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*Full Length Research Paper*

# Phytochemical and nutrient compositions of the leaves of *Ocimum canum* Sims

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***Ocimum canum* Sims is an underutilized medicinal plant that grows in south-western Nigeria. The leaf is used for the treatment of gastrointestinal problems and also for the preparation of local soups. The phytochemical and nutrient compositions of its leaves were investigated using established analytical procedures. The phytochemical analysis of the plant revealed high concentrations of flavonoids (10.00%), saponins and tannins, but low levels of phenolics and alkaloids. The leaves showed high carbohydrate content (639.6 g/kg), ash, crude fat and crude fiber, but very low in protein content. The study reveals high concentration of calcium (50.72 g/kg) with appreciable levels of potassium, sodium, phosphorous and magnesium. In addition, the plant was found to be a good source of iron, zinc and manganese. Furthermore, the concentrations of cadmium (0.01 g/kg) and lead (0.02 g/kg), which are toxic metals were very low, while the vitamin C content of the leaves was found to be high (0.05 g/kg). The result of this study therefore revealed that the leaves of *O. canum* are a good source of phytochemicals and nutrients that can be harnessed to combat nutritional deficiencies, especially in the rural communities.**

**Key words:** *Ocimum canum*, phytochemicals, proximate, mineral and nutrient composition.

## INTRODUCTION

The increase in human populations, high prices of food items and increasing rate of poverty are the major causes of food insecurity in most developing countries of the world today (Weaver, 1994). The Food and Agricultural Organization of the United Nations has suggested a 4% increase in the production of nutritious food yearly, as a means of combating food scarcity for the world's projected population (FAO, 2008). This has necessitated the need to incorporate non convectional food plants (wild) into human diets as a means of alleviating nutritional deficiencies especially in rural communities. Studies have revealed that many wild plants posses high minerals, vitamins, fiber and phytochemical contents that make them nutritionally important (Tukan et al., 1998; Roja and Rao, 2000). However, many of these plants are underutilized today because of the inadequate scientific knowledge of their nutritional potentials. One of such plants is *Ocimum canum.*

*Ocimum canum* Sims is a semi-woody plant of about 40 cm high that belongs to the family of Lamiaceae. It is an aromatic plant and native to tropical Africa (Steel, 2006). It is popularly called African basil, but known as "Efinrin elewe dudu" in south-western Nigeria where it is used locally for the treatment of gastrointestinal problems, piles, dysentery, as an infusion for nursing mothers and as insect repellant. The leaf is used for the treatment of diabetes in Ghana (Hogarh, 1996; Nyarko et al., 2002). The plant is reported to be rich in volatile essential oils of therapeutic importance (Ekundayo et al., 1989). The leaves of *O. canum* are used in the preparation of delicious local soups as well as flavoring agent in yam and cocoyam porridges in the Yoruba tribe of Nigeria. The plant is also used as a local condiment because of its aromatic properties (Bassole et al., 2005). Despite its nutritive and therapeutic values, this plant has not been scientifically examined for its nutritive components.

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This study is therefore aimed at providing information on the phytochemical compositions of *O. canum* leaves in an attempt to justify and validate its suitability as edible vegetable and as an herbal drug for its various acclaimed pharmacological properties, and to also provide information on the proximate, macro and micro nutrient compositions of the leaves of the plant.

## MATERIALS AND METHODS

### Collection and identification of plant materials

The leaves of *O. canum* were collected on May 2011, from a local farmland near Orin Ekiti, Nigeria. The plant was identified and authenticated by Mr Omotayo (herbarium curator) at the Department of Plant Science, University of Ado Ekiti, Nigeria. A voucher specimen (Aluko 09) was deposited in the herbarium of the Department. The leaves were air dried for 10 days and pulverized into fine powder using an electric blender. One hundred and fifty grams (150 g) of the powdered sample was used for the phytochemical, proximate, mineral and vitamin C analysis.

### Chemicals

Ammonium hydroxide, amyl alcohol, ethanol, butanol, sodium chloride, acetic acid, methanol, Folin-Denis reagent, sodium carbonate, tannic acid, diethyl ether, nitric acid, ascorbic acid and hydrochloric acid were purchased from Merck USA. All chemicals used in this study were of analytical grade.

### Phytochemical screening

The qualitative determination of phytochemical constituents of the *O. canum* leaves was carried out using the method of Parekh and Chanda (2009), while the quantitative analysis of phenolics, saponins, alkaloids, flavonoids and tannins were done in triplicates as follows.

### Determination of total phenolics

Briefly, 2.5 g of the powdered leaves was boiled with 25 ml of ether for 15 min. Then 5 ml of the extract was pipette into a 50 ml volumetric flask and 10 ml of distilled water, 2 ml ammonium hydroxide and 5 ml amyl alcohol were added sequentially. The solution was made up to the mark with distilled water and allowed to stand for 30 min for color development. The absorbance of the solution was read at 505 nm using a UV spectrophotometer and tannic acid was used as the standard (Obadoni and Ochuko, 2001).

### Determination of saponin content

The determination of saponin was done using the method of Obadoni and Ochuko (2001). Five grams of powdered plant sample was dispersed in 50 ml of ethanol prepared in distilled water (20% v/v). The mixture was heated over a hot water bath for 4 h at 55°C with continuous stirring. The residue collected after filtration was re-extracted with 50 ml ethanol (20%) and reduced to 20 ml over a boiling water bath. This was shaken with 10 ml diethyl ether in a separating funnel. The aqueous layer was collected and the process was repeated. Then, 20 ml of butanol was added to the filtrate and washed with 10 ml 5% w/v aqueous sodium chloride.

The whole mixture was evaporated to dryness on a hot water bath and later oven-dried to constant weight at 40°C.

### Determination of alkaloids

A volume of 200 ml of 10% acetic acid in ethanol was added to 5 g of the powered leaves. This was covered with a watch glass and allowed to stand for 4 h. It was then filtered and the filtrate was concentrated to ¼ of the original volume on a water bath. Concentrated ammonium hydroxide was added drop wise for complete precipitation and the solution was allowed to settle. The collected precipitates were washed with dilute ammonium hydroxide and then filtered. Finally, the residue was dried and weighed (Edeoga et al., 2005).

### Determination of flavonoids

Ten grams of the powdered sample was extracted repeatedly with 100 ml of 80% aqueous methanol. The solution was filtered and the filtrate was evaporated to dryness over a hot water bath (Obadoni and Ochuko, 2001).

### Determination of tannins

The determination of tannin was done using the method of Edeoga (2005) with some modifications. One gram of the ground leaves was added to 40 ml of 50% methanol. The mixture was shaken vigorously and placed in a hot water bath at 80°C for 1 h. The extract was filtered into a 100 ml volumetric flask, then 20 ml distilled water, 2.5 ml Folin-Denis reagent and 10 ml of 17% sodium carbonate was added and shaken vigorously. The mixture was then made up to the mark with distilled water and allowed to stand for 20 min for full color development. The absorbance was read at 760 nm on a UV spectrophotometer and tannic acid of concentrations ranging from 0- 10 ppm was prepared and used as standard. A standard graph was plotted and the calibration curve $Y = 0.0593x – 0.0485$, $R^2 = 0.9826$ was obtained, where x was the absorbance and Y was tannic acid equivalent.

### Proximate analysis

The proximate analysis of the dried leaves was carried out using a modified method of Antia et al. (2006). The crude fat was determined by exhaustive extraction of 5 g of the powdered leaves with 100 ml of diethyl ether twice. The solvent was removed from the sample by filtration and the residue was taken as the lipid free sample. The ash content was determined by the incineration of 5 g of the sample in a muffle furnace (Naber Industrieofenbau, Bremen, Germany) at 550°C for 12 h. Determination of crude fiber involved the digestion of the sample with 100 ml of 1.25% (v/v) sulphuric acid and 100 ml of 1.25% (w/v) sodium hydroxide. This was followed by incineration in muffle furnace.

The fiber content was calculated from the loss of weight on ignition. The digestion of the sample for the determination of protein content was done using the micro Kjeldahl method of AOAC (1990). The total nitrogen content in the leaves was determined according to the colorimetric method described by Okalebo et al. (2002). The value of nitrogen was multiplied by a conversion factor (6.25) to give the amount of protein present in the leaves. The carbohydrate content was calculated by subtracting the sum of the ash, fiber, fat and protein from 100. The values for all parameters were reported in g/kg of the whole sample. Each experiment was replicated three times.

**Table 1.** The phytochemical constituents of *Ocimum canum* leaves.

| Phytochemical | Composition (%) |
|---|---|
| Phenolics | 0.40 ±0.04 |
| Saponins | 6.00 ± 0.71 |
| Alkaloids | 2.00 ± 0.25 |
| Flavonoids | 10.00 ±0.02 |
| Tannins | 4.73 ± 0.01 |
| Terpenoids | Not detected |
| Steroids | Not detected |
| Cardiac glycosides | Not detected |

Data are expressed as mean ± SD.

### Mineral analysis

The powdered *O. canum* leaves (0.5 g) was digested in a reaction mixture of 9 ml of 55% (v/v) nitric acid and 3 ml of 32% (v/v) hydrochloric acid for 60 min in a microwave digester. The mixture was allowed to cool and was made up to 50 ml with distilled water in a volumetric flask. The digested sample was analyzed for macro and micro elements using the Inductively Coupled Plasma-Atomic Emission Spectrometry (ICP-OES, Varian 710 –ES series). The instrument was set at the following working conditions: power 1.00 KW, plasma flow 15.0 L/min, auxiliary flow 1.50 L/min, nebulizer flow 200 KPa, sample uptake delay 30 s, rinse rate 10 s, pump rate 15 rpm, replicate read time 5.00 s, instrument stabilizing delay 15 s. The phosphorus content was analyzed on the flow analyzer. The experiment was done in triplicates.

### Determination of vitamin C content

The vitamin C content of the leaves was determined by iodine titration method of Suntornsuk et al. (2002). Briefly, freshly prepared iodine solution was titrated against 10 ml of aqueous extract (1 mg/ml) of the dry *O. canum* leaves and various concentrations of ascorbic acid (0.02- 1 mg/ml) using 10 drops of 1% starch as an indicator. Ascorbic acid was used as the standard. The experiment was replicated three times.

## RESULTS AND DISCUSSION

The result of the phytochemical screening of *O. canum* leaves revealed the presence of phenolics, saponins, alkaloids, flavonoids and tannins (Table 1). The phytochemicals in medicinal plants have been reported to be the active principles responsible for the pharmacological potentials of medicinal plants (Edeoga et al., 2005). The presence of these chemicals in the leaves of *O. canum* justifies the local use of this plant for the treatment of various ailments. The leaves are rich in flavonoids, saponins and tannins, with considerable amount of phenolics and alkaloids. Flavonoids are polyphenolic compounds that are biologically active against liver toxins, microorganisms, inflammation, tumor and free radicals (Okwu, 2004). They have also been reported to inhibit the growth of cataracts in diabetic patients (Okwu and Omodamiro, 2005). The high level of flavonoids (10.00 %) in the leaves might be responsible for the use of the plant by traditional healers to treat diabetes.

Saponins are natural glycosides that act as hypoglycemic, antifungal and serum cholesterol lowering agents in animals (Sapna et al., 2009). They are essential elements in ensuring hormonal balance and synthesis of sex hormones (Okwu, 2003). The saponin content (6.00%) of *O. canum* leaves is considerably high. This substantiates its use as a local condiment for the nursing mothers in some tribes in Nigeria. Tannins are bitter polyphenolic compounds that hasten the healing of wounds. They also possess anti-diuretic and anti-diarrhea properties (Okwu, 2004). The significant amount of tannins (4.73 %) in the leaves of *O. canum* might be responsible for its use by the local herbalists to treat gastrointestinal disorders. Conversely, condensed tannins can inhibit herbivore digestion by binding to consumed proteins, thereby making it indigestible for animals. Its concentration in the leaves might be the reason why animals do not graze on this plant (Edeoga et al., 2006). However, to neutralize this effect, it is necessary for humans to cook the leaves before consumption. Phenolic compounds are potent antioxidants and free radical scavengers with inhibitory activities against some pathogenic microorganisms (Khoobchandani et al., 2010). The concentration of phenolics in the leaves of *O. canum* (0.40%) as observed in this study was higher than that of *Ocimum gratissimum* (0.049%*) and Hyptis suaveolens* (0.05%) as reported by Edeoga et al. (2006). *O. canum* is therefore likely to be a better source of antioxidants than some other wild vegetables. Alkaloids are chemicals which help plants to repel some predators. The concentration of alkaloid in the leaves as shown in this study (2.00 %) could contribute to the use of the leaves as an insect repellant.

The proximate analysis of the leaves *O. canum* revealed a considerable high level of carbohydrate, which represents about 63.96% of the total nutrients (Table 2). Carbohydrate is a pivotal nutrient that is required in adequate amounts in the diet to produce energy for the smooth functioning of the body. Its content in *O. canum* leaves as observed in this study makes the plant a rich source of vegetable carbohydrate over some convectional vegetables such as *Amaranthus hybridus* (52.18%) and *Ipomea batatas* (51.95%) as reported by some researchers (Akubugwo et al., 2007; Antia et al., 2006). The incorporation of the leaves of *O. canum* into diet will provide a non starchy adequate source of carbohydrate for diabetic and obese patients. However, *O. canum* leaves may not be a good source of protein because the level (0.4 ± 0.1 g/kg) observed in this study is very low when compared with other leafy vegetables like *O. gratissimum* leaves (1.21%) and *Cabbage* (1.40%). This observation is in agreement with the findings of Tull (1996) who reported that leafy vegetables are usually poor sources of proteins.

**Table 2.** The proximate composition of *Ocimum canum* leaves.

| Content | Composition (g/kg) |
|---|---|
| Carbohydrates | 639.6 ± 30.9 |
| Crude protein | 0.40 ± 0.10 |
| Ash | 120.0 ± 28.3 |
| Crude fiber | 170.0 ± 14.2 |
| Crude fat | 70.0 ± 14.1 |

Data expressed as mean ± SD.

**Table 3.** The mineral composition of *Ocimum canum* leaves.

| Element | Composition (g/kg) |
|---|---|
| Calcium | 50.72 ± 1.77 |
| Potassium | 18.76 ± 0.12 |
| Magnesium | 4.26 ±0.01 |
| Sodium | 9.58 ± 0.03 |
| Iron | 1.85 ± 0.02 |
| Phosphorous | 7.59 ± 0.05 |
| Manganese | 0.10 ± 0.01 |
| Zinc | 0.13 ± 0.01 |
| Lead | 0.02 ± 0.01 |
| Cadmium | 0.01 ± 0.01 |
| Na/K[1] | 0.51 |
| Ca/P[1] | 6.68 |
| Vitamin C[2] | 0.05 ± 4.51 |

Data expressed as mean ± SD.[1]Expressed as ratio; [2]Expressed as g/Kg.

Furthermore, the ash content, which is an indication of the mineral in the leaves, was higher than that of *O. gratissimum* leaves (6.88%) as reported by Edeoga et al. (2006), but the same with *Leonotis leonorus* (12.0%) according to Jimoh et al. (2010). This was an indication of the richness of *O. canum* leaves in minerals of nutritional importance. The crude fiber in the diet has been reported to lower the blood cholesterol and sugar levels, reduce the risk of hypertension and other cardiovascular related diseases (Ishida et al., 2000). The high crude fiber content implies that the leaves of *O. canum* can be a good dietary supplement especially for the hypertensive and obese patients. Dietary fats are flavoring agents that are used to increase the palatability of food (Antia et al., 2006). The fat content (70.0 ± 14.1 g/kg) of the leaves is high which contributes to the use of the plant as local condiment and flavoring agents. The finding of this study suggests that *O. canum* leaves can be a good source of vital nutrients even better than some of the convectional vegetables such as *Glechoma hederaceae*, *Origanum vulgare* and *Thymus mastichina* (Lillian et al., 2011).

Calcium is the most abundant macro element in the leaves of *O. canum* (Table 3). It is a vital element in building and maintaining strong bones and teeth. It is essential for blood clotting, maintenance of blood pressure and as a cofactor in enzymatic processes (Akubugwo et al., 2007). The high concentration of calcium in *O. canum* leaves makes it a good source of nutrients for the elderly who are predisposed to osteoporosis. The potassium and sodium levels of the leaves are higher than that of *O. gratissimum* leaves (850 and 6.0 mg/100 g) respectively as reported by Adepoju and Oyewole (2008). These macro elements are required for the normal functioning of the nervous system and play a vital role in regulating blood pressure. It has been recommended that Na/K ratio less than 1 will prevent high blood pressure (FDN, 2002).

The result of this study produced a Na/K ratio of 0.51 which is in conformity with the recommended ratio. The concentrations of magnesium, phosphorous, iron, manganese and zinc in *O. canum* leaves are higher than those reported for other *Lamiaceae* such as *L. leonorus* (Jimoh et al., 2010) and *O. gratissimum* leaves (Adepoju and Oyewole, 2008). These elements are essential components of immune system and are vital for the synthesis of hemoglobin (Akubugwo et al., 2007). A food sample is considered to be good if the calcium-phosphorus (Ca/P) ratio is greater than 1 and poor if it is less than 0.5 (Adeyeye and Aye, 2005). This is because a Ca/P ratio above 2 helps in the absorption of calcium in the small intestine. The Ca/P ratio of *O. canum* leaves was 6.68, which portrays that the leaves can promote the absorption of calcium in the intestinal lumen. Zinc serves as a cofactor in many enzymatic processes while magnesium influences cardiac output by regulating the fluid balance in the body (Ozcan, 2004).

Iron is involved in oxygen binding to hemoglobin thereby playing a vital role in blood formation. According to WHO (1997), two billion of the world's population are living with iron deficiency anemia. The result obtained in this study revealed an iron content that is higher than the recommended dietary allowance for males (1.37 mg/day) and females (2.94 mg/day) (FAO/WHO, 1988). Hence, the supplementation of the diet with *O. canum* leaves can help to combat the problem of iron deficiency anemia. Although, lead and cadmium are poisonous metals but their content in the leaves are very low and insignificant. Moreover, vitamin C is a water soluble antioxidant whose high dietary intake is associated with reduced risk of colon cancer and inflammatory diseases (Pattison et al., 2004). The sufficient amount of vitamin C in the leaves *O. canum* is an indication of the ability of the leaves to prevent the formation of carcinogens and scavenge free radicals which are formed during metabolic processes in humans.

## Conclusion

The result obtained in this study revealed that *O. canum* leaves contain vital compounds of nutritional importance.

The high concentration of micronutrients such as zinc, manganese and iron in the plant could be harnessed to combat micronutrient deficiencies especially in the rural communities. The use of convectional drugs has posed a lot of severe side effects on human health. Therefore, there is a need to incorporate edible phytochemicals into human diets as they are more effective, accessible and inexpensive. The high level of polyphenolic compounds (especially flavonoids and saponins) and vitamin C detected in *O. canum* leaves makes it a good source of dietary antioxidant. However, further studies are been carried out to access the efficacy of various extracts of the leaves to validate its ethnomedical use as acclaimed by traditional healers.

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