



Phytoconstituent Screening of Roselle (*Hibiscus sabdariffa*), Moringa (*Moringa oleifera*), Ginger (*Zingiber officinale*) and Fluted pumpkin (*Telfairia occidentalis*) Leaves

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ABSTRACT: There is a growing concern associated with the safety of some medicinal plants as plant-based medicine stages a comeback in the last few decades. The phytoconstituent and acute toxicity of some selected food plants eaten as vegetables or spices in Nigeria and some other tropics of the world were evaluated in this study. The acute toxicity of the ethanolic extracts of roselle, moringa, ginger and fluted pumpkin was tested using albino rats (*Rattus norvegicus*). The extracts were then screened to identify the phytonutrients and phytochemicals in them, using standard protocols. The acute toxicity study shows the extracts were nontoxic to the rats, even at a high dose of 2000 mg/kg body weight. The phytonutrients in roselle extract are calcium, iron, zinc, magnesium, vitamin A and vitamin C, while ginger extract has zinc, magnesium, vitamin A and vitamin C. Moringa and fluted pumpkin have all the tested nutrients. The phytochemicals in roselle extract are alkaloids, tannins, glycosides, and reducing sugars, while moringa contains all the tested phytochemicals except flavonoids and phlobatanins. Ginger extract has glycosides, reducing sugars, saponins, and flavonoids, while fluted pumpkin extract has all the tested phytochemicals except reducing sugars and phlobatanins. The findings of the study show the food plants are rich in nutrients and antioxidants, but contain traces of potentially toxic chemicals whose long-term use safety levels need to be evaluated. © JASEM

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Studies have shown that diets rich in fruits and vegetables are beneficial to human health and can even reduce the risk of many serious health conditions (Harper, 2011). Fruits and vegetables contain certain compounds known as phytochemicals and phytonutrients (Edward, 2016), and may reduce the risk of cancer, heart diseases and other illnesses (USFDA, 2009).

Phytochemicals and phytonutrients are often used interchangeably, however the two are quite different. While phytochemicals are non-nutritious natural chemicals formed during plants' normal metabolic processes, phytonutrients are nutrients such as minerals and vitamins derived from plants (Okigbo *et al.*, 2009). Phytochemicals include plant compounds that are beneficial as well as those that are detrimental, while phytonutrients specifically refers to compounds that have a positive effect. In other words, all phytonutrients are phytochemicals, but not all phytochemicals are phytonutrients (Edward, 2016). Classes of phytochemicals include the alkaloids, flavonoids, coumarins, glycosides, lycopenes, gums, carotenoids, polysaccharides, phenols, tannins, terpenes, and terpenoids (Densie, 2013). The varieties of phytonutrients are wide, and they are found in fruits, vegetables, legumes, grains,

nuts and teas. High amounts of phytonutrients and phytochemicals can be obtained from fruits and vegetables that have strong colours (Jackie, 2009).

In the last few decades, there is a renewed interest in plant-based medicine attributable to its efficacy and affordability (Yahaya *et al.*, 2012). However, despite the growing interest in plant-based medicine, there are concerns associated with not only their use, but their safety. Less than 10 % of herbal products in the world market are truly standardized to known active components and strict quality control measures are not always diligently adhered to (Obidike and Salawu, 2013). It is therefore necessary to carry out phytoconstituent analysis and toxicity studies of medicinal plants, even though they have been used for decades, to determine acceptable from non-acceptable toxicity levels. This study evaluates the phytoconstituent and toxic levels of some selected food plants eaten as spices or vegetables in Nigeria.

MATERIALS AND METHODS

Source of the Plant Materials: The plant materials-roselle, moringa, ginger and fluted pumpkin were purchased from Ketu in Lagos, Nigeria. They were identified by a curator, Mr. Odewo T. Kolawole, in the Department of Botany, University of Lagos, Nigeria. The voucher numbers of the authenticated

samples are LUH 4394, LUH 4558, LUH 4396 and LUH 4395 for roselle, moringa, ginger and fluted pumpkin, respectively.

Preparation of the Plant Materials: Fresh leaves of the plant materials were washed gently to remove impurities and air-dried under shade for one week. The dried leaves were milled into powder using a laboratory mill manufactured by Norris Limited, Poole, England. The ground plant materials were then stored in desiccators before use.

Preparation of the Plant Extracts: The bioactive compounds were extracted from the plant materials using the method of Sasidharan *et al.* (2011). Fifty grams (50 g) powder of each plant material were put in 500 ml 95 % cold ethanol and was allowed to stand for 72 hours. The extracts thus obtained were filtered with muslin cloth and evaporated to dryness at a temperature of 38°C. The resulting dried extracts of each plant material yielded 6.6 g, 6.5 g, 6.2 g, 5.9 g of roselle, moringa, ginger and fluted pumpkin, respectively. These dry extracts were reconstituted in water and were the decoctions used for the experiment.

Acute Toxicity Test: The acute toxicity of the ethanolic extracts of the plants was measured using the 'Classical LD₅₀' method described by Gabriel *et al.* (2008). Albino rats (30) of both sexes weighing between 183 and 205 g were used for the studies. The rats were randomly distributed into five groups of 6 rats each and were made to fast for 12 hours before commencing the study. The control group received only distilled water, while the test groups were orally administered doses of 200, 400, 500, 700, 1500, and 2000 mg kg⁻¹ of the extracts. The general symptoms of toxicity were monitored and recorded for each group within 24 hours.

Phytonutrients Analysis of the Plant Extracts: The phytonutrients present in the plant extracts were determined using thin layer chromatography (TLC) method as described by Sasidharan *et al.* (2011).

TLC was used to determine the compounds in the plant extracts by comparing the refractive values of the compounds in the extracts with the refractive values of their standards on the same TLC plate. A TLC plate which serves as the stationary phase was a sheet of glass coated with a thin layer of a solid adsorbent made of silica, while the mobile phase consisted of ethanol. In principle, the components will differ in solubility and in the strength of their adsorption to the adsorbent thereby different compounds will have different refractive values.

Phytochemical Screening of the Plant Extracts: The phytochemicals present in the plant extracts were identified using standard procedures as described by Sasidharan *et al.* (2011).

Test for Alkaloids: The presence of alkaloids in the extracts was tested using the Wagner Dragendoff's test. About 0.2 g of the extracts was heated with 2 % H₂SO₄ for two minutes. The mixture was filtered and few drops of Dragendoff's reagent were added. An orange-red precipitate shows the presence of alkaloids.

Test for Tannins: The tannins was tested in the plant extracts using the Ferric chloride test. Few quantities of the extracts were mixed with water and heated on a water bath. The mixture was filtered and ferric chloride was added to the filtrate. A dark-green solution shows the presence of tannins.

Test for Glycosides: The Felling test was used to detect the presence of glycosides in the plant extracts. The extracts were hydrolyzed with HCl solution and neutralized with NaOH solution. A few drops of Fehling's solution A and B were added. A red precipitate signals the presence of glycosides.

Test for reducing sugars: The reducing sugars were tested in the extracts using the Felling test. The extracts were shaken with distilled water and filtered. The filtrate was boiled with drops of Fehling's solution A and B for 5 minutes. An orange-red precipitate shows the presence of reducing sugars.

Test for Saponins: The Frothing test was used to detect the presence of saponins. About 0.2 g of the extracts was shaken with 5 ml of distilled water and then heated to boil. Frothing (appearance of a creamy mass of small bubbles) shows the presence of saponins.

Test for Flavonoids: The sodium hydroxide test was used to detect the presence of flavonoids in the extracts. Extract of about 0.2 g was dissolved in diluted NaOH and HCl was added. A yellow solution that turns colourless signals the presence of flavonoids.

Test for Phlobatanins: The presence of phlobatanins in the extracts was carried out using the hydrogen chloride test. The extract (0.5 g) was dissolved in distilled water and filtered. The filtrate was boiled with 2 % HCl solution. A red precipitate shows the presence of phlobatanins.

RESULTS AND DISCUSSION

The acute toxicity study of the plant extracts showed no mortality within 24 hours after administering the

plant extracts, which implies the extracts were non-toxic. The rats that received roselle extract displayed readiness to take more; they were licking the cannular used to administer the extract. The rats that received ginger extracts initially showed restlessness, but soon returned to normal. Moringa, and fluted pumpkin fed rats showed no any noticeable abnormal sign.

Table 1 shows the phytonutrients found in the plant extracts. Roselle extract contains calcium, iron, zinc, magnesium, vitamin A and vitamin C, while ginger extract has zinc, magnesium, vitamin A and vitamin

C. Moringa and fluted pumpkin extracts have all the tested nutrients.

Table 2 shows the phytochemicals present in the plant extracts. Roselle extract contains alkaloids, tannins, glycosides, and reducing sugars, while moringa contains all the tested phytochemicals except flavonoids and phlobatanins. Ginger extract has glycosides, reducing sugars, saponins, and flavonoids, while fluted pumpkin extract has all the phytochemicals except reducing sugars and phlobatanins

Table 1: The phytonutrients present in the plant extracts

Extract	lle	Moringa	er	Fluted pumpkin
Phytonutrient				
Calcium		++		+
Iron		++		++
Zinc		++		+
Magnesium		++		++
Vitamin A		+		+
Vitamin C		++	+	+
Protein		++		++

Key: - not present; + present in moderate amount; ++ present in abundant amount.

Table 2: The phytochemicals present in the plant extracts

Extract	Roselle	Moringa	Ginger	Fluted pumpkin
Phytochemical				
Alkaloids	+	+	-	+
Tannins	+	+	-	+
Glycosides	++	++	++	+
Reducing sugars	+	++	+	-
Saponins	-	+	+	+
Flavonoids	-	-	+	+
Phlobatanins	-	-	-	-

Key: - not present; + present in moderate amount; ++ present in abundant amount.

Phytonutrients and phytochemicals are nutrients and biologically active compounds, respectively, present in plants. These nutrients and chemicals have been shown by scientists to be necessary for sustaining human life (Harper, 2011). The short-term nontoxic property of the tested food plants confirmed the earlier belief that most medicinal plants are safe. The present of phytonutrients and phytochemicals in the plant extracts underscores the therapeutic and bioprotective properties of the selected food plants reported in several studies. Yahaya *et al.* (2012) reported that roselle, moringa, ginger and fluted pumpkin significantly protected the health of rats

exposed to cement dust compared with the equally exposed rats that received distilled water only.

Although the plants were nontoxic to the rats following treatment for 24 hours, the presence of secondary metabolites in the plants such as alkaloids, glycosides and tannins show the plants could produce side effects at an abnormally high dose or when taken too frequently. Many studies have reported cases of animal poisoning following consumption of plants containing secondary metabolites. For instance, consumption of groundsel plant containing the alkaloid senecionine, has resulted in many recorded

cases of livestock fatalities due to liver failure (PL, 2015). Short-term use of a commercial *Cascara sagrada* product containing anthraquinone glycosides has been reported to cause cholestatic hepatitis with subsequent portal hypertension, prolonged prothrombin time, and ascites (Nadir *et al.*, 2000). Tannins (commonly referred to as tannic acid) are water-soluble polyphenols that are present in many plant foods. They have been implicated in the decreases in feed intake, growth rate, feed efficiency, net metabolizable energy, and protein digestibility in experimental animals (Chung, 1998). Saponins are anabolic compounds, which are synthetic derivatives of testosterone, and have been shown to have many side effects, including liver damage, mood swings, and impotence (Leigh *et al.*, 1998).

Conclusion: The study shows the plants are nontoxic and are rich in nutrients and antioxidants. However, the presence of secondary metabolites in the plants is an indication that the plants can cause side effects if taken at abnormally high doses or too frequently.

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