Performance, Gut Microbial Status and Meat Quality of Weaned Pigs Fed Diets Supplemented with Fresh Waterleaf

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Target Audience: Animal nutritionists, livestock researchers, pig farmers, student in animal science, veterinary professionals

ABSTRACT

The demand for meat has increased due to the growing human population, thus putting intense production pressure on the meat industry. Thus, this study was conducted to evaluate the growth performance, gut microbial status, and meat quality of pigs fed diets supplemented with varying levels of waterleaf (Talinum triangulare). Fresh waterleaf was sourced and air dried under room temperature to wither before incorporation into the pigs diets at 0, 5 and 10 g/kg, designated as Diets 1, 2, and 3 respectively. Eighteen (18) weaned, (8weeks old) unsexed pigs were randomly assigned to the three dietary treatments following a completely randomized design. The proximate composition of the diets were determined while data on growth performance were collected, and after eight weeks, nine pigs were randomly selected and slaughtered for gut microbial and meat quality assessment. All data were subjected to appropriate statistical analyses. Results showed that Diet 2 (5 g/kg waterleaf) had the highest crude protein (19.17%), crude fibre (4.26%). Pigs fed Diet 2 recorded the highest weight gain (14.31 kg), daily feed intake (723.07 g), and best feed conversion ratio (2.82). Total bacterial counts in the ileum and fungal counts in the duodenum significantly decreased ($P < 0.05$) from 8.50×10^5 CFU/ml (Diet 1) to 2.50×10^5 CFU/ml (Diet 3) and 3.50×10^5 CFU/ml (Diet 1) to 0.00×10^5 CFU/ml, respectively. Identified gut microbes included Lactobacillus spp, Faecalis, Aspergillus niger, and Rhizopus stolonifer. Lipid oxidation of meat differed significantly ($P < 0.05$) at day 0, increasing from 0.30 mg MDA/kg (from pigs fed Diet 1) to 1.16 mg MDA/kg (pigs fed Diet 3), while values at day 8 were not significantly different ($P > 0.05$). Thawing and cooking losses significantly decreased ($P < 0.05$) from pigs fed Diet 1 (1.36%, 40.40%) to Diet 3 (0.78%, 33.55%), and fat content showed a non-significant decrease ($P > 0.05$) across diets. Overall, feeding waterleaf did not compromise pig performance, gut microbial balance, or meat quality. Supplementation at 5 g/kg (Diet 2) was most effective, suggesting that waterleaf can serve as a viable alternative feed supplement at this level. Its adoption by pig farmers could support sustainable and efficient pig production.

Keywords: *Weaned pig; Talinum triangulare; Ileal bacteria count; moisture retention*

DESCRIPTION OF THE PROBLEM

Pig (*Sus scrofa*), is one of the most important sources of animal protein in globally, with over 140% increase in productions since the 1960s (1). The pig industry in Nigeria is an important arm of the livestock industry providing food security for human sustenance, poverty eradication, draught power, income savings, and inorganic manure for crop farming for both urban and rural economy (2, 3). Over the years, pig production in Nigeria has increased 4 folds with an

estimated annual production of approximately 3.1% according to (4).

Pork accounts for more than one-quarter of total protein consumed worldwide and for ~35% of all meat productions (5, 6). The demand for meat and animal protein will likely further increase, firstly because the world population is expected to grow further and secondly because, in low- and middle-income countries, more people are expected to have higher incomes that enable increased meat consumption (7). The utilization of feed supplements is on the rise in the swine industry recently to support the most effective production. Feed supplements are known to have the ability to enhance growth performance, nutrient digestibility, utilization and gut health of livestock species, (8). They contain compound that serves as flavouring, antibacterial, antifungal, antiviral, hepatoprotective, immunological modulating and physiological effects, all of which are critical to their ability to improve performance in animals (9).

Waterleaf production is economically attractive due to low cost for seeds, pesticides, and herbicides for producing the crop. It is a good source of calcium, magnesium, potassium, vitamins, crude protein, α-tocopherols, β-tocopherols, total lipids, essential oils and ascorbic acid, which helps to prevent scurvy. Waterleaf is rich in indigestible fibre, which aids in digestion (10). The objective of this study is thus to evaluate the impact of varying levels of fresh waterleaf supplementation in the diets of weaned pigs on their growth performance, gut microbial profile, and meat quality characteristics.

MATERIALS AND METHOD

Eighteen (18) weaned pigs from the piggery unit of the Teaching and Research Farm of the Institution were used for this study. The pigs were randomly allotted to the dietary treatments following a Completely Randomized Design (CRD). They were assigned to three dietary treatments (Diets 1, 2, and 3), with six replicates per treatment. The composition of basal diet were as follows: Diet 1: Basal diet without waterleaf (control), Diet 2: Basal diet + 5 g/kg fresh waterleaf, Diet 3: Basal diet + 10 g/kg fresh waterleaf. The basal diets were offered to the pigs at 7% of their body weight.

Data on growth performance were collected weekly. Parameters measured included body weight, weight gain, fee intake, and feed conversion ratio. At the end of the feeding trial nine pigs – three per treatments- were randomly selected and slaughtered for gut microbial assessment and meat quality evaluation. The pigs was dissected, and digesta samples were obtained from the duodenum and ileum. Samples were transported immediately to the microbiology laboratory for microbial enumeration and identification. With the samples from the duodenum and ileum, subsequent serial dilutions up to 10^{-5} were made. The total viable counts of the samples were obtained using nutrient agar. The enumeration of the bacteria was carried out by using pour plate technique. The inoculated plates on nutrient agar were incubated at 37ºC for 24 hours after which the resultant microbial colonies were counted using Gaellenkamp colony counter.

Meat samples were collected from the longissimus dorsi muscle for meat quality assessment and stored in the freezer for 2 weeks before carrying out the thaw loss. Proximate composition of the diets, including crude protein, crude fibre, ash, moisture and ether extract, was determined using standard AOAC

methods. Mineral analysis for calcium, potassium, magnesium, iron, zinc and phosphorus was also conducted. Meat quality parameters assessed included lipid oxidation (measured as malondialdehyde content), thawing losses, cooking loss and fat content and all laboratory analysis were carried out using established procedures as described in AOAC (11), and other relevant standard method. Data collected were subjected to one - way analysis of variance (ANOVA) using appropriate SPSS. Treatment means were separated using Duncan's Multiple Range Test, and significant differences were declared at ($P<0.05$).

RESULTS

Proximate Composition of Diets Containing Varied Supplementation Levels of Waterleaf

The proximate composition of diets containing varied levels of fresh waterleaf is presented in Table 1. All parameters evaluated were significantly ($P<0.05$) different. The highest dry matter (87.59%) and metabolizable energy (3144.05MJ/kg) were obtained in Diet 1 while the least (86.81% and 3106.61MJ/kg) were from Diet 2. The crude protein values obtained were from 18.53% to 19.17%, with the highest value in Diet 2 and the least in Diet 1. The ash content in the diets ranged from 3.76% (Diet 2) to 4.63% (Diet 1). The crude fat content decrease with the supplementation of waterleaf with values from 6.06% to 5.93%, from Diet 3 to Diet 1 Crude fibre ranged from 3.20% to 4.26%, the highest value observed in Diet 2 while the least value in diet 3. The highest nitrogen free extract content was obtained in Diet 3 (56.35%) while the least value (53.57%) was observed in Diet 2.

Table 1: Proximate composition of diets containing varied supplementation levels of waterleaf

Parameters	Diet 1	Diet 2	Diet 3	SEM	P-Value
Dry matter (%)	87.59^{a}	86.81^{c}	87.06^{b}	0.11	0.01
Crude protein (%)	18.53^{b}	19.17^{a}	16.95^{c}	0.33	0.01
Crude fat (%)	6.27^{a}	6.06^{b}	5.93^{c}	0.05	0.02
Crude fibre (%)	3.46^{b}	4.26^{a}	3.20^{c}	0.16	0.01
Ash (%)	4.53^{b}	3.76^{c}	4.63^{a}	0.14	<0.01
Nitrogen free extract (%)	54.80^{b}	53.57^{c}	56.35^{a}	0.40	<0.01
Metabolizable energy (Kcal/Kg)	3144.05^{a}	3106.61^{c}	3112.65^{b}	5.81	0.01

Means with different superscripts along the same row are significantly ($P<0.05$) different
SEM: Standard error of mean
Diet 1: Basal diet without waterleaf; Diet 2: Basal diet + 5 g/kg waterleaf; Diet 3: Basal diet + 10 g/kg waterleaf

Growth Performance Characteristics of Pigs Fed Diets Containing Various Supplementation Levels of Waterleaf

Table 2 shows the growth performance characteristics of pigs fed diets containing varied supplementation levels of waterleaf. It was observed that all parameters evaluated were not significantly ($P>0.05$) different among treatment groups. Final weight (18.82 kg) and weight gain (14.31 kg) were numerically highest in pigs fed Diet 2 while those on Diet 3 had the least final weight (15.79 kg) and weight gain (11.29 kg) respectively. Pigs fed Diet 2 had the highest daily weight gain 255.48g

compared to other experimental pigs. The total feed intake (40.49 kg) and daily feed intake (723.07g) were higher in pigs fed Diet 2 while those on Diet 3 had the least total feed intake (34.18 kg) and daily feed intake (610.39g) respectively. Pigs fed Diet 2 had the least feed conversion ratio value 2.82 compared to other experimental groups.

Table 2 Growth performance characteristics of pigs fed diets containing various inclusion levels of waterleaf

Parameters	Diet 1	Diet 2	Diet 3	SEM	P-Value
Initial weight (kg)	4.45	4.51	4.50	0.13	0.98
Final weight (kg)	17.68	18.82	15.79	0.73	0.25
Weight gain (kg)	13.23	14.31	11.29	0.67	0.18
Daily weight gain (g/day)	236.31	255.48	201.67	11.90	0.18
Total feed intake (kg)	38.86	40.49	34.18	17.07	0.33
Daily feed intake (g/day)	693.90	723.07	610.39	30.49	0.33
Feed conversion ratio	2.95	2.82	3.04	0.05	0.24

Means with different superscripts along the same row are significantly (P<0.05) different.
SEM: Standard error of mean
Diet 1: Basal diet without waterleaf; Diet 2: Basal diet + 5 g/kg waterleaf; Diet 3: Basal diet + 10 g/kg waterleaf

Bacteria Counts in the Duodenum and Ileum of Pigs Fed Diets Containing Various Supplementation Levels of Waterleaf

The bacteria count in the duodenum and ileum of pigs fed diets containing various levels of waterleaf is presented in Table 3. It was observed that bacteria count in duodenum was not significantly different (P>0.05) while in the ileum it was significantly (P<0.05). Bacteria count in the ileum was significant (P<0.05) among the varying dietary treatments. It showed that pigs fed Diet 3 had the least bacteria count (2.50×10^{-5} CFU/ml) been significantly lower than the pigs fed Diet 1 (8.50×10^{-5} CFU/ml) in the duodenum of experimental pigs, bacteria ranged from (1.50×10^{-5} CFU/ml) to (2.00×10^{-5} CFU/ml).

It was observed that fungi count in

Table 3: Bacteria counts ($\times 10^{-5}$ CFU/ml) in the duodenum and ileum of pigs fed diets containing varied supplementation levels of waterleaf

Parameters	Diet 1	Diet 2	Diet 3	SEM	P-Value
Duodenum	2.00	1.50	1.50	0.33	0.85
Ileum	8.50^{c}	5.50^{b}	2.50^{a}	2.77	0.04

Means with different superscripts along the same row are significantly (P<0.05) different.
SEM: Standard error of mean
Diet 1: Basal diet without waterleaf; Diet 2: Basal diet + 5 g/kg waterleaf; Diet 3: Basal diet + 10 g/kg waterleaf.

Fungi Counts in the Duodenum and Ileum of Pigs Fed Diets Containing Various Supplementation Levels of Waterleaf

Microbial count in the duodenum and ileum of pigs fed diets containing various levels of waterleaf is presented in Table 4. duodenum was significantly differently (P<0.05) while in the ileum was not significantly different (P>0.05). The fungi count in the duodenum of experimental pigs were significantly different (P< 0.05) among dietary treatment. In the ileum fungi count was

highest in pigs fed Diet 1 (3.5 x 10^{-5} CFU/ml) and lowest in pigs fed Diet 3 (0.00 × 10^{-5} CFU/ml).

Table 4: Fungi counts (× 10^{-5} CFU/ml) in the duodenum and ileum of pigs fed diets containing varied supplementation levels of waterleaf

Parameters	Diet 1	Diet 2	Diet 3	SEM	P-Value
Duodenum	3.50^{c}	2.00^{b}	0.00^{a}	0.65	0.01
Ileum	3.00	2.50	2.00	0.72	0.91

Means with different superscripts along the same row are significantly (P<0.05) different.
SEM: Standard error of mean
Diet 1: Basal diet without waterleaf; Diet 2: Basal diet + 5 g/kg waterleaf; Diet 3: Basal diet + 10 g/kg waterleaf.

Microbial Isolates Identified in the Duodenum and Ileum of Pigs Fed Diets Containing Varied Supplementation Levels of Waterleaf

Presented in Table 5 is the microbial isolate identified in the duodenum and ileum of pigs fed diets containing different levels of waterleaf supplementation. Identified fungi in the duodenum of the pigs include *Aspergillusniger, Penicillium sp., Mucormucedo, Rhizopus stolonifera,* and *Neurosporacrassa.* Similarly, *Rhizopus stolonifera*, *Mucor mucedo* were identified in the ileum of all the pigs. Also, *Lactobacillus sp., E. coli, Streptococcus* and *faecalis* were isolated in the duodenum of pigs fed Diet 1 and 3 while *Lactobacillus sp.,* was absent in the duodenum of pigs fed Diet 2. Bacteria isolated in ileum of all the pigs include *E. Coli.,* and *Lactobacillus sp.,* except those fed Diet 3 where *Pseudomonas sp*. was absent in their ileum.

Table 5: Microbial isolates (× 10^{-5} CFU/ml) in the duodenum and ileum of pigs fed diets containing varied supplementation levels of waterleaf

	Fungi		Bacterial	
Treatment	Duodenum	Ileum	Duodenum	Ileum
Diet 1	*Aspergillus niger, Penicillium sp., Mucor mucedo, Rhizopus stolonifera, Neurospora crassa*	*Rhizopus stolonifera, Mucor mucedo*	*Lactotobacillus sp., E. coli, Streptococcus faecalis*	*E. Coli., Pseudomonas sp., Lactobacillus sp*
Diet 2	*Aspergillus niger, Penicillium sp., Rhizopus stolonifera,*	*Rhizopus stolonifera, Mucor mucedo*	*E. coli, Streptococcus faecalis*	*E. Coli., Pseudomonas sp., Lactobacillus sp*
Diet 3	Nill	*Rhizopus stolonifera, Mucor mucedo*	*Lactotobacillus sp., E.coli, Streptococcus faecalis*	*E. Coli., Lactobacillus sp*

Diet 1: Basal diet without waterleaf; Diet 2: Basal diet + 5 g/kg waterleaf; Diet 3: Basal diet + 10 g/kg waterleaf

Moisture Retention Properties and Fat Content of Meat from Pigs Fed Diets Containing Varied Supplementation Levels of Waterleaf

Moisture retention properties from pigs fed diets containing varying supplementation levels of waterleaf is presented in Table 6. It was observed that all parameters evaluated were significantly (P<0.05) different except the moisture content. The moisture content of the meat ranged from 71.82% to 74.28%. Thaw and cooking loss of the meat decreased with the increase in the supplemental levels of waterleaf from 1.36% and 40.40% in pig fed Diet 1 to 0.78% and 33.55% in pig fed Diet 3 respectively. It was observed that fat content was not significantly (P>0.05) different. The value of fat content in the meat ranged from 11.90% to 17.57%, the least value was observed in meat of pigs fed Diet 2 and the highest from pigs fed Diet 1.

Table 6: Moisture retention properties and fat content of meat from pigs fed diets containing varied supplementation levels of waterleaf

Treatment	Moisture Content (%)	Thaw Loss (%)	Cooking Loss (%)	Fat Content (%)
Diet 1	72.66	1.36[a]	40.40[a]	17.57
Diet 2	71.83	0.98[b]	35.69[b]	11.90
Diet 3	74.28	0.78[b]	33.55[b]	15.50
SEM	0.74	0.12	1.69	1.78
P-Value	0.43	0.03	0.02	0.45

Means with different superscripts along the same column are significantly (P<0.05) different, SEM: Standard error of mean, Diet 1: Basal diet without waterleaf; Diet 2: Basal diet + 5 g/kg waterleaf; Diet 3: Basal diet + 10 g/kg waterleaf

Lipid Oxidation of Meat from Pigs Fed Diets Containing Varied Supplementation Levels of Waterleaf

The lipid oxidation of meat from pigs fed diets containing varying supplementation levels of waterleaf is shown in Table 7. The lipid oxidation of meat on day 0 was significantly different (P<0.05) while that of Day 8 was not. On day 0, the lipid oxidation of the meat increased with increased in supplementation levels of witted waterleaf in their diet from 0.30% to 1.16%. The lipid oxidation of the meat on Day 8 ranged from 2.40% to 2.71%, the highest value is observed in pigs fed Diet 1 while the least was observed in meat of pigs fed Diet 2.

Table 7: Lipid oxidation (MDA/Kg) of meat from pigs fed diets containing varied supplementation levels of waterleaf

	Day 0	Day 8
Diet 1	0.30[b]	2.71
Diet 2	0.48[b]	1.53
Diet 3	1.16[a]	2.40
SEM	0.16	0.24
P-Value	0.04	0.12

Means with different superscripts along the same column are significantly (P<0.05) different.
SEM: Standard error of mean
Diet 1: Basal diet without waterleaf; Diet 2: Basal diet + 5 g/kg waterleaf; Diet 3: Basal diet + 10 g/kg waterleaf

DISCUSSION

The dry matter values obtained for proximate composition of experimental diets are comparable to those reported by (12), who recorded 86.50% – 88.10% in pig diets supplemented with leaf meals. High dry matter values are desirable as they indicate higher nutrient density and improve feed storage stability (13). The crude protein levels obtained in this study are higher than the minimum requirement of 14% – 16% recommended for growing and finishing pigs (14). This implies that all the formulated diets contained sufficient protein for growth and tissue development, adequate dietary protein is essential for muscle accretion, enzyme synthesis, and overall performance in pigs (15).

In this study, it was observed that all parameters evaluated were not significantly ($P>0.05$) different among treatment groups. Pigs fed Diet 2 (basal diet + 5 g/kg waterleaf) recorded the highest weight gain (14.31 kg) compared to Diet 1 (13.23 kg) and Diet 3 (11.29 kg). This superior performance can be attributed to the balanced nutrient composition, particularly crude protein and digestible energy, which support growth and metabolic functions. The trend aligns with the findings of (16), who reported that weight gain in pigs is largely influenced by the protein level, diet palatability, nutrient digestibility, and dry matter intake. Daily weight gain followed a similar trend, with pigs fed Diet 2 achieving the highest gain of 255.48 g/day, reflecting the superior utilization of nutrients in this diet.

Feed conversion ratio (FCR), which reflects the efficiency of converting feed into body mass, was lowest in pigs fed Diet 2 (2.82), indicating the most efficient feed utilization. In contrast, pigs on Diet 3 had the highest FCR (3.04), suggesting that higher supplementation levels of waterleaf may reduce the efficiency of feed utilization. A lower FCR is desirable as it indicates that less feed is required to produce a unit of weight gain (17).

Total feed intake and daily feed intake followed the same trend, with pigs fed Diet 2 consuming more feed (40.49 kg total; 723.07 g/day). This implies that moderate supplementation of waterleaf improves feed palatability and encourages voluntary feed intake. Factors such as diet composition, nutrient density, age, breed, and environmental conditions also contribute to variations in feed intake, FCR, and growth performance (18, 19).

Bacteria count suggests that waterleaf supplementation did not negatively affect the microbial population in the duodenum, which is consistent with the observations of (20), who reported stable duodenal bacterial counts in pigs fed diets supplemented with leafy vegetables. In contrast, the ileum bacterial counts were significantly affected by dietary treatments ($P<0.05$). Pigs fed Diet 1 had the highest ileal bacterial count (8.50×10^5 CFU/ml), while pigs fed Diet 3 recorded the lowest count (2.50×10^5 CFU/ml). This reduction in ileal bacterial load with increasing waterleaf supplementation may be attributed to the functional properties in waterleaf, which can exhibit antimicrobial properties and modulate gut microflora (21, 22).

The reduction in duodenal fungal population with increasing waterleaf supplementation is beneficial for pigs, as high fungal load in the upper small intestine can compete with host digestive enzymes and reduce nutrient digestibility (23). The complete suppression of fungi in the duodenum at 10 g/kg waterleaf demonstrates the potential of waterleaf as a natural antifungal feed additive,

supporting better gut health and improving nutrient absorption.

Bacterial isolates showed more consistent patterns across diets. *Lactobacillus sp.* was present in both the duodenum and ileum across all diets. This genus plays a crucial role in gut health by fermenting carbohydrates into lactic acid, lowering intestinal pH, inhibiting pathogenic microbes, and enhancing nutrient absorption and feed efficiency (24). *Escherichia coli* was detected in all gut regions; while many strains are commensal, some can be opportunistic pathogens if overpopulated, potentially causing diarrhea or growth retardation (25). *Streptococcus faecalis* was isolated from the duodenum and contributes to lactic acid production, supporting intestinal pH regulation and suppressing harmful microbes (26). *Pseudomonas sp.* appeared in the ileum, which is generally opportunistic but may play minor roles in nutrient competition without significant harm under normal gut conditions (27).

Reduction in water loss can be attributed to the antioxidant and polyphenolic compounds present in waterleaf, which may stabilize muscle cell membranes, reduce protein denaturation, and improve the water-holding capacity of meat (28, 29). Improved water-holding capacity is beneficial for meat processing, as it reduces drip and cooking losses, which not only preserves juiciness and tenderness but also minimizes economic losses during storage and handling (30).

The fat content in pigs fed waterleaf aligns with reports by (31), who observed reduced fat deposition in pigs fed diets supplemented with plant-derived bioactive compounds. Similarly, studies by Surai (32) demonstrated that phytochemicals from plants reduce lipid accumulation and improve oxidative stability in meat, supporting the trend observed in the current study. These results suggest that incorporating moderate levels of waterleaf in pig diets can improve meat nutritional quality, enhance oxidative stability, and contribute to healthier meat for consumers.

The initial higher MDA in Diet 3 may reflect increased polyunsaturated fatty acids or enhanced oxidative substrates due to waterleaf supplementation, which contains phytochemicals capable of interacting with lipids (33). However, the differences at day 0 were significant ($P < 0.05$), indicating that waterleaf supplementation can influence the initial oxidative status of meat. By day 8, MDA levels increased across all treatments, reflecting normal lipid oxidation during storage. The meat of pigs fed basal diet reached 2.71 mg/kg, although differences at day 8 were not statistically significant ($P > 0.05$), the trend suggests that moderate supplementation of waterleaf (5 g/kg) may have a protective effect against lipid oxidation during storage. These results are in line with previous studies reporting that plant-based feed additives can modulate meat oxidative stability. For example, dietary supplementation with green leafy vegetables or herbal extracts rich in antioxidants has been shown to reduce MDA formation in pork and other meats during storage (34, 35).

CONCLUSION

Fresh waterleaf (*Talinum triangulare*) can be used as a beneficial dietary supplement in feeding of weaned pigs. Supplementation at 5 g/kg (Diet 2) produced the best results in terms of growth performance, gut microbial balance, and overall meat quality. Higher inclusion levels (10 g/kg) showed moderate benefits but did not outperform

the 5 g/kg supplementation level. The study indicates that fresh waterleaf enhances nutrients supply, supports healthy gut microbial populations, and maintains acceptable meat quality characteristics. It use as a low cost, readily available feed resources is therefore recommended for pig farmers seeking to improve productivity and reduce feed cost.

APPLICATION

From this study, it could therefore be recommended that:

1. Moderate supplementation of waterleaf (5 g/kg diet) is recommended for growing pigs to optimize growth performance, nutrient utilization, and meat quality.
2. Further studies should investigate the long-term effects of waterleaf supplementation on carcass traits, immune function, and reproductive performance in pigs.
3. Research into the optimal combination of waterleaf with other plant-based feed additives may enhance synergistic effects on oxidative stability and gut microbiota.

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