Nigerian J. Anim. Sci. 2017 (1):144 - 156

Effect of dietary supplementation of guinea hen weed (*Petiveria alliacea*) leaf and root meals on nutrient utilization and intestinal morphology of finishing broiler chicken.

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Target audience: Feedmillers, livestock farmers, extension agents, animal nutritionists

Abstract

A 56- day feeding trial was carried out to evaluate the effect of feeding guinea hen weed leaf and root meals as phytobiotics on nutrient digestibility and intestinal morphology of finishing broiler chickens using 192 day old chicks. Eight treatment groups were arranged in a 2 × 4 factorial arrangements of 2 plant parts; Petiveria leaf meal (PLM) and Petiveria root meal (PRM) at 4 levels (0mg/kg, 500mg/kg, 1000mg/kg and 1500mg/kg). Each group was replicated three times with 8 birds per replicate. Digestibility parameters were influenced ($p<0.05$) by petiveria plant parts. Crude protein, ash and NFE digestibility of birds fed diet containing PRM were higher compared to birds fed PLM. Birds fed 1500mg/kg had the highest value of crude protein digestibility compared to other dietary treatments. The interaction of plant parts and inclusion levels on the nutrient utilization showed that highest crude protein, ether extract, crude fibre and NFE values were observed in birds on 1500mg/kg PLM compared to other dietary treatments. Intestinal morphology of finishing broiler chickens revealed that duodenal apical width, basal width and Jejunal villi height values were higher ($p<0.05$) in birds fed diet containing PLM. It was observed that supplementation of finishing broiler diets elicited improved nutrient digestibility and intestinal morphology.

Key Words: Guinea hen weed, broiler, digestibility, gut morphology, root meal, leaf meal

Description of the problem

Broiler birds face several challenges which disturb the normal functioning organisms in the gut, resulting in impaired absorption of nutrients, reduced performance and increased mortality. Antibiotics are commonly used to supplement diets to make birds

perform better under harsh conditions during rearing. The use of antibiotic-based growth promoters is presently facing serious criticism and has raised global concern as some reports revealed side effects such as the development of microbial resistance by the birds and their potentially harmful effects on human health (1). The use of plant and plant bioactive compounds dates back thousands of years to the ancient era (2,3). There has been an increasing interest in the utilization of growth promoters from natural origin (4,5). Medicinal plants and herbs are one of the natural feed additives currently used in poultry diets to enhance the performance and immune response of birds (1). Herbs and plant extracts are potential substitutes currently used as antibiotic growth promoting compounds (6, 7). Phytobiotics are plant–derived compounds and natural bioactive compounds that can be incorporated into diets in order to enhance the performance and well–being of animals. Their effects have been proven to improve feed palatability and quality, growth, gut function and nutrient digestibility, gut micro flora (8). The beneficial multifunction aspect of most phytobiotics is derived from their specific bio-active components. Experiments have been conducted showing the implication of phytobiotics. It has reported that herbs, spices and their extracts can stimulate appetite and endogenous secretions of digestive enzymes (9). Supplementation of 200 ppm essential oil extract from oregano with combination of cinnamon and pepper improved the apparent faecal digestibility of dry matter and crude protein in broiler finisher diet (10). They also concluded that 5000 ppm Labiate extract from sage, thyme and rosemary gave same results in broiler finisher diet. In addition, (11) found that feeding cinnamon to broilers significantly reduced the concentration of pathogenic microorganisms in the ileum, caecum and colon, which was accompanied by an increased weight gain of birds. However, (12) did not observe any positive effects on growth performance or macronutrient digestibility in female broiler chickens when diets were supplemented with thymol or cinnamon. Guinea hen weed (*Petiveria alliacea)*, a perennial shrub, is used as herbs in the treatments of ailments such as pile, worm, cancer. The influence of guinea hen weed, however, as a phytobiotic or growth promoter in poultry species has not been fully explored. Therefore, the present study was designed to evaluate the effects of guinea hen weed **(***Petiveria alliacea*) leaf and root meals on nutrient digestibility and intestinal morphology of finishing broiler chicken.

Materials and Methods

Experimental site and test ingredients

This experiment was carried out at the Teaching and Research Farm, Federal University of Agriculture, Abeokuta, Ogun State. The area lies on latitude 7^{0}10'N and longitude 3^{0} 2'E. It is 76m above sea level. The climate is tropical humid with a mean annual rainfall of 1037mm,34.7^{o}C temperature and relative humidity of 82%. (13). The plant

Petiveria alliacea was obtained within the premise of Federal University of Agriculture, Abeokuta (FUNAAB). It was uprooted completely; the leaves and the roots were cut off from the stalk separately, chopped into bits, washed to remove debris. The leaves and roots were spread separately on polythene bags, the leaves were air dried under a shade (29±2° C) and the roots were sundried (≤ 90% DM) this was done for 14 days until they become crispy and easy to break and without altering the greenish colour of the leaf. They were milled (1mm sieve) into powdered form using a laboratory mill and stored separately in air tight containers at room temperature till it is ready for use: as *Petiveria* leaf meal (PLM) and *Petiveria* root meal (PRM). Proximate (14) and phytochemical (15) analysis of PLM and PRM were carried out according to standard procedures (Table 1).

Table 1. Proximate composition and phytochemical screening of PLM and PRM

Measurements	PLM	PRM
Proximate composition (%)		
Dry matter	90.40	90.30
Crude protein	19.82	8.03
Crude fibre	11.56	23.62
Ether extract	4.03	0.76
Ash %	15.57	6.69
NFE %	39.45	51.30
Phytochemical screening (%)		
Terpenoids	24.50	14.50
Flavonoids	7.77	2.97
Tannin	3.75	1.50
Alkaloids	9.90	10.00
Phytate	4.50	6.20
Phenol	4.65	2.50
Antioxidant	31.60	23.60
Oxalate	0.33	0.13
Carotenoid	63.50	13.50
Trypsin inhibitor	0.04	0.02

Experimental birds management and dietary treatments

A total of 192 day-old broiler chicks of commercial strain (ANAK 2000) were purchased from a reputable commercial hatchery in Abeokuta. The poultry house and equipments were thoroughly washed and disinfected before the arrival of chicks. The birds were raised on deep litter system for 56-days and reared in two phases; starter and finisher phases. The birds were alloted to eight treatment groups of 24 birds each. A basal diet which met the necessary nutrient requirement of broilers were formulated for starter and finisher phases (Table 2) of the birds. The experiment was laid out in a 2 × 4 factorial

arrangement of diets. Eight dietary treatments were formulated such that each of the *Petiveria* meal obtained from either leaf or root was supplemented at day old in the basal diet at 0, 500, 1000 and 1500mg/kg, respectivelys. Each treatment containing 24 birds were replicated thrice with 8 birds per replicate. Proximate analysis of experimental diets was carried out according to the methods of (14).

Table 2: Percentage (%) Composition of basal diet for broiler starter (0-4 weeks) and finisher (5-8 weeks)

Ingredients	(0-4 weeks)	(5-8 weeks)
Maize	52.00	56.00
Wheat Offal	6.30	8.30
SBM	22.00	16.00
PKC	1.50	2.50
GNC	10.00	9.00
FM (72%)	3.00	3.00
Bone meal	2.50	2.50
Oyster shell	2.00	2.00
Lysine	0.10	0.10
Methionine	0.10	0.10
Salt	0.25	0.25
*Premix	0.25	0.25
Total	100	100
Composition		
ME (Kcal/Kg)	2920.30	2933.20
Crude Protein (%)	22.90	20.74
Crude Fibre (%)	3.39	3.41
Fat (%)	3.95	4.00
Calcium (%)	1.53	1.52
Phosphorus (%)	0.51	0.50

*For starter diets, Vitamin Mineral premix provided (per kg of diet): Vit A 11500IU, Vit D3 1600IU, Riboflavin 9.9mg, Biotin 0.25mg, Pantothenic acid 11.0mg, Vitamin K 3.0mg, Vit B2 2.5mg, Vit B6 0.3mg, VitB12 8.0mg, Nicotininc acid 8.0mg, Iron 5.0mg, Manganase 10.mg, Zinc 4.5mg, Cobalt 0.02mg, Selenium 0.01

*For finisher diets, Vitamin Mineral premix provided (per kg of diet): Vit A 11500IU, Vit D3 1600IU, Riboflavin 9.9mg, Biotin 0.25mg, Pantothe nic acid 11.0mg, Vitamin K 3.0mg, Vit B2 2.5mg, Vit B6 0.3mg, VitB12 8.0mg, Nicotininc acid 8.0mg, Iron 5.0mg, Manganase 10.mg, Zinc 4.5mg, Cobalt 0.02mg, Selenium 0.01m.

Data Collection

Apparent nutrient digestibility

At the expiration of 56 days, two broilers from each pen (making a total of 10 birds per treatment) were selected and arranged in clean, separate and disinfected metabolic cages. Three days of acclimatization were allowed before the commencement of the digestibility study. A known weight of feed, which matched their previous daily feed intake, was fed during the metabolic trial. Excreta collection was done daily for a period of four days. The daily excreta

voided for each bird was dried overnight (at 55 ^{o} C) while total collections per bird were pooled at the expiration of 4 days metabolic trial. Proximate compositions of dried faecal samples were determined according to (14).

Gut morphometry

About 0.5 cm portion taken at the medium part of each of the three intestinal segments (duodenum, jejunum and ileum) was used for histological measurements. The samples were opened longitudinally, rinsed with cold saline and fixed in a buffered formalin solution for about 4 h. Histo-morphological analysis was done according to the procedures of (16). The preparations were mounted between slide and strip. Intestinal villi with their crypts were, individually, separated under a dissecting microscope while the length and width of the villi were measured according to the procedures described by (17).

Statistical Analysis

Data obtained in this experiment were laid out in a 2 × 4 factorial arrangement and analysed using (18) to determine main and interaction effects. Level of probability was expressed at 5% and significant means seperated using Duncan Multiple Range Test (19). The statistical model is as described below:

$$Y_{ijk} = \mu + D_i + E_j + (DE)_{ij} + \Sigma_{ijk}$$

Where:

Y_{ijk}=Observed value of dependent variable

μ=Population mean

D_i=Main effect *Petiveria* plant parts (i =Leaf, Root)

E_j=Main effect of inclusion level in diet (j =1, 2,3,4)

$(DE)_{ij}$ = Interaction effect of *Petiveria* plant parts and inclusion level

Σ_{ijk} =Random residual error

Results

Main effect of Petiveria plant parts and level of inclusion on the nutrient utilization

Table 3 shows the main effect of *Petiveria* plant parts and level of inclusion on the nutrient utilization of finishing broiler chicken. All parameters measured with the exception of dry matter digestibility were influenced ($p<0.05$) by the main effect of *petiveria* plant parts. Crude protein, ash and NFE digestibility of birds fed diet containing PRM were higher compared to birds fed PLM. Meanwhile, birds fed diet containing PLM showed improved ($p<0.05$) ether extract and crude fibre digestibility. Main effect of graded levels of inclusion of *Petiveria* meal in the diet were significant ($p<0.05$) for crude protein and ether extract digestibility. Crude protein digestibility of birds fed diet containing 500 mg/kg had the least ($p<0.05$) value compared to other dietary treatments. All birds fed diet supplemented with *Petiveria* meal had higher ($p<0.05$) ether extract digestibility when compared with the control. Increased ($p<0.05$) ash and NFE digestibility were obtained with birds fed diet supplemented with 500 and 1500 mg/kg when compared to those fed other dietary treatments.

Interaction effects of Petiveria plant parts and varying inclusion levels of Petiveria meal on the nutrient digestibility

The interaction effects of *Petiveria* plant

parts and varying inclusion levels of *Petiveria* meal on the nutrient utilization of finishing broiler chickens are shown in Table 4. Birds fed diet supplemented with 500, 1000 mg/kg PRM and 1500 mg/kg PLM showed higher ($p<0.005$) crude protein and ether extract digestibility than other treatments. Meanwhile, birds fed diet supplemented with 1000 mg/kg PLM and1000 mg/kg PLM showed the least ($p<0.05$) ash digestibility. Birds fed diet supplemented with 1500 mg/kg PRM and 500 mg/kg PLM recorded the highest ($P<0.005$) NFE digestibility.

Table 3: Main effect of *Petiveria* plant parts and levels of inclusion on the nutrient digestibility of finishing broiler chickens

	Plant parts			Levels of inclusion (mg/kg)				
Measurement	**PLM**	**PRM**	**SEM**	**0**	**500**	**1000**	**1500**	**SEM**
Dry matter	85.23	73.59	3.427	79.58	75.65	81.95	80.50	4.847
Crude protein	67.13^{b}	74.95^{a}	1.723	74.88^{a}	59.84^{b}	71.40^{a}	77.87^{a}	2.437
Ether extract	56.70^{a}	56.24^{b}	3.463	37.40^{b}	69.61^{a}	57.40^{a}	61.51^{a}	4.898
Crude fibre	47.10^{a}	46.10^{b}	3.419	31.40^{b}	63.50^{a}	36.24^{b}	55.23^{a}	4.835
Ash	40.88^{b}	57.50^{a}	3.946	32.40^{b}	67.02^{a}	34.83^{b}	62.41^{a}	5.580
NFE	60.55^{b}	67.82^{a}	2.754	57.30^{b}	69.39^{a}	53.98^{b}	76.24^{a}	3.894

$^{a-b}$ Means on the same row with different superscripts are significantly different ($p<0.05$)
SEM- Standard Error of Mean ; PLM; *Petiveria* leaf meal; PRM; *Petiveria* root meal; NFE; Nitrogen free extract;

Table 4: Interaction of *Petiveria* plant parts and level of inclusion (ppm) on the nutrient digestibility of finishing broiler chickens

	PLM				PRM				
Measurements	**0**	**500**	**1000**	**1500**	**0**	**500**	**1000**	**1500**	**SEM**
Dry matter	84.05	82.72	90.86	83.30	75.11	68.60	73.04	77.64	2.455
Crude protein	75.50^{b}	40.76^{c}	65.11^{bc}	86.90^{a}	74.30^{b}	78.90^{ab}	77.64^{ab}	68.95^{bc}	2.883
Ether extract	40.71^{c}	57.94^{b}	51.94^{bc}	83.21^{a}	40.99^{c}	81.30^{a}	62.86^{ab}	39.81^{d}	4.168
Crude fibre	27.05^{c}	53.54^{ab}	35.22^{c}	72.50^{a}	27.00^{c}	73.44^{a}	37.25^{b}	37.97^{b}	4.023
Ash	56.10^{ab}	55.86^{ab}	23.50^{c}	75.50^{a}	56.15^{ab}	78.20^{a}	46.20^{bc}	49.34^{b}	5.178
NFE	60.60^{b}	56.66^{bc}	46.59^{c}	85.40^{a}	60.89^{b}	81.87^{a}	61.40^{b}	67.13^{ab}	3.085

$^{a-d}$ Means on the same row with different superscripts are significantly different ($p<0.05$)
SEM- Standard Error of Mean ; PLM; *Petiveria* leaf meal; PRM; *Petiveria* root meal; NFE;
Nitrogen free extract

Main effect of Petiveria plant parts and level of inclusion on intestinal morphology

Table 5 shows the main effect of *Petiveria* plant parts and level of inclusion on the intestinal morphology of finishing broiler chickens. Results revealed that duodenal villi height, laminal proria depth, jejunal apical width and ileum basal width were not affected ($p>0.05$) by the main effect of *Petiveria* plant parts. However, duodenum apical width, basal width and Jejunum villi height values were ($p<0.05$) higher in birds fed diet containing PLM compared those on PRM. Main effect of graded levels of inclusion of *Petiveria* meal in diets of broiler chickens influenced ($p<0.05$) the duodenal villi height, apical width, jejunal villi height, basal width, laminal propria depth and ileum villi height and laminal propria depth. Birds fed diet supplemented with 1500mg/kg *Petiveria* meal had improved duodenal apical value than other dietary treatments. Birds on control diet had improved duodenal villi height than

Table 5: Ma in effect of *Petiveria* plant parts and level of inclusion on the intestinal morphology of finishing broiler chickens

	Plant parts			Levels of inclusion (ppm)				
Measurements	**PLM**	**PRM**	**SEM**	**0**	**500**	**1000**	**1500**	**SEM**
Duodenum (µm)								
Villi height	547.50	706.30	63.899	777.50^{a}	542.50^{b}	477.50^{c}	710.00^{ab}	90.367
Apical width	48.75^{a}	45.00^{b}	0.884	45.00^{b}	45.00^{b}	47.50^{ab}	50.00^{a}	1.250
Basal width	126.30^{a}	100.00^{b}	3.920	105.00	122.50	107.50	117.50	5.543
Laminal propria depth	386.30	277.50	37.274	252.50	362.50	405.00	307.50	52.713
Jejunum (µm)								
Villi height	900.00^{a}	815.00^{b}	36.472	692.50^{b}	730.00^{b}	765.00^{b}	1242.00^{a}	51.579
Apical width	56.30	41.30	2.976	50.00	47.50	50.00	47.50	4.208
Basal width	97.50^{b}	111.30^{a}	4.050	105.00^{ab}	87.50^{b}	90.00^{b}	135.00^{a}	5.728
Laminal propria depth	195.00^{b}	217.50^{a}	8.447	175.00^{b}	202.50^{ab}	185.00^{b}	262.50^{a}	11.946
Ileum (µm)								
Villi height	906.30^{b}	1229.00^{a}	85.620	1288.00^{a}	1080.00^{ab}	1072.50^{ab}	1170.00^{a}	121.085
Apical width	45.00^{b}	52.00^{a}	2.282	57.50	42.50	50.00	45.00	3.227
Basal width	96.30	122.50	7.790	115.00	97.50	107.50	117.50	11.016
Laminal propria depth	192.50^{b}	252.50^{a}	10.508	255.00^{a}	187.50^{b}	217.50^{ab}	230.00^{a}	14.860

$^{a-b}$ Means on the same row with different superscripts are significantly different ($p<0.05$)
SEM- Standard Error of Mean ; PLM; *Petivera* leaf meal; PRM; *Petiveria* root meal;

Table 6: Interaction effects of *Petiveria* plant parts and level of inclusion (ppm) on the intestinal mo rphology of finishing broiler chickens

Measurements	PLM				PRM				SEM
	0	500	1000	1500	0	500	1000	1500	
Duodenum (µm)									
Villi height	550.00	510.00	560.00	570.00	550.00	575.00	595.00	850.00	54.064
Apical width	45.00^{ab}	50.00^{a}	50.00^{a}	50.00a	45.00^{ab}	40.00^{b}	45.00^{ab}	50.00^{a}	0.893
Basal width	120.00^{b}	130.00^{ab}	135.00^{ab}	150.00^{a}	120.00^{b}	115.00^{b}	80.00^{c}	85.00^{c}	5.507
Laminal propria depth	230.00	350.00	490.00	475.00	275.00	375.00	320.00	140.00	31.893
Jejunum (µm)									
Villi height	710.00^{c}	685.00^{c}	830.00^{c}	1375.00^{a}	675.00^{c}	775.00^{c}	700.00^{c}	1110.00^{b}	53.866
Apical width	60.00	55.00	60.00	50.00	40.00	40.00	40.00	45.00	2.450
Basal width	100.00^{b}	75.00^{c}	115.00^{ab}	100.00^{b}	100.00^{b}	100.00^{b}	65.00^{c}	170.00^{a}	6.582
Laminal propria depth	165.00^{c}	195.00^{b}	175.00^{c}	225.00^{ab}	165.00^{c}	210.00^{ab}	195.00^{b}	300.00^{a}	9.641
Ileum (µm)									
Villi height	1435.00^{ab}	850.00^{bc}	600.00^{c}	740.00^{c}	1400.00^{ab}	1310.00^{b}	865.00^{bc}	1600.00^{a}	86.161
Apical width	65.00^{a}	35.00^{c}	50.00^{ab}	45.00^{b}	65.00^{a}	50.00^{ab}	50.00^{ab}	45.00^{b}	2.113
Basal width	105.00	100.00	85.00	95.00	125.00	95.00	130.00	140.00	5.991
Laminal propria depth	300.00^{a}	155.00^{c}	210.00^{bc}	195.00^{bc}	300.00^{a}	220.00^{b}	225.00^{b}	265.00^{ab}	5.731

$^{a-c}$ Means on the same row with different superscripts are significantly different ($p<0.05$)

SEM- Standard mean of error; PLM; *Petiveria* leaf meal; PRM; *Petiveria* root meal

others. Jejunal villi height, basal width and laminal propria depth for birds fed diet supplemented with 1500 mg/kg *Petiveria* meal recorded the highest values compared to other levels of inclusion.

Interaction effects of Petiveria parts and levels of inclusion on the gut morphology of finishing broilers

The interaction effects of *Petiveria* plant parts and levels of inclusion on the gut morphology of finishing broiler chickens are presented on Table 6. Duodenal apical width numerical values observed did not follow a definite pattern nevertheless birds fed diet containing 1500mg/kg PLM and PRM respectively had the highest value. In the duodenum, as the levels of PLM inclusion increased, duodenal basal width increased. Conversely, duodenal basal width decreased with increasing PRM. 1500mg/kg PLM supplementation recorded highest value of Jejunal villi height while, 1500mg/kg PRM revealed highest jejunal basal width and lamina propria depth with other values following no particular trend.

Discussion

The highest crude protein and ether extract digestibility recorded with birds fed diet supplemented with 500, 1000 mg/kg PRM and 1500 mg/kg PLM indicated improved nutrient utilisation. Herbs, spices and their extracts have been reported to stimulate appetite and endogenous secretions such as enzymes (9). Birds fed diet supplemented with 1500 mg/kg PRM and 500 mg/kg PLM also showed the highest NFE digestibility. (20) and (21) reported that phytogenics have stimulating effects on the output of digestive enzymes from the pancreas, gut mucosa, and increased bile flow and such effects might lead to improved nutrient utilization in the gut. The numerically higher nutrient digestibility reported in petriveria root meal (PRM) compared to petriveria leaf meal (PLM) might be due to the fact that the root meal contains more of the enzyme stimulating substances than the leaf meal. The observed improved crude protein and ether extract digestibility observed in birds fed graded levels of supplementation plant parts could also be linked to a synergetic effect of increasing levels of test ingredient resulting in enhanced digestion of the nutrients. (22) reported that phytogenic additives sped up digestion and enhanced the secretion of protein digesting enzyme. Also, (10) observed that supplementation of 200mg/kg extracts of some phytobiotics improved digestibility of dry matter and crude protein in broiler finisher diets. The result was also in *consonance with the findings of* (23) who reported that probiotics improved digestion, absorption and availability of nutrients with a positive effect on intestinal activity and increasing digestive enzymes. Additionally, (24) opined that there was evidence to suggest that herbs, spices, and various plant extracts have appetite and digestion-stimulating properties and antimicrobial effects . However, (12) did not observe any positive effects on macronutrient digestibility in female broiler chicken when diets were supplemented with

phytobiotics. Furthermore, the result did not agree with findings of (25) who reported that phytogenic feed additives did not alter the apparent ileal digestibility of nutrients in broiler chickens. Nevertheless, (20) reported that phytogenic plant substances improved gut function. The increase in duodenal apical width, basal width; jejunal villi height, basal width, laminal propria depth; ileum illi height, apical width and laminal propria depth observed in birds fed graded levels of PLM and PRM could be due to greater efficiency in the utilization of feed resulting in enhanced development of intestinal morphology in poultry (26). The result corroborates the work of (27) who showed that dietary supplementation of 0.2 g/kg pure curcumin derived from turmeric in a corn-soybean based diet increased the villi height and width of duodenum and jejunum. Meanwhile, feeding broiler chickens with phytogenic products has been shown to cause increase or decrease intestinal segments (28*, 29, 30 and 31*). In addition, (32) indicated that the villi height and crypt depth in all segments of the small intestine were significantly increased in diets containing a probiotic. The improved intestinal morphology observed in finishing broilers fed 1500mg/kg of PLM and PRM is in harmony with the report of (33) that showed that a probiotic treatment significantly increased the villi height in the jejunum and ileum at 21 or 42 d compared with a non-supplemented basal diet.

Conclusion

The findings from the study indicated that supplementation of finishing broiler diets with petiveria leaf and root meals respectively elicited improved nutrient digestibility and intestinal morphology with highest improvement in 1500mg/kg supplementation.

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