

EVALUATION OF THE PHYTOCHEMICALS, MINERAL CONSTITUENTS AND ANTIOXIDANT ACTIVITY OF THE ETHANOL EXTRACT OF *BAPHIA NITIDA*

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ABSTRACT

Baphia nitida (camwood) is a widely distributed plant most commonly found in the interior and coastal regions of tropical Africa. Known by diverse local names across Africa, extracts from its stem, leaves and roots have been found to possess several medicinal values. This study was designed to investigate the phytochemical constituents, mineral elements composition and antioxidant properties of the ethanol extract of *Baphia nitida* leaves. *Baphia nitida* leaves was identified at the herbarium unit of Botany Department, University of Lagos with herbarium number: LUH 5614. The ethanol extract was screened for the presence of phytochemicals, antioxidant assays were carried out using DPPH scavenging activity and free radical antioxidant power and the mineral content was determined using Atomic Absorption Spectroscopy (AAS). Phytochemical screening of the extract revealed the presence of tannins, saponins, steroids, reducing sugar, glycosides, phlobotannins, terpenoids, flavonoids and alkaloids. There was high reducing power and it inhibited 2, 2-diphenyl-1-picrylhydrazyl (DPPH), indicating its antioxidant activity. Minerals detected include calcium, potassium, sodium, copper, iron, manganese, magnesium, zinc and cobalt. These were in varying concentrations with iron having the highest concentration and cobalt the lowest. The analysis shows that *Baphia nitida* is a promising herbal remedy with strong antioxidant property.

Keywords: *Baphia nitida*, 2, 2-diphenyl-1-picrylhydrazyl, Atomic absorption, Spectroscopy.

1 Introduction

Since the dawn of history, man has been faced with the challenges of eliminating ailments, a need which has not been met. Therefore, different generations have been adopting different measures to help check ailments. However from creation, plants have always been among the most effective primary measures for this check (Porter, 1997). Plants have been proven scientifically to have the ability to cure ailments, by providing some necessary nutrients which may be lacking in the body or by attacking the causative organism themselves. Medicinal plants have provided modern medicine with numerous plant-derived therapeutic agents (Evans, 2000; Oladunmoye *et al.*, 2009). These plants

have been the mainstay of traditional herbal medicine amongst rural dwellers worldwide since antiquity.

The medicinal value of a plant is due to the presence of some chemical substances (bioactive ingredients) also referred to as phytochemical or phytoconstituent that produce a definite physiological action in the human body (Mbagwu, 2005). They are also responsible for protecting the plant against microbial infections or infestations by pests (Nweze *et al.*, 2004; Doughari *et al.*, 2009). It is very important to have sufficient knowledge regarding herbs not only because of their widespread uses, but also because they have the potentials to cause toxic reactions or interaction with other drugs (Okigbo *et al.*, 2008).

Baphia nitida is a widely distributed plant most commonly found in the interior and coastal regions of tropical Africa and very abundant in under wood in the African dense forests (Ake-Assi, 1984; Lebrun and stork, 1997). It is locally known as *Igi osun*, *Abosi* and *Magiji* in the South-West, South-East and North West parts of Nigeria respectively. It belongs to Fabaceae family of suborder Caesalpinieae. Commonly known as Camwood, it appears mainly as shrubs or small trees with variable leaves pointed at the tip.

Baphia nitida has a rich history of use as dyewood and as shade provider in addition to its use across West Africa in the treatment of various ailments (Daziel, 1937; Irwine, 1961). Its leaves have been shown to possess diverse pharmacological properties acting against sprain, nosebleed, arthritis, rheumatism and asthma (Neuwinger, 1996; Poorter *et al.*, 2004; Ouattara, 2006). The leaf extract has been reported to possess a dose dependent analgesic activity (Onwukaeme and Lot, 1991). *Baphia nitida* produces a red gum which is very much used in folk medicine for treating various pathologies, including skin disorders, skin wounds, cicatrization and inflamed and infected umbilical cords (Onwukaeme and Lot, 1991).

The key to a successful discovery of naturally occurring therapeutic agents rests on the screening of extracts of natural products. This has had an impressive history of identifying active agents (Simon, 2007). In line with this, this study was geared towards investigating the phytochemical constituents, mineral elements composition and antioxidant properties of the ethanol extract of *Baphia nitida* leaves.

MATERIALS AND METHODS

Sample Collection and Identification

Fresh leaves of *Baphia nitida* were purchased from Mushin market in Lagos, Nigeria. It was authenticated at the herbarium unit of the Department of Botany of the University of Lagos by Mr. Odewo (Herbarium number: LUH 5614).

Extract Preparation

The dried leaves of *Baphia nitida* was powdered using blender (Blender/Miller III, model MS-223, Tapei, Taiwan). One hundred and twenty two grammes (122 g) of the powdered leaves were soaked in 98 % (v/v) ethanol for 72 hrs. The ethanol extract was recovered from three consecutive extractions by sieving. The crude ethanol extract was concentrated in rotary evaporator to obtain a weight of 3.46 % w/w g.

Phytochemical Screening

The methods of Trease and Evans (1996) and Parekh and Chanda (2007) were used to determine the presence of the following phytochemicals in the extract: saponins, phlobatannins, flavonoids, cardiac glycosides, steroids, alkaloids, terpenoids and tannins and reducing sugars.

Mineral Analysis

One gramme of the dried leaves was weighed and digested in a mixture of 5 ml of HCl, 2 ml of concentrated H₂SO₄ and 20 ml of concentrated HNO₃ in a conical flask under a fume hood. The content was mixed and heated gently at 180 °C for 30 min on a hot plate. The content was continuously heated until dense white fumes appeared. It was then finally heated strongly for about 30 min and then allowed to cool before making up to the mark in 50 ml volumetric flask with the acid mixture. Atomic Absorption Spectrophotometer (model Buck 210, VGR, USA) was then used to determine Ca, K, Na, Fe, Mn, Mg, Cu, Zn, and Co in the digested dried powdered (ashed) samples (A.O.A.C, 1990)

Antioxidant Activities

DPPH Radical Scavenging Activity

This was carried out using the method of Brand-Williams *et al.*, (1995) and ascorbic acid as a standard antioxidant. The free radical scavenging activity of the extract against 2, 2 – diphynyl -1 picryl – hydrazil (DPPH) radical was measured at 517 nm, as an index to its antioxidant activity. The concentration of the extract and ascorbic acid used were 0.4, 0.8, 1.2, 1.6 and 2.0 µg /ml.

Reducing Power

The reducing power of the ethanol extract of *Baphia nitida* was determined with slight modification of the method of Oyaizu (1986) using ascorbic acid as a standard antioxidant. Substances which have reduction potential react with potassium ferrocyanide (Fe²⁺) which then reacts with ferric chloride to form ferrous complex that has an absorption maximum at 700 nm. Increase in absorbance of the reaction mixture indicates increase in reducing power.

Statistical Analysis

All statistical comparisons were made with one-way analysis of variance (ANOVA) followed by Turkey Multiple Comparison Test using SPSS 17 data package. A value of $p < 0.05$ indicates significant differences in all cases.

RESULTS

Qualitative Phytochemical Analysis

Table 1 shows the results for the phytochemical analysis of the ethanol extract of *Baphia nitida*. The extract revealed the presence of tannins, phlobatannins, saponins, steroids, reducing sugar, glycosides, alkaloids, terpenoids, flavonoids.

Table 1: Phytochemical Screening of the ethanol extract of *Baphia nitida*

Contents	Indicators
Reducing Sugar	+ve
Tannins	+ve
Saponins	+ve
Steroids	+ve
Glycosides	+ve
Alkaloids	+ve
Phlobatannins	+ve
Terpenoids	+ve
Flavonoids	+ve

Keys: +ve: indicates positive

Mineral Analysis

Table 2 shows the results of the mineral analysis of the ethanol extract of *Baphia nitida*. The extract contains iron (35.6 mg/100 g), calcium (35.1 mg/100 g), potassium (21.05 mg/100 g), magnesium (11.1 mg/100 g), sodium (5 mg/100 g), copper (4.4 mg/100 g), zinc (4.4 mg/100 g) and cobalt (1.6 mg/100 g).

Table 2: Concentration (mg/100 g) of metals in *Baphia nitida* extract

S/N	MINERAL	CONCENTRATRATION (mg/100 g)
1	Calcium (Ca)	35.10
2	Potassium (K)	21.05
3	Sodium (Na)	5.00
4	Copper (Cu)	4.40
5	Iron (Fe)	35.60
6	Manganese (Mn)	5.50
7	Magnesium (Mg)	11.10
8	Zinc (Zn)	4.40
9	Cobalt (Co)	1.60

DPPH Scavenging Activity

Figure 1 shows the percentage inhibition of DPPH by the ethanol extract of *Baphia nitida* and ascorbic acid. There was a concentration-dependent increase in percentage inhibition of DPPH by the ethanol extract of *Baphia nitida* and ascorbic acid. The extract showed a significantly ($p < 0.05$) lower percentage inhibition of DPPH when compared to the standard ascorbic acid used at each of the concentrations.

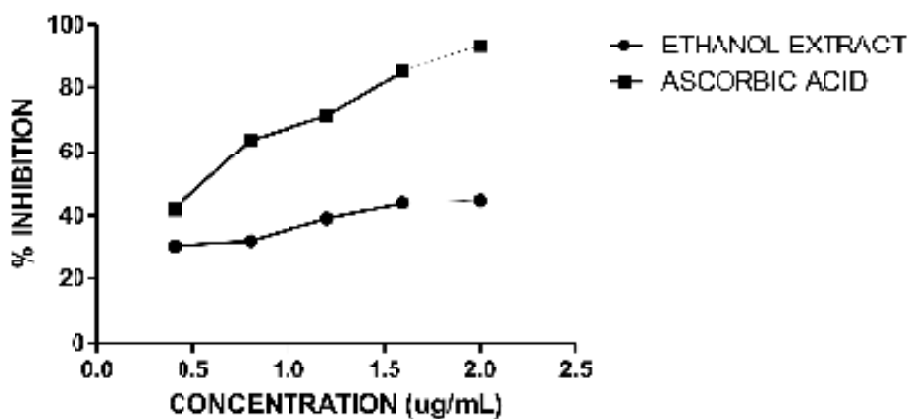


Fig. 1: Percentage inhibition of DPPH by ethanol extract of *B. nitida* & ascorbic acid

Values are mean \pm SD, n = 2. *Significant change in comparison with ascorbic acid at $p < 0.05$

Reducing Power Assay

Figure 2 shows the reducing power of the ethanol extract of *Baphia nitida* and ascorbic acid. There was a concentration-dependent increase in reducing power of the ethanol extract of *Baphia nitida* and ascorbic acid. The extract showed a significantly ($p < 0.05$) lower reducing power when compared to the standard ascorbic acid used at each of the concentrations.

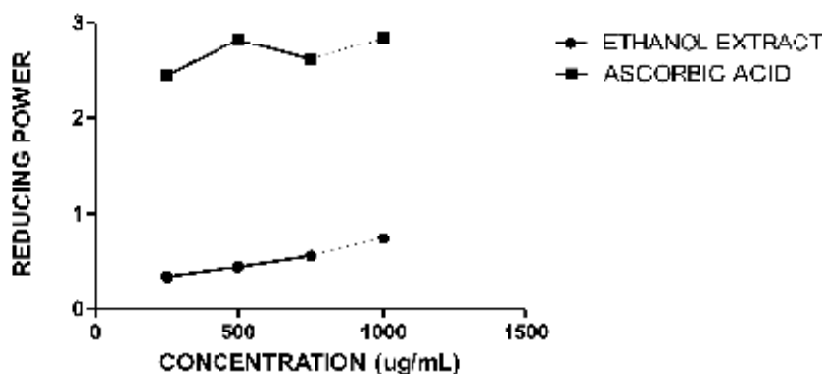


Fig.2: Reducing power of ethanol extract of *B. nitida* and ascorbic acid

Values are mean \pm SD, n = 2. *Significant change in comparison with Ascorbic acid at $p < 0.05$

4. Discussion

The medicinal value of herbal remedies lies in those chemical substances that produce definite physiological actions on the human body such as alkaloids, flavonoids and tannins (Akanke *et al.*, 2011). The phytochemical analysis of the ethanol extracts of *Baphia nitida* leaves reveals the presence of tannins, phlobatannins, saponins, steroids, reducing sugar, glycosides, alkaloids, terpenoids, and flavonoids. These phytochemicals may account for the medicinal value attributed to the plant leaves (Parivugunada *et al.*, 2008). Some examples of such medicinal values include treating sprain, nosebleed, skin disorders, arthritis, rheumatism and asthma (Poorter *et al.*, 2004; Ouattara, 2006).

The medicinal values of some plant species used in homeopathic system have been traced to the presence of elements such as calcium, chromium, copper, iron, magnesium, potassium and zinc in plants (Perman *et al.*, 1993). Elements equally contribute to neurochemical transmissions, and act as co-factors for various enzymes and metabolic processes (Mayer and Vyklicky, 1989). Analysis of the extract using atomic absorption spectrophotometry (AAS) reveals the presence of some mineral elements at varying concentrations. The accumulation of elements in medicinal plants have been reported to depend on climatic factors, plant species, air pollution and other environmental factors (Sovljanski, *et al.*, 1989). The plant used in the study was cultivated in Lagos, Nigeria and the soil is loam-sandy. The extract is very rich in iron (35.6 mg/100 g), calcium (35.1 mg/100 g) and potassium (21.05 mg/100 g) but with lower levels of magnesium (11.1

mg/100 g), sodium (5 mg/100 g), copper (4.4 mg/100 g), zinc (4.4 mg/100 g) and cobalt (1.6 mg/100 g). The result of this present study agrees with earlier studies of elemental distribution in medicinal plant species (Kim *et al.*, 1994). We observed a high level of macro elements accumulation in the sampled plant leaf except in very few cases where the concentration is considerably low.

The percentage inhibition of DPPH and reducing power activity are associated with antioxidant activity and may serve as a significant reflection of the antioxidant activity. The ethanol extract of *Baphia nitida* showed a concentration-dependent increase in percentage inhibition of DPPH, thereby indicating its antioxidant property (Figure 1). The reducing power of the extract of *Baphia nitida* leaves was found to be remarkable, which increased gradually with an increase in the concentration of the extract (Figure 2). Compounds with reducing power indicate that they are electron donors and can reduce the oxidized intermediates of lipid peroxidation processes, so that they can act as primary and secondary antioxidants (Bhandari *et al.*, 2012). Compared to the standard ascorbic acid, the extract showed a significantly lower percentage inhibition of DPPH and reducing power activity at each of the concentrations used. This shows that a higher concentration of the ethanol extract will be required to reflect an equivalent antioxidant activity with the standard ascorbic acid.

The ethanol extract of *Baphia nitida* leaves is a good source of important phytochemicals and minerals, and therefore may have therapeutic properties hitherto accredited to it. It has antioxidant properties and could serve as supplements of macro and micro elements in the body. However, further studies are needed to validate the bioactivity of the extract in vitro and in vivo. It may also be necessary to isolate and characterize its active components having potential for prevention or treatment of chronic diseases.

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