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Phytochemical Screening and Mineral Contents of Leaves of Some Nigerian Woody Plants

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Abstract: In this study, leaves of woody plants (2-4 species) obtained in Nigeria between November and December 2001 was evaluated for their phytochemical and mineral contents using standard methods. To determine minerals, 0.5 g of each samples were dry ashed, dissolved in distilled water with few drops of conc. HCl and read on an atomic absorption spectrophotometer. Phytate and phytate P were analyzed by first extracting the samples followed by titrating with FeCl_3 , the values obtained were multiplied with standard factors (1.95 and 3.56, respectively) and qualitative methods were used for other tests. The study revealed presence of alkaloids, tannins, resins, saponins, flavonoids, glycosides, carbohydrate, sterols and flobatanin in most of the samples. The levels of phytate (mean = 692, SD = 212, CV% = 30.7) and phytate phosphorous (mean = 188, SD = 51, CV% = 27.4) were generally high and all leaves had more than 30% of their total phosphorus linked to phytate. These results compared with literature values. The concentrations of the mineral were found to be high. The result showed that with high phytate contents the bioavailability of minerals might be relatively low.

Key words: Phytochemical, phytate, mineral composition, therapeutic compositions, bioavailability

INTRODUCTION

Woody plants have been of wise use in Nigeria. They have contributed to sustainable development and benefit to many people especially the poor, who depend on these for their livelihoods. There are about 900 different kinds of trees in Nigeria, some are easily recognized but many can only be named with certainty when flowers or fruit as well as leaves, are available (Aju and Popoola, 2005). Fruits, barks and leaves, body are appreciated, not only for texture and flavor, but also for their chemical and nutritional properties (Abulude *et al.*, 2004).

The woody plants are versatile material plants having a wide range of local therapeutic applications. The leaves, roots, barks and seeds are found to be active antipyretic laxative, analgesic, antifungal, antibacterial and anti-inflammatory (Faruq, 2004; Olafimihan *et al.*, 2004; Hassan *et al.*, 2004; Ojukwe *et al.*, 2004). Some of the woody plants have also been reported as therapeutic foods, useful in preventing diseases such as hypertension, hypercholesterolemia and cancer (Lexicon, 1983). Fragrant gum resins obtained from some of them are sometimes used as fumigants (Hassan *et al.*, 2004). These functional characteristics are mainly due to the presence of phytochemicals. Despite the extensive use of these woody plants, reports on detailed phytochemical investigation are scanty. Out of many potent phytochemicals, tannin, glycosides, saponin, sterol, resin, flobatannins, flavonoids, alkaloids and phytate were initially chosen for this research.

The aim of this research was to extend knowledge on mineral and phytochemical qualities of woody plant leaves. It is hoped that the results would add to composition table.

MATERIALS AND METHODS

The different varieties (2-4 species) of wood plant leaves were obtained from the Federal College of Agriculture Campus in Akure, Ondo State, Nigeria between November and December, 2004, identified and labeled (Irvine, 1961). The samples under investigation are shown in Table 1.

Experimental

One hundred gram of the leaves were washed with distilled water, oven dried at 60°C for 6 h, then finely ground, sieved (2 mm sieve), mixed thoroughly, stored in plastic containers and kept in ambient temperature prior analysis.

Mineral Analysis

Total phosphorus (P) was analyzed from solutions obtained by first dry-ashing 0.6 g of the samples at 550°C to constant weight and dissolving the ash in volumetric flasks using distilled water with a few drops of concentrated HCl. Phosphovanadomolybdate method was used for the determination (AOAC, 1990). A Perkin Elmer atomic absorption spectrophotometer was used for the determination of Ca, K, Na and Mg.

Phytate Analysis

Four grams of ground sample was soaked in 100 cm³ of 2% HCl for 3 h and then filtered through two layers of filter paper 25 cm³ of the filtrate was placed in a 250 cm³ conical flask and 5 cm³ of 0.3% NH₄SCN solution was added as an indicator, 53.5 cm³ of distilled water was then added to reach the proper acidity. This mixture was titrated against FeCl₃ solution, which contains about 0.00195 g of Fe iron per cm³ of FeCl₃ solution. The result was multiplied by factor 1.95 to obtain phytate P. phytate P result was multiplied by factor 3.55 to convert to phytate.

Table 1: Common, scientific, family and vernacular names of the investigated woody plants

Code	Common name	Scientific name	Family name	Vernacular name
WP 1	Christmas Bush	<i>Alchornea cordifolia</i>	Euphorbiaceae	-
WP 2	Gliricidia	<i>Gliricidia sepium</i>	Papilionaceae	Agunmaniye
WP 3	Hop bush	<i>Dodonaea viscosa</i>	Sapindaceae	-
WP 4	Neem	<i>Azadirachta indica</i>	Meliaceae	Dongoyaro
WP 5	Mango	<i>Mangifera indica</i>	Anacardiaceae	Mangoro
WP 6	Cashew	<i>Anacardium occidentale</i>	Anacardiaceae	Kaju
WP 7	Hog plum	<i>Spondias monbin</i>	Anacardiaceae	Iyeye
WP 8	Teak	<i>Tectona grandis</i>	Verbenaceae	-
WP 9	Indian almond	<i>Prunus amygdalus</i>	Rosaceae	-
WP 10	Arabia coffee	<i>Coffea arabica</i>	Rubiaceae	-
WP 11	Wild mango	<i>Irvingia gaboneusis</i>	Irvingiaceae	Oro
WP 12	Kolanut	<i>Cola nitida</i>	Struculiaceae	Obi
WP 13	Pear	<i>Persea Americana</i>	Lauraceae	-
WP 14	Rubber	<i>Hevea brasiliensis</i>	Euphorbiaceae	-
WP 15	Pawpaw	<i>Carica papaya</i>	Caricaceae	Ibepe
WP 16	Grape	<i>Citrus paradise</i>	Rutaceae	Osan
WP 17	Abbe walker	<i>Dracaena fragrans</i>	Agaraceae	Peregum
WP 18	Fig	<i>Ficus capensis</i>	Moraceae	Opoto
WP 19	-	<i>Bombax buonoposcence</i>	Bombacaceae	Poporo
WP 20	African teak	<i>Chlorophora excelsa</i>	Moraceae	Iroko
WP 21	Tallow tree	<i>Detarium microcarpum</i>	Gaesalpinaceae	-
WP 22	Lemon-Scented gum	<i>Encalyptus citriodora</i>	Myrtaceae	-
WP 23	Sand paper	<i>Ficus asperifolia</i>	Moraceae	Epinpin
WP 24	Sour sop	<i>Aumona muricata</i>	Annonaceae	-
WP 25	Alee apple	<i>Blighia sapoda</i>	Sapindaceae	-
WP 26	Ringworm shrub	<i>Cassia alata</i>	Caesalpinaceae	-
WP 27	White star apple	<i>Chrysophyllum albidum</i>	Sapotaceae	Agbalumo
WP 28	White butter	<i>Butyrospermum parkii</i>	Sapotaceae	Ori

^A Vernacular-Yoruba name

Phytochemical Analysis

Extraction

One hundred and fifty centiliter of water was added to 20 g of ground sample in a conical flask. The mixture was covered and allowed to stand for 3 h with occasional stirring. The mixture was filtered with a whatman No. 2 filter paper. The filtrate was stored in plastic containers and kept in ambient temperature prior to analyses (Abulude, 2001).

Test for Carbohydrate

Fehling's Test

5 cm³ of mixture of equal volumes of Fehlings A and B was added to 2 cm³ of each extract in a test tube. The resultant mixture was boiled for 2 min. A brick red precipitation of copper oxide was observed.

Test for Tannins

Two drops of 5% FeCl₃ was added to 1 cm³ of extract. A dirty-green precipitate was observed in each extract.

Test for Glycosides

10 cm³ of 50% H₂SO₄ was added to 1 cm³ of each extract in a test tube. The mixture was heated in boiling water for 5 min. 10 cm³ of Fehlings solution (5 cm³ of each solution A and B) was added and boiled. A brick red precipitate indicating presence of glycosides was observed.

Tests for Saponins

Frothing Test

2 cm³ of each extract in a test tube was vigorously shaken for 2 min. Frothing indicating presence of saponin was noted.

Test for Resins

5 cm³ of copper was added to 5 cm³ of each extract. The resulting solution was shaken vigorously and allowed to separate. A green colored precipitate indicating presence of resin was noticed.

Test for Phlobatanins

5 cm³ of distilled water was added to 5 cm³ of each extract and boiled with 5 cm³ of 1% HCl for 2 min. No visible reaction was obtained indicating absence of phlobatanins.

Test for Flavonoids

2 cm³ of extract was heated with 10 cm³ of ethyl acetate on a water bath and cooled. The layers were allowed to separate and the color of the NH₃ layer noted (red coloration formed).

Test for Alkaloids

1 cm³ of 1% HCl was added to 3 cm³ of each extract in a test tube. Each extract was treated with few drop of Meyer's reagent. A creamy white precipitate was observed indicating presence of alkaloids.

RESULTS

Table 2 shows total P; phytate P, phytate, phytate P expressed as percentage of total P. Phytate content ranged from 391-1108 mg 100 g⁻¹ DM. Total P was the highest in neem leaves and lowest in

Table 2: Total phosphorus (P), phytate P, phytate (mg100 g⁻¹ DM) and phytate P (as % of total P) in the woody plant leaves examined

Code	Samples	Total P (mg100 g ⁻¹)	Phytate P (mg100 g ⁻¹)	Phytate (mg100 g ⁻¹)	Phytate P (as % of total P)
WP 1	<i>Alchornea cordifolia</i>	250.0	110.0	391.0	44.0
WP 2	<i>Gliricidia sepium</i>	290.0	240.0	852.0	83.0
WP 3	<i>Dodonaea viscosa</i>	320.0	160.0	568.0	50.0
WP 4	<i>Azadirachta indica</i>	450.0	210.0	746.0	47.0
WP 5	<i>Mangifera indica</i>	297.0	125.0	444.0	42.0
WP 6	<i>Anacardium occidentale</i>	310.0	110.0	391.0	36.0
WP 7	<i>Spondias monbin</i>	218.0	121.0	430.0	56.0
WP 8	<i>Tectona grandis</i>	314.0	135.0	479.0	43.0
WP 9	<i>Prunus amygdalus</i>	291.0	114.0	405.0	39.0
WP 10	<i>Coffea arabica</i>	315.0	215.0	763.0	68.0
WP 11	<i>Irvingia gabonensis</i>	420.0	200.0	710.0	48.0
WP 12	<i>Cola nitida</i>	410.0	270.0	959.0	66.0
WP 13	<i>Persea americana</i>	325.0	240.0	852.0	74.0
WP 14	<i>Hevea brasiliensis</i>	285.0	200.0	1065.0	70.0
WP 15	<i>Carica papaya</i>	217.0	212.0	1108.0	98.0
WP 16	<i>Citrus paradise</i>	225.0	190.0	675.0	84.0
WP 17	<i>Dracaena fragrans</i>	330.0	182.0	646.0	55.0
WP 18	<i>Ficus capensis</i>	360.0	310.0	1101.0	86.0
WP 19	<i>Bombax buonoposence</i>	410.0	240.0	852.0	59.0
WP 20	<i>Chlorophora excelsa</i>	310.0	245.0	870.0	79.0
WP 21	<i>Detarium microcarpum</i>	321.0	128.0	454.0	40.0
WP 22	<i>Encalyptus citriodora</i>	245.0	135.0	479.0	55.0
WP 23	<i>Ficus asperifolia</i>	336.0	164.0	582.0	49.0
WP 24	<i>Annona muricata</i>	218.0	182.0	646.0	84.0
WP 25	<i>Blighia sapida</i>	328.0	210.0	746.0	64.0
WP 26	<i>Cassia alata</i>	323.0	210.0	746.0	65.0
WP 27	<i>Chrysophyllum albidum</i>	278.0	176.0	625.0	63.0
WP 28	<i>Butyrospermum parkii</i>	252.0	221.0	785.0	88.0
	X	309.0	188.0	692.0	62.0
	±SD	61.0	51.0	212.0	17.0
	CV (%)	19.6	27.4	30.7	27.6

Table 3: Phytochemical analysis of the woody plant leaves examined

Label	Sample	Carbo- hydrate	Tannins	Glyco- sides	Sapon nins	Sterols	Resins	Flobat- anin	Flavo- noids	Alkaloids
WP 1	<i>Alchornea cordifolia</i>	+	+	-	+	+	+	-	+	+
WP 2	<i>Gliricidia sepium</i>	+	+	+	+	+	+	-	+	+
WP 3	<i>Dodonaea viscosa</i>	+	+	+	+	+	+	-	+	+
WP 4	<i>Azadirachta indica</i>	+	+	+	+	+	+	-	+	+
WP 5	<i>Mangifera indica</i>	+	+	+	+	+	+	-	+	+
WP 6	<i>Anacardium occidentale</i>	+	+	+	+	+	+	-	+	+
WP 7	<i>Spondias monbin</i>	+	+	+	-	+	+	-	+	+
WP 8	<i>Tectona grandis</i>	+	+	+	+	+	+	-	+	+
WP 9	<i>Prunus amygdalus</i>	+	+	-	+	+	+	-	+	+
WP 10	<i>Coffea arabica</i>	+	+	+	+	+	-	-	+	+
WP 11	<i>Irvingia gabonensis</i>	+	+	+	+	+	+	-	+	+
WP 12	<i>Cola nitida</i>	+	+	-	+	+	+	-	+	+
WP 13	<i>Persea americana</i>	+	+	+	+	+	+	-	+	+
WP 14	<i>Hevea brasiliensis</i>	+	+	-	+	+	+	-	+	+
WP 15	<i>Carica papaya</i>	+	+	+	+	+	+	-	+	+
WP 16	<i>Citrus paradisi</i>	+	+	+	+	+	+	-	+	+
WP 17	<i>Dracaena fragrans</i>	+	+	-	+	+	-	-	+	+
WP 18	<i>Ficus capensis</i>	+	+	-	+	+	+	-	+	+
WP 19	<i>Bombax buonoposence</i>	+	+	+	-	+	+	-	+	+
WP 20	<i>Chlorophora excelsa</i>	+	+	+	+	+	+	-	+	+
WP 21	<i>Detarium microcarpum</i>	+	+	+	+	+	+	-	+	+
WP 22	<i>Encalyptus citriodora</i>	+	+	+	+	+	+	-	+	+
WP 23	<i>Ficus asperifolia</i>	+	+	+	+	+	+	-	+	+
WP 24	<i>Annona muricata</i>	+	+	-	-	+	+	-	+	+
WP 25	<i>Blighia sapida</i>	+	+	+	+	+	+	-	+	+
WP 26	<i>Cassia alata</i>	+	+	+	+	+	+	-	+	+
WP 27	<i>Chrysophyllum albidum</i>	+	+	+	+	+	+	-	+	+
WP 28	<i>Butyrospermum parkii</i>	+	+	+	+	+	+	-	+	+

+ Present, - Absent

Table 4: Mineral composition in leaves investigated (mg100 g⁻¹ DM)

Label	Sample	Ca	Na	K	Mg
WP 1	<i>Alchornea cordifolia</i>	600	75	672	199
WP 2	<i>Gliricidia sepium</i>	420	45	717	74
WP 3	<i>Dodonaea viscosa</i>	513	125	625	92
WP 4	<i>Azadirachta indica</i>	768	183	425	93
WP 5	<i>Mangifera indica</i>	460	247	470	82
WP 6	<i>Anacardium occidentale</i>	815	158	620	120
WP 7	<i>Spondias monbin</i>	358	80	525	134
WP 8	<i>Tectone grandis</i>	620	112	473	98
WP 9	<i>Prunus amygdalus</i>	680	118	327	110
WP 10	<i>Coffea arabica</i>	490	80	490	120
WP 11	<i>Irvingia gabonensis</i>	754	95	972	79
WP 12	<i>Cola nitida</i>	800	76	812	82
WP 13	<i>Persea Americana</i>	752	221	621	91
WP 14	<i>Itevea brasiliensis</i>	727	125	561	94
WP 15	<i>Carica papaya</i>	681	232	655	191
WP 16	<i>Citrus paradisi</i>	625	242	472	120
WP 17	<i>Dracaena fragrans</i>	481	252	724	80
WP 18	<i>Ficus capensis</i>	567	189	625	140
WP 19	<i>Bombax buonoposence</i>	888	88	754	170
WP 20	<i>Chlorophora excelsa</i>	479	70	785	120
WP 21	<i>Detarium microcarpum</i>	450	68	777	116
WP 22	<i>Encalyptus citriodora</i>	410	99	625	118
WP 23	<i>Ficus asperifolia</i>	624	121	872	147
WP 24	<i>Ammonia muricata</i>	578	100	612	152
WP 25	<i>Blighia sapida</i>	574	97	525	89
WP 26	<i>Cassia alata</i>	620	135	621	143
WP 27	<i>Chrysophyllum albidum</i>	717	148	348	127
WP 28	<i>Butyrospermum parkii</i>	824	193	562	184
	X	617	746	617	120
	±SD	140	78	150	35
	CV (%)	23	53	24	29

pawpaw leaves. Phytate P expressed as percentage of total P ranged between 36% (*Anacardium occidentale*) and 98% (*Carica papaya*). Coefficient of variation in percent (CV%) of all the results were high among the samples with total P being the least (20%) while phytate (31%) showed the highest variation.

The results in Table 3 showed that carbohydrate, alkaloids, glycosides, sterols, saponins, flavonones, tannin and resin were present in most of the extracts of leaves examined. Phlobatanins was absent.

All the minerals were highly concentrated. Calcium, potassium and sodium were highly concentrated with means (mg 100 g⁻¹) of 617 (SD = 140, CV = 23%), 617 (SD = 150, CV = 24%) and 746 (SD = 78, CV = 53%), respectively (Table 4).

DISCUSSION

The values of total P, phytate P, phytate, phytate P expressed as percentage of total P were higher than those reported for some varieties of mushrooms (Abulude *et al.*, 2001) and some foods of major consumption in Nigeria (Adeyeye *et al.*, 2000). Levels of phytate were comparable to levels reported for vegetables (Abulude, 2001) and varieties of lupin seeds (Trugo *et al.*, 1993). Phytate contents vary considerably depending on the environmental conditions, maturation and processing procedures (Griffiths and Thomas, 1981). Phytate chelates with mineral elements thereby have significant effects on the utilization of the minerals and also interfere with basic residues of proteins. Phytate has been shown to play a role in preventing colorectal carcinoma, hypercholesterolaemia and renal calculi (Marounek *et al.*, 2000). Phosphorus assists calcinm in many body reactions although it

also has its own independent functions (Fleck, 1976). Calcium in conjunction with phosphorus, magnesium, manganese, vitamins A, C and D, chlorine and proteins are all involved in bone formation. Results of Phytate P expressed as percentage of total P were consistent with earlier results on lesser-known leguminous crops seeds (Balogun and Fetuga, 1989). Coefficients of variation of the parameters were not similarly distributed in all its samples.

Results of the phytochemical analyses compared well with literatures (Hassan *et al.*, 2004; Faruq *et al.*, 2004; Olafimihan, 2004). The classes of compounds are known to show curative activity against several pathogens and is therefore not surprising that the plant leaves extracts are used traditionally as an analgesic, antimicrobial and soothing herbs (Hassan *et al.*, 2004). It is documented that presence of saponins can control human cardiovascular disease and reduce blood cholesterol. Tannins may provide protection against microbial degradation of dietary proteins in the rumen (Aletor, 1993).

The results recorded in this research were in total agreement with results obtained by Abulude *et al.* (2001) for mushrooms. Despite the fact that there is no recommended dietary allowance for potassium and sodium, it is recommended that the intake should be the same to counteract the effect of sodium in raising blood pressure. The leaves may serve as good supplements in the body supply of calcium, magnesium, potassium and sodium. Calcium is important in blood clotting, muscles contraction and in certain enzymes metabolic processes. It is very important that the normal calcium level in the diet should be balanced throughout life. Magnesium is an activator of many enzyme systems and maintains the electrical potential in nerves. Sodium and potassium are required to maintain osmotic balance of the body fluid, the pH of the body, regulate muscle and nerve irritability and control of glucose absorption.

The nutritive values of the leaves were high, since they were rich in mineral elements, efforts to reduce their loss through processing should be ensured. The mineral bioavailability may be low due to high phytate contents. The results obtained for the identified phytochemical compounds showed the therapeutic compositions of the leaves.

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