



Phytochemical and Microbial screening of the Aerial parts of *Vernonia Pauciflora* (willd.) Less

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ABSTRACT

The medicinal plant *Vernonia pauciflora* (willd.) Less (Asteraceae) is known for its medicinal potential. Preliminary phytochemical screening of the sample of the herb showed positive test for Alkaloid, Carbohydrate, Triterpenes, Cardiac glycoside, Saponins, Tannins and Flavonoids. The antimicrobial activity of the plant extracts namely Methanol, Ethyl acetate and Hexane extracts were tested on gram positive bacteria (*Methicillin Rest. Staph. aureus*, *Staphylococcus aureus*, *Staphylococcus pyogene*, *Staphylococcus pneumoniae*), gram negative bacteria (*Vancomycin Rest. Enterococci*, *Shigella dysenteriae*, *Salmonella typhi*, *Escherichia coli*, *Neisseria gonorrhoeae*), using disc diffusion method while fungi (*Candida albicans* and *Candida krusel*), tested using sabroiddextroxe broth were found to be effective on all the pathogen when compared with activity of the standard antimicrobial except *Strep. pyogene*, *Vancomycin Rest. Staph. enterococci*, *Shigella dysenteriae* and *Candida.tropicalis* that did not show activity. The plant exhibited significant antimicrobial potency. This study concluded that *Vernonia pauciflora* (Willd.) Less used as a traditional medicinal plant has antimicrobial activity against pathogenic microorganisms. To the best of our knowledge there is not any report on phytochemical and antimicrobial results on this plant.

Key words: *Vernonia pauciflora* (Willd.) Less, Phytochemical analysis, antimicrobial activity.

INTRODUCTION

Nature is the paradise of medicinal principles offers to the humanity through plants which act as richest source of phytochemicals since time immemorial. An impressive number of modern drugs have been isolated from the floristic resources; many being tapped based on their use in the treatises of traditional medicines. Various medicinal plants have been used for years in daily life to combat diseases, world over [1].

Medicinal plants are of great importance to the health of individuals and communities. The medicinal value of these plants lies in some chemical substances that produce a definite physiological action on the human body [2]. The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids, and phenolics compounds [3]. Many of these indigenous medicinal plants are used as spices and food plants. They are also sometimes added to foods meant for pregnant and nursing mothers for

medicinal purposes [4, 5]. This field of natural products research is currently being carried out intensively though it remains far from exhaustion. An attempt to obtain bioactive agents from plants is a worthwhile exercise since only 10% of all plants have been investigated in detail [6].

According to World Health Organization medicinal plants would be the best source to obtain a variety of drugs. In developed countries about 80% of plants are used in traditional medicine. Therefore, such plants have been investigated for better understanding of their medicinal properties. The antimicrobial properties of many plants have been investigated by a number of researcher's in worldwide [7].

Research into traditional plants and herbs received further boost due to the increasing resistance to many orthodox medicine and thus a search for new organic molecules of plants with antimicrobial properties [8]. The use of plant extracts and phyto-products is gaining attention due to their

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availability, cost effectiveness, proven nature of specificity, biodegradability, low toxicity, and minimum residual toxicity in the ecosystem [9]. A lot of work has been carried out to prove that several plant species possess antifungal and antibacterial properties [10, 11].

MATERIALS AND METHODS

Plant Material: Fresh plant parts of *Vernonia pauciflora* was collected from Adoka village, Benue State, in the month of October, 2013. The plant was identified in Herbarium unit of Biological Sciences, Ahmadu Bello Univeristy, Zaria – Nigeria, and the Voucher number was given as 1403.

Preparation of Plant Powder: The collected plant parts were dried separately under shade for 7 days and then coarse powder by hand mill. The fine powder of plant parts were used for phytochemical and antimicrobial studies.

Extraction of plant samples: The powdered plant material (800g) was successively extracted with Hexane, Ethyl acetate and Methanol using cold maceration till exhaustion. All the extracts were carefully evaporated in a rotary evaporator under controlled temperature and reduced pressure to get the extract [12].

Phytochemical Screening: The freshly prepared Hexane, Ethyl acetate and Methanol extracts of *Vernonia pauciflora* was qualitative tested for the presence of chemical constituents. Phytochemical screening of the extracts was performed using the following reagents and chemicals: Alkaloids with Drangendorff’s reagents; Carbohydrates with Molish’s reagents; Steroid and Triterpenes with Lieber-bucher reagents; Cardiac glycogen with keller-kiliani reagents; Anthraquinone with bontragger’s reagents; Saponin glycoside with fronthing reagents; Tanins with ferric chloride and Flavonoids with Sodium hydroxide. These were

identified by characteristic color changes using standard procedures [13].

Microorganisms: Bacterial (Methicillin Rest. Staph. aureus, *Staphylococcus aureus*, *Streptococcus pyogene*, *Streptococcus pneumonia*, Vancomycin Rest. Enterococci, *Shigella dysenteriae*, *Salmonella typhi*, *Escherichia coli*, *Neisseria gonorrhoeae*), Fungi (*Candida albicans*, *Candida krusel*, *Candida tropicalis*), where procured from the department of Medical Microbiology A.B.U. Teaching hospital, Zaria - Nigeria.

Antimicrobial screening (activity) of the extracts

Sensitivity test: The disc diffusion method was used [14]. The antimicrobial activities of the n-hexane, ethyl acetate and methanol extract of the stem bark from *Vernonia pauciflora* was determine using stock concentration of 100 mg/ml. the standardize inoculation of the isolates were uniformly streaked unto freshly prepared Mueller Hinton agar plates with the aid of a sterile swab stick using a sterile cork borer (8 mm in diameter). Appropriately labeled wells were punched into each agar plate; 0.2 ml of the appropriate extract concentration was placed in each well and then allowed in diffuse into the agar. The plates were incubated at 37 °C for 24hours. While for the Fungi, Sabroiddextroxe broth was used and the incubation period was 48hours.

RESULTS AND DISCUSSION

Phytochemical Analysis: The plant extract shows the presence of Alkaloid, Carbohydrate, cardiac glycoside and triterpene were found to be present in the n-Hexane, Ethyl acetate and Methanol extracts. Flavonoids, Tannins, Cardiac Glycoside and Triterpenes were present only in the Ethyl acetate and Methanol extracts. Saponin was only present in the methanol extract. Anthraquinones and steroids were absent in the three extracts: n-Hexane, Ethyl acetate and Methanol extracts (Table 1).

Table 1: phytochemical screening of the extracts from *Vernonia pauciflora* (Willd.) Less

Secondary metabolite	MeOH	EtOAc	Hexane
Alkaloids	++	+	+
Carbohydrate	++	++	+
Anthraquinones	-	-	-
Steroid	-	-	-
Triterpenes	++	+	+
Cardiac glycogen	++	+	+
Tannins	+	+	-
Flavonoid	++	+	-
Saponin	++	-	-

Key: + Presence, ++Appreciable present,- Absence

Results of extraction: (Table 2) revealed the results of weight of solute extracted from 800g of powdered aerial part of *Vernonia pauciflora* and the percentage yield of the crude extract using different solvents. The results indicate that

methanolic extract has the highest percentage yield with 48.60g representing 6.1% followed by ethyl acetate extract with 16.18g having 2.0% and the least is n-hexane extract which has 12.08g representing 1.5%.

Table 2: Extraction of the aerial part of *Vernonia pauciflora*

Solvent	Weight of the powdered Sample (g)	Mass of Extracts(g)	Percentage yield(%)
n-hexane	800	12.08	1.5
Ethyl acetate	800	16.18	2.0
Methanol	800	48.60	6.1

The results of the sensitivity test revealed that the extracts were active against eight of the eleven microorganisms tested while three of the microorganisms were resistant to the extracts. The results of the zone of inhibition showed that the plant extracts had remarkable activity against the

tested microorganisms with inhibition zones ranging from 17 mm – 28 mm. Hexane extract was the least active against all the tested organisms while the Ethyl acetate was the most active against the tested organisms (Table 3).

Table 3. Zone of inhibition (mm) of *Vernonia pauciflora* against microorganism (mg/ml)

TEST ORGANISM	EtOAc	MeOH	n-Hexane	Ciprofloxacin	Fluconazole
<i>Methicillin</i> <i>Rest.staph.aureus(MRSA)</i>	27	25	20	32	0
<i>Vancomycin</i> <i>Rest.enterococci(VRE)</i>	0	0	0	0	0
<i>Staphylococcus aureus</i>	24	22	18	31	0
<i>Streptococcus pyogene</i>	0	0	0	35	0
<i>Sreptococcus pnemoniae</i>	28	23	20	33	0
<i>E.coli</i>	24	20	18	37	0
<i>Neisseria gonorrhoeae</i>	25	21	17	0	0
<i>Salmonella typhi</i>	22	20	18	42	0
<i>Shigella dysenteriae</i>	0	0	0	40	0
<i>Candidas albicans</i>	24	20	17	0	35
<i>Candidas krusei</i>	23	20	18	0	37

The results of the Minimum Inhibitory Concentration (MIC) of the extracts were shown in Table 4. It revealed that the extracts could inhibit the growth of eight out of the eleven microorganisms tested. The result of the Minimum Microbicidal Concentration (MMC) of Ethyl acetate extract, Methanol extract, and n-Hexane extracts was shown in Table 4. The MMC for n-Hexane extract is 60 mg/ml for all the sensitive microbes, the MMC for ethyl acetate extract shows that *S.aureu* and *E.coli* are (30 mg/ml), *S.typhii* (60 mg/ml) and 15 mg/ml for *S.pnemoniae*. The MMC for Methanol extract shows that *S.aureus*, *S.typhii*, *E.coli* and *C.albicans* was 60 mg/ml and 30 mg/ml for *MRSA* and *S.pnemoniae* and the highest MMC is 60mg/ml while the lowest was 15mg/ml.

CONCLUSION

Verninia pauciflora is a plant used in traditional medicine for the treatment of several diseases such as diarrhea, Gonorrhoea, skin infection, typhoid fever, abdominal pain etc. Phytochemical screening revealed the presence of some secondary metabolites which are responsible for the observed antimicrobial activity seen against the tested microorganisms. So, the claim made by the local practitioners was verified to be true to some extent.

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Table 4. Minimum inhibitory and bactericidal concentration of *Vernonia pauciflora* (Willd.) Less against test organisms.

Test Organism	Antimicrobial parameter (mg/ml)	EtOAc extract	MeOH extract	Hexane extract
MRSA	MIC	7.5	15	15
	MMC	30	30	30
VRE	MIC	–	–	–
	MMC	–	–	–
S.aureus	MIC	15	15	30
	MMC	30	60	60
S.pyogene	MIC	–	–	–
	MMC	–	–	–
S.pneumoniae	MIC	7.5	15	15
	MMC	15	30	60
E.coli	MIC	15	15	30
	MMC	30	60	60
N.gonorrhoeae	MIC	15	15	30
	MMC	30	60	60
S.typhi	MIC	15	15	30
	MMC	60	60	60
S.dysenteriae	MIC	–	–	–
	MMC	–	–	–
C.albicans	MIC	15	15	30
	MMC	60	60	60
C.krusei	MIC	15	15	30
	MMC	60	60	60

MIC = Minimum inhibitory concentration; MBC = minimum bactericidal concentration; - = not sensitive.

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