended the use of *Moringa* seed as an animal feed, and it is an excellent source of amino acid content for monogastric animals.

### 4.2. Growth performance

Birds fed MOSP at 0.75% with enzyme had significantly higher body weight (BW) than control with enzyme and other dietary treatments. In this respect, all levels of germinated *Moringa oleifera* seed (GMOS) (0.25, 0.50, and 0.75%) significantly increased BW and BWG at 14, 28 and, 42 days of age compared to the control group [24]. These results are compatible with those observed by Abbas and Ahmed [6] who used *Moringa oleifera* seed powder at levels 0.37, 0.75, 1.5%, and found that level of 0.37% increased live body weight compared to control.

Results obtained herein are in line with Ochi *et al.*, [7] who assured that the enrollment of *Moringa oleifera* seed powder (MOSP) at levels of 0.5, 1, and 2% in the diet of the broilers significantly ( $P < 0.05$ ) enhanced their body weight, weight gain and feed consumption.

The improved weight gain and feed conversion ratio (FCR) of birds fed on MOSP may be due to the *Moringa oleifera* seeds are a good source of fat, protein, antioxidants, and minerals as mentioned by Compaore *et al.*, [2]. Also, Toye *et al.*, [25] found that the improvement of body weight gain and FCR of broilers having MOSP may be attributed to nutrients rich content of germinated *Moringa oleifera* seed, while Fahey *et al.*, [26] stated that the main cause is the antimicrobial properties of *Moringa*.

The results showed improved in the gizzard weight in all broilers were fed on MOSP. The results are in agreement with Molepo [27] who mentioned that *Moringa* seed meal supplementation improved the gizzard weight of female Ross 308 broiler chickens aged at 42 days of ages. Akouango *et al.*, [28] also found that *Moringa* seed meal supplementation increased the gizzard weights of broiler chickens.

Abdominal fat was significantly decreased in broilers fed MOSP at both 0.75% and 1% with an enzyme, compared to the control group. However, this result is not consistent with Compaore *et al.*, [2] and Akouango *et al.*, [28] who reported that *Moringa* seed meal supplementation increased the fat pad of broiler chickens aged at 42 days of ages.

Results showed that MOSP at 1% with enzyme recorded significantly ( $P < 0.05$ ) a larger percentage of the lymphoid organs (Table 7). These results may be attributed to the antioxidant activities of some components of *Moringa oleifera* like vitamins C and E [29] and phenols especially flavonoids [30] as well as to the capacity of plants polysaccharides to modulate the immune system [31]. In general, all the lymphoid organs are in a normal range and parallel to that reported by Cazaban and Gardin [32], being ranged between 0.18 and 0.30%.

### 4.3. Blood analysis

It is well known that heterophils play an indispensable role in the immune defense of the avian host, functionally; heterophils rapidly kill bacterial pathogens by phagocytosis, degranulation, and generation of an oxidative burst [33,34] because of their early response and ability to kill pathogens. Therefore, heterophils are a useful functional biomarker for evaluating the innate immune competence in poultry. Their numbers increase during mildly or moderately stressful conditions and consequently, the heterophil/lymphocyte ratio can be used to detect the presence of physiological stress for most stressors [35].

It is worth mentioning that HDL cholesterol is strongly and inversely associated with the occurrence of cardiovascular events. Due to that MOSP has antioxidative and anti-inflammatory properties; it reduces LDL cholesterol [36], especially at 0.75% with an enzyme.

Changes in the AST level are possible due to the changes in liver metabolism and to the significant muscle development that usually happens during the growth period, as noted by Szabo *et al.*, [37]. Almeida *et al.*, [38] noted that the higher level for AST was at 35 days of age, wherein this phase there is rapid body development due to higher feed intake. It is well known that ALT enzyme in a higher amount in the liver is used to identify acute liver failures [39], although MOSP did not cause negative effects compared to the control (Table 7).

### 4.4. Newcastle disease titer

The results agree with Walter *et al.*, [12] who suggested that MOSM has beneficial effects on Salmonella and Escherichia so, it should increase overall immunity. Also, Eze *et al.*, [40] recommended using *Moringa oleifera* extract as an immune-booster against ND in non-vaccinated birds.

It is worth to mention that no mortality had been recorded all over the experimental period. In this respect, Ochi *et al.*, [7] mentioned that no adverse effect on mortality rate had been observed on birds receiving dietary MOSP. Seemingly, Walter *et al.*, [12] noticed that *Moringa oleifera* seed extract reduced bacteria numbers which cause water-borne diseases, thus reducing mortality rates in chickens. The absence of deaths of chicks having MOSP might be attributed to the presence of antioxidants in Moringa which enhance the immune system of chickens [41,42].

## 5. Conclusion

Under the current research conditions, it could be concluded that *Moringa oleifera* seed Powder (MOSP) improved performance, immune response and, blood constituents of Cobb 500 broiler chicks either at 0.75% or 1%, with 0.75% plus phytase being the best.

### Ethics approval of research

Animal housing and handling procedures during experimentation were approved by the Institutional Animal Care and Use Committee (CU-IACUC), Cairo University, Egypt, with approval number CU II F 16 20.

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## Conflict of interest

No potential conflict of interest was reported by the authors.

## References

1. Makkar HPS, Becker K (1999) Plant toxins and detoxification methods to improve feed quality of tropical seeds review Asian-Australian. *AJAS* 12: 467–480.
2. Compaore WR, Nikiema PA, Bassole HIN (2011) Chemical composition and antioxidative properties of seeds of *moringa oleifera* and pulps of parkiabiglobosa and adansonia digitata commonly used in food fortification in burkinafaso. *Curr Res J Biol Sci* 3: 64–72.
3. Abbas TE (2013) The use of *Moringa oleifera* seeds in poultry diet. *Turk J Vet Anim Sci* 37: 492–496.
4. Ferreira PMP, Farias DF, Oliveira JTA, et al. (2008) *Moringa oleifera*: bioactive compounds and nutritional potential. *Rev. Nutr* 21: 431–437.
5. Anwar F, Rashid U (2007) Physico-chemical characteristics of *moringa oleifera* seeds and seed oil from a wild provenance of pakistan. *Pak J Bot* 39: 1443–1453.
6. Abbas TE, Ahmed ME (2012) Use of *moringa oleifera* seeds in broilers diet and its effects on the performance and carcass characteristics. *Int J Appl Poult Res* 1: 1–4.
7. Ochi EB, Elbushra ME, Fatur M, et al. (2015) Effect of Moringa (*Moringa oleifera* Lam) seeds on the performance and carcass characteristics of broiler chickens. *J Nat Sci Res* 5: 66–73.
8. Qwele K, Muchenje V, Oyedemi SO, et al. (2013) Effect of dietary mixtures of moringa (*Moringa oleifera*) leaves, broiler finisher and crushed maize on anti-oxidative potential and physicochemical characteristics of breast meat from broilers. *Afr J Biotechnol* 12: 290–298.
9. Ghebremichael KA, Gunaratna KR, Henriksson H, et al. (2005) A simple purification and activity assay of the coagulant protein from moringa oleifera seed. *Water Res* 39: 2338–2344.
10. Jamil A, Shahid M, Khan MM, et al. (2007) Screening of some medicinal plants for isolation of antifungal proteins and peptides. *Pak J Bot* 39: 211–221.
11. Gaia S (2005) Wonder tree 100 facts moringa fact 04 exceptional animal feed moringa as livestock feed & pet food. Moringa mission trust. Available from: <http://gaiathelivingplanet.blogspot.com/2005/06/wondertree-100-facts-moringa-fact-04.html>.
12. Walter A, Samuel W, Peter A, et al. (2011) Antibacterial activity of *Moringa oleifera* and *Moringa stenopetala* methanol and n-hexane seed extracts on bacteria implicated in water borne diseases. *Afri J Microbiol Res* 5: 153–157.
13. Yang RY, Chang LC, Hsu JC, et al. (2006) Nutritional and functional properties of moringa leaves—from germplasm, to plant, to food, to health. *Moringa and other highly nutritious plant resources: Strategies, standards and markets for a better impact on nutrition in Africa*. Accra, Ghana. 1–9.

14. Chollom SC, Agada GOA, Gotep JG, et al. (2012) Investigation of aqueous extract of *Moringa oleifera* lam seed for antiviral activity against Newcastle disease virus in ovo. *J Med Plants Res* 6: 3870–3875.
15. Broiler performance and nutrition supplement (2018) Available from: <https://cobbstorage.blob.core.windows.net/guides/5a171aa0-6994-11e8-9f14-bdc382f8d47e>.
16. AOAC (1990) (Association of official analytical chemists): animal feed. Official methods of analysis; arlington 4<sup>th</sup> edition.
17. AOAC (2012) Official methods of analysis of the AOAC International No. 994.12. 19<sup>th</sup> chapter 4, 18–19, official journal of the European Communities 19.9.98, Gaithersburg, Maryland, USA.
18. AOAC (2005) (Association of official analytical chemists): animal feed. Official methods of analysis; arlington 16<sup>th</sup> edition.
19. Allan WH, Gough RE (1974) A standard hemagglutination inhibition test for Newcastle disease. A comparison of macro and micro methods. *Vet Rec* 95: 120–123.
20. SAS (1990) Statistical analysis system, users guide statistics, as, institute Cary, North Carolina, USA.
21. Duncan DB (1955) Multiple range and multiple F tests. International Biometric Society. *Biometrics* 11: 1–42.
22. Francis G, Makkar H, Becker K (2012) Products from little researched plants as aquaculture feed ingredients. *Agrippa-FAO online journal*. Available from: <http://www.fao.org/Agrippa>.
23. EI-Massry FHM, Mossa MEM, Youssef SM et al. (2013) *Moringa oleifera* plant “value and utilization in food processing”. *Egypt J Agric Res* 91: 1597–1609.
24. Mousa MAM, Riry FHS, Kout Elkloub M, et al. (2016) Effect of using germinated *Moringa oleifera* seeds on japans quail growth performance. *Egypt Poult Sci* 36: 561–571.
25. Toye AA, Sola-Ojo FE, Olaniyan OM, et al. (2013) Nutrigenetic effect of *Moringa oleifera* seed meal on the biological growth programme of young broiler chickens. *Agrosearch* 13: 149–163.
26. Fahey JW, Zakmann AT, Talalay P (2001) The chemical diversity and distribution of glucosinolates and isothiocyanates among plants. *Phytochemistry* 56: 5–51.
27. Molepo LS (2014) Effect of moringa seed meal supplementation on productivity and carcass characteristics of ross 308 broiler chickens. Thesis (MSC. Agriculture (Animal Production)) University of Limpopo. Available from: <http://hdl.handle.net/10386/1363>.
28. Akouango F, Ngokaka C, Ewomango P, et al. (2010) Caractérisation morphométrique et reproductive des taureaux et vaches N'Dama du congo. *Anim Genet Resour* 46: 41–47.
29. Rocha JSR, Lara LJC, Baiao NC, et al. (2010) Antioxidant properties of vitamins in nutrition of broiler breeders and laying hens. *World's Poult Sci J* 66: 261–270.
30. Diallo A, Eklu-Gadegkeku K, Mobio T, et al. (2009) Protective effect of *moringa oleifera* lam. and *lanneakerstingii* extracts against cadmium and ethanol-induced lipid peroxidation. *J Pharmacol Toxicol* 4: 160–166.
31. Dong XF, Gao WW, Tong JM, et al. (2007) Effect of polysavone (alfalfa extract) on abdominal fat deposition and immunity in broiler chickens. *Poult Sci* 86: 1955–1959.
32. Cazaban C, Gardin Y (2012) Part 1: Bursa of fabricius is a visual indicator. *Poultry World* online. Available from: <https://www.poultryworld.net/Broilers/Health/2011/12/Part-1-Bursa-of-Fabricius-is-a-visual-indicator-WP009780W/>.

33. Genovese LL, Lowry VK, Genovese KJ, et al. (2000) Longevity of augmented phagocytic activity of heterophils in neonatal chickens following administration of salmonella enteritidis-immune lymphokines to chickens. *Avian Pathol* 29: 117–122.
34. Kogut MH, Pishko EJ, Kaspers B, et al. (2001) Modulation of functional activities of chicken heterophils by recombinant chicken IFN- $\gamma$ . *J Interferon Cytokine Res* 21: 85–92.
35. Maxwell MH, Robertson GW (1998) The avian heterophil leucocyte: a review. *World's Poult Sci J* 54: 155–178.
36. Norata GD, Pirillo A, Catapano, AL (2006) Modified HDL. Biological and physiopathological consequences., *Nutr Metab Cardiovasc Dis* 16: 371–386.
37. Szabo A, Mezes M, Horn P, et al. (2005) Developmental dynamics of some blood biochemical parameters in the growing turkey (*Meleagris Gallopavo*). *Acta Vet Hung* 53: 397–409.
38. Almeida JG, Vieira SL, Gallo BB, et al. (2006) Period of incubation and posthatching holding time influence on broiler performance. *Braz J Poult Sci* 8: 153–158.
39. Orlewick MS, Vovchuk E (2012) Alanine aminotransferase online. Available from: <http://emedicine.medscape.com/article/2087247-overview>.
40. Eze DC, Okwor EC, Okoye JOA, et al. (2013) Immunologic effects of *moringa oleifera* methanolic leaf extract in chickens infected with Newcastle disease virus (kudu 113) strain. *Afr J Pharm Pharmacol* 7: 2231–2237.
41. Yang RY, Tsou SC, Lee TC, et al. (2006) Moringa, a novel plant rich in antioxidant, bio-available iron and nutrients. ACS Symposium Series. American Chemical Society. 925: 224–239.
42. Du PL, Lin PH, Yang RY, et al. (2007) Effects of dietary supplementation of *moringa oleifera* on growth performance, blood characteristics and immune response in broilers. *J Chin Soc Anim Sci* 36: 135–146.



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