

EFFECTS OF SOME LEAFY VEGETABLES AND THEIR COMBINATIONS AS FEED ADDITIVES ON PERFORMANCE, EGG QUALITY AND SHELF- LIFE

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ABSTRACT

This study was aimed at exploiting the nutritive and preservative potentials of leafy vegetables on egg quality and shelf-life. 189 Lohman Brown Classic point-of-lay chickens at sixteen weeks with average weight of 1550-1620g were used in a 12-week feeding trials. The birds were allotted on weight equalization basis to seven feeding treatments (T1-T7). T1 was white maize without additive, T2 was yellow maize without additives while T3-T7 were white maize, fortified with synthetic yolk colourant (T3); fortified with *Corchorus olitorius* (T4); fortified with *Talinum fruticosum* (T5); fortified with *Chromolaena odorata* (T6); and fortified with a blend of *Corchorus*, *Talinum* and *Chromolaena* at 0.33% each (T7) respectively. Results obtained from this study showed that the dietary treatments had effects on the performance parameters. Chickens on T3 recorded the highest ($P<0.05$) Hen-day production followed by T1 and T7 with values ranging from 69.95-89.57%. Similar pattern was observed with Feed/dozen egg. Chickens on T2 had the heaviest egg (63.41 g) followed by T3 (59.45 g) and T1 (58.56 g). The pH values of eggs from T1, T2, T4 and T6 increased as the eggs age while it decreases in T3 and T7. The microbial load in eggs from chickens fed with T1 and T5 increased, T3 remain stable whereas eggs from T2, T4, T6 and T7 decreased with storage days. This study revealed that the blend of *Corchorus*, *Talinum* and *Chromolaena* can be used as alternative source of feed additive without deleterious effects on laying performance, quality characteristics and storage life.

Keywords: Additives, Microbial load, Performance, Point-of-lay chickens, pH value.

INTRODUCTION

The movement of eggs from farms to end users usually take a period of time due to bad road, use of unserviceable vehicles, vehicular breakdown, poor logistics and the situation could be more apparent during a period of lockdown as experienced with the Novel Coronavirus disease (COVID-19) pandemic currently ravaging the whole world (WHO, 2020) as well as banditry and kidnapping in the country. As the lockdown played the major role to counter the spread of COVID-19, it however caused significant damages owing to the restrictions on economic activities, reduction in demand of different commodities, wastage of the produce due to the closure of transport and market chains, distress sale of the produce (Mayur et al., 2020). It should be noted that as an egg is laid, the internal quality of eggs begins to deteriorate due to loss of moisture, carbon dioxide and entrance of bacteria via the eggshell pores (Nongtaodum et al., 2013). The quality of the egg, once laid, cannot be improved. Hence, efforts to maintain its quality must start right at the moment as eggs stored longer than necessary may impact negatively on egg quality due to the transition period. Therefore, the improvement of quality and extension of shelf life of edible eggs are parts of the strategies employed to safeguard public health and increase profitability in egg production. Various research

efforts have been made to improve the quality and shelf life of edible eggs through oil coatings (Atabo, et al., 2018), vitamin-mineral premixes and use of phyto-additives (Balogun and Odunsi, 2020), antimicrobial coating, ozone and ultrasound applications (Muhammed and Cengiz, 2021).

The egg yolk colour is one of the main indicators of egg quality affecting consumer's preference. Phyto-genic additives are commonly used as colourings in laying hens to affect the egg yolk colour as laying hens cannot synthesize egg yolk pigments (Englmaierova et al., 2014). The source of these pigments may be natural or synthetic colourings (ethyl ester of β -apo-8'-carotenoic acid and canthaxanthin known as Carophyll yellow and Carophyll Red) that are more economical but also potentially dangerous to human health. Hence, there is need to embrace natural colourings like carotenoid which can be sourced from herbs, spices and other shrubs which has positive effect on laying performance and egg quality (Gerzilov et al., 2015). *Chromolaena odorata* (Siam) weed is a perennial shrub that belongs to the aster family. It is a highly successful plant that has colonized diverse ecological area of tropical lands. It is generally regarded as poisonous to animals and thus not recommended as a livestock feed (Anyanwu et al., 2017). However, some studies showed its benefits in low concentrations, such as up to 5% for egg-laying chickens which also improved yolk colour (Fasuyi et al., 2005). *Corchorus olitorius* (jute) leaves are rich sources of potassium, iron, copper, manganese and zinc as well as high energy values essential in human and animal nutrition. Oboh et al. (2009) also reported jute leaves as a rich source of carotene. *Talinum fruticosum* (water leaf) is one of the underrated and undervalued plants in Nigeria. It flourishes more during the rainy season. It has been reported that a dose of the methanol extracts of *Talinum* suppresses oxidative damage in the liver cells and cerebral functions in Swiss Albino mice (Ezekwe et al., 2013). Additionally, they possess a stimulatory effect on the digestive system through increasing the production of digestive enzymes and improving feed utilization efficiency by enhancing liver functions (Abou-Elkhair et al., 2014). Based on the findings by these researchers, the hypothesis that these additives may positively affect laying performance and egg quality is most likely to be valid. However, the number of in-vivo studies in laying hens is still limited.

In other to exploit the nutritive potentials (proteins, amino acids, fats, vitamins, and minerals), eggs must be conserved over the marketing period. It usually takes weeks from the time the eggs are laid and purchased, before they are eventually consumed. These time frame may impact negatively on the egg qualities as the internal qualities can be altered as the eggs age (Monira et al., 2003). Therefore, quality loss in eggs is an unavoidable phenomenon that takes place continuously over time and that can be worsened by several factors (Giampietro-Ganeco et al., 2015). Most studies (Lana et al., 2018) employed storage duration of 30 days maximum. However, in this study, freshly-laid eggs were stored to a maximum of 56 days under room temperature with the aim of evaluating the effect of *Corchorus olitorius*, *Talinum fruticosum* and *Chromolaena odorata* (used singly and in combination as feed additives) on performance, egg quality and shelf-life extension amidst restrictions on economic activities.

MATERIALS AND METHODS

Experimental Site

This study was carried out at the Teaching and Research Farm of Yaba College of Technology, Epe, Nigeria. The farm is situated along Epe-Ijebu Ode road on Latitude 6.58 °N, Longitude 3.98 °E. It lies in the low land rain forest vegetation zone within the savannah agro ecological zones with altitude of 42 m above the sea level. The average rainfall of 1694 mm and temperature of 27.10 °C exist during the experimental period (Google Earth, 2019).

Preparation of the Test Ingredients

The test ingredients were *Corchorus olitorius*, *Talinum fruticosum* and *Chromolaena odorata*, and. The *Corchorus olitorius* used in this study were planted in the college farm. *Chromolaena odorata* and *Talinum fruticosum* were harvested before flowering, chopped and air-dried. The air-dried samples were milled individually at 0.2 mm diameter sieve using Kenwood blender mode and stored in plastic containers.

Determination of Proximate Composition

Moisture content was determined by placing the samples in an oven for 24 h at 60 °C. Proximate compositions of the various samples were determined according to the standard procedures of AOAC (2005). Samples were analyzed for crude fibre, ether extract and ash. The nitrogen fraction of the samples

was determined using the Kjeldahl method and crude protein determined by multiplying the N value by 6.25.

Phytochemical Screening

Tests were carried out on the powdered specimens using standard procedures to identify the phytochemical constituents according to Harborne (1994).

Experimental Bird Management and Design

One hundred and eighty-nine (189) Lohman Brown Classic point-of-lay hens at sixteen weeks with average weight of 1550-1620 g were used in this study. The house and cages were washed and disinfected before the arrival of the hens. The layers were allotted on weight equalization basis in a complete randomized design. Each treatment was replicated nine times with three birds per replicate. The experimental birds were housed replicate-wise in randomly allotted tiers of the battery cages (each cage of dimension 1.2 × 0.9 × 0.5 m) equipped with separate waterers and feeders with three birds per cell). The cages were placed in a well-ventilated building under natural lighting and uniform management. Standard management practices were followed during the experiment. Birds were fed with experimental diets and clean water provided based on daily requirement. The feeding trials lasted for 12 weeks. Diets were formulated according to the nutrient requirement of laying chicken (NRC, 1994). Birds were initially given standard grower mash to acclimatize and to achieve 10% production before introducing experimental diets.

Experimental Diets and Treatments

Seven experimental diets (T1-T7) were formulated with inclusion of 1% of the test ingredients as stated below:

T1 = white maize (without colourant)

T2 = Yellow maize (without colourant)

T3 = white maize and synthetic yolk colourant.

T4 = white maize and Corchorus leaf meal at 1% inclusion

T5 = white maize and Talinum leaf meal at 1% inclusion

T6 = white maize and Chromolaena leaf meal at 1% inclusion.

T7 = white maize and a blend of (Corchorus, Talinum and Chromolaena) at 0.33% each.

Routine medication and vaccination were followed throughout the experimental period. Table 1 shows the gross composition of the experimental diet.

Table 1: Gross composition of experimental diet.

Components	T1	T2	T3	T4	T5	T6	T7
Maize	50.00	50.00	50.00	50.00	50.00	50.00	50.00
SBM	21.00	21.00	21.00	21.00	21.00	21.00	21.00
Wheat Offal	16.00	16.00	16.00	16.00	16.00	16.00	16.00
Fish (72%)	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Limestone	8.00	8.00	8.00	8.00	8.00	8.00	8.00
Bone meal	3.00	3.00	3.00	3.00	3.00	3.00	3.00
Table salt	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Premix 1 (without colourant)	0.30	0.30	-	-	-	-	-
Methionine	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Lysine	0.20	0.20	0.20	0.20	0.20	0.20	0.20
Premix 11 (with colourant)	-	-	0.30	-	-	-	-
CLM	-	-	-	1.00	-	-	-
TLM	-	-	-	-	1.00	-	-
CHLM	-	-	-	-	-	1.00	-
CTCHLM	-	-	-	-	-	-	1.00
Total	100	100	100	100	100	100	100
Determined analysis (%)							
Crude protein	16.98	16.98	16.98	16.98	17.00	16.98	17.00
Crude fibre	3.42	3.47	3.47	3.48	3.50	3.51	3.55
Ether extract	3.29	3.30	3.30	3.29	3.30	3.29	3.31
Calculated analysis (%)							
Calcium	3.66	3.66	3.66	3.66	3.66	3.66	3.66
Phosphorus	0.53	0.54	0.54	0.53	0.54	0.53	0.54
Lysine	1.04	1.04	1.04	1.04	1.04	1.04	1.04
Methionine	0.48	0.48	0.48	0.48	0.48	0.48	0.48
ME (Kcal/kg)	2539	2558	2539	2539	2539	2539	2539

Layers premix: Vit. A 10,000,000 iu, Vit. D₃ 2, 000,000 iu, Vit. E 23,000 Mg, Vit. K₃ 2, 000mg, Vit. B₁ 3000mg, Vit. B₂ 6000mg, Niacin 50,000mg, Calcium Pantothenate 10,000mg, Vit. B₆ 5,000mg, Vit. B₁₂ 25mg, Folic acid 1000mg, Biotin, Chloride 400,000mg, Mn 120,000mg, Fe 100,000mg, Zn 80,000mg, Cu 8,500mg, Co 1500mg, Se 300mg, Anti-oxidant 120,000mg. CLM- Corchorus leaf meal, TIM- Talinum leaf meal, CHLM- Chromolaena leaf meal, CTCHLM- Corchorus, Talinum and Chromolaena leaf meal

Data collection

Performance

Weight Gain

Birds in each replicate were weighed at the beginning of the experiment, while subsequent weighing was done on weekly basis. The weight gains were determined by the difference between the two consecutive weighing.

Feed Intake

Known quantity of feed was given to each replicate group at the beginning of each week and the left over at the end of seven days period was subtracted from the amount supplied. The differences were recorded as feed intake and by dividing the number of birds in each of the replicate to obtain the feed intake per bird.

Feed/dozen Egg

Feed consumed in relation to egg laid, were calculated for each of the treatments.

Laying performance

Records of daily egg production on replicate basis were kept starting from the two weeks in lay to 8 weeks. Weekly egg production per replicate were pooled and expressed as percentage Hen-day egg production. Hen-day at a given period of time were calculated as the percentage of the ratio of number of eggs laid to the number of hen days (NAPRI, 2002). Record of feed intake was taken and used to calculate feed to dozen egg laid.

$$\text{Hen-day egg production (\%)} = \frac{\text{Number of eggs laid}}{\text{Number of birds housed}} \times 100$$

Determination of internal and external egg qualities

Nine (9) eggs from each treatment were picked at random weekly, egg qualities assessment was done within 24 h of lay on external and internal qualities for a period of five weeks.

Internal egg qualities

The height of the egg white (albumen height) was measured off the chalazae at a point above mid-way between the inner and other circumference of the thick white with a P6085 spherometer having an accuracy of 0.01 mm. The yolks were separated from the albumen using a plastic egg separator. Yolk weight was measured using sensitive weighing scale (Saltex ® electronic balance with sensitivity of 0.01 g). The albumen weight was taken as the difference between the eggs weight and the combined weight of the yolk and dry egg shell for individual egg sample. Yolk colours were scored for individual egg yolk (on treatment basis) by comparing the colour of yolk with the colour of the chips of a Hoffman-La Roche yolk colour fan (Hoffman-LaRoche, 1984). Haugh unit was calculated using the egg weight and albumen height for individual egg sample using the formula described by (Haugh, 1973).

$$\text{H.U.} = 100 \log (\text{H} + 7.5 - 1.7 \text{W}^{0.37})$$

Where:

H.U. = Haugh unit

H = Albumen height

W = Weight of egg

Determination of external egg qualities

The weights of the eggs laid were determined with a sensitive weighing scale (saltex® electronic balance) to the nearest 0.01 g on treatment basis. The egg lengths were taken as the longitudinal distance between the narrow and the broad ends using a digital vernier caliper with an accuracy of 0.1 mm. The diameter of the widest cross-sectional region was taken as the egg breadth using a digital vernier caliper with an accuracy of 0.1 mm. Egg shape index was calculated from the measurement of egg length and egg breadth from individual egg sampled. It is the ratio of egg breadth to its length as expressed below:

$$\text{Egg shape index} = \frac{\text{Breadth of egg}}{\text{Length of egg}} \times 100$$

Shell weight egg shells were air-dried for 72 h in egg trays and were weighed using weighing scale (Saltex® electronic balance with sensitivity of 0.01 g) while shell thickness was measured with a digital micrometer screw gauge to the nearest 0.01 mm.

Determination of pH of the egg samples

Raw eggs were collected from the production line on treatment basis (12x7) on same day and labelled with dates at interval of 14, 28, 42 and 56 days respectively. The eggs were stored at room temperature. The pH and microbial load determination were carried out at interval of two weeks. The eggs were broken separately and homogenized in a piston-gap homogenizer at 100 bars. pH was measured using a pre-calibrated pH meter (TESTO 206) on the liquid egg.

Total heterotrophic count of bacteria in the egg samples

One mL of each homogenised sample (1 – 7) was added to 9 mL of sterile distilled water in a tube. 1 mL of this was transferred to a second tube also containing 9 mL of sterile distilled water. The procedure was repeated up to the 8th tube to give 10⁻⁸ dilution. One mL each from dilutions 10⁻⁶ and 10⁻⁸ was placed in sterile petri dish and about 10 mL of plate count agar (PCA) was added. The plates were swirled gently for even distribution of the organisms and were allowed to set. The plates were incubated at 35 °C for 24 – 48 hours in an inverted position (Reasoner, 2004).

Statistical data analysis

Data generated from performance and egg quality traits were subjected to Analysis of Variance (ANOVA) while data on pH and microbial load were analyzed using Minitab (version 17.0) and Significant Means were compared using Duncan's multiple range test (SAS,2002).

RESULTS

Effect of phytogenic meals on the performance of laying bird

Performance of hen fed experimental diets supplemented with CLM, TLM and CHLM is show in Table 2. The results obtained showed a significant (P<0.05) increase in the daily feed intake compared to T1. Feed/dozen egg production was highest (P<0.05). Hen day production of hens in T3 was higher compared to other treatments though T6 and T7 had similar HDP. T3 laid highest number of eggs followed by T1 while T6 and T7 were similar. Feed/dozen egg of hens on T3 was the heaviest followed by T1 while the least was recorded for hens on T5.

Table 2: Effects of phytogenic meals on the performance of laying birds

Parameters	T1	T2	T3	T4	T5	T6	T7	SEM	P value
Hen-day egg production (%)	79.08 ^b	61.48 ^d	89.57 ^a	58.87 ^e	41.00 ^f	69.00 ^c	69.95 ^c	3.21	<.0001
Total egg produced	1190 ^b	943.33 ^d	1353.33 ^a	860.00 ^e	623.33 ^f	1056.67 ^c	1060 ^c	48.79	<.0001
Daily Feed Intake (g/bird)	115.56 ^e	116.25 ^d	116.25 ^d	116.57 ^b	115.61 ^e	116.48 ^c	116.80 ^a	0.1	<.0001
No of Egg Laid (%)	44.68 ^b	34.00 ^d	50.03 ^a	33.37 ^d	22.93 ^e	37.66 ^e	38.85 ^c	1.8	<.0001
Total feed intake (g/bird)	6047	6510	6528	6458	6523	6541	1.36	0.46	
Feed/Dozen Egg	1.73 ^d	2.29 ^b	1.56 ^e	2.34 ^b	3.38 ^a	2.07 ^c	2.01 ^c	0.12	<.0001

Means on the same row having different superscript are significantly different ($P < 0.05$)

T1 = white maize (without colouration), T2 = Yellow maize (without colouration), T3 = Synthetic yolk colourant based on manufacturer requirement, T4 = Corchorus leaf meal at 1% inclusion, T5 = Talinum leaf meal at 1% inclusion, T6 = Chromolaena leaf meal at 1% inclusion, T7 = CTC leaf meal (Corchorus, Talinum and Chromolaena) at 0.33% each.

Effect of CLM, SLM and TLM on egg qualities of laying bird

Effect of CLM, TLM and CHLM supplementation on external and internal qualities traits are presented in Table 3. Results obtained showed that heaviest average egg mass (63.41 g) was laid by hens in T2 while the least of 55.79 g was recorded in T5. There were no significant effects of the additive on egg shape index (egg breadth ad length). Shell thickness was significant affected by fed diets, T2, T6 and T7 had thicker shells compared to other hens. The inclusion of CLM, TLM and CHLM affected the shell weight, shell thickness, yolk weight and yolk colour. The highest shell weight was observed with hen on CHLM diet (T5) and a combination of the three leaf meals (T7). Yolk colour improved significantly ($P < 0.05$) at the inclusion of CLM, CHLM and TLM respectively. Yolk pigmentation in hens on T4, T6 and T7 laid eggs with yellow yolk, hens on T2 had a deeper yellow yolk and hens fed T3 laid a golden yellow yolk while hens on T1 laid egg with pale yolk. Egg weight, length, height, breadth, albumen weight, albumen height, and haugh unit were not affected.

Table 3: Effects of phytogenic meals on the egg qualities of laying birds

Parameters	T1	T2	T3	T4	T5	T6	T7	SEM	P-value
Egg Weight(g)	58.56	63.41	59.45	58.21	55.79	59.13	56.93	0.93	0.51
Egg Breadth(mm)	44.18	41.31	43.27	42.87	44.22	43.90	44.39	0.54	0.8
Egg Length(mm)	54.34	54.10	56.53	56.06	55.25	56.88	56.6	0.34	0.11
Albumen Height(mm)	9.58	9.60	9.69	8.73	9.00	9.65	9.82	0.14	0.38
Albumen Weight(g)	36.04	37.36	34.78	35.60	36.82	36.43	38.01	0.39	0.4
Yolk colour	1 ^e	6 ^b	8 ^a	5 ^c	5 ^c	3 ^d	5 ^c	0.46	<.0001
Yolk Weight(g)	13.74 ^b	13.28 ^b	13.50 ^b	13.50 ^b	13.56 ^b	14.86 ^a	13.10 ^b	0.16	0.07
Shell Thickness(mm)	0.46 ^b	0.51 ^b	0.46 ^b	0.48 ^b	0.44 ^b	0.57 ^a	0.49 ^b	0.01	0.01
Shell Weight(g)	6.03 ^b	6.09 ^{ab}	6.03 ^b	6.12 ^{ab}	6.48 ^a	6.11 ^{ab}	6.21 ^{ab}	0.05	0.22
Haugh Unit	91.66	92.76	99.41	99.48	96.25	100.06	99.45	1.23	0.33

Means on the same row having different superscript are significantly different ($P < 0.05$)

T1 = white maize (without colouration), T2 = Yellow maize (without colouration), T3 = Synthetic yolk colourant based on manufacturer requirement, T4 = Corchorus leaf meal at 1% inclusion, T5 = Talinum leaf meal at 1% inclusion, T6 = Chromolaena leaf meal at 1% inclusion, T7 = CTC leaf meal (Corchorus, Talinum and Chromolaena) at 0.33% each.

Effects of phytogenic meals on egg pH and microbial load over storage duration.

pH values of eggs were significantly affected by number of days in storage. It was observed that the pH values of eggs from T1, T2, T4 and T6 increased as the eggs age while the values in T3 and T7 decreased with aging. Variation were between slightly acidic to alkaline pH (Table 4). The microbial load in eggs fed T1 and T5 increased with storage days, eggs from T2, T4, T6 and T7 decreased while that of eggs T3 remain stable for the period of storage (Table 5).

Table 4: Effects of phytogenic meals on pH value of eggs at storage interval

Treatments	pH values at intervals of days				SEM	P value
	14	28	42	56		
T1	6.98 ^f	7.76 ^c	7.96 ^b	7.98 ^c	0.03	0.00
T2	7.18 ^{ef}	8.01 ^a	8.23 ^a	8.51 ^a	0.03	0.00
T3	8.20 ^b	7.67 ^d	7.12 ^e	6.74 ^e	0.14	0.00
T4	7.22 ^e	7.32 ^e	7.86 ^c	8.03 ^c	0.02	0.00
T5	8.83 ^a	7.93 ^b	7.66 ^d	7.56 ^e	0.04	0.00
T6	7.57 ^d	7.66 ^d	7.98 ^b	8.24 ^b	0.04	0.00
T7	8.05 ^c	7.98 ^{ab}	7.88 ^c	7.86 ^d	0.02	0.00

Means on the same row having different superscript are significantly different ($P < 0.05$)

T1 = white maize (without colouration), T2 = Yellow maize (without colouration), T3 = Synthetic yolk colourant based on manufacturer requirement, T4 = Corchorus leaf meal at 1% inclusion, T5 = Talinum leaf meal at 1% inclusion, T6 = Chromolaena leaf meal at 1% inclusion, T7 = CTC leaf meal (Corchorus, Talinum and Chromolaena) at 0.33% each.

Table 5: Effects of phytogetic meals on microbial load in eggs at storage interval

Treatments	Microbial load x 10 ⁵ (cfu/ml) at intervals of days				SEM	P value
	14	28	42	56		
T1	0.00 ^d	5.60 ^a	8.90 ^a	9.60 ^a	0.18	0.00
T2	4.00 ^b	3.40 ^c	3.23 ^c	2.10 ^c	0.73	0.00
T3	0.50 ^{cd}	0.50 ^g	0.50 ^d	0.60 ^d	0.18	0.00
T4	1.50 ^c	1.40 ^e	0.60 ^d	0.10 ^d	0.13	0.00
T5	0.00 ^d	2.60 ^d	5.50 ^b	8.00 ^b	0.13	0.00
T6	1.50 ^c	1.00 ^f	0.20 ^e	0.10 ^d	0.13	0.00
T7	6.00 ^a	4.30 ^b	0.60 ^d	0.20 ^d	0.65	0.00

Means on the same row having different superscript are significantly different ($P < 0.05$)

T1 = white maize (without colouration), T2 = Yellow maize (without colouration), T3 = Synthetic yolk colourant based on manufacturer requirement, T4 = Corchorus leaf meal at 1% inclusion, T5 = Talinum leaf meal at 1% inclusion, T6 = Chromolaena leaf meal at 1% inclusion, T7 = CTC leaf meal (Corchorus, Talinum and Chromolaena) at 0.33% each.

DISCUSSION

The performance parameters showed that hen-day production, egg produced, percentage egg production, feed intake and feed-dozen egg were influenced by dietary treatment. T3 performed better than other treatments. This could be due to the presence of essential nutrients such as crude protein, essential fatty acids, minerals and carbohydrates in stimulating yolk formation and ovulation (Cabuk et al., 2006). However, none of the egg quality parameters were influenced except yolk weight and colour, shell weight and thickness. These observations are in line with Zhao et al., (2005) who reported a significant improvement in eggshell quality and bone mass after supplementation with daidzein (a phytoestrogen component) as a result of the regulation of calcium metabolism for shell formation (Arjmandi et al., 2002). In similar studies, Ghasemi et al., (2010) reported that no significant effects were obtained by supplementation of 5 and 10 g kg⁻¹ garlic powder on albumen index, egg shell thickness, and Haugh unit values in layer hen but yolk colour was increased.

The values obtained for hen-day egg production in this study were similar to the values of 75-79% reported by Dey et al., (2011) who worked with neem leaf meal as a hypocholesteromic dietary additives on laying pullets. Olayemi et al., (2019) reported a value range of 78-80 for percentage hen-day production at mid-laying phase when Moringa leaf meal was used as replacement for soybean at 10% in laying birds. The results of this study showed production was economical in agreement with Oluyemi and Robert, (2007) that hen-day egg production becomes uneconomical when below 65%. The feed consumed per dozens of eggs is the yardstick used for the measurement of feed efficiency (Oluyemi and Robert, 1979). The best feed efficiency was recorded at T3 (Synthetic yolk colourant) inclusion while the poorest feed efficiency was recorded for T4 (Corchorus leaf meal). This could be as a result of phytochemical constituents in the leaf meal as earlier reported (Olayemi et al., 2019). As observed feed intake was normal when compared with control. This implies that feed intake may not be negatively affected at these inclusion levels. The total feed consumed per dietary treatment also followed the same trend.

The weight of the eggs obtained in this study is consistent with the standard egg value of 58 g and slightly higher in T2 (63.41), T6 (59.13) and T3 (59.45) according to Olugbemi et al. (2009). The similarity in egg weight values obtained in this study by the experimental birds indicated that the dietary inclusion of CLM, TLM and CHLM in the diets had no negative effect on the weight of the eggs. Aro et al., (2009) in their work on siam weed leaf meal as egg yolk colourant for laying hens also reported similar egg weight of 58.4-

63.9g. Therefore, the range of values reported for egg weight for hens in this study implied that the diets were adequately balanced. The egg shell thickness values of $\geq 0.38\text{mm}$ falls within normal range as reported by Fasuyi et al. (2005). The result obtained for shell weight revealed that birds dietary leaf meal had better values of shell weight than those on T1 (control), T2 and T3. The values in this study ranged from 6.03-6.48g which is higher than values obtained by Garba et al. (2010). This implies that eggs with strong shells are less likely to crack during egg collection, storage and transportation. The egg shell thickness values of $\geq 0.38\text{mm}$ falls within normal range as earlier documented.

The result obtained for shell weight revealed that birds dietary leaf meal had better values of shell weight than those on T1(control), T2 and T3. The values in this study ranged from 6.03-6.48g and were higher than values obtained by Garba et al. (2010). This implied that eggs with strong shells are less likely to crack during egg collection, storage and transportation. The range of albumen weight values obtained (35.6-38.01g) was similar to the values of 35.87-37.27g reported by Esonu et al. (2005). This showed that inclusion of Corchorus, Talinum and Chromolaena in the diets of the birds had no negative effect on the albumen weight, since there was no abnormal decrease or increase in the values. The yolk weights (13.1-14.86g) obtained in this study were similar to range of 14.21-14.85g obtained earlier in laying birds fed neem leaf diets (Olayemi et al., 2019). Therefore, it could be noted that the addition of CLM, TLM and CHLM in the diet of the birds did not have a drastic effect on the yolk weight of the eggs of the birds. The highest value for yolk colour was observed in T3 with 8, while the lowest was obtained in the control with 1 on the colour fan chart. The colour score of eggs from CLM, TLM and CHLM diets indicate that there was improvement in yolk colouration due to the xanthophylls content in them as compared to the control. This agreed with the report of Kaijage (2003) on sunflower seed meal with moringa in egg strain chicken who observed deeper yolk colour. The colour score of egg yolk indicated improved pigmentation in the diets due to its rich xanthophylls content which agrees with studies conducted by Akintomide and Onibi (2018). However, this might be attributed to the presence of xanthophylls and carotenoid pigments in the leaf meal diets as confirmed by (Moyo et al., 2011).

The range of values obtained for Haugh unit in this study was higher than values of 83.92-84.78 reported by (Olabode and Okelola, 2014). The average values of Haugh unit obtained in this study conformed to standard commercial egg production guides. Oluyemi and Roberts (2007) also reported that Haugh unit score of 72 and above has been graded as the best quality. Haugh units of 72 and above are indications of freshness in eggs – an index of ability of albumen to remain viscous (Uchegbu et al., 2011). The non-significant effect noticed in the Haugh unit values was an indication of an improvement in the albumen height and quality caused by feeding CLM, TLM and CHLM to the laying birds. The results agreed with the findings of Austic and Nesheim (1990) that Haugh unit was not influenced mainly by diet but by albumen consistency which was dependent on the age of the hen, genetics and storage of the eggs. The height of the albumen determines the Haugh's unit of the egg. When the albumen height is high, the value of the Haugh unit is greatly enhanced and better is the quality of the egg (Oluyemi and Robert, 2007). This suggests that the eggs produced in this study are of better quality. However, the result also showed that inclusion of CLM, TLM and CHLM had no negative effect on the external and internal egg quality characteristics of the laying birds.

Effect of inclusion of the different additives on pH values and microbial loads as reported in Tables 4 and 5 shows that measures of food safety, hygiene and egg freshness ranked higher in guaranteeing quality in meeting consumer expectations. pH changes are related to the thinning of the albumen as evident sometime after lay, depending primarily on storage time and temperature. Eggs are relatively pH neutral with an initial pH value that can be as low as 7.6 at time of lay but with increasing alkalinity as the egg ages, and can reach pH of 9.2 (Stadelman and Cotterill, 1995). As the weeks in storage increases egg loss water, carbon dioxide and a subsequent increase in the pH and weakens the vitelline layer (Jin et al., 2011). As reported in Table 4, the diets had significant ($P < 0.05$) effect on pH values measured overtime. The results obtained were in agreement with the findings of Ryu et al. (2011) that albumen pH value ranges from 7.6 to 8.7. In this study, only eggs laid by hens on T3, T5 and T7 had pH range within the freshness category after eight weeks of storage. The stability in oxidation reactions could be as a result of phenolic compounds in herbal plants (Yulianti and Muharliien, 2020). As reported in Table 5, reduction in microbial load in eggs from hens fed diets supplemented with herbal plants could be as a result of the antimicrobial properties contained in herbal

plants that work through the mechanism to decrease proliferation of pathogenic bacteria as well as antioxidant compounds which play a role in protecting feed lipids from damage caused by oxidation reactions thus increasing stability in egg contents Yulianti and Muharlieni, 2020).

CONCLUSION

This study has shown that the supplements had no negative influence on the parameters monitored. The leaf meals improved egg yolk colouration and could be used as egg yolk colourant with better shell thickness which is important during handling and transportation. pH values and microbial load in eggs can be maintained over a storage period. Therefore, the blend of *Corchorus olitorius*, *Talinum triangulare* and *Chromolaena odorata* can be used as an alternative feed additive without deleterious effect on performance and egg quality characteristics as well as consumers' acceptability.

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