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# ANTIMICROBIAL ACTIVITY OF VERNONIA AMYGLADINA AND VERNONIA COLORART ROOT BARK

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## ABSTRACT

The antimicrobial activity of Vernonia amygladina and Vernonia colorata root bark extracts was carried out using Agar well diffusion method on selected food borne pathogens of medical importance. Both ethanol and aqueous extracts of these root bark extracts were tested against Escherichia coli, Staphylococcus aureus, Bacillius cereus, Salmonella typhi and Shigella dysenteriae, with the ethanol extract showing greater activity against these test microbes. The antimicrobial activity of these extracts is concentration dependent as it increases with increased concentration of the extracts. V. colorata extracts showed greater antimicrobial activity than the V. amygladina extracts. The work therefore provides scientific evidence for the local use of these two plant species against cough, pneumonia and diarrhea. The work also shows that the root bark extracts contain the following classes of organic compounds; alkaloids, glycosides, flavonoids and polphenols. The antimicrobial activity of these roots are therefore attributed to the presence of these phytochemicals.

**KEYWORDS:** Vernonia amygladina, Vernonia colorata, root bark, antimicriobial activity.

## INTRODUCTION

*Vernonia amygladina* and *Vernonia colorata* are leafy vegetables consumed in Nigeria and some other parts of the world. *V. amygladina* leaf has a deeper green colour and a more bitter taste than *V. colorata* leaf. The root of both species have the same use in herbal medicine. The root bark infusion is used to treat malaria, diarrhea and dysentry. Their roots are also chewed to increase appetite and to treat recalcitrant cough (Oliver-Bever, 1986, Etukudo, 2003). The roots are also used locally as blood purifier and utreus toner (Oliver-Bever, 1986). A lot of work has been reported on the leaves but these is scanty information on the roots. The present work is therefore focused on the proximate composition, phytochemicals and antimicrobial activites of the root bark of these two *Vernonia* species on some selected food borne pathogens of clinical importance.

## MATERIALS AND METHODS

Fresh root of *Vernonia amygladina* and *V. colorata* were harvested from the University of Calabar Campus. Both plants were authenticated by the head of herbarium unit (Frank Adejoye) of the Botany Department, University of Calabar, Calabar. The roots were washed and the root bark peeled out, dried in the oven of $40^0$C for three hours and powdered.

The powdered dry root bark (20g) was extracted with ethanol ($100cm^{-3}$) in a Soxhlet extractor to get the ethanol extract. Another 20g of each root bark was soaked in distilled water for about twenty four hours, filtered and the filtrate distilled down to get the aqueous extract (1.0g). Moisture content was determined by separately heating each of the fresh root bark (2g) in an oven at $100^0$C to a constant weight. Ash, crude protein, lipid, fibre and carbohydrate contents were determined using standard methods (Pomeranz and Meloan, 1996). The presence of the following phytochemicals were tested: alkaloids, glycosides, flavonoids, reducing compounds, polphenols, phlobatanins, saponins, tannins, anthraquinones and hydroxymethyl anthraquinones using standard methods (Harbone, 1973, Sofowora, 1993, Trease and Evans, 1989).

Antimicrobial activity of the root barks were tested *against Escherichia coli, Staphylococcus aureus, Bacillus cereus, Salmonella typhi* and *Shigella Dysenteriae* which were obtained from the microbiology unit of the University of

Calabar Teaching Hospital. The bacteria were cultured and maintained by the method of Cruikshank *et al* (1977). The antimicrobial sensitivity test was by disc diffusion method (Baver *et al*, 1994). The plant extract (5g) was mixed with sterile water 50cm$^3$ to get a 100mgcm$^{-3}$ concentration. This was diluted to give 0.8,1.2, 1.6, 2.0, 2.4, 2.8. 3.2, 3.6, 4.0 and 4.4mgcm$^{-3}$ solutions of each root bark extract. A sterile swab stick dipped into the appropriate test organism suspended in normal saline, was used to uniformly seed previously prepared Mueller Hinton agar plates. Sterilized cork borer (5mm diameter) was used to make wells on the agar plate. The different solutions of the extracts were used to fill each well using steralised Pasteur pipette. The plates were incubated at 37$^0$C for eighteen hours. The zone of inhibition around each well was measured and used to determine the microbial sensitivity to each extract (Olowosulu and Ibrahim, 2006). The experiment was carried out in triplicate.

## RESULTS AND DISCUUSION

Table 1 shows the proximate composition of the root bark of *V. amygladina* and *V. colorata.* Both root barks contain about the same amount of total carbohydrate. *V. amygladina* root bark has higher ash and lipid contents while that of *V. colorata* has higher protein content.

The results of phytochemical screening of *V. amygladina* and *V. colorata* root barks are shown in figure 2. The result shows that both root barks are rich poyphenols and alkaloids. *V. colorata* is richer in flavonoids while *V. amygladina* is richer in glycosides. Phlobatannins, anthraquinones, tainnins and hydroxymethyl anthraquinones are absent in both root barks.

The antibacterial activity of the aqueous extract of *V. amygladina* and *V. colorata* on selected pathogenic micro be is show in Table 3 while that of their ethanol extracts is shown on Table 4. The microbes were shown to be more sensitive to the ethanol extracts than the aqueous extracts. This indicates that the antimiocrobial principles are more soluble in ethanol than in water. The observed activity is concentration dependent as it increases with an increase in the concentration of the extracts. With exception of *B. cereus*, the ethanol extract of *V. colorata* has higher activity against the test organisms than the *V. amygladina* ethanol extract. In the case of aqueous extracts, *V. colorata* root bark extract showed higher antimicrobial activity than the *V. amygladina* root bark extract. With the observed zone of inhibition, the susceptibility of the tested microbes to the root bark extract will be described as resistance. Since the susceptibility is concentration resistant, increase in the level of the extract will definitely improve their susceptibility.

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***Table 1: Proximate Composition of V. amygladina and V. colorata Root barks (g/100gDM)***

| Sample | Moisture content % | Ash content | Crude protein content | Lipid content | Fibre content | Total carbohydrate |
|---|---|---|---|---|---|---|
| *V. colorata* | 47.52± 0.02 | 3.10± 0.1 | 7.20± 0.1 | 2.40± 0.02 | 4.40 ± 0.02 | 87.30 ± 0.02 |
| *V. amygladina* | 52.50± 0.2 | 4.40±0.1 | 4.73±0.2 | 3.10±0.1 | 6.47±0.02 | 87.67±0.03 |

***Table 2: Phytochemical Screening of V. amygladina and V. colorata Root Barks***

| S/N | Phytochemicals | *V. colorata* | | *V. amydradina* | |
|---|---|---|---|---|---|
| | | Ethanol extract | Aqueous extract | Ethanol extract | Aqueous extract |
| 1 | Alkaloids | + | ++ | ++ | + |
| 2 | Glycosides | + | + | + | ++ |
| 3 | Saponins | - | + | - | ++ |
| 4 | Tannins | - | - | - | - |
| 5 | Flavonoids | ++ | ++ | + | ++ |
| 6 | Reducing compounds | + | + | + | ++ |
| 7 | Phlobatannins | - | - | - | - |
| 8 | Polyphenols | ++ | +++ | ++ | +++ |
| 9 | Anthraquinones | - | - | - | - |
| 10 | Hydroxymethyl anthraquinones | - | - | - | - |

Key: +++ = very strong; ++ strong presence; += slight presence; -=not present

***Table 3: Antimicrobial activity of Aqueous Extract of V. amygladina and V. colorata Root Barks Against Test organisms (Zones of Inhibition).***

| Conc. mgcm$^{-3}$ | *E. coli* | | *S. aureus* | | *B. cereus* | | *S. typhi* | | *S. dysenterie* | |
|---|---|---|---|---|---|---|---|---|---|---|
| | *V. amygladina* | *V. colorata* | *V. amygladina* | *V. colorata* | *V. amygladina* | *V . colorata* | *V. amygladina* | *V. colorata* | *V. amygladina* | *V. colorata* |
| 0.8 | 1.1±0.05 | 1.5±0.10 | 1.0±*0.33* | 1.0±0.31 | 0.00±0.00 | 0.0±0.00 | 0.0±0.00 | 0.00±0.00 | 0.0±0.00 | 0.0±0.00 |
| 1.2 | 2.0±0.00 | 2.0±0.01 | 1.5±0.11 | 2.0±0.33 | 1.0±0.36 | 1.0±0.01 | 0.0±0.00 | 0.00±0.00 | 0.0±0.00 | 0.0±0.00 |
| 1.6 | 2.5±1.00 | 3.5±0.02 | 2.5±0.86 | 3.5±0.15 | 2.0±0.22 | 2.0±0.16 | 0.5±0.10 | 1.0±0.10 | 0.0±0.00 | 0.0±0.00 |
| 2.0 | 3.0±0.20 | 3.9±0.05 | 3.5±0.33 | 3.8±0.22 | 2.2±0.31 | 2.4±0.05 | 1.5±0.14 | 2.0±0.36 | 0.5±0.00 | 1.0±0.01 |
| 2.4 | 3.2±0.05 | 4.2±1.00 | 3.7±0.19 | 4.0±0.11 | 2.5±0.63 | 2.6±0.22 | 1.7±0.72 | 2.1±0.14 | 0.9±0.14 | 1.2±0.05 |
| 2.8 | 3.5±0.00 | 4.5±0.02 | 3.9±0.39 | 4.3±0.66 | 2.6±0.09 | 2.8±0.17 | 1.9±0.33 | 2.4±0.32 | 1.2±0.16 | 1.5±0.16 |
| 3.2 | 3.9±0.00 | 4.7±0.20 | 4.1±0.05 | 4.5±0.63 | 2.8±0.16 | 3.1±0.31 | 2.1±0.54 | 2.6±0.62 | 1.4±0.14 | 1.7±0.10 |
| 3.6 | 4.2±0.02 | 5.1±0.03 | 4.3±0.16 | 4.7±0.14 | 3.0±0.39 | 3.3±0.11 | 2.3±0.62 | 2.8±0.18 | 1.8±0.16 | 1.9±0.14 |
| 4.0 | 4.3±1.12 | 5.3±0.14 | 4.5±0.51 | 4.9±0.16 | 3.1±0.16 | 3.6±0.50 | 2.6±0.71 | 3.1±0.04 | 2.1±0.04 | 2.1±0.16 |
| 4.4 | 4.5±0.88 | 5.7±0.39 | 4.6±0.57 | 5.2±0.69 | 3.3±0.18 | 3.9±0.01 | 2.9±0.10 | 3.4±0.34 | 2.4±0.16 | 2.4±0.15 |

***Table: 4: Antimicrobial Activity of the Ethanol Extract of V. amygladina and V. colorata Root Barks Against Test Organisms (Zone of Inhibition)***

| Conc. mgcm$^{-3}$ | *E. coli* | | *S. aureus* | | *B. cereus* | | *S. typhi* | | *S. dysenterie* | |
|---|---|---|---|---|---|---|---|---|---|---|
| | *V. amygladina* | *V. colorata* | *V. amygladina* | *V. colorata* | *V. amygladina* | *V . colorata* | *V. amygladina* | *V. colorata* | *V. amygladina* | *V. colorata* |
| 0.8 | 1.5±0.22 | 2.0±0.01 | 1.1±0.12 | 2.0±0.03 | 0.0±0.00 | 1.5±0.01 | 0.0±0.00 | 1.0±0.01 | 0.0±0.00 | 1.0±0.02 |
| 1.2 | 2.0±0.12 | 2.5±0.10 | 2.0±0.04 | 2.5±0.14 | 1.5±0.50 | 2.0±0.22 | 0.0±0.00 | 1.5±0.20 | 0.0±0.00 | 1.5±0.04 |
| 1.6 | 3.0±0.01 | 3.5±0.09 | 3.0±0.06 | 3.5±0.32 | 3.0±0.17 | 2.5±0.31 | 1.1±0.03 | 2.0±0.17 | 1.0±0.01 | 2.0±0.03 |
| 2.0 | 3.6±0.34 | 4.2±0.14 | 4.0±0.16 | 4.6±0.12 | 3.5±0.02 | 3.0±0.16 | 1.5±0.20 | 2.5±0.24 | 1.5±0.31 | 2.5±0.05 |
| 2.4 | 4.0±0.52 | 5.0±0.51 | 4.8±0.52 | 5.6±0.10 | 3.9±0.36 | 3.4±0.10 | 2.0±0.14 | 2.9±0.30 | 2.0±0.32 | 2.9±0.16 |
| 2.8 | 4.4±0.30 | 5.8±0.31 | 5.3±0.33 | 6.7±0.04 | 4.3±0.14 | 4.0±0.14 | 2.2±0.36 | 3.4±0.01 | 2.2±0.00 | 3.3±0.08 |
| 3.2 | 4.9±0.11 | 6.6±0.00 | 5.9±0.11 | 7.4±0.34 | 4.7±0.16 | 4.3±0.62 | 2.5±0.39 | 3.7±0.11 | 2.6±0.67 | 3.7±0.12 |
| 3.6 | 5.3±0.10 | 7.6±0.01 | 6.4±0.12 | 8.4±0.18 | 5.1±0.09 | 4.7±0.18 | 2.9±0.33 | 4.0±0.16 | 2.9±0.16 | 4.0±0.15 |
| 4.0 | 6.0±0.39 | 8.4±0.37 | 6.9±0.15 | 9.5±0.17 | 5.9±0.57 | 5.0±0.40 | 3.2±0.51 | 4.2±0.16 | 3.2±0.23 | 4.2±0.14 |
| 4.4 | 6.3±0.36 | 9.3±0.34 | 7.3±0.01 | 9.6±0.14 | 6.4±0.22 | 5.2±0.15 | 3.7±0.22 | 4.4±0.14 | 3.7±0.34 | 4.4±0.09 |