Comparative phytochemical screening of three medicinal plants: Acacia nilotica; Ambrosia maritima; and Moringa oleifera

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Abstract: Many of the pharmacological purpose currently available to physicians have history of use as herbal remedies. Many important drug chemicals are derived from plants. This study aimed to screen *Moringa oleifera*, *Acacia nilotica, and Ambrosia maritima* contents of some important phytochemicals. Materials and methods: 10 g of each powdered plant sample was refluxed with 100 ml of 80% of ethanol four 4 hours. This prepared extract (PE) was used for the various tests. Phytochemical screening for the active constituents (Triterpenes; Alkaloids; Flavonoids; Tannins; Saponins; and Glycosides) was carried out using the standard methods described in literature with some explained modifications. Results: Acacia represented high amount of Tannins and Sterols. Moringa showed high levels of Flavonoids, Coumarins, and Tannins, few Alkaloids and Saponins, but no Glycosides were detected. Ambrosia maritima expressed copious amounts of Triterpenes and Tannins, but no Sterols were seen. None of the plants shown to contain Cyanogenes. Conclusion: Acacia showed to have the most versatile chemical contents, and moringa showed rich content of Tannins and Flavonoids, while Ambrosia was rich in Triterpenes. The three plants models were in lack of Cyanogenes and only trace amounts of glycosides were observed in Acacia extract.

Keywords: Acacia, Moringa oleifera, Ambrosia, Phytochemistry.

1. INTRODUCTION

The use of herbs to treat disease is almost universal among non – industrialized societies; many of the pharmacological purpose currently available to physicians have history of use as herbal remedies, including opium, aspirin, digitalis and quinine. The world health organization (WHO) estimated that 80 percent of the population of some Asian and African countries presently uses herbal medicine for some aspect of primary health care [1]. In fact, according to the (WHO), approximately 25% of modern drugs used in the United Stat have been derived from plants [1]. Among the 120 active compounds currently isolated from the higher plants and widely used in modern medicine today, 80 percent show a positive correlation between their modern therapeutic use and the traditional use plants from which they are derived. At least 35,000 of the plants species have medicinal value and 7,000 plant chemical (phytochemical) compounds [2]. These are some important phytochemicals derived from plants.

Alkaloids: Chemical compounds with heterocyclic a nitrogen ring like amino acids and polyamine. They are produced by plants and animals and by a large variety organism including bacteria and fungi.

Polyphenols: Compounds which contain phenol rings such that give grapes their purple color.

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Glycosides: Molecules in which a sugar is bound to a non- carbohydrate moiety, usually a small organic molecule. In animal and human poisons are often bound to sugar molecules a part of their elimination from the body.

Terpenes: a strong smelling substances produced by a variety of plants. Teroids (derivatives of the terpenes which play role in steroid synthesis) is an example.

Saponins: A class of chemical compounds found in particular abundance in various plant species, like soap foaming they produce when shaken in a solution.

Triterpenoids saponins: Triterpenoid are type of terpense contain Ten Carbons; C10. Cholesterol and phytosterol are triterpense, which reduced surface tension of water with foaming and will break down lipid.

Tannins: It is a high molecular weight compounds used to treat diarrhea and superficial injuries. It is a phenol in structure.

Sterols: Also known as steroid alcohols are a subgroup of steroids, an important class of organic molecules. They occur naturally in plants, animals and fungi with the most familiar type of animal sterol being cholesterol.

Coumarin: A benzo pyrone chemical class, which is colorless and crystalline with a sweet odor. It's a precursor reagent in the synthesis of a number of synthetic anticoagulant pharmaceuticals such to dicourmarol and warfarin.

Flavonoids: They are polyphenol that act as secondary metabolites. Flavonoids were referred to as vitamin β chemically they have the general structure of 15 –carbon skeleton.

This study aimed to report the phytochemistry screening of *Moringa oleifera*, *Acacia nilotica*, *and Ambrosia maritima*, with concrn to the abovementioned chemicals, and to report a quantitative and qualitative comparison between the three plants.

2. MATERIALS AND METHODS

• *Preparation of the extract:*

10 g of each powdered plant sample was refluxed with 100 ml of 80% of ethanol four 4 hours. The cooled solution was filtered and enough 80% ethanol was passed through the volume of the filtrate to 100 ml. This prepared extract (PE) was used for the various tests.

• *Phytochemical screening:*

Phytochemical screening for the active constituents was carried out using the methods described by Martinez and Valencia, 2003 [3]; Sofowora, 1993 [4]; Harborne, 1984 [5]; and Wall *et al.*, 1952 [6] with many few modifications.

• Test for Unsaturated Sterols and Triterpenes:

10 ml of the plant extract (PE) was evaporated to dryness on a water path and residue was washed several times with petroleum ether to remove most of the coloring materials. The residue was then extracted with 20 ml chloroform. Chloroform solution was dehydrated over anhydrous sodium sulphate . 5 ml portion of the chloroform solution was mixed with 0.5 ml of acetic anhydride followed by 2 drops of conc. Sulphuric acid. The gradual appearance of green, blue pink to purple color was taken as an evidence of the presence of sterols (green to blue) and or triterpenes (pink to purple) in the sample.

• Test for Alkaloids:

7.5 ml of the PE was evaporated to dryness on a water bath.5 ml of 2 NHcl was added and stirred while heating on the water bath for 10 minutes, cooled filtered and divided into two test tubes. To one test tube few drops of Mayer's reagent was added while to the other tube few drops of Valser's reagent was added. A slight turbidity or heavy precipitate in either of the tow test tubes was tanked as presumptive evidence for the presence of alkaloids.

• Tests for Flavonoids:

17.5 ml of the PE was evaporated to dry ness on water bath, cooled and the residue was defatted by several extractions with petroleum ether, the defatted residue was dissolved in 30 ml of 80% ethanol and filtered. The filtrate was used for following tests: -

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A/ to 3 ml of the filtrate in a test tube 1ml of 1% aluminum chloride solution was in methanol was added. Formation of a yellow color indicated the presence of Flavonoids. Flavones or and Chalcone.

B/ to 3 ml of the filtrate in a test tube 1ml of 1% potassium hydroxide solution was added. A dark yellow color indicated the presence of Flavonoids compounds (flavones or flavonenes) chalcone and or flavonols.

C/ to 2 ml of the filtrate 0.5 g of magnesium turnings were added flowed by 1 ml of con hydrochloric acid. Producing of defiant color to pink or red was taken as presumptive evidence that flavonenes were present in the plant sample.

• Tests for Tannins:

For this test 7 ml quantity of the PE was evaporated to dryness on water bath. The residue was extracted several times with n-hexane and filtered. The insoluble residue was stirred with 10ml of hot saline solution. The mixture was cooled, filtered and the volume of the filtrate was adjusted to 10 ml with more saline solution. 5 ml of this solution was treated with few drops of gelatin salt reagent. Formation of immediately precipitate was taken as evidence for the presence of tannin in the plant sample. To another portion of this solution, few drops of ferric chloride test reagent were added. The formation of blue, black or green was taken as an evidence for the presence of tannins.

• Test for Saponins:

1 g of the original dried powder plant material was placed in a clean test tube.10 ml of distilled water was added and the tube was stoppard and vigorously shaken for about 30 seconds. The tube was then allowed to stand and observed for the formation of (honeycomb) the appearance of honeycomb, which persisted for least an hour, was taken as evidence for presence of Saponins.

• *Test for cyanogenic glycosides:*

3 g of the powdered plant sample were placed in Erlenmeyer flask and sufficient water was added to moisten the sample, followed by 1ml of chloroform (to enhance every activity). A piece of freshly prepared sodium picrate paper was carefully inserted between a split crock which was used to stopper the flask, a change in color of the paper from yellow to various shades of red was taken as an indication of the presence of cyanogenic glycoside.

• Test for Coumarins:

3 g of the original powdered plant sample boiled with 20 ml distilled water in test tube and filter paper attached to the test tube to be saturated with the vapor after a spot of 0.5 N KOH put on it. Then the filter paper was inspected under UV light, the presence of coumrins was indicated if the spot have found to be adsorbed the UV light.

• Results exhibition:

Results were expressed as follows: - Negative; + Trace; ++ Moderate; + ++ High. A bar chart with multiple bars for each plant was also blotted for visible comparison.

3. RESULTS

As described in Table (1), Acacia represented high amount of Tannins and Sterols, Moderate amount of Triterpenes, Flavonoids, Saponins, and Coumarins, few amount of Alkaloids and Glycosides, but there were no detected Cyanogenes. Moringa showed high levels of Flavonoids, Coumarins, and Tannins, median amounts of Triterpenes and Sterols, few Alkaloids and Saponins, but no Cyanogenes nor Glycosides were detected. On the other hand, Ambrosia maritima expressed copious amounts of Triterpenes and Tannins, fewer Alkaloids, Flavonoids, Saponins, and Coumarins, but no Sterols were seen, and, as in Moringa, also there were no Cyanogenes Glycosides; see also Figure (1).

Test	Sample			Observation
	Acacia	Moringa	Ambrosia	
Alkaloids	+	+	+	Turbidity
Sterols	+++	++	-	Green color
Triterpenes	++	++	+++	Purple color
Flavonoids	++	+++	+	Yellow color

 Table: (1) Results of phytochemical screening of the three plants.

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Saponins	++	+	+	Foam
Coumarins	++	+++	+	UV adsorption
Tannins	+++	+++	+++	Green color/ turbidity
Cyanogenic	-	-	-	No observation
Glycosides	+	-	-	Yellow color to red

Key: - Negative; + Trace; ++ Moderate; + ++ High

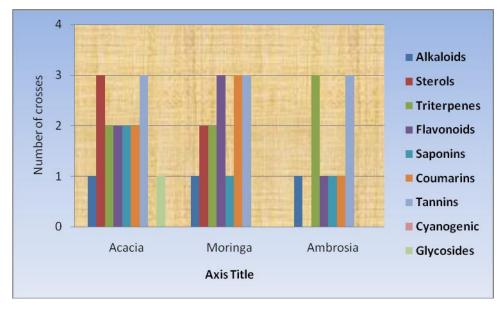


Figure 1: A bar chart showing the level of each chemical in the different plants; empty column designates the absence of this particular chemical.

4. DISCUSSION

The study of chemical compounds derived from plants is called phytochemistry. Many drugs are derived from plant chemicals, and in this article three plants were included in this study: *Acacia nilotica; Ambrosia maritime;* and *Moringa oleifera*. They had been chosen for their pharmacological importance and abundance in Sudan.

Many alkaloids are toxic to other organisms, and used on medication like local anesthesia and hyperion agents [7]. The role of alkaloids for living organism that produce them is still under question [8]. All the three plant showed mild content of alkaloids within their leaves.

The terpenoids, sometimes called Isoprenoids, are derived from the five Carbon isoprene unit. They play an important role in traditional herbal remedies and are under investigation for antibacterial and other pharmaceutical function. Isoprene plays a role in vitamin A, D, E, and K biosynthesis. Steroids and sterols in animal are biologically produced from terpenoid precursors. [9]. While Acacia and Moringa expressed good amounts of Sterols, Ambrosia contained no sterols but at the same time it had a large amount of triterpenes; a steroid precursor.

The aglycone (Glycoside – free) portions of saponins are termed sapogenins. Saponins may enhance nutrient absorption and aid in animal digestion. Saponins were promoted commercially as dietary supplements and nutraceuticals. Saponins are used widely for their effect on ammonia, as they inhibit urease enzyme and splits up urea in feces [10]. The three plants gave a mild to moderate results of Saponins content.

Tannins have the ability to precipitate protein. They are high molecular weight compounds used to treat diarrhea and superficial injuries. They are phenols in structure. The common one is polyphenol which not soluble in organic solvents and soluble in water. Tannins precipitates amylase enzyme which is produced from the salivary gland. Tannins are hydrolyzed to produce Gallic and Ellagic acid [11]. They are equally hih in concentration in the three plants.

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Cholesterol is vital to animal processor to fat- soluble vitamins and steroid hormones [12]. Phytosterol is a sterol of plants where it serves a role similar to cholesterol in animal cells. Phytosterol have been shown in clinical trials to block cholesterol absorption sites in humans intestine, thus helping to reduce cholesterol in humans

[13]. Sterols are highly abundant in Acacia, and to a lower extent in Moringa, but was not detected in Ambrosia in this study.

Coumarin has a bitter taste and animals tend to avoid feeding on. It is used in treatment of asthma [14]. Coumarin is moderately toxic to the liver and kidneys, with a median lethal dose (LD 50). Some 100 natural coumarins have been isolated. Coumarin itself has been found in about 150 species belonging to over 30 different families. This powerful anticoagulant and hemorrhagic agent can cause the death of animals consuming the spoiled fodder [15]. The European food safety authority in 2008 consider toxicity of coumarin known to causes liver and kidney damage in high concentrations 1mg of coumarin per kg of body weight [16]. In this study coumarin concentration was highest in Moringa, lesser in Acacia, but was quite few in Ambrosia.

Flavonoids are important plant pigments. In vivo studies with rats showed that Flavonoids have anti-oxidant activity [17]. The chemical composition of the above plants is interesting and yet to be further researched. The amount of flavonoids was as coumarin in this report; concentration was high in Moringa, moderate in Acacia and mild in Ambrosia.

This report showed that Acacia was the most versatile and rich source of Phytochemicals, since it had only one component missing, while Moringa had two, and Ambrosia three missing. Acacia was also the most abundant in amount.

5. CONCLUSION

The phytochemicals within plants determines their chemical importance, ability to produce drugs and nutrients metabolites, and also their toxic potentials. This study compared the contents of some important phytochemicals in three plants: *Acacia nilotica, Moringa oleifera* and *Ambrosia maritima*. Acacia showed to have the most versatile chemical contents, and moringa showed rich content of Tannins and Flavonoids, while Ambrosia was rich in Triterpenes. The three plants models were in lack of Cyanogenes and only trace amounts of glycosides were observed in Acacia extract. Tannins were the most detected chemical in the plant extracts.

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