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RESEARCH ARTICLE

ANTIBACTERIAL POTENTIAL OF VERNONIA AMYGDALINA PLANT AGAINST STAPHYLOCOCCUS AUREUS AND ESCHERICHIA COLI

***CHEA, Sampson K. P., VESELEE, Keselebah, MULBAH, Edmund, BORLAY Allen J. and JAVA, Merfee M.**

Department of Biological Sciences, T. J. R. Faulkner College of Science and Technology, University of Liberia

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ABSTRACT

Medicinal plants are plants in which one or more of its parts contain substances that can be used for therapeutic purposes or which are precursors for the synthesis of useful drugs. Chemical substances obtained from these plants have potential bacteriological activities, and the use of these plants for primary health is a common practice in Asia, Latin American and Africa. *Vernonia amygdalina* plant is a medicinal herb of the family Asteraceae. It is one of the plants commonly grown and harvested wildly across centuries in Africa. Phytochemical components observed to be present within this plant make it a valuable access because of various biological effects in humans. This research was carried out to assess the antibacterial potential from aqueous extract of *Vernonia amygdalina* (Bitter leaf) against two most common bacteria strains namely, the gram-positive *Staphylococcus aureus* and the gram-negative *Escherichia coli*, and to also investigate its aqueous extract via phytochemical Analysis against various reagents. Fresh samples of leaves of *Vernonia amygdalina* plant were harvested and allowed to sun dried for one week. The dried leaves were mashed and subsequently weighed. Twenty-five (25) grams of powder was soaked into 200 ml of distilled water, and the soaked powder was stored at 4˚C for 24hrs and then filtered. The aqueous extracts obtained was stored in an airtight container at 4˚C for about 7 days, and it used for the phytochemical Analysis and antibacterial Susceptibility testing *V. amygdalina* contains very active chemical agents that have potential antibacterial activities. The antibacterial potential of *V. amygdalina* extract makes it better suited as an alternative remedy for cheaper, affordable and a safe treatment of bacterial infection. The aqueous extracts obtained was stored in an airtight container at 4˚C for about 7 days, and it used for the phytochemical Analysis and antibacterial Susceptibility testing.

INTRODUCTION

Medicinal plants are plants in which one or more of its parts contain substances that can be used for therapeutic purposes or which are precursors for the synthesis of useful drugs (WHO, 2007). The local use of natural plants for primary health is a common practice in Asia, Latin American and Africa (Unnikrishnan P, 2010). Although many plants are consumed as food without an in-depth knowledge of their exact chemical composition and contribution to health, their utilization through several generations appear to justify their use (Atana et al, 2015). The use of plant extracts or their active principle may serve as a source of new drugs or sources of intermediate compounds for synthesizing analog drugs (Si-Yuan Pan, 2013).

****Corresponding author:*** **CHEA, Sampson K. P.,**
Department of Biological Sciences, T. J. R. Faulkner College of Science and Technology, University of Liberia.

Medicinal plants known for playing vital roles in the health of individuals and the communities and the medicinal value of some plants lies in some chemical substances that produce definite physiological actions in the human body. *Vernonia amygdalina* plant is a medicinal herb of the family Asteraceae. It is one of the plants commonly grown and harvested wildly across centuries in Africa. Phytochemical components observed to be present within this plant make it a valuable access because of various biological effects in humans. Traditional health workers across Africa recommend the leaf aqueous extract of *Vernonia amygdalina* plant as treatment for anti-parasitic, anti-bacterial, anti-inflammatory and anti-helminthes, a laxative, an appetizer, an aphrodisiac or for body weight loss. (Aminu Mohammed, 2014). The *Vernonia amygdalina* plant grows very well under full sunlight and humid environmental conditions, the shrub of the plant grows to a height of 1-3 m with petiole leaves of about 6 mm in diameter and about 20 cm long, and elliptic in shape. (Igile et al., 1995). The leaves of *V. amygdalina* plant are dark green colored with a characteristic odor and have a bitter taste.

The bark of the plant is gray or brown and has a rough texture that is flaked. The flowers of the plant are white, small, tiny, and clustered with small fruits that are slightly hairy, with small nuts inside (Gill et al., 1992). During the dry season, only the leaves of the plant are harvested without cutting the shoot since there is a scarcity of water for the plant to regrow. All parts of the *Vernonia amygdalina* plant have numerous uses, especially in treatment of bacterial infections which is one most common cause of illness and diseases. (Akinjogunla *et al*. 2011). Drugs used in treatment of bacterial infections are costly and unaffordable for the poor in many countries, and Liberia is no exception. Bacteria resistance against existing drugs, lead to the used of different antibiotics which post the host at risk of developing other side effects. Therefore, the need to investigate the bacteriological activity of medicinal plant so as to unearth new and novel potential chemical agents that can be used to combat the immerging menace of drug resistance. This research was carried out to assess the antibacterial potential from aqueous extract of *Vernonia amygdalina* (Bitter leaf) against two most common bacteria strains namely, the gram-positive *Staphylococcus aureus* and the gram-negative *Escherichia coli*.

MATERIALS AND METHODS

A experimentally designed study and conducted to investigate the Phytochemicals composition and Antibacterial potential of *Vernonia amygdalina* (Bitter leaf) plant. The research was conducted in Fendall, Louisiana, upper Montserrado County at the Leon Q. Ludlem Central Vetenarian Diagnostic Laboratory near the University of Liberia. Campus.

Preparation of Plant Extracts: Fresh samples of leaves of *Vernonia amygdalina* plant were harvested from the back-yard garden in Fendall, upper Montserrado County and taken to the laboratory. The leaves were washed thoroughly with distilled water and sun dried for one week. The dried leaves were mashed using mortar and pestle and subsequently weighed. Twenty-five (25) grams of powder was obtained and then subsequently soaked into a conical flask using 200 ml of distilled water. The soaked powder was stored at 4˚ C for 24hrs. The solution was filtered using Whatman No.1 filter paper obtaining the aqueous extract of the leaves of the plant. The aqueous extracts obtained was stored in an airtight container at 4˚ C for about 7 days, and it used for the phytochemical Analysis and antibacterial Susceptibility testing.

Test for Saponins: 2 ml of distilled water was added to 2 ml of aqueous extract, the solution was shocked for 15minutes, and a foam appeared and foam appeared indicating the presence of Saponins.

Test for Tannins: 2ml of 10 % Potassium hydroxide was added to 2ml of aqueous extract and well mixed, the presence of darkly white precipitation indicated the presence of Tannins.

Test for Steroid: Few drops of acetic acid was added to 2ml of aqueous extract and it was warmed and cooled, then 2ml of sulphuric acid was added, a green color appeared indicating the presence of steroid.

Test for Flavonoid: 2ml of 10% sodium Hydroxide was added to 2ml of aqueous extract, a red color appeared indicating the presence of Flavonoid.

Test for Quinones: Sodium Hydroxide 2ml was added to aqueous extract of 2ml, a green color appeared indicated the present Quinone.

Test for Glycoside (Reducing sugar): 5ml of dilute sulphuric acid was added to 2ml of the aqueous extract, the solution was boiled for 10minutes, then cooled. 2ml of 10% sodium hydroxide was added along with 2ml Fehling solution to neutralize. A brick red precipitation indicated the present of Glycoside.

Test for Alkaloids: 3ml of 1% HCL was added to 2ml of aqueous extract and steam bath for 10minutes, then cooled and filtered. Two drops of picric acid was added and a cream precipitation appeared, indicating the presence of alkaloids. Antibacterial Susceptibility of the Aqueous Extract of *V. amygdalina*

Quality Zone Diameter Test: The agar well method was used to test the antibacterial activity of the aqueous extract of *V. amygdalina* plant. The Mueller-Hinton Agar (Kirby-Bauer method) was used to prepare two separate dishes, solidified and swabbed with S. aureus and *E. coli* over the entire plates respectively. After 5 minutes when the surface dried, extract was dropped in/on the test organism. The plates were inoculated at 37°C, the diameter of the zone of inhibition for each disk was observed and measured within 18 to 24hrs using the metric ruler. The inhibition zones were in millimeter from one edge to the other.

RESULTS AND DISCUSSION

The results of the phytochemical screening and antibacterial susceptibility of the Aqueous Extract of V. amygdalina were obtained and the data are illustrated in the tables below. Table 1, shown plant extract tested against various reagents by standardize phytochemical analysis. Each component tested are present in the *V. amygdalina* plant, indicating that there are bioactive components with in the *Vernonia amygdalina* (Bitter leaf) plant, which are major constituents of the plant activities. *Vernonia amygdalina* is one of the most common medicinal plant known to contain bioactive compounds such as Saponins, Tannins, Steroid, Flavonoid, Quinones, Glycosides and alkaloid that make the plant suitable source for treatment of various microbial infections. The presence of these phytochemical compounds has been detected in the roots and bark and leaf extracts of *Vernonia amygdalina* (Eyong et al, 2011; Akinjogunla *et al*. 2011)).

The zone diameter quality testing in table 2, using the well diffusion method, shown the extract zone at 21 mm against *S. aureus* and 16 mm against *E. coli*. This result revealed that the extract was sensitive against *S. aureus* and intermediate against *E. coli*, in accordance with the standard of agar well diffusion method using extract. Gram-positive bacterium (*S. aureus*) has a relatively thick membrane consisting of layers of peptidoglycan, but peptidoglycan is fully permeable to many substances which makes it sensitive to the extract. The gram-negative bacterium (*E coli*), has thin layer of peptidoglycan but contains an outer membrane and lipid bilayer embedded with carrier proteins called porins. These proteins allow passage of certain small molecules or ions either into or out of the cell periplasm. The size of the porin channel particularly determines the size of the molecule that

Figure 1. Fresh leaves of *V.amygdalina* plant

Figure 2. Sun dried leaves

Figure 3. Mashed powder leaves

Figure 4. Aqueous extract of *V. amygdalina*

Figure 5. *Vernonia amygdalina* (Bitter Leaf)

Table 1. Qualitative Analysis of phytochemical compounds

Compound	Indication	Result
Alkaloid	Appearance of cream precipitation	+
Flavonoid	Appearance of red color	+
Glycoside (reducing sugar)	Appearance of Brick red color	+
Quinones	Appearance of Green color	+
Saponins	Appearance of Foam	+
Steroids	Appearance of Green precipitation	+
Tannins	Appearance of White precipitation	+

Key: + means present; - means not present

Table 2. Quality (Zone Diameter) Test

Microbe	Inhibition Zone (mm)	Interpretation
S. aureus	> = 19 (21mm)	Sensitive
E. coli	15- 18 (16 mm)	Intermediate

can pass through the active compounds may not be able to pass into the cells, making them inactive. The outer membrane serves as a barrier to the passage of many molecules and excludes many toxic compounds, which made the extract less sensitive to it being Intermediate (Nester el al., 2004). Although *S. aureus* usually acts as a commensal of the human microbiota it can also become an opportunistic pathogen, being a common cause of skin infections including abscesses, respiratory infections such as sinusitis, and food poisoning, *Staphylococcus aureus* is both a commensal bacterium and a human pathogen (Masalha et al ,2001). Antibacterial susceptibility testing is used in pathology to determine the resistance of certain bacterial strains in pharmacology research, as it is used to determine the efficacy of novel antimicrobials from biological extracts against different microorganisms (Das et al. 2010).

Conclusion

V. amygdalina (Bitter leaf) plant contains very active chemical agents that have potential antibacterial activities. The presence of these components in the plant, makes it suitable as a pharmaceutical reservoir for the harvesting of potent novel compounds that can serve as drug candidates in the pharmaceutical industry. Moreover, the antibacterial potential of *V. amygdalina* makes it better suited as an alternative remedy for cheaper, affordable and a safe treatment of bacterial infection.

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