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COMPARISON OF THE NUTRITIVE VALUE AND BIOLOGICAL ACTIVITIES OF THE ACETONE, METHANOL AND WATER EXTRACTS OF THE LEAVES OF *BIDENS PILOSA* AND *CHENOPODIUM ALBUM*

ADEOLU ADEDAPO[1]*, FLORENCE JIMOH[2] and ANTHONY AFOLAYAN[2]

[1]Center for Cardiovascular Diseases, College of Pharmacy and Health Sciences, Texas Southern University, Houston TX, USA
[2]Department of Botany, University of Fort Hare, Alice 5700, South Africa

Abstract: A resurgence of interest has developed in wild vegetables for their possible medicinal values in diets. Wild plant species provide minerals, fibre, vitamins and essential fatty acids and enhance taste and color in diets. For this reason, the nutritional, phytochemical, antioxidant and antibacterial activities of the acetone, methanol and water extracts of the leaves of *Bidens pilosa* and *Chenopodium album* were investigated. The proximate analysis showed that the leaves of the plants contained appreciable percentage of moisture content, ash content, crude protein, crude lipid, crude fibre and carbohydrate. Elemental analysis in mg/100 g dry weight (d.w.) indicated that the leaves contained sodium, potassium, calcium, magnesium, iron, zinc, phosphorus, copper, manganese, and nitrogen. The chemical composition in mg/100 g d.w. showed the presence of alkaloid, saponins, and phytate. The extracts also caused DPPH radical scavenging activities which were comparable to those of ascorbic acid. This was also the same for BHT scavenging activity. With respect to the polyphenols, the extracts of these two plants also contained appreciable levels of these phytochemicals. The extracts of these plants also caused varied inhibition of the bacterial strains used in this study.

Keywords: antibacterial properties, antioxidant activities, *B. pilosa*, *C. album*, nutritional value

Modern agricultural technology and marketing have caused a reduction in the genetic diversity of plant species, especially in vegetables, worldwide (1–3). Wild plants with a desired gene (resistance to diseases etc.) may be used in breeding programmes. Williams (4) emphasized the need to preserve new plant resources to broaden the biological diversity in human nutrition.

A resurgence of interest has developed in wild species for their possible medicinal values in diets. Wild plant species provide minerals, fibre, vitamins and essential fatty acids and enhance taste and color in diets. In addition, they have antibacterial, hepatoprotective and anticarcinogenic properties, and therefore having medicinal values (5–7).

It is interesting to note that few vegetables species were consumed in South Africa by the general populace. The interview conducted during the course of our research in Alice and its surrounding villages indicated that many of these species, though known, are considered as weeds and were not eaten by the people. This is in spite of the fact that these vegetables grow spontaneously and in abundance around the rural homesteads. Many workers (8–12) have reported the compositional evaluation and functional properties of various types of edible wild plants in use in the developing countries. The consumption of vegetables has also been linked to reduction in the incidence of oxidative stress related diseases such as cancer and diabetes among others (13). This is due to beneficial health functionality of phenolic antioxidants present in them. Some plant phenolics have also shown antimicrobial effects (14).

Bidens pilosa L. is a species in the plant family Asteraceae. It is considered a weed in some tropical habitats. However, in some parts of the world it is a source of food (15). It is a small, erect annual herb that grows to 1 m high. It has bright green leaves with serrated, prickly edges and produces small, yellow flowers and black fruit. The plant has a long history of use among the indigenous people of the Amazon, and virtually all parts of the plant are used. Generally the whole plant is uprooted and pre-

* Corresponding author: e-mail: adedapo3a@yahoo.co.uk.

pared in decoctions or infusions for internal use, and/or crushed into a paste or poultice for external use. In Peruvian herbal medicine *Bidens pilosa* (Picăo preto) is employed to reduce inflammation, increase urination, and to support and protect the liver. It is commonly used there for hepatitis, conjunctivitis, abscesses, fungal infections, urinary infections, as a weight loss aid, and to stimulate childbirth. Phytochemically, the plant is rich in flavonoids, terpenes, phenylpropanoids, lipids, and benzenoids (16–20).

Chenopodium album L. is a fast-growing weedy annual plant in the genus *Chenopodium*. It belongs to the family Amaranthaceae. The standard British name is Fat-hen and it is also known as lamb's quarters. The leaves and seeds of all members of this genus are more or less edible. However, many of the species in this genus contain saponins, though usually in quantities too small to do any harm. Although toxic, saponins are poorly absorbed by the body and most pass straight through it without any problem. They are also broken down to a large extent in the cooking process. In nitrogen-rich soils, the plants can also concentrate hydrogen cyanide (21). In small quantities, hydrogen cyanide has been shown to stimulate respiration and improve digestion, it is also claimed to be of benefit in the treatment of cancer. The leaves are anthelmintic, antiphlogistic, antirheumatic, mildly laxative and odontalgic (22–24).

These two plants were identified as wild vegetables and the study was therefore aimed at assessing their nutritional quality and possible biological activities.

MATERIALS AND METHODS

Plant collection and extract preparation

Fresh plant materials of *Bidens pilosa* and *Chenopodium album* were collected in November 2006 from the wild around the University of Fort Hare campus (Alice, South Africa). The area falls within the latitudes 30°00'–34°15'S and longitudes 22°45'–30°15'E. It is bonded by the sea in the east and the drier Karoo (semi-desert vegetation) in the west (25). These areas consist of villages which are generally classified as rural and poor. Professor D. Grierson of the Department of Botany, University of Fort Hare, authenticated the species. A voucher specimen was prepared and deposited in the herbarium of the Department of Botany (Jimoh Med. 2006/7). The plant material was allowed to air-dry at ambient temperature (± 24°C) and then milled. Twenty grams each of the sample were extracted with 200 mL each of acetone, methanol, and water, respectively, at ambient temperature, with agitation for 18–24 h. Each extract was filtered using Whatman no. 1 filter paper and concentrated under reduced pressure to dryness below 40°C. The water extract was freeze-dried. The extract yields (w/w) were: acetone – 2.8%, methanol – 8.6% and water – 9.8%, respectively, for *B. pilosa* but acetone –2.5%, methanol – 8.2% and water – 8.3%, respectively, for *C. album*. The dried extracts thus obtained were used directly for the determination of the antioxidant and antibacterial activities (26). Determinations of chemical and nutritive values of these plants were carried out using the dried samples that were ground into powder form.

Chemicals

1,1-Diphenyl-2-picrylhydrazyl (DPPH), 2,2'-azino-bis-3-ethylbenzothiazoline-6-sulfonic acid (ABTS), 3-(2-pyridyl)-5,6-diphenyl-1,2,4-triazine-4',4''-disulfonic acid, potassium ferricyanide; catechin, butylated hydroxytoluene (BHT), ascorbic acid, catechin, tannic acid, quercetin and $FeCl_3$ were purchased from Sigma Chemical Co. (St. Louis, MO, USA)., vanillin from BDH; Folin-Ciocalteu phenol reagent and sodium carbonate were from Merck (Damstadt, Germany). All other chemicals used including the solvents, were of analytical grade.

Determination of total phenolics

Total phenol contents in the extracts were determined by the modified Folin-Ciocalteu method (27). An aliquot of the extract was mixed with 5 mL of Folin-Ciocalteu reagent (previously diluted with water 1:10 v/v) and 4 mL (75 g/L) of sodium carbonate. The tubes were vortexed for 15 s and allowed to stand for 30 min at 40°C for color development. Absorbance was then measured at 765 nm using the Hewlett Packard UV-VS spectrophotometer. Samples of extract were evaluated at a final concentration of 0.1 mg/mL. Total phenolic contents were expressed as mg/g tannic acid equivalent using the following equation based on the calibration curve: $y = 0.1216x$, $r^2 = 0.9365$, where x was the absorbance and y was the tannic acid equivalent (mg/g).

Determination of total flavonoids

Total flavonoids were estimated using the method of Ordonez et al. (28). To 0.5 mL of sample, 0.5 mL of 2% $AlCl_3$ ethanol solution was added. After one hour at room temperature, the absorbance was measured at 420 nm. A yellow color indicated

the presence of flavonoids. Extract samples were evaluated at a final concentration of 0.1 mg/mL. Total flavonoid contents were calculated as quercetin (mg/g) using the following equation based on the calibration curve: $y = 0.0255x$, $r^2 = 0.9812$, where x was the absorbance and y was the quercetin equivalent (mg/g).

Determination of total flavonols

Total flavonols in the plant extracts were estimated using the method of Kumaran and Karunakaran (29). To 2.0 mL of sample (standard), 2.0 mL of 2% $AlCl_3$ ethanol and 3.0 mL (50 g/L) sodium acetate solutions were added. The absorption at 440 nm was read after 2.5 h at 20°C. Extract samples were evaluated at a final concentration of 0.1 mg/mL. Total flavonoid content was calculated as quercetin (mg/g) using the following equation based on the calibration curve: $y = 0.0255x$, $r^2 = 0.9812$, where x was the absorbance and y was the quercetin equivalent (mg/g).

Determination of total proanthocyanidins

Determination of proanthocyanidin was based on the procedure reported by Sun et al. (30). A volume of 0.5 mL of 0.1 mg/mL of extract solution was mixed with 3 mL of 4% vanillin-methanol solution and 1.5 mL hydrochloric acid; the mixture was allowed to stand for 15 min. The absorbance was measured at 500 nm. Extract samples were evaluated at a final concentration of 0.1 mg/mL. Total proanthocyanidin contents were expressed as catechin equivalents (mg/g) using the following equation based on the calibration curve: $y = 0.5825x$, $r^2 = 0.9277$, where x was the absorbance and y is the catechin equivalent (mg/g).

Determination of antioxidant activity

ABTS radical scavenging assay

For ABTS assay, the method of Re et al. (31) was adopted. The stock solutions included 7 mM ABTS solution and 2.4 mM potassium persulfate solution. The working solution was then prepared by mixing the two stock solutions in equal quantities and allowing them to react for 12 h at room temperature in the dark. The solution was then diluted by mixing 1 mL ABTS solution with 60 ml of methanol to obtain an absorbance of 0.706 ± 0.001 units at 734 nm. Fresh ABTS solution was prepared for each assay. Plant extracts (1 mL) were allowed to react with 1 mL of the ABTS solution and the absorbance was taken at 734 nm after 7 min using the spectrophotometer. The ABTS scavenging capacity of the extract was compared with that of BHT and percentage inhibition calculated as ABTS radical scavenging activity (%) = $[(Abs_{control} - Abs_{sample})]/(Abs_{control})] \times 100$, where $Abs_{control}$ is the absorbance of ABTS radical + methanol; Abs_{sample} is the absorbance of ABTS radical + sample extract /standard.

DPPH radical scavenging assay

The effect of extracts on DPPH radical was estimated using the method of Liyana-Pathirana and Shahidi (32). A solution of 0.135 mM DPPH in methanol was prepared and 1.0 mL of this solution was mixed with 1.0 mL of extract in methanol containing 0.02–0.1 mg of the extract. The reaction mixture was vortexed thoroughly and left in the dark at room temperature for 30 min. The absorbance of the mixture was measured spectrophotometrically at 517 nm. Ascorbic acid and BHT were used as references. The ability to scavenge DPPH radical was calculated by the following equation: DPPH radical scavenging activity (%) = $[(Abs_{control} - Abs_{sample})]/(Abs_{control})] \times 100$ where $Abs_{control}$ is the absorbance of DPPH radical + methanol; Abs_{sample} is the absorbance of DPPH radical + sample extract /standard.

Total antioxidant activity (FRAP assay)

A modified method of Benzie and Strain (33) was adopted for the FRAP assay. The stock solutions included 300 mM acetate buffer (3.1 g $C_2H_3NaO_2 \cdot 3H_2O$ and 16 mL $C_2H_4O_2$), pH 3.6, 10 mM TPTZ (2,4,6-tripyridyl-*s*-triazine) solution in 40 mM HCl, and 20 mM $FeCl_3 \cdot 6H_2O$ solution. The fresh working solution was prepared by mixing 25 mL of acetate buffer, 2.5 mL TPTZ, and 2.5 mL $FeCl_3 \cdot 6H_2O$. The temperature of the solution was raised to 37°C before using. Plant extracts (150 µL) were allowed to react with 2850 µL of the FRAP solution for 30 min in the dark condition. Readings of the colored product (ferrous tripyridyltriazine complex) were taken at 593 nm. The standard curve was linear between 200 and 1000 µM $FeSO_4$. Results are expressed in µM Fe (II)/g dry mass and compared with that of BHT, ascorbic acid and catechin.

Proximate analysis

The recommended methods of the Association of Official Analytical Chemists (AOAC, 1999) were used for the determination of moisture, ash, crude lipid, crude fibre and nitrogen content.

Mineral analysis

The automated procedure for determining cations in the plant materials utilizes the reaction

Table 1. Polyphenol contents of the acetone, methanol and water extracts of the leaves of *Bidens pilosa* and *Chenopodium album* (n = 3, X ± SEM).

Phenolics	*Bidens pilosa*			*Chenopodium album*		
	Acetone	Methanol	Water	Acetone	Methanol	Water
Total polyphenol	32.53 ± 1.10[a]	27.08 ± 2.90[a]	6.09 ± 0.28[a]	13.11 ± 0.21[a]	8.61 ± 0.76	7.50 ± 0.80
Flavonoids	2.01 ± 0.07[a]	0.93 ± 0.02	0.77 ± 0.01	1.82[a] ± 0.04	0.80 ± 0.07	0.78 ± 0.02
Proanthocyanidins	2.42 ± 0.13[a]	1.77 ± 0.10	1.69 ± 0.83	4.51 ± 0.51[a]	3.74 ±0.36[a]	1.43 ± 0.50
Total Flavonol	0.31 ± 0.010	0.79 ± 0.06[a]	0.14 ± 0.06	1.34 ± 0.04[a]	0.98 ± 0.41	0.10 ± 0.08

Total polyphenol is expressed as mg tannic acid/g of dry plant material. Flavonoids, proanthocyanidins and total flavonol are expressed as mg quercetin/g of dry plant material.* significantly different from the other at $p < 0.05$.

Table 2. FRAP activity of the acetone, methanol and water extracts of *Bidens pilosa and Chenopodium album.*

Extracts/Standards	*Bidens pilosa*	*Chenopodium album*
Acetone	2431.93 ± 143.95	263.96 ± 29.75
Methanol	561.68 ± 10.57	57.47 ± 2.24
Water	35.70 ± 0.08	23.67 ± 0.00
Ascorbic acid	1632.1 ± 16.95	1632.1 ± 16.95
BHT	63.46 ± 2.49	63.46 ± 2.49
Catechin	972.02 ± 0.61	972.02 ± 0.61
Quercetin	3107.29 ± 31.28	3107.29 ± 31.28

FRAP is expressed in units of µmol Fe(II)/g.

Table 3. Proximate analysis of the leaves of *Bidens pilosa* and *Chenopodium album*.

Constituents	*Bidens pilosa*	*Chenopodium album*
Moisture	80.48 ± 0.48	84.80 ± 1.55
Ash	15.75 ± 0.25	23.25 ± 0.25
Protein	19.13 ± 0.06	26.44 ± 0.20
Fat	6.0 ± 1.0	4.25 ± 0.25
Carbohydrate	37.64 ± 0.52	29.41 ± 0.1
Crude fibre	21.48 ± 2.0	16.65 ± 3.55
Energy (kcal)	281.08 ± 0.62	261.65 ± 0.2

between a particular cation and molybdovanadate to form a complex. The complex is then measured colorimetrically at 420 nm. The elements comprising sodium, calcium, potassium, magnesium, iron, zinc, copper, manganese, potassium, nitrogen and phosphorus were determined in this way.

Other analyses

Determination of alkaloid and saponins were as described by Obadoni and Ochuko (34). Phytate was estimated by the method of Wheeler and Ferrel (35).

Bioassay

The bacterial cultures used in this study were obtained from the Department of Biochemistry and Microbiology, Rhodes University, South Africa. They consisted of five Gram-positive and five Gram-negative strains (Tables 6 and 7). Each organism was maintained on nutrient agar plates and was recovered for testing by growth in nutrient broth for 24 h. Before use, each bacterial culture was diluted 1:100 with fresh sterile nutrient broth (36).

Test organisms were streaked in a radial pattern on sterile nutrient agar plates containing filtered extracts at final concentrations of 0.1, 0.5, 1.0, 2.5 and 5.0 mg/mL (14, 37). Plates containing only nutrient agar and another set containing nutrient agar and the respective solvents served as controls. After inoculation, the plates were incubated at 37°C for 24 to 48 h. Each treatment was performed in triplicate and complete inhibition of bacterial growth was required for an extract to be declared bioactive.

Statistical analysis

The experimental results were expressed as the mean ± standard deviation (SD) of three replicates. Where applicable, the data were subjected to one way analysis of variance (ANOVA) and differences between samples were determined by Duncan's Multiple Range test using the Statistical Analysis System (SAS, 1999) program. Values of $p < 0.05$ were regarded as significant.

RESULTS AND DISCUSSION

At 1 mg/mL, the acetone, methanol, water and ascorbic acid caused DPPH radical scavenging activity at 95.7, 94.2, 91.7 and 99.0%, respectively, for *B. pilosa* while for *C. album* at 1 mg/mL; the results were 62.4, 87.2, 81.7 and 100% for acetone, methanol, water and ascorbic acid, respectively. For BHT, at 1 mg/mL, the DPPH radical scavenging activity was 99.3% (Fig. 1 and 3). The effect of antioxidants on DPPH is thought to be due to their hydrogen donating ability (38). The DPPH radical scavenging abilities of the acetone extract of *B. pilosa* at 1 mg/mL was 95.7% and slightly less than those of ascorbic acid (99%) and BHT (99.3%), showing that the extract has the proton-donating ability and could serve as free radical inhibitor or scavenger, acting possibly as primary antioxidant. The results for the extracts of *C. album* were not as high.

At 1 mg/mL, the acetone extract of *B. pilosa* caused 98.8% ABTS radical scavenging inhibition while the methanol, water, and BHT caused inhibition at 99.1, 95.7 and 99.3%, respectively. For *C. album* at the same concentration, the results were 93.1, 99.4, 95.0 and 99.3% for acetone, methanol, water and BHT, respectively. It must be stated, however, that at 0.75 mg/mL, the percentage inhibition for methanol extract of *B. pilosa* was 99.9% while

Table 4. Macro and micro elements constituents of the leaves of *Bidens pilosa* and *Chenopodium album.*

Macro and micro elements (mg/100 g d.w.)	*Bidens pilosa*	*Chenopodium album*
Magnesium	0.640	0.719
Calcium	1.971	2.172
Potassium	3.285	6.938
Phosphorus	0.519	0.317
Sodium	0.053	0.370
Iron (ppm)	986	255
Zinc	51	50
Copper	24	13
Manganese	115	118
Total Kjeldahl nitrogen	3.05	4.23

Table 5. Analysis of antinutrients contents of *Bidens pilosa* and *Chenopodium album.*

Anti-nutrients	*Bidens pilosa*	*Chenopodium album*
Alkaloids	0.83 ± 0.40	1.8 ± .45
Saponins	4.5 ± .02	5.25 ± 0.01
Phytate	5.59 ± 0.02	18.06 ± 0.01

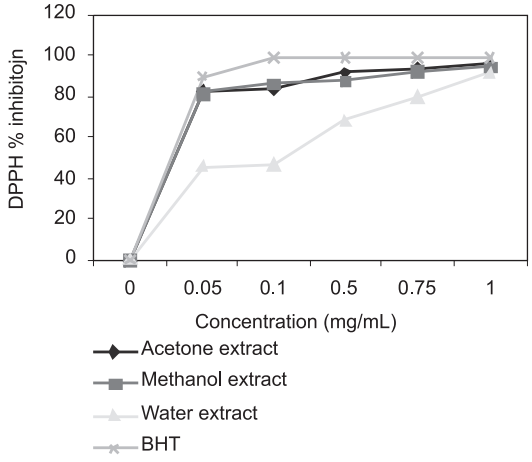

Figure 1. DPPH radical scavenging activity of *B. pilosa*

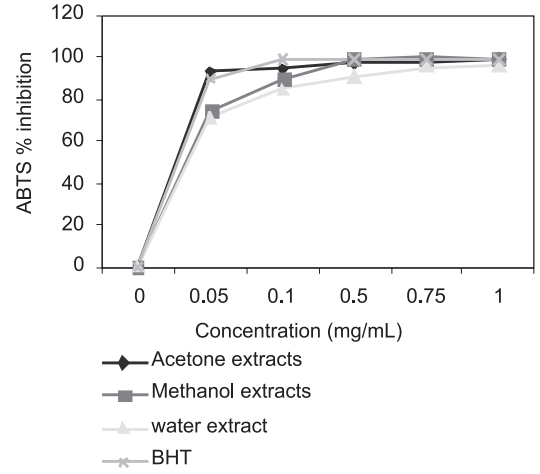

Figure 2. ABTS radical scavenging activity of *B. pilosa*

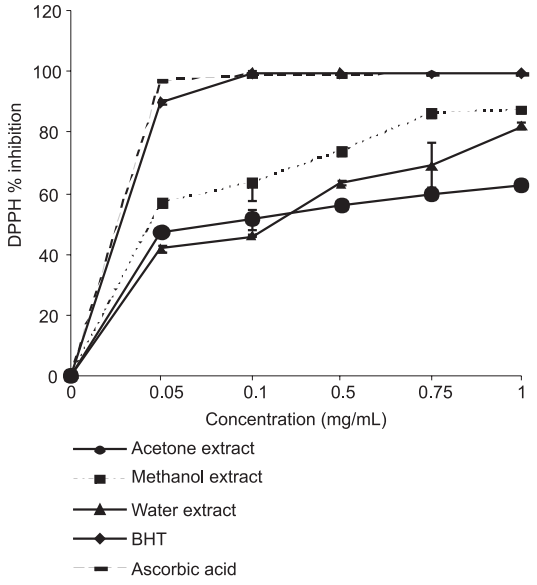

Figure 3. DPPH radical scavenging activity of *C. album*

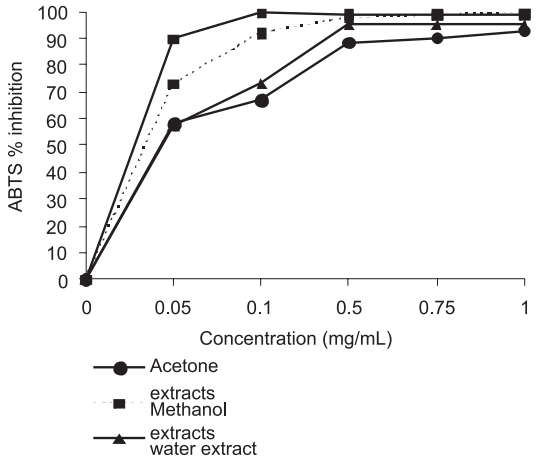

Figure 4. ABTS radical scavenging activity of *C. album*

for the acetone extract it was 98.6% (Figs. 2 and 4). Proton radical scavenging is an important attribute of antioxidants. ABTS, a protonated radical, has characteristic absorbance maximum at 734 nm which decreases with the scavenging of the proton radicals (39). Higher concentrations of the extracts were more effective in quenching free radicals in the system. At 1 mg/mL concentration, all the extracts for the two plants produced similar or equal ABTS radical scavenging activity. The scavenging of the ABTS radical by the extracts at 1 mg/mL was found to be slightly higher than that of DPPH radical. Factors like stereoselectivity of the radicals or the solubility of the extract in different testing systems have been reported to affect the capacity of extracts to react and quench different radicals (40). Wang et al. (41) found that some compounds which have ABTS scavenging activity did not show DPPH scavenging activity.

With respect to the total polyphenol, flavonoids and proanthocyanidins, the acetone and methanol extracts of *B. pilosa* and *C. album* had higher content of this constituent than those of water extracts. Even for flavonols, the pattern is the same except that the methanol extracts of *B. pilosa* was

higher than that of acetone (Tab. 1). Polyphenols are the major plant compounds with antioxidant activity. This activity is believed to be mainly due to their redox properties (42), which play an important role in adsorbing and neutralizing free radicals, quenching singlet and triplet oxygen, or decomposing peroxides. The results strongly suggest that phenolics are important components of these plants, and some of their pharmacological effects could be attributed to the presence of these valuable constituents. The results from this study showed that the polyphenolic contents of *B. pilosa* were higher than those of *C. album*. This may have accounted for relatively higher antioxidant activities of *B. pilosa* over *C. album*.

The ferrous reducing antioxidant power (FRAP) value for the plants showed that acetone extracts had much higher activity than those of methanol and water extracts. The FRAP value for the acetone extract of *B. pilosa* (2431.9) was higher than those of BHT (63.5), ascorbic acid (632) and catechin (972) but lower than those of quercetin (3107.3). The FRAP value for water extracts of both plants was the lowest (Tab. 2). The antioxidant potentials of the extracts of the leaves of the plants were estimated from their ability to reduce TPTZ-Fe(III) complex to TPTZ-Fe(II). According to recent reports, a highly positive relationship between total phenols and antioxidant activity appears to be the

Table 6. Antibacterial activity of the leaves extracts of *Bidens pilosa*.

Bacterial species	Gram +/–	Minimum inhibitory concentration (mg/mL)				
		Acetone	Methanol	Water	Ch.	St.
Bacillus cereus	+	0.1	0.5	2.0	< 2	< 2
Staphylococcus epidermidis	+	1.0	na	2.0	< 2	< 2
Staphylococcus aureus	+	1.0	na	5.0	< 2	< 2
Micrococcus kristinae	+	0.1	1.0	2.0	< 2	< 2
Streptococcus pyrogens	+	0.1	1.0	na	< 2	< 2
Escherischia coli	–	0.1	1.0	na	< 2	< 2
Salmonella pooni	–	0.5	na	na	< 2	< 2
Serratia marcescens	–	1.0	5.0	5.0	< 2	< 2
Pseudomonas aeruginosa	–	na	na	na	< 20	< 5
Klebsiella pneumonae	–	na	na	na	< 2	< 2

na = not active; Ch. = chloramphenicol; St. = streptomycin

Table 7. Antibacterial activity of the leaves extracts of *Chenopodium album*.

Bacterial species	Gram +/–	Minimum inhibitory concentration (mg/mL)				
		Acetone	Methanol	Water	Ch.	St.
Bacillus cereus	+	2.0	5.0	0.5	<2	<2
Staphylococcus epidermidis	+	na	5.0	0.5	<2	<2
Staphylococcus aureus	+	na	na	1.0	<2	<2
Micrococcus kristinae	+	1.0	5.0	0.5	<2	<2
Streptococcus pyrogens	+	na	na	0.5	<2	<2
Escherischia coli	–	0.1	na	1.0	<2	<2
Salmonella pooni	–	5.0	5.0	1.0	<2	<2
Serratia marcescens	–	na	na	1.0	<2	<2
Pseudomonas aeruginosa	–	na	na	na	<20	<5
Klebsiella pneumonae	–	na	na	na	<2	<2

na = not active; Ch. = chloramphenicol; St. = streptomycin

trend in many plant species (27). The result showed that FRAP activity was the highest in the acetone extract followed by methanol and least in water extract of the plants. It thus meant that these plants exhibited similar activity.

The proximate analysis showed the percentage moisture content, ash content, crude protein, crude lipid, crude fibre and carbohydrate of the leaves of *B. pilosa* as 80.5, 15.8, 19.1, 6.0, 37.6 and 21.5%, respectively, while its calorific value was 281.1 Kcal/100 g (Tab. 3). Elemental analysis in mg/100 g d.w. indicated that the leaves contained sodium (0.05), potassium (3.2), calcium (2.0), magnesium (0.6), iron (986), zinc (51), phosphorus (0.5), copper (24), manganese (115), and nitrogen (3.1) (Tab. 4). In the case of *C. album*, analysis showed the percentage moisture content, ash content, crude protein, crude lipid, crude fibre and carbohydrate of the leaves as 84.8, 23.3, 26.4, 4.3, 29.4 and 16.6%, respectively, while its calorific value was 261.7 Kcal/100 g (Tab. 3). Elemental analysis in mg/100 g d.w. indicated that the leaves of *C. album* contained sodium (0.4), potassium (6.9), calcium (2.2), magnesium (0.7), iron (255), zinc (50), phosphorus (0.3), copper (13), manganese (118), and nitrogen (4.2) (Tab. 4). The results of proximate composition of the leaves of *B. pilosa* (80.5) and *C. album* (84.8) show high moisture content (Tab. 3). This is within the reported range (81.4–90.3%) in some Nigerian green leafy vegetables (13). Ash content, which is an index of mineral contents in biota, is 15.8% d.w. in *B. pilosa* and 23.3% d.w. in *C. album*. These compare favorably with the values reported for *Ipomea batatas* (11.10%), *Vernonia colorate* (15.86%) and *Moringa oleifera* (15.09% d.w.) (8, 43). The value for *C. album* is also, higher than that of some Nigerian leafy vegetable such as *Ocimum gratissium* (18.00% d.w.) *and Hibiscus esculentus* (8.00% d.w.) (9). The crude protein content of *B. pilosa* (19.1% d.w.) and *C. album* (26.4% d.w.) are higher than protein content of *Momordica foecide* (4.6%) leaves consumed in Nigeria and Swaziland (11, 44, 45). The crude protein content of *C. album* compared favorably with those of *I. batatas* (24.85% d.w.), *Amaranthus candatus* (20.5% d.w.), *Piper guineeses* (29.78% d.w.) and *T. triangulare* (31.00% d.w.) (9, 43, 46). According to Pearson (47), plant food that provides more than 12% of its calorific value from protein is considered a good source of protein. Therefore, the protein content of the leaves of the investigated plants will go a long way in meeting the protein requirement of the local people.

The crude lipid content of *B. pilosa* (6.0 % d.w.) and *C. album* (4.3% d.w.) is slightly lower than the reported values (8.3–27.0% d.w.) in some vegetables consumed in West Africa (48, 49). However, it compares favorably with 4.2% reported for *Calchorus africanum* leaves and 1.85–8.71% d.w. in some edible green leafy vegetables of Southern India and Nigeria (50, 51). The carbohydrate content of *B. pilosa* (37.6% d.w.) and *C. album* (29.4% d.w.) is higher than 20 and 23.7% reported for *Senna obtusfolia* and *Amaranthus incurvatus* leaves, respectively (11, 52). This is, however, lower than reported values for *Corchorus tridens* (75.0% d.w.) and sweet potatoes leaves (82.8%) (53). The crude fibre content of *B. pilosa* (21.5% d.w.) and *C. album* (16.6% d.w.) is high when compared to *Ipomea batatas* (7.20%), *T. triangulare* (6.20%) *P. guineensis* (6.40%), *Corchorus olitorius* (7.0%), and *Vernonia amygdalina* (6.5%) (9, 43). Adequate intake of dietary fibre can lower the serum cholesterol level, risk of coronary heart disease, hypertension, constipation, diabetes, colon and breast cancer (54, 55). The RDA of fibre for children, adults, pregnant and lactating mothers are 19–25, 21–38, 28 and 29 g, respectively. The estimated calorific value for *B. pilosa* (281.1 Kcal/100 g d.w.) and *C. album* (261.7% d.w.) leaves compare favorably to 248.8–307.1 Kcal/100 g d.w. reported in some Nigerian vegetables (13, 43, 56). Asibey–Berko and Tayie (53) also reported comparable energy content in some Ghanaian green leafy vegetables. Thus, the calorific value agreement with general observation that vegetables have low energy values (57).

The mineral composition of *B. pilosa* and *C. album* is shown in Table 4. The Na/K ratio in the body is of great concern for prevention of high blood pressure. Na/K ratio less than one is recommended (58). Therefore, consumption of *B. pilosa* and *C. album* would probably reduce high blood pressure diseases because their Na/K is less than one. Iron content of the leaves of *B. pilosa* (986 mg/100 g) and *C. album* (255 mg/100 g) is very high when compared with the value reported in *I. batatas* (16.00 mg/100 g) (43). Iron is an essential trace element for hemoglobin formation, normal functioning of the central nervous system and in the oxidation of carbohydrates, protein and fats (13, 59). The zinc content of *B. pilosa* (51 mg/100 g) and *C. album* (50 mg/100 g) compares favorably to most values for green leafy vegetables reported in the literature (11, 60). Zinc is involved in normal function of immune system.

The chemical composition in mg/100 g d.w. for alkaloid, saponins, and phytate were 0.8, 4.5 and 5.6, respectively, for *B. pilosa*. In the case of *C.*

album, these values were alkaloids (1.8), saponins (5.3) and phytate (18.1) (Tab. 5). Analysis of the antinutrient contents of the plant showed that alkaloid level in *B. pilosa* (0.8) and *C. album* (1.8 mg/100g) is lower than the values reported for the leafy vegetables like *Aspilia africana*, *Bryophyllum pinnatum*, *Cleome rutidosperma* and *Emilia coccinea* consumed in Nigeria (10, 13, 61). The levels of saponins in these plants are much less than the value reported for some medicinal plants used in Nigeria. Although the phytate level in *C. album* (18.1 mg/100 g) is higher than the value reported for *I. batatas* and *G. africana* leaves (43, 62); this value is still within the tolerable limits and can easily be detoxified by soaking, boiling or frying (13, 63–65). The acetone extract of *B. pilosa* has activity against all the organisms used in this study except *Pseudomonas aeruginosa* and *Klebsiella pneumoniae*. The water extract of *C. album* also showed similar activity. While the methanol extract of *B. pilosa* showed activity against 5 organisms, the water extract showed activity against all Gram positive strains except *Streptococcus pyrogens*. The acetone and methanol extract of C. album are not as active (Tabs. 6 and 7). The high activity shown by the extracts of these 2 plants may have justified their use for medicinal purposes. It has also been shown that the plant extracts were active against most of the Gram-positive strains and less of the Gram-negative strains. This observation therefore supports the fact that, in general, the Gram-negative bacteria are less susceptible to antibacterial effect than the Gram positive ones (66, 67). Since these extracts show some activity against some of the organisms used in this study; the use of this plant for medicinal purpose may be justified.

The results of this study showed that the leaves of *B. pilosa* and *C. album* contain appreciable amount of proteins, fat, fibre, carbohydrate and calorific value, mineral elements, polyphenols, and generally low level of toxicants. Their antioxidant and antibacterial activities further lend credence to the biological value of this plant. Thus, it can be concluded that *B. pilosa* and *C. album* leaves can contribute significantly to the nutrient requirements of man and should be used as a source of nutrients to supplement other major sources. The plants also have high biological activities hence may be of great medicinal value.

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