

Effects of blending a phytobiotic (*Dichrostachys glomerata*) to an enzyme and/or a probiotic on growth performances of Japanese quails

Pimagha Moffo Hermann Joël^{1*}, Chongsi Margaret Mary Momo¹,
Noumbissi Marie Noël Bertine¹, Edie Nounamo Langston Wilfried¹,
Tchouan Deffo Gilchrist¹, Ngatcha Yonkeu Lynda¹, Nyembo Kondo Camile¹,
Kana Jean Raphaël¹

¹Department of Animal Sciences, Animal Production and Nutrition Research Unit, Faculty of Agronomy and Agricultural Sciences, University of Dschang, PO Box: 70 Dschang, Cameroon

*Corresponding author: hermannmoffo@yahoo.fr

ABSTRACT

The present study was carried out to assess the effects on Japanese quails of the combination of a phytobiotic (*Dichrostachys glomerata*), an enzyme (Enziblend Plus[®]) and a probiotic (Thepax[®]) on growth performance, carcass characteristics, feed component digestibility and blood parameters. For this purpose, 256 two-week-old quails weighing 55±15 g were housed in cages following a completely randomised design with 8 treatments including 2 controls repeated 4 times each (8x4= 32 experimental units). Each treatment consisted of 32 quails, 8 per replicate (4 males and 4 females). The birds in group 1 without additives represented the negative control and those in group 2 supplemented with antibiotic, the positive control. Animals in groups 3, 4 and 5 received 4 g *D. glomerata* per kg feed, 1 g enzyme per kg feed and 0.4 ml of probiotic/l of drinking water. Finally, the animals in groups 6, 7 and 8 were fed blend *D. glomerata* (4 g) with enzyme (1 g), probiotic (0.4 ml/l of drinking water) and the blend of enzyme (1 g) + probiotic (0.4 ml/l of drinking water) respectively. The main results showed that the highest live weight (218.43 g) and the lowest feed conversion ratio (7.49) were obtained with blend *D. glomerata*-enzyme and blend *D. glomerata*-probiotic respectively. The latter also registered the highest apparent digestive utilisation coefficient for crude protein (87%). Carcass characteristics were unaffected, apart from abdominal fat, which dropped to 0.67 g with blend *D. glomerata* -probiotic, and relative liver weight, which increased slightly with the different additive combinations. With regard to haematological parameters, only the mean corpuscular volume and haematocrit level increased significantly ($p<0.05$) with the different additive combinations; the significantly highest haematocrit level was recorded with the blend of *D. glomerata*-enzyme. In terms of biochemical profile, only ALT, urea, creatinine, triglyceride and LDL cholesterol levels were significantly ($p<0.05$) lower with the additive combinations. In conclusion, blending *D. glomerata* with the enzyme (Enziblend Plus[®]) / probiotic (Thepax[®]) or both improve live weight and digestibility of crude protein in the ration; these blending have no toxic effects on Japanese quails.

Key words: *Dichrostachys glomerata*, Digestibility, Enzyme, Growth performance, Japanese quails, Probiotic

Date of Submission: 02-08-2023

Date of Acceptance: 12-08-2023

I. INTRODUCTION

The ban on the use of antibiotics as feed additives in livestock farming has led to a deteriorating effect in animal health, resulting in a drop in farm yields (1). This situation has forced researchers in the livestock sector to develop alternative strategies. In response to this challenge, several substitute products such as prebiotics, probiotics, symbiotics, enzymes and phytobiotics or phytochemicals have emerged. Phytobiotics are plant-derived products that, when added to animal feed, improve digestive health and growth performance (2). Several of their chemical compounds have beneficial effects such as modulating the intestinal flora by reducing the number of certain pathogenic bacteria such as salmonella, increasing digestive secretions, improving the digestibility and absorption of nutrients, and modulating the immune system (3). Among spices, the fruit of mimosa small bell (*Dichrostachys glomerata*) is rich in flavonoids, phenols and alkaloids, which confer antimicrobial, anti-inflammatory, antioxidant properties and regulate the intestinal flora of animals (4). (5) have shown that its incorporation into broiler chicken rations induces better growth performance compared with control rations without supplements. (6) recently reported that its incorporation at a rate of 4 g/kg feed improved weight gain and carcass yield in Japanese quail. Enzymes are biological catalysts used in animal feed to increase the accessibility

of nutrients contained in feed, to compensate for the absence in animals of enzymes capable of hydrolysing particular chemical bonds and to compensate for the lack of enzymes in the digestive tract of young animals. In so doing, they improve the digestibility and availability of nutrients and thus contribute to better assimilation of the ration and, consequently, improve animal performance (7).

Probiotics (as described by Fuller in 1989) are living microorganisms that affect their host by improving intestinal balance. In fact, they modify the composition of the intestinal microflora, providing the body with microorganisms that modify the acidity of the intestinal contents, stabilise the intestinal mucosa and support digestion, thus helping to maintain good intestinal health by eliminating entero-pathogens. They also stimulate the body's defence mechanisms (8, 9). Probiotics have already been used in a number of studies where their effects have been demonstrated. For example, (10) found that supplementing broiler rations with probiotics (*Lactobacillus fermentum* and *Saccharomyces cerevisiae*) improved growth performance.

Given the multiple beneficial effects of the various additives, their combination could have more pronounced synergistic effects on the growth performance of the animals. It was in this perspective that the present study was initiated, with the general aim of contributing to the search for growth-promoting antibiotic substitutes in livestock farming and more specifically, to assess the effects of blending in feed a phytobiotic (*Dichrostachys glomerata*), an enzyme (EnziBlend Plus[®]) and a probiotic (Thepax[®]) on growth performance, feed components digestibility, carcass characteristics and haemato-biochemical markers of Japanese quail.

II. Materials and methods

Area of study

This study was carried out at the Animal Production and Nutrition Research Unit of the University of Dschang, Cameroon. It is located at 5°26' North latitude, 10°26' East longitude and has an average altitude of 1,420 m. Dschang is located in the agro-ecological zone of the highlands of Western Cameroon, with an equatorial climate at high altitude.

Animal material and feed additives

Two hundred and fifty-six (256) two-week-old Japanese quails (*Coturnix japonica*) with an average weight of approximately 55±15g were used in this study. These birds were housed in cages following a completely randomised design of 32 birds (16 males and 16 females) per treatment. The additives used were the antibiotic Doxycycline[®], the enzyme EnziBlend Plus[®], the probiotic Thepax[®] and the dried fruit powder of *Dichrostachys glomerata*. They were used to formulate five diets as follows:

T0: Basal diet (without additives);

T0+: T0 + 1 g antibiotic (Positive control);

T1: T0 + 4 g *D. glomerata*;

T2: T0 + 1 g Enzyme;

T3: T0 + 0.4 ml probiotic/l drinking water;

T4: T0 + 4 g *D. glomerata* + 1 g Enzyme;

T5: T0 + 4 g of *D. glomerata* +0.4 ml of probiotic/l of drinking water;

T6: T0 + 4 g *D. glomerata* + 1 g Enzyme +0.4ml probiotic/l drinking water.

The basal diet (T0) was formulated as presented in table 1.

Table 1: Composition of basal diet

Ingredients	Quantity (%)
Maize	60
Wheat bran	4
Soybean meal	22
Groundnut meal	4
Fish meal	4
Bone meal	0,5
Oyster shell	0.5
*Premix 5%	5
Total	100
Analysed chemical composition	
Dry Matter (%)	92.54
Crude protein (%DM)	19.78
Ash (%DM)	8.28
Crude cellulose (%DM)	7.41
Fat (%DM)	2.06
Calculated chemical composition	
Metabolizable energy (kcal/kg DM)	3117.16
Lysine (%)	1.33
Méthionine (%)	0.46
Calcium (%)	1.04
Available phosphorus (%)	0.54
Sodium (%)	0.03

* Vitamin premix provided per kilogram of diet: vitamin A: 3000000 IU; vitamin D3: 600000 IU; vitamin E: 4000 mg; vitamin K: 500 mg; vitamin B1: 200 mg; vitamin B2, 1000 mg; vitamin B6: 400 mg; vitamin B12: 4 mg; Mn: 80 mg; Fe: 8000 mg; Zn: 10000 mg; Cu: 2000 mg; Methionine: 200000 mg; Lysine: 78000 mg; Se: 20 mg. DM: Dry Matter

Data collection

- Growth performances

At the start of the trial and every 7 days thereafter, the birds were weighed in groups to assess changes in live weight, and weight gain was calculated by doing the difference between two consecutive weekly live weights. Feed intake was calculated at the end of the week (7 days) by doing the difference between the quantity of feed served at the start of the week and the refusal at the end of the same week. Feed conversion ratio was calculated as the ratio of feed intake to weight gain. At the end of the 35-day experiment, 12 quails per treatment (6 males and 6 females) were randomly selected and fasted for 24 hours. They were then weighed, bled, plucked and eviscerated to assess carcass yields and relative organ weights.

- Feed Component Digestibility

At the end of the experiment, 24 quails of comparable average weight per treatment were selected to assess the digestibility of the feed components. For this purpose, digestion sheets were placed under their cages to collect the faeces for a period of 3 days, during which a defined quantity of feed was served, and then faeces were collected and weighed. These faeces were taken to the animal nutrition laboratory to determine dry matter, organic matter, ash, crude protein and crude cellulose contents in order to calculate the apparent digestive coefficients (aDC) of feed with respect to the treatment received. The aDC was calculated using the formular below:

$$\text{Feed component aDC} = \frac{\text{Ingested feed component (g)} - \text{Excreted feed component (g)}}{\text{Ingested feed component (g)}} \times 100$$

- Haematological and Biochemical analysis

Blood from birds sacrificed at 35 days to assess carcass characteristics was collected in test tubes containing anticoagulant and without anticoagulant in order to assess respectively haematological parameters (white blood cell, red blood cell, haemoglobin, hematocrite, platelets and plateletcrite) using an automatic blood cell haematometer and biochemical parameters (ASAT, ALAT, Creatinine, Urea, Total Protein, Albumin, Globulin, Total Cholestérol, Triglycérides, HDL-cholesterol and LDL-cholesterol) using commercial Chronolab® kits.

Statistical analysis

Data collected were submitted to a one-way analysis of variance (ANOVA); where there was a significant difference between means, Duncan's test at the 5% threshold was used to separate them. SPSS 20.0 was used to perform these analyses.

III. Results

Effects of blend *D. glomerata*-Enziblend Plus®-Thepax® on growth performance of Japanese quails

The effects of in-feed blend *D. glomerata*- Enziblend Plus®- Thepax® on cumulative feed intake, live weight, weight gain and feed conversion ratio of Japanese quails are summarised in Table 2. It can be seen that the various parameters studied were not significantly ($p>0.05$) affected, with the exception of water intake, which dropped significantly ($p>0.05$) with treatments T2, T3, T5 and T6 as compared to the other treatments; the highest data was recorded in birds receiving *D. glomerata* and enzyme (T4) and the lowest in those receiving only the enzyme (T2).

Table 2: Effects of blend *D. glomerata*- Enziblend Plus® -Thepax® on growth performance of Japanese quails.

Parameters	Treatments								P
	T0	T0+	T1	T2	T3	T4	T5	T6	
Feedintake (g)	903.79± 100.43	835.2± 105.98	940.56± 84.92	884.57± 60.92	873.88± 116.71	880.75± 47.15	897.37± 93.24	919.15± 130.00	0.87
Water intake (ml)	1586.25± 30.57 ^{ab}	1511.42± 43.79 ^{bc}	1514.80± 67.12 ^{bc}	1287.93± 59.38 ^d	1448.78± 45.09 ^c	1626.23± 38.08 ^a	1466.56± 41.94 ^c	1446.46± 77.91 ^c	0.00
Live weight (g)	210.56± 15.81	211.91± 6.29	215.99± 11.68	197.72± 10.31	203.57± 14.25	218.43± 8.92	214.85± 5.68	206.79± 11.85	0.20
Weight gain (g)	116.00± 19.74	110.47± 16.92	122.33± 13.78	103.44± 10.86	109.83± 11.27	120.83± 23.48	120.64± 14.75	109.59± 16.42	0.68
Feed conversion ratio	7.89± 0.94	7.59± 0.21	7.73± 0.64	8.83± 1.17	8.10± 1.90	7.49± 1.40	7.47± 0.48	8.49± 1.52	0.76

a, b, c, d: means bearing the same letter on the same line are not significantly different ($p>0.05$)

T0: Basal diet without additives (Negative Control); T0+: T0 + 1 g Doxycycline® /kg feed (Positive Control); T1: T0 + 4 g *D. glomerata* /kg feed; T2: T0 + 1 g Enzyme /kg feed; T3: T0 + 0.4 ml probiotic/l of drinking water; T4: T0 + 4 g *D. glomerata* + 1g Enzyme/kg feed; T5: T0 + 4 g *D. glomerata* +0.4 ml probiotic/l drinking water; T6: T0 + 4 g *D. glomerata* + 1g Enzyme/kg feed + 0.4 ml probiotic/l drinking water; p: probability.

Effects of blend *D. glomerata*-Enziblend Plus®-Thepax® on feed components digestibility

Table 3 shows that the incorporation of the enzyme alone (T2) in feed resulted in a significant increase ($p<0.05$) in the apparent digestibility of crude fibre compared to all the other treatments; the rest of the treatments were comparable. However, the lowest digestive utilisation coefficient for crude fibre was recorded with birds fed T4 ration. Birds fed the control and T1 rations registered the lowest ($p<0.05$) crude protein digestibility values. The various feed additives used alone or blend induced a non-significant increase on dry matter and organic matter digestibility. However, the digestibility of these feed components remained comparable in birds fed supplemented rations compared to the control.

Table 3: Effects of blend *D. glomerata*-Enziblend Plus®-Thepax® on feed components digestibility of Japanese quails

Digestibility parameters (%)	Treatments								P
	T0	T0+	T1	T2	T3	T4	T5	T6	
DM aDC	69.40± 4.94	70.62± 4.56	72.37± 1.75	71.50± 3.95	71.52± 3.05	66.46± 3.26	71.73± 3.05	70.16± 5.88	0.670
CC aDC	73.90± 2.36 ^b	76.67± 2.02 ^{ab}	76.94± 1.20 ^{ab}	79.42± 2.07 ^a	76.58± 2.86 ^{ab}	69.88± 1.65 ^c	74.26± 1.63 ^b	75.76± 2.59 ^{ab}	0.003
CP aDC	75.36± 3.80 ^b	77.39± 3.53 ^b	79.09± 1.40 ^b	88.43± 1.60 ^a	85.44± 1.60 ^a	86.58± 1.18 ^a	87.55± 0.59 ^a	87.16± 2.02 ^a	0.000
OM aDC	70.33± 4.65	71.70± 4.50	73.37± 1.59	72.59± 3.85	72.56± 2.89	68.07± 3.67	72.70± 1.41	71.27± 5.68	0.740

a, b, c: means bearing the same letter on the same line are not significantly different ($p>0.05$)

T0: Basal diet without additives (Negative Control); T0+: T0 + 1 g Doxycycline® /kg feed (Positive Control); T1: T0 + 4 g *D. glomerata* /kg feed; T2: T0 + 1 g Enzyme /kg feed; T3: T0 + 0.4 ml probiotic/l of drinking water; T4: T0 + 4 g *D. glomerata* + 1g Enzyme/kg feed; T5: T0 + 4 g *D. glomerata* +0.4 ml probiotic/l drinking water; T6: T0 + 4 g *D. glomerata* + 1g Enzyme/kg feed + 0.4 ml probiotic/l drinking water; p: probability. aDC: apparent Digestibility Coefficient; DM: Dry Matter; CC: Crude Cellulose; CP: Crude Protein; OM: Organic Matter.

Effects of blend *D. glomerata*-Enziblend Plus®-Thepax® on carcass characteristics of Japanese quails

Carcass yield and relative weights of the head, leg and heart were not significantly ($p>0.05$) affected by the incorporation of *D. glomerata*, Enziblend Plus® enzyme and their blending in feed (Table 4). Supplementing feed with antibiotic (T0+) and the blending of different additives (T6) resulted in a significant ($p<0.05$) increase in relative weight of the liver compared to all the other treatments. The inclusion of *D. glomerata* (T1) and blend

D. glomerata-enzyme (T4) resulted in a significant ($p < 0.05$) increase in the weight of the abdominal fat compared to the enzyme and probiotic supplemented rations.

Table 4: Effects of blending *D. glomerata*-Enziblend Plus®-Thepax® on carcass characteristics of Japanese quails

Parameters (%BW)	Treatments								P
	T0	T0+	T1	T2	T3	T4	T5	T6	
Carcass yield 1	65.32± 2.98	67.46± 7.40	63.95± 8.02	66.59± 3.84	64.74± 4.31	65.34± 3.33	64.89± 4.37	65.60± 4.16	0.79
Carcass yield 2	76.61± 3.42	78.76± 8.37	74.48± 9.01	77.38± 3.48	75.73± 4.00	76.40± 2.72	76.45± 4.25	76.33± 4.18	0.74
Head	4.72± 0.97	4.55± 1.00	4.62± 0.60	4.65± 0.49	4.59± 0.57	4.66± 1.15	5.07± 0.83	4.43± 0.72	0.75
Legs	2.04± 0.33	2.08± 0.31	1.87± 0.36	2.04± 0.19	2.09± 0.17	2.01± 0.27	1.96± 0.19	1.93± 0.16	0.41
Heart	0.90± 0.18	0.88± 0.13	0.89± 0.17	0.85± 0.10	0.84± 0.14	0.84± 0.12	0.84± 0.11	0.85± 0.08	0.89
Liver	1.73± 0.50 ^{bc}	2.20± 0.48 ^{ab}	1.62± 0.43 ^c	1.71± 0.33 ^{bc}	1.88± 0.42 ^{abc}	1.75± 0.39 ^{bc}	1.83± 0.27 ^{abc}	2.23± 0.77 ^a	0.03
Pancreas	0.26± 0.05	0.27± 0.07	0.23± 0.08	0.25± 0.08	0.22± 0.08	0.21± 0.05	0.24± 0.09	0.20± 0.03	0.36
Abdominal fat	0.89± 0.16 ^{ab}	0.89± 0.32 ^{ab}	1.10± 0.30 ^a	0.68± 0.14 ^{bc}	0.61± 0.36 ^c	1.01± 0.30 ^a	0.67± 0.29 ^{bc}	0.90± 0.38 ^{ab}	0.00

a, b, c: means bearing the same letter on the same line are not significantly different ($p > 0.05$). Carcass yield 1=100*(Dressed weight/live body weight); Carcass yield 2={ 100*(Dressed weight+legs+heart+liver+head+gizzard)/live body weight }.

T0: Basal diet without additives (Negative Control); T0+: T0 + 1 g Doxycycline® /kg feed (Positive Control); T1: T0 + 4 g *D. glomerata* /kg feed; T2: T0 + 1 g Enzyme /kg feed; T3: T0 + 0.4 ml probiotic/l of drinking water; T4: T0 + 4 g *D. glomerata* + 1 g Enzyme/kg feed; T5: T0 + 4 g *D. glomerata* +0.4 ml probiotic/l drinking water; T6: T0 + 4 g *D. glomerata* + 1 g Enzyme/kg feed + 0.4 ml probiotic/l drinking water; p: probability.

Effects of blend *D. glomerata*-Enziblend Plus®-Thepax® on the relative weight of organs

The effects of incorporating *D. glomerata*, enzyme and their blending in feed on the development of digestive organs are shown in Table 5. This table shows that there was no significant ($p > 0.05$) effect on the evaluated digestive organs whatever the additives used.

Table 5: Effects of blend *D. glomerata*-Enziblend Plus®-Thepax® on the digestive organs of Japanese quails

Digestive organs	Treatments								P
	T0	T0+	T1	T2	T3	T4	T5	T6	
Gizzard(%BW)	1.59± 0.24	1.71± 0.35	1.77± 0.56	1.67± 0.34	1.69± 0.35	1.51± 0.31	1.50± 0.26	1.66± 0.26	0.49
Intestinal weight (g)	6.35± 2.03	6.25± 1.59	6.47± 1.35	7.14± 2.09	6.18± 1.89	6.06± 1.90	6.01± 1.72	7.01± 1.66	0.70
Intestinal length (cm)	58.25± 6.47	59.09± 5.00	62.00± 6.75	62.17± 7.46	61.42± 7.93	63.00± 9.35	63.92± 8.68	62.18± 4.95	0.56
Intestinal density	0.11± 0.02	0.11± 0.02	0.10± 0.02	0.11± 0.03	0.10± 0.03	0.09± 0.02	0.10± 0.02	0.11± 0.02	0.35

T0: Basal diet without additives (Negative Control); T0+: T0 + 1 g Doxycycline® /kg feed (Positive Control); T1: T0 + 4 g *D. glomerata* /kg feed; T2: T0 + 1 g Enzyme /kg feed; T3: T0 + 0.4 ml probiotic/l of drinking water; T4: T0 + 4 g *D. glomerata* + 1 g Enzyme/kg feed; T5: T0 + 4 g *D. glomerata* +0.4 ml probiotic/l drinking water; T6: T0 + 4 g *D. glomerata* + 1 g Enzyme/kg feed + 0.4 ml probiotic/l drinking water; p: probability.

Effects of blend *D. glomerata*-Enziblend Plus®-Thepax® on haematological parameters of Japanese quails

Incorporating the different additives alone or blend had no significant ($p > 0.05$) effect on haematological parameters (Table 6) with the exception of mean corpuscular volume and haematocrit level which increased significantly ($p < 0.05$) respectively with the inclusion of blend *D. glomerata* - Enziblend Plus® enzyme - probiotic (T6) and *D. glomerata* - Enziblend Plus® enzyme combination (T4) in the feed compared to the control treatments (T0 and T0+).

Table 6: Effects of blend *D. glomerata*-Enziblend Plus®-Thepax® on haematological parameters

Parameters	Treatments							p	
	T0	T0+	T1	T2	T3	T4	T5		T6
WBC (10 ³ /µl)	93.40±	91.53±	96.75±	92.33±	101.77±	93.70±	97.50±	95.50±	0.51
	2.40	6.83	2.90	5.77	9.65	1.27	0.71	2.12	
RBC (10 ⁶ /µl)	3.59±	3.29±	3.75±	3.36±	3.56±	3.75±	3.63±	3.51±	0.40
	0.24	0.54	0.41	0.38	0.33	0.32	0.32	0.35	
Hb (g/dl)	21.02±	19.18±	22.03±	20.27±	20.78±	21.96±	22.10±	21.52±	0.47
	1.47	3.11	2.16	2.83	2.31	2.41	2.60	2.24	
HCT (%)	67.76±	63.33±	71.67±	66.83±	69.13±	74.20±	71.33±	72.33±	0.00
	4.05 ^{bcd}	3.63 ^d	4.39 ^{abc}	2.93 ^{cd}	3.32 ^{abc}	5.26 ^a	1.81 ^{abc}	3.88 ^{ab}	
MVC (fL)	189.40±	191.58±	191.33±	199.52±	194.17±	197.82±	196.50±	207.00±	0.00
	7.64 ^c	8.36 ^{bc}	4.89 ^{bc}	8.33 ^{ab}	3.76 ^{bc}	3.65 ^{bc}	5.45 ^{bc}	9.49 ^a	
MCH (pg)	58.52±	58.23±	58.80±	60.18±	59.95±	58.10±	60.75±	61.25±	0.28
	2.46	0.97	2.06	3.44	2.53	3.12	2.28	2.27	
MCHC (g/dL)	31.10±	30.43±	30.82±	30.25±	30.00±	29.56±	30.93±	29.63±	0.67
	2.36	1.49	1.93	1.24	0.97	1.50	0.85	1.77	
PLT (10 ³ /µl)	70.00±	64.67±	74.67±	64.20±	66.17±	67.00±	73.75±	67.83±	0.20
	9.14	5.79	4.80	11.43	9.95	1.00	9.54	1.60	
PCT (%)	0.06±	0.05±	0.07±	0.06±	0.06±	0.06±	0.07±	0.06±	0.35
	0.01	0.01	0.01	0.01	0.01	0.01	0.01	0.01	

a, b, c, d: means bearing the same letter on the same line are not significantly different (p>0.05)

T0: Basal diet without additives (Negative Control); T0+: T0 + 1 g Doxycycline® /kg feed (Positive Control); T1: T0 + 4 g *D. glomerata* /kg feed; T2: T0 + 1 g Enzyme /kg feed; T3: T0 + 0.4 ml probiotic/l of drinking water; T4: T0 + 4 g *D. glomerata* + 1 g Enzyme/kg feed; T5: T0 + 4 g *D. glomerata* +0.4 ml probiotic/l drinking water; T6: T0 + 4 g *D. glomerata* + 1g Enzyme/kg feed + 0.4 ml probiotic/l drinking water; p: probability. WBC=white blood cells; RBC=red blood cells; Hb=haemoglobin; HCT=haematocrit; MCV=mean corpuscular volume; MCHC=mean corpuscular haemoglobin concentration; PLT= platelets; PCT=plateletcrit.

Effects of blend *D. glomerata*- Enziblend Plus®-Thepax® on biochemical parameters

Whatever the blending of feed additives considered (Table 7), serum ALAT levels reduced significantly (p<0.05) with the different treatments compared to birds fed the control ration (T0). However, there was no significant difference (p>0.05) between treatments on serum AST, total protein, albumin, globulin and albumin / globulin ratio. On the other hand, serum urea level was significantly (p<0.05) lower with the different additives compared to birds fed antibiotic ration. Creatinine level dropped significantly (p<0.05) with treatments T2, T3 and T6. In the same line, serum LDL-cholesterol and triglyceride levels were significantly (p<0.05) lower in birds fed T1 and T2 respectively compared to the other treatments.

Table 7: Effects of blend *D. glomerata*- Enziblend Plus®-Thepax® on biochemical parameters

Parameters	Treatments								p
	T0	T0+	T1	T2	T3	T4	T5	T6	
ASAT	170.41± 60.93	170.00± 55.94	143.00± 40.65	153.67± 36.90	168.22± 64.42	173.40± 48.51	118.56± 34.55	155.13± 52.20	0.40
ALAT	56.44± 4.04 ^a	43.83± 6.91 ^{bcd}	33.43± 2.99 ^d	33.98± 10.44 ^d	39.75± 7.94 ^{cd}	53.25± 13.78 ^{ab}	45.50± 6.87 ^{bc}	43.09± 9.40 ^{bcd}	0.00
Creatinine	0.02± 0.01 ^{abc}	0.02± 0.01 ^{bc}	0.03± 0.02 ^{ab}	0.02± 0.01 ^c	0.04± 0.01 ^a	0.03± 0.02 ^{ab}	0.02± 0.01 ^c	0.02± 0.01 ^c	0.01
Urea	15.95± 2.42 ^{abc}	17.67± 0.52 ^a	17.00± 0.58 ^{ab}	14.69± 0.54 ^c	14.59± 1.88 ^c	15.98± 2.07 ^{abc}	15.50± 1.74 ^{bc}	14.55± 1.90 ^c	0.01
Total Protein	3.39± 0.68	3.46± 0.51	3.62± 0.24	3.44± 0.32	3.53± 0.32	3.74± 0.47	3.49± 0.71	3.35± 0.51	0.81
Albumin	1.36± 0.29	1.41± 0.17	1.38± 0.18	1.31± 0.12	1.32± 0.11	1.33± 0.25	1.28± 0.55	1.32± 0.15	0.98
Globulin	2.03± 0.47	2.04± 0.41	2.24± 0.17	2.12± 0.23	2.21± 0.24	2.41± 0.36	2.21± 0.65	2.03± 0.48	0.67
Albumin / Globulin	0.69± 0.15	0.71± 0.12	0.62± 0.10	0.62± 0.05	0.60± 0.06	0.56± 0.12	0.63± 0.29	0.68± 0.19	0.55
Total Cholestérol	210.40± 54.32	197.90± 28.72 ^c	194.37± 27.12	220.40± 60.27	217.44± 68.02	214.72± 29.54	186.19± 29.98	223.52± 42.28	0.65
Triglycerides	216.50± 33.75 ^{ab}	236.53± 33.08 ^a	220.05± 28.70 ^a	120.29± 37.14 ^c	172.10± 42.75 ^b	203.22± 42.47 ^{ab}	202.29± 54.19 ^{ab}	201.69± 46.54 ^{ab}	0.00
HDL-Cholesterol	121.70± 28.63	119.83± 27.37	126.70± 26.23	127.61± 22.50	128.47± 30.24	111.36± 14.31	115.28± 12.18	110.40± 22.29	0.64
LDL-Cholesterol	43.00± 6.50 ^{cd}	46.49± 3.82 ^{bc}	27.29± 6.63 ^c	58.73± 11.19 ^a	54.71± 9.34 ^{ab}	62.87± 6.93 ^a	35.20± 4.76 ^{de}	49.16± 4.43 ^{bc}	0.00

a, b, c, d: means bearing the same letter on the same line are not significantly different (p>0.05)

ALAT= Alanine aminotransferase; ASAT= Aspartate aminotransferase; HDL: High density lipoproteins; LDL: Low density lipoproteins; T0: Basal diet without additives (Negative Control); T0+: T0 + 1 g Doxycycline® /kg feed (Positive Control); T1: T0 + 4 g *D. glomerata* /kg feed; T2: T0 + 1 g Enzyme /kg feed; T3: T0 + 0.4 ml probiotic/l of drinking water; T4: T0 + 4 g *D. glomerata* + 1 g Enzyme/kg feed; T5: T0 + 4 g *D. glomerata* +0.4 ml probiotic/l drinking water; T6: T0 + 4 g *D. glomerata* + 1 g Enzyme/kg feed + 0.4 ml probiotic/l drinking water; p: probability.

IV. Discussion

The incorporation of *D. glomerata*, Enziblend Plus®, the probiotic (Thepax®) and their blending in ration did not significantly affect growth performance of quails. This result is in agreement with those of (11) who recorded no significant effect on feed intake with the incorporation of thyme (5 g/kg feed) in broiler feed. (6, 12) also reported that the incorporation of *D. glomerata* in feed did not significantly affect feed intake in Japanese quail. However, this result contradicts that reported by (13) who recorded a significant increase in live weight in broilers receiving a probiotic. (14) also recorded better weight gain and feed efficiency in quails fed diet supplemented with a probiotic and enzymes.

The incorporation of the additives (*D. glomerata*, enzyme or probiotic) alone or blend resulted in a statistically comparable improvement in the digestibility of organic and dry matter meanwhile a significant improvement in the digestibility of cellulose and crude protein was recorded. This improvement in digestibility could be attributed to the enzymes and micro-organisms used, which promoted the breakdown of cellulose and proteins, leading to better nutrient availability. These results are partially consistent with those obtained by (15), where supplemented enzymes and spice, used alone or blend in broiler ration, had no effect on the digestibility of feed components. (16) reported significant increase in feed components digestibility in laying quails following supplementation with Avizyme. In the same line, (17) registered an increase in feed digestibility when quails were fed different levels of enzyme.

Carcass yield and the relative weight of the head, legs and heart were not significantly affected irrespective of the additive used. The same results were obtained by (18) on quail fed ration supplemented with turmeric and enzymes. (19) also found a non significant effect on the carcass characteristics of quails fed parsley and enzymes. These results contradict those obtained by (20) who reported an increase in carcass yield when using a phytobiotic, a probiotic and their blending in the broiler ration.

The incorporation of these studied additives alone or blend in the ration had no significant effect on the development of digestive organs. This result is comparable to those of (13) following probiotic and symbiotic supplementation in broiler rations, and (21) with the incorporation of sunflower meal and a multienzyme in quail's feed.

The various additives used did not affect the haematological parameters studied, apart from haematocrit and mean corpuscular volume, which showed a significant increase compared to the controls treatments. This result is consistent with that of (22) who reported an increase in haematocrit with a medicinal plants mixed infusion (20 ml/L) in drinking water in broilers. This increase in haematocrit remains within the standard for a healthy quail, implying the good nutritional value of the feed used. However, the present work contradicts those of (22, 23) who reported a significant increase in red blood cell count and haemoglobin level with aqueous extracts of ginger and garlic respectively. (24) also reported a significant effect on the haematological parameters studied in quails by supplementing their rations with enzyme.

The incorporation of additives alone or blend had a significant effect on serum levels of ALAT, creatinine, urea and triglycerides. These results are in line with those obtained by (19), who supplemented the ration of quails with parsley and enzymes. Meanwhile, (5) reported that incorporating increasing levels of *D. glomerata* in broiler feed had no significant effect on serum urea, total protein and ALAT levels. Also, (15) had no effect on these parameters when broilers were fed an enzyme supplemented feed. (25) found significant effects on these parameters in quails supplemented with black pepper and turmeric.

V. Conclusion

Blending *D. glomerata* (4 g/kg feed) the enzyme (Enziblend Plus[®]) / probiotic (Thepax[®]) or both improves the live weight and crude protein digestibility. These blending have no side effects on Japanese quails.

References

- [1]. Chardon H, Brugere H, 2014. Usages des antibiotiques en élevage et filières viandes. Cahiers Sécurité Sanitaire Santé Animale. Centre d'Information des Viandes, 36P.
- [2]. Jacela J Y, DeRouchey J M, Tokach M D, Goodband R D, Nelssen J L, Renter D G, Dritz S S. 2010. Feed additives for swine: Fact sheets - prebiotics and probiotics, and phytogenics. *J Swine Health Prod.* 18(3):132-136.
- [3]. Gong J, Yin F, Hou Y, Yin Y. 2014. Review: Chinese herbs as alternatives to antibiotics in feed for swine and poultry production: Potential and challenges in application. *Canadian Journal of Animal Science.* , 94(2) : 223-241.
- [4]. Jane T N, Matthias O A, Ikechukwu F U. 2014. Antioxidant and Hepatoprotective Activity of Fruit Extracts of *Tetrapleura tetraptera* (Schum & Thonn) Taubert. *Jordan Journal of Biological Sciences*, 7(4): 251 - 255.
- [5]. Kana J R, Mube K H, Ngouana T R, Tsafong F, Komguep R, Yangoue A, Tegua A. 2017. Effect of Dietary Mimosa Small Bell (*Dichrostachys glomerata*) Fruit Supplement as Alternative to Antibiotic Growth Promoter for Broiler Chicken. *J. World. Poul. Res*, 7(1)27-34.
- [6]. Ebile Dayan A, Kana J R, Pimagha Moffo H J, Edie Nounamo L W, Nguefack Djieufu G, Ngouana Tadjong R, Fonteh A F. 2018. Effects of *Dichrostachys glomerata* Feeding Regimes on Growth Performance, Gut Microbiota and Haemato-Biochemical profile of Japanese Quails. *J. Anim. Res. Nutr.*, 3(25): 1-8.
- [7]. Cowieson A J, Hruby M, Pierson E E M. 2006. Evolving enzyme technology: impact on commercial poultry nutrition. *Nutrition Research Reviews*, 19: 90-103.
- [8]. Lessard M. 2004. Utilisation de probiotiques chez le porc- modulateurs potentiels de la santé intestinale. *Centre de référence en agriculture et agroalimentaire du Québec.* 14P.
- [9]. Ohimain, E I, Ofongo, R T S. 2012. The effect of probiotic and prebiotic feed supplementation on chicken health and gut microflora: a review. *International Journal of Animal and Veterinary Advances*, 4: 135-143.
- [10]. Bai S P, Wu A M, Ding X M, Lei Y, Bai J, Zhang K Y, Chio J S. 2013. Effects of probiotic-supplemented diets on growth performance and intestinal immune characteristics of broiler chickens. *Poul. Sci.* 92 :663-670.
- [11]. Majid T, Mohsen T, Abas A G, Sayed A T. 2010. Performance, immunity, serum biochemical and hematological parameters in broiler chicks fed dietary thyme as alternative for an antibiotic growth promoter. *African Journal of Biotechnology*, 9(40): 6819-6825.
- [12]. Ebile Dayan Agwah, Kana Jean Raphaël, Edie Nounamo Langston Wilfried, Pimagha Moffo Herman Joël, Nguefack Djieufu Gildas, Ngouana Tadjong Ruben, Mube Kuetchie Hervé, Fonteh Anyangwe Florence. 2018. Growth Performance, Gut Microbiota and Haemato-Biochemical Profile of Quails Fed Diet Supplemented with Graded Levels of *D. glomerata* Fruit Powder. *Animal and Veterinary Sciences*. 6(5): 80-87. doi: 10.11648/j.avs.20180605.13.
- [13]. Awad, W A, Ghareeb K, Abdel-Raheem S, Bohm J. 2009. Effects of dietary inclusion of probiotic and synbiotic on growth performance, organ weights, and intestinal histomorphology of broiler chickens. *J. Poul. Sci.* 88: 49-55.
- [14]. Chimote M J, Barmase B S, Raut A S, Dhok A P, Kuralkar S V. 2009. Effect of Supplementation of Probiotic and Enzymes on Performance of Japanese Quails. *Vet. World.*2(6): 219-220.

- [15]. Al-Harthy M A. 2006. Impact of supplemental feed enzymes, condiments mixture or their combination on broiler performance, nutrients digestibility and plasma constituents. *Int. J. Poul. Sci.* 5(8): 764-771.
- [16]. Alagawany M, Attia A. 2015. Effects of feeding sugar beet pulp and avizyme supplementation on performance, egg quality, nutrient digestion and nitrogen balance of laying Japanese quail. *Av. Biol. Res.*, 8(2): 79-88.
- [17]. Rabie M H, Abo El-Maaty H M A. 2015. Growth performance of Japanese quail as affected by dietary protein level and enzyme supplementation. *Asian J. Anim. Vet. Adv.*, 10: 74-85.
- [18]. Kilany O E, Mahmoud M M A. 2014. Turmeric and Exogenous Enzyme Supplementation Improve Growth Performance and Immune Status of Japanese quail. *World'sVeterinary Journal.* 4(3): 20-29.
- [19]. Bahnas M S, Ragab M S, Asker N E A, Emam R M S. 2009. Effects of using parsley or its by-product with or without enzyme supplementation on performance of growing japanese quails. *Egypt. Poul. Sci.*, 29(1): 241-262.
- [20]. Ferdous M F, Arefin M S, Rahman M M, Ripon M M R, Rashid M H, Sultana M R, *et al.* 2019. Beneficial effects of probiotic and phytobiotic as growth promoter alternative to antibiotic for safe broiler production. *J. Adv. Vet. Anim. Res.*, 6(3):409-15.
- [21]. Tüzün A E, Olgun O, Yıldız A O, Sentürk E. 2020. Effect of Different Dietary Inclusion Levels of Sunflower Meal and Multi-Enzyme Supplementation on Performance, Meat Yield, Ileum Histomorphology, and Pancreatic Enzyme Activities in Growing Quails. *Animals*, 10:680.
- [22]. Javed Y, Khan S, Chand N, Mushtaq M, Sultan A, Rafiullah, Tanweer A J. 2012. Comparative efficacy of different schedules of administration of medicinal plants mixed infusion on hematology of broiler chicks. *Sarhad J. Agric.* 28(2):327-331.
- [23]. Oleforuh-Okoleh, Harriet M N-F, Solomon O O, Joesph O U. 2015. Evaluation of Growth Performance, Haematological and Serum Biochemical Response of Broiler Chickens to Aqueous Extract of Ginger and Garlic. *Journal of Agricultural Science*, 7: 167-173.
- [24]. Shehab A E, Kamelia M Z, Khedr N E, Tahia E A, Esmail F A. 2012. Effect of Dietary Enzyme Supplementation on Some Biochemical and Hematological Parameters of Japanese Quails. *J Anim Sci Adv.* 2(9): 734-739.
- [25]. Asmaa S E, Reham A M A, Eman A E. 2021. Complementary effect of black pepper and turmeric on productive performance and physiological responses of japanese quail. *Egypt. Poul. Sci. J.*, 41(1): 77-91.