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Comparative vitamin and mineral composition of horned melon as a substitute for commercial premix and egg quality characteristics of the fed layers

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ABSTRACT: Horned melon (*Cucumis metuliferus*), is a fruit rich in various phytochemical components important in the daily diet and has a high economic value which has not been fully exploited in poultry feeding. The objectives of the study are to evaluate proximate analysis, vitamin, and mineral content and determine the quality of eggs of layers fed horned melon meal. The horned melon fruits were harvested, oven-dried and milled into a horned melon meal (HMM). Proximate, vitamin, and mineral contents of horned melon were carried out using AOAC methods. Two hundred and forty Isa brown layers were grouped into four treatments with four replicates of fifteen birds each. Diets were formulated to contain HMM at 0, 25, 37.5 and 50% in replacement of commercial premix representing the four experimental diets. Two eggs were selected per replicate every week and cracked with the dull end of the knife into the yolk separator and egg qualities were analysed. The results showed that HMM had higher values ($p<0.05$) of vitamins A, D, E, K, B_1 B_2, B_{12}, Folic acid and Biotin (10940.00, 16670.00, 13280, 17190, 10510, 9410.00, 5110.00, 10830.00 and 4550.00 mg/kg, respectively), and Cobalt (51400.00 mg/kg), Iron (26202 mg/kg) and Cupper (5200 mg/kg), while the commercial premix values were 1680.00, 35.00, 3600.00, 960.00, 720.00, 2000.00, 3.50, 280.00, 20.00, 100.00, 16,000 and 3200 mg/kg, respectively. The results of egg quality analysis showed no significance ($p>0.05$) across the treatments. The ranges for egg length, egg width, egg weight, shell thickness, shell weight, yolk weight, yolk height, yolk colour score and albumen weight were 5.03 – 5.29, 3.94 – 4.48, 58.07 – 60.40, 0.43 – 0.45, 0.67 – 0.77, 28.67 – 31.00, 1.26 – 1.79, 4.06 – 6.75 and 18.08 – 22.40, respectively. It was concluded that horned melon meal is a promising feedstuff that can replace commercial premix in layer diets and its use of up to 50% replacement level did not adversely affect egg quality of the fed layers.

Keywords: Egg weight, horned melon, minerals, shell thickness vitamins, yolk weight.

INTRODUCTION

Balancing minerals and vitamins needed in poultry diets has become increasingly important to optimize diet compositions and meet the demands of modern poultry production. Vitamin and mineral nutrition play a crucial role in bird well-being and performance, along with supporting crucial body functions like growth, fertility, and immunity (Ewing and Charlton, 2007) and deficiencies of minerals and vitamins can even lead to death (Öztürk *et al.*, 2024). The main dietary inclusion of vitamins and minerals in poultry production is usually through premixes, though certain quantities are obtained from other major ingredients. Premixes are nutritional feed supplements added into the feed of farm animals to enhance their productivity and growth; it is a combination of vitamins, minerals and some other chemicals (Leeson and Summer, 2005).

In poultry, mineral deficiencies can be characterized by poor rate of hatch, inadequate skeletal development,

reduced embryo mineral content, or increased mortality rate (Öztürk *et al.*, 2024). Dietary minerals can be available in inorganic and organic forms. Inorganic trace mineral sources are the most common sources used in poultry diets. The benefits of inorganic trace minerals are improved growth rate, health, and reproduction (Boo *et al*., 2007; Nollet *et al*., 2008). Organic trace minerals are more bioavailable in several species of livestock and poultry compared to inorganic trace minerals. Also, organic mineral sources have been reported to enhance intestinal absorption of trace elements as they decrease interference from agents that form insoluble complexes with the ionic microelements (Brown and Zeringue, 1994).

Minerals are usually classified into two main categories; the micro- and macro-minerals depending on their required daily intake rather than their relative importance or physiological functions (Gupta and Gupta, 2014; Morris and Mohiuddin, 2023). Macro-minerals are typically needed at levels higher than 100 mg/day examples are calcium (Ca), phosphorus (P), magnesium (Mg), sulfur (S), sodium (Na) and potassium (K). On the other hand, micro-minerals are needed in amounts lower than 100 mg/day and include elements such as iron (Fe), zinc (Zn), iodine (I), selenium (Se), manganese (Mn), chromium (Cr), copper (Cu), molybdenum (Mo), fluorine (F), boron (B), cobalt (Co), silicon (Si), aluminium (Al), arsenic (Ar), tin (Sn), lithium (Li) and nickel (Ni) (Mohamed *et al*., 2023; Morris and Mohiuddin, 2023).

Vitamins are active, organic compounds that are essential for body functions and have a crucial impact with only small amounts in the feed to maintain physiological functions. A few vitamins play a role as co-enzymes (e.g. B-vitamins), and also as precursors of co-enzymes, in metabolic processes. The difference between vitamins and minerals is that vitamins do not provide energy (e.g. P is necessary for building up ATP, which is the energy source in living organisms), nor act as building substances (Jeroch *et al.,* 2008; Ewing and Charlton, 2007).

Vitamin and mineral specification for laying chicken according to Leeson and Summer (2005) is as follows Vitamin A, Vitamin D3, Vitamin E, Vitamin K, Thiamin, Riboflavin, Pyridoxine, Pantothenic acid, Folic acid, Biotin, Niacin, Choline, Vitamin B12, Manganese, Iron, Copper, Zinc, Iodine, Selenium (8,000 I.U, 3,500 I.U, 50 I.U, 3 I.U, 2 mg, 5 mg, 3 mg, 10 mg, 1 mg, 100 µg, 40 mg, 400 mg, 10 µg, 60 mg, 30 mg, 5 mg, 50 mg, 1 mg and 0.3 mg, respectively).

Vitamins and minerals are found in plant and animal-based feed materials fed to poultry; however, the quantity does not meet the requirements of the birds. Nowadays, vitamin supplements for chickens are frequently produced synthetically in the form of premixes and added to poultry diets in order to achieve a sufficient level of vitamins and minerals and prevent birds from becoming malnourished. A poultry premix completes the vitamin and mineral requirements of a diet (www.biomin.net>species>poultry). However, there are some fruits that are very high in minerals, vitamins and some other useful phytochemicals, examples include cucumbers and horned melon.

Horned melon, (*Cucumis metuliferus* E. Mey. Ex. Naudin) boasts an array of vitamins and minerals, many of which play a role in its ability to positively impact health (Achikanu *et al.*, 2020). *Cucumis metuliferus* is an exotic, peculiar-looking fruit from the central and southern regions of Africa. It's also known as horned melon, *kiwano* and African horned cucumber. When ripe, the thick outer skin is bright orange and covered in small spiny projections or horns. The inner flesh consists of a gelatinous, lime-green or yellow substance that houses a multitude of edible seeds (Šeregelj *et al*., 2022).

A matured 100 grams of wet horned melon has 44 kcal (calories) of energy. The other nutrients in the fruit are thiamine (0.025 mg), riboflavin (0.015 mg), niacin (0.565 mg), pantothenic acid (0.183 mg), vitamin B6 (0.063 mg), copper (0.020 mg), manganese (0.039 mg) and zinc (0.48 mg). Also, 100 grams of horned melon contains approximately 7.56 grams of carbohydrate, 1.26 grams of fat, 1.78 grams of protein, 88.97 grams of water, 3 micrograms of folate, 5.3 grams of vitamin C, 13 grams of calcium, 1.13 grams of iron, 40 grams of magnesium, 37 grams phosphorus, 123 grams of potassium, 2 grams of sodium (Achikanu *et* al., 2020).

Due to the presence of essential nutrients, minerals and organic compounds in the fruit, it got recognition by WHO, stating kiwano to be an essential fruit in fighting against illness and malnutrition. The health benefits of the kiwano are in its nutrients, including vitamins C, Fe and K. It also has smaller amounts of P, Mg, Zn, Ca, Cu, and Na. The seeds contain linoleic and oleic acid. A couple of anti-oxidants recognized in kiwano seeds are α-tocopherol as well as β-tocopherol. The yellowish-green coloured pigments are located in the seeds. Its pulp contains carotenoid and beta-carotene (Bina *et al*., 2019).

Egg quality defines those characteristics of an egg that affect consumer acceptability and preference. Components of quality include external and interior characteristics. The quality of the egg once it is laid cannot be improved. Egg quality has a genetic basis, but it is also affected by the laying hens' age, diets, and the hens' housing (Singh *et al.,* 2009; Silversides *et al.,* 2006). Proteins, fats, carbohydrates, vitamins, minerals and water are essential for a quality egg, but vitamins have an additional dimension. They are required at adequate levels to enable the animal to efficiently utilize all other nutrients in the feed. Therefore, optimum nutrition occurs only when the bird is offered the correct mix of macro- and micronutrients in the feed and is able to efficiently utilize those nutrients for its growth, health, reproduction and survival (McGee, 2004).

MATERIALS AND METHODS

The experiment was carried out at the Poultry Unit of

Table 1. Gross composition of diets replacing commercial premix with *Cucumis metuliferus* meal.

Ingredients	Treatment 1 0% HMM	Treatment 2 25% HMM	Treatment 3 37.5% HMM	Treatment 4 50% HMM
Premixes	0.3	0.225	0.1975	0.15
Horned melon meal	0.00	0.08	0.1125	0.15
Maize	53.00	53.00	53	53
Soybean meal	23.00	23.00	23.00	23.00
Corn bran	13.50	13.50	13.50	13.50
Oyster shell	8.00	8.00	8.00	8.00
Di-Calcium Phosphate	1.50	1.50	1.50	1.50
Common salt	0.30	0.30	0.30	0.30
Methionine	0.20	0.20	0.20	0.20
Lysine	0.20	0.20	0.20	0.20
Total	**100.00**	**100.00**	**100.00**	**100.00**
Calc. Energy	2608.50	2608.50	2608.50	2608.50
Calc. Protein	16.57	16.57	16.57	16.57

HMM = Horned Melon Meal.

Teaching and Research farm of Oyo State College of Agriculture and Technology, Igboora, situated at 80 kilometres (50 mi) north of Lagos with Latitude 7° 26' 1.79" N and Longitude 3° 17' 16.37" E. The town's average annual rainfall is 139.39 mm, humidity 76% and temperature of 22 – 29°C (https://weatherandclimate.com/nigeria/oyo/igbo-ora).

Horned melon processing

Fruits of horned melon were harvested from a farm in Ibadan, Oyo State, Nigeria. The fruits were thoroughly washed, slashed and oven dried for two days. Dried horned melon fruits were milled with a Laboratory milling machine (MG1803MIXER/GRINDER) and stored in a polythene bag for chemical analysis and incorporation into layers feed in replacement of commercial premix.

Feeding trial

Two hundred and forty (240), 12 weeks old birds were purchased from a reputable farm and raised to the laying phase. The birds were grouped into 4 treatments with four replicates of fifteen birds each**.** The experimental design adopted was a completely randomized design. The composition of the feed ingredients in the four experimental diets is shown in Table 1.

Chemical analysis of horned melon meal

Proximate analysis

Ash, crude fibre and moisture were determined according to the methods of AOAC (2000). Crude protein was determined using micro-Kjeldahl method. Nitrogen Free Extract (NFE) was determined using the method reported by Onyeike *et al.* (2001); % Non-Fat Extract = 100 – (% moisture + % crude fibre + %ash + % crude fat + % crude protein).

Vitamin analysis

The vitamin contents were determined using the modified method of AOAC (2005). One gram of sample was macerated with 200 ml of petroleum ether for 10 minutes and allowed to stand for 1 hour with intermittent shaking every 1 minute. The mixture was centrifuged for 5 minutes and 3 ml of the supernatant was transferred into three separate test tubes. Each supernatant in the test tube was evaporated to dryness and the residue re-dissolved with 0.2 ml of acetic anhydride/chloroform (1:1) and 2 ml of 50% trichloroacetic acid (TCA) in chloroform. The absorbance of the resulting solution was measured at 620 nm at 15 seconds and again after 30 seconds against a blank (mixture without the sample). A sample (5 g) was weighed into a test tube and 20 ml of petroleum ether was added and shaken for 5 minutes. The supernatant was decanted into another test-tube and the absorbance was read at 450 nm.

Analysis of mineral elements

The dried sample (5g) was dried ashed, and heated with 5.0 cm^3 of HNO3 and HCl (15 cm^3) was added. It was made up to 100 cm^3 with deionised water after filtration and mineral elements were determined using Microplasma Atomic Emission Spectrophotometer (MPAES).

Chemical composition of the commercial premix

Laboratory analysis was not carried out on the commercial premix. Vitamin and mineral constituents indicated on the label of the premix were compared against the result of the chemical analysis of the horned melon. International Unit (IU) values on the label were converted to mg/kg according to DSID (2023).

Egg quality analysis

Two eggs were selected at random from each replicate every week for five consecutive weeks to determine egg characteristics. The eggs were weighed and measured vertically and longitudinally with the aid of Venier callipers and then cracked with a dull edge knife centrally. The yolk was separated with a yolk separator and yolk color was measured with Roche yolk score. After the eggs were broken, the eggshell thickness was measured with a QCT shell thickness micrometre (TSS, England) at the equatorial area after the removal of shell membranes. The eggshell weight was determined after drying.

Data analysis

Results of the chemical analysis of horned melon meal were compared with values indicated on the label of the commercial premix with t-test analysis. However, data obtained on egg analysis were subjected to a one-way analysis of variance. Means showing significant ($p<0.05$) differences were separated using the Duncan Multiple Range Test (Duncan, 1955).

RESULTS

The result of the proximate analysis of the horned melon is presented in Table 2. The result revealed that horned melon meal has a very high moisture content of 14.23%, also crude protein, fat and fibre were also high (13.84, 9.55 and 9.80%, respectively). Ash content was 3.88, while carbohydrate (NFE) was 57.70%.

Vitamin and mineral contents of horned melon were analysed and compared with the values presented on the label of the commercial premix as shown in Table 3. The results showed that vitamins A, D, E, K, B_1 B_2, B_{12}, Folic acid and Biotin in the commercial premix (1680.00, 35.00, 3600.00, 960.00, 720.00, 2000, 3.5, 280 and 20.00mg/kg, respectively) were significantly lower ($p<0.05$) than values recorded by horned melon meal (10940.00, 16670.00, 13280, 17190, 10510, 9410.00, 5110.00, 10830.00 and 4550.00mg/kg, respectively). However, the pantothenic (2400 mg/kg) content of the commercial premix was higher ($p<0.05$) than values recorded for horned melon (1500 mg/kg), while values of vitamin B_{12} were similar ($p>0.05$) for the commercial premix and the horned melon.

Table 2. Proximate composition of horned melon meal (HMM).

Nutrients	Composition (%)
Moisture	14 23 ± 0.52
Crude fat	9.80 ± 0.33
Crude fibre	9.55 ± 0.76
Ash	3.88 ± 0.22
Protein	13.84 ± 0.81
NFE	57.70 ± 0.53

NFE = Non-Fat Extract.

Results of the mineral contents showed that Manganese (38400.00 mg/kg) and Zinc (24000.00 mg/kg) contents of the commercial premix were significantly higher ($p<0.05$) than analysed values of the horned melon meal (910.00 and 5190.00 g/kg, respectively). The horned melon meal had the highest values ($p<0.05$) for Cobalt (51400.00 mg/kg), Iron (26202 mg/kg) and Copper (5200 mg/kg) compared to those of commercial premix that were 100.00, 16000.00 and 3200 mg/kg, respectively. The antioxidant content of the horned melon meal (20300) was significantly higher ($p<0.05$) than the antioxidant content of the commercial premix. Other important constituents of horned melon meal that were not indicated on the label of the commercial premix are Calcium (365.83 mg/kg), Phosphorus (3756.96 mg/kg), Potassium (589.92 mg/kg), and Sodium (517.24 mg/kg),

Results for egg qualities obtained were presented in Tables 4 and 5 and revealed that there were no significant differences in all the parameters considered. However external egg quality of birds fed the control diet had the highest values of egg breath (4.68 cm), egg weight (60.40 g), eggshell weight (7.08 g), eggshell index (98.84), egg surface area (71.96 cm^2) and egg shape index (9.85). Treatment 3 (37.5% horned melon meal) had the lowest values for egg breath (3.93 cm), eggshell index (72.17) and egg surface area (66.03 cm^2) while those fed 50% horned melon recorded the lowest values for egg shape index (6.83) and highest value for egg length (5.29 cm)

Internal qualities shown in Table 5 revealed that the mean values for all the parameters were not influenced by the dietary treatments as there were no significant differences ($p>0.05$) across the treatments. Treatment 4 (50% horned melon meal) recorded 1.79 cm and 6.75 for yolk height and yolk colour score, respectively that were similar ($p>0.05$) to 1.26 cm recorded by treatment 1 for yolk height and 4.06 recorded by treatment 3 for yolk colour score.

DISCUSSION

The proximate result revealed that horned melon meal contained high protein, carbohydrate and fat. These nutrients will contribute to the proximate value of the feed.

Table 3. Mineral and vitamin content of the commercial premix and horned melon meal.

Nutrients	Commercial premix (mg/kg)	Horned melon fruit (mg/kg)	P-value
Vitamin A	1680.00[b]	10940[a]	0.00
Vitamin D	35.00[b]	16670[a]	0.00
Vitamin K	960.00[b]	17190[a]	0.00
Vitamin E	3600.00[b]	13280[a]	0.00
Vitamin B_1	720.00[b]	10510[a]	0.00
Vitamin B_2	2000.00[b]	9410[a]	0.01
Biotin	20.00[b]	4550[a]	0.00
Pantothenic Acid	2400.00[a]	1500[b]	0.02
Vitamin B_6	720.00	610.00	0.68
Vitamin B_{12}	3.50[b]	5110[a]	0.00
Folic Acid	280.00[b]	10830[a]	0.00
Vitamin C	NI	28101	
Cobalt	100.00[b]	51400[a]	0.00
Manganese	38400.00[a]	910[b]	0.00
Iron	16000.00[b]	26202[a]	0.02
Zinc	24000.00[a]	5190[b]	0.00
Copper	3200.00[b]	5200[a]	0.01
Antioxidant	50[b]	20300[a]	0.00
Choline chloride	150.00	ND	
Phosphorus	NI	365.83	
Potassium	NI	3756.96	
Calcium	NI	589.92	
Sodium	NI	517.24	

[ab] means differently subscripted along the same row are significantly different from one another ($p<0.05$). ND = Not Determined; NI = Not Indicated.

Table 4. External egg characteristics of layers fed the four experimental diets.

Parameters	T1 (0% HMM)	T2 (25% HMM)	T3 (37.5% HMM)	T4 (50% HMM)	SEM	P-value
Egg length (cm)	5.03	5.25	5.03	5.29	0.11	0.85
Egg breadth (cm)	4.68	4.13	3.94	4.03	0.16	0.40
Egg weight (g)	60.40	58.67	59.18	59.00	1.39	0.91
Egg shell thickness (cm)	0.43	0.45	0.44	0.43	0.63	0.61
Egg shell weight (g)	7.08	6.67	6.67	6.67	0.41	0.25
Egg shell index	98.84	78.72	72.17	76.25	5.75	0.51
Egg surface area (cm^2)	71.96	70.49	66.03	69.15	1.88	0.75
Egg shape index	9.85	9.44	9.75	6.83	0.55	0.27

T = treatment; HMM = Horned Melon Meal.

This attribute compares horned melon with some vitamin/mineral concentrates. The high moisture content of the horned melon may be related to its hygroscopic nature, hence, should be processed and stored in an air-tight package as obtainable for the commercial premix. The proximate values obtained from this study were higher than the values reported by Zheng and Wang (2001), but similar to values obtained by Achikanu *et al.* (2020).

Vitamin and mineral contents vary significantly between commercial premix and horned melon meal; horned melon had higher values for majority of the nutrients. Also, nutrients in horned melon may be more reliable because organic trace minerals are more bioavailable in several species of livestock and poultry compared to inorganic trace minerals (Brown and Zeringue, 1994). Mineral and vitamin contents of horned melon are higher than vitamin and mineral specification for laying chicken according to Leeson and Summer (2005). Calcium (365.83 mg/kg),

Table 4. Internal egg qualities of layers fed the four experimental diets.

Parameters	T1 (0% HMM)	T2 (25% HMM)	T3 (37.5% HMM)	T4 (50% HMM)	SEM	P-value
Yolk weight (g)	28.67	31.00	30.11	30.34	0.52	0.19
Yolk height (cm)	1.26	1.58	1.59	1.79	0.06	0.60
Yolk color score	6.50	5.33	4.06	6.75	0.47	0.15
Albumen weight (g)	20.25	21.00	22.40	18.67	2.27	0.46

T = treatment; HMM = Horned Melon Meal.

Phosphorus (3756.96 mg/kg), Potassium (589.92 mg/kg), Iron (26202 mg/kg) and Sodium (517.24 mg/kg) reported in this study were higher than previously reported values. Rudrappa (2019) reported that fresh fruits of horned melon contain 123 mg/100g of K, 1.13 mg/100g of Fe, 37 mg/100g of P and 2 mg/100g of Na. While Achikanu *et* al. (2020) reported 13 mg calcium, 1.13 mg iron, 40 mg magnesium, 37 mg phosphorus, 123 mg potassium, and 2 mg sodium per 100 grammes of fresh horned melon. The values presented by these authors were from fresh horned melon. The benefits of high K intake in animal bodies include the prevention of stroke, coronary heart disease and hypertension (Weaver, 2013) while Iron plays a role in oxidation metabolism, transport and cellular proliferation (Nair and Iyengar, 2009).

The results obtained from the egg quality study proved that horned melon provided enough vitamins and minerals that support egg production and the maintenance of egg quality. The rich vitamins and minerals content of horned melon might have assisted in supporting the quality of the eggs of the fed birds. Major nutrients contained in horned melon (Iron, Magnesium, Vitamin C, Phosphorus, and Vitamin B6) assisted the performance of birds-fed diets containing horned melon. Other minor nutrients that might have contributed include Vitamin A, Vitamin B5, Vitamin B3, Vitamin B2 and beta-carotene (Usman *et al.,* 2015).

The obtained results in this study showed that the vitamin/mineral content of horned melon was enough to support the egg characteristics of layers. Horned melon provides nutrients (magnesium, vitamin C, and zinc) that support bone remodelling and maintenance of bone strength (Šeregelj *et al*., 2022). The vitamin C and water in horned melon may support collagen production, wound healing, and protection from sun damage (Usman *et al*., 2015). Horned melon is a rich source of magnesium and potassium. These minerals can reduce inflammation, prevent the accumulation of arterial plaque, and help regulate blood pressure. Horned melon also offers multiple nutrients that are vital for a healthy immune system, including vitamin C, zinc, iron, and magnesium (Usman *et al*., 2015).

Conclusion

The vitamin and mineral profile of horned melon meal presented it as a promising feedstuff that can replace commercial premix in layer diet. Replacement of commercial layer premix up to 50% with horned melon meal did not adversely affect both the internal and external quality of eggs of the fed experimental birds. These proved that horned melons provided enough vitamins and minerals that support egg production and the maintenance of egg quality.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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