

Effect of mixed vegetable based diets on haematology and serum biochemistry of meat type chickens

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Target Audience: Researchers, Poultry Farmers, Physiologists

Abstract

The poultry industries main challenges in Africa include inadequate supply and high price of conventional feedstuff; this necessitated an experiment conducted to determine the effect of mixed vegetable diets (*Moringa oleifera* and *Telferia occidentalis*) as partial substitutes for soya bean meal on the haematological and serum biochemical indices of growing meat type-chickens. A total of one hundred and sixty day- old, unsexed broiler chicks (Anak Strain) were reared for 56 days. The birds were allotted to 4 dietary treatments with 40 birds per treatment, replicated 4 times. The experimental diets were formulated with *Moringa oleifera* and *Telferia occidentalis* (Fluted pumpkin) at (0%, 5%, 10% and 15%) inclusion levels in broiler ration represented as T1, T2, T3, and T4 respectively. Data obtained were subjected to ANOVA in a Completely Randomized Design (CRD) using SAS (1994); significant means were separated using Duncan's Multiple Range Test of the same statistical package. The result revealed that birds fed diets containing mixed vegetables at 0%, 5% and 10% inclusion levels had the highest ($P<0.05$) PCV, Hb, RBC. The growing birds fed 5%, 10%, 15% had the least WBC and MCH. The results of the serum biochemistry showed that birds on 0% and 5% had the highest Total protein, sodium and chloride. Other parameters measured were not significant for the treatment. The study reveals that up to 10% inclusion level of mixed vegetable diets had the highest ($p<0.05$) PCV, Hb, RBC with the least WBC resulting in optimum performance and without any adverse effect on haematology and serum biochemical parameters which is an indication that it could replace soya bean meal in a typical broiler's ration for improved performance.

Keywords: Haematological indices, Blood chemistry indices, Anak Strain chicken, *Moringa oleifera* and *Telferia occidentalis*

Description of Problem

Vegetable protein source is increasingly being used by the poultry industries around the world for many reasons which include its cheap cost, abundance and its ability to synthesize amino acids from a wide range of available primary materials such as water, carbon dioxide, and atmospheric nitrogen (31). (14) reported that the utilization of plant and leaf extract in animal production has found

scientific and commercial wide spread acceptance as a strategy to improve the health status and performance of the animals. According to (46, 4, 2, 23) the leaf is a highly valued plant with multipurpose effects which could complement the inadequacies of most feedstuffs. (40, 28) reported that leaf protein has the potential to supplying good quality food compared to what is obtainable from cereals, legumes, and oil seeds. In addition,

they are also rich in potassium, calcium, and magnesium.

Recently, there has been interest in the utilization of *Moringa oleifera* commonly called *Zogali* in the native language (Hausa), where it originated and in non-native countries (35,28), due to its nutritional, therapeutic, and prophylactic properties (10) as a protein source for livestock, (39). (40) reported that moringa foliage is a potential inexpensive protein source for livestock feeding. The advantages of using moringa leaf meal as protein source are numerous and the fact that, is a perennial plant that can be harvested several times in a growing season and has the potential to reduce feed cost. Moringa tree is considered as one of the world's most useful trees, as almost every part of the Moringa tree can be used for food, medication, and industrial purposes (19). People use its leaves, flowers, and fresh pods as vegetables while others use it as livestock feed (3). (18) found that the inclusion of *Moringa oleifera* leaf meal at amounts up to 10% did not produce significant ($P > 0.05$) effects on feed consumption, body weight, feed conversion ratio and carcass weight in broiler chickens. Many researchers have reported a major effect of the genotype on live weight (29, 34) feed conversion, carcass composition (22, 25) carcass weight (37) and abdominal fat (5, 11). *Telferia occidentalis* (Fluted pumpkin) is a leaf vegetable that is widely cultivated in the tropics and subtropics characteristically contribute dietary energy value of about 3121kcal/kg (16). According to (20) *Telferia occidentalis* leaves contain 30.5% CP, 3.0% crude lipid, 8.3% CF, and 8.47 total Ash. (20) noted that the leaves have low level of tannin ($4.75 + 2.10\text{mg}/100\text{g DM}$) and oxalate ($0.45 + 2.10\text{mg}/100\text{g DM}$) but high level of phytic acid ($20.5 + 2.10\text{mg}/100\text{g DM}$).

The examination of blood constituents help to assess the physiological, nutritional and pathological status of animals (8). Blood constituents were reported to change in

relation to the physiological conditions of the animals (42). Haematological components such as red blood cells, white blood cells or leucocytes, mean corpuscular volume, mean corpuscular haemoglobin and mean corpuscular haemoglobin concentration are valuable in monitoring feed toxicity as well as the health status of farm animals (32).

In view of the high crude protein content of these unconventional feed stuffs (*Moringa oleifera* and *Telferia occidentalis*), this research seek to evaluate the resultant effect of combining these vegetables since there is no or limited information on their combination as alternative protein supplement in poultry diets.

Materials and Methods

Experimental Site

The experiment was carried out at the Poultry Unit of the Teaching and Research Training center, Federal University, Dutse, Jigawa State.

Collection and Processing of Test Ingredients

Moringa oleifera and *Telferia occidentalis*

Fresh *Moringa oleifera* and *Telferia occidentalis* leaves were collected and air dried until they were crispy to touch while retaining their greenish coloration. The leaves were then milled to obtain a product referred to as moringa leaf meal (MLM) and *Telferia occidentalis* leaf meal (TLM) were stored in sacks until they were needed for inclusion in the formulated diets. Four (4) experimental diets were formulated using 0%, 5%, 10%, and 15%, inclusion levels of dried combination of moringa and fluted pumpkin at ratio 50: 50. The dried leaf meal moringa and fluted pumpkin were mixed with other feed ingredients at varying levels of inclusion.

Experimental Design

One hundred and sixty (160) unsexed day-old Anak broiler chicks were allotted to four

(4) dietary treatments of birds replicated 4 times with 10 birds each arranged in a completely randomized design. Four experimental diets were formulated for the starter and finisher phases of the study which consisted of four dietary supplementations of equal mixtures of moringa and pumpkin leaf meals at varying levels of 0%, 5%, 10%, and

15% and are represented as T₁, T₂, T₃, and T₄ respectively. The experimental diets for starter and finisher phases are balanced such that the crude protein and metabolizable energy contents of the feeds for each phase of the broilers are balanced within the recommended range (24).

Table 1: Composition of Experimental Diets for Starter Broiler chickens fed mixed Vegetable Based Diets.

Ingredients (%)	T ₁	T ₂	T ₃	T ₄
Maize	47.00	47.00	47.00	47.00
Wheat offal	15.00	15.00	15.00	15.00
Fish meal	3.00	3.00	3.00	3.00
Soya bean	16.00	11.00	6.00	1.00
Groundnut cake	14.60	14.60	14.60	14.60
Bone meal	1.80	1.80	1.80	1.80
Oyster shell	1.50	1.50	1.50	1.50
Premix	0.30	0.30	0.30	0.30
Salt	0.25	0.25	0.25	0.25
Lysine	0.25	0.25	0.25	0.25
Methionine	0.30	0.30	0.30	0.30
MLM + PPLM	-	5.00	10.00	15.00
Total	100	100	100	100
Determined Nutrients				
ME (MJ/kg)	11.29	11.26	11.35	11.31
Dry matter	94.45	96.12	95.64	95.71
Crude Protein	22.89	22.62	22.57	22.52
Crude Fiber	6.10	5.89	5.89	5.79
Ether extract	2.51	2.42	2.35	2.57
Ash	6.61	6.62	6.54	6.52
NFE	56.33	55.68	58.31	57.01

MLM- Moringa Leaf meal, PPLM – Pumpkin leaf meal, ME- Metabolizable energy, Vitamin premix supplied per kg diet to supply ;VitA 8,000.00iu, D3,1440iu;VitE,21.6mg;VitK3,2.7mg;VitB1,1.8mg;VitB2,3.6mg;VitB6,2.7mg;Niacin,21.6mg;VitB12,0.018mg;FolicAcid,0.54mg;Panthothenic acid,9.0mg;Biotin,0.036mg;Choline chloride,270mg;Zinc,27mg;Mn,108mg;Fe,18mg;I2,0.72mg;Se,0.072mg;Cu,1.44mg;Co,0.144mg.

Table 2: Composition of Experimental Diets for Finisher Broiler chickens fed mixed Vegetable Based Diets.

Ingredients (%)	T₁	T₂	T₃	T₄
Maize	55.00	55.00	55.00	55.00
Wheat offal	15.00	15.00	15.00	15.00
Fish meal	1.00	1.00	1.00	1.00
Soya bean	15.00	10.00	5.00	00.00
GNC	10.00	10.00	10.00	10.00
Bone meal	1.50	1.50	1.50	1.50
Oyster shell	1.50	1.50	1.50	1.50
Premix	0.30	0.30	0.30	0.30
Salt	0.25	0.25	0.25	0.25
Lysine	0.20	0.20	0.20	0.20
Methionine	0.25	0.25	0.25	0.25
MLM + PPLM	-	5.0	10.0	15.0
Total	100	100	100	100

Determined Nutrients				
ME MJ/kg	11.32	11.31	11.21	11.51
Dry Matter	94.56	96.24	96.70	96.24
Crude Protein	20.50	21.18	21.73	21.82
Crude Fiber	5.63	4.95	4.89	4.79
Ether extract	2.18	2.25	2.45	2.29
Ash	5.90	5.14	6.69	6.72
NFE	60.35	66.72	68.94	63.35

MLM- Moringa Leaf meal, PPLM – Pumpkin leaf meal, ME- Metabolizable energy, Vitamin premix supplied per kg diet to supply ;vit A 10,000.00iu, D3,1440iu;VitE,21.6mg;VitK3,2.7mgVitB1,1.8mg;VitB2,3.6mg;VitB6,2.7mg;Niacin,21.6mg;VitB12,0.018mg;FolicAcid,0.54mg;Panthothenic acid,9.0mg;Biotin,0.036mg;Choline chloride,270mg;Zinc,27mg;Mn,108mg;Fe,18mg;I2,0.72mg;Se,0.072mg;Cu,1.44mg;Co,0.144mg.

Data Collection and Analysis

Blood sample collection and analysis

At 56 day of the study, blood samples were collected from 12 randomly selected birds per treatment (4 per replicate) to determine the blood serum chemistry. Blood collection was done through brachial vein puncture (12) using needles and syringes. Each blood sample was emptied into 2 sets of well labeled sample bottles; the sample containing anti-coagulant was used for the analysis of hematological traits while the plane bottle without anti-coagulant was used to analyze the serum bio-chemical traits of the birds. The hematological traits analyzed were: Packed

Cell Volume (PCV), Haemoglobin (Hb), Red Blood Cell (RBC) and White Blood Cell (WBC) and biochemical traits: Blood glucose, the total serum protein, albumin and globulin using bromocresol green method (44). Serum creatinine (6) and serum uric acid concentration (45) was determined according to standard procedures. Serum enzymes: alanine transaminase (ALT) and aspartate serum transaminase (AST) were analysed using the commercial kits (Qualigens India. Pvt. Ltd., Catalogue number 72201-04). Mean Corpuscular Volume (MCV), Mean Corpuscular Hemoglobin (MCH) and Mean Corpuscular Hemoglobin Concentration

(MCHC) were determined using the method of (15). Erythrocyte indices which include the mean corpuscular volume (MCV) and mean corpuscular hemoglobin concentration (MCHC) were computed in accordance with the standard formulae of (41, 17)

Chemical Analysis

The proximate compositions of the diets were determined using the methods of (1) while metabolizable energy was calculated.

Statistical Analysis

All data generated were subjected to one-way Analysis of Variance (ANOVA) using (38). Significant means at 5% level of probability were separated using (9).

Results

The proximate composition of the experimental diets fed at both starter and finisher phases are shown in Tables 4. The dry matter contents of the experimental diets ranged between 96.12 for birds on 5% inclusion and 94.45 for birds 0 % (control diets) fed at the starter meanwhile 96.70 for birds on 10% inclusion to 94.24 for birds on 15% inclusion level at finisher phase. The crude protein, moisture, crude fiber, ether extract, ash, and NFE for experimental diets ranged from 25.42 to 20.52, 5.55 to 3.88, 6.10 to 5.19, 2.51 to 1.75, 6.62 to 5.40, 62.01 to 55.68, and 21.82 to 18.18, 5.76 to 3.30, 5.63 to 3.95, 2.45 to 1.59, 8.69 to 5.14, 66.72 to 53.35 at both starter and finisher phases respectively.

Table 3: Proximate composition of the experimental test ingredients

Parameters (%)	<i>Moringa oleifera</i> leaf	<i>Telferia occidentalis</i> leaf
Dry Matter	90.23	92.13
Moisture	9.77	7.87
Crude Protein	24.21	40.19
Crude Fiber	5.83	12.50
Ether Extract	1.09	1.26
Ash	11.30	9.34
Nitrogen Free Extract	57.80	28.87
Sodium (Na)	26.32	50.50
Potassium (K)	11.45	17.63
Calcium (Ca)	64.33	69.33
Magnesium (Mg)	52.99	36.18
Phosphorus (P)	55.86	38.79
Iron (Fe)	25.92	35.60
Manganese (Mn)	1.73	2.20
Zinc (Zn)	0.02	0.13
Nitrogen (N)	2.27	6.45
Oxalate	1.42	1.89
Tannine	4.28	4.55
Phytase	1.98	2.68

Table 4: Proximate Composition of the Experimental Diets for Starter and Finisher phases

Parameter	STARTER				FINISHER			
	1	2	3	4	1	2	3	4
DM (%)	94.45	96.12	95.64	95.71	94.56	96.24	96.70	96.24
Moisture (%)	5.55	3.88	5.36	4.29	5.44	3.76	3.30	5.76
CP (%)	22.89	22.62	22.57	22.52	20.50	21.18	21.73	21.82
CF (%)	6.10	5.98	6.01	5.19	5.63	3.95	4.89	4.29
EE (%)	2.51	2.42	2.35	1.75	2.18	2.25	2.45	1.59
Ash (%)	6.61	6.62	5.40	6.25	5.90	5.14	8.69	7.21
NFE (%)	56.33	55.68	58.31	62.01	60.35	66.72	58.94	53.35

DM- Dry matter, CP- crude protein, CF- crude fibre, EE- ether extract, NFE- nitrogen free extract

Table 5: Effect of Diets on Hematological Parameters of Growing Meat Type Chicken.

Parameters	Inclusion levels of mixed vegetable leaves				±SEM
	T1	T2	T3	T4	
	0%	5%	10%	15%	
PCV %	29.67 ^{ab}	30.00 ^a	28.67 ^{ab}	27.67 ^c	0.33
Hb g/L	10.0 ^a	9.67 ^{ab}	9.0 ^{bc}	8.67 ^c	0.18
WBC (X10 ⁶)/L	7.12 ^a	5.68 ^c	6.60 ^b	6.44 ^{bc}	0.17
RBC (X10 ⁹)/L	5.57 ^{ba}	5.67 ^a	5.23 ^{ab}	5.03 ^c	0.10
MCV	53.25 ^c	52.89 ^d	54.80 ^a	54.34 ^b	0.24
MCH	18.11 ^a	17.03 ^c	17.19 ^b	15.55 ^d	0.28
MCHC	34.02	25.88	31.37	31.31	1.62

^{abc}Means on the same row having different superscripts are significantly different ($P < 0.05$). SEM = standard error mean. Hb: Hemoglobin, PVC: packed Cell Volume, WBC: White Blood Cell, RBC: Red Blood Cell, MCV: Mean Corpuscular Volume, MCH: Mean Corpuscular Hemoglobin, MCHC: Mean Corpuscular Concentration.

Influence of dietary inclusions levels of mixed vegetables on the haematological parameters of meat-type chickens.

The effect of the dietary inclusions of mixed vegetables on the hematological parameters of meat-type chickens is presented in Table 5. Results revealed that birds fed with 5% inclusion of mixed vegetables recorded the highest ($p < 0.05$) PCV values. Birds fed 0% (control diet) inclusion of mixed vegetables recorded higher ($p < 0.05$) but statistically similar in value to those obtained for birds fed

(10%) inclusion while the least PCV was recorded for birds on 15% diet. Birds on control diet recorded a significantly ($p < 0.05$) higher Hb than those fed 15% diet. Although, the value was statistically similar ($p > 0.05$) to those fed 5% inclusions of mixed vegetables. The highest values of RBC was recorded for birds on control and 5% diet which were statistically similar ($p < 0.05$) to the value recorded for birds on 10% inclusion while birds on 15% diets recorded the least value for RBC. Birds fed diet containing (0%) control

diet recorded the highest ($P<0.05$) value of WBC. Meanwhile, birds on 10 and 15% recorded slightly higher ($p<0.05$) value which were statistically similar compared birds on 5% diet for WBC. Mean corpuscular haemoglobin concentration (MCHC) did not show any significant ($p<0.05$) difference for the treatment imposed on the birds across the dietary treatment. The result for mean corpuscular volume (MCV) revealed that control diet, 10% and 15% recorded significantly higher values than birds on 5% inclusion level of mixed vegetable leaves. The same trend was observed for MCH though birds fed 15% recorded the least of MCH.

Influence of dietary inclusion levels of mixed vegetable leaves on serum chemistry of growing meat-type chicken are presented in Table 6.

Values obtained for Total protein, Na and Cl ($p<0.05$) were significantly affected by the

dietary treatment. Total protein decreased with increasing inclusion levels of mixed vegetables. The birds on 0% and 5% diet recorded a similar significant ($p<0.05$) value for TP. The values obtained were however higher than birds fed 10 and 15% diets. Birds fed 10% diet recorded the least ($P < 0.05$) value for Na meanwhile higher ($P < 0.05$) values of Na enzyme were obtained for birds fed control diet, 5% and 15% inclusion levels of mixed vegetable leaves. Birds on Control diet had the highest value for Cl. Meanwhile, birds on 5% diet and 15% diet recorded slightly higher ($p<0.05$) value over bird fed 10% diet.

The albumin, globulin, ALT, ALP, serum creatinine, uric acid and serum glucose were not affected ($p>0.05$) by the inclusion of dietary treatments. However, K and CO_2 were not affected ($p>0.05$) by dietary treatments imposed on them.

Table 6: Effect of diets on serum biochemical values of growing meat type chickens
Inclusion levels of mixed vegetable leaves

Parameters	Inclusion levels of mixed vegetable leaves				±SEM
	T1 0%	T2 5%	T3 10%	T4 15%	
Total Protein g/L	36.67 ^a	35.33 ^a	27.00 ^b	27.00 ^b	1.61
Albumin g/L	15.00	13.67	14.00	13.33	0.45
Globulin g/L	21.67	21.66	13.00	13.67	1.16
Glucose mmol/L	11.37	10.77	18.60	15.17	1.50
ALP u/L	688.7	652.7	599.7	467.3	44.23
ALT u/L	12.67	16.67	15.67	18.33	1.04
AST u/L	145.00	145.33	143.33	149.33	1.98
Sodiummmol/l	149.33 ^a	140.67 ^{ab}	137.00 ^b	142.00 ^a	0.96
Potassium mg/dl	5.31	4.23	4.66	4.43	0.29
Carbon dioxide	20.33	20.33	22.33	23.00	0.62
Chloride	114.00 ^a	109.00 ^b	104.00 ^c	109.67 ^b	1.19
Uric acid mm	0.883	0.900	0.700	0.900	0.09
Creatininemmol/l	26.33	21.33	29.33	24.33	1.97

^{abc} Means on the same row having different superscripts are significantly different ($P<0.05$) SEM=standard error mean., ALP = alkaline phosphate, ALT=Alanine aminotransferase, AST= Aspartate aminotransferase, CRT: Creatinine.

Discussion

The proximate composition of pumpkin leaf showed that the test ingredient was a rich source of crude protein. The crude protein value recorded here was at variance with the report of (21, 20). These researchers reported a crude protein of 21.31% and 30.5% respectively. The crude fiber reported here agreed with the report of (30). However, ether extract of 1.26% reported here was lower compared to values reported by (21, 30, 20).

The nitrogen free extract (NFE) content was normal for a carbohydrate rich feed ingredient. This agreed with report of (21). The Iron content value of 35.60% reported here was higher than values reported by (21, 30). The Oxalate and Tannine reported here agreed with the earlier work of (21, 20). The phytase content recorded here was at variance with the report of (21) (51.50%), (20) (20.5%).

The proximate composition of Moringa leaf showed that the test ingredient was rich source of crude protein. The crude protein value recorded here was at variance with the report of both (23, 27) and (26) respectively. This variation could be attributed to the planting location and stage of harvesting of the Moringa leaves. The crude fiber content and ether extract of the forage in this study were lower than 9.75% and 8.02% reported by (27).

The high PCV, Hb, RBC obtained for birds fed 5% and 10% inclusion which was statistically similar with values obtained for birds fed control diet implied that birds on mixed vegetable supplements had high oxygen carrying capacity (7). It is an indication that the nutritional profile of the diet was more enriched when supplemented with combined inclusion of moringa and pumpkin. Nutrition was reported to influence the haemoglobin level of the blood (43). Pellet and Young (33) confirmed that haemoglobin levels are positively correlated with protein quality and level in the diets. This work agreed with the

independent report of (36) on moringa and (21) on pumpkin who reported high RBC, Hb, when fed solely graded levels of these dietary vegetables.

Increasing levels of mixed vegetables in diets decreased WBC and MCH. The reduced value of WBC parameter implies the absence of foreign bodies which could trigger or raise the leucocyte count.

The indices of total protein measured decreased with increased level of mixed vegetables. This affirmed the report of (36, 10) the researchers reported decreased TP as the *Moringa oleifera* increases across the dietary treatments. Serum biochemical analysis is used to determine the level of heart attack, liver damage and to evaluate protein quality and amino acid requirements in animals as reported by (13).

Biochemical indices of the meat type chicken in the present study revealed that at 15% inclusion the sodium (Na) level obtained was significantly affected by the dietary treatments. This implies that combinations of mixed vegetables at 15% inclusion level could maintain the concentration of this mineral without adverse effect. This study revealed that most of the serum biochemical components of the meat type chickens were not significant ($p < 0.05$) and falls within normal range for poultry. Moreover, the result revealed a strong positive influence on haematological parameters with no adverse effect on the meat type chickens.

Conclusion and Application

1. It can be concluded that supplementation of mixed vegetables of high protein content such as *Moringa oleifera* and *Telferia occidentalis* could be used as partial substitute for soya bean meal up to 10% inclusion level and enhance broiler production.
2. The study also revealed that inclusion of mixed vegetables up to 10% inclusion

level improved PCV, Hb, RBC and total protein with decrease in WBC, Sodium, Chloride and MCH.

3. It was also shown that at these same levels, mixed vegetables have potentials to replace soya bean meal in a typical broiler's ration for optimum performance and without any adverse effect on haematology and serum biochemistry parameters measured in this research.
4. It is therefore recommended that mixed vegetables such as *Moringa oleifera* and *Telferia occidentalis* leaves could be used as partial replacement for soya bean meal in a typical broiler ration formulation at 10% inclusion to reduce cost of feeding and competition for the limited resources.

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