Anti-Oxidant and Anti-Microbial Activities of the Root and Leaf Extracts of *Ageratum conyzoides* L.

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Summary

Plants and plant-based pharmaceuticals are the basis of many of the modern days drugs used for treatment of various ailments. The present investigations assess the qualitative and quantitative phytochemicals, DPPH antioxidant and antimicrobial activities of the root and leaf extracts of *Ageratum conyzoides*. The qualitative screening of the phytochemicals allows us to describe alkaloids, flavonoids, tannins, phenols, terpenoids, saponins, steroids and glycosides. The antimicrobial activity test of the root and leaf extracts showed considerable antibacterial and antifungal activities against clinical isolates of *Staphylococcus aureus*, *Bacillus subtilis* (gram positive), *Escherichia coli*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Salmonella typhi* (gram negative) and strains of fungi i.e. *Candida albicans*, *Aspergillus niger*, *Rhizopus stolon* and *Penicilliumnotatum*. The root extracts showed higher antibacterial and antifungal activities on the test organisms than the leaf extracts. The phenolic content, flavonoid concentrations and antioxidant activity (free radical scavenging assay) were carried out using Folin-Ciocalteu reagent, aluminum chloride and 2,2 diphenyl-1-picrylhydrazyl (DPPH). The total phenolic content ranged from 20.43±0.49 to 105.4±0.3 µg/mL for the root extracts and 15.31±0.1 to 94.34±0.6 µg/mL for the leaf extracts expressed as gallic acid equivalents. Flavonoid concentrations ranged from 38.92±0.18 to 243±0.32 µg/mL and 32.65±0.33 to 182.45±0 µg/mL for the root and leaf extracts expressed as quercetin equivalents respectively. The IC_{50} antioxidant activities ranged from 7.34 to 14.62µg/mL and 9.18 to 15.88 µg/mL for the root and leaf extracts respectively.

Key words

phytochemicals, phenolic content, flavonoid concentration

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Introduction

Worldwide, there is a prevalence of infectious diseases which accounts for over a third of death cases. Plants are great reservoir of chemical compounds which are of high benefits to mankind. Although orthodox medical practice is generally acceptable, alternative health care is still relied on in many parts of the world (O'Brien, 2004; Leckridge, 2004). In developing countries, traditional herbal medicine is often used side by side with western therapies. Due to the ease of accessibility and affordability, herbal medicine is sometimes preferred over western medicine in developing countries (Busia, 2005; Igoli et al, 2005). The rising prevalence of multi-antibiotics resistant strain of pathogenic organisms worldwide has led to the search for alternative antimicrobial agents and tremendous efforts have been expended to discover and evaluate new antioxidants, antibacterial and antifungal compounds from different kinds of natural sources like soil, microorganisms, animals and plants (Nagesh et al., 2012).

Plant phenolics constitute one of the major groups of compounds acting as primary antioxidants or free radical terminators. Phenolic and polyphenolics (polymeric phenolic) can provide relief from certain physical ailments and degenerative diseases in humans, including the reduction of cardiovascular diseases and certain cancers (Scalbert et al., 2002; Arts and Hollman, 2005). Therefore, it is not surprising that the extraction and analysis of phenolics from plants and other food sources have been extensively studied (Naczk and Shahidi, 2004; Dai and Mumper, 2010). The phenols contain hydroxyls that are responsible for the radical scavenging effect mainly by redox reactions (Rice-Evans et al, 1997). Plants consumed by humans may contain thousands of different phenolic compounds, the effects of which have generated current interest, due to their antioxidative and possible anticarcinogenic activities. Phenolics may inhibit carcinogenesis by affecting the molecular events in the initiation, promotion, and progression stages of oxidation (Malencic et al., 2007).

Ageratum conyzoides is an erect, annual, branched, slender, hairy and aromatic herb that grows in the tropics. It is very common in West Africa, Australia, some parts of Asia and South America.

The weed has been known since ancient times for its curative properties and has been utilized for treatment of ailments like burns, wounds, infectious diseases, bacterial infections, arthritis, headaches and dyspnea, pneumonia, inflammatory, asthmatic, spasmodic and haemostatic effects, stomach ailments, gynaecological diseases, leprosy and other skin diseases (Marques et al., 1988; Gbolade et al., 1999; Oladejo et al., 2003). Therefore, the antioxidant activities and antimicrobial potential of *Ageratum conyzoides* extracts are studied in this research.

Materials and Methods

Collection and Authentication of Plant Materials

Fresh roots and leaves of *Ageratum conyzoides* were collected from the premises of the University of Ilorin, Ilorin, Nigeria. The plant samples were authenticated at the herbarium of the Department of Plant Biology, Faculty of Life Sciences, University of Ilorin, Nigeria and a specimen copy (UIL001/1108) was deposited. The plant materials were air dried at room temperature for about two weeks and pulverized.

Preparation of Extracts

The pulverized sample (300 g) was extracted with cold n-hexane in a stopper glass container for five days with frequent agitation (Ncube et al., 2008). The extract was thereafter decanted, filtered by cotton-plugged funnel and filter paper. The extract was concentrated in vacuum. The dried concentrated extract was stored in an opaque glass bottle for further works. Subsequent extraction using the above procedure was done to obtain crude ethyl acetate and ethanol extracts successively and respectively.

Phytochemical Screening

Qualitative Analysis

Phytochemical screening of the n-hexane, ethylacetate and ethanol extracts of *Ageratum conyzoides* root and leaf were carried out using standard procedures to identify the secondary metabolites present in the plant extracts (Harborne, 1973; Sofowora, 1993). Small quantities of the extracts were used in the analysis and the major secondary metabolites analysed were alkaloids, saponins, terpenoids, tannins, polyphenols, flavonoids, steroids and cardiac glycoside.

Quantitative Analysis

Estimation of Total Phenolic Content

The concentration of phenolic compounds in the plant extracts was determined using spectrophotometric method (Singleton et al., 1999). Methanolic solution of the extracts in concentrations of 1, 5 and 10 µg/mL was used in the analysis. The reaction mixture was obtained by mixing 0.5 mL of methanolic solution of extract with 2.5 mL of 10% Folin-Ciocalteu's reagent dissolved in water and add 2.5 mL 7.5% $NaHCO_3$. A blank was concomitantly prepared, containing 0.5 ml methanol, 2.5 mL of 10% Folin-Ciocalteu's reagent dissolved in water and 2.5 mL of 7.5% of $NaHCO_3$. The samples thereafter underwent incubation in a thermostat water bath at 45°C for 45 min. The absorbance of the sample mixture was then determined using spectrophotometer at λmax = 765 nm. The samples were prepared in triplicate for each analysis and the mean value of absorbance was obtained. The stock solution of gallic acid was prepared in methanol (5.0 mg/mL). Dilutions were made to obtain concentrations of 25, 12.5, 6.25, 3.13, 1.56 and 0.78 µg/mL, and the calibration line was constructed on graph. Based on the measurement of the absorbance, the concentrations of phenolic compounds were determined (µg/mL) from the calibration curve. The phenolic compounds content of the extracts was expressed in gallic acid equivalent (µg of GA/g of extract).

Estimation of Flavonoid Concentrations

Total flavonoids in the plant extracts were determined by spectrophotometric method (Quettier et al., 2000). Quercetin was used as standard and flavonoid concentrations were measured as quercetin equivalent. Methanolic solutions of the extracts were made of concentrations 1.0, 5.0 and 10.0 µg/mL which were used in the analysis. 1 ml of the prepared methanolic solution

of the extracts was added to 1 ml of 2% $AlCl_3$ solution prepared in methanol. The samples underwent incubation for an hour at room temperature. The absorbance was then determined using spectrophotometer at λmax = 415 nm. The samples were prepared in triplicate for each analysis and the mean value of the absorbance was calculated. The quercetin calibration curve was constructed by preparing quercetin solutions of concentrations varying from 10 to 100µg/mL in methanol. Based on the measured absorbance, the concentration of flavonoids was determined (µg/mL) on the calibration line and flavonoid contents in extracts were expressed in terms of quercetin equivalent (µg of QE/g of extract).

Evaluation of Antioxidant Activity

The ability of the plant extract to scavenge DPPH (2,2-diphenyl-1-picrylhydrazyl) free radicals was assessed by the standard method (Tekao et al., 1994; Kumarasamy et al., 2007). The stock solution of extracts was prepared in methanol to obtain a concentration of 1 mg/mL. Dilutions were then made to obtain concentrations of 500, 250, 125, 62.5, 31.25, 15.62, 7.81, 3.90, 1.99 and 0.97 µg/mL of the extract. Diluted solutions (1 ml) were mixed with 1 ml of methanolic solution of DPPH of 1 mg/mL concentration. After 30 min incubation in darkness at room temperature (23°C), the absorbance was measured at 517 nm. A control sample containing all the reagents except the extract was set alongside. Percentage inhibition was calculated using the equation below, whilst IC_{50} values were estimated from the % inhibition versus concentration plot, using a non-linear regression algorithm.

$$\% \ inhibition = ((A_{\ of\ control} - A_{\ of\ sample}) / A_{\ of\ control}) \times 100$$

Antimicrobial Assay

Cultures of six human pathogenic bacteria made up of four gram negative and two gram positive were used for this antibacterial assay. These were: *Salmonella typhi* (UCH 4801), *Escherichia coli* (UCH 0026), *Pseudomonas aeruginosa* (UCH 1102) and *Klebsiellaepneumoniae* (UCH 2894) as the gram-negative bacteria, while *Bacillus subtilis* (UCH 7423) and *Staphylococcus aureus* (UCH 2473) were the gram-positive bacteria used. Four fungi, which includes *Candida albicans*, *Aspergillus niger*, *Rhizopus stolon* and *Penicilliumnotatum* were utilized for antifungal assay. All the microorganisms used were clinical strains from the Medical Microbiology Laboratory of the University College Hospital, Ibadan, University of Ibadan, Nigeria. Gentamycin (10 µg/mL) and Tioconazole (0.7 µg/mL) were used as reference (standard) compound in the study for antibacterial and antifungal assay respectively.

Agar Diffusion-Pour Plate Method (bacteria)

An overnight culture of each of the organisms was prepared by taking a loop full of the organism from stock and inoculated each into the sterile nutrient broth of 5 mL and then each incubated for 18-24 hours at 37°C. From overnight culture, 0.1 mL of each organism was taken and put into 9.9 mL of sterile distilled water to get 1:100 (10^{-2}) of the dilution of the organism. From the diluted organism (10^{-2}), 0.2mL was taken into the prepared sterile nutrient agar which was at 45°C, and aseptically poured into sterile Petri dishes and allowed to solidify for about 45-60 minutes. Using a sterile cork borer of 8- mm diameter, wells were made according to the number of graded concentration of the sample. In each well, different graded concentrations of the sample were produced, and this was done in duplicates. The plates were allowed to stay on the bench for 2 hours to allow pre-diffusion. The plates were incubated uprightly in the incubator for 18-24 hours at 37 °C (Oloyede and Onocha, 2010).

Agar Diffusion-Surface Plate Method (fungi)

A sterile sabouraud dextrose agar (62 g/L) was prepared accordingly and aseptically poured into the sterile plates in duplicates and left to solidify properly. 0.2 mL of the 10^{-2} of the organism was spread on the surface of the agar using Petri dish to cover the whole surface of the agar. 8 wells were bored using a sterile corkborer of 8- mm diameter. The graded concentrations of the extract were put into the wells accordingly, including the controls. All the plates were left on the bench for 2 hours to allow the extract to diffuse properly into the agar (pre-diffusion). The plates were incubated at 25°C for 72 hours. Solvent of extraction, n-hexane, ethyl acetate and ethanol were used as control, while gentamycin (10µg/mL) and tioconazole (0.7µg /mL) were used as standard reference drugs in the study (Bayer et al., 1986).

Results and Discussion

Phytochemical Screening

Qualitative Analysis

The phytochemical constituents of n-hexane, ethylacetate and ethanol extracts of the root and leaf of *Ageratum conyzoides* are shown in Table 1. Alkaloids, flavonoids, tannins, phenols and glycosides were present in all extracts, while steroids and terpenoids were absent in the root extracts.

Phytochemicals act in numerous ways to assist the body in combating diseases and health challenges. They combine with some biomolecules to neutralize the activity of scavenging free radicals before they can cause damage within the body system (Okaka and Okaka, 2001).

Alkaloids have been used as analgesic, antispasmodic and bactericidal agent (Okwu and Okwu, 2004), while flavonoids have shown antibacterial, anti-inflammatory, antiallergic as well as the ability to scavenge hydroxyl radicals, super oxide anions and lipid peroxy radicals (Okwu and Okwu, 2004; Alan, 1996). Tannins have astringent properties, hasten the healing of wounds and inflamed mucous membrane (Okwu and Okwu, 2004). The presence of tannins and saponins in the present study could account for the use of *Ageratumconyzoidesin* treating wounds and prevention of blood loss etc (Iwu, 1993; Onwuka, 2005). Saponins have both hypocholesterolemic and antidiabetic properties while triterpenoids have analgesic and anticancer properties (Ali et al., 2008). Glycosides present in drugs could be dangerous but the concentration found in the *Ageratum conyzoides* extracts is very low and should not pose a problem to consumers when prescribed by traditional healers. Steroidal compounds are available in leaf extracts in low concentration and are important in pharmacy due to their relationship with sex hormones (Okwu, 2001).

Table 1. Phytochemicals in the root and leaf extracts of *Ageratum conyzoides*

Phytochemicals	Leaf extract			Root extract		
	n-H	E.A	ETOH	n-H	E.A	ETOH
Alkaloids	+	+	++	+	+	++
Flavonoids	+	+	++	+	+	++
Tannins	+	+	+++	+	+	+
Steroids	+	+	+	-	-	-
Saponins	+	+	+	-	+	+
Terpenoids	+	+	+	-	-	-
Phenols	+	+	+	+	+	+
Glycosides	+	+	+	+	+	+

Key: n-H n-hexane, E.A Ethyl acetate, ETOHEthanol
+++ = High concentration, ++ = Moderate concentration, + = Low concentration, – = Absent

The presence of phenol and other phytochemicals in all the investigated samples indicated that the plant, *Ageratum conyzoides* can be used as an antioxidant and antimicrobial agents (Okwu and Okwu, 2004).

Quantitative Analysis

Total Phenolic Compounds

Total phenolic content of the crude extracts of *Ageratum conyzoides* root and leaf using Folin-Ciocalteu's reagent was expressed in gallic acid equivalent. The absorbance of serial concentrations of gallic acid was plotted against concentration to yield a linear calibration curve of gallic acid (y = 0.0053x) with correlation coefficient, R^2 = 0.9842 (Figure 1). From the standard curve equation, y denotes the absorbance and x denotes the total phenolic contents in the extracts at different concentrations. The values obtained were expressed as µg of GA/g of extracts (Table 2).

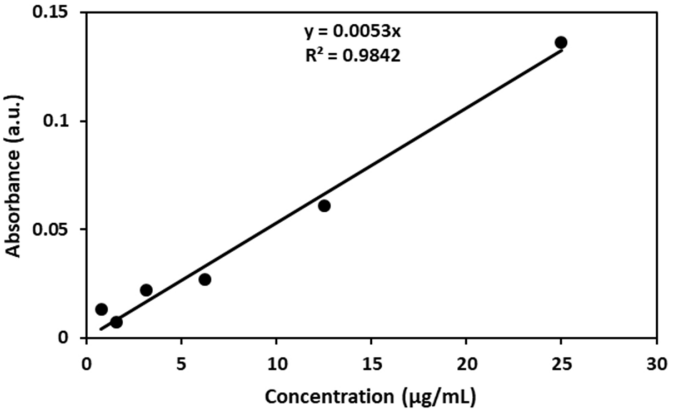

Figure 1. Linear curve of concentration of gallic acid standard (µg/mL) versus absorbance for determination of phenolic content of the root and leave extracts of *Ageratum conyzoides*

Table 2. Total phenolic content of roots and leaf of *Ageratum conyzoides* expressed in terms of gallic acid equivalent (µg/g of extract)

Concentration (µg/mL)	Solvents	Extracts	
		Root	Leaf
1.0	n-hexane	20.75±0.1	15.31±0.1
	Ethyl acetate	25.14±0.1	16.79±0.3
	Ethanol	41.98±0.7	37.73±0.3
5.0	n-hexane	33.84±0.2	21.57±0.2
	Ethyl acetate	38.74±0.3	25.78±0.3
	Ethanol	56.67±0.5	49.18±0.4
10.0	n-hexane	51.05±0.3	40.43±0.2
	Ethyl acetate	60.06±0.1	58.24±0.1
	Ethanol	105.4±0.3	94.34±0.6

Total phenolic contents of the examined extracts ranged from 20.75±0.1 to 105.4±0.3 µg/mL for the root extracts and 15.31± 0.1 to 94.34±0.6 µg/mL for the leaf extracts. The ethanolic root extract at 10.0 µg/mL recorded the highest phenolic content of 105.41±0.3 µg/mL followed by the ethanolic leaf extract with 94.34±0.6 µg/mL at the same concentration. As the polarity of the solvent extraction decreases, the quantity of phenolic compounds extracted decreases, hence ethyl acetate extract has higher phenolics content than n-hexane extract but less than that of ethanol. This trend was observed for leaf extracts also. Hexane extracts of the root and leaf recorded the lowest phenolic contents, 20.75±0.1 µg/mL and15.31±0.1 µg/mL respectively. The total phenolic contents of *Ageratum conyzoides* extracts depend on its concentration and polarity of extraction solvent.

High solubility of phenolic compounds in polar solvents provides high concentration of these compounds in polar extracts (Mohsen and Ammar, 2008; Zhou and Yu, 2004). The ethanol extracts of the root and leaf showed the highest concentration of phenolic compounds and as the polarity of solvent of extraction reduces, the quantity of extracted phenolic compounds reduces, too. The rich endowment of the root of *Ageratum conyzoides* with phenolic compounds is elucidated by high antimicrobial and antioxidant activities displayed by root extracts over those of leaf.

The high phenolic content of the ethanol root extract of *Ageratum conyzoides* from this study can explain its high free radical scavenging activity.

Flavonoids Concentration

The absorbance of serial concentration (10, 20, 30, 40, 50,100) of quercetin standard was plotted against concentration to yield a linear curve of quercetin equivalent (y = 0.0034x and R^2 = 0.9918) of QE/g of extract (Figure 2). Total flavonoid concentration in the examined extracts ranged from 38.92±0.2 to 243.02±0.3 µg/mL for root extracts and 32.65±0.3 to 182.45±0.1 µg/mL for leaf extracts respectively (Table 3). The total flavonoid concentration of *Ageratum conyzoides* is concentration and polarity dependent (the higher the polarity of the solvent of extraction and concentration of extracts, the higher the flavonoid concentration obtained). This observation is supported by previous work as reported by Benavente-Garcia et al. (1997).

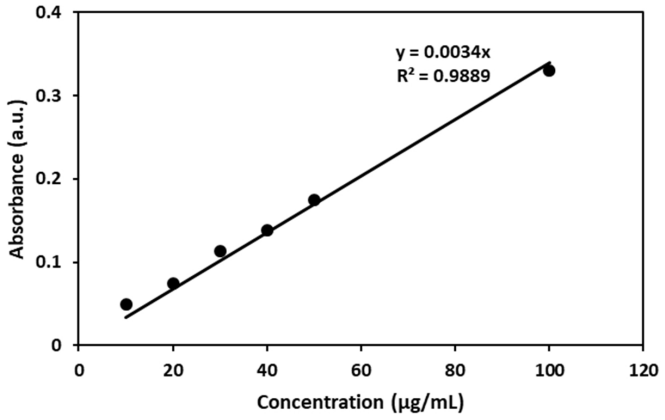

Figure 2. Linear curve of quercetin concentration (µg/mL) versus absorbance for determination of Flavonoid concentration of the root and leaf extracts of *Ageratum conyzoide*

Flavonoids are the most common and widely distributed group of phenolic compounds found in plants, fruits and vegetable. The antioxidant activities of flavonoids in *Ageratum conyzoides* may operate on several different mechanisms such as scavenging of free radicals, chelation of metal ions and inhibition of enzymes responsible for free radical generation (Shan et al., 2005). Several studies reported that flavonoids present in herbs significantly contributed to their antioxidant properties (Sathyaprabhaa et al., 2011). Depending on their structure, flavonoids are able to scavenge practically all known reactive oxygen species (ROS). Many pharmacological effects of phenolic compounds and flavonoids are linked to their ability to act as strong antioxidants

Table 3. Flavonoid concentration of root and leaf of *Ageratum conyzoides* expressed in terms of quercetin equivalent (µg/g of extract)

Concentration (µg/mL)	Solvents	Extracts	
		Root	Leaf
1.0	n-hexane	38.92±0.2	32.65±0.3
	Ethyl acetate	56.27±0.4	49. 90±0.4
	Ethanol	70.49±0.2	68. 92 ±0.2
5.0	n-hexane	95.0±0.1	45.38±0.2
	Ethyl acetate	99. 90±0.2	76. 96±0.5
	Ethanol	137.84±0.2	136.61±0.2
10.0	n-hexane	131.18±0.4	55.49±0.2
	Ethyl acetate	138. 92±0.4	134.04±0.2
	Ethanol	243.02±0.3	182.45±0.1

and free radical scavengers, to chelate metals, and to interact with enzymes, adenosine receptors (Siddique et al., 2010). The high contents of flavonoid concentration in *Ageratum conyzoides* (Table 3) explained its high radical scavenging activity which is in agreement with other studies reporting flavonoid concentration as a function of antioxidant activities in different plant parts (Blois, 1958).

Antioxidant Assay

The ability of the root and leaf extracts of *Ageratum conyzoides* to scavenge DPPH free radicals was expressed by IC_{50} value (µg/mL), i.e. the minimum concentration of the extract capable to scavenge the DPPH radical by 50%. Sample with IC_{50} less than 50 µg/mL is regarded as a very strong antioxidant sample, 50-100 µg/mL is a strong antioxidant, 101-150 µg/mL is a medium antioxidant, and sample with IC_{50} greater than 150 µg/mL is a weak antioxidant (Canadanovic-Brunet et al., 2008). The largest capacity to neutralize DPPH radicals was found for ethanol extracts (root and leaf) which neutralized 50% of free radicals at the inhibition concentrations of 7.34 and 9.18µg/mL respectively (Figures 3 and 4). IC_{50} of 11.63 and 12.97 µg/mL recorded for ethyl acetate extracts for root and leaf extracts respectively were equally high, while the inhibition activities reported for the n-hexane extracts of root and leaf were 14.62 and 15.88µg/mL respectively. In comparison to IC_{50} of ascorbic (5.66 µg/mL), ethanol root extract of *Ageratum conyzoides* manifested the strongest capacity for neutralization of DPPH radicals. Numerous investigations of qualitative composition of plant extracts revealed the presence of high concentrations of phenols in the extracts obtained using polar solvents (Tosun et al., 2009). The extracts with the highest antioxidant activities have the highest concentration of phenols (Table 2). Phenols are very important plant constituents because of their scavenging ability on free radicals by hydroxyl groups. Therefore, the phenolic quantity of the plant may contribute directly to its antioxidant ability.

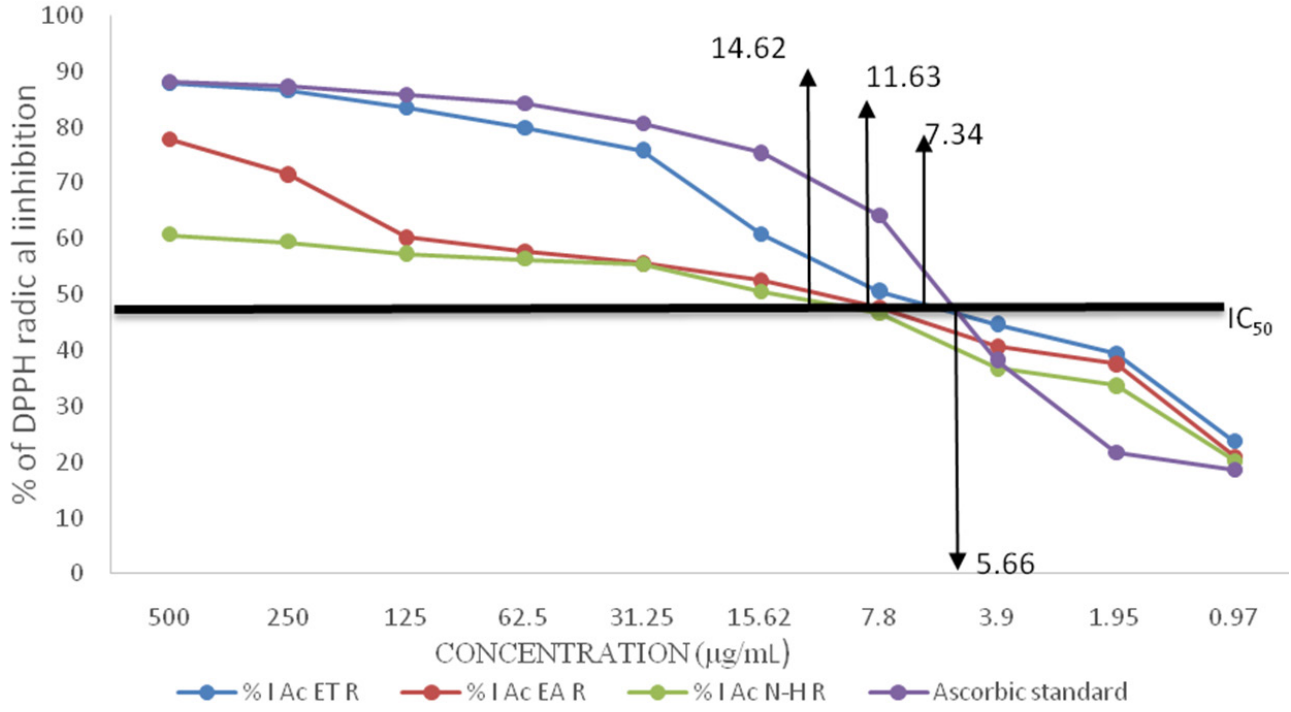

Figure 3. Plot of % Inhibition of DPPH radical versus concentrations used to evaluate the antioxidant activity of Root extracts of *Ageratum conyzoides*. Key

%IAcETR: Percentage inhibition of *Ageratum conyzoides* of ethanolic root
%IAcEAR: Percentage inhibition of *Ageratum conyzoides* of ethyl acetate root
%IAcN-HR: Percentage inhibition of *Ageratum conyzoides* of n-hexane root

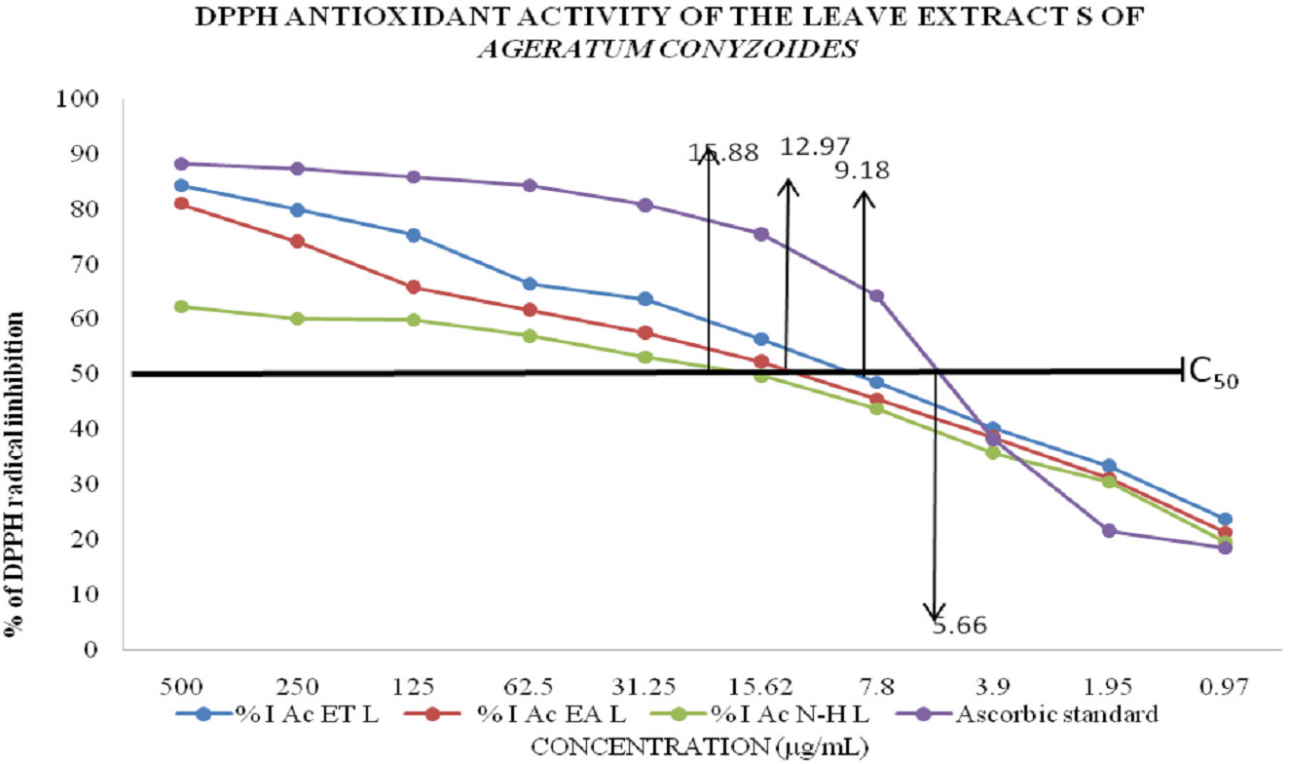

Figure 4. Plot of % inhibition of DPPH radical versus concentrations used to evaluate the antioxidant activity of Leave extracts of *Ageratum conyzoides*. Key:

%IAcETL: Percentage inhibition of *Ageratum conyzoides* of ethanolic leave
%IAcEAL: Percentage inhibition of *Ageratum conyzoides* of ethyl acetate leave
%IAcN-HL: Percentage inhibition of *Ageratum conyzoides* of n-hexane leave

Antimicrobial Sensitivity Assay

The results of antibacterial activities of *Ageratum conyzoides* root and leaf are shown in figure 5. Ethanol extracts of the root and leaf of the plant exhibited higher inhibition on six test organisms than the n-hexane and ethyl acetate extracts. All the bacteria strains were sensitive to the three root extracts of *Ageratum conyzoides* at concentrations ranging from 12.5 to 200 µg/mL using the agar broth cup diffusion procedure.

Ethanol extract of *Ageratum conyzoides* root exhibited higher inhibition against the growth of *Staphylococcus aureus*, *Escherichia coli*, *Bacillus subtilis*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae* and *Salmonellatyphi* than ethyl acetate and hexane extracts of the plant, compared with leaf extracts using the same solvents.

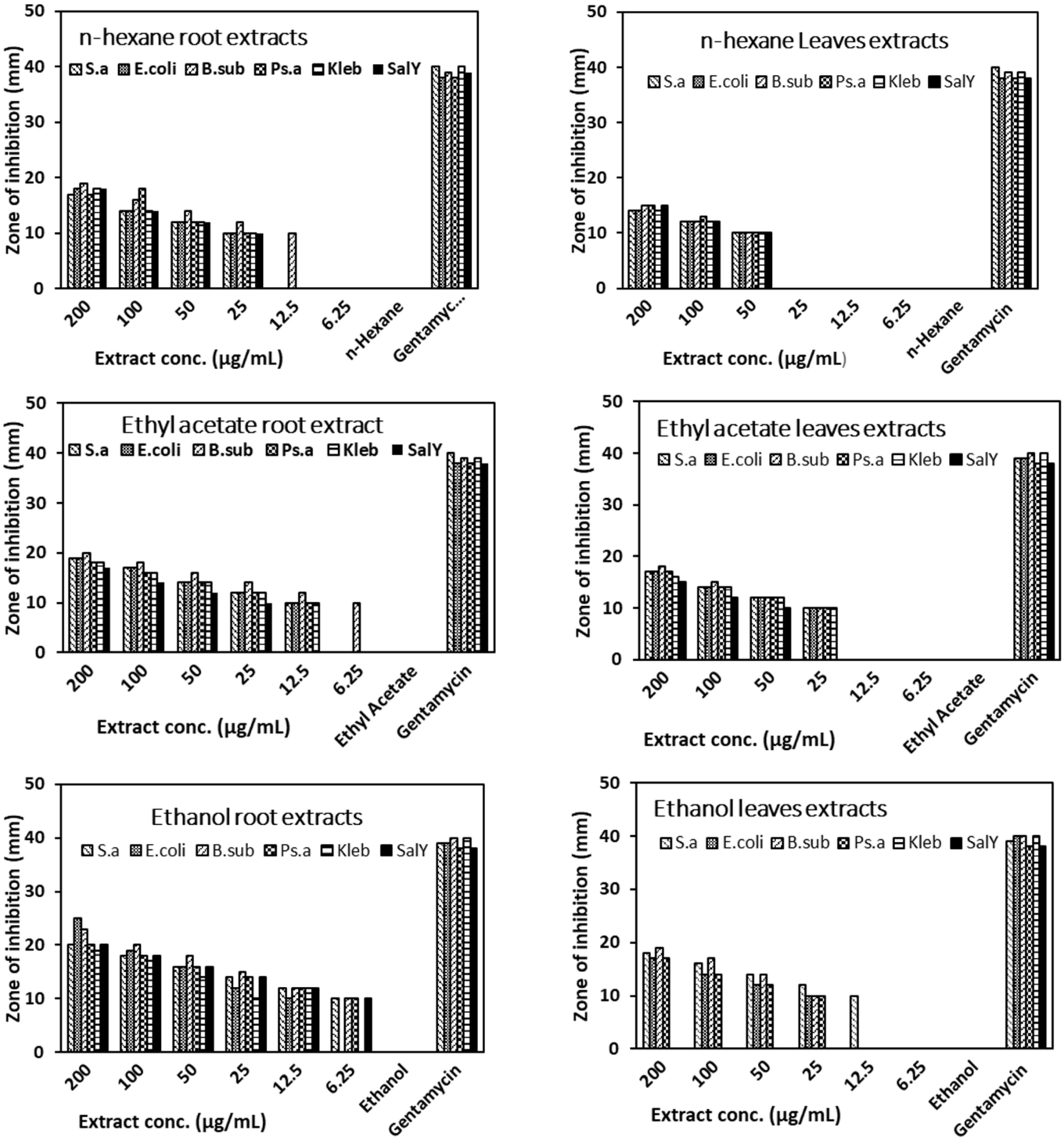

Figure 5. Antibacterial activity of n-hexane, ethyl acetate and ethanolic leaf and root extracts of *A.conyzoides*
Key: S.a: *Staphylococcus aureus*; E.coli: *Escherica coli*; B.sub: *Bacillus subtilis*; Ps.a: *Pseudomonas aeruginosa*; Kleb: *Klebsiellae pneumonae*; SalY: *Salmonella typhi*

The results of the antifungal activities of n-hexane, ethyl acetate and ethanol extracts of *Ageratum conyzoides* root and leaf at concentrations between 200 and 6.25 µg/mL are presented in figure 6. Four clinical strains of human pathogenic fungi were used in the study (*Candida albicans*, *Aspergillus niger*, *Rhizopus stolon* and *Penicilliumnotatum*). *Candida albicans* (*C.a*) and *Aspergillus niger* (*A.n*) showed higher sensitivity with n-hexane, ethyl acetate and ethanol extracts than *Rhizopus stolon* (*Rhiz*) and *Penicillumnotatum* (*Pen*). Also, ethanolic *extract* of the roots revealed higher antifungal activities on *Aspergillus niger* than ethanol extracts of the leaf, as compared with both n-hexane and ethyl acetate extracts of *Ageratum conyzoides* (roots and leaf). The study further revealed that *Penicillumnotatum* showed no sensitivity with n-hexane extracts of both root and leaf but were sensitive to ethyl acetate and ethanol extracts. Ethanol extract of the roots showed higher inhibition on *Aspergillus niger* than other test fungi at concentrations between 25 and 200 µg/mL. However, the sensitivity of the tested bacteria and fungi to all the extracts were concentration dependent, activity being higher at higher concentration of the extracts. Antibacterial and antifungal properties of *Ageratum conyzoides* extracts suggest the use of the plant for the treatment of infectious diseases caused by fungi and bacteria.

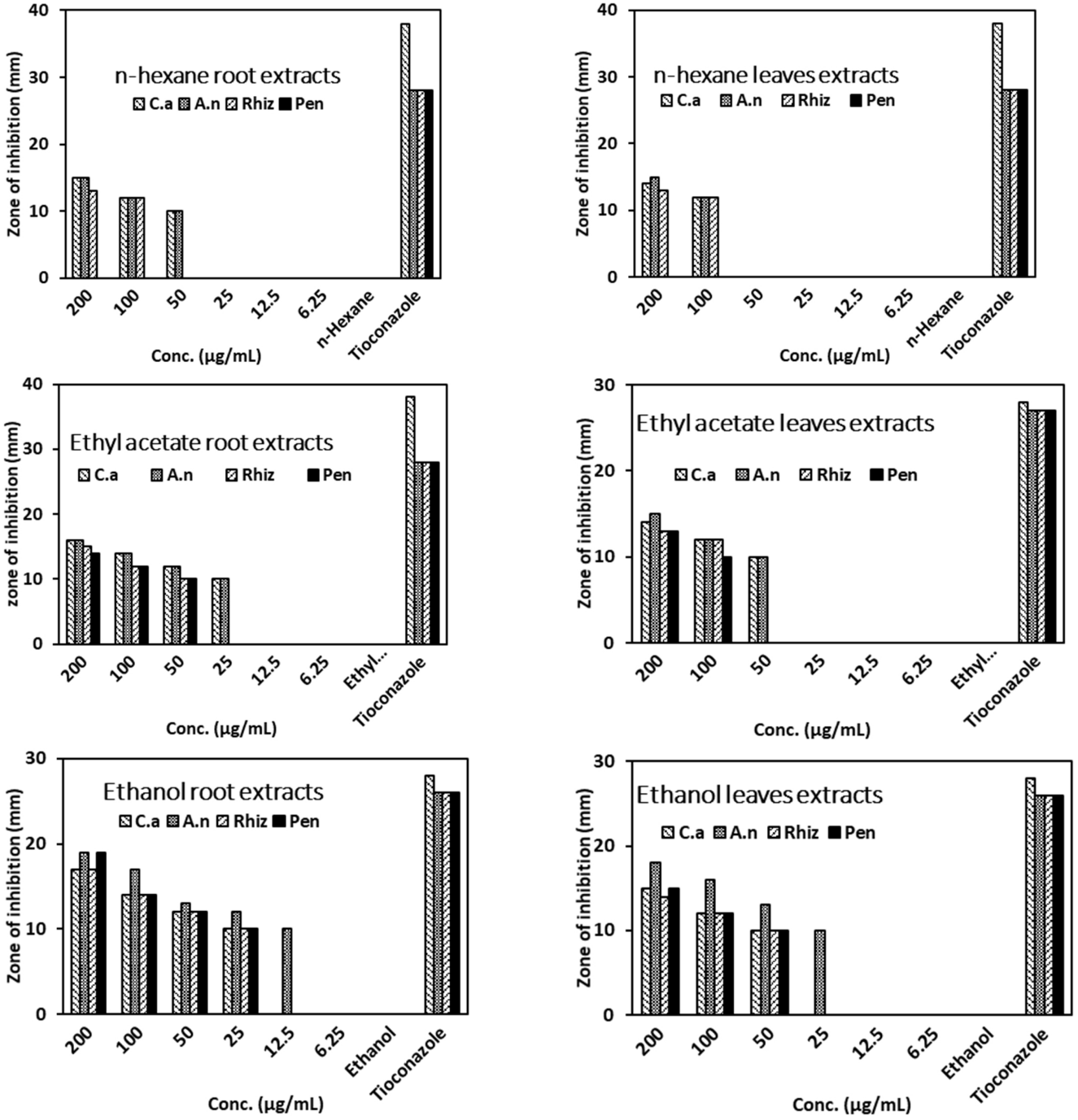

Figure 6. Antifungal activity of n-hexane, ethylacetate and ethanolic leaf and root extracts of *A. conyzoides*
Key: C.a: *Candida albicans*; A.n: *Aspergillus niger*; Rhiz: *Rhizopus stolon*; Pen: *Penicillum notatum*

Conclusion

Ageratum conyzoides is a natural source of antioxidant substances of great importance. The higher antibacterial and antifungal properties of ethanolic extracts of *Ageratum conyzoides* support the use of the plant in alternative traditional medicine for the treatment of bacteria infectious diseases like gonorrhea, syphilis, typhoid; and treatment of fungi diseases such as skin diseases and craw-craw. High contents of flavonoids and phenolics in the extracts of *Ageratum conyzoides* in solvent polarity dependent manner contributed to the high antioxidant activity of the polar solvent which is similar in polarity to that of aqueous extract or alcoholic extract as practised by traditional healers. This study shows that the highest concentration of flavonoids and phenolics in the extracts were obtained using solvents of high polarity. Ethanol whose polarity is very close to that of water manifested the greatest extraction for flavonoids and phenolics.

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