



Research Article

PHYTOCHEMICAL CONSTITUENTS OF SOME LEAVES EXTRACTS OF ALOE VERA AND AZADIRACHTA INDICA PLANT SPECIES

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ARTICLE INFO	ABSTRACT
Received 10 th March, 2015 Received in revised form 13 th April, 2016 Accepted 20 th May, 2016 Published online 28 th June, 2016 Keywords: Phytochemicals, Neem, <i>Aloe vera</i> , Extraction Yields, Bioactive Compounds.	Chloroform and water extractions of <i>Aloe vera</i> and neem <i>Azadirachta indica</i> leaves were carried out so as to quantify the phytochemical yields in such samples, and to identify their main constituents of active compounds. The percentage yields obtained through the soxhlet (chloroform) extraction for the <i>A. vera</i> and neem plants were 8.6% and 14.3%, while the percentage yields for the aqueous extracts of the two plants were 5.4% and 6.2% respectively. The result of the phytochemical screening showed the presence of tannins, flavonoids, terpenoids, carbohydrates, and alkaloids for the <i>A. vera</i> plants with neem plant contains flavonoids, steroids, carbohydrates, glycosides, antiquinone, terpenoides and alkaloids, with saponins, tannins, phlobatannins been absent. The occurrence of these biologically active chemicals in Neem and <i>A. vera</i> plants may justify their wide usage in traditional medicine.
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INTRODUCTION

Aloe vera L. (Liliaceae) is a semi tropical plant that has been used by herbalists for the treatment of different human diseases (McCouley, 1990; Shellon, 1996; Davis and Robson, 1999). It is related to other members of the Lily family such as onion and garlic. The *A. vera* plant is made up of fibrous roots, short stem and a spiral greenish leaves. The leaf is made of a gel, which is colourless, viscous liquid consisting primarily of water and polysaccharides and has a bitter taste (Brinelson, 1995). Over 250 species of the genus *Aloe* are existing with only two species grown on commercial basis (*Aloe barbadensis* and *Aloe arborescens*). Regarding chemical constituents, *A. vera* contains amino acids, lipids, sterols, tannins, enzymes chromones (flavonoids) and mannoses-6-phosphate (Brinelson, 1995; Davis and Robson, 1999; Santrel International, 1998; Davis and Robson, 1999). Therefore, this plant had been found useful in the treatment of wound, burns, skin disorders and anti-inflammatory activity (McCouley, 1990; Shellon, 1996; Davis and Robson, 1999).

The mean (*Azadirachta indica* A. Juss) is an evergreen robust tree, belongs to the family Meliaceae. It is mostly found in tropics and sub-tropical areas of the world (Africa and Asia). Occur in medium to large size and have brown to dark grey bark and a dense rounded brown of pinnate leaves (Ogbuwu, 2008). Have an extensive deep root system which is

responsible for their survival in arid and semi arid area of the world.

The chemical constituent of the neem plant is a blend of 3 to 4 actively related compounds, with over 20 lesser ones. Presently over 250 compounds has been identified. The important classes included triterpenoids and limonoids: saladucin, valassin, meliacin, Nimbin nimbinicin, geducin, Azadirachtin, etc (Koul, *et al.*, 1989; Uko and Kamalu, 2001; Lale, 2002). Accordingly, all parts of this plant are useful and have been used in treatment of diseases ranging from teeth decay, ulcers, swollen liver, malaria, dysentery, diarrhea, etc (Ogbuwu, 2008; Allameh *et al.*, 2002; Mossini, *et al.*, 2004; Solowora, 1993; Haller, 1990).

The aim of the study was to quantify the chloroform and water extracts of *Aloe vera* and *Azadirachta indica* leaves, and to identify their constituents of bioactive compounds (viz, glycosides, antiquinones, phlobatannins, carbohydrates, alkaloids, terpenes, saponins, tannins, steroids, flavonoids), which believed to be responsible for their medicinal uses.

METHODS USED

Sample Collection

The leaves of *Aloe vera* and neem (*A. indica*) were obtained from Lokoja area in Kogi State, Nigeria. They were washed to remove dirt and impurities. The gel in the leaves were removed and kept in the refrigerator. Some of the leaves were dried for two weeks under low sun intensity, crushed in a mortar and

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further ground into a coarse powder by using an automated grinder. They were stored in polythene bag, imbedded and kept in the oven to be used as samples for the extraction.

Procedures used for the extraction

Two methods of extraction were adopted as follows:

- ✓ Soxhlet extraction method using chloroform as the solvent.
- ✓ Aqueous extraction method using water as the solvent (Ojo et al., 2005; Sofowora, 1993).

Extraction yields

Procedure for phytochemical test

Phytochemical screening for alkaloids, steroids, triterpenoids, glycosides, carbohydrates, flavonoids, tannins, phlobatannins, antiquinones and saponins were carried out as described below (Sofowora, 1993; Harbome, 1973; Ogbuwu, 2008).

Test for tannins

About 2ml of the aqueous extract was stirred with 2ml of distilled water and few drops of FeCl₃ solution (5% w/v) were added. The formation of a green precipitate was an indication for the presence of tannins.

Test for saponins

5ml of aqueous extract was shaken vigorously with 5ml of distilled water in a test tube and warmed. The formation of stable foam, honey comb in shapes, was taken as an indication for presence of saponins.

Test for phlobatannins

About 2ml of aqueous extract was added to 2ml of 1% HCl and the mixture was boiled. Deposition of a red precipitate was taken as an evidence for the presence of phlobatannins.

Test for flavonoids

To 1ml of aqueous extract, 1ml of 10% lead acetate solution was added. The formation of a yellow precipitate was taken as a positive test for flavonoids.

Test for anthraquinones

(a) Borntrager's test: 3ml of aqueous extract was shaken with 3ml of benzene, filtered and 5ml of 10% ammonia solution was added to the filtrate. The mixture was shaken and the presence of a pink, red or violet colour in the ammonical (lower) phase indicates the presence of free anthraquinones.

(b) 3ml of the aqueous extract was boiled with 3ml of aqueous sulphuric acid and filtered while hot. 3ml of benzene was added to the filtered and shaken. The benzene layer was separated and 3ml of 10% HN₃ added. A pink, red or violet colouration in the ammonical (lower) phase indicates the presence of anthraquinone derivatives.

Test for terpenoids

2ml of the organic extract was dissolved in 2ml of chloroform and evaporated to dryness. 2ml of concentrated sulphuric acid was then added and heated for about 2min. A grayish colour indicates the presence of terpenoids.

Test for steroids

- A red colour produced in the lower chloroform layer when 2ml of the extract dissolved in 2ml of chloroform and 2ml concentrate sulphuric acid added in test tube indicates the presence of steroids.
- The development of a greenish colour when 2ml of the organic extract was dissolved in 2ml of chloroform and treated with sulphuric and acetic acids indicates the presence of steroids.

Test for alkaloids

3ml of aqueous extract was stirred with 3ml of 1% HCl on a steam bath. Mayer's and Wagner's reagents were then added to the mixture. Turbidity of the resulting precipitate was taken as evidence for the presence of alkaloids.

Test for carbohydrates

- Molisch's test: 3ml of the aqueous extract was added to 2ml of Molisch's reagent and the resulting mixture shaken properly, then 2ml of concentrated H₂SO₄ was poured carefully down the side of the test tube. A violet ring at the interphone indicates the presence of carbohydrate.
- 3ml of the aqueous extract was measured into test tube and 1ml of iodine solution was added. A purple coloration at the interphone indicates the presence of carbohydrates.

Test for Glycosides

Liebermann's test

2ml of the organic extract was dissolved in 2ml of chloroform, where 2ml of acetic acid was added carefully. A colour change from violet to blue to green indicates the presence of a steroidal nucleus (i.e. a glycone portion of glycoside).

Salkowski's test

2ml of each extract was dissolved in 2ml of chloroform. 2ml of sulphuric acid was added carefully and shaken gently. A reddish brown colour indicates the presence of a steroidal ring (i.e. a glycone portion of glycoside).

Keller-Kiliani test

2ml of each extract was dissolved in 2ml of glacial acetic acid containing 1ml of concentrated H₂SO₄ characteristic of cardenolides.

RESULTS AND DISCUSSION

The results of extraction yields of chloroform and water extracts of *A. vera* and *A. indica* leaves were presented in Table 1. Accordingly, the soxhlet extract procedure using the chloroform solvent showed that the crude extract of the *Aloe vera* leaves (8.6%) was lower than that obtained by the neem leaves extract (14.3%). On the other hand, the aqueous extraction method gave a value of 5.4% for the *A. vera* leaves and 6.2% for the neem leaves, with little outweigh of the latter plant. The reasons why chloroform extraction gave superior yields may be attributed to the followings: (i) The use of organic solvent that can easily be

evaporated than water, (ii) The high temperature used (heating), (iii) Most components of this plant are organic compounds which easily dissolves in an organic solvent. However, percentage yield of the crude extract was observed to be generally low, and might even be smaller when these bioactive agents are to be obtained in their pure form, this situation give an evidence that are at low concentration is very low in the plant.

Table 1 The results of chloroform and aqueous extraction yields for Aloe vera and the neem leaves

Plant species	Chloroform leaves extract (%)	Aqueous leaves extract (%)
Aloe vera	8.6	5.4
Neem	14.3	6.2

Table 2 Results of phytochemical analysis of the Aloe vera leaves and the neem leaves

Variable tested	Aloe vera leaves	Neem leaves
Alkaloids	+	+
Tannins	+	-
Steroids	-	+
Flavonoids	+	+
Saponins	-	-
Glycosides	-	+
Terpenoids	+	+
Carbohydrates	+	+
Phlobatannins	-	-
Antiquinones	-	+

It is clear that chloroform extracts gave higher yield percentage than water extracts in both plant samples. The phytochemical screening results from table 2 show that the *A. vera* leaves contains alkaloids, tannins, flavonoids, carbohydrates, and terpenoids while the neem plant contain alkaloids, steroids, flavonoids, carbohydrates, glycosides and terpenoids. These compounds may be responsible for their medicinal uses.

CONCLUSION

Bioactive compounds such as alkaloids, flavonoids, steroids, terpenoids glycosides, carbohydrates and tannins were detected to be present in the leaves of *Aloe vera* and *Azadirachta indica* plants. Since these two plants had been used in the treatment of different ailment such as malaria, dysentery, diarrhea, skin burn etc, the medicinal roles of these plants could be related to such identified bioactive compounds. Efforts should be geared up at characterizing entire bioactive agents present, in the two plants for their full utilization.

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