



NUTRITIONAL AND ANTI-NUTRITIONAL FACTORS OF PURPLE ORCHID (*Piliostigma reticulatum*) (DC.) SEED AS A POTENTIAL PLANT PROTEIN

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ABSTRACT

The study aimed to assess the nutritional profile of purple orchid (*Piliostigma reticulatum*) seeds as a potential alternative plant protein source for poultry feed. Ground seed samples were analyzed for proximate composition, mineral content, amino acid profile, and anti-nutritional factors using standard analytical procedures. Proximate analysis indicated that *Piliostigma reticulatum* seeds contain considerable amounts of crude protein (28.34), ash (12.51), and nitrogen-free extract (19.34%). The amino acid analysis revealed the presence of all 20 essential and non-essential amino acids, with glutamic acid (11.08), aspartic acid (8.90), and phenylalanine (4.09%) being the most abundant. The seeds were also rich in both macro and micro minerals, particularly potassium (8.91%), phosphorus (0.86%), manganese (147.80), zinc (25.63), iron (15.06), and iodine (5.60 mg/kg). Phytochemical screening identified several anti-nutritional compounds, including phenols (71.32), alkaloids (6.00), lectins (3.69), and trypsin inhibitors (1.15 mg/100g). The findings suggest that, with appropriate detoxification, *Piliostigma reticulatum* seed could serve as an affordable and effective plant protein supplement in poultry nutrition.

Keywords: Amino acids, Anti-nutrients, Minerals, *Piliostigma reticulatum*, Proximate

INTRODUCTION

The global poultry industry has experienced rapid expansion over the past three decades (Sugioto and Toana, 2021). This growth has intensified the demand for feed ingredients, particularly protein-rich raw materials. However, the overreliance on conventional plant protein sources, such as soya bean, combined with rising population pressure, urbanization, and limitations in local production, has contributed to increasing feed costs. Consequently, poultry products have become less affordable for many households (Chala, 2024). Given these challenges, there is a pressing need to identify alternative, underutilized plant protein sources that are affordable, readily available, and nutritionally comparable to soya bean. One promising plant is purple orchid, camel's foot, monkey bread (*Piliostigma reticulatum* DC.) seed commonly known in Hausa as “*kargo* or *kalgo*”. Purple orchid (*P. reticulatum*) is a leguminous shrub or small tree belonging to the family *Caesalpiniaceae* (Soufianou and Issoufou, 2024). It is widely distributed across arid region of tropical Africa, particularly in the Sahel and Sudan zones, including Senegal, Mali, Burkina Faso, Niger, Nigeria, Chad, Sudan, and Ethiopia (Wezel and Bocker, 1999). Adapted to harsh dry climates and nutrient-poor soils, the species is commonly found in savannah ecosystems, fallow lands, and agroforestry systems, where it plays an important role in soil fertility improvement, erosion control, and biodiversity conservation (Bayala *et al.*, 2015).

Morphologically, *P. reticulatum* grows up to 15 m in height, with a spreading crown and characteristic bilobed leaves resembling a camel's hoof. It produces woody pods that remain on the tree after leaf fall and contain multiple hard seeds (FAO, 2017). Several species within the genus have been described, including *P. thonningii* (Schumach.), *Bauhinia reticulata* (DC.), and *P. pyrrhocarpum* (Hochst.), but the two African species most widely distributed are *P. reticulatum* and *P. thonningii* (Haruna *et al.*, 2015). While the two share similar characteristics, *P. thonningii* typically exhibits larger, pubescent leaves and is more common in less arid environments (Burkill, 1995; Iliyasu *et al.*, 2020). Beyond its ecological importance, *P. reticulatum* is valued as a multipurpose species providing



fodder, firewood, and raw materials (Keay, 1989; Nouhoun *et al.*, 2013). The seeds, in particular, are of interest as a potential feed resource. Previous studies have reported crude protein contents ranging from 30.30% in raw seeds to 40.40% in defatted seeds (Akin-Osanaiye *et al.*, 2009), and 34.20% in heat-processed seeds (Musa and Bichi, 2015), values comparable to those of soya bean (38 - 44%; Aduku, 1992). Despite this nutritional potential, *P. reticulatum* seeds also contain anti-nutritional factors, including *saponins*, *tannins*, *cyanides*, *oxalates*, *phytates*, and *phenols*, which may reduce nutrient utilization and animal performance (Jimoh and Oladiji, 2005; Haruna *et al.*, 2015).

To mitigate these limitations, processing techniques such as boiling and other heat treatments have been evaluated. Haruna *et al.*, (2015) demonstrated that thermal processing substantially reduced anti-nutritional compounds, with traditional boiling emerging as the most effective method, in some cases eliminating specific factors entirely. In light of its nutritional potential and adaptability to marginal environments, *P. reticulatum* represents a viable non-conventional protein source for poultry diets. This study was therefore undertaken to evaluate the chemical composition and nutritional potential of *P. reticulatum* seeds, with a view to promoting their utilization as an affordable supplement in poultry nutrition.

MATERIALS AND METHODS

Procurement and Identification of Purple Orchid (*Piliostigma reticulatum*) Seed

Mature *Piliostigma reticulatum* seeds were collected from the outskirts of Potiskum, Yobe State, Nigeria. Potiskum lies between latitude 11°42' 50.08'N and longitude 11°04' 51.89'E, with an altitude of approximately 426 m above sea level. The region has an average annual temperature of 24.9 °C and receives about 713 mm of rainfall per annum (Abdurrahman *et al.*, 2017). Pods were manually opened to extract seeds, which were subsequently winnowed to remove debris and stored in clean containers. Species identification was carried out at the Department of Forestry, College of Agriculture and Technology, Gujba, Yobe State, Nigeria. The seeds were then ground to a particle size of 2-3 mm for subsequent analyses.

Proximate Analyses of Purple Orchid (*Piliostigma reticulatum*) Seed

Proximate composition, including dry matter (DM), crude protein (CP), crude fiber (CF), ether extract (EE), nitrogen-free extract (NFE), and ash, was determined using the standard procedures of the Association of Official Analytical Chemists (AOAC, 2006).

Minerals Analyses of Purple Orchid Seed

Mineral content was determined following the procedure of Shahid *et al.* (1999). Briefly, ground seed samples were subjected to dry ashing at 550 °C in clean crucibles. The ash was dissolved in 5 ml of nitric acid and hydrochloric acid (HNO₃/HCL, 1:2) and heated gently until brown fumes disappeared and a whitish residue remained. The solution was filtered into a 100 ml volumetric flask and diluted to volume with deionized water. Mineral concentrations were quantified using a flame atomic absorption spectrophotometer (Model VGP 210, Buck Scientific, USA) and expressed as mg/100g.

Amino Acid Profile of Purple Orchid Seed

The amino acid profile was determined using high-performance liquid chromatography (HPLC) as described by Kaur *et al.* (2021). Approximately 100 mg of the ground seed sample was hydrolyzed with 6 N HCl at 110 °C for 24 hours in sealed digestion bottles under vacuum. Hydrolysates were extracted with 1 ml of 0.1 N HCl. Separation was carried out using a reverse-phase HPLC system equipped with a UV detector at 338 nm, a C18 column (2.5 × 200 mm, 5 µm), and a mobile phase of sodium phosphate buffer (100 mM, pH 7.2), acetonitrile, and methanol in the ratio of 1:1:2. The flow rate was maintained at 0.45 ml/min and column temperature at 40 °C. Identification of amino acids was based on retention times compared with authenticated standards (Thermo-Scientific, NC1018) as reported by Manpreet *et al.* (2021).

Phytochemical Screening of Purple Orchid Seed

Phytochemical screening was performed to detect secondary metabolites, including *tannins*, *saponins*, *oxalates*, *phytates*, *phenolics*, *cyanides*, *trypsin* inhibitors, *lectins*, and *alkaloids*, using standard procedures described by Olademeji *et al.* (2012) and Olawale and Cyril (2015).



RESULT AND DISCUSSION

Proximate Composition of Purple Orchid (*Piliostigma reticulatum*) Seed

The proximate composition of **purple orchid** (*Piliostigma reticulatum*) seeds is presented on Table 1. The analysis demonstrates that the seeds are nutritionally rich, with crude protein constituting a substantial proportion (28.34%). The levels of other components were recorded as follows: moisture (11.13%), crude fiber (25.30%), ether extract (3.38%), ash (12.51%), and nitrogen-free extract (19.34%). The dry matter content (88.87%) was slightly lower than that previously reported for *Piliostigma thonningii* (93.29%) and baobab (*Adansonia digitata*) seeds (94.12%), as noted by Jimoh and Oladiji (2005) and Shehu *et al.* (2021). The comparatively higher dry matter content reported in these species has been associated with improved storage potential and extended shelf life (Jimoh and Oladiji, 2005).

The crude protein (CP) content obtained in this study (28.34%) was comparable to values previously reported for *Piliostigma reticulatum* seed (30.30 and 34.40%) by Akin (2009) and Musa and Bichi (2015) respectively, as well as for *Piliostigma thonningii* (30.33%) noted by Jimoh and Oladiji (2005). However, the CP value recorded here was higher than those reported for *Ziziphus mauritiana* seeds (19.27%) and *Vigna unguiculata* seeds (22.66%) Mufwan *et al.* (2021); Vivian and Wilson (2024). In contrast, the result was lower than values reported for other non-conventional protein sources, including *Hibiscus sabdariffa* (sorrel) seeds (43.15%) and baobab (*Adansonia digitata*) seeds (38.71%) (Shehu *et al.*, 2021), as well as beni seeds (*Sesamum indicum*), which contained 48.65% CP (Nwosu *et al.*, 2020). The relatively high crude protein content of *P. reticulatum* seeds highlights their potential economic value as an alternative plant protein supplement. The metabolizable energy content (2008.93 kcal/kg) observed in this study were closely aligned to the values reported by Shehu *et al.* (2021) for sorrel seed and baobab seed cakes (2675.66 and 2720.44 kcal/kg, respectively), but lower than that reported for *Z. mauritiana* seeds (3025.40 kcal/kg) by Mufwan *et al.* (2021).

The crude fiber (CF) content was notably high (25.30%) compared to the value (6.05%) reported by Musa and Bichi (2015). It also exceeded levels found in several non-conventional plant protein sources, including moringa seed cake (9.47%), roselle seed cake (8.75%), and baobab seed cake (Afolayan *et al.*, 2020; Shehu *et al.*, 2021). However, the CF content was relatively close to that of *Cucurbita pepo* seed meal (21.58%) reported by Wafar *et al.* (2017). Although moderate levels of dietary fiber can improve digestibility, excessive fiber may reduce nutrient digestibility and cause gastrointestinal irritation (Jimoh and Oladiji, 2005). Nonetheless, dietary fiber plays an important physiological role, influencing gastrointestinal function through bile acid elimination, modulation of fecal water content, and intestinal transit time, while also reducing the risk of colon cancer (Ogbemudia *et al.*, 2018; Bayero *et al.*, 2019).

Table 1: Proximate Composition (%) of Raw Purple Orchid (*Piliostigma reticulatum*) Seed

Parameters	Proportions
ME (Kcal/kg)	2008.93
Crude protein (%)	28.34
Crude fibre (%)	25.30
Ether extract (%)	3.38
Ash (%)	12.51
NFE (%)	19.34
Dry matter (%)	88.87

ME = Metabolisable energy, NFE = Nitrogen Free Extract

Amino Acid Profile of Purple Orchid (*Piliostigma reticulatum*) Seed

The amino acid analysis (Table 2) revealed that glutamic acid (11.08), aspartic acid (8.96), and phenylalanine (4.09%) were the predominant amino acids in *P. reticulatum* seeds, consistent with reports by Manpreet *et al.* (2021), who identified glutamic and aspartic acids as abundant in various oilseeds. Essential amino acids detected included glycine (2.45%), methionine (1.17%), tryptophan (1.40%), threonine (1.80%), arginine (1.80%), leucine (3.40%), isoleucine (2.27%), valine (2.40%),

and histidine (0.98%). These values were generally lower than those reported for full-fat soybean (Sergey *et al.*, 2020), but comparable to values recorded for *Prosopis* seeds (Yusuf *et al.*, 2008) and groundnut cake (Ajah and Asuquo, 2018).

The result noted, the levels of lysine (2.15%), methionine (1.17%) and threonine (1.19%) were relatively low compared to soya bean cake (6.62, 1.00 and 3.81%) respectively; Sergey *et al.* (2020) and groundnut cake (3.26, 0.93 and 1.48%, respectively; Ajah and Asuquo, 2018). Since lysine, methionine and threonine are among the most limiting amino acids in poultry nutrition, this suggests that diets incorporating *P. reticulatum* seeds for broiler chickens would require supplementation especially methionine and threonine to ensure optimal growth and productivity. Knowledge of the amino acid profile is therefore essential in determining the nutritive value and potential applications of the seed as an alternative protein resource.

Table 2: Amino Acids Profile of Raw **Purple Orchid** (*Piliostigma reticulatum*) Seed

Parameters	Proportions (%)
Lysine	2.15
Methionine	1.17
Tryptophan	1.43
Threonine	1.19
Arginine	1.85
Glycine	1.50
Histidine	0.98
Isoleucine	2.27
Leucine	3.43
Aspartic acid	8.96
Phenylalanine	4.09
Valine	2.40
Serine	3.80
Proline	3.60
Alanine	2.38
Glutamic acid	11.08
Tyrosine	3.07
Cysteine	2.56

Minerals Content of Purple Orchid (*Piliostigma reticulatum*) Seed

The results of the mineral analysis of *Piliostigma reticulatum* seeds revealed the presence of both macro (Ca, Mg, P, K, S) and trace (Mn, Zn, I, Fe) elements. The mineral profile was comparable to that reported for *Piliostigma thonningii* seeds by Jimoh and Oladiji (2005). However, the concentrations of K, P, Mn, Zn, iodine, and Fe were considerably higher than those previously reported for *P. reticulatum* seeds. Conversely, the mineral values obtained were lower than those observed in soya bean seeds, as documented by Ogbemudia *et al.* (2018) and Riya *et al.* (2024).

The relatively high levels of Fe and Zn are of particular nutritional significance, as these elements play essential physiological roles. Iron is a crucial component of hemoglobin, which facilitates oxygen transport in red blood cells, while zinc contributes to immune function and is necessary for protein and DNA synthesis. Most of the mineral concentrations recorded in this study meet the minimum nutrient requirements recommended for meat-producing and laying birds (NRC, 1994; Abbass *et al.*, 2023).

Table 3: Minerals Composition of Raw Purple Orchid (*Ptilostigma reticulatum*) Seed

Parameters	Proportions
Macro nutrients	
Calcium (%)	1.32
Magnesium (%)	0.26
Phosphorus (%)	0.86
Potassium (%)	8.91
Sodium (mg/kg)	11.50
Micro nutrients	
Manganese (mg/kg)	147.88
Zinc (mg/kg)	25.63
Iodine (mg/kg)	5.60
Iron (mg/kg)	15.05

Anti-nutritional Factors in Purple Orchid (*Ptilostigma reticulatum*) Seed

The results of anti-nutritional factor analysis are presented on Table 2. The findings indicate the presence of several anti-nutritional compounds, including saponin, oxalate, phytate, tannin, phenol, cyanide, trypsin inhibitor, lectin, and alkaloid. Among these, phenol (71.32), alkaloid (6.0), lectin (3.69), and trypsin inhibitor (1.15 mg/100g) were the most abundant, whereas oxalate (0.06), cyanide (0.24), and tannin (0.20 mg/100g) were present in the lowest concentrations. These results align with the earlier reports of Haruna *et al.* (2015) and a related specie *Ptilostigma thonningii* by Jimoh and Oladiji (2005). The saponin content (0.28 mg/100g) of *P. reticulatum* seeds was slightly higher than that reported by Haruna *et al.*, (2015), yet lower than the value recorded for sorrel (*Hibiscus sabdariffa*) seeds (3.0 mg/100g) by Lamalang *et al.* (2024). Similarly, the levels of oxalate (0.06), phytate (0.32), and tannin (0.20 mg/100g) were lower than those reported for *H. sabdariffa* (Lamalalang *et al.*, 2024) but consistent with the findings of Haruna *et al.* (2015). The cyanogenic content (0.24) of *P. reticulatum* was slightly higher than the (0.17) reported by Haruna *et al.* (2015) but lower than the 0.27 mg/100g reported for *Cucurbita pepo* by Wafar *et al.* (2017).

Overall, the levels of anti-nutritional factors in *P. reticulatum* do not pose a significant threat to ruminant animals. However, processing is necessary to detoxify the seeds before inclusion in poultry feeds. This is crucial because several of the measured values exceeded the permissible limits recommended by WHO/FAO, as noted by Olan *et al.*, (2024). If not adequately managed through processing or supplementation, anti-nutritional compounds can adversely affect poultry health and productivity, leading to reduced feed intake and nutrient utilization (Chala, 2024).

Table 4: Anti-nutritional Factors in Raw Purple Orchid (*Ptilostigma reticulatum*) Seed

Parameters	Proportions
Saponin (mg/g)	0.28
Oxalates (mg/g)	0.06
Phytate (mg/g)	0.32
Tannin (mg/g)	0.20
Phenolic (mg/GAE/g)	71.32
Cyanide (mg/g)	0.24
Trypsin inhibitors (mg/g)	1.15
Lactin (mg/g)	3.69
Alkaloids (mg/g)	6.00

CONCLUSION AND RECOMMENDATION

It can be concluded from the results obtained that, purple orchid seed have a good nutritional profile with appreciable quantities of protein and other nutrients, which could save as a substitute to conventional plant protein supplement in poultry production. Moreover, the study also established the presence of amino acids, minerals and anti-nutritional factors in purple orchid seed, which could be a cheap, affordable and locally available source of non-conventional plant protein supplement in poultry production if appropriately detoxified.



Further investigation on the effect of incorporation of purple orchid seed in different species of poultry is therefore recommended.

ACKNOWLEDGMENTS

The authors wish to acknowledge the immense contribution of Tertiary Education Trust Fund (Tetfund) for their sponsorship. Desert Research Monitoring and Control Center and Yobe State University in particular for their support.

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Nigerian Journal of Agriculture and Agricultural Technology (NJAAT)
Volume 5, Number 4A, December, 2025
ISSN (Print): 2811-1885; ISSN (Online): 2811-1893



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