

## The effect of *Adhatoda vasica* as a herbal supplement on the performance, nutrient digestibility, and blood profile of weaned pigs

Alagbe Olujimi John

Department of Animal Nutrition and Biochemistry, Sumitra Research Institute, Gujarat, India

### Abstract

A 60-day trial was conducted to gain insight into how *Adhatoda vasica* as a herbal supplement affected the performance, nutrient digestibility, and blood profile of weaned pigs. 40 crossbred male pigs (Yorkshire × Landrace) with an initial body weight of  $10.21 \pm 0.03$  kg were grouped into four treatment groups of ten pigs each. Upon arrival, the animals were quarantined for two weeks and fed a basal diet that is balanced in all nutrients, according to NRC (2012). Pigs in treatment 1 were fed a basal diet without *Adhatoda vasica* leaf powder, whereas those in treatments 2, 3, and 4 were provided the same food supplemented with *Adhatoda vasica* leaf powder at doses of 200 g, 400 g, and 600 g/kg. A completely randomised design approach was used, and the animals had unlimited access to clean, fresh water and food. The concentration of phyto-constituents in *Adhatoda vasica* leaf powder was found to be greater in flavonoids (102.1 mg/g) and lowest in steroids (30.41 mg/g). The average daily weight gain was higher for pigs fed treatments 3 and 4, intermediate for treatment 2, and lowest for treatment 1. The method of treatment significantly altered average daily feed intake, feed conversion ratio, and mortality ( $P < 0.05$ ). The treatment had a significant effect ( $P < 0.05$ ) on digestibility of dry matter, ether extract, crude protein, and crude fibre. There were significant differences ( $P < 0.05$ ) in red blood cell, pack cell volume, mean platelet volume, mean corpuscular volume, mean corpuscular haemoglobin, white blood cell, and total protein levels among treatments, with the exception of aspartate amino transferase, alanine amino transferase, and alkaline phosphatase ( $P > 0.05$ ). However, all levels fell within the recommended range for healthy pigs. Finally, *Adhatoda vasica* leaf powder can be added to pigs' diets without affecting their growth or health.

**Key Words:** *Adhatoda vasica*, phytochemicals, pigs, blood, nutrient, herbs

### Introduction

With the rise of antimicrobial resistance and the more frequent identification of multidrug resistance strains, as well as a lack of research into the development of new antimicrobial agents, scientists are increasingly interested in tests of active substances derived from plants due to their ease of availability, safety, and cost effectiveness (Albert, 2021). The use of medicinal plants in animal nutrition has increased due to their positive properties such as bactericidal, fungicidal, antiviral, antioxidant capacity, growth promoting efficacy, immune stimulating effects, stimulation of digestive enzyme secretion, and nutrient absorption (Cross et al., 2007; Wojcikowski et al., 2004). A medicinal plant, as defined by the World Health Organisation in 2000, is any plant that contains chemicals that can be employed for purposes of medicine or that serve as substrates for chemo-pharmaceutical semi-synthesis. Such a plant's parts, such as leaves, roots, rhizomes, stems, barks, flowers, fruits, grains, or seeds, are used in the control or treatment of a disease condition, and thus include medically active chemical components or phytochemicals (Singh et al., 2022). Phytochemicals are naturally occurring substance that serve as the foundation for modern pharmaceuticals. Alkaloids, flavonoids, saponins, terpenoids, tannins, and phenolic chemicals, among others, have several biological or pharmacological actions in animals (Alagbe, 2022).

*Adhatoda vasica*, a possible medicinal plant, is an evergreen perennial shrub from the Acanthaceae family that grows throughout Asia, including India (Rahman et al., 2004; Gupta et al., 1977). In folk medicine, the leaves and roots are traditionally used in the treatment of various respiratory disorders like bronchitis, asthma, whooping cough, tuberculosis, skin infections, gives relief in pyorrhoea and bleeding gums, malarial, quick ejaculation, headache, hypertension, dysentery, premature ageing, memory improvement, blood cleansing, chronic venous insufficiency, mental function, minor burns, scars, scieroderma, skin ulcers, varicose veins and wound healing (Petel and Venkata, 1984). The plant contains pyrroloquinazoline alkaloids such as vasicine, vasicol, adhatonine, vasicinone, vasicinol, and vasicinolone, which are its main ingredients. These bioactive compounds have demonstrated a wide range of medicinal and pharmacological activity, including anti-malarial, anti-inflammatory, antioxidant, antidiabetic, antibacterial, and anti-cancer properties (Chakrabarty and Brantner, 2001). *Adhatoda vasica* leaf and extracts of roots has been shown to hinder the activity of *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Enterococcus faecalis*, *Staphylococcus aureus*, *Bacillus subtilis*, *Penicillium notatum*, *Candida albicans*, and other bacteria (Vinothapooshan and Sundar, 2010).



The potential functions or works of medicinal plants or herbs in animals for growth promotion include changes in the intestinal microbiota, increased digestibility and nutrient absorption, immune response improvement, and morphological and histological changes of the gastrointestinal tract (Hashemi et al., 2008; Hashemi et al., 2009). However, little or no research is available on the effects of *Adhatoda vasica* as a herbal supplement on weaned pig performance, nutritional digestibility, and blood profile. This study is crucial because it will assist to protect antibiotics for future usage in animal agriculture and ensure food security.

## Materials and methods

### *Experimental Area*

This experiment had been carried out at Sumitra Research Institute's livestock section in Gujarat, India, between 28° 18' N and 70° 35' E, from September to November 2021. The examination followed the rules and animal methodology established by the institute's Animal Department.

### *Collection of Adhatoda vasica leaves and their processing*

Mature leaves of *Adhatoda vasica* were gathered in Orathur village, Kancheepuram district, Tamil Nadu, India, and sent to the Sumitra Research Institute's Nomenclature Department in Gujarat for certification by a licensed taxonomist. The leaves were then air-dried for ten days before being ground into powder with an electric blender and stored in an airtight container for future study. 500 grammes of grinded *Adhatoda vasica* were sent to the department of biochemistry at Sumitra Research Institute in Gujarat for quantitative analysis of its phyto-constituents.

### *Experimental design and animal management*

40 crossbred male pigs (Yorkshire × Landrace) with an initial body weight of  $10.21 \pm 0.03$  kilogrammes were obtained from a renowned breeder farm in Gujarat, separately kept in sections measuring 1.5 m by 2.2 m by 0.5 m (length × width × height) and confined for two weeks. During the adjustment period, pig's were dewormed with Albendazole plus® (1 tablet per kg weight), and fed with basal diet rich in all nutrients needed for pig's according to NRC in 2012. Thereafter, animals weight were balanced before being randomly assigned to four treatment groups. Treatment one was fed basal diet without *Adhatoda vasica* powder while treatment two, three and four were fed same diet with *Adhatoda vasica* powder at 200 grams, 400 grams and 600 grams per kilogram diet. Proper management was thoroughly observed during the 60 days' experimental period and a completely randomized design was adopted. Animals had unlimited access to fresh water and feed. Feed intake, and body weight gain was recorded per pen. Weight was recorded on weekly basis before feeding the animals with an automatic digital scale. Body weight gain was estimated by subtracting initial body weight from the final body weight expressed in kilogram. Total feed intake was calculated as the difference between the left over and feed served.

### *Blood collection and analysis*

On the 60<sup>th</sup> day of the experiment, 5 mL of blood samples were collected from the Jugular vein of five randomly selected rabbits per treatment for haematological and serum biochemical analysis. Blood for haematology (2.5 mL) was collected into bottles with anticoagulant while those for serum indices were placed in bottles without anticoagulant (2.5 mL). Labotronics haematology analyzer (Model- LB -20 HEA) was used to analyze: red blood cell, haemoglobin, pack cell volume, white blood cell and its differentials using triangle laser scattering, flow cytometry differentiation, impedance and cyanide free technique. For precision in the results, the kit is adjusted to a processing speed of 60 test per hour and operating temperature (10 to 30°C) before final outcome were displayed on the monitor.

Serum biochemical indices was carried out using Labdex automatic biochemistry analyzer (Model LX101ABA) adjusted to a sample volume (2 -70 µL), reagent volume (20 – 350 µL), wavelength of about 1000 nm and absorbance (0 to 3 Abs) after calibration to ensure precision in results.

### *Digestibility trial*

Digestibility study was carried out at the end of the trial. Four pigs were randomly selected from each treatment and housed individually before the commencement of the experiment. A known quantity of feed was served to each pigs mixed with chromium oxide (an indigestible marker) daily for five days. Daily fresh excreta were collected and dried at a temperature of 65 °C for 3 days before it was taken to the laboratory for the determination of dry matter, crude protein, crude fibre, ether extract.

### *Evaluation of phyto-constituents in Adhatoda vasica leaf powder*

Phyto-constituents were analyzed according to the procedures recently published by Alagbe (2024). 200 grams of *Adhatoda vasica* leaf powder was analyzed using GC-MS 6800 gas chromatography/mass spectrometer. To ensure precision, gas chromatography chamber where the sample is first injected is maintained at an inlet temperature of



450 °C, column temperature (4 – 450 °C, pressure range (0 – 100 psi ± 0.002 psi) and heating rate up to 1201/min while the mass spectrometer unit is adjusted at an ion source temperature (100 – 350 °C), stability (± 0.10 amu/48 hours), mass range (1.5 – 1000 amu), scan rate (up to 10000 amu/sec) and ionization energy (5 eV – 250 eV). Each phyto-constituents were quantified at different optical density (alkaloids, 500 nm), flavonoids (460 nm), terpenoids (370 nm), alkaloids (550 nm), tannins (480 nm), steroids (250 nm), saponins (360 nm) and phenols (670 nm).

### Proximate composition of experimental diet

Proximate composition of experimental diet was determined using diode array based near infra-red reflectance and trans reflectance analyzer (Model NIRSTM DA 1650). 200 g of feed sample is placed in the collector after the machine was calibrated following the manufacturers lay down procedures. To maintain further precision, it was adjusted at an optical bandwidth (8.75 nm), spectral resolution (1.0 nm), absorbance ranges (up to 2 AU), wavelength accuracy (less than 0.05 nm), photometric noise (400 – 700 nm less 50 micro au; 700 – 2500 nm less than 20 micro au).

### Analysis of data

All data on growth performance, nutrient digestibility, and blood metabolites were analysed using one-way ANOVA in Statistical Package for Social Sciences (version 25). Differences between treatment means (P<0.05) were assessed using the Duncan multiple range test in the same statistical program.

## Results and discussion

Table 1. Ingredients and chemical makeup of the experimental diet

| Ingredients                        | Diet 1 | Diet 2 | Diet 3 | Diet 4 |
|------------------------------------|--------|--------|--------|--------|
| Corn                               | 53.00  | 53.00  | 53.00  | 53.00  |
| Rice bran                          | 11.00  | 11.00  | 11.00  | 11.00  |
| Soybean meal                       | 20.50  | 20.50  | 20.50  | 20.50  |
| Groundnut meal                     | 5.00   | 5.00   | 5.00   | 5.00   |
| Fish meal (Imported: 72%)          | 5.00   | 5.00   | 5.00   | 5.00   |
| Limestone                          | 1.50   | 1.50   | 1.50   | 1.50   |
| Bone meal                          | 3.00   | 3.00   | 3.00   | 3.00   |
| Methionine                         | 0.25   | 0.25   | 0.25   | 0.25   |
| Lysine                             | 0.20   | 0.20   | 0.20   | 0.20   |
| **Mineral/Vitamin Premix           | 0.25   | 0.25   | 0.25   | 0.25   |
| Salt                               | 0.25   | 0.25   | 0.25   | 0.25   |
| Toxin binder                       | 0.05   | 0.05   | 0.05   | 0.05   |
|                                    | 0.00   | 2.00   | 4.00   | 6.00   |
| Total                              | 100.0  | 100.0  | 100.0  | 100.0  |
| Calculated analysis                |        |        |        |        |
| Crude protein (%)                  | 16.50  | 16.50  | 16.50  | 16.50  |
| Crude fibre (%)                    | 4.34   | 4.34   | 4.34   | 4.34   |
| Ether extract (%)                  | 3.21   | 3.21   | 3.21   | 3.21   |
| Ash (%)                            | 2.87   | 2.87   | 2.87   | 2.87   |
| Calcium (%)                        | 1.46   | 1.46   | 1.46   | 1.46   |
| Phosphorus (%)                     | 0.51   | 0.51   | 0.51   | 0.51   |
| Nitrogen free extract (%)          | 48.64  | 48.64  | 48.64  | 48.64  |
| Metabolizable energy (MJ/kg)       | 10.00  | 10.00  | 10.00  | 10.00  |
| Determined analysis (% dry matter) |        |        |        |        |
| Crude protein                      | 18.33  | 18.33  | 18.33  | 18.33  |
| Crude fibre                        | 4.00   | 4.00   | 4.00   | 4.00   |
| Ether extract                      | 3.80   | 3.80   | 3.80   | 3.80   |
| Ash                                | 3.10   | 3.10   | 3.10   | 3.10   |
| Calcium                            | 1.55   | 1.55   | 1.55   | 1.55   |
| Phosphorus                         | 0.63   | 0.63   | 0.63   | 0.63   |
| Nitrogen free extract              | 50.17  | 50.17  | 50.17  | 50.17  |
| Metabolizable energy (MJ/kg)       | 10.60  | 10.60  | 10.60  | 10.60  |

\*\*Mineral-vitamin premix (2.5 kg) contains; Thiamine, 8000 mg, riboflavin, 12,000 mg, pyridoxine, 5000 mg, cyanocobalamine, 5000 mg, niacin, 20,000 mg, D-panthotenate, 10,000 mg, folic acid, 500 mg, biotin, 2000 mg, cholecalciferol, 3,000,000 iu., tocopherol acetate, 25,000 iu., ascorbic acid, 62,000 mg, manganese, 56mg, iron, 70,200 mg, 300 mg, iodine, 200 mg, selenium, 85 mg, choline chloride, 46,000 mg



Table 2 shows that the largest concentration of phyto-constituents in *Adhatoda vasica* leaf powder was flavonoids (102.1 mg/g), subsequently followed by phenols (87.61 mg/g), terpenoids (61.53 mg/g), saponins (51.88 mg/g), tannins (47.12 mg/g), alkaloids (36.04 mg/g), and steroids (30.41 mg/g). The findings demonstrate that *Adhatoda vasica* leaf powder has therapeutic properties or exerts numerous biological effects in animals, including antimicrobial, antifungal, antioxidant, immune-stimulatory, hepatoprotective, and others (Singh et al., 2022; Adewale et al., 2022). They can also be used historically to treat a variety of diseases because they are less harmful, beneficial to the environment, and require no withdrawal period when administered (John, 2024; Shittu and Alagbe, 2022). The phyto-constituent results obtained in this study are consistent with those reported by Kokati et al. (2013) and Harsukhet et al. (2020). The phenolic component content found in this study was higher than that found in *Bryophyllum pinnatum* leaves (18.4 mg/g), *Terminalia bellerica* leaves (29.6 mg/g), and *Xanthium strumarium* leaves (71.6 mg/g) reported by Yadav and Munin (2011). Iqbal et al. (2011) found higher levels of alkaloids, flavonoids, and saponin in the leaves of *Ranunculus arvensis* (0.025 mg/g, 17.69 mg/g, 24.1 mg/g), *Equisetum ravenis* (0.039 mg/g, 10.34 mg/g, 16.07 mg/g), *Carathamus lanatus* (0.017 mg/g, 5.617 mg/g, 25.1 mg/g), and *Fagonia critica* (0.022 mg/g, 9.86 mg/g, 8.223 mg/g). Regional location, species, plant age, and processing methodologies have all been shown to alter phyto-constituents in medicinal plants (John, 2024; Alagbe, 2022).

Table 2: Phyto-constituent in *Adhatoda vasica* leaf powder

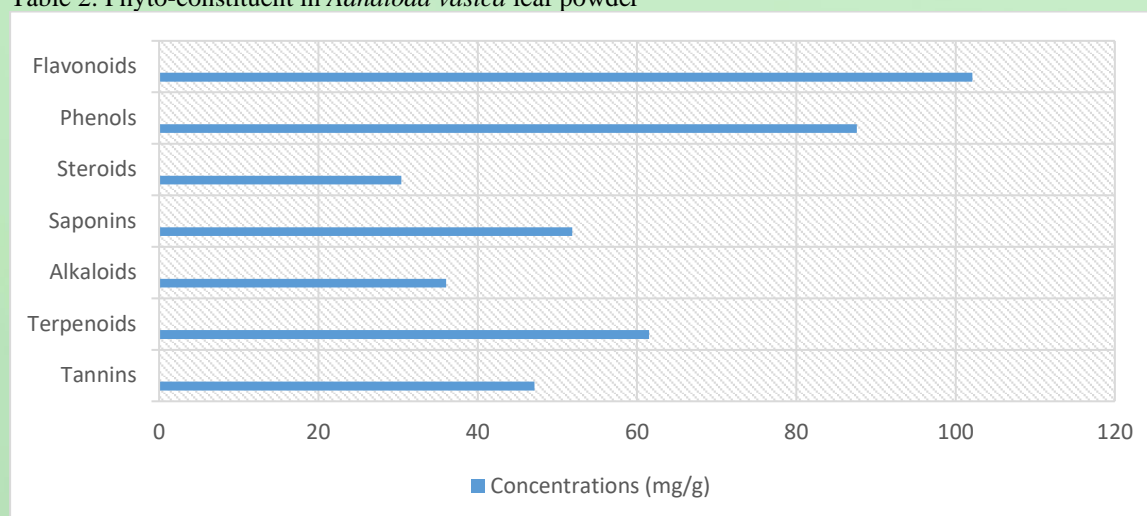


Table 3 shows how *Adhatoda vasica* leaf powder affects the growth performance of weaned pigs. Pigs fed diet 3 (basal diet + 400 g *Adhatoda vasica* leaf powder/kg) gained similar daily weight ( $P>0.05$ ) to those fed diet 4 (basal diet + 600 g *Adhatoda vasica* leaf powder/kg), but significantly higher ( $P<0.05$ ) than those fed diet 2 (basal diet + 200 g *Adhatoda vasica* leaf powder/kg) and diet 1 (basal diet without *Adhatoda vasica* leaf powder/kg). The average daily weight gain found in this investigation with dietary supplementation of diet 3 *Adhatoda vasica* leaf powder (0.33 - 0.51 kg) was comparable to the outcomes of John et al. (2024) and Olujimi et al. (2024), who recorded an average daily weight gain range of 0.24 - 0.50 kg in weaned pigs fed *Cordyline fruticosa* leaf powder. This result was higher than that obtained by Upadaya and Kim (2015) when essential oil and yeast culture were added to the feed of weaned pigs.

The results indicate that supplementation with of *Adhatoda vasica* leaf powder may enhance pig performance through elevating digestive enzyme secretion, decreasing the number of undesirable microbes in the digestive tract, modifying intestinal morphology functions, and ultimately positively affecting animal productivity (Shittu et al., 2022; John, 2024). In this study, pigs fed *Adhatoda vasica* leaf powder (diets 2, 3, and 4) had higher average daily feed intake compared to diet 1 ( $P<0.05$ ). The result could be due to enhanced flavours due to the presence of phyto-constituents such as flavonoids, which have been demonstrated to enhance gut structure and influence barrier porosity to protect from endotoxin damage. This also clarifies the reason no mortality was observed among animals fed *Adhatoda vasica* leaf powder (Li et al., 2012; Mizumoto et al., 2012).

*Adhatoda vasica* leaves have been shown in studies to have both local and systemic anti-inflammatory properties as a result of immunomodulatory action (Subhashini et al., 2011). The results of this investigation are consistent with the findings of Guoqi et al. (2018), who supplemented weaned pig diets with plant essential oil. The feed conversion ratio range observed in this experiment with dietary supplementation of *Adhatoda vasica* leaf powder (2.10 - 2.84) was similar to the results of a study by Zeng et al. (2015) and Li et al. (2012), who discovered that dietary supplementation of essential oil improved feed conversion ratio in weaned pigs. This result was lower than those presented by John (2024) who found out that feed conversion ratio ranged from 1.73 – 2.40 in weaned pigs fed *Cordyline fruticosa* leaf powder.

Table 3: effect of *Adhatoda vasica* leaf powder on the growth performance of weaned pigs



| Variables                                   | Diet 1             | Diet 2             | Diet 3             | Diet 4             | SEM   |
|---|--------------------|--------------------|--------------------|--------------------|-------|
| Number of animals                           | 10                 | 10                 | 10                 | 10                 | -     |
| Experimental days                           | 60.0               | 60.0               | 60.0               | 60.0               | -     |
| Average Initial body weight (kg)            | 10.23              | 10.21              | 10.21              | 10.21              | 0.02  |
| Final body weight (kg)                      | 30.17 <sup>c</sup> | 35.04 <sup>b</sup> | 40.12 <sup>a</sup> | 40.82 <sup>a</sup> | 0.06  |
| Weight gain (kg)                            | 19.94 <sup>c</sup> | 24.83 <sup>b</sup> | 29.91 <sup>a</sup> | 30.61 <sup>a</sup> | 0.04  |
| <sup>a</sup> Average daily weight gain (kg) | 0.33 <sup>c</sup>  | 0.41 <sup>b</sup>  | 0.50 <sup>a</sup>  | 0.51 <sup>a</sup>  | 0.001 |
| Total feed intake (kg)                      | 56.8 <sup>b</sup>  | 63.12 <sup>a</sup> | 63.34 <sup>a</sup> | 63.21 <sup>a</sup> | 0.72  |
| <sup>b</sup> Average daily feed intake (kg) | 0.95 <sup>b</sup>  | 1.05 <sup>a</sup>  | 1.05 <sup>a</sup>  | 1.05 <sup>a</sup>  | 0.01  |
| <sup>c</sup> Feed conversion ratio          | 2.84 <sup>a</sup>  | 2.54 <sup>b</sup>  | 2.11 <sup>c</sup>  | 2.10 <sup>c</sup>  | 0.01  |
| Mortality (%)                               | 1.00               | -                  | -                  | -                  | 0.001 |

<sup>a</sup>weight gain/60 days; <sup>b</sup>total feed intake/60days; <sup>c</sup>average daily feed intake/average daily weight gain; SEM – standard error of mean; diet 1: basal diet without *Adhatoda vasica* leaf powder; diet 2: basal diet supplemented with 200 g *Adhatoda vasica* leaf powder per kg; diet 3: basal diet supplemented with 400 g *Adhatoda vasica* leaf powder per kg; diet 4: basal diet supplemented with 600 g *Adhatoda vasica* leaf powder per kg.

Effect of *Adhatoda vasica* leaf powder on the nutrient digestibility of weaned pigs is presented in Table 4. Dry matter, crude protein and ether extract digestibility of pigs fed diet 3 was similar ( $P>0.05$ ) to those fed diet 4 but significantly higher ( $P<0.05$ ) than the other treatments. Crude fibre value was significantly ( $P<0.05$ ) influenced by the treatments. Dry matter, crude protein and ether extract digestibility recorded in this experiment with the dietary supplementation of *Adhatoda vasica* leaf powder which varied from 69.83 – 85.06 %, 57.88 – 70.28 % and 49.04 – 60.93 % respectively is similar to the result of a study by Maenner et al. (2011) who found out that dry matter, crude protein and ether extract of weaned pigs ranged from 68.84 – 89.02 %, 60.08 – 71.20 % and 40.00 – 62.00 %. Outcome on crude fibre values is also in agreement with the findings of Wan et al. (2017). Potential pathways for improved nutrient digestibility by *Adhatoda vasica* leaf powder supplementation include the ability to enhance desire to eat, saliva formation, intestinal generation of mucus, bile acid production, and activity of digestive enzymes (Mojca, 2020). It may also promote uterine health by increasing calcium stores and pancreatic secretions, hence improving nutrition digestion (John, 2024; Smith et al., 2011). Flavonoids can also help the small intestine absorb nutrients more effectively (Musa et al., 2020; Muritala et al., 2022).

Table 4. effect of *Adhatoda vasica* leaf powder on the nutrient digestibility of weaned pigs

| Variables expressed in percentage | Diet 1             | Diet 2             | Diet 3             | Diet 4             | SEM  |
|-----------------------------------|--------------------|--------------------|--------------------|--------------------|------|
| Dry matter                        | 69.83 <sup>c</sup> | 76.08 <sup>b</sup> | 84.10 <sup>a</sup> | 85.06 <sup>a</sup> | 0.51 |
| Crude protein                     | 57.88 <sup>c</sup> | 63.60 <sup>b</sup> | 70.21 <sup>a</sup> | 70.28 <sup>a</sup> | 0.47 |
| Crude fibre                       | 35.22 <sup>b</sup> | 40.60 <sup>a</sup> | 44.93 <sup>a</sup> | 45.09 <sup>a</sup> | 0.15 |
| Ether extract                     | 49.04 <sup>c</sup> | 56.11 <sup>b</sup> | 60.50 <sup>a</sup> | 60.93 <sup>a</sup> | 0.26 |

SEM – standard error of mean; diet 1: basal diet without *Adhatoda vasica* leaf powder; diet 2: basal diet supplemented with 200 g *Adhatoda vasica* leaf powder per kg; diet 3: basal diet supplemented with 400 g *Adhatoda vasica* leaf powder per kg; diet 4: basal diet supplemented with 600 g *Adhatoda vasica* leaf powder per kg.

Effect of *Adhatoda vasica* leaf powder on haematological indices of weaned pigs is presented in Table 5. Red blood cell, haemoglobin, pack cell volume, mean platelet volume, mean corpuscular volume, mean corpuscular haemoglobin and mean corpuscular haemoglobin concentrations follow similar pattern as pigs fed diet 2 (basal diet + 200 g *Adhatoda vasica* leaf powder/kg) were similar ( $P>0.05$ ) to ones given diet 3 (basal diet + 400 g *Adhatoda vasica* leaf powder/kg) and diet 4 (basal diet + 600 g *Adhatoda vasica* leaf powder/kg) but significantly higher ( $P<0.05$ ) than those in diet 1 (basal diet without *Adhatoda vasica* leaf powder). The improvement in this values could be attributed to the presence of phyto-constituents (as presented in Table 2) capable of supplying the required nutrients to animals and were within the tolerable level for their optimum performance. Haemoglobin (142.7 – 158.1 g/L), pack cell volume values (29.18 – 33.14 %) and red blood cell [(5.61 – 7.18 ( $\times 10^{12}$ /L))] observed in this experiment was within the normal range 95.99 – 154.22 g/L, 20.88 – 35.86 % and [4.19 – 7.00 ( $\times 10^{12}$ /L)] reported by (Czech et al., 2018). The results obtained suggests that the animals have sufficient oxygen in the blood to convey absorbed nutrients round the body (Grace and Alagbe, 2019). The mean platelet volume achieved in this experiment falls within the usual range (7.05 - 13.08 fl) reported by Casas et al. (2015). Mean platelet volume is a key indicator of platelet size and function in the body (Omokore and Alagbe, 2019). Elevated mean platelet volume values imply an inflammatory disease or other health issues (Alagbe, 2021). The mean corpuscular volume, mean corpuscular haemoglobin, and mean corpuscular haemoglobin concentrations ranged from 35.89 to 46.11 fl, 20.80 to 27.17 pg, and 211.3 to 270.1 g/L, respectively, and were within Lindsay's (1977) baseline values. The results for mean corpuscular volume, mean corpuscular haemoglobin, and mean corpuscular haemoglobin concentrations show that the animals were not anaemic (Pampori and Iqbal, 2007). White blood cells produce antibodies and protect the body from pathogens (Mitruka and Rawnsley, 1977). Thrall (2007) and Czech et al. (2018) reported values ranging from 6.36 to 26.30 ( $\times 10^9$ /L).

Table 5. Effect of *Adhatoda vasica* leaf powder on haematological indices of weaned pigs

| Variables measured | Diet 1 | Diet 2 | Diet 3 | Diet 4 | SEM |
|--------------------|--------|--------|--------|--------|-----|
|--------------------|--------|--------|--------|--------|-----|



|  |                    |                    |                    |                    |      |
|--|--------------------|--------------------|--------------------|--------------------|------|
| Red blood cell ( $\times 10^{12}/L$ )            | 5.61 <sup>b</sup>  | 7.06 <sup>a</sup>  | 7.12 <sup>a</sup>  | 7.18 <sup>a</sup>  | 0.02 |
| Haemoglobin (g/L)                                | 142.7 <sup>b</sup> | 152.7 <sup>a</sup> | 156.3 <sup>a</sup> | 158.1 <sup>a</sup> | 4.93 |
| Pack cell volume (%)                             | 29.18 <sup>b</sup> | 31.40 <sup>a</sup> | 32.80 <sup>a</sup> | 33.14 <sup>a</sup> | 0.08 |
| Mean platelet volume (fl)                        | 9.65 <sup>b</sup>  | 11.00 <sup>a</sup> | 11.06 <sup>a</sup> | 11.62 <sup>a</sup> | 0.03 |
| Mean corpuscular volume (fl)                     | 35.89 <sup>b</sup> | 45.11 <sup>a</sup> | 46.07 <sup>a</sup> | 46.11 <sup>a</sup> | 0.04 |
| Mean corpuscular haemoglobin (pg)                | 20.80 <sup>b</sup> | 26.82 <sup>a</sup> | 27.10 <sup>a</sup> | 27.17 <sup>a</sup> | 0.02 |
| Mean corpuscular haemoglobin concentration (g/L) | 211.3 <sup>b</sup> | 262.7 <sup>a</sup> | 268.9 <sup>a</sup> | 270.1 <sup>a</sup> | 9.06 |
| White blood cell ( $\times 10^9/L$ )             | 7.18 <sup>c</sup>  | 11.32 <sup>b</sup> | 17.02 <sup>a</sup> | 17.18 <sup>a</sup> | 0.03 |

SEM – standard error of mean; diet 1: basal diet without *Adhatoda vasica* leaf powder; diet 2: basal diet supplemented with 200 g *Adhatoda vasica* leaf powder per kg; diet 3: basal diet supplemented with 400 g *Adhatoda vasica* leaf powder per kg; diet 4: basal diet supplemented with 600 g *Adhatoda vasica* leaf powder per kg.

The effect of *Adhatoda vasica* leaf powder on blood biochemical parameters in weaned pigs is shown as shown in Table 6. Pigs fed diets 4 and 5 had greater total protein, albumin, and globulin values, while diet 2 had intermediate values and diet 1 had lower levels ( $P < 0.05$ ). The values obtained in this investigation were within the normal range of 24.49 - 43.61 g/L, 20.0 - 35.00 g/L, and 35.0 - 80.0 g/L, as published by Cooper et al. (2014) and Perri et al. (2017) for albumin, globulin, and total protein. This outcome shows that the pigs were not underweight or inflamed (Musa et al., 2022). Aspartate amino transferase, alanine amino transferase and alkaline phosphatase values which varied from 31.67 – 40.11 (U/L), 103.6 – 115.8 (U/L) and 25.90 – 27.92 (U/L) were not significantly ( $P > 0.05$ ) altered by the treatment. The result obtained suggests that dietary supplementation of *Adhatoda vasica* leaf powder up to 600 g/kg was not toxic to the animals and integrity of the liver was not compromised or damaged (Clark and Coffey, 2008). The values found in this investigation were comparable to those reported by Fang et al. (2016) when essential oil was fed to weanling pigs.

Table 6. Effect of *Adhatoda vasica* leaf powder on serum biochemical indices of weaned pigs

| Variables                         | Diet 1             | Diet 2             | Diet 3             | Diet 4             | SEM  |
|-----------------------------------|--------------------|--------------------|--------------------|--------------------|------|
| Total protein (g/L)               | 46.12 <sup>c</sup> | 57.49 <sup>b</sup> | 73.80 <sup>a</sup> | 74.05 <sup>a</sup> | 0.93 |
| Albumin (g/L)                     | 26.00 <sup>c</sup> | 30.41 <sup>b</sup> | 41.80 <sup>a</sup> | 42.05 <sup>a</sup> | 0.41 |
| Globulin (g/L)                    | 20.12 <sup>c</sup> | 27.08 <sup>b</sup> | 32.00 <sup>a</sup> | 32.09 <sup>a</sup> | 0.32 |
| Aspartate amino transferase (U/L) | 31.67              | 39.08              | 40.05              | 40.11              | 0.04 |
| Alanine amino transferase (U/L)   | 103.6              | 110.4              | 112.6              | 115.8              | 4.31 |
| Alkaline phosphatase (U/L)        | 25.90              | 26.17              | 27.88              | 27.92              | 0.86 |

SEM – standard error of mean; diet 1: basal diet without *Adhatoda vasica* leaf powder; diet 2: basal diet supplemented with 200 g *Adhatoda vasica* leaf powder per kg; diet 3: basal diet supplemented with 400 g *Adhatoda vasica* leaf powder per kg; diet 4: basal diet supplemented with 600 g *Adhatoda vasica* leaf powder per kg.

## Conclusion

In the final analysis, *Adhatoda vasica* leaf powder contains numerous phyto-constituents with medicinal effects, which are harmless and do not require a withdrawal time. The results of this investigation demonstrated that *Adhatoda vasica* leaf powder can be supplied in the feed of weaned pigs at a dose of up to 600 g/kg without adversely affecting animal performance or health.

## References

- Adewale, A.O., Alagbe, J.O., Adeoye, Adekemi. O. (2021). Dietary Supplementation of Rauvolfia Vomitoria Root Extract as A Phytogetic Feed Additive in Growing Rabbit Diets: Haematology and serum biochemical indices. International Journal of Orange Technologies, 3(3): 1-12.
- Alagbe John Olujimi, Ramalan Sadiq Muhammad., Shittu Muritala Daniel and Olagoke Olayemi Christiana (2022). Effect of Trichilia monadelpha stem bark extract on the fatty acid composition of rabbit's thigh meat. Journal of Environmental Issues and Climate Change 1(1): 63-71.
- Alagbe Olujimi John (2024). Novel phytogenics' impact on weaned pigs growth performance, haematology and serum biochemical indicators. Black Sea Journal of Agriculture, 7(2): 82-89.
- Alagbe Olujimi John, Daniel Nnadozie Anuore, Shittu Muritala Daniel, Emiola Adewale, Akande Taiwo and Adegoke Adegbite Emmanuel (2023). Impact of dietary supplementation of Carica papaya essential oil on the blood chemistry of broiler chickens. Science Letters, 11(3): 105 - 110.
- Alagbe Olujimi John, Jubril, E.A., Matthew, B., Effiong, E and Taiwo, K.O. (2024). Supplementing broiler chicken diet with Uvaria chamae leaf meal: effects on immune response, gut microbial population and growth performance. Science Letters, 12(1): 10-19.



- Alagbe Olujimi John., Anuore, Daniel Nnadozie., Shittu Muritala Daniel and Ramalan, Sadiq Mohammad (2023). Growth performance and physiological response of weaned pigs fed diet supplemented with novel a phyto-genics. *Brazilian Journal of Science*, 3(1): 43-57.
- Alagbe, J.O (2021). Dietary Supplementation of Rauvolfia Vomitoria Root Extract as A Phyto-genic Feed Additive in Growing Rabbit Diets: Growth Performance and Caecal Microbial Population. *Concept in Dairy and Veterinary Sciences*. 4(2):2021.
- Alagbe, J.O (2022). Use of medicinal plants as a panacea to poultry production and food security: A review. *Gospodarka I Innowacje* 22(2022): 1-12.
- Alagbe, J.O (2023). Investigating the effects of dietary supplementation of Eucalyptus camaldulensis essential oil on the growth performance, nutrient digestibility and caecal fermentation of weaned rabbits. *Research in: Agricultural and Veterinary Sciences*, 7(3): 139 - 148.
- Alagbe, J.O. (2023). Bioactive compounds in ethanolic extract of Strychnos innocua root using gas chromatography and mass spectrometry (GC-MS). *Drug Discovery*, 2023; 17:e4dd1005.
- Alagbe, J.O., Shittu, M.D and Tanimomo, Babatunde K. (2022). Influence of Anogeissusleio carpus stem bark on the fatty acid composition in meat of broiler chickens. *European Journal of Life Safety and Stability* 14(22): 13-22.
- Albert, N. (2022). Strategies for diarrhea control in poultry with natural phyto-biotics. *International Poultry Production*. 5(1): 2-4
- Casas-Diaz, E., Closa, S., Marco, I., Lavin, S., Bach-Raich, E and Cuenca, R. (2015). Haematological and biochemical reference intervals for wild boar captured by cage trap. *Veterinary Clinical Pathology*, 44: 215-222.
- Chakrabarty, A. and Brantner, A.H. (2001). Study of alkaloids from Adhatoda vasica Nees on their anti-inflammatory activity. *Phytother. Research*, 15: 532-534.
- Copper, C.A., Moraes, I.E., Murray, I.D and Owens, S.D. (2014). Haematological and serum biochemical reference values for specific pathogen 6 weeks old Hampshire and Yorkshire crossbred pigs. *Journal of Animal Science and Biotechnology*, 5(1): 25-30.
- Cross D.E., McDevitt R.M., Hillman K. and Acamovic T. (2007): The effect of herbs and their associated essential oils on performance, dietary digestibility and gut microflora in chickens from 7 to 28 days of age. *British Poultry Science*, 48:496–506
- Czech, A., Grela, E., Klebaniuk, R., Ognik. K., Samolinska, W. (2018). Polish crossbred pigs blood haematology parameters depending on their age and physiological state. *Annals of Warsaw University Life Sciences*, 56: 185-195.
- Edeoga, H.A., Okwu, D.E and Mbaebie, B.O. (2005). Phytochemical constituents of some Nigerian Medicinal Plants, *African Journal of Biotechnology Academic Journals*. 4: 685-688.
- Fang T, Liu G, Cao W, Wu X, Jia G, Zhao H, Chen X, Wu C, Wang J. (2016). Spermine: new insights into the intestinal development and serum antioxidant status of suckling piglets. *RSC Advancement*, 6:31323–35
- Grace, F.R and Alagbe, J.O (2019). Effect of *Albizia lebbeck* seed oil dietary supplementation on the haematological and serum biochemical parameters of weaner rabbits. *Sumerianz Journal of Agriculture and Veterinary*. 2(10): 96 -100.
- Gupta, O.P., Sharma, M.L., Ghattak, B.J.R. and Atal, C.K. 1977. Pharmacological investigation of vasicine and vasinone- The alkaloids of Adhatoda vasica. *Indian Journal of Medical Research*, 66: 680 691.
- Hashemi, S., Zulkifli, I., Zunita, Z., Hair-bejo, M., Somchit, M., Kok, P and Davoodi, H. (2009). Effects of dietary supplementation with Euphorbia hirta and acidifier on performance and Salmonella colonization in broiler chickens. *Proceedings of the 30th Malaysia Society of Animal Production Annual Conference*, 2009.
- Hashemi,S., Zulkifli, I., Farida, A. and Somchit, M. (2008). Acute toxicity study and phytochemical screening of selected herbal aqueous extract in broiler chickens.
- Iqbal, H., Riaz, U., Rooh, U., Muhammad, K., Naseem, U., Abdul, B., Farhat, A.K., Muneeb, R., Mohammed, Z., Jehangir, K and Naeem, K. (2011). Phytochemical analysis of selected medicinal plants. *African Journal of Biotechnology*, 10(38): 7487-7492.
- John, A.O. (2024). Clerodendron splendens leaf extract supplementation in weaner rabbits: impact on growth performance, haematology and intestinal microbial population. *Cerrado: Agricultural and Biological Research*, 1(1): 21-31.
- John, A.O. (2024). Effect of coconut shell extract on the growth performance and some haemato-biochemical parameters of broiler chicken. *Brazilian Journal of Science*, 3(6): 82-95.
- John, A.O. (2024). Effect of performance, serum biochemistry and heamatological components of feeding Japanese quails phyto-genic feed additions comparing Megaphrynium macrostachyum leaves. *Brazilian Journal of Science*, 3(5): 51-64.



- John, A.O. (2024). Growth performance, haemato-biochemical indices of broiler chicken fed *Aristochia indica* as a phyto-genic feed additive. *Cerrado: Agricultural and Biological Research*, 1(1): 42-53.
- John, A.O. (2024). Impact of *Cordyline fruitcosa* leaf meal supplemented diet on growth performance, egg production, egg quality and some haematological indices of laying hen. *Brazilian Journal of Science*, 3(9): 19-32
- John, A.O. (2024). Impact of dietary supplementation of *Rhamnus prinoides* leaf extract on the growth performance, nutrient retention and intestinal microbial count of Japanese quails. *Brazilian Journal of Science*, 3(5): 40-50.
- John, A.O., Muritala, S.D., Aduragbemi, Y.A., Kadiri, C.M., Bamiloje, S.O, Jummai, C and Effiong, E. (2024). The approximate mineral and phytochemical content of the leaves, stem bark and roots of *Pterocarpus erinaceus* in India. *Cerrado: Agricultural and Biological Research*, 1(1): 32-41.
- Li P, Piao X, Ru Y, Han X, Xue L, Zhang H. (2012). Effects of adding essential oil to the diet of weaned pigs on performance, nutrient utilization, immune response and intestinal health. *Asian-Australas Journal of Animal Science*, 25:161
- Li SY, Ru YJ, Liu M, Xu B, Péron A, Shi XG. (2012). The effect of essential oils on performance, immunity and gut microbial population in weaner pigs. *Livestock Science*, 145:119–23.
- Lindsay, D. B. (1977). The effect of feeding patterns and sampling on blood parameters. *British Journal of Nutrition*, 56, 99–120.
- Maenner K, Vahjen W, Simon O. (2011). Studies on the effects of essential-oil-based feed additives on performance, ileal nutrient digestibility, and selected bacterial groups in the gastrointestinal tract of piglets. *Journal of Animal Science*, 89:2106 –12
- Mitruka, H. M and Rawnsley, S. K. (1977). *Chemical, Biochemical and Haematological Reference in Normal Experimental Animals*. Mason, N.Y., pp. 287–380
- Mizumoto R, Kawarada Y. (2012) Effects of broccoli extract and various essential oils on intestinal and faecal microflora and on xenobiotic enzymes and the antioxidant system of piglets. *Open Journal of Animal Science*, 2:78–98.
- Mohana, D.V, Lee, S.I and Kim, I.H. (2015). Effect of phyto-genics on growth performance, fecal score, blood profiles, gas emission, digestibility and intestinal morphology of weanling pigs challenged with *E. coli*. *Polish Journal of Veterinary Sciences*, 18(3): 557-564.
- Mojca O.M (2020). Benefits of phyto-genics for laying hens during environmental challenges. *International Poultry Production*. 31(3): 12-13.
- Muritala, Daniel Shittu., Alagbe, J.O., Ojebiyi, O.O., Ojediran, T.K and Rafiu, T.A. (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. *Animal Science and Genetics* 18(2): 57-71.
- Musa, B., Alagbe, J.O., Adegbite Motunrade Betty, Omokore, E.A. (2020). Growth performance, caeca microbial population and immune response of broiler chicks fed aqueous extract of *Balanites aegyptiaca* and *Alchornea cordifolia* stem bark mixture. *United Journal for Research and Technology*, 2(2):13-21.
- National Research Council (2012). *Nutrient Requirements of Poultry*, eighth ed. National Academic Press, Washington, DC. 1994.
- Olujimi, J.A. (2024). Impact of *Cordyline fruiticosa* leaf meal supplemented diet on growth performance, egg production, egg quality and some haematological indices of laying hen. *Discovery Agriculture*, 2024, 10(21) e14da1570.
- Omokore, E.O and Alagbe, J.O. (2019). Efficacy of dried *Phyllanthus amarus* leaf meal as an herbal feed additive on the growth performance, haematology and serum biochemistry of growing rabbits. *International Journal of Academic Research and Development*. 4(3): 97-104
- Pampori, Z. A and Iqbal, S. (2007). Haematology, Serum Chemistry and Electrocardiographic Evaluation in Native Chicken of Kashmir. *International Journal of Poultry Science*, 6(8), 578–582
- Patel, V.K. and Venkata-Krishna- Bhatt, H (1984). In vitro study of anti- microbial activity of *Adhatoda vasica* (L) (Leaf extract) on gingival inflammation- A preliminary report. *Indian Journal of Medical Science*, 38: 70-72.
- Perri, A.M., Sullivan, T.L., Harding, J.C and Wood, R.D. (2017). Haematology and serum reference value of Ontario commercial nursery pigs close to the same weanling. *Canadian Veterinary Journal*, 58: 371-376.
- Puvaca, N., Stanacev V., Glamocic D., Levic J., Peric L., Stanacev V and Milic D. (2013): Beneficial effects of phyto additives in broiler nutrition. *World Poultry Science Journal*, 69(1): 27–34.
- Rahman, S.M.M., Sen, P.K., Afroz, F. and Sultana, K. 2004. In vitro propagation of *Adhatoda vasica* from shoot tip. *Molecular Biology and Biotechnology Journal*, 2(1&2): 33-35.
- Shittu, M.D., Alagbe, J.O., Adejumo, D.O., Ademola, S.G., Abiola, A.O., Samson, B.O and Ushie, F.T. (2021). Productive Performance, Caeca Microbial Population and Immune-Modulatory Activity of Broiler





- Chicks Fed Different Levels Sida Acuta Leaf Extract in Replacement of Antibiotics. *Bioinformatics and Proteomics Open Access Journal* 5(1): 000143.
- Singh Sharma., Alagbe Olujimi John., Liu Xing., Sharma Ram and Kumar Amita (2022). Comparative analysis of ethanolic *Juniperus thurifera* leaf, stem bark and root extract using gas chromatography and mass spectroemetry. *International Journal of Agriculture and Animal Production*, 2(6): 18-27.
- Smith AG, O'Doherty JV, Reilly P, Ryan MT, Bahar B, Sweeney T. (2011). The effects of laminarin derived from *Laminaria digitata* on measurements of gut health: selected bacterial populations, intestinal fermentation, mucin gene expression and cytokine gene expression in the pig. *Brazilian Journal of Nutrition*, 105:669–77.
- Subhashini S, Kantha D. Arunachalam (2011). Investigation of the phytochemical activities and wound healing properties of *Adhathoda vasica* leave in swiss albino mice. *African Journal of Plant Science*. 5(2): 133-145
- Thrall, M. A. (2007). *Hematologia e Bioquimica Clinica Veterinaria*. Philadelphia: Lippincott Williams & Wilkins, Sao Paulo: Roca, 582 pp
- Upadhaya, S.D and Kim, I.A. (2015). Effects of essential oil and yeast culture supplements on growth performance, nutrient digestibility and blood characteristics of weaning pigs. *Indian Journal of Animal Sciences*, 85(9): 1017-1020.
- Upadhaya, S.D and Kim, I.H. (2015). Effect of essential oil and yeast culture supplementation on growth performance, nutrient digestibility and blood metabolites in weanling pigs. *Indian Journal of Animal Sciences*, 85(9): 1017-1020.
- Vinothapooshan, G. and Sundar, K. (2010). Hepatoprotective activity of *Adhatoda vasica* leaves against carbontetrachloride induced toxicity. *Pharmacologyonline*, 2: 551-558.
- Wan J, Jiang F, Xu Q, Chen D, Yu B, Huang Z, He J. (2017). New insights into the role of chitosan oligosaccharide in enhancing growth performance, antioxidant capacity, immunity and intestinal development of weaned pigs. *RSC Advancement*, 7(16):9669–79
- Wojcikowski, K., Johnson, D.W and Gobé, G. (2004). Medicinal herbal extracts -- renal friend or foe? Part one: the toxicities of medicinal herbs. *Nephrology*. 9 (5): 313-31
- World Health Organization, (WHO) (2000). *Promoting the Role of Traditional Medicine in Health Systems: A Strategy for the African Region 2001-2010*. Harare, World Health Organization. pp.10- 286.
- Yadav, N.P and Dixit, V.K. (2008). Recent approaches in herbal drug standardization. *International Journal of Intergrative Biology*. 2(3): 195-203
- Yadav, R.N.S and Munin, A. (2011). Phytochemical analysis of some medicinal plants. *Journal of Phytology*, 3(12): 10-14.
- Zeng Z, Xu X, Zhang Q, Li P, Zhao P, Li Q, Liu J, Piao X. (2015). Effects of essential oil supplementation of a low-energy diet on performance, intestinal morphology and microflora, immune properties and antioxidant activities in weaned pigs. *Animal Science Journal*, 86:279–85

