

INFLUENCE OF ETHANOLIC EXTRACT OF *ASPILIA AFRICANA* LEAF ON THE PERFORMANCE AND EGG QUALITIES OF JAPANESE QUAILS

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ABSTRACT

A 16-week trial evaluated the effects of ethanolic extract of *Aspilia africana* leaf (EeAaL) on the growth performance, carcass and egg characteristics of growing and laying quails. One hundred and eighty (180), one-week old Japanese quail chicks were assigned to six experimental diets (0, 2.5, 5, 7.5, 10% EeAaL or 0.02% oxytetracycline). Each treatment had 30 birds which were further subdivided into three replicates of 10 birds. Each replicate was housed in a pen under the deep litter system; feed and water were provided *ad libitum*. Growth performance and egg characteristics (egg quantity and quality) indices were subjected to the one-way ANOVA in a completely randomized design (CRD). No significant ($P > 0.05$) effect of EeAaL was observed between treatments on the final body weight and daily weight gains of quails. Feed conversion ratio (4.95 – 5.28) was significantly ($P < 0.05$) improved in quails fed 2.5 – 7.5% EeAaL diets. Zero mortality was observed in quails fed 5 – 10% EeAaL diets compared to other treatments. EeAaL supplementation reduced ($P < 0.05$) the amount of fat deposition in quail carcass, thus improving the carcass quality. The efficacy of EeAaL was more pronounced in laying than growing quails where significant ($P < 0.05$) differences were observed on some external and internal egg characteristics, whereas egg composition did not differ ($P > 0.05$) between treatments. Egg number, hen day production, albumen weight, yolk weight, shell thickness and yolk colour were significantly improved following dietary EeAaL supplementation, indicative of strong growth promoting and stimulatory effects. Therefore, for improved carcass and egg performances up to 7.5% EeAaL should be supplemented into quail diets.

KEYWORDS: Leaf extract, growth rate, egg characteristics, bush marigold, quails

INTRODUCTION

Dietary manipulations involving nutritional, sensory, chemical, physical and physiological characteristics of feed materials are of the innovative strategies developed for improving the quality of animal products (Runjaic-Antic *et al.*, 2010; Agiang *et al.*, 2011). In the developing countries such as Nigeria, animal protein shortage with its increasing demand has remain a major challenge for the Livestock sector and several research efforts are geared towards ameliorating this situation. Diversification of the livestock industry and exploitation of locally, available cheap feed resources are the current research focus (Oko *et al.*, 2011; Abang *et al.*, 2017; Malik *et al.*, 2018).

The Japanese quail (*Coturnix coturnix japonica*) popularly known as the “quintessential micro-livestock” is one of the animal species that is gaining attention in food security programmes (FAO, 2012) due to its suitability in producing meat and eggs at the shortest time (Oko *et al.*, 2012; Egbeyale *et al.*, 2013; Akintunde *et al.*, 2017). Quail breeding is also known as

coturniculture (Mondry, 2016). Lee *et al.* (2005) further noted that quail meat and eggs are of public health significance because of their high protein value and low caloric contents thus, the ideal food for people with several health issues.

The suitability of a number of unconventional feed ingredients including *Aspilia africana* leaf (Oko *et al.*, 2011, 2012, 2013), kenaf seed (Odetola *et al.*, 2017), mango seed (Abang *et al.*, 2017), pigeon pea seed (Akintunde *et al.*, 2017), cassava peel meal (Malik *et al.*, 2018) and sweet potato peels (Edache *et al.*, 2018) in improving quail productivity are currently being explored. *Aspilia africana* (Bush marigold) an obnoxious plant is successfully being incorporated into animal diets (Etim *et al.*, 2017) including poultry (Agiang *et al.*, 2011; Oko *et al.*, 2011) due to its numerous nutritional properties. Its leaf meal and aqueous extract showed significant effects as protein source and growth promoter, respectively in poultry (Agiang *et al.*, 2011; Oko *et al.*, 2011). Egg qualities including yolk colour improved significantly following *Aspilia africana* supplementations.

Thus, this present study further evaluated the effects of

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various levels of ethanolic extract of *Aspilia africana* leaf on the performance and egg characteristics of laying Japanese quails.

Materials and Methods

The research was conducted at the Poultry unit of the University of Calabar, Teaching and Research farm, Calabar, Nigeria. The site is located in the rain forest zone of South-south Nigeria on Latitude 4^o57¹N and Longitude 8^o 19¹E and 42m above sea level. The area is of humid climate with a mean annual rainfall of 1,830mm and temperature of 25^o - 31^oC (Google Earth, 2016).

Test ingredient

Aspilia africana plants used in this study were harvested within the vicinity of the University of Calabar, Teaching and Research farm. The plant was obtained by

harvesting whole field plant above 3cm stubble height within the mid vegetative to flowering stage of development as previously described (Oko *et al.*, 2010). The leaves were sorted, air dried for 72 hrs. and the dried leaves were ground using a hammer mill fitted with 1mm screen into *Aspilia africana* leaf meal. Exactly 200g of leaf meal was measured into a conical flask and soaked in 600ml of 80% ethanol for 24 hrs. at room temperature. The slurry was filtered (using Whatman filter paper No.4) into a 250ml conical flask. The filtrate was then lyophilized to produce gel-like ethanolic extract of *Aspilia africana* leaf (EeAaL). Extract was prepared frequently as required.

Experimental diets

Basal (antibiotic-free) diets were formulated to meet the nutrient requirements of growing and laying quails (Table 1).

Table 1: Composition of basal diets

Ingredients	Growing Period	Laying Period
Maize	41.57	37.80
Soyabean meal	35.43	41.70
Crayfish dust	5.00	5.00
Wheat offal	10.00	4.00
Palm kernel cake	4.00	2.00
Dicalcium phosphate	2.50	4.00
Limestone	0.00	4.00
*Vitamin premix	0.50	0.50
Salt	0.50	0.50
Lysine	0.30	0.30
Methionine	0.20	0.20
Total	100	100
Calculated Analysis		
Crude protein (%)	24.00	24.00
Calcium (%)	0.80	3.00
Metabolisable energy (Kcal/kg)	2,750.00	2,650.00

*Vitamin BCP (premix; 0.25% vitamins and Trace elements)

To determine the optimum level of ethanolic extract supplementation with the best growth promoting effects, two control diets; negative control (diet 1 - basal diet) and positive control (diet 2 - basal + 0.02g/kg oxytetracycline) were formulated according to the methods of Denli *et al.* (2004), and estimated equivalent of 2.5, 5, 7.5 and 10% of ethanolic extract of *Aspilia africana* leaf (EeAaL) were supplemented into the basal diets to represent diets 3, 4, 5 and 6, respectively. Therefore, a total of six dietary treatments were studied.

Management of experimental birds

A total of 180, one-week old quail chicks were assigned to the six experimental diets in a completely randomized design (CRD). Thirty (30) chicks were allotted to each treatment which was further divided into three replicates of 10 birds each. Each replicate was housed in a pen, feed and water were provided *ad libitum*. The birds were managed under the deep litter system, according to the Federation of Animal Science Societies' Principles of Animal Care in experimentation set throughout the 16 weeks duration.

Data collection and analysis

For growing quails, their growth rate (body weight, weight gain, feed intake, feed conversion ratio and mortality) was monitored weekly from weeks 1- 6 of experiment while their carcass quality in terms of dressed, organ and intestinal weights were evaluated at the sixth week. In the laying phase, layers were separated from the males based on their body/phenotypic characteristics. According to Mondry (2016) the female Japanese quail is slightly larger than the male and has a lighter throat with black speckles while the male has a darker, caramel - brown throat. Quail eggs from layers were collected (weeks 7 - 16) twice daily (9.00 and 16.00h) and feed intake, egg weight, egg size and egg quality were measured weekly. Total amount of eggs laid throughout the period were recorded and hen day production was calculated. On a weekly basis, three freshly laid eggs were randomly picked from each replicate and were used for egg quality determination throughout the ten weeks of collection. Egg weights were measured thereafter, broken individually into a flat plate to assess the internal and external egg quality characteristics. Each shell was

washed with distilled water in order to eliminate adhering albumen particles, the shell was then air-dried for 24 hours and weighed.

Egg weights were measured using a digital electronic scale (G & G). The proportion of albumen, yolk and shell were determined as % of egg weight. The vernier caliper (RS 548-718, Mitutoyo Corporations, Japan) was used in measuring the length, diameter, height of the albumen, yolk and egg. Egg shell thickness including shell membrane was measured at three points; on the end, equatorial region and at the pointed end using a Micrometer Screw Guage (PM 025, Draper Tools Ltd., Eastleigh, UK). These measurements were recorded as mean of pooled values. The number of eggs cracked per treatment were recorded. Egg shape index was calculated as the egg diameter divided by the egg length multiplied by 100. Yolk visual colour score was determined by matching the yolk with one of the 15 bands of Roche yolk colour fan.

Data collected were subjected to the one-way analysis of variance in a completely randomized design using the generalized linear model and significant means were separated using the Tukey's methods of the GENSTAT (2011) software package.

Results and Discussion

Each 2.5kg contained;
 Vitamin A 10,000,000 I.U, Vitamin D 2,000,000 I.U,
 Vitamin E 20,000I.U, Vitamin K 2,250mgr, Thiamine 1,750mgr, Riboflavin B₂ 5,000mgr, Pyridoxine B₆ 2,750 mgr, Niacin 27,500 mgr, Vitamin B₁₂ 15 mgr, Pantothenic acid 7,500 mgr, Biotin 50 mgr, Cholin chloride 400gr, Antioxidant 125 gr, Manganese 80 gr, Zinc 50 gr, Iron 20 gr, Copper 5 gr, Iodine 1.20 gr, Selenium 200 gr, Cobalt 200 gr

Diet	Level of supplementation	of Statement
1	Negative Control	Basal diet
2	Positive Control	Basal diet + 0.02g/kg oxytetracycline
3	2.5% EeAaL	Basal diet + 4.25 g/kg EeAaL
4	5.0% EeAaL	Basal diet + 8.49 g/kg EeAaL
5	7.5% EeAaL	Basal diet + 12.74 g/kg EeAaL
6	10.0% EeAaL	Basal diet + 16.98 g/kg EeAaL

Table 2. Performance of quails on EeAaL diets

Parameter	NC	PC	2.5% EeAaL	5.0% EeAaL	7.5% EeAaL	10.0% EeAaL	sem	LSD
Initial weight, g	16.52	16.48	16.54	16.46	16.50	16.53	0.01	
Final weight, g	129.74	130.00	132.88	133.22	130.74	129.93	0.56	6.78
Daily, feed intake g/d	15.70 ^a	15.03 ^b	13.76 ^c	13.77 ^c	13.77 ^c	13.84 ^c	0.16*	0.44
Daily weight gain, g/d	2.70	2.70	2.77	2.78	2.72	2.70	0.91	1.81
Feed conversion ratio	5.82 ^a	5.57 ^b	4.97 ^d	4.95 ^d	5.06 ^{cd}	5.13 ^c	0.02*	0.14
Mortality, %	4.00 ^a	2.67 ^b	1.33 ^c	0.00 ^d	0.00 ^d	0.00 ^d	0.14*	0.21
Carcass yield, %LW	58.90 ^{ab}	59.32 ^{ab}	59.42 ^a	55.29 ^c	58.22 ^b	54.70 ^c	0.56*	1.13
Organ Weight, %LW:								
Gizzard	3.19 ^a	2.75 ^b	2.24 ^c	2.61 ^b	2.36 ^c	2.39 ^c	0.08*	0.23
Heart	0.86 ^{ab}	0.81 ^b	0.90 ^a	0.65 ^c	0.43 ^d	0.76 ^b	0.03*	0.07
Liver	2.36 ^c	2.49 ^c	2.79 ^b	2.45 ^c	2.02 ^d	3.09 ^a	0.10*	0.26
Abdominal fat	0.34 ^b	0.67 ^a	0.31 ^b	0.32 ^b	0.24 ^c	0.21 ^c	0.06*	0.08
Intestine	5.69 ^a	4.90 ^b	5.77 ^a	4.66 ^b	3.31 ^c	4.90 ^b	0.28*	0.45

Table 2 presents the impact of dietary ethanolic extract of *Aspilia africana* leaf (EeAaL) supplementation on the growth performance of growing quails. No significant (P>0.05) effect of EeAaL was observed between treatments on the final body weight and daily weight gains of quails. Whereas, daily feed intake, feed conversion ratio and mortality rate were significantly (P<0.05) different between dietary treatments. Daily feed intake (13.76 – 15.70g/d) was highest in quails fed the control diet without antibiotics or EeAaL supplements, while quails on EeAaL diets had lower feed intake. Feed conversion ratio (4.95 – 5.28) was significantly (P<0.05) improved in quails fed 2.5 – 7.5%

EeAaL followed by those on 10% EeAaL, 0.02g/kg oxytetracycline with the least in quails fed the control diet.

This result indicated that EeAaL supplementation could stimulate digestion process thus improving feed conversion ratio. The inclusion of 5 – 10% EeAaL in quail diets led to a zero mortality compared to other dietary treatments suggesting that EeAaL exerted some antibiotic effects in the animals, thus its ability to improve the immunity and disease resistance status of the quails. Results of this present study are consistent with previous studies (Alicicek *et al.*, 2004; Agiang *et al.*, 2011) that plant extracts improved the growth

performance of growing quails. This report showed that up to 7.5% EeAaL in quail diets exerted strong antibiotic and growth promoting effects on quails.

Table 2 showed that carcass characteristics of quails fed varying levels of ethanolic extracts of *Aspilia africana* leaf did not differ ($P>0.05$) between treatments. Carcass yield of 54.70 – 59.42% was obtained with quails on 2.5% EeAaL recording the highest value while the least value was obtained in quails fed 10% EeAaL. Apart from abdominal fat pad, no significant ($P>0.05$) effects of dietary treatments were observed on the relative organ weight of quails. At increasing levels of EeAaL supplementation, abdominal fat pad reduced

significantly ($P<0.05$), suggesting that EeAaL could reduce the amount of fat deposition in quail carcass, thus improving the carcass quality. This observation is in line with reports on the use of plant products in poultry diets (Alicicek et al., 2004; Agiang et al., 2011; Oko et al., 2012). Higher carcass yield of 76.11 – 90.55% was reported by Malik et al. (2018) in quails fed fermented cassava peels.

The influence of dietary ethanolic extract of *Aspilia africana* leaf (EeAaL) supplementation on the egg characteristics of Japanese quails are presented in Table 3.

Table 3. Effects of EeAaL diets on the egg quality traits of Japanese quails

Parameter	NC	PC	2.5% EeAaL	5.0% EeAaL	7.5% EeAaL	10.0% EeAaL	sem	LSD
External qualities:								
Egg size, g	9.99 ^b	9.72 ^c	10.37 ^a	9.86 ^c	9.41 ^d	10.13 ^b	0.08*	0.23
Egg width, mm	24.08 ^a	23.27 ^b	23.85 ^{ab}	24.11 ^a	22.57 ^c	23.61 ^b	0.16*	0.45
Egg length, mm	30.95 ^a	30.30 ^{ab}	30.87 ^a	31.32 ^a	29.11 ^b	31.07 ^a	0.28*	1.36
Shape Index, %	77.80	76.80	77.26	76.98	77.53	75.99	0.27	2.32
Egg surface area, mm	22.38	21.93	23.02	22.16	21.40	22.62	0.24	2.41
Shell thickness, mm	0.29 ^d	0.30 ^c	0.29 ^d	0.35 ^a	0.31 ^b	0.29 ^d	0.03*	0.01
% Crack	0.87	0.85	0.87	0.84	0.81	0.79	0.01	0.18
Total egg laid	1288 ^c	1453 ^b	1584 ^{ab}	1626 ^a	1738 ^a	1557 ^b	0.79*	221.00
Hen day production, %	61.33 ^e	69.19 ^d	75.43 ^{bc}	77.43 ^b	82.76 ^a	74.14 ^c	1.19*	2.45
Internal qualities:								
Albumen weight, %	47.79	51.95	52.08	52.04	52.05	51.99	0.22	4.55
Yolk weight, %	30.65 ^e	31.03 ^d	31.71 ^c	32.77 ^a	32.79 ^a	32.73 ^{ab}	0.01*	0.03
Shell weight, %	21.56 ^a	17.02 ^b	16.21 ^c	15.19 ^d	15.16 ^d	15.28 ^d	0.04*	0.12
Yolk diameter, mm	22.06	20.98	23.51	21.04	22.43	22.63	0.15	2.56
Yolk height, mm	8.96	10.04	9.38	9.25	9.35	8.82	0.13	1.38
Yolk index, %	41.00	48.00	40.00	44.00	42.00	39.00	0.16	9.52
Yolk colour	3.03 ^e	4.31 ^d	4.75 ^c	5.30 ^b	5.65 ^a	5.80 ^a	0.02*	0.18
Composition:								
Dry matter, %	30.74	30.02	30.90	31.34	31.30	31.28	1.20	1.41
Crude protein, %DM	46.83	45.68	45.48	43.45	43.60	43.56	0.24	3.46
Ash, %DM	1.22	1.20	1.10	1.12	1.18	1.00	0.35	0.11
Crude fat, %DM	38.14	39.47	38.50	38.43	39.26	41.35	1.94	3.43
Nitrogen Free extract, %DM	13.81	13.65	14.92	17.40	16.46	16.09	1.25	4.45

Different superscripts (a, b, c, d and e) indicate significant ($P<0.05$) differences between treatments

NC: Negative control; PC: Positive control

There were significant ($P<0.05$) differences between treatments on some external and internal egg characteristics, whereas egg composition did not differ ($P>0.05$) between treatments. Egg size (9.72 – 10.37g) was largest ($P<0.05$) in quails on 2.5% EeAaL and least in those fed 7.5% EeAaL diet. Egg size, width, length, shell thickness, total egg laid and hen day production were influenced ($P<0.05$) by dietary treatments. There were no effects ($P>0.05$) of dietary treatments on egg shape index, surface area and % crack. Shell thickness was significantly ($P<0.05$) higher in quails fed 5% EeAaL followed by those on 7.5%, 0.02g/kg oxytetracycline and least in those fed control, 2.5% as well as 10% EeAaL diets. % cracked eggs reduced ($P>0.05$) at increasing level of EeAaL supplementation. Compared to the control, hen day production was higher ($P<0.05$) in quails fed diets supplemented with either synthetic or plant extract. There was increasing trend in hen day

production (82.76%) up to 7.5% EeAaL supplementation implying that EeAaL could stimulate more egg production. Lower hen day production were reported by Akinfenwa et al., 2011 (68.60 – 75.78%), Tuleun and Dashe, 2010 (62.71 - 77.02%), and Edache et al. 2018 (73.09 – 78.11%) in quails fed dietary lysine, mucuna seed and sweet potato meal, respectively.

On internal qualities, the impact of dietary treatments was observed on the yolk and shell weights as well as

yolk colour. Shell weight was lower ($P<0.05$) in quails fed EeAaL and oxytetracycline compared to those on the control diet, whereas yolk weight was significantly higher in EeAaL fed quails than in the control. This result indicated that EeAaL supplementation exerted significant influence on yolk weight. There was a rising trend in yolk colour as level of supplementation increases. The present results are consistent with previous observations reported (Agiang et al., 2011; Akintunde et al., 2017; Edache et al., 2018; Malik et al., 2018) on quail egg quality following various supplementations. Quail egg composition did not differ

($P > 0.05$) between dietary treatments, indicating that EeAaL supplementation had no adverse effects on egg composition in line with previous reports (Oko et al., 2013).

CONCLUSION

The inclusion of varying levels of ethanolic extract of *Aspilia africana* leaf (EeAaL) on quail diets exerted significant effects on the carcass and egg quality traits of Japanese quails. Though, quails could tolerate 10% EeAaL in their diets, improved egg characteristics were obtained at 7.5% supplementation. This study therefore recommends that up to 7.5% EeAaL should be supplemented into quail diets.

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