



NOVEL PHYTOGENICS' IMPACT ON WEANED PIGS' GROWTH PERFORMANCE, HEAMATOLOGY AND SERUM BIOCHEMICAL INDICATORS

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Abstract: This experiment was carried out to investigate a novel phytochemicals (PCCPR) an acronym for (peppermint, celery, coriander, parsley and rosemary leaf meal mixture) impact on weaned pigs' performance, haemato-biochemical indicators. A total of forty cross bred weaned male pigs (Large white × Landrace) with an initial body weight of 7.33 ± 0.38 weaned at 28 days of age were individually housed in an open sided pen were randomly divided into four treatment group of six pigs each. Basal diet were adequate in all nutrients (NRC, 2012). The animals were fed as follows; basal diet with antibiotics (neomycin – 1.5 g/kg) in treatment one, treatment two, three and four were fed basal diet supplemented with PCCPR at 5 g, 10 g and 15 g/kg respectively. The experiment lasted for two months and all necessary management practices were observed. Average daily weight gain and average daily feed intake were similar in diet three and four compared to the other groups ($P < 0.05$). Dietary supplementation of PCCPR resulted in a numerical increase in pack cell volume, haemoglobin and red blood cell, white blood cell and its differentials (monocytes, eosinophils, basophils and leucocytes) and a remarkable improvement in mean corpuscular volume mean corpuscular haemoglobin and mean corpuscular haemoglobin concentrations. All the serum biochemical parameters were significantly affected by the treatments ($P < 0.05$). However, all values were within the normal physiological range for healthy pigs. It was concluded that the use of PCCPR up to 15 g/kg could boost swine productivity at weaned stage without causing any negative impact on the health status of animals.

Keywords: Phytochemicals, Haematology, Food safety, Swine, Management

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1. Introduction

Interest in pig nutrition and the widespread use of antibiotic-free feeding systems has been sparked by public outrage over possible risks associated with antibiotic resistance to the health of humans. As a result, feed additives that can be employed as in-feed antibiotic substitutes in pig feeding regimens have been developed (Caroline, 2019). Due to their wide variety of efficacies and their impacts on sustainability and safety, phytochemicals are one of the most important substitutes for antibiotics because they have an exciting potential in animal feeding (Jan et al., 2016). Phytochemical feed additions include a variety of plants, including herbs, spices, and essential oils obtained from plants (Luis, 2012; Singh et al., 2022).

Due to the substantial amount of phytochemicals in them, they are well known for having good benefits on animals in the form of flavoring, antioxidant, anti-inflammatory, antiviral, antifungal, and antibacterial qualities (Manu, 2006; Alagbe et al., 2022a). The variances in efficiency between phytochemical feed additives are explained by the chemical makeup of these additives, which highlights some variations due to their ingredients and other

influencing factors like climate, location, harvest stage, and storage conditions (Jan et al., 2016). Additionally, several natural plant bioactives can have a growth-promoting effect by favorably affecting the morphology and physiology of the gastrointestinal tract and most likely by either stimulating or inhibiting a specific metabolic pathway (Sandra, 2020).

The effects of plant chemicals (phytochemicals) directly on gut tissues and digestive enzyme secretions or indirectly through stimulatory or inhibitory effects on microbial populations, immune modulation, anti-inflammatory and antioxidant effects have been observed may affect nutrient digestion and absorption (Chris, 2010). The release of brush border enzymes and pancreatic enzymes can both be favourably influenced by mixtures of various phytochemical feed additives (Manu, 2018). Trypsin, an enzyme involved in breaking down proteins, increased by 13% when phytochemicals (spices and herbs) were added to the diet of monogastric animals in vivo. Amylase, which breaks down starch, sucrase, and maltase, which breaks down the disaccharide maltose, also showed increases in their respective carbohydrate-digesting capacities (Manu, 2006).



With all of these advantages of phyto-genic feed additives eliminate the risks that conventional antibiotics offer to animal health (antimicrobial resistance), and harmful residues in animal products. To avoid toxicity and improve food safety, it is necessary to investigate the usage of additional medicinal plants with pharmacological qualities and perhaps conduct study on their tolerable levels. This experiment was designed to examine a novel phyto-genics' impact on weaned pigs' growth performance, heamatology and serum biochemical indicators.

2. Materials and methods

2.1. Location of the Test

The research was conducted at livestock department of Sumitra Research Institute in Gujarat, India, which is situated at 23° 13' N and 72° 41' E and has a 1600 km coastline (Bose Ashish, 1991). The study was carried out between January to March, 2022.

2.2. Gathering and Preparation of Phyto-genic Feed Additive

Leaves of Rosemary (*Rosmarius officinalis*), peppermint (*Mentha piperita*), celery (*Apium graveolens*), coriander (*Coriandrum sativum*) and parsley (*Petroselinum crispum*) were harvested from Sumitra Research Institute gardens in Gujarat. The leaves were identified and authenticated by a certified taxonomist, washed with running tap water separately and the placed in different plastic sieve for the water in each leaf to drain for 10 minutes. Afterwards, they were shade dried separately under room temperature for 16 days until each leaf attain a constant weight. Each leaf is powdered using an electric blender (Kenwood 500 Watts commercial high speed blender, Japan) and stored in a labeled zip lock bags stored in the refrigerator (Haier Thermocool, Model: HRF-95EX, Nigeria) at a temperature of 4 degrees Celsius. 200 grams of each powdered leaf sample was transferred into another electric blender and agitated for 10 minutes to allow a homogenous mixture to form a new phyto-genic feed additive (PCCPR) an abbreviation for peppermint, celery, coriander, parsley and rosemary leaf. This was thereafter taken to the laboratory for additional testing. The following standard laboratory techniques were used to determine the phyto-constituents in PCCPR.

2.3. Chemicals and Reagents for Analysis

Aluminium chloride, Sodium hydroxide, Sodium carbonate solution, concentrated sulfuric acid, Potassium hexacyanoferrate solution, acetic acid solution, ethanol, vanillin solution, sodium nitrate.

2.4. Equipment Used During Analysis

YSTE – UV5600 UV/VIS Spectrophotometer which uses imported flanged deuterium lamp with low stray light and a rigid die-cast aluminum base as its optical mount. It is capable of setting up various standard curves. It has the following technical specifications; wave length range (190 – 1100 nm), band width (2nm), wavelength accuracy (± 0.8 nm), wavelength repeatability (0.3nm),

wavelength setting: auto, photometric accuracy (± 0.3 % T), photometric repeatability (± 0.02 A/h), stray light (≥ 0.05 % T), detector (silicon photodiode).

2.5. Estimation of Flavonoids

Using catechin as a reference, the total flavonoid content was calculated using the aluminium chloride technique. After adding 0.1 mL of aluminum chloride and 0.2 mL of 5% sodium nitrite, 0.5 grams of PCCPR was added. 2 mL of 1 M sodium hydroxide was added to the reaction mixture after the mixture had been incubated for 6 minutes at room temperature. Immediately, 10 mL of distilled water were added to the final volume. Using a spectrophotometer, the reaction mixture's absorbance at 410 nm was evaluated in comparison to a blank.

2.6. Total Phenolic Content

The Folin-reagent Ciocalteu's was used to determine the total phenolic contents in PCCPR. In the method, 0.5 grams of PCCPR and 0.4 mL of 1:10 v/v diluted FCR were combined. 4 mL of sodium carbonate solution was added after 5 minutes. The tubes were filled to their final content with 10 mL of distilled water and left to stand at room temperature for 90 minutes. A spectrophotometer was used to test the sample's absorbance at 850 nm in comparison to the blank. The phenolic content of the oil was expressed as milligrams of catechol per dry gram of dry weight, and a calibration curve was created using catechol solution as the standard with the standard graph given by and the total phenolic content of the sample was reported as milligrams of catechol per dry gram of dry weight (Otlés and Yalcin, 2012).

2.7. Estimation of Saponins

Using a vanillin and concentrated sulfuric acid colorimetric technique, saponin was quantified. 0.4 milliliters of 77% sulfuric acid, 0.5 milliliters of freshly made vanillin solution, and 0.2 milliliters of PCCPR were combined. The mixture was allowed to cool to room temperature before being heated in a water bath for 15 minutes at 60 degrees centigrade. A spectrophotometer was used to detect the absorbance at 545 nm.

2.8. Estimation of Total Steroid Content

In accordance with reports by Madhu *et al.* (2016), 0.5 grams of PCCPR was added to a 10 mL volumetric flask. Potassium hexacyanoferrate solution (0.5% w/v, 0.5% w/v, 2 mL) was added after that. Prior to getting diluted with distilled water to the right concentration, the substance was heated for 20 minutes at 40-50 degrees centigrade in a steam bath with regular shaking. The absorbance was calculated at 380 nm and compared to a reagent blank.

2.8. Alkaloids Complex

Using the gravimetric technique, the alkaloids content of PCCPR was determined (Adeniyi *et al.*, 2009). Alkaloids were precipitated by mixing 20 mL of acetic acid solution in ethanol (10% w/v) with 0.5 grams of PCCPR and placing the mixture on a water bath. The alkaloids were then precipitated by adding drops of extremely concentrated ammonium hydroxide. After the precipitate reached a constant weight, it was transferred to

desiccators and reweighed.

2.9. Total Tannins Estimation

The Folin-Ciocalteu technique was employed to determine the total tannin concentration (Biswas et al., 2020). 0.5 mL of metaphosphoric acid, 1.5 mL of 90 % ethanol, 1.5 mL of 1.0 mol/mL Na₂ CO₃, and 1.5 mL Folin-Ciocalteu were added to 1.0 grams of PCCPR to dilute it (100 mL). The combination was thoroughly blended and then let to cool for 20 minutes at room temperature. Then, using a spectrophotometer, the absorbance of the standard curve and PCCPR were compared to a blank at 880 nm.

2.10. Mineral Composition of PCCPR (novel phytochemicals)

Composition of calcium, phosphorus, potassium, magnesium, manganese, zinc, iron, sodium, copper, chromium, nickel and boron were analyzed using trace series atomic absorption spectrometer (Model HD-A11200P, USA). The equipment has the following specifications; wave length (185 – 900nm), wavelength scan series (300 nm/min), grating (1800 lines/min) with automatic gas control with auto ignition, optimization and change over, heating (transversely heated graphite tube up to 3800 K/s heating rate, sensitivity (2 mg/L Cu: Abs ≥ 0.4, RSD < 0.5 %), standard universal auto-sampler for F/GF and VG random access and 8-lamp 2D motorized array with automatic lamp selection, positioning and alignment.

2.11. Experimental Design and Livestock Management

A total of forty cross bred weaned male pigs (Large white × Landrace) with an initial body weight of 7.33 ± 0.38 weaned at 28 days of age were procured from a reputable farm in Gujarat, India and were individually housed in an open-sided pens with standard dimensions (2.0 × 1.5 × 1.2 m). Animals were transported to the experimental site in the early hours of the morning and given a mixture of water and glucose (10 grams of glucose to 5 liters of water) on arrival to the teaching and research farm. Before the start of the experiment, pigs were placed on two weeks acclimatization period where they were prophylactically treated with antibiotics (Oxytrox L.A) administered subcutaneously on each animal according to the manufacturers recommendation and ivermectin against external and internal parasites (0.1 mL per kilogram of animal). After the adjustment period, the pigs were balanced for their weight to ensure that the initial weight of each group were similar and randomly divided into four treatment group of ten pigs each with one animal per replicate in a completely randomized design. The experiment lasted for 2 months and all necessary management practices were observed according to the ethical guidelines outlined by the ethical guidelines for the use of animal in research.

2.12. Experimental Diet and Design

Basal diet was adequate in all nutrients according to National Research Council (2012) (Table 1). Weaners in treatment one was given basal diet with antibiotics

growth promoter (neomycin at 1.5 g/kilogram), and those in treatment two through four were fed basal diet supplemented with 10 grams, 20 grams, and 30 grams of PCCPR respectively.

Table 1. Experimental diets’ total composition

Components	Amount (kilogram)
Yellow maize	40.00
Dried cassava peel	4.20
Wheat offal	6.50
Palm kernel cake	21.00
Soya meal	12.00
Groundnut cake	10.00
Fish meal (Imported)	1.00
Bone meal	3.00
Limestone	1.50
Lysine	0.15
Methionine	0.15
*Mineral/Vitamin Premix	0.25
Salt	0.30
Total	100.05
Determined analysis (%)	
Crude protein	19.19
Crude fibre	5.97
Ether extract	3.88
Calcium	1.04
Phosphorus	0.63
Lysine	1.00
Methionine +Cysteine	0.79
Metabolizable energy (kcal/kg)	2865.1

*Mineral/Vitamin premix supplied per kg diet: - vit A, 9,000 I.U; vit E, 8.91 mg; vit D3, 2500I.U, vit K, 3.2mg; vit B2, 5.0mg; Niacin, 40 mg; vit B12, 25 mg; choline chloride, 100 mg; Mn, 5.0 mg; Zn, 35.1mg; Cu, 2.0g; folic acid, 2.5mg; Fe, 5.8g; pantothenic acid, 10mg; biotin, 30.5g; antioxidant, 56mg (starter’s mash)

2.13. Performance Characteristics

The amount of feed ingested was determined by subtracting the feed refused from the feed supplied. The feed conversion ratio, or the quantity of feed necessary to produce one unit of gain, was calculated as the ratio of average feed intake to average body weight growth. The weight gain was calculated using the difference between the initial and final body weights. The average daily body weight was determined by dividing the amount of weight gained for each treatment by the number of trial days.

2.14. Measurement of Blood Sample

Six pigs per treatment were chosen for hemato-biochemical evaluation on the eighth weekday of the study’s duration. 4 ml of blood was drawn from the sampled birds using 20 to 100 sterilized metallic needles to take blood from a culinary vein. Two mL were put into vials that had been treated with ethylene diamine tetra acetate for hematological testing, and the other two mL were utilized for serum testing. Using an automated Sysmex analyzer (model XN-3100, China), hamatological variables were measured. For the serum indices, the

remaining 2 mL of blood were drawn and placed in bottles without anticoagulant. Technical details of the apparatus include optical flow (20 L quartz), reaction volume (300–800 L), and photometric range (– 0.10–3.00 abs), and filters (6 interference filters: 380–425–516–508, 600–700 nm).

2.15. Statistical Analysis

Using Statistical Package of Social Sciences (SPSS version 23.0), one way analysis of variance (ANOVA) was used to examine all of the data. Using Duncan’s multiple range test of the same package, means were sorted.

3. Results

3.1. Mineral Composition of PCCPR (Novel phytogetic mixture)

Phyto-constituents of PCCPR (novel phytogetic mixture) reveals the major compounds in the sample. Phenols had the highest concentration (2107.18 mg/g) closely followed by flavonoids (1093.86 mg/gram), alkaloids (485.91 mg/gram), tannins (282.10 mg/gram), saponins (112.75 mg/gram), steroids (100.14 mg/gram) and oxalates (12.30 mg/gram) correspondingly (Table 2).[^]

Table 2. Phyto-constituents of PCCPR (Novel phytogetic mixture)

Constituents	Concentration (milligram /gram)
Flavonoids	1093.86
Alkaloids	485.91
Tannins	282.10
Steroids	100.14
Phenols	2107.18
Saponins	112.75
Oxalates	12.30

Mineral composition of PCCPR (novel phytogetic mixture) (Table 3). Calcium (1712.81 milligram /100 gram), phosphorus (973.68 milligram /100 gram), potassium (877.82 milligram /100 gram), magnesium

(509.57 milligram /100 gram), manganese (100.40 milligram /100 gram), zinc (209.38 milligram /100 gram), iron (95.17 mg/100g), sodium (315.60 mg/100g), selenium (80.05 mg/100g), chromium (0.03 milligram /100 gram) and nickel (0.41 milligram /100 gram). Calcium had the highest concentration while chromium had the least value.

3.2. Effects of Supplemented Diet with PCCPR (Novel Phytogetic Mixture) on Growth Performance of Weaned Pigs

Effects of supplemented diet with PCCPR (Novel phytogetic mixture) on growth performance of weaned pigs (Table 4). Initial body weight varies from 7.30 – 7.34 kg, final body weight (19.18 – 25.17 kg), weight gain (11.85 – 17.87 kg), average daily weight gain (0.20 – 0.30 kg), total feed intake (37.14 – 42.05 kg), average daily feed intake (0.60 – 0.75 kg) and feed conversion ratio (2.34 – 3.30). There was effect of treatments on in weight gain, average daily feed intake and feed conversion ratio (P<0.05).

Table 3. Mineral composition of PCCPR (Novel phytogetic mixture)

Constituents	Concentrations (milligram /100 gram)
Calcium (Ca)	1712.81
Phosphorus (P)	973.68
Potassium (K)	877.82
Magnesium (Mg)	509.57
Manganese (Mn)	100.40
Zinc (Zn)	209.38
Iron (Fe)	95.17
Sodium (Na)	315.60
Copper (Cu)	87.10
Selenium (Se)	80.05
Chromium (Cr)	0.03
Nickel (Ni)	0.41

Table 4. Effects of supplemented diet with PCCPR (Novel phytogetic mixture) on growth performance of weaned pigs

Parameters	Diet 1	Diet 2	Diet 3	Diet 4	SEM
Initial body weight (kg /pig)	7.33	7.32	7.34	7.30	0.21
Final body weight (kg /pig)	19.18 ^c	21.40 ^b	25.00 ^a	25.17 ^a	0.89
Weight gain (kg /pig)	11.85 ^c	14.08 ^b	17.66 ^a	17.87 ^a	0.75
Av. daily weight gain (kg/pig)	0.20 ^c	0.25 ^b	0.32 ^a	0.32 ^a	0.02
Total feed intake (kg/pig)	37.14 ^b	40.80 ^a	41.72 ^a	42.05 ^a	1.98
Av. daily feed intake (kg/pig)	0.66 ^b	0.73 ^a	0.74 ^a	0.75 ^a	0.01
Feed conversion ratio	3.30 ^a	2.92 ^b	2.31 ^b	2.34 ^b	0.02
Mortality (%)	-	-	-	-	-

Average in rows having various characters vary markedly (p< 0.05); Initial body weight, Final body Weight gain, Average daily weight gain, Average daily feed intake; diet 1: basal diet + 1.5 g/kg Neomycin; PCCPR was added to basal diet in the following: diet 2: 5

g/kg; diet 3: 10 g/kg ; diet 4: 15 g/kg respectively. SEM: standard error of mean.

3.3. Effects of Supplemented Diet with PCCPR (Novel Phytogenic Mixture) on Hematological Indices of Weaned Pigs

Effects of supplemented diet with PCCPR (Novel phytogenic mixture) on hematological indices of weaned pigs (Table 5). Pack cell volume values varies from (29.93 – 34.15 percent), haemoglobin (10.71 to 13.92 gram/dL), red blood cell [4.72 to 8.86 (x10⁶)], mean corpuscular volume (50.80 to 68.01 fl), mean corpuscular haemoglobin (18.50 to 21.00 pg), mean corpuscular haemoglobin concentration (29.81 to 34.02 g/dL), white blood cell [7.64 to 12.02 (x10³)], monocytes (3.00 to 6.10

percent), lymphocytes (47.80 to 61.27 percent), basophils (1.02 to 1.16 percent) and eosinophil's (0.68 to 0.81 percent). Pack cell volume, red blood cell, haemoglobin, mean corpuscular volume, mean corpuscular haemoglobin and mean corpuscular haemoglobin concentration values follow similar pattern as the parameters were highest in diet two through four and lowest in diet one (P<0.05). White blood cell, monocytes and lymphocytes values were maximum at diet three and four relative to other treatments (P<0.05). Basophils and eosinophils values were not affected by the treatments (P>0.05).

Table 5. Effects of supplemented diet with PCCPR (Novel phytogenic mixture) on hematological indices of weaned pigs

Parameters	Diet 1	Diet 2	Diet 3	Diet 4	SEM
Pack cell volume (%)	29.93 ^b	33.21 ^a	34.03 ^a	34.15 ^a	0.11
Haemoglobin (g/dL)	10.71 ^b	12.19 ^a	13.80 ^a	13.92 ^a	0.06
Red blood cell (x10 ⁶)	4.72 ^c	8.10 ^a	8.51 ^a	8.86 ^a	0.02
Mean corpuscular volume (fl)	50.80 ^b	66.00 ^a	67.12 ^a	68.01 ^a	0.27
Mean corpuscular heamoglobin (pg)	18.50 ^b	20.80 ^a	20.40 ^a	21.00 ^a	0.09
Mean corpuscular heamoglobin conc. (g/dL)	29.81 ^b	32.75 ^a	33.64 ^a	34.02 ^a	0.15
White blood cell (x10 ³)	7.64 ^c	9.96 ^b	12.40 ^a	12.02 ^a	0.04
Monocytes (%)	3.00 ^b	3.08 ^b	5.74 ^a	6.10 ^a	0.02
Lymphocytes (%)	47.80 ^c	50.05 ^b	60.10 ^a	61.27 ^a	0.35
Basophils (%)	1.20	1.02	1.09	1.16	0.03
Eosinophils (%)	0.74	0.68	0.73	0.81	0.02

Values in cells with various characters vary markedly (P<0.05); Diet 1: basal diet + 1.5 g/kg Neomycin; PCCPR was added to basal diet in the following: diet 2: 5 g/kg; diet 3: 10 g/kg; diet 4: 15 g/kg respectively. SEM: standard error of mean

3.4. Effects of Supplemented Diet with PCCPR (novel phytogenic mixture) on Serum Biochemical Indices of Weaned Pigs

Table 6 revealed the effects of supplemented diet with PCCPR (novel phytogenic mixture) on serum biochemical indices of weaned pigs. Total protein values varied from (57.18 to 73.25 g/L), globulin (25.10 to 38.01 g/L), albumin (32.08 to 34.24 g/L), urea (3.71 to 5.06 mmol/L), creatinine (68.51 to 73.28 μmol/L), cholesterol

(3.06 to 4.02 mmol/L), glucose (2.08 to 3.92 mmol/L), alanine transaminase (30.15 to 45.02 U/L), alanine serum transaminase (42.06 to 61.74 U/L), alanine phosphatase (112.1 to 155.9 U/L), calcium (2.02 to 3.40 mmol/L), phosphorus (1.80 to 2.17 mmol/L), sodium and chloride [98.10 to 120.9 mmol/L; 93.82 to 105.1 mmol/L] respectively. All the values were significantly (P<0.05) affected by the treatments.

Table 6. Effects of supplemented diet with PCCPR (Novel phytogenic mixture) on serum biochemical indices of weaned pigs

Parameters	Diet 1	Diet 2	Diet 3	Diet 4	SEM
Total protein (g/L)	57.18 ^c	68.04 ^b	70.16 ^a	73.25 ^a	0.56
Globulin (g/L)	25.10 ^b	36.00 ^a	37.12 ^a	38.01 ^a	0.19
Albumin (g/L)	32.08 ^b	33.04 ^b	33.04 ^b	34.24 ^a	0.15
Urea (mmol/L)	3.71 ^c	4.92 ^b	5.00 ^a	5.06 ^a	0.02
Creatinine (μmol/L)	68.51 ^b	70.83 ^a	72.19 ^a	73.28 ^a	0.55
Cholesterol (mmol/L)	4.02 ^a	4.00 ^a	3.00 ^b	3.06 ^b	0.01
Glucose (mmol/L)	3.92 ^a	3.83 ^a	3.76 ^a	2.08 ^b	0.02
Alanine transaminase (U/L)	30.15 ^b	40.08 ^a	43.89 ^a	45.02 ^a	0.19
Alanine serum transaminase (U/L)	42.06 ^c	43.72 ^c	50.70 ^b	61.74 ^a	0.42
Alanine phosphatase (U/L)	112.1 ^c	143.8 ^b	150.6 ^a	155.9 ^a	1.37
Calcium (mmol/L)	2.02 ^b	2.48 ^b	3.39 ^a	3.40 ^a	0.01
Phosphorus (mmol/L)	1.80 ^b	1.96 ^b	2.00 ^a	2.17 ^a	0.01
Sodium (mmol/L)	98.10 ^b	116.8 ^a	120.2 ^a	120.9 ^a	0.38
Chloride (mmol/L)	93.82 ^b	100.2 ^a	103.7 ^a	105.1 ^a	0.32

Values in cells with various characters vary markedly (P<0.05); Diet 1: basal diet + 1.5 g/kg Neomycin; PCCPR was added to basal diet in the following: diet 2: 5 g/kg; diet 3: 10 g/kg; diet 4: 15 g/kg respectively. SEM: standard error of mean.

4. Discussion

The presence of multiple phyto-constituents or phytochemicals in PCCPR gives the sample a higher affinity to function as; antioxidant, anti-fungal, antiviral, anti-helminthic, hepato-protective, immune-modulatory and immune-stimulatory amongst others (Uphadhyay et al., 2010; Igbal et al., 2015). The study's increased flavonoid concentration lends support to PCCPR's anti-inflammatory, immune-stimulating, and antioxidant capabilities. According to Mbaebe et al. (2012), flavonoids exhibits several pharmacological effects and are capable to scavenging free radicals in the body, thus preventing infections. The flavonoid content recorded in this experiment was higher than those recorded for *Hypochaeris radicata* leaves (14.31 mg/g), *Rumex crispus* leaves (130.4 mg/g) and *Mesua ferrea* leaves (140.82 mg/g) by Narender et al. (2012); Jamuna et al. (2012). According to Jamuna et al. (2012), tannins are crucial for both the treatment of inflamed tissues and the prevention of cancer. For example, alkaloids act as agents with analgesics and anti-plasmodic activities whereas saponins operate as antifungal agents (Govindappa et al., 2011). Both alkaloids and saponins are recognized to have ethno-pharmacological benefits (Govindappa et al., 2011).

Minerals are inorganic elements required in animals' feed for adequate development and functioning. Calcium, magnesium and phosphorus values recorded in PCCPR were higher than values reported for *Terminalia sericea* leaf (759.20, 560.70 and 102.1 mg/100g) respectively by Chivandi et al. (2013). Mineral composition of medicinal plants are influenced by age of plant or stage of growth, climatic condition and cultivar within plant species (Oluwafemi et al., 2020). However, PCCPR values recorded were within the NRC (2012) daily requirement for weaned pigs. Potassium are responsible for proper fluid balance, nerve contraction, heart beat regulation and membrane potential (Arinola et al., 2008). Sodium are required for proper fluid and pH balance, nerve transmission and contraction of the muscles (NHWC, 2002). Strong bones, blood pressure regulation and strengthening of the immune system are the role of calcium (Afisu et al., 2016). Copper are needed for the creation of haemoglobin, regulating neuro-transmitters and clean up free radicals (Abu et al., 2016). Zinc is immune boosters, healing of wounds, needed for making protein, blood clotting and genetic materials (NHWC, 2002). Iron is part of haemoglobin which carries oxygen in the body needed for energy metabolism and part of protein (a protein in muscle cells) (Arinola et al., 2008). Selenium and manganese are responsible for the functioning of antioxidant enzymes and metabolism of amino acid, cholesterol and carbohydrates (Alagbe et al., 2023a, Alagbe et al., 2023b). Nickel is responsible for the absorption of iron and other elements in the body (NHWC, 2002).

The results on performance indicated some beneficial effects of the experimental diets on the growth

performance of weaned pigs which is reflected in a numerically increased average daily weight gain among animals in diet two (5 g/kg PCCPR), three (10 g/kg PCCPR) and four (15 g/kg PCCPR) and a significant improvement in average daily feed intake and feed conversion ratio among the treatments respectively. The experiment clearly shows that PCCPR has the potential to enhance the growth of animals due its phyto-genic constituents. For instance, parsley, peppermint, rosemary, coriander and celery leaves which are the major components of the novel phyto-genic material has apiol, menthol, cineole, linalool and borneol as well as phtalides as active compounds with medicinal properties such as; Appetite and digestive stimulant, antiseptic, antioxidant, anti-diarrhoeal, anti-inflammatory amongst others (Luis, 2012). A synergy in these compounds could inhibit the development of pathogenic bacteria, support the proliferation of *lactobacilli spp* as well as improving the secretion of digestive enzymes which in turn positively influence the growth patterns and feed conversion ratio (Singh et al., 2022). Phyto-genics have also been reported to support normal liver metabolism and protection of kidney cells against osmotic fluctuations to ensure an outstanding performance in animals (Yang et al., 2015). It can also improve the flavor and palatability of feed as well as its retention time (Singh et al., 2022). The outcome of this research confirms the earlier results by Zhang et al. (2012) when phyto-genic consisting of turmeric and clove was fed to weaned pigs at 20 g/kg. Yan et al. (2011) also recorded a positive outcome when *Houttuynia cordata* and *Taraxacum officinale* extract powder was fed to pigs at 15 g/kg.

In situation of nutrient insufficiency and health situation, heamatological data could be used as a point of reference for analysis in animals (Etim et al., 2014). Results of the experiment revealed that the dietary supplementation of PCCPR lead to a numerical elevation in pack cell volume, red blood cell, haemoglobin concentration and a remarkable increase in white blood cell, lymphocytes and monocyte values. Effective distribution of oxygen supply and nutrient round the body will be more prominent among pigs fed diet two through four relative to diet one. Pack cell volume are used for the detection of presence or absence of anemia or polycythemia (Oyawoye and Ogunkunle, 2004). White blood cell and lymphocytes were maximum in diet three and four compared to the other treatments. The outcome helps to strengthen the immune system in these groups. Phyto-genic mixture has been reported to exhibit immune-modulatory properties and immune-globulin secretion, thus preventing mortality among animals (Zhou et al., 2013; Olafadehan et al., 2020). Eosinophils and basophils are strong defense mechanisms against parasites and diseases, however, values were not affected by the treatments and are within the normal range specified.

The results on the serum biochemical indices agree that the dietary addition of PCCPR resulted in a significant

increase in total protein (albumin plus globulin). According to Olafadehan et al. (2020), nutrition can greatly influence the total serum protein. Experimental diets were adequate in all nutrients, however, dietary supplementation of PCCPR could stimulate the activities of bile and digestive enzymes (Agubosi et al, 2022; Muritala et al., 2022). This is reflected among pigs fed diet three (10 g/kg PCCPR) and four (15 g/kg PCCPR) compared to the other treatments ($P < 0.05$). Urea and creatinine values recorded are within the normal physiological range for pigs. Elevated values are clear sign of renal failure which can lead to death at extreme cases (Alagbe et al., 2022b). Elevated glucose could be triggered during period of stress from starvation or malnutrition, insufficient clean water as well as poor handling during farm operations (Olafadehan et al., 2020). It is important to note that pigs fed on diet three and four had lower values. This could as a result of the active compound in PCCPR especially those with antioxidant properties. Values for cholesterol rose as PCCPR levels improved across the group, this demonstrates unequivocally that PCCPR hypocholesteromic characteristics. As the concentration of PCCPR increased among the group, serum enzyme levels dropped. The existence of harmful substance in the blood causes abnormal readings to be activated (Rafiu et al., 2013). Electrolytes are capable of activating enzymatic activities, helps the cells in the uptake of nutrients and release hormones as well as acting as buffer in the blood (Etim et al., 2013).

5. Conclusion

At the end of the experiment, it was concluded that the dietary inclusion of PCCPR (novel phytochemicals) in feed optimizes the performance by competing with harmful gut flora and by stimulating the immune system of the animal and therefore, increasing its resistance to infectious agents. PCCPR may positively impact the secretion of digestive juices and nutrient absorption when included up to 15 g/kg without causing any deleterious effect on the health of the animal.

Author Contributions

The percentage of the author contributions is presented below. The author reviewed and approved the final version of the manuscript.

	A.O.J.
C	100
D	100
S	100
DCP	100
DAI	100
L	100
W	100
CR	100
SR	100
PM	100
FA	100

C=Concept, D= design, S= supervision, DCP= data collection and/or processing, DAI= data analysis and/or interpretation, L= literature search, W= writing, CR= critical review, SR= submission and revision, PM= project management, FA= funding acquisition.

Conflict of Interest

The author declared that there is no conflict of interest.

Ethical Consideration

Permissions were obtained from the Sumitra Research Institute Ethics Committee (protocol code: 2010/63/EU and date: March 01, 2022).

References

- Abu NE, Ezeomeke S, Azegba P, Davidson G. 2016. Phytochemical nutritional and anti-nutritional properties of leaves stems bark and roots of trees used in popular medicine for the treatment of malaria in South Eastern Nigeria. *J Medicinal Plants Research*, 10(38): 662-668.
- Adeniyi SA Orjiekwe CL, Ehiagbonare JE. 2009. Determination of alkaloids and oxalates in some selected food samples in Nigeria. *African J Biotechnol*, 8(1): 110-112.
- Afisu B, Kehinde OS, Funsho O. 2016. Comparative proximate minerals composition and antinutritional factors of *Walteria indica* leave root and stem. *Annals Food Sci Technol*, 17(2): 478-484.
- Agubosi OCP, Alexander J, Alagbe JO. 2022. Influence of dietary inclusion of Sunflower (*Helianthus annuus*) oil on growth performance and oxidative status of broiler chicks. *Central Asian J Med Nat Sci*, 2(7): 187-195.
- Agubosi OCP, Imudia Fand Alagbe JO. 2022. Evaluation of the nutritional value of air dried and sun-dried sweet potato (*Ipomoea batatas*) peels. *European J Life Safe Stab*, 14(22): 43-51.
- Alagbe JO Ramalan SM, Shittu MD, Olagoke OC. 2022a. Effect of *Trichilia monadelpha* stem bark extract on the fatty acid composition of rabbit's thigh meat. *J Environl Issues Clim Chan*, 1(1): 63-71.
- Alagbe JO, Shittu MD, Tanimomo Babatunde K. 2022b. Influence of *Anogeissusleio carpus* stem bark on the fatty acid composition in meat of broiler chickens. *European J Life Safety Stab*, 14(22): 13-22.
- Alagbe OJ, Oluchi CA, Rufus AO, Taiwo AO, Adegoke EA, Emiola IA. 2023b. Haemato-biochemical indices and intestinal

- microbial population of broiler chickens fed diet supplemented with *Prosopis africana* (African mesquite) essential oil. *Brazilian J Sci* 2(9): 98-110.
- Alagbe OJ, Oluchi CP, Rufus AO. 2023a. Histopathology of broiler chickens fed diet supplemented with *Prosopis africana* (African mesquite) essential oil. *Brazilian J Sci*, 2(9): 49-59.
- Arinola OG, Olaniyi JA, Abibinu MO. 2008. Elemental trace elements and metal binding proteins in Nigerian consumers of alcoholic beverages. *Pakistan J Nutri*, 7(6):766-769.
- Bose A. 1991. Demographic diversity of India census state and district level data. BR Publication, URL: <http://www.popline.org/node/317128> (accessed date: March 17, 2022).
- Caroline CV. 2019. Improving feed efficiency and gut integrity in *Escherichia coli* challenged weaned pigs. *Inter Pig Magaz*, 1(4): 8-10
- Chivandi E, Davidson BC, Erlwanger KH. 2013. Proximate mineral fibre phytate-phosphate vitamin E amino acid and fatty acid composition of *Terminalia sericea*. *South Africa J Botan*, 88: 96-100.
- Chris C. 2010. Gut flora and immune modulation by plant extracts. *Inter Poultry Magaz*, 7(2): 1-2.
- Etim NN, Enyenihi GE, Akpabio U, Offiong EEA. 2014a. Effects of nutrition on haematology of rabbits: A Review. *European Sci J*, 10(3): 413-424.
- Etim NN, Williams ME, Enyenihi GE, Udo MD, Offiong EEA. 2013. Haematological parameters: indicators of the physiological status of farm animals. *British J Sci* 10(1): 33-45.
- Govindappa M, Bharath N, Shruthi HB, Sadananda TS, Sharanappa P. 2011. Antimicrobial antioxidant and in vitro anti-inflammatory activity and phytochemical screening of *Crotalaria pallida* Aiton. *African J Pharma Pharmacol*, 5(21): 2359-2371.
- Jamuna S, Paulsamy S, Karthika K. 2012. Screening of in vitro antioxidant activity of methanolic leaf and root extracts of *Hypochoeris radicata* L(Asteraceae). *J Applied Pharmaceutical Sci*, 2(7): 149-154.
- Jan D, Van K, Ester V. 2016. Phytochemicals: be one step ahead with plant derived feed additives. *Inter Poultry Magazine* 5(1): 3-5.
- Luis MM. 2012. Essential oils and their digestive properties in pigs. *Inter Pig Magaz*, 6(2): 4-6.
- Madhu M, Sailaja V, Satyadev TN. 2016. Quantitative phytochemical analysis of selected medicinal plant species by using various organic solvents. *J Pharmac Phytochem*, 5(2): 25-29
- Manu DL. 2006. Support gut health with a unique combination of fatty acid and phytochemicals. *Inter Poultry Magaz*, 7(8): 6-8
- Manu DL. 2018. Support gut health with unique combination of fatty acids and phytochemicals. *Inter Poultry Magaz*, 1(3): 4-6
- Mbaebe BO Edeoga HO Afolayan AJ. 2012. Phytochemical analysis and antioxidant activities of aqueous stem bark extract of *Schotia latifolia* Jacq. *Asian Pacific J Tropical Biomed*, 2(2): 118- 12.
- Muritala DS, Alagbe JO, Ojebiyi OO, Ojediran TK, Rafiu TA. 2022. Growth performance and haematological and serum biochemical parameters of broiler chickens given varied concentrations of *Polyalthia longifolia* leaf extract in place of conventional antibiotics. *Anim Sci Genet*, 18(2): 57-71.
- Narender PD, Ganga R, Sambasiva E, Mallikarjuna T, Praneeth VS. 2012. Quantification of phytochemical constituents and in vitro antioxidant activity of *Mesua ferrea* leaves. *Asian Pacific J Trop Biomed*, 2(Suppl 2): S539-S542.
- National Health and Wellness Club. 2002. Smart Nutrition: the essential mineral vitamin and supplement reference guide. NHWC Minnetonka, URL: www.healthandwellnessclub.com (accessed date: March 17, 2022).
- Olafadehan OA, Oluwafemi RA, Alagbe JO. 2020. Carcass quality nutrient retention and caeca microbial population of broiler chicks administered Rolfe (*Daniellia oliveri*) leaf extract as an antibiotic alternative. *J Drug Discov*, 14(33): 146-154
- Olafadehan OA, Oluwafemi RA, Alagbe JO. 2020. Performance haemato-biochemical parameters of broiler chicks administered Rolfe (*Daniellia oliveri*) leaf extract as an antibiotic alternative. *Adv Res Rev*, 2020: 1-4.
- Oluwafemi RA, Isiaka O, Alagbe JO. 2020. Recent trends in the utilization of medicinal plants as growth promoters in poultry nutrition- A review. *Res Agri Vet Sci*, 4(1): 5-11.
- Otles H, Yalcin B. 2012. Phenolic compounds analysis of root stalk and leaves of Nettle. *Sci World J*, 2012: 564367DOI: 10.1100/2012/564367.
- Oyawoye BM, Ogunkunle HN. 2004. Biochemical and haematological reference values in normal experimental animals. *Masson New York*, 2004: 212-218.
- Rafiu TA, Adennola OA, Akinwumi AO, Alabi TA, Shittu MD. 2013. Performance and blood chemistry of broiler chickens fed *Moringa oleifera* leaf meal. *Proceedings of the 18th Annual Conference of the Nigerian*, 21-25 March, Owerri, Nigeria, pp: 294.
- Sandra C. 2020. Phytochemicals: how to improve pig production efficiency with plants. *Inter Pig Magaz*, 8(1): 1-3.
- Singh S, Alagbe JO, Liu X, Sharma R, Kumar A. 2022. Comparative analysis of ethanolic *Juniperus thurifera* leaf stem bark and root extract using gas chromatography and mass spectrometry. *Inter J Agri Anim Prod*, 2(6): 18-27.
- Upadhyay NK, Kumar MS, Gupta A. 2010. Antioxidant cytoprotective and antibacterial effect of sea buckthorn (*Hippophae rhamnoides* L) leaves. *Food Chem, Toxicol*, 48: 3443-3448.
- Yan L, Meng QW, Kim IH. 2011. The effects of dietary *Houttuynia cordata* and *Taraxacum officinale* extract powder on growth performance nutrient digestibility blood characteristics and meat quality in finishing pigs. *Livestock Sci*, 141: 188-193
- Yang C, Chowdhury MAK, Hou Y, Gong J. 2015. Phytochemical compounds as alternatives to in-feed antibiotics: potentials and challenges in application. *Pathogens*, 4: 137-156.
- Zhang S, Jung JH, Kim HS, Kim BY, Kim IH. 2012. Influences of phytoncide supplementation on growth performance nutrient digestibility blood profiles diarrhea scores and fecal microflora shedding in weaning pigs. *Asian-Aust J Anim Sci*, 25: 1309-1315.
- Zhou TX, Zhang ZF, Kim IH. 2013. Effects of dietary *Coptis chinensis* herb extract on growth performance nutrient digestibility blood characteristics and meat quality in growing-finishing pigs. *Asian-Australian J Anim Sci*, 26: 108-115.