Efficacy of *Parquetina nigrescens* Leaf Extract on Growth Performance and Gastrointestinal Response of Japanese Quails

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Abstract

Japanese quail has the potential to complement the gap in the protein need of Nigerians. This study aimed to evaluate the effects of varying inclusion levels of Parquetina nigrescens leaf extract (PNLE) on the growth performance, gastrointestinal response, and plasma response of Japanese quails (Coturnix coturnix japonica). Fresh leaves of P. nigrescens were harvested. Two-hundred day old of unsexed Japanese quails were weighed and randomly allocated to 5 dietary treatment groups with forty birds at 4 replicates of 10 chicks, respectively. The leaf was obtained by harvesting and blending the leaves, using 50 g of leaves in 1000 mL of water. The treatment were as follows: (T1) which is the control did not receive any PNLE, T2, T3, T4 and T5 administered 0.2, 0.4, 0.6, 0.8 mL of PNLE per 500 mL of water, respectively, for 6 week of experiment. Feed and water were provided ad libitum and mortality was recorded as it occurred. Parameters measured were performance (feed intake, weight gain, feed conversion ratio (FCR) and gastrointestinal response (Internal organ of GIT and morphometrics). All data were subjected to analysis of variance using Duncan's multiple range test in SAS (2010) where p value < 0.005 was considered to be statistically significant. For performance, the results showed that the administration of PNLE did not have significant influence (p > 0.05) on all the parameters in the starter and finisher phases. It was observed that when all the parameters were put together without considering the phases, all the parameters considered were not significant (p > 0.05) except feed conversion ratio (FCR) with the highest value being in T1 (5.041) and the lowest value being T4 (3.183). For the gastrointestinal response parameters, the inclusion of PNLE had significant effect (p < 0.05) on crop pH, proventriculus temperature, proventriculus pH, gizzard weight, gizzard temperature, relative gizzard weight, jejunum length and ileum temperature. It was concluded that the inclusion of PNLE had no detrimental effect on growth performance, gastrointestinal response and plasma of Japanese quail, hence it can be used up to 0.4 mL into 500 mL of water.

Keywords: gastrointestinal response, growth performance, Japanese quail, Parquetina nigrescens

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INTRODUCTION

The continuous rise in population in developing countries necessitates the establishment of alternative sources of animal protein (El-Katcha *et al.*, 2015). The shortage of animal protein intake among the ever increasing human population in the third world countries has long been recognized and remains one of the greatest issues of concern today (Delgado, 2003; Akintunde *et al.*, 2011; Henchion *et al.*, 2017). Because of their resilience and short generation interval, Japanese quails are therefore a viable and

sustainable source of animal protein, particularly in developing nations. The use of natural plants and products as alternative therapeutic agents for a variety of illnesses and disease conditions has recently acquired popularity on a global scale (Atanasov *et al.*, 2015; Ugboko *et al.*, 2020). It has been demonstrated that medicinal plants contain an abundance of phytochemicals that are advantageous to the health of livestock (Akintunde *et al.*, 2021).

The medicinal plant's work is related to improving the gut environment and enhancing the body's immunity by inhibiting or causing a hindrance to the growth of pathogenic bacteria. It has been used as food additives to demonstrate the high ability to improve the physiological, immunological, and reproductive traits of poultry (Ali *et al.*, 2021). The use of phytogenics also has been proven to enhance poultry's ability to produce eggs and meat (Nichol and Steiner, 2010; Kirana *et al.*, 2024).

Nigerian traditional medicine uses a perennial plant called Parquetina nigrescens, which is a member of the Periplocaceae family and grows in and around villages in both Senegal and Nigeria (Oluwafemi and Debiri, 2008; Guede et al., 2010; Ayoola et al., 2011). It is useful in the treatment of sickle cell and anemia (Imaga et al., 2010) and gastro-intestinal disorder (GID) (Odetola et al., 2006). Scientifically proven and well-documented benefits of P. nigrescens leaf hematinic, extracts include antidiabetic, cardiotonic, anti-ulcerative, and antioxidant properties (Datté and Ziegler, 2001; Saba et al., 2010; Ozaslan, 2011; Ayoola et al., 2011). The analgesic, antiinflammatory and antipyretic effects of P. nigrescens leaf extract have been documented (Owoyele et al., 2009).

Olumide et al. (2022) reported that the leaf extracts of P. nigrescens had high concentration of moisture (7.80%), crude fibre (9.38%), crude protein (8.40%), ether extract (9.38%), and ash (6.90%). Mineral and vitamin analysis showed that P. nigrescens leaf extract contained macro minerals (%) such as sodium (0.36), calcium (29.96), phosphorus (6.88), potassium (23.21), magnesium (4.05), and micro minerals such as silicon (23.71 ppm), aluminium (4.34%), iron (3.59%), manganese (1.46 ppm) and chlorine (0.33%) and high content of vitamins A (2.27 mg/100 g), B1 (270.25 mg/100 g), B2 (850.26 mg/100 g), B3 (325.20 mg/100 g), C (16.20 mg/100 g) and E (0.015 mg/100 g) respectively (Olumide al., et 2022). Phytochemical evaluation revealed that P. nigrescens leaf extract have high contents of alkaloids (8.27 mg/100 g), flavonoids (2.25 mg/100 g), glycosides (0.06 mg/100 g), saponin (5.20 mg/100 g), steroids (0.20 mg/100 g), phenols (0.86 mg/100 g), terpenoids (0.52 mg/100 g), tannin (6.30 mg/100 g) and

anthraquinones (1.55 mg/100 g) (Olumide *et al.*, 2022). Olumide *et al.* (2022) however concluded that *P. nigrescens* leaf extract are of high nutritional quality due to high crude protein, vitamins and mineral contents especially calcium and potassium with the resultant phytochemicals attributes that could serve as feed additives in monogastric animal production. Akintunde *et al.* (2023^a) also reported that *P. nigrescens* leaf extracts has good antioxidant potential thus improving the health status of broiler chickens.

There is limited information on the digestive physiology of Japanese quails especially as it relates to the utilization of phytogenics hence the study is aimed at evaluating the growth performance and gastrointestinal response of Japanese quails to administration of *P. nigrescens* leaf extract.

MATERIALS AND METHODS

Ethical Approval

All of the protocol related with experimental animals were performed according to the guidelines and approved by Babcock University Health Research Ethics with the reference number BUHREC793/22.

Study Period and Location

The study was carried out from January to February, 2023 at the poultry unit of Babcock University farm house, Ilishan-Remo, Ogun State, Nigeria. Ilishan-Remo is in the rain forest zone of Nigeria, having a mean temperature of 27°C.

Fresh *P. nigrescens* leaves were collected from Ilisan-Remo, Ikenne Local Government Area in Ogun State, Nigeria. The plant was identified and authenticated by a botanist in the Department of Basic Sciences, Babcock University, Ilishan-Remo, Ogun State, Nigeria.

Management of Experimental Birds and Design

The house was cleaned, disinfected and dried for two weeks before the arrival. Before the arrival of the day-old quail, drinkers and feeders will be properly cleaned and disinfected. A total of 200-day-old quail was obtained from National Veterinary Research Institute, Ikire, Office, Osun State, Nigeria. The quails' initial weight was taken before they were randomly assigned to one of five treatments (T1, T2, T3, T4 and T5) with four replicates of 10 quails each in a completely randomized design. The birds were given feed and water *ad libitum* for 6 weeks.

Preparation of Test Ingredient

The fresh leaves of *P. nigrescens* were harvested, thereafter, it was washed. 50 g of fresh leaves harvested was blended with 1000 mL of water using a blender. The blending was done for about 4 minutes after which the blended samples will be filtered using standard filter paper (Whatman paper No. 1). The filtrate was then administered to the quails.

Experimental Treatments

Isonitrogenous and isocaloric diets were formulated for all the birds. T_1 which was the control had no administration of *P. nigrescens* in its water, T_2 , T_3 , T_4 , T_5 had 0.2 mL, 0.4 mL, 0.6 mL, and 0.8 mL of *P. nigrescens* extract per 500 mL of water respectively. Table 1 showed the gross composition of experimental diets at starter and finisher phases.

Data Collection

Data were collected on performance (feed intake, weight gain, feed conversion ratio (FCR), gastrointestinal response (Esophagus, Crop, Gizzard, Proventriculus, Small intestine, large intestine and gut morphology), Plasma Concentration (Plasma protein, Albumin, Globin) and visceral organ morphometrics.

Feed Intake

Feed intake was calculated weekly. This was done by subtracting the amount of feed left in the feeder from the initial amount of feed given in the previous day as feed intake for the day.

Feed intake (g) = Feed offered (g) - Feed leftover (g) (1)

Average feed intake = $\frac{\text{feeed offerd in } (g) - \text{feed leftover } (g)}{\text{Total nuber of birds in the group}}$ (2)

Water Intake

Water intake was calculated weekly. This was done by subtracting the volume of water left in the drinker from the initial volume of water given in the previous day.

Water intake = Volume of water giving (mL) - Volume of water left (m) (3)

Gastrointestinal Study

At the end of the starter phase (21 days) and finisher phase (42 days), two birds from each replicate were randomly selected and sacrificed for gastro-intestinal tracts developmental studies. The contents of the guts at each phase of the GIT were excised and be carefully examined for a detailed study on their developmental digestive physiology.

Gut Morphology

On day 42, two birds from each replicate were randomly selected and sacrificed for anatomical examination to predict the nutrient absorption abilities of the bird in response to the varying inclusions of *P. nigrescens* leaf extract. Internal organs of the gastrointestinal tract such as duodenum, jejunum, and ileum were identified and removed. The morphometric variables to examine include villus height, villus width, crypt depth, villus height: crypt depth and muscle thickness. Intestinal histology measurements were done according to the procedures of Yu *et al.* (1998).

Visceral Organ Evaluation

Four birds were randomly selected from each treatment for visceral organ evaluation. The selected birds were starved overnight and their weights were taken. The birds were then eviscerated and weighed. The following parameters were observed and measurements were recorded: Gizzard, Liver, Kidney, Heart, Crop, Preventiculus, Spleen, Pancreas, Small intestine, large intestine, Caecum.

Data Analysis

Data collected was subjected to Analysis of Variance (ANOVA) according to the procedure of SAS (2002). Significant difference between the treatment's means was separated using Duncan multiple range test.

RESULTS AND DISCUSSION

Table 2 showed that there was no significant difference (p > 0.05) in the initial weight, body weight, total feed intake, total water intake (TWI), weight gain and feed conversion ratio at starter phase. Table 3 showed that there was no significant difference (p > 0.05) in the initial weight, final body weight gain, feed intake, water intake, weight gain and feed conversion ratio at the finisher phase. Table 4 showed that there was

significant difference (p < 0.05) in crop pH, proventriculus temperature, proventriculus pH, gizzard weight, gizzard temperature, relative gizzard weight, duodenum length, jejunum length, jejunum pH and ileum temperature but no significant difference (p > 0.05) in crop weight, temperature, relative crop crop weight, proventriculus weight, relative proventriculus weight, gizzard pH, duodenum temperature, duodenum pH, jejunum temperature, ileum length, ileum pH, caecum length, caecum temperature, caecum pH, heart weight and liver weight at the end of the starter phase.

Ingredient	Starter	Finisher
Maize	48.00	59.00
Soybean	33.00	30.00
Wheat offal	6.00	5.64
Fishmeal	4.00	-
Palm oil	-	3.00
Vegetable oil	4.00	-
Meat-bone meal	2.50	-
Limestone	1.00	-
Dicalcium phosphate	0.50	1.56
Oyster shell	-	1.00
Salt	0.40	0.25
Methionime	0.20	0.25
Lysine	0.10	0.05
Avatec	-	0.06
CP (%)	23.00%	19.33%
ME(Kcal/kg)	2946.64	3062.95

Table 2. Performance characteristics of Ja	panese quail given P. nigrescens at starter p	ohase
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Parameters	T1	T2	Т3	T4	Т5
Initial Weight (g)	7.0 ± 0.00	7.2 ± 0.00	7.1 ± 0.02	7.0 ± 0.00	7.2 ± 0.00
Body Weight (g)	74.1 ± 3.35	83.1 ± 4.77	75.6 ± 3.30	81.8 ± 3.29	73.9 ± 2.50
TFI/Bird (g)	195.9 ± 11.69	166.4 ± 5.88	190.3 ± 13.40	175.7 ± 7.00	191.2 ± 11.82
TWI/Bird (mL)	542.1 ± 23.51	579.9 ± 118.86	549.7 ± 32.68	500.5 ± 5.98	522.3 ± 12.36
Weight Gain (g)	67.1 ± 3.35	75.9 ± 4.77	68.5 ± 3.31	74.8 ± 3.29	66.7 ± 2.50
FCR	3.1 ± 0.39	2.5 ± 0.27	3.1 ± 0.05	2.5 ± 0.34	2.8 ± 0.22

TFI = Total Feed Intake, TWI = Total Water Intake, FCR = Feed Conversion Ratio.

Table 5 showed the gastrointestinal response of male Japanese quails to administration of *P*. *nigrescens* leaf extract at finisher phase. There was significant difference (p < 0.05) in live bird weight, slaughtered bird weight, proventiculus pH, duodenum weight, relative duodenum weight, jejunum length, caecum weight and relative caecum weight. There was no significant difference (p > 0.05) in crop weight, crop temperature, crop pH, proventiculus weight, proventiculus temperature, gizzard weight, relative gizzard weight, gizzard temperature, gizzard temperature, gizzard pH, duodenum length, jejunum weight, ileum weight, relative ileum weight, ileum length, caecum length, liver weight, relative liver weight, spleen weight, heart weight and relative heart weight.

Table 6 showed the gastrointestinal response of female Japanese quails to administration of P. nigrescens leaf extracts at finisher phase. There was significant difference (p < 0.05) in the crop weight, relative crop weight, crop pH and gizzard weight. There was no significant difference (p > p)0.05) in the live weight, slaughtered weight, crop temperature, proventiculus pH, relative gizzard weight, gizzard temperature, gizzard pH, duodenum weight, relative duodenum weight, duodenum length, jejunum weight, relative jejunum weight, jejunum length, ileum weight, relative ileum weight, ileum length, caecum weight, relative caecum weight, caecum length, liver weight, relative liver weight, spleen weight, heart weight and relative heart weight.

Table 7 showed the histomorphometrics of Japanese quails to administration of *P. nigrescens* leaf extracts. At the duodenum, it was observed that there is significant difference (p > 0.05) villi height, villi width, cryptal depth, cryptal depth, Muscle thickness, villi height, villus height: villi depth, villi height, villi width, cryptal depth. At the jejunum, there was a significant difference (p > 0.05) in villi height, villi width, cryptal depth, cryptal width, muscle thickness, villi height, cryptal depth, cryptal width, muscle thickness, villi height: crypt depth. In the ileum, there was a significant difference (p > 0.05) in villi height, villi width, muscle thickness, villi height, villi width, cryptal depth, cryptal depth, cryptal depth, cryptal width, muscle thickness, villi height, villi width, cryptal depth, cryptal depth, cryptal width, muscle thickness, villi height, villi width, cryptal depth, cryptal depth, cryptal width, muscle thickness, villi height, villi width, cryptal depth, cryptal depth, cryptal depth, cryptal width, muscle thickness, villi height, villi width, cryptal depth, cryptal depth, cryptal depth, cryptal width, muscle thickness, villi height, villi width, cryptal depth, cryptal depth, cryptal width, muscle thickness, villi height, villi width, cryptal depth, cryptal width, muscle thickness, villi height, cryptal depth, cryptal width, muscle thickness, villi height, cryptal depth, cryptal width, muscle thickness, villi height, and crypt depth.

Nutrient digestion and absorption play an important role in improving growth performance. The results showed that the administration of the extracts of *P. nigrescens* did not significant influence performance parameters of Japanese quails at starter phase. The results of this study are in agreement with the observations of Akintunde *et al.* (2023^b), who reported no significant differences in body weight, feed intake, and

weight gain at the starter phase in Japanese quails administered an aqueous egg-lime-molasses mixture. The results obtained from this study was however in contrast with the observations of Ustundag and Ozdogan (2023) who studied the effects of mulberry leaves on growth performance, carcass characteristics, and meat quality of Japanese quail. They reported that the inclusion of the leaves significantly influenced all the growth parameters at the starter phase. The variation might be due to the differences in the test ingredients. The results were also in contrast with the findings of Oreagba (2022) who studied the performance and immunological response of broiler chickens to P. nigrescens leaf extracts and reported that the administration of P. nigrescens significantly influenced body weight, weight gain and feed conversion ratio at the end of the starter phase. The variation could be as a result of different poultry specie used in the present study.

A study by Asghar *et al.* (2022) investigated the effect of dietary supplementation of black cumin seeds (*Nigella sativa*) on performance, carcass traits, and meat quality of Japanese quails (*Coturnix coturnix japonica*). They observed improvements in body weight, feed intake, and feed conversion ratio in the treated quails. Similarly, a study by Al-Tekreeti and Allaw (2022) investigated the effect of adding nano garlic to the quail's diet on productive performance. There was also improvements in body weight gain, feed intake, and FCR in the treated quails. The variations in results might be as a result of variations in the test ingredients.

The results obtained from this study was in agreement with the observations of Akintunde et al. (2023^b) who reported that there was no significant difference in body weight, feed intake and feed conversion ratio at the finisher phase in Japanese quails to the administration of aqueous egg-lime-molasses mixture. The results obtained from this study were in contrast with the study of Reda et al. (2020) who investigated the effect of dietary supplementation of biological curcumin nanoparticles on growth and carcass traits, antioxidant status, immunity and caecal microbiota of Japanese quails.

Table 3. Performance characteristic of Japan	nese quail administered	P. nigrescen at finisher phase
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Parameters	T1	T2	Т3	T4	
Initial Weight (g)	74.1 ± 3.35	83.1 ± 4.77	75.6 ± 3.30	81.8 ± 3.29	73.9 ± 2.50
BW (g)	132.0 ± 4.90	149.3 ± 4.62	148.4 ± 4.03	153.3 ± 3.72	142.0 ± 13.34
TFI/Bird (g)	373.1 ± 49.31	313.4 ± 14.49	394.2 ± 51.61	304.0 ± 7.52	374.8 ± 53.47
TWI (mL)	1391.6 ± 56.24	1311.0 ± 27.44	1453.2 ± 112.41	1303.9 ± 66.36	1387.6 ± 68.92
Weight Gain (g)	57.9 ± 4.72	66.1 ± 7.43	72.8 ± 5.49	71.5 ± 5.49	68.1 ± 13.64
FCR	7.6 ± 0.94	4.2 ± 0.21	5.9 ± 1.18	3.9 ± 0.40	9.0 ± 3.11

TFI = Total Feed Intake, TWI = Total Water Intake, FCR = Feed Conversion Ratio.

Table 4. Gastrointestinal response of Japanese quail after administration of P. nigrescens leaf extracts at starter phase

Parameters	T1	T2	Т3	T4	T5
Live Weight (g)	70.0 ± 7.00	90.5 ± 5.50	83.0 ± 4.00	82.0 ± 9.00	80.5 ± 3.50
Crop Weight (g)	0.0 ± 0.00	0.0 ± 0.00	0.5 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Relative Crop Weight (%)	0.0 ± 0.00	0.0 ± 0.00	0.6 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Proventriculus Weight (g)	0.0 ± 0.00	0.0 ± 0.00	1.0 ± 0.01	0.0 ± 0.00	0.0 ± 0.00
Relative Proventriculus					
Weight (%)	0.0 ± 0.00	0.0 ± 0.00	1.3 ± 0.03	0.0 ± 0.00	0.0 ± 0.00
Gizzard Weight (g)	$2.0\pm0.00^{\text{b}}$	0.0 ± 0.00^{a}	$3.0\pm1.00^{\text{b}}$	$2.0\pm0.00^{\text{b}}$	3.0 ± 0.00^{b}
Relative Gizzard Weight (%)	$2.9\pm0.29^{\text{b}}$	0.0 ± 0.00^{a}	3.7 ± 1.38^{b}	$2.5\pm0.27^{\text{b}}$	3.7 ± 0.162^{b}
Duodenum Length (cm)	9.0 ± 1.50^{a}	13.8 ± 2.25^{ab}	12.5 ± 1.00^{ab}	9.6 ± 1.55^{ab}	14.9 ± 0.65^{b}
Jejunum Length (cm)	14.5 ± 0.50^{b}	$14.7\pm2.15^{\text{b}}$	12.8 ± 0.50^{ab}	$14.3\pm0.25^{\text{b}}$	$9.0 \pm 1.00^{\rm a}$
Ileum Length (cm)	8.1 ± 1.90	7.3 ± 0.75	7.8 ± 1.25	11.5 ± 1.00	7.5 ± 2.50
Ceacum Length (cm)	9.3 ± 0.75	9.0 ± 1.00	12.0 ± 4.00	10.2 ± 2.15	9.0 ± 1.00
Heart Weight (g)	0.0 ± 0.00	0.0 ± 0.00	1.0 ± 0.00	0.0 ± 0.00	1.0 ± 0.00
Relative Heart Weight (%)	0.0 ± 0.00	0.0 ± 0.00	1.2 ± 0.01	0.0 ± 0.00	1.2 ± 0.03
Liver Weight (g)	2.0 ± 0.00	3.5 ± 0.50	1.5 ± 0.50	2.0 ± 0.00	2.0 ± 0.00
Relative Liver Weight (%)	2.9 ± 0.05	3.9 ± 0.04	1.8 ± 0.00	2.4 ± 0.02	2.5 ± 0.03
Temperature (°C)					
Crop Temperature	27.8 ± 0.75	28.1 ± 0.60	34.1 ± 4.00	30.2 ± 0.10	30.9 ± 0.80
Proventriculus Temperature	28.4 ± 0.10^{ab}	$27.6\pm0.30^{\rm a}$	$31.3 \pm 1.80^{\text{b}}$	29.6 ± 0.45^{ab}	30.1 ± 0.05^{ab}
Gizzard Temperature	28.1 ± 0.00^{a}	$27.2\pm0.30^{\rm a}$	$30.2\pm0.90^{\text{b}}$	$30.0\pm0.10^{\text{b}}$	29.9 ± 0.05^{b}
Duodenum Temperature	28.8 ± 1.50	29.2 ± 2.75	30.0 ± 2.10	32.4 ± 1.10	29.4 ± 1.10
Jejunum Temperature	28.2 ± 0.70	27.8 ± 0.15	30.2 ± 1.70	30.8 ± 0.30	30.3 ± 0.15
Ileum Temperature	$28.1\pm0.10^{\rm a}$	$27.9\pm0.85^{\rm a}$	30.3 ± 1.15^{ab}	$30.8\pm0.25^{\text{b}}$	31.1 ± 0.60^{b}
Caecum Temperature	28.6 ± 0.10	28.3 ± 1.00	30.7 ± 0.95	30.8 ± 0.45	30.9 ± 0.50
рН					
Crop pH	6.7 ± 0.25^{b}	$6.2\pm0.05^{\rm a}$	$6.1\pm0.10^{\rm a}$	$6.3\pm0.05^{\rm a}$	6.1 ± 0.20^{a}
Proventriculus pH	$6.2\pm0.00^{\text{b}}$	6.3 ± 0.30^{b}	5.4 ± 0.65^{ab}	$4.8\pm0.25^{\rm a}$	4.9 ± 0.00^{a}
Gizzard pH	4.3 ± 1.05	5.0 ± 0.60	4.3 ± 0.15	4.5 ± 0.00	4.7 ± 0.25
Duodenum pH	5.9 ± 0.05	6.2 ± 0.10	5.3 ± 0.30	5.7 ± 0.50	5.7 ± 0.05
Jejunum pH	6.0 ± 0.10^{ab}	6.3 ± 0.10^{b}	5.5 ± 0.40^{ab}	5.9 ± 0.25^{ab}	$5.4\pm0.10^{\rm a}$
Ileum pH	6.4 ± 0.25	6.7 ± 0.15	5.9 ± 0.35	6.5 ± 0.35	6.0 ± 0.10
Caecum pH	6.3 ± 0.55	6.5 ± 0.35	5.8 ± 0.15	6.4 ± 0.45	5.9 ± 0.15

^{a,b,ab} Means along the same row with different superscript are significant different (p < 0.05).

They observed that the group of chickens fed with a diet containing the highest level of curcumin nanoparticles had the highest body weight gain but least feed intake compared to the control group. Similarly, a study by Abu Hafsa *et al.* (2020) investigated the effect of dietary *Moringa oleifera* leaves on the performance, ileal microbiota and antioxidative status of broiler chickens. The authors found that the chicks fed 0.5% and 1% *M. oleifera* leaves exhibited a higher final body weight (FBW), greater weight gain (BWG), and better average daily gain (ADG), feed intake gradually decreased as the inclusion rate of *M. oleifera* leaves in the broilers' diet increased to 5% while the best feed conversion ratio and performance index (PI) were observed in broilers that were fed the *M. oleifera* leaves diet.

Table 5. Gastrointestinal response of Japanese quail after administration of P. nigrescens leaf extracts at finisher phase

Parameters	T1	T2	Т3	T4	T5
Live Bird Weight (g)	136.0 ± 4.00^{c}	$120.0\pm0.00^{\text{b}}$	125.0 ± 5.00^{bc}	126.0 ± 0.00^{bc}	102.5 ± 1.50^{a}
Slaughtered Bird Weight (g)	126.5 ± 2.50^{c}	111.5 ± 0.50^{b}	117.0 ± 5.00^{bc}	117.5 ± 1.50^{bc}	97.0 ± 2.00^{a}
Crop Weight (g)	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Proventriculus Weight	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Gizzard Weight (g)	4.0 ± 0.00	3.5 ± 0.50	4.0 ± 1.00	3.5 ± 0.50	3.5 ± 0.50
Relative Gizzard Weight (%)	2.9 ± 0.09	2.9 ± 0.42	3.2 ± 0.67	2.8 ± 0.40	3.4 ± 0.54
Duodenum Weight (g)	$2.0\pm0.00^{\text{b}}$	$0.5\pm0.05^{\rm a}$	$0.0\pm0.00^{\mathrm{a}}$	$0.0\pm0.00^{\rm a}$	0.0 ± 0.00^{a}
Relative Duodenum Weight	$1.5\pm0.04^{\text{b}}$	$0.4\pm0.04^{\rm a}$	$0.0\pm0.00^{\mathrm{a}}$	$0.0\pm0.00^{\rm a}$	0.0 ± 0.00^{a}
(%)					
Duodenum Length (cm)	12.8 ± 2.25	9.2 ± 0.85	12.5 ± 1.50	10.5 ± 0.50	10.5 ± 0.50
Jejunum Weight (g)	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Jejunum Length (cm)	$14.0 \pm 1.00^{\text{b}}$	11.4 ± 1.40^{ab}	13.5 ± 0.50^{b}	$9.8\pm0.75^{\rm a}$	$10.0\pm0.01^{\rm a}$
Ileum Weight (g)	0.0 ± 0.00	0.3 ± 0.02	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Relative Ileum Weight (%)	0.0 ± 0.00	0.2 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Ileum Length	10.5 ± 1.50	9.8 ± 0.75	7.5 ± 0.50	7.5 ± 1.50	7.3 ± 0.25
Caecum Weight (g)	1.0 ± 0.00^{ab}	$0.0\pm0.00^{\rm a}$	$0.0\pm0.00^{\rm a}$	$0.0\pm0.00^{\mathrm{a}}$	2.0 ± 0.00^{b}
Relative Caecum Weight (%)	$0.7\pm0.01^{\rm a}$	0.0 ± 0.00^{a}	$0.0\pm0.00^{\rm a}$	$0.0\pm0.00^{\rm a}$	1.9 ± 0.03^{b}
Caecum Length (cm)	8.8 ± 3.25	8.5 ± 2.50	8.3 ± 1.75	7.3 ± 1.05	8.0 ± 1.00
Liver Weight (g)	3.0 ± 0.00	2.0 ± 0.00	2.0 ± 0.10	2.0 ± 0.00	2.0 ± 0.00
Relative Liver Weight (%)	2.2 ± 0.06	1.7 ± 0.00	1.6 ± 0.74	1.6 ± 0.00	1.9 ± 0.03
Spleen Weight (g)	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Heart Weight (g)	2.0 ± 0.00	1.5 ± 0.01	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Relative Heart Weight (%)	1.5 ± 0.04	1.3 ± 0.01	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Temperature (°C)					
Crop Temperature	31.6 ± 0.65	30.5 ± 0.90	32.6 ± 1.55	32.9 ± 0.20	33.1 ± 0.25
Proventriculus Temperature	30.8 ± 0.65	31.4 ± 0.50	32.1 ± 0.65	32.2 ± 0.55	31.3 ± 1.00
Gizzard Temperature	31.9 ± 0.65	31.5 ± 0.55	31.6 ± 0.45	33.3 ± 0.05	33.9 ± 2.45
рН					
Crop pH	5.8 ± 0.20	5.9 ± 0.30	5.9 ± 0.05	6.0 ± 0.10	5.8 ± 0.00
Proventriculus pH	5.6 ± 0.05^{ab}	5.7 ± 0.10^{ab}	$5.9\pm0.05^{\text{b}}$	5.8 ± 0.20^{ab}	$5.5\pm0.15^{\rm a}$
Gizzard pH	5.7 ± 0.10	4.7 ± 0.30	5.4 ± 0.40	5.7 ± 0.30	5.4 ± 0.30

a,b,c,ab,abc Means along the same row with different superscript are significant different (p < 0.05).

The study aimed to investigate the gastrointestinal response of Japanese quails *given P. nigrescens* leaf extract at different concentrations. The live bird weight, slaughtered bird weight, crop weight, crop temperature and

pH, proventriculus weight, temperature and pH, gizzard weight, temperature and pH, duodenum weight and length, jejunum length, ileum weight, length, caecum weight, length, liver weight, and spleen weight were evaluated. Other studies have

investigated the effects of different plant extracts on the gastrointestinal parameters of quails. Owolabi *et al.* (2017) studied the effects of aqueous *M. oleifera* leaf extracts on gut morphology and pH of hubbard broiler chickens and found that the extract had a significant effect on the pH of the gizzards, duodenum, jejunum, ileum, caecum, kidney and lung. Another study by Abudabos *et al.* (2018) investigated the effect of phytogenics on growth traits, blood biochemical and intestinal histology in broiler chickens exposed to *Clostridium perfringens* challenge. The study found that the thyme extract improved the weight of the pancreas and gizzard and increased the length of the small intestine but these values were not significant for the male and female Japanese quails in the present study and this could be due to differences in the test ingredient and variation in the species of birds used in both studies.

Table 6. Gastrointestinal response of female Japanese quail after administration *of P. nigrescens* leaf extracts at finisher phase

Parameters	T1	T2	Т3	T4	Т5
Live Weight (g)	145.5 ± 0.50	157.5 ± 8.50	147.0 ± 9.00	154.5 ± 3.50	136.0 ± 9.00
Slaughtered Weight (g)	142.0 ± 1.00	145.5 ± 7.50	140.5 ± 10.50	146.5 ± 3.50	127.5 ± 7.50
Crop Weight (g)	$0.0\pm0.00^{\rm a}$	$0.0\pm0.00^{\rm a}$	$1.5\pm0.50^{\text{b}}$	$0.0\pm0.00^{\rm a}$	0.5 ± 0.00^{ab}
Relative Crop Weight (%)	0.0 ± 0.00^{a}	$0.0\pm0.00^{\rm a}$	$1.0\pm0.24^{\rm b}$	$0.0\pm0.00^{\rm a}$	0.3 ± 0.03^{ab}
Proventriculus Weight (g)	0.0 ± 0.00	0.0 ± 0.00	1.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Relative Proventriculus	0.0 ± 0.00	0.0 ± 0.00	0.7 ± 0.02	0.0 ± 0.00	0.0 ± 0.00
Weight (%)					
Gizzard Weight (g)	4.5 ± 0.50^{ab}	5.0 ± 0.00^{ab}	5.5 ± 0.50^{b}	$4.0\pm0.00^{\rm a}$	5.0 ± 0.00^{ab}
Relative Gizzard Weight (%)	3.1 ± 0.33	3.2 ± 0.17	3.8 ± 0.57	2.6 ± 0.06	3.7 ± 0.24
Duodenum Weight (g)	2.5 ± 0.50	2.0 ± 0.00	1.0 ± 0.01	2.5 ± 0.50	1.5 ± 0.01
Relative Duodenum Weight	1.7 ± 0.35	1.3 ± 0.07	0.6 ± 0.04	1.6 ± 0.36	1.2 ± 0.11
(%)					
Duodenum Length (cm)	12.8 ± 2.25	14.3 ± 0.75	14.0 ± 1.00	15.5 ± 0.50	10.5 ± 2.50
Jejunum Weight (g)	1.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Relative Jejunum Weight	0.7 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
(%)					
Jejunum Length (cm)	16.0 ± 4.00	14.5 ± 1.50	14.0 ± 1.00	12.0 ± 0.00	12.0 ± 2.00
Ileum Weight (g)	4.5 ± 0.50	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Relative Ileum Weight (%)	3.1 ± 0.02	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Ileum Length (cm)	11.5 ± 0.55	11.6 ± 5.55	11.5 ± 1.50	11.5 ± 0.50	10.0 ± 1.00
Ceacum Weight (g)	1.0 ± 0.00	2.0 ± 0.00	0.0 ± 0.00	1.5 ± 0.05	0.0 ± 0.00
Relative Caecum Weight (%)	0.7 ± 0.00	1.2 ± 0.20	0.0 ± 0.00	0.9 ± 0.03	0.0 ± 0.00
Caecum Length (cm)	10.5 ± 0.50	9.6 ± 1.60	9.5 ± 1.50	10.5 ± 1.50	9.5 ± 1.50
Liver Weight (g)	3.0 ± 1.00	4.0 ± 1.00	3.5 ± 0.50	3.5 ± 0.50	4.5 ± 0.50
Relative Liver Weight (%)	2.1 ± 0.69	2.6 ± 0.77	2.4 ± 0.49	2.3 ± 0.37	3.3 ± 0.15
Spleen Weight (g)	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Heart Weight (g)	2.0 ± 0.00	1.5 ± 0.01	0.0 ± 0.00	0.0 ± 0.00	0.0 ± 0.00
Temperature (°C)					
Crop Temperature	33.3 ± 0.00	32.7 ± 0.95	33.8 ± 1.20	32.0 ± 0.80	32.8 ± 0.30
Proventriculus Temperature	31.3 ± 1.15	32.7 ± 0.25	31.9 ± 0.35	33.5 ± 0.55	33.0 ± 0.00
Gizzard Temperature	31.5 ± 1.45	32.9 ± 1.60	32.9 ± 0.20	31.9 ± 0.55	33.9 ± 1.05
рН					
Crop pH	$5.3\pm0.25^{\rm a}$	6.0 ± 0.10^{b}	6.0 ± 0.20^{b}	$5.9\pm0.05^{\text{b}}$	6.1 ± 0.20^{b}
Proventriculus pH	5.3 ± 0.80	6.0 ± 0.00	6.1 ± 0.15	6.1 ± 0.05	5.9 ± 0.15
Gizzard pH	3.4 ± 1.50	5.7 ± 0.30	5.7 ± 0.10	5.7 ± 0.15	5.6 ± 0.20
^{a,b,ab} Means along the same ro					

^{a,b,ab} Means along the same row with different superscript are significant different (p < 0.05).

Parameters	T1	T2	T3	T4	T5
Duodenum					
Villi Height	2283.2 ± 31.75^a	2448.9 ± 24.02^{b}	2319.2 ± 21.58^{a}	2229.7 ± 27.71^{a}	$2577.9 \pm 33.99^{\circ}$
Villi Width	$222.5\pm2.27^{\mathrm{b}}$	210.1 ± 0.03^{a}	217.8 ± 4.23^{ab}	$235.9\pm2.31^{\circ}$	$243.6\pm2.37^{\rm c}$
Cryptal Depth	471.9 ± 3.88^{b}	$452.4\pm9.05^{\text{b}}$	$467.0\pm18.51^{\text{b}}$	412.3 ± 6.55^a	$562.4\pm14.70^{\rm c}$
Cryptal Width	225.6 ± 5.23^{ab}	$213.1\pm2.92^{\rm a}$	221.2 ± 6.04^{ab}	234.1 ± 3.48^{b}	238.9 ± 7.93^{b}
Muscle	$177.4\pm16.35^{\mathrm{a}}$	$179.8\pm6.22^{\rm a}$	324.6 ± 12.24^{b}	311.0 ± 0.99^{b}	$299.8\pm18.64^{\text{b}}$
Thickness					
Villus Height:	$4.8\pm0.03^{\rm a}$	5.4 ± 0.15^{b}	4.9 ± 0.24^{ab}	$5.4\pm0.06^{\text{b}}$	$4.6\pm0.06^{\rm a}$
Crypt Depth					
Jejunum					
Villi Height	$1954.9\pm13.17^{\mathrm{a}}$	1973.9 ± 9.00^{a}	$2256.9\pm7.54^{\rm c}$	2147.9 ± 5.92^{b}	2396.9 ± 5.24^d
Villi Width	$226.0\pm3.05^{\rm c}$	$198.7\pm0.57^{\rm a}$	$217.1\pm0.49^{\text{b}}$	232.6 ± 0.96^{d}	193.6 ± 2.32^{a}
Cryptal Depth	463.3 ± 8.69^{b}	352.8 ± 11.97^{a}	$387.8\pm2.40^{\mathrm{a}}$	$453.2\pm13.01^{\text{b}}$	362.0 ± 17.69^{a}
Cryptal Width	$222.3\pm2.65^{\mathrm{b}}$	$200.1\pm5.82^{\rm a}$	$222.3\pm4.66^{\text{b}}$	234.3 ± 0.58^{b}	$192.3\pm3.01^{\mathrm{a}}$
Muscle	$193.4\pm12.02^{\mathrm{a}}$	173.2 ± 6.61^{a}	$285.2\pm14.48^{\rm c}$	347.3 ± 10.78^{d}	242.4 ± 16.34^{b}
Thickness					
Villus Height:	$4.2\pm0.05^{\rm a}$	$5.6\pm0.18^{\text{b}}$	5.8 ± 0.02^{b}	$4.8\pm0.15^{\rm a}$	$6.7\pm0.34^{\circ}$
Crypt Depth					
Ileum					
Villi Height	$1641.1 \pm 19.38^{\circ}$	1039.8 ± 41.14^{a}	1794.4 ± 3.80^d	1926.8 ± 31.68^{e}	1383.7 ± 8.26^{b}
Villi Width	187.3 ± 0.67^{b}	$208.9\pm4.24^{\rm c}$	192.2 ± 0.60^{bc}	223.1 ± 0.26^{d}	158.2 ± 12.01^{a}
Cryptal Depth	$268.0\pm5.59^{\mathrm{a}}$	$352.9\pm0.81^{\text{b}}$	$408.5 \pm 11.71^{\rm c}$	$544.7\pm3.11^{\text{d}}$	$269.8\pm5.53^{\mathrm{a}}$
Cryptal Width	182.1 ± 2.05^{b}	$215.3\pm4.92^{\rm c}$	$192.9\pm0.40^{\text{b}}$	$226.1 \pm 1.51^{\circ}$	$150.2\pm7.09^{\rm a}$
Muscle					
Thickness	$199.9\pm15.24^{\circ}$	129.0 ± 4.60^{a}	282.5 ± 8.96^{d}	338.3 ± 3.35^{e}	$158.8\pm1.95^{\text{b}}$
Villus Height:					
Crypt Depth	$6.1 \pm 0.18^{\text{e}}$	$2.9\pm0.12^{\rm a}$	$4.4\pm0.13^{\circ}$	3.5 ± 0.05^{b}	5.1 ± 0.10^{d}

^{a,b,c,d,e} Means along the same row with different superscript are significant different (p < 0.05).

The birds' digestive system and gut conditions are responsible for digestion and absorption. The length and weight of the small intestines differ between the different species of birds (Hassouna, 2001; Hamid et al., 2018; Wardhana et al., 2021). Thus, the digestive system tends to regulate itself depending on the physiological requirements of the bird. This is dependent on a number of factors including intestinal pH (Rahmani et al., 2005). The health of the chicken and the kind of nutrients consumed affect the pH level in the digestive system of the chicken (Rahmani et al., 2005). The pH level in particular parts of the GIT influences the growth of microbes. Additionally, pH level affects digestibility and absorption of nutrients (Fikri and Purnama, 2020). Yamauchi and Isshiki (1991) found that broiler chickens which are bred for rapid growth have a higher rate of small intestine

development. Longer intestines are assumed to digest feed efficiently and provide a greater surface area for nutrient absorption. The increase in small intestine weight allows broiler chickens to reach a heavier body weight faster compared to indigenous chickens (Jamroz, 2005; Kurniawati *et al.*, 2024). This implied that the administration of *P. nigrescens* leaf extracts up to 0.4 mL/bird would ensure better feed absorption thus, providing greater surface area for more efficient nutrient absorption at both the starter and finisher phases.

Similarly, another study by Mustafa *et al.* (2021) investigated the effect of Adding different levels of turmeric powder and curcumin in the diet on broiler performance, carcass traits, immunity and gut morphology of broiler chicken under normal and heat stress condition. They found that increasing the level of dietary turmeric

under normal condition increased the weight of the gizzard and decreased the weight of the small intestine, which is also consistent with the findings of this study.

Dietary pH for monogastric animals is usually reported to vary between 5.5 and 6.5 (Ao *et al.*, 2008), and changes as digesta transit different segments of the GIT. The range of crop pH observed in this study was 5.25 to 6.10 with the control having significant least value for crop pH. The range in this study was similar to the crop pH level of 6.084 of the broiler chickens reported by Mabelebele *et al.* (2014). These findings are in contrast with the work reported by Hinton *et al.* (2000) that the pH values or levels in the crops of broiler chickens were not acidic. The contrast could be as a result of variation in poultry species used for the study.

Digesta pH drops gradually as digesta reach the proventriculus or glandular stomach, where hydrochloric acid and pepsinogen are secreted and mixed with digesta through muscular movements in the gizzard (Svihus, 2014). Feed form may also play a role in the digesta pH in the stomach. Svihus (2011) reported that average values in stomach pH for broiler chickens ranged between 3 and 4 for normal pelleted diets.

The study investigated the effect of P. nigrescens leaf extracts on the histomorphometric parameters of the duodenum, jejunum, and ileum of Japanese quails. The examination of the small intestine's structure under a microscope is known as intestinal morphology. Some of the microscopic structure includes villus height and crypt depth. The development, health, and functionality of the gut are primarily indicated by the villus height and crypt depth, which affect nutrient digestion and absorption (Biasato et al., 2018; Kartikasari et al., 2019). The findings of the current study demonstrated that the gut's ability to expand and develop was improved when PNLE was administered at the highest doses (0.60 and 0.80 mL). By lowering the crypt depth and raising the villus height of the duodenum and jejunum, the present study's findings demonstrated that PNLE in the jejunum and ileum could enhance small intestine development.

The health and functional status of the chicken intestine are explained by the condition of the crypt depth. The rate at which the intestinal epithelium renews itself depends on their sizes (Zhang *et al.*, 2019; Prakatur *et al.*, 2019). According to Kaunitz and Akiba (2019), the crypts are where intestinal cell proliferation occurs most frequently. A larger, deeper crypt indicates higher food requirements for intestinal care and lower chicken productivity. A successful strategy to lower the expense of intestinal care for chickens is evidenced by the lower (shallow) crypt depth observed in this study for Japanese quails administered with PNLE.

The crypt is the villi factory, and a large crypt denotes a faster tissue turnover and a greater demand for new tissue, according to Xu et al. (2003) and Prakatur et al. (2019). Once more, the decreased crypt depth observed in this study, particularly at higher levels of PNLE administration (0.60 and 0.80 mL), may be an indication of an efficient tissue turnover, a healthy gut, and increased chicken body weight. Additionally, some findings suggested that the intestinal mucosa's villi and microvilli may have an impact on the generation of digesting enzymes (Wu et al., 2013). The study found that higher PNLE administration increased the height of the villi and the height-to-crypt ratio of Japanese quails. The higher enzymatic bio-activities of the gastrointestinal contents were related to the better Improved mucosal morphology. intestinal morphology leads to greater disease resistance, reduced increased food absorption, gastrointestinal output, and improved overall performance (Ebrahimzadeh et al., 2018). The chickens in the control group, who did not receive PNLE in the duodenum and jejunum, had the lowest ratio of villi height to crypt depth. Higher villi are associated with the enterocytes' efficient absorption ability, while shorter villi reduce the surface area for nutrient absorption, claim Parsaie et al. (2007) and Ensari and Marsh (2018). However, the crypt is where the villi's epithelial cells originate, and a large crypt exhibits rapid enterocyte turnover and a greater demand for maintaining mucosal tissue (Ayoola et al., 2015). The rapid enterocyte production and the epithelial cell renewal rate impact the small intestinal mucosa's protein and energy requirements, reducing the animal's feed efficiency (Luo *et al.*, 2009; Wardhana *et al.*, 2019; Arike *et al.*, 2020). Reduced villi height and deepened crypts increased secretion in the GI tract, resulting in birds' poor growth performance (Parsaie *et al.*, 2007; Prakatur *et al.*, 2019).

Kim and Khan (2013) reported that goblet cells are the source of the mucus deposit in the small intestine, which allows for gastric secretion and creates a protective obstruction against physical and chemical harm from ingested feed, microorganisms, and pathogens (Hollingsworth and Swanson, 2004; Sobolewska et al., 2017). Goblet cells, on the other hand, are modified epithelial cells that secrete mucus on the mucous membrane surface of the small intestine e.g., epithelium layer and an underlying lamina propria of loose connective tissue. The mucous membrane secretes mucous, a viscous fluid that shields the body from viruses and contaminants and keeps tissues from drying up. Muscularis is a thin layer of smooth muscle that supports mucous and provides it to move and fold. It is responsible for the segmental contractions and peristaltic movement in the GIT. These muscles cause feed to move and mix together with the digestive enzyme down the GIT. In the present study, muscularis thickness for the duodenum, jejunum and ileum were affected by the administration of PNLE.

There are other studies on the effect of natural substances on the small intestines of animals that can provide valuable insights into the potential benefits of *P. nigrescens* leaf extracts. For instance, a study by Okanlawon *et al.* (2020) investigated the effect of turmeric powder on the histomorphometric parameters of the small intestines of broiler chickens. The study found that the administration of turmeric powder increased villi height, villi width, and muscle thickness while reducing cryptal depth and cryptal width, similar to the findings of the study *on P. nigrescens* leaf extracts.

CONCLUSION

The study demonstrated that *P. nigrescens* can be administered to Japanese quails during both the starter and finisher phases without impairing their performance or gastrointestinal response. For best output an inclusion of 0.4/bird of *P. nigrescens* into their water is required to improve performance and better nutrient utilization.

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AUTHORS' CONTRIBUTIONS

AOA, OEO and SIA: Management of experimental animals, data collection and data management. AOA: Conceptualization, design of the experiments, manuscript writing and data analysis. LCN, OEA, OAA and ROA: Visualization, manuscript review and final approval of manuscript.

COMPETING INTERESTS

The authors declare that they have no competing interests.

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