

## Nutrients, Phytochemical Composition and Antioxidant Analysis of Selected Agricultural Wastes as Potential Livestock Feed Ingredients

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### ABSTRACT

This study was carried out to analyse the proximate, phytochemical composition and antioxidant activity of three agricultural wastes (Melon husk, moringa pod and mango endocarp). The proximate parameters of the samples were determined using the standard analytical method of Association of Official Analytical Chemists. The antioxidant activity was determined by the radical scavenging activity of the methanol extract of samples against DPPH (Sigma Aldrich) by UV-Visible Spectrophotometer at 517 nm. The results revealed that the three samples are high in crude fibre: 51.61% (melon seed husk), 58.10% (moringa pod) and 49.47% (mango endocarp). The crude protein content of the samples ranged from 3.45% to 12.22%. The mineral analysis showed that melon seed husk contains; Ca (14.94 mg/kg), Mg (52.26 mg/kg), Fe (21.2mg/kg), P (106.7 mg/kg), Zn (0.70 mg/kg); moringa pod contains Ca (243.95 mg/kg), Mg (61.44 mg/kg), Fe (3.60 mg/kg), P (33.75 mg/kg), Zn (0.35 mg/kg); mango endocarp; Ca(6.14 mg/kg), Mg (13.00 mg/kg), P (24.25 mg/kg), Zn (0.17 mg/kg). Phytochemical screening of the three samples showed small amount of antinutrient like saponin, oxalate, flavonoid etc. The results obtained from this study indicate that the agricultural wastes may serve as energy and mineral supplier and thus may be utilized as a potential feed ingredient in livestock feed, coupled with their low antinutrient content.

**Keywords:** Antioxidant, Mango endocarp, Melon husk, Moringa pod, Nutritional

### INTRODUCTION

Agricultural wastes are all forms of plant-derived or animal-derived materials that are considered useless either because they have no known positive economic importance or because they are not grown/raised for any specific purpose. These include woods, herbaceous plants, crops and forest residues, animal wastes and so on (Adeyi, 2010). In Nigeria, large quantities of these wastes are produced annually and are vastly underutilised. Many agricultural products such as soybean and groundnut used as ingredients in the formulation of feed for livestock are also staple food for many people (Ogbe and George, 2012). In Africa corn/maize is a staple food for many people and soybean is not readily available, as soybean is utilized for making soybean milk which is taken as fresh drink by many people. Soybeans are not readily available (Mutayoba *et al.*, 2011). Thus, there is competition for these feed ingredients between animals and humans, making them more expensive and their inclusion in poultry diet

also increases the cost of poultry production (Mutayoba *et al.*, 2011). Under normal conditions the cost of poultry feed have been reported to account for over 70% of the total cost of poultry production (Mutayoba *et al.*, 2011; Ogbe and George, 2012). Consequently, a search for alternative sources of raw material for livestock feed will go a long to mediate the competition between human food sector and animal food sector without compromising quality (Ogbe and George, 2012). Nutritionists and research scientists are turning attention to alternative sources of feed ingredients such as agricultural and industrial by-products. An agrarian country like Nigeria is rich in many agricultural by-products, majority of which are laying waste as litters in the environment.

Melon (*Citrullus lanatus*) is a cucurbit crop that belongs to the family cucurbitaceae. Melon seeds 'Egusi' are grown, harvested and processed by removing the seeds from the fruits followed by washing and sun-drying. The flesh is inedible but the seeds are valuable in

Africa. They found usage either as food supplement (soup condiment) or also for medicinal purposes (Abdulrazak *et al.*, 2014). They are used in the treatment of urinary tract infection, hepatic congestion, intestinal worms and abnormal blood pressure (Moerman, 1998). Melon husks are shells that are discarded after shelling of melon seeds. Melon husk is a lignocellulosic plant biomass and a waste product from the food industry, having proteins, celluloses and lipids coated on its surface (Adelagun *et al.*, 2014). Mango (*Mangifera indica* L.), is an Indian or a tropical evergreen (deciduous) tree which produces green fruits when unripe but green to light green or yellowish to reddish (sweet, juicy and succulent) fruits when ripe (Morton, 1987; Bally, 2006; Seifu, 2010). Among the major producers of mango in the World, Nigeria ranks 8<sup>th</sup> (Morton, 1987; FAO, 2004; Yusuf and Salau, 2007), and Benue ranks first (1<sup>st</sup>) in the league of states that produce mangos in Nigeria (Yusuf and Salau, 2007). The other mango producing states in Nigeria include; Jigawa, Plateau, Kebbi, Niger, Kaduna, Kano, Bauchi, Sokoto, Adamawa Taraba and the Federal Capital Territory (FCT) (Yusuf and Salau, 2007). Mango pulp can be consumed at ripeness or in immaturity although most of the fruit is eaten fresh, and a wide range of foods can be prepared with the pulp (Ribeiro and Schieber, 2010). It can be canned, frozen as concentrates, mashed, dehydrated, minimally processed, or prepared as juices and jams (Masibo and He, 2009). It is estimated that 35-60% of the fruit is discarded as waste after processing (O'Shea *et al.*, 2012), in the particular case of the seed, more than one million tons of mango seeds are annually produced as wastes, and these are not currently utilized for any commercial purposes (Leanpolchareanchai *et al.*, 2014). After consumption of the pulp, the mango seed endocarps are usually dumped carelessly and thereby constituting a nuisance to the environment, especially during its season.

*Moringa Oleifera* seeds are large and circular-shaped, and grow inside the lengthy pods of the *Moringa Oleifera* tree (Adejumo and

Abayomi, 2012). Moringa seed pods can reach well over a foot in length and each pod can provide over a dozen large Moringa seeds. Moringa trees produce seed pods on an annual basis, much like other similar species in the plant kingdom. Moringa trees give off incredible volume of seed pods during their reproduction months. An average-sized Moringa tree of fifteen to twenty feet in height can produce hundreds or even thousands of seed pods, yielding countless Moringa seeds every year (Adejumo and Abayomi, 2012). The fresh moringa seeds have long been used by people as a tasty vegetable and when dried, it is used as water purifier because of its coagulant properties (Ayotunde *et al.*, 2011). However, large quantities of the melon husks, moringa pod and mango endocarp are discarded and burnt, which pollute the environment. To the best of the authors' knowledge, there is no comprehensive information for the nutritional and antioxidant assessment of moringa pod and as such the study would provide not only baseline data for literature but also accord industrialists a feedstock base to make choices.

The aim of this study therefore is to determine the nutritional, phytochemical composition and antioxidant properties of melon seed husk, mango endocarp and moringa seed pod. The main objective is to evaluate their potential as possible feed ingredient for livestock.

## **MATERIALS AND METHODS**

### **Materials**

The materials for this analysis are: Melon husk, Mango endocarp, Moringa pod. The melon husk and mango seed were sourced in Sheda village, Kwali Area Council of FCT, Nigeria. The moringa pods were obtained from the moringa plantation of Sheda Science and Technology Complex (SHESTCO), Abuja, Nigeria. All other reagents are of high analytical grade and were products of Sigma.

### **Preparation of Samples**

The mango seed integument and seed leaf were removed from the seeds, leaving the mango endocarp. The samples (melon husk, mango endocarp and moringa seed pod) were

air-dried for three days and pulverized to smaller particles using blender (for the melon husk) and pulverizing machine (for the moringa pod and mango endocarp). The pulverized samples were sieved, then packaged into air tight polythene bag and kept until when needed.

### Methods

The method of AOAC (1990) was used for proximate analysis including determinations of moisture content, ash content, crude lipid, crude protein and carbohydrate.

### Antioxidant capacity

The antioxidant activity was evaluated by discolouration of 2, 2-diphenyl-1-picrylhydrozyl radical (DPPH) in methanol by a slightly modified method of Brand-Williams *et al.* (1995). The following concentrations of the extract were tested (0.5, 0.25, 0.125, 0.0625, 0.03125 mg/ml). The absorbance was monitored at 517nm. Vitamin C was used as the antioxidant standard at concentrations (0.5, 0.25, 0.125, 0.0625, 0.03125 mg/ml). A blank solution was prepared containing the same amount of methanol and DPPH. The radical scavenging activity was calculated using the following formula:

$$\text{Inhibition (\%)} = \frac{A_b - A_s}{A_b} \times 100$$

Where  $A_b$  = Absorbance of the blank  
 $A_s$  = The absorption of the sample extract.

### Quantitative Phytochemical Determination

This analysis was carried out to test for the presence and quantity of phenols (AOAC, 1995), alkaloids (Manjunath *et al.*, 2012), tannins (Van-Burden and Robinson, 1981), flavonoids (Vabkova and Neugebauerova, 2012), saponins (Obadoni and Ochuko, 2001), Oxalate (Ukpabi and Ejidoh (1989).

### Determination of Gross energy

The gross energy content was determined by calculation from fat, carbohydrate and protein contents using the Atwater's conversion factors; 16.7 kJ/g (4kcal/g) for protein, 37.4 kJ/g (9 kcal/g) for fat and 16.7 kJ/g (4 kcal/g) for

carbohydrates and expressed in calories (Guyot *et al.*, 2007).

### Mineral analysis

Two grams (2 g) of each of the samples was weighed into a beaker and 20 ml of nitric acid was added to the sample. This was heated on the hotplate at 60°C for 30 min. It was then removed from heat and allowed to cool. The solution was diluted with distilled deionized water and made up to 50 ml in a volumetric flask. A blank was prepared in the same manner and poured into a polypropylene bottle. The samples were analysed for metals on Thermo Scientific iCE 3000AA02134104 Atomic Absorption Spectrometer using appropriate working standards (Emmanuel *et al.*, 2016). Phosphorus was determined by vanadomolybdate colorimetric method (Ologhobo and Fetuga, 1983)

### Data Analysis

All data generated were analyzed using descriptive statistic as described by Olawuyi (1996). Statistical values that were calculated include mean and standard deviation.

## RESULTS AND DISCUSSION

Analysis of proximate composition provides information on the basic nutritional composition of the samples. The compositions are moisture, ash, crude fat, protein, crude fibre, and carbohydrate. These components are crucial to the assessment of the nutritive quality of the samples being analysed. The results of the proximate analysis and gross energy content are shown in Table 1. The moisture level of the samples ranged from 6.73% to 9.52%. The moisture content of foods or its processed products gives an indication of its freshness and shelf life and high moisture content subjects food items to increased microbial spoilage, deterioration and short shelf life (Tressler *et al.*, 1980; Adepoju and Onasanya, 2008). The ash content can provide an estimate of the mineral content of a product. The values of the ash content of the samples range from 3.59% to 7.69% which may be an indication of high mineral content of the samples. The ash content of samples is similar

to a range of ash content values reported by Adeyi (2010) for some agricultural wastes. The result shows that the agriculture wastes are high in crude fibre (Mango endocarp, 49.47%; Melon seed husk, 51.61%; Moringa pod, 58.10%), comparably higher than coconut husk, cocoa pods, kola nut pod, plantain wastes flour reported by Adeyi (2010) and Okareh *et al.* (2015). High fibre content in diets have been reported to result in increased removal of carcinogens, potential mutagens, steroids, bile acids and xenobiotics by binding or absorbing to dietary fibre components and be rapidly excreted, hence these wastes will have health promoting benefits for the ruminants and non-ruminants (Ayoola and Adeyeye, 2009). Thus, the high fibre content of the samples may make them desirable to be used as components in feed formulation. The result showed that melon husk has the highest amount of crude protein (12.22%), moringa pod (7.4%) and mango endocarp (3.45%). The appreciable amount of crude protein of the samples, especially melon seed husk, indicates that they could be preferably used as source of protein for animal feed enrichment and or formulation. Protein is an essential component of diet needed for survival of animals and human beings, their basic function in nutrition is to supply adequate amount of required amino acids. Protein deficiency causes growth retardation, muscle wasting, oedema, abnormal swelling of the belly and collection of fluids in the body (Mounts, 2000). The proximate analysis results of melon seed husk obtained from this research work are in agreement with the study by Abdulrazaq *et al.* (2014) who reported a similar range of values. In livestock diets, energy is one of the most expensive nutritional components of feed formulation.

The amount of gross energy content is shown in Table 1 with melon husk having the highest energy content (245.69J). This may be due, in part, to the high lipid content of the samples as lipids have been shown to be concentrated energy source, inclusion of lipids is known to affect growth rate and feed efficiency (Brian *et al.*, 2015). The presence of appreciable quantity of energy in the samples in this study may make them fit for consideration in formulation of poultry and animal feeds in general as sources of energy.

Table 2 shows the mineral composition of the test samples. The results of mineral analysis showed the presence of calcium, Ca (6.14 mg/kg-243.95 mg/kg), phosphorus, P (24.25 mg/kg-106.70 mg/kg); Magnesium, Mg (13 mg/kg-61.44 mg/kg); iron, Fe (3.60 mg/kg-29.32 mg/kg); zinc, Zn (0.17 mg/kg-0.70mg/kg). The presence of cobalt, nickel and copper are negligible in all the samples. From the results obtained, moringa pod possess the highest amount of calcium and magnesium (243.95 mg/kg and 61.44 mg/kg respectively). The result also showed that melon husk possessed the highest amount of iron (21.26 mg/kg). The presence of these essential nutrients and minerals imply that the samples from this study could be utilized as sources of essential nutrients in feed ingredients for livestock. Minerals, which are essential nutrients are said to be present in small amounts in the body or in several parts per million (Gafar and Itodo, 2011). They are essential because each of them plays important role in metabolic processes of the body and their absence can cause deficiency symptoms in animals (McDonald *et al.*, 1995; Gafar and Itodo, 2011).

**Table 1:** Proximate composition of test samples and Gross energy content

Sample	Moisture content (g/100g)	Ash content (g/100g)	Crude Protein (g/100g)	Crude Lipid (g/100g)	Crude Fibre (g/100g)	Carbohydrate (g/100g)	Gross Energy (J)
Melon husk	8.49±0.04	3.59±0.05	12.22±0.25	20.09±0.28	51.61±0.27	4.00±0.23	245.69±4.40
Moringa pod	9.52±0.07	5.08±0.09	7.40±0.11	11.75±0.31	58.10±0.14	8.15±0.17	167.95±3.91
Mango endocarp	6.73±0.15	7.69±2.22	3.45±0.18	9.41±0.25	49.47±0.32	23.25±0.23	191.49±3.80

\*Data are mean values ± standard deviation (SD) of triplicate results

**Table 2:** Mineral composition of samples (mg/kg)

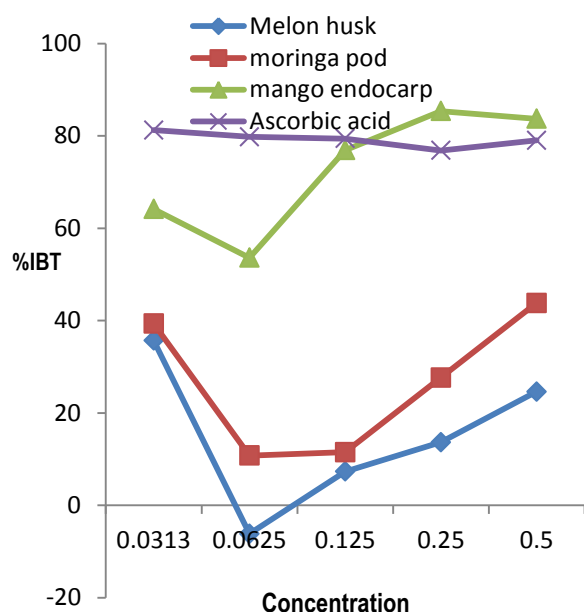
Sample	Iron	Magnesium	Calcium	Zinc	Copper	Manganese	Nickel	Phosphorus
Melon husk	21.28	52.26	14.94	0.7	0.16	1.15	0.00	106.70
Moringa pod	3.60	61.44	243.95	0.35	0.03	1.22	0.00	33.75
Mango endocarp	29.32	13.00	6.14	0.17	0.00	1.59	0.00	24.25

The essential mineral elements of nutritional importance are the macro (major) elements such as Ca, P, K, Na, and Mg. The micro (trace) elements are Fe, Zn, Cu, Mn, and Co (Ogbe and George, 2012).

Calcium and phosphorus are very important in the formation of strong bones and teeth, for growth, normal nerve and muscle action, blood clotting, heart function and cell metabolism (Roth and Townsend, 2003; Rolfe *et al.*, 2009). Some minerals are required for normal body growth, activities of muscles and skeletal development (such as calcium), cellular activity and oxygen transport (copper and iron), chemical reaction in the body and intestinal absorption (magnesium), as well as the regulation of acid-base balance (phosphorus). Iron is useful in prevention of anaemia and other related diseases (Oluyemi *et al.*, 2006). Manganese plays a role in energy production and in supporting the immune system (Muhammad *et al.*, 2011). It also works with vitamin K to support blood clotting, and with B complex vitamins to control the effects of stress (Muhammad *et al.*, 2011). Zinc is useful for protein synthesis, normal body development and recovery from illness (Muhammad *et al.*, 2011). Results from this study also show that the samples contain some of the antioxidant minerals such as Ca, Zn and Mn which are important for antioxidant enzymes *in vivo* and hence protect the body from diseases caused by oxidative stress such as cancer. This is in agreement with the report of Elsayed and Amany (2016). The result of macro elements of mango endocarp in this study is also in range with that reported by Elegbede *et al.* (1995) and Odunsi (2005).

Figure 1 shows the DPPH scavenging capacity of the samples. Mango endocarp displayed the highest antioxidant capacity (53.61%-85.37%);

Melon seed husk (-6.16%-35.65%); Moringa pod (10.79%-43.80%). The antioxidant assay was carried at a concentration range of 0.03125mg/ml-0.5mg/ml of the samples ethanolic extract. Almost all chronic diseases are associated with the danger of free radicals, which are also considered a cause of advanced aging (Mustafa *et al.*, 2010). The free radical scavenging assay is the most widely used analysis in evaluating antioxidant activity in phytochemistry. It is a simple and rapid test that is based on the reaction rate between a stable free radical, 2, 2-diphenyl-2-picrylhydrazyl (DPPH) and antioxidants. The availability of DPPH radicals commercially is another reason for its broad utilization (Paixão *et al.*, 2007). Historically, antioxidants have been added to commercial feeds to prevent lipid peroxidation and oxidative rancidity during production, processing and storage of feeds. More importantly, the current trend of formulating diets with polyunsaturated fatty acids (PUFAs)-rich ingredients has heightened the use of antioxidants in animal feeds. Indeed, PUFA-rich foods and feeds are highly susceptible to lipid peroxidation (Decker *et al.*, 2012). The samples in this study may not only serve as source of antioxidant for preservation of animal feeds but a natural source of antioxidants in keeping the livestock in good health, as natural antioxidants have been shown to possess broad spectra of biological actions, including pharmacological and therapeutic activity against free radicals and oxidative stress (Bagchi *et al.*, 2000). The result on mango endocarp antioxidant activity obtained in this study is in agreement with reports by Saito *et al.* (2008) and Abdullah *et al.* (2014) that mango seed endocarp possess powerful antioxidants of high antioxidant capacity. Mango endocarp was also shown to compete favourably with the standard (Ascorbic acid) used in the study.



IBT = Inhibition

**Figure 1:** DPPH free radical scavenging activities of Melon husk, Moringa pod and Mango endocarp

The result of the antinutrient screening of the samples is shown in Table 3. Antinutrients are compounds that limit the wide use of many plants due to their ubiquitous occurrence. They are compounds capable of eliciting deleterious effect in man and animals (Kubmarawa *et al.*, 2008). The presence of these anti nutrients, especially in high quantity reduces the bioavailability of nutrients in the food and plants (Akindahunsi and Salawu, 2005). Oxalate for example tends to render calcium unavailable by binding to the calcium ion to form complexes (calcium oxalate crystals). These oxalate crystals formed prevents the absorption and utilization of calcium. The calcium crystals may also precipitate around the renal tubules thereby causing renal stones (Ladeji *et al.*, 2004). The amount of oxalate in the samples was negligible.

Tannin was not detected in melon husk and moringa pod, while mango endocarp contained 0.52% tannin (Table 3). Tannins are plant polyphenols, which have ability to form complexes with metal ions and with macromolecules such as protein and polysaccharides (Dei *et al.*, 2007). Excess of consumptions of tannins in human could be toxic (Ferreira *et al.*, 2008). This is because tannins are metal ions-

chelators and tannin-chelated metal ions are not bioavailable hence could decrease the bioavailability of iron leading to anaemia.

The saponin content of melon husk (0.014%), moringa pod (0.014%) is negligible while it was not detected in mango endocarp. Saponins are glucosides, which includes steroid saponins and triterpenoid saponins (Dei *et al.*, 2007). Saponin in excess could cause hypocholesterolemia because it binds cholesterol making it unavailable against red blood cell (Khahlil and Eladomy, 1994). Saponin-protein complex formation can reduce protein digestibility (Shimoyamada *et al.*, 1998). The presence of saponin in the body can help to fight infection and microbial inversion (Sodipo *et al.*, 2000). High levels of saponins in feed affect feed intake and growth rate in poultry (Dei *et al.*, 2007).

**Table 3:** Phytochemical/Antinutrient activity of Melon husk, Moringa pod and Mango endocarp (%)

Constituents	Melon husk	Moringa pod	Mango endocarp
Alkaloids	0.26	0.17	0.25
Flavonoids	0.38	0.38	0.33
Tannins	ND	ND	0.52
Saponin	0.01	0.01	ND
Phenols	ND	ND	0.09
Oxalate (10 <sup>-6</sup> )	6.55	6.55	6.55

ND- Not detected

Alkaloid content of the samples is: melon husk (0.26%), moringa pod (0.17%) and mango endocarp (0.25%). Despite the medical uses of alkaloids, they cause gastrointestinal upsets and neurological disorders (Ogbuagu, 2008).

The antinutrient components of the samples in this study were very low in concentration; this is in agreement with report of antinutrient content of melon husk by Ogbu and George (2012). The presence of nutrients, such as protein, fibre and lipid, in appreciable quantity in the samples in this study implies they could be utilized as feed ingredient in poultry diets. Nutrients are known to improve the performance and health of birds. Nutrients are

required for proper bone development and improved eggs quality (Ogbe and George, 2012).

## CONCLUSION

The results of the samples analysed in this study indicate that the agricultural wastes could serve as potential ingredient in formulation of feedstock for animal feed. This is shown in the high level of fibre, considerable quantity of crude protein, carbohydrate, crude fibre, ash content, crude lipid and low levels of antinutrients factors. The results also showed that the samples contain some of the antioxidant minerals such as Ca, Zn and Mn which are important for antioxidant enzymes *in vivo* and hence could protect the body from diseases caused by oxidative stress.

Thus, rather than been allowed to constitute environmental pollutants, they (melon husk, mango endocarp and moringa pod) may be used as nutritive components in formulation of animal feeds.

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