

Evaluation of Synbiotic Effect of Curry Leaf (*Ocimum Canum*) with Probiotic (*Lactobacillus subtilis*) on the Performance and Gut Morphology of Broiler Chickens

*Salihu, E.A.¹, Bawa, G.S.¹, Omage, J.J.¹, Buba, W.², Tanko, S.Y.^{1,3}, Yunana, Y.L.⁴ and Akanbi, O.M.¹

¹Department of Animal Science, Ahmadu Bello University, Zaria

²National Agricultural Extension and Research Liaison Services, Ahmadu Bello University, Zaria

³Nuhu Bamalli Polytechnic, Zaria

⁴Department of Animal Science, Bayero University, Kano

*Corresponding Author: salihuezekiel@gmail.com, +2347060638200

Target Audience: Animal Nutritionist, Feed Manufacturers, Policy Makers, Poultry Farmer, Researchers

Abstract

*The withdrawal of antibiotic growth promoters (AGPs) in poultry production due to antimicrobial resistance concerns has necessitated sustainable alternatives. This study evaluated the synbiotic effects of curry leaf (*Ocimum canum*) combined with probiotic (*Lactobacillus subtilis*) on the growth performance and gut morphology of broiler chickens. A total of 360 day-old chicks were randomly assigned to six dietary treatments in a completely randomised design with three replicates of 20 birds each. Treatment diets comprised: a control (no additives), *Ocimum canum* at 800 g/100 kg or 1200 g/100 kg, synbiotics (*Ocimum canum* at 800 g/100 kg or 1200 g/100 kg + *Lactobacillus subtilis*), and oxytetracycline (positive control). Data were collected on growth performance and villi morphometric traits. Results showed that the highest final body weight (953.42 g/bird), daily weight gain (32.69 g/bird/day), and best FCR (1.72) were recorded in birds fed diet containing oxytetracycline, and those fed diet containing 800 and 1200 g/100kg diet of *Ocimum canum* followed by group fed the synbiotic diet with 800 and 1200 g/100 kg *O. canum* plus *L. subtilis*. Feed cost per kilogram gain was lowest in the same group with similar trend. At the finisher phase, the highest final body weight (3221.57 g/bird), daily weight gain (110.32 g/bird/day), and best FCR (1.72) were recorded in birds fed the diet containing oxytetracycline, followed closely by those fed the synbiotic diet with 800 g/100 kg *Ocimum canum* plus *L. subtilis* (final body weight 3212.75 g/bird, daily weight gain 109.94 g/bird/day, FCR 1.74), and then the *O. canum*-only groups at 800 g/100 kg and 1200 g/100 kg. Feed cost per kilogram gain was lowest in the oxytetracycline and synbiotic (800 g/100 kg) groups. Hence, poultry farmers should incorporate *Ocimum canum* at 800 g/100 kg diet combined with *Lactobacillus subtilis* as a synbiotic feed additive to replace antibiotic growth promoters.*

Keywords: Antibiotic Growth Promoters, *Lactobacillus subtilis*, Microbiota, *Ocimum canum*, Prebiotics, Probiotics, Synbiotic

Description of Problem

The intensive broiler poultry industry has historically depended on antibiotic

growth promoters (AGPs) to optimize feed efficiency, accelerate growth rates, and mitigate subclinical enteric challenges, thereby supporting high

productivity under commercial conditions [1,2,3]. However, the extensive use of AGPs has been increasingly linked to the global rise in antimicrobial resistance (AMR), including the dissemination of multidrug-resistant pathogens, potential transfer of resistance determinants to human microbiota, and detectable antibiotic residues in poultry meat and eggs, posing significant risks to public health and food safety [4,5]. In response, regulatory bodies worldwide have progressively restricted or banned AGPs in animal feed; the European Union implemented a full ban in 2006, followed by similar measures in numerous countries, with ongoing global efforts (including in Asia, Africa, and Latin America) to phase out non-therapeutic antibiotic use amid calls from organizations such as the FAO and WHO to curb AMR through a One Health approach [3,4,5]. The withdrawal of AGPs has often resulted in compromised broiler performance, including reduced body weight gain, elevated feed conversion ratios (FCR), increased incidence of enteric disorders and higher overall production costs [6,7], emphasizing the critical need for efficacious, safe, and sustainable alternatives that preserve gut integrity, nutrient utilization, and immune competence throughout the production cycle [6, 8,9,10].

In this context, synbiotics, a synergistic combinations of probiotics (live beneficial microorganisms) and prebiotics (selectively fermentable substrates) have emerged as one of the most promising non-antibiotic strategies

for broiler nutrition [11]. Synbiotics promote a balanced gut microbiota by enhancing the survival and colonization of beneficial bacteria, increasing short-chain fatty acid production, strengthening the intestinal barrier, improving nutrient digestibility and absorption, modulating immune responses, and suppressing pathogenic overgrowth, frequently yielding performance benefits comparable to or exceeding those of AGPs in experimental settings [12,13]. These effects are particularly valuable across both the starter phase (when chicks establish gut microbiota and immunity) and the finisher phase (when rapid growth and feed efficiency are maximized), and can contribute to sustained improvements in live weight, FCR, carcass quality, and resilience to environmental stressors.

Medicinal plants rich in bioactive secondary metabolites provide accessible, cost-effective, and locally sourced prebiotic- or phytobiotic-like options, especially in tropical regions like sub-Saharan Africa where such herbs are abundant and traditionally utilized [6]. *Ocimum canum* Sims (commonly referred to as curry leaf, African sweet basil, or wild basil) is a perennial herb of the Lamiaceae family, widely distributed in Nigeria and used in traditional medicine and cuisine for its potent antioxidant, antimicrobial, anti-inflammatory, and digestive-enhancing properties, primarily attributed to high levels of phenolic compounds, flavonoids, and essential oils [6,14]. Related *Ocimum* species (e.g., *O. gratissimum*, *O. basilicum*) have demonstrated positive effects in poultry,

including improved growth performance, enhanced immune function, reduced oxidative stress, and modulation of gut microbiota under heat stress or pathogenic challenge, positioning *O. canum* as a viable candidate for supporting intestinal health in broiler chickens [6,11,15].

Complementing this, *Lactobacillus subtilis* (often aligned with *Bacillus subtilis* strains in modern probiotic applications due to spore-forming robustness and stability in feed) is a well-established probiotic in poultry nutrition. It colonizes the gastrointestinal tract, produces antimicrobial peptides (e.g., bacteriocins), competitively excludes pathogens, enhances endogenous enzyme activity, improves villus architecture and crypt depth for better nutrient uptake, and bolsters systemic and mucosal immunity, leading to superior body weight gain, feed efficiency, and reduced inflammation across production phases [16,17,18].

The strategic combination of *O. canum* (as a natural prebiotic-like modulator) with *Lactobacillus subtilis* (as a resilient probiotic) in a synbiotic formulation could exert amplified, complementary benefits: phytochemicals from the plant may selectively nourish or protect the probiotic, exert direct antimicrobial effects against pathogens, and synergistically optimize gut microflora balance, villus morphology, nutrient absorption, and overall performance [19]. Although individual applications of *Ocimum* species and *Bacillus/Lactobacillus*-based probiotics have shown promise in broilers, research

on the specific synbiotic potential of *Ocimum canum* combined with *Lactobacillus subtilis* remains scarce, particularly regarding comprehensive impacts on growth performance metrics and gut morphology. Hence, this study evaluates synbiotic effect of *Ocimum canum* with *Lactobacillus subtilis* on the growth performance and gut morphology of broiler chickens.

Materials and Methods

Experimental site

The experiment was conducted at the Poultry Unit, Department of Animal Science Teaching and Research Farm, Ahmadu Bello University, Zaria, Kaduna State. Zaria is in the Northern Guinea Savannah zone, located on longitude 11° 09' 01.78" N and 7° 39' 14.79" E 671m above sea level. The climate is characterized by well-defined dry and wet seasons. The area has three distinct seasons; hot dry season from March to May, the warm rainy season from June to September, and a cool dry season from November to February. The climate is relatively dry with a mean annual rainfall of 700-1400 mm [20]. The area has an average relative humidity of 36.00 % during the dry season and 78.50 % for the wet season and an average minimum and maximum temperatures of 13.30 and 38.50 °C respectively [21,22].

Ethical approval

Ethical approval was obtained from Ahmadu Bello University Committee on Animal Use and Care (ABUCAUC) before the commencement of the

experimental trials (ABUCAUC/2024/Animal Science/073).

Source and processing of test materials

Ocimum canum leaves were purchased from Sabo market in Sabon-Gari Local Government Area of Kaduna State. The leave samples were removed from the stem and dried under shade for five (5) days, crushed by grinding using hammer mill and included as part of the experimental diet. *Lactobacillus spp* was procured from Animal Care, Kaduna.

Source of experimental birds

The experimental birds (Ross 308 day-old broiler chicks) were purchased from Agrited Hatchery, Ibadan, Nigeria for the study.

Experimental diets

Six treatment diets were formulated as shown in Tables 1 and 2 for the starter and finisher phases, respectively, as follows:

Table 1: Composition of broiler starter diets containing *Ocimum canum* with *Lactobacillus subtilis*

Ingredients (%)	0	<i>Ocimum canum</i> (g/100kg Diet)		<i>Ocimum canum</i> (g/100kg Diet) + LS		Oxytet
		800	1200	800+LS	1200+LS	
Maize	56.80	56.00	55.60	56.00	55.60	56.80
Soyabean cake	29.00	29.00	29.00	29.00	29.00	29.00
Groundnut cake	10.00	10.00	10.00	10.00	10.00	10.00
Curry leaf	0.00	0.80	1.20	0.80	0.80	0.00
Bone Meal	3.00	3.00	3.00	3.00	3.00	3.00
Limestone	0.40	0.40	0.40	0.40	0.40	0.40
Common Salt	0.25	0.25	0.25	0.25	0.25	0.25
Methionine	0.20	0.20	0.20	0.20	0.20	0.20
Lysine	0.10	0.10	0.10	0.10	0.10	0.10
Vit-min Premix ^A	0.25	0.25	0.25	0.25	0.25	0.25
Total	100.00	100.00	100.00	100.00	100.00	100.00
Calculated Analysis						
ME (Kcals/Kg)	2,956	2,939	2,930	2,939	2,930	2,956
Crude Protein (%)	23.02	23.01	23.00	23.01	23.00	23.02
Crude Fibre (%)	3.50	3.54	3.56	3.54	3.56	3.50
Ether Extract (%)	3.22	3.22	3.22	3.22	3.22	3.22
Calcium (%)	1.31	1.31	1.31	1.31	1.31	1.31
Phosphorus (%)	0.87	0.86	0.86	0.86	0.86	0.87
Methionine (%)	0.55	0.54	0.54	0.54	0.54	0.55
Lysine (%)	1.26	1.26	1.26	1.26	1.26	1.26
Feed Cost (₦/kg)	370.98	384.58	390.83	390.58	396.83	387.48

^A Vitamin- mineral premix provide per kg of diet: Vit. A, 10,000,000 IU; Vit. D₃, 2, 000,000 IU; Vit. E, 40,000mg; Vit. K₃, 2000mg; Vit. B₁, 1,500mg; Vit. B₂, 5,000mg, Vit. B₆, 4,000mg; Vit. B₁₂, 20mg; Niacin, 40,000mg; Calpan, 10,000mg; Folic acid, 1,000mg; Biotin, 100mg; Choline Chloride, 30,000mg; Manganese, 80,000mg; Iron, 40,000mg; Zinc, 60,000mg; Copper, 80,000mg; Iodine, 800mg; Cobalt, 300mg; Selenium, 200mg. Oxytet = Oxytetracycline, LS: *Lactobacillus subtilis*.

Table 2: Composition of broiler finisher diets containing *O. canum* with *Lactobacillus subtilis*

Ingredients (%)	<i>O. canum</i> (g/100kg diet)			<i>O. canum</i> (g/100kg diet) +LS		Oxytet
	0	800	1200	800+LS	1200+LS	
Maize	58.00	58.00	58.00	58.00	58.00	58.00
Maize offal	7.00	6.20	5.80	6.20	5.80	7.00
Soyabean cake	21.00	21.00	21.00	21.00	21.00	21.00
Groundnut cake	9.80	9.80	9.80	9.80	9.80	9.80
Curry leaf	0.00	0.80	0.80	0.80	0.80	0.00
Bone Meal	3.00	3.00	3.00	3.00	3.00	3.00
Limestone	0.40	0.40	0.40	0.40	0.40	0.40
Common Salt	0.25	0.25	0.25	0.25	0.25	0.25
Methionine	0.20	0.20	0.20	0.20	0.20	0.20
Lysine	0.10	0.10	0.10	0.10	0.10	0.10
Vit-min Premix ^A	0.25	0.25	0.25	0.25	0.25	0.25
Total	100.00	100.00	100.00	100.00	100.00	100.00
Calculated Analysis						
ME (Kcals/Kg)	3,012	3,004	3,001	3,004	3,001	3,012
Crude Protein (%)	20.04	20.02	20.01	20.02	20.01	20.04
Crude Fibre (%)	3.91	3.91	3.92	3.91	3.92	3.91
Ether Extract (%)	3.53	3.53	3.53	3.53	3.53	3.53
Calcium (%)	1.29	1.29	1.29	1.29	1.29	1.29
Phosphorus (%)	0.85	0.85	0.85	0.85	0.85	0.85
Methionine (%)	0.50	0.50	0.50	0.50	0.50	0.50
Lysine (%)	1.02	1.02	1.02	1.02	1.02	1.02
Feed Cost (₦/kg)	359.74	373.44	379.69	379.44	385.69	376.09

^A Vitamin- mineral premix provide per kg of diet: Vit. A, 10,000,000 IU; Vit. D₃, 2, 000,000 IU; Vit. E, 40,000mg; Vit. K₃, 2000mg; Vit. B₁, 1,500mg; Vit. B₂, 5,000mg; Vit. B₆, 4,000mg; Vit. B₁₂, 20mg; Niacin, 40,000mg; Calpan, 10,000mg; Folic acid, 1,000mg; Biotin, 100mg; Choline Chloride, 30,000mg; Manganese, 80,000mg; Iron, 40,000mg; Zinc, 60,000mg; Copper, 80,000mg; Iodine, 800mg; Cobalt, 300mg; Selenium, 200mg. Oxytet = Oxytetracycline, LS: *Lactobacillus subtilis*.

Diet 1: Standard diet without experimental materials (Control)

Diet 2: Standard diet with curry leaf feed additive 800g/100kg

Diet 3: Standard diet with curry leaf feed additive 1200 g/100kg

Diet 4: Standard diet with curry leaf feed additive 800g/100kg and *Lactobacillus* spp

Diet 5: Standard diet with curry leaf feed additive 1200g/100kg and *Lactobacillus* spp

Diet 6: Standard diet with Oxytetracycline as feed additive (Tetracin at 111 g/100kg diet).

Management of experimental birds and data collection

Three hundred and sixty (360) day-old Ross 308 broiler chicks were allotted to six dietary treatments with 3 replicates of 20 birds each in a completely randomized design (CRD). The birds were housed in deep litter pens and managed with all necessary routine management practices. Feed and water were supplied *ad libitum* throughout the experimental period.

Growth study

Initial and final weights of birds were taken at the beginning and at the end of both starter and finisher phases. Weight

gain and feed intake were measured weekly while feed conversion ratio and cost per Kg gain were computed for both phases. Mortality was recorded as they occur.

Villi morphometric study

Intestinal segment samples (approximately 2 cm in length) were taken from the birds used for carcass evaluation to ascertain villi morphometric traits. Intestinal tissues were harvested and fixed in 10 % formolsaline. They were histologically processed according to the method of Bancroft and Stevens [23]. They were dehydrated through ascending grades of alcohol (70 %, 90 % and 100 %) for 2 hours each. The tissues were cleared in xylene for 2 hours then were impregnated and embedded in paraffin wax. They were sectioned at 5 micron thickness using Rotary microtome machine (Leica RT 25 made in England). Sectioned tissues were mounted on slides, dried and stained using Hematoxylin and Eosin (H and E) stain. Stained slides of the tissues were photomicrographed using Amscope Digital Camera for microscope version 2.0, made in Japan. Histomorphometric analysis for the villus was carried out using Digimizer image analysis software version 4.5 made in USA [23]. The morphometric indices that were evaluated include villi height, villi width, crypt depth, surface area, perimeter and the ratio of villi height to villi depth at the Histology Laboratory, Department of Human Anatomy, Ahmadu Bello University, Zaria.

Data analysis

All data obtained from the study were statistically analysed using the General Linear Model Procedure of Statistical Analysis Systems software package while significant difference between treatment means were separated using Tukey Procedure [24].

Experimental model

$$Y_{ij} = \mu + T_i + e_{ij}$$

Y_{ij} = Performance of the chickens (j^{th} observation),

μ = Population mean,

T_i = Effect of treatment (i^{th} treatment),

e_{ij} = Error term.

Results and Discussion

Growth performance of broiler chicks fed diets containing *Ocimum canum* with *Lactobacillus subtilis*

Table 3 shows the growth performance of broiler chicks fed diets containing *Ocimum canum* with *Lactobacillus subtilis* from 0–4 weeks. The results showed that there significant ($P < 0.05$) differences in final weight, weight gain, feed conversion ratio and feed cost per kg weight gain. Feed intake, average daily feed intake, and mortality showed no significant ($P > 0.05$) differences across treatments. Broiler chickens on diets supplemented with *Ocimum canum* at 800 g/100 kg and 1200 g/100 kg had the highest final body weights of 949.17 g/b and 931.67 g/b, respectively, along with the greatest weight gains of 911.08 g/b and 893.57 g/b, and average daily weight

gains of 32.54 g/b/d and 31.91 g/b/d; these values were statistically similar ($P > 0.05$) to each other and to the oxytetracycline-supplemented group (final body weight 953.42 g/b, weight gain 915.34 g/b, average daily weight gain 32.69 g/b/d). The synbiotic combinations at 800 g/100 kg and 1200 g/100 kg *Ocimum canum* plus *Lactobacillus subtilis* produced intermediate values (final body weights 876.67 g/b and 865.00 g/b, weight gains 838.58 g/b and 826.93 g/b, average daily weight gains 29.95 g/b/d and 29.53 g/b/d), which remained significantly ($P < 0.05$) higher than the control group (final body weight 841.54 g/b, weight gain 803.44 g/b, average daily weight gain 28.69 g/b/d).

Feed conversion ratio showed a significant ($P < 0.05$) improvement with supplementation; the lowest value of 1.72 was obtained in birds fed diets containing oxytetracycline, while values of 1.78 (800g of *O. canum*), 1.76 (1200g of *O. canum*), 1.81 (800g of *O. canum* + LS), and 1.83 (800g of *O. canum* + LS) in the supplemented groups were better than the value (1.93) obtained for birds in the control group. Feed cost per kg gain was significantly ($P < 0.05$) reduced in most supplemented treatments, with the lowest value of ₦1153.05/kg in the oxytetracycline group, followed by ₦1179.46/kg and ₦1179.56/kg in the *Ocimum canum*-only groups at 800g and 1200g, respectively, and intermediate reductions in the synbiotic groups with values of ₦1206.30/kg and ₦1233.80/kg,

for 800g+LS and 1200g+LS groups, respectively, compared with ₦1257.35/kg in the control.

These results indicate that *Ocimum canum* supplementation, particularly at 800 g/100 kg and 1200 g/100 kg, markedly enhanced growth performance and feed efficiency in broiler chickens during the starter phase, with effects comparable to oxytetracycline. The synbiotic inclusion of *Lactobacillus subtilis* provided additional benefits over the unsupplemented control, though the phytogetic-only treatments performed numerically better. Such improvements in final body weight, weight gain, average daily weight gain, and feed conversion ratio implies better nutrient utilisation and intestinal function, while the reductions in feed cost per kg gain highlight economic advantages from supplementation. The findings align with evidence that *Ocimum canum* and related *Ocimum* species improve growth performance, weight gain, and feed efficiency in broiler chickens [6]. *Bacillus subtilis* supplementation similarly enhances body weight gain, feed conversion ratio, and overall performance through gut modulation [9, 12, 13, 14]. Synbiotic combinations further support these outcomes by promoting superior growth and efficiency comparable to antibiotic growth promoters [10, 19]. The absence of differences in feed intake suggests that performance gains arose primarily from enhanced nutrient utilization and metabolic efficiency rather than increased consumption [1, 3].

Table 3: Growth performance of broiler chicks fed diets containing *Ocimum canum* with *Lactobacillus subtilis* (0-4 weeks)

Parameters	Control	<i>Ocimum canum</i>		<i>Ocimum canum</i>		Oxytet	SEM
		(g/100kg diet)		(g/100kg diet) + LS			
		800	1200	800	1200		
Initial weight (g/b)	38.10	38.08	38.10	38.08	38.07	38.08	0.09
Final weight (g/b)	841.54 ^c	949.17 ^a	931.67 ^a	876.67 ^b	865.00 ^{bc}	953.42 ^a	15.56
Weight gain (g/b)	803.44 ^c	911.08 ^a	893.57 ^a	838.58 ^b	826.93 ^{bc}	915.34 ^a	15.57
ADWG (g/b/d)	28.69 ^c	32.54 ^a	31.91 ^a	29.95 ^b	29.53 ^{bc}	32.69 ^a	0.55
Feed intake (g/b)	1548.34	1617.75	1574.00	1514.92	1515.42	1570.81	60.35
ADFI (g/b/d)	55.30	57.78	56.21	54.10	54.12	56.10	2.09
FCR	1.93 ^c	1.78 ^{ab}	1.76 ^{ab}	1.81 ^b	1.83 ^b	1.72 ^a	0.04
Feed cost (₦/kg)	652.62	663.98	669.66	667.18	672.86	671.98	NA
FC/kg gain (₦/kg)	1257.35 ^d	1179.46 ^{ab}	1179.56 ^{ab}	1206.30 ^{bc}	1233.80 ^{cd}	1153.05 ^a	20.46
Mortality (%)	3.33	0.00	0.00	0.00	0.00	1.67	1.79

^{abc}Means with different superscripts along the same row are significantly ($P \leq 0.05$) different, Oxytet: oxytetracycline, SEM: standard error of means, NA: Not analyzed, , LS: *Lactobacillus subtilis*, FCR: Feed Conversion Ratio

Growth performance of broiler chickens fed diets containing *Ocimum canum* with *Lactobacillus subtilis*

Table 4 shows the growth performance of broiler chickens fed diets containing *Ocimum canum* with *Lactobacillus subtilis* during the finisher phase (5–7 weeks). Final body weight, weight gain, average daily weight gain, feed intake, average daily feed intake, feed conversion ratio, and feed cost per kg gain were significantly ($P < 0.05$) affected by dietary treatments. Broiler chickens fed diet containing synbiotic combinations of *Ocimum canum* at 800 g/100 kg with *Lactobacillus subtilis* had the highest final body weight of 3212.75 g/b, weight gain of 2308.83 g/b, average daily weight gain of 109.94 g/b/d, and the lowest feed conversion ratio of 1.74 along with the lowest feed cost per kg gain of ₦1087.24/kg; these values were statistically similar ($P > 0.05$) to the oxytetracycline-supplemented group (final body weight 3221.57 g/b, weight gain 2316.67 g/b, average daily weight gain 110.32 g/b/d, feed conversion ratio

1.72, feed cost per kg gain ₦1082.64/kg); and groups fed diets containing *Ocimum canum* alone at 800 g/100 kg and 1200 g/100 kg had intermediate values (final body weights 3125.49 g/b and 3096.08 g/b, weight gains 2221.57 g/b and 2194.12 g/b, average daily weight gains 105.79 g/b/d and 104.48 g/b/d, feed conversion ratios 1.81 and 1.88, feed cost per kg gain ₦1124.43/kg and ₦1182.07/kg). The synbiotic at 1200 g/100 kg *Ocimum canum* with *Lactobacillus subtilis* had intermediate growth performance traits (final body weight 2978.43 g/b, weight gain 2076.47 g/b, average daily weight gain 98.88 g/b/d, feed conversion ratio 1.96, feed cost per kg gain ₦1238.42/kg), which were better than the control group (final body weight 2668.63 g/b, weight gain 1767.65 g/b, average daily weight gain 84.17 g/b/d, feed conversion ratio 2.11, feed cost per kg gain ₦1284.77/kg).

Feed intake and average daily feed intake were significantly ($P < 0.05$) higher in all supplemented groups (ranging from

3978.43–4131.18 g/b and 189.45–196.72 g/b/d) compared with the control (3736.27 g/b and 177.92 g/b/d), and these values were statistically similar ($P > 0.05$) among the supplemented treatments. Mortality showed no significant ($P > 0.05$) differences across treatments. These results reveal that the synbiotic combination, especially *Ocimum canum* at 800 g/100 kg with *Lactobacillus subtilis*, resulted in the strongest enhancements in final body weight, weight gain, average daily weight gain,

feed efficiency, and economic performance during the finisher phase, and were similar to the oxytetracycline group while markedly outperforming the unsupplemented control. The increased feed intake in supplemented groups, coupled with superior feed conversion ratio, indicates improved nutrient utilisation and digestive efficiency [11]. Higher intake may be attributed to the aromatic nature of the essential oils in *Ocimum* spp that has been reported to have appetite stimulating effect [6,11].

Table 4: Growth performance of broiler chickens fed diets containing *Ocimum canum* with *Lactobacillus subtilis* (5-7 weeks)

Parameters	Control	<i>Ocimum canum</i> (g/100kg diet)		<i>Ocimum canum</i> (g/100kg diet) + LS		Oxytet	SEM
		800	1200	800	1200		
Initial weight (g/b)	900.98	903.92	901.96	903.92	901.96	904.90	5.42
Final weight (g/b)	2668.63 ^c	3125.49 ^{ab}	3096.08 ^{ab}	3212.75 ^a	2978.43 ^b	3221.57 ^a	106.15
Weight gain (g/b)	1767.65 ^d	2221.57 ^{ab}	2194.12 ^b	2308.83 ^{ab}	2076.47 ^c	2316.67 ^a	55.52
ADWG (g/b/d)	84.17 ^d	105.79 ^{ab}	104.48 ^b	109.94 ^{ab}	98.88 ^c	110.32 ^a	2.70
Feed intake (g/b)	3736.27 ^b	4021.57 ^a	4131.18 ^a	4020.59 ^a	4075.29 ^a	3978.43 ^a	102.05
ADFI (g/b/d)	177.92 ^b	191.50 ^a	196.72 ^a	191.46 ^a	194.06 ^a	189.45 ^a	5.13
FCR	2.11 ^d	1.81 ^{ab}	1.88 ^{bc}	1.74 ^a	1.96 ^c	1.72 ^a	0.07
Feed cost (₦/kg)	607.83	621.15	627.81	624.35	631.01	630.43	NA
FC/kg gain (₦/kg)	1284.77 ^d	1124.43 ^{ab}	1182.07 ^{bc}	1087.24 ^a	1238.42 ^{cd}	1082.64 ^a	30.35
Mortality (%)	0.00	0.00	0.00	0.00	0.00	0.00	0.00

^{abcd}Means with different superscripts along the same row are significantly ($P \leq 0.05$) different, Oxytet: oxytetracycline, SEM: standard error of means, NA: Not analyzed, , LS: *Lactobacillus subtilis*, FCR: Feed Conversion Ratio.

More so, *Ocimum canum* supplementation alone resulted into better growth performance and feeding efficiency than the control, though the addition of *Lactobacillus subtilis* amplified these benefits in the synbiotic formulation at 800g+LS. This indicates the complementary actions of the plant's bioactive compounds that support microbial balance and the probiotic's role in enhancing gut function and performance. The findings align with evidence that *Ocimum gratissimum* and

related species improve weight gain, feed conversion ratio, and overall performance in broiler chickens [6]. In a related study, *Bacillus subtilis* supplementation was reported to enhance body weight gain, feed conversion ratio, and growth performance through intestinal modulation and nutrient assimilation [9, 12, 13, 14]. Synbiotic approaches have similarly been reported to yield superior final body weight, weight gain, feed efficiency, and reduced costs compared with controls or antibiotics alone [10, 19].

The elevated feed intake without compromising efficiency suggests optimised digestion and energy utilisation in supplemented broiler chickens [1, 3].

Villi morphometric traits of broiler chickens fed diets containing *Ocimum canum* with *Lactobacillus subtilis*

The villi morphometric traits of broiler chickens fed diets containing *Ocimum canum* with *Lactobacillus subtilis* is shown in Table 5. The results showed that villi area, perimeter, height, width and crypt depth were significantly ($P < 0.05$) affected by dietary treatments. Broiler chickens on the synbiotic diets of *Ocimum canum* at 800 g/100 kg with *Lactobacillus subtilis* and at 1200 g/100 kg with *Lactobacillus subtilis* had the highest villi areas of 23,895.55 μm^2 and 22,764.19 μm^2 , respectively, along with the greatest perimeters of 909.13 μm and 879.77 μm , villi heights of 413.50 μm and 394.69 μm , villi widths of 119.18 μm and 110.53 μm , and crypt depths of 127.07 μm and 130.69 μm ; these values were statistically similar ($P > 0.05$) to each other and to the oxytetracycline-supplemented group (villi area 21,639.72 μm^2 , perimeter 887.24 μm , height 398.48 μm , width 115.66 μm , crypt depth 131.39 μm).

Ocimum canum supplementation alone at 800 g/100 kg and 1200 g/100 kg had higher values that were statistically similar ($P > 0.05$) to those of the symbiotic groups and the oxytetracycline-supplemented group (areas 21,738.46 μm^2 and 22,011.03 μm^2 , perimeters 857.21 μm and 863.08 μm , heights 403.62 μm and 396.17 μm , widths 113.29 μm and 118.64 μm , crypt depths 119.95 μm and 127.88 μm , respectively) which were significantly ($P < 0.05$) higher than the control group (area 15,623.57 μm^2 ,

perimeter 738.66 μm , height 326.45 μm , width 86.41 μm , crypt depth 102.33 μm) for all these parameters except crypt depth in the 800 g/100 kg *Ocimum canum* group. The villus height to crypt depth ratio showed no significant ($P > 0.05$) differences across treatments.

These results revealed that supplementation with *Ocimum canum*, particularly when combined with *Lactobacillus subtilis* in synbiotic form, substantially improved intestinal villi architecture in broiler chickens. The larger villi dimensions and deeper crypts in supplemented treatments indicate an expanded absorptive surface area and greater proliferative activity in the intestinal mucosa, which likely contributed to the superior nutrient absorption and growth performance observed in this study. The synbiotic at 800 g/100 kg *Ocimum canum* plus *Lactobacillus subtilis* consistently ranked among the top performers, suggesting synergistic benefits from the plant's bioactive compounds and the probiotic's gut-modulating effects. *Ocimum canum* alone also markedly improved these morphometric traits over the control, confirming its role in supporting intestinal development. The findings align with evidence that *Ocimum canum* and related *Ocimum* species enhance villi morphometric traits resulting in improved intestinal microbiota in broiler chickens [6]. *Bacillus subtilis* supplementation similarly promotes villus height, crypt depth, and overall intestinal morphology, leading to better nutrient uptake [9, 12, 13, 14]. Superior villi morphometric traits reported in synbiotic interventions further implies structural improvements, and may have contributed to enhanced gut health and performance comparable to the control and antibiotic-supplemented diets [10, 19]. The lack of differences in villus

height to crypt depth ratio suggests that supplementation primarily increased absolute dimensions without

disproportionately altering the balance between absorptive and proliferative zones [1, 3].

Table 5: Villi morphometric traits of broiler chickens fed diets containing *Ocimum canum* with *Lactobacillus subtilis*

Parameters	Control	<i>Ocimum canum</i> (g/100kg diet)		<i>Ocimum canum</i> (g/100kg diet) + LS		Oxytet	SEM
		800	1200	800	1200		
Area (μm^2)	15,623.57 ^c	21,738.46 ^b	22,011.03 ^a	23,895.55 ^a	22,764.19 ^a	21,639.72 ^b	962.00
Perimeter (μm)	738.66 ^b	857.21 ^a	863.08 ^a	909.13 ^a	879.77 ^a	887.24 ^a	50.35
Villi height (μm)	326.45 ^b	403.62 ^a	396.17 ^a	413.50 ^a	394.69 ^a	398.48 ^a	26.93
Villi Width (μm)	86.41 ^b	113.29 ^a	118.64 ^a	119.18 ^a	110.53 ^a	115.66 ^a	12.45
Crypt depth (μm)	102.33 ^c	119.95 ^b	127.88 ^{ab}	127.07 ^{ab}	130.69 ^a	131.39 ^a	5.42
VH:CD	3.19	3.36	3.10	3.25	3.02	3.03	0.22

^{abc}Means with the same superscripts along the rows are not significantly different ($p>0.05$), SEM: standard error of means, Oxytet-Oxytetracycline, VH:CD: Villi Height/Crypt Depth.

Conclusion

This study demonstrates that the synbiotic combination of *Ocimum canum* (curry leaf) at 800 g/100 kg diet with *Lactobacillus subtilis* significantly enhances growth performance parameters and intestinal villi morphometric traits in broiler chickens across starter and finisher phases, comparable to oxytetracycline while outperforming the unsupplemented control.

Application

Poultry farmers in sub-Saharan Africa should incorporate *Ocimum canum* at 800 g/100 kg diet combined with *Lactobacillus subtilis* as a synbiotic feed additive to replace antibiotic growth promoters, thereby reducing antimicrobial resistance risks and production costs while maintaining optimal broiler performance.

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