

## THE PROXIMATE, MINERAL AND PHYTOCHEMICAL ANALYSIS OF THE LEAVES OF *Ocimum gratissimum* L., *Melanthera scandens* A. and *Leea guineensis* L. AND THEIR MEDICINAL VALUE

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**ABSTRACT :** The chemical composition of *Ocimum gratissimum*, *Melanthera scandens* and *Leea guineensis* were investigated. The proximate analysis in % showed that *Melanthera scandens* had the highest amount of ash content of 7.73 and moisture content 6.54 while *Ocimum gratissimum* had the lowest amount of ash content 5.11 and moisture content 5.04. *Ocimum gratissimum* had the highest amount of fat 7.75 and carbohydrate 56.16 while *Melanthera scandens* had the least amount of fat 6.87 and carbohydrate 50.0. *Leea guineensis* had the highest amount of crude protein 19.3 while *Melanthera scandens* had the highest amount of crude fibre 12.66. The mineral analysis in mg/100g indicated that the leaves contained calcium, sodium, potassium, magnesium, zinc, iron, manganese, and phosphorus. The Phytochemical analysis of the plants showed that the three medicinal plants contained alkaloid, tannins, saponins, steroid, phlobatanin/ terpenoid, flavonoid cardiac glycoside, while phlobatanin was not found in *Melanthera scandens*. The medicinal plants also contained antinutrient phytin phosphorus, oxalate, phytic acid and polyphenol.

**Keywords:** Proximate, Polyphenol, Phytopathogenic fungi, Plant extract, Radial mycelia growth.

### INTRODUCTION

Medicinal plants are plants which contain substances that could be used for therapeutic purposes or which are precursors for the synthesis of useful drugs (Sofowora, 2008). The medicinal value of these plants lies in bioactive Phytochemical constituents that produce definite physiological action on the human body (Akinmoladun *et al.*, 2007). *Ocimum gratissimum* belongs to the family Lamiaceae and found mostly in the tropical countries including: Nigeria, India, North and South America, Mexico and Brazil where it is popularly known as alfavaca-cravo, alfavacao, alfavaca (Brenan, 1996). The local names are Efinrin, Efinrin aaja, Erumaba (Yoruba), Daidoyatagida (Hausa), Esewon (Edo-Akoko), Nehonwu, Nchanwu (Igbo) and Menthesauvage (French). It is traditionally used to relief pains and also used in the treatment of rheumatism, diarrhea, high fever, convulsions, diabetes, eczema, piles and as a repellent (Chitwood, 2003, Hotlets *et al.*, 2003, Pessoa *et al.*, 2003). The decoction of the stem is inhaled for the treatment of catarrh and bronchitis (Gills, 1992).

*Melanthera scandens* belongs to the plant family, Asteraceae and found mostly in the tropical regions including Nigeria, Mexico, Central America, South America, Sub-saharan Africa (Brenan, 1996). The common local names are Aboyunrinyun and Ounje-ehoro (Yoruba), vine (English). The herbalists use the juice of leaves to stop dysentery. The plant is popularly used as further for animals (Gills, 1992). It is used in the treatment of skin infections, gastroenteritis, stomachache, cuts, wounds, inflammation, diuretic and also for hypertensive activities. It is also used against piles, diarrhea and hemorrhoid (Omotayo, 2007). *Leea guineensis* is an evergreen shrub, up to 20ft high locally abundant in moist shaded places which belongs to the family Leaceae (Hutchinson *et al.*, 1968). It has an English name called Red tree vine or Hansid-hapan. The local names are Kojiya, arigbokuta and Iya-kekere. The aqueous Methanolic extract of *Leea guineensis* exhibits potential in-vivo anti-tumor and antioxidant activity (Falodun *et al.*, 2007). It is used in the treatment of enlarged spleen in children, pregnancy detection, purgative, toothache, gonorrhea, general weakness, skin lesions, skin rash, ulcer, herpes and boils. The plant is fungistatic and bacteriostatic (Gills, 1992).

The aim and objectives of this study was to know the constituent of the leaves of *O. gratissimum*, *M. scandens* and *L. guineensis* through proximate, mineral and phytochemical analysis.

## MATERIALS AND METHODS

### Collection of Plant materials

The fresh plants were collected from a local farm in Ado-Ekiti, Ekiti State, Nigeria. Identification and authentication were carried out in the herbarium section of the Department of Plant Science, Ekiti State University, Ado-Ekiti, Ekiti State, Nigeria.

### Processing of Plant Materials

The fresh leaves of the following plants *O. gratissimum*, *M. scandens* and *L. guineensis* were air dried at 28°C for 30 days. They were grounded into fine powder using an electric blender and stored in a cool dry container until use.

**Phytochemical analysis:** Quantitative phytochemical screening to determine the presence of alkaloids, tannins, saponins, steroids, phlobatannin, terpenoids, flavonoid and cardiac glycosides using standard methods as described by Trease and Evans (1985), Harbone (1984) and Sofowora (2008) were carried out.

**Proximate analysis:** The proximate analysis of the samples for moisture, ash, fibre and fat were done by the method of AOAC (2005). The nitrogen was determined by micro-Kjeldahl method as described by Pearson (1976) the percentage Nitrogen was converted to crude protein by multiplying 6.25. All determinations were performed in triplicates.

**Mineral analysis:** The minerals of the samples were analyzed using the solution obtained by dry ashing the sample at 550°C and dissolving it in HCl (25ml) and 5% Lanthanum chloride (2ml), boiling, filtering and making up to standard volume with deionized water. Mn, Cu, Co, Zn, Fe, Mg, Na, and Ca were determined with a Buck Atomic Absorption Spectrometer (Buck Scientific, Model 200A/200, Inc. East Norwalk, Connecticut, U.S.A). Sodium was measured with a Corning 405 flame photometer (Corning Halstead, Essex, UK, Model 405) (AOAC, 2005). The detection limits had previously been determined using the methods of Techtron (1975) as Mn 0.01, Cu 0.005, Co 0.05, Zn 0.005, Fe 0.02, Mg 0.002, Ca 0.04, Na 0.001, ppm (all for aqueous solutions).

The optimum analytical range was 0.5 to 10 absorbance units with coefficient of variation of 0.05 to 0.40% phosphovanado-molybdate method using a Spectronic 20 colorimeter (Gallenkamp, London, UK) (AOAC, 2005). All chemicals were BDH analytical grade.

## RESULTS AND DISCUSSION

The results of proximate analysis (in %) of *O. gratissimum*, *M. scandens* and *L. guineensis* leaves are shown on Table 1. The plants contained higher amount of carbohydrates content which were 56.16, 50.08 and 50.71 respectively. These results are similar to that reported for *A. sativum* (57.28) (Hussain et al., 2009) but are higher than that of *Senna obtusifolia* (23.70) and *Amaranthus incurvatus* (39.05) (Faruq et al., 2002). It is however lower than the value reported for *P. fistulosus* (62.39) (Hussain et al., 2009). Carbohydrate constitutes a major class of naturally occurring organic compounds which are essential for the maintenance of life in plant and animals and also provide raw materials for many industries (Ebun-Oluwa and Alade, 2007). The plant is a good source of carbohydrate when consumed because it meets the Recommended Dietary Allowance (RDA) values (FND, 2002).

**Table 1: Results of proximate analysis of three Medicinal plants (Percentage of dry samples)**

TEST	A	B	C
Ash content	5.11	7.73	7.43
Moisture content	5.04	6.54	5.69
Crude protein	14.6	16.19	19.3
Fat	7.57	6.81	7.28
Crude fibre	11.38	12.66	9.61
Carbohydrate	56.16	50.08	50.7

Key : A = *Ocimum gratissimum*

B = *Melanthera scandens*

C = *Lea guineensis*

The crude protein content (%) of *O. gratissimum*, *M. scandens* and *L. guineensis* were 14.60, 16.19 and 19.30 respectively. These are higher than the protein content of *Telfaria occidentalis* (7.00) and *Momordica balsania* L. (11.29) (Isong and Idong, 1997), but however lower than those of *Piper guineensis* (29.78) and *Talinum. triangulare* (31.00) (Akindahunsi and Salawu, 2005). However, it compared favourably with the value reported for *A. viridus* (16.41) and *S. oleraceae* (23.74) (Pandey et al., 2006). The plants are considered as a good source of protein because it provides more than 12% of calorific value from protein (Pearson, 1976).

The ash content (in %) of *O. gratissimum*, *M. scandens* and *L. guineensis* leaves were 5.11, 7.73 and 7.43 which is lower than the values reported for the leaves of *A. viridus* 22.84 (Pandey et al., 2006), *Ipomea batatas* with 11.10 and *Moringa deifera* with 15.09 (Antia et al., 2006). They are however higher than that of *A. sativum* with 4.84 (Hussain et al., 2009). The ash content is a reflection of the amount of mineral elements present in the samples; therefore, the plants contained a good amount of minerals.

The moisture content (in %) values for the leaves of *O. gratissimum* (5.04), *M. scandens* (6.54) and *L. guineensis* (5.69) were relatively low, therefore it would hinder the growth of micro organisms and life span of stored samples would be high. This is good for the long preservation and will prevent early spoilage. The moisture content of the plant is low when compared to that of *Xylophia aethiopia* (16.04) (Abolaji et al., 2007) and *Acalypha hispida* (11.91) Iniaghe et al., (2009).

The values of the crude fat (in %) for the leaves of *O. garanissimu*, *M. scandens* and *L. guineeensis* were 7.57, 6.81 and 7.28 respectively which were moderate in amount when compared to those of *Talinum triangulare* (5.09), *Amarantus hybridus* (4.80) (Akindahunsi and Salawu 2005) and *Gnetum africanum* (3.15) (Abolaji et al., 2007). Dietary fat increases the palatability of food by absorbing and retaining flavour (Antia et al., 2006). A diet providing 1.20% of its caloric of energy as fat is said to be deficient for human being as excess fat consumption is implicated in certain cardiovascular disorders (Antia et al., 2006)

The crude fibre values (in %) for the leaves of *O. gratissimum*, *M. scandens* and *L. guinersis* were 11.38, 12.66 and 9.61 which are low when compared to that of *A. esculentus* with 14.71, *M. charantia* with 16.62 (Hussain et al., 2009) *P. thonningii* with 35.03 (Ene-Obong and Carnovale, 1992) but are higher than that of *Gnetum africanum* (4.60), *M. ureans* (4.00) and *Parinari polyandra* (Ekpo, 2007). The plants are good source of crude fibre when consumed because adequate intake of dietary fibre can lower the serum cholesterol level, heart disease, hypertension, constipation, diabetes and breast cancer (Ishida et al., 2000).

The mineral composition (in mg/100g) of *O. grantissimum*, *M. scandens* and *L. guineensis* leaves were shown in Table 2. The values of sodium in the plants varied from 26.94 (*M. scandens*) to 39.38 (*O. gratissimum*) while that of potassium varied from 32.21 (*L. guineensis*) to 47.01 (*O. granitissimum*). The ratio of sodium to potassium is less than 1 (0.8); therefore consumption of the plants would reduce high blood pressure disease because Na:K is less than one as recommended by FND (2002).

**Table 2: Results of Mineral analysis of the three medicinal plants (mg/100g)**

TEST	A	B	C
Sodium	39.38	26.94	31.51
Potassium	47.01	33.70	31.21
Calcium	51.44	39.98	36.29
Magnesium	31.84	33.60	28.68
Zinc	44.47	39.49	30.18
Iron	10.93	8,88	5.08
Lead	-	-	-
Manganese	1.04	0.77	1.22
Copper	-	-	-
Phosphorus	68.31	27.40	35.53

Key : A = *Ocimum gratissimum*

B = *Melanthera scandens*

C = *Lea guineensis*

The value of calcium and phosphorous in the leaves of *O. gratissimum*, *M. scandens* and *L. guineensis* varied from 36.29 (*L. guineensis*) to 51.44 (*O. gratissimum*) for calcium and from 27.40 (*M. scandens*) to 68.31 (*O. gratissimum*) for phosphorous. Calcium and phosphorous are associated with each other for growth and maintenance of bones, teeth and muscles (Okaka et al., 2006). The calcium level in the leaves studied compared favourably with the values reported in some green leafy vegetables consumed in Nigeria and some wild edible leaves grown in Eastern Amatoria, Turkey (Ladan et al., 1996). The phosphorous content compared favorably with that of *Ipomea batatas* with 37.28 (Antia et al., 2006) but low for *M. scandens*. Therefore, *O. gratissimum*, *M. scandens* and *L. guineensis* are good sources of calcium and phosphorous which aids intestinal absorption because the ratio of Ca: P in leaves was (0.6) close to unity (Gull-Guerrero et al., 1998).

Magnesium content of *O. gratissimum*, *M. scandens* and *L. guineensis* varied from 28.68 (*L. guineensis*) to 33.60 (*M. scandens*). These are high when compared to *Xylopiya aethiopia* (2.42) (Abolaji et al., 2007). Magnesium is a composition of chlorophyll and it is an important content in connection with Ischemic heart disease and calcium metabolism in bones (Ishida et al., 2000).

Zinc content of *O. gratissimum*, *M. scandens* and *L. guineensis* varied from 30.18 (*L. guineensis*) to 44.47 (*O. gratissimum*). These are low when compared to the mineral analyzed for in *Pilostigma thioningi* (70.10) (Elegbede, 1998). Zinc is involved in normal functioning of immune system (Ibrahim et al., 2001) and is associated with protein metabolism. The leaves are a good source of zinc because it is far above 6.23 recommended by RDA (Borgert et al., 1975).

Iron content of *O. gratissimum*, *M. scandens* and *L. guineensis* varied from 5.08 (*L. guineensis*) to 10.93 (*O. gratissimum*). These values compared favourably with the values reported for *Ipomea batata* 16.00 (Antia et al., 2006) but low when compared to the values of other green leafy vegetables as reported by Ibrahim et al., (2001). Iron is an essential trace element for haemoglobin formation, normal functioning of central nervous system and in the oxidation of carbohydrates, protein and fats (Adeleye and Otokiti, 1999). This perhaps justifies the already locally established function of the plant in the regulation of haemoglobin level.

The values of manganese in the leaves of *O. gratissimum*, *M. scandens* and *L. guineensis* varied from 0.77 (*M. scandens*) to 1.22 (*L. guineensis*). This suggest that *O. gratissimum* does not contribute or rather cannot be used as a substitute for other blood forming leafy vegetables while *M. scandens* and *L. guineensis* are involved in the boosting of the immune system and are antioxidant micronutrient (Talwar et al., 1989).

Copper and Lead were absent in *O. gratissimum*, *M. scandens* and *L. guineensis* and therefore has also been reported that lead and copper are highly toxic even at low concentrations (Asaolu et al., 1997).

The result of phytochemical analysis of the leaves of *O. gratissimum*, *M. scandens* and *L. guineensis* are shown in Table 3.

**Table 3: Results of Phytochemical analysis of the three medicinal plants**

TEST	A	B	C
Alkaloid	+	+	+
Tannin	+	+	+
Saponin	+	+	+
Steroid	±	+	+
Phlobatannin	+	-	+
Terpenoid	+	+	+
Flavonoid	+	+	+
Cardiacglycoside	+	+	+

+ = Presence of constituents

• = Absence of constituents

± = Slightly present

The three medicinal plants contained alkaloids, tannis, saponins, steroids, phlobatannin terpenoids, flavonoids and cardiac glycosides while steroid was slightly present in *O. gratissimum* and phlobatannin but was absent in *Melanthera scandens*. Alkaloids has been found to have microbiocidal effect and the major anti-diarrheal effect is probably due to their effects on small intestine and antihypertensive antifungal, anti-inflammatory, antifibrogenic effect (Ghosal et al., 1996). However, the result of this work is similar to the findings of Awoyinka et al., (2007) who reported the presence of alkaloid in *Cnidioscolus aconitifolius*. Some alkaloids are useful against HIV infection as well as intestinal infection associated with AIDS (McDevith et al., 1996). The presence of alkaloids in the three medicinal plants make them recommendable for patient as alkaloids posses a significant pharmacological property.

Tannin is non toxic and can generate physiological responses in animals that consume them (Scalbert, 1991). Tannin can be toxic to filamentous fungi, yeast and bacterial. The presence of tannin in the medicinal plant suggests the ability of these plants to play major roles as antifungal antidiarrheal, antioxidant and antihemorrhoidal agent (Asquith and Butter, 1986).



Saponins showed a positive result in the leaves of *O. gratissimum*, *M. scandens* and *L. guineensis*. This compound has been reported to have antihyper-cholesterol, anti-inflammatory, cardiac depressant properties (Trease and Evans 1985) and appear to kill or inhibit cancer cells without killing the normal cells in the process (Lewis and Elvin-Lewis, 1995).

Steroid also showed a positive result in the leaves of *O. gratissimum*, *M. scandens* and *L. guineensis* which are of importance and interest in pharmacy due to their relationship with such compounds as sex hormones (Okwu, 2001) and promote immune function in the skin and also reduce inflammation (Bell, 2008). Iniaghe et al., (2009) reported the presence of steroids in *Acalypha hispida*.

Phlobatanin were detected in the three medicinal plants. The report of this work is similar to the findings of Iniaghe et al., (2009) who reported the presence of phlobatannin in *A. hispida* and *A. racemosa*. This compound inhibit the growth of many microorganisms like fungi, yeast, bacteria and viruses (Scalbert, 1991).

The *O. gratissimum*, *M. scandens* and *L. guineensis* contained terpenoid. Terpenoids called petalostemumol, showed excellent activity against *Bacillus subtilis* and *Staphylococcus aureus* and lesser activity against gram negative bacteria as well as *Candida albicans* (Hufford et al., 1993).

In this study, the leaves of *O. gratissimum*, *M. scandens* and *L. guineensis* contained flavonoid. It modifies the body's reaction to allergens, virus and caranogens. It has been reported to show anti inflammatory and antimicrobial activity (Cushnie and Lamb, 2005). Akubugwo et al., (2007) reported the presence of flavonoids in *A. hybridus*.

Cardiac glycoside showed positive result in the leaves *O. gratissimum*, *M. scandens*, *L. guineensis*. The cardiac glycoside has been used for over two centuries as stimulant in cases of cardiac failure and diseases (Trease and Evans, 1978; Olayinka et al., 1992). This perhaps justifies the already locally established function of the plant in the treatment and management of hypertension (Taiwo et al., 2009).

The polyphenol content of the three medicinal plants were 2.22%, 1.96% and 1.55% respectively shown in

Table 4. It is considered bacteriostatic against both fungi (Duke, 1985) and bacteria (Thomson, 1978).

The values of oxalate content for the leaves of *O. gratissimum*, *M. scandens* and *L. guineensis* were 2.21mg/g, 1.53mg/g and 1.98mg/g respectively but are higher than *Musa sapientum* with 0.72mg/g (Baiyeri, 2000), *Butyrospermum parkii* with 1.5mg/g (Taiwo et al., 2009), *Spondias mombin* with 0.9mg/g (Igwe et al., 2009) and lower than the reported values for cotton leaves .22mg/g (Taiwo et al., 2009). High oxalate level in food may reduce the bio availability of such metal as calcium.

**Table 4: Results of anti nutrient analysis of the three medicinal plants**

TEST	A	B	C
Phytin phosphorus (mg/g)	2.90	2.44	3.71
Oxalate content (mg/g)	2.21	1.53	1.93
Phytic acid content (mg/g)	10.29	8.65	13.18
Tannic acid (%)	6.19	5.34	5.81
Saponin (%)	2.14	1.57	2.25
Alkaloid (%)	6.19	5.34	5.81
Flavonoid (%)	0.65	0.55	0.36
Polyphenol (%)	2.22	1.96	1.55

Key : A = *Ocimum gratissimum*

B = *Melanthera scandens*

C = *Lea guineensis*

The phytic acid content of *O. gratissimum*, *M. scandens* and *L. guineensis* were 10.29mg/g, 8.65mg/g and 13.18mg/g respectively which are high when compared to the values reported for *Dacryodes edulis* with 4.00mg/g (Ibanga and okon, 2009). Bambara groundnut with 0.29mg/g, pigeon pea with 0.2mg/g (Igbedioh et al., 1994)

The phytin phosphorus values for the leaves of *O. gratissimum*, *M. scandens* and *L. guineensis* were 2.9mg/g, 2.44mg/g and 3.71mg/g respectively which are low compared to the values reported for *Zingiber officinale* 28.83mg/g (Aletor and Omodara, 1994) but higher than wild yam tubers with 1.7mg/g (Adeniji et al., 2007). The phytic acid intake of 4.9mg/g is said to decrease Iron absorption by 4.5 folds in humans. Therefore, these medicinal plants can be consumed because the levels of antinutrient do not reach lethal dosages. (A.O.A.C, 2005).

It is established that only high content of these antinutrients prevent the absorption of mineral like iron, magnesium, potassium and calcium which are essential for metabolism in the body. Reduction of antinutrients in foods may be necessary especially when their levels are higher than those generally regarded as safe for human consumption. This can be accomplished through different hydrothermal treatments, which also enhances the nutritional qualities: increase palatability and digestibility of foods (Adeniji *et al.*, 2007).

## CONCLUSION

Plants have contributed immensely to the medical field. It has been the source of most drugs used for combating infections. The three plants used in this study were found to contain the important constituent needed to combat various kinds of infection in human.

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