

Phytochemical Screening and Antibacterial Activity of *Solenostemma argel*: A Medicinal Plant

Hatim MY Hamadnalla* and Mohamed M El-Jack

Department of Biochemistry, College of Applied Sciences, University of Bahri, Sudan

*Corresponding Author: Hatim MY Hamadnalla, Department of Biochemistry, College of Applied Sciences, University of Bahri, Sudan.

Received: April 05, 2019; Published: May 03, 2019

DOI: 10.31080/ASAG.2019.03.0462

Abstract

The main objectives of the present paper was to verify antibacterial activity of *Solenostemma argel* a medicinal plant used in folk medicine in Sudan for cure of many diseases. Five solvents with grade polarity were used to extract *Solenostemma argel* (petroleum ether, chloroform, ethyl acetate, ethanol and distilled water). These different plant extracts showed the presence of flavonoid, saponin and cardiac glycoside. *Solenostemma argel* leaves extracts were subjected to antibacterial assay using cup-plate agar diffusion method against one Gram positive bacterium *Staphylococcus aureus* and one Gram negative bacterium *Escherichia coli*. Four extracts except petroleum ether showed clear inhibitory action against both bacteria. The antibacterial activity is due to presence of phytoconstituents as saponin and flavonoid in different plant extracts which confirmed by Thin Layer Chromatography (TLC).

Keywords: *Solenostemma argel*; Phytochemical Screening; Antibacterial Activity

Introduction

Plant have traditionally served as man's most important weapon against pathogens. Several plant species are used as source of medicine as direct therapeutic agents. Digdoxin, strophanthin, morphine and atropine are still unsurpassed in their respective field. Plants also are used as starting materials for manufacturing of drugs, example saponin ingredient, which chemically altered to produce saponins; necessary for manufacturing of steroid drugs. Plants can also serve as precursors for substances that can be used as model for the production of new synthetic compounds; for example cocaine for coca nut plants. *Erythroxylum coca* has served as model for the synthesis of a number of local anesthetics drugs; such as procaine.

The important medicinal plants are mostly extracted from herbs, shrubs and even trees. Herbs are defined as small plants with soft stem (People have discovered the benefit of using plants for medical purpose along time age [1]. An estimated large population of half billion people, mostly in the third world use medicinal plants in various traditional ways [2]. Herbal medicine is becoming popular now a day. Simply because they are available, cheap and have no side effect, on one hand. In addition the massive poor population cannot afford the expensive and sometimes infective imported medicine available in the market [2].

The Sudan is a large country with varied topography and different ecological conditions. These factors have created suitable condition for the growth of a large number of various species and varieties of plants of which a high percentage are of medical value [3], for the majority of people in the Sudan traditional medicine remains the main sources of health care, or even the only; specially in remote rural area. (El Shazali, 1994). In many parts of the world herbal medicine has been shown to have utility and that about 80% of the rural population depend on them as their primary health care [2]. Various herbal plant parts were used for curing different disease with remarkable success. Among the enormous number of these medicinal plants is *Solenostemma argel*, which belong to the family *Sclepiadacea*. Extract from the leaves of *S. argel* was used for treatment of Diabetes mellitus and renal inflammation (El Shazali, 1994). Other uses of *S. argel* are purgative, antipyretic, for treatment of cough, colds, renal and colic, and inflammation as well as treatment of chaps, internally. Externally the powder of *S. argel* leaves is claimed to treat inflammatory wound (El Shazali, 1994). It is leaf is reported to be rich in saponin, flavonoid and glycoside [4], and [5] which showed a clear positive antimicrobial activity [6]. The problems of unavailability of pharmaceutical drugs in remote and rural areas, fake drugs and increasing rate of resistance of bacteria to available medicine have lead to the initiation of such work.

The main objective of this work was

- To carry out research and laboratory work to obtain information about antibacterial activity of the Sudanese medicinal plant.
- To verify the clinical of certain Sudanese medicinal plants used in traditional medicine as antibacterial agent.
- To determine most potent agent by Thin Layer Chromatography (TLC)
- To select the most potent antibacterial extract and to subject it to further microbiological studies and test it against pathogenic bacteria.

Material and Methods

Plant materials

Leaves of *S. argel* were obtained from Attarat El timan shop in Omdurman and were identified at the herbarium of Department of Botany, Faculty of Science, University of Khartoum.

Microbial cultures

The bacteria *Staphylococcus aureus* and *Escherichia coli* isolated from stool samples of patients in Khartoum Teaching Hospital were used for determination of antimicrobial activities. The organisms were maintained on blood agar slopes and sub cultured for 24 hours before used.

Extraction

The dried leaves were milled in to a fine powder, using a Waring Blender (Mill MX 391 N). The ingredients of the powdered leaves (100g) were then extracted with four different solvents of grade polarity petroleum ether, ethyl acetate, chloroform, ethanol 80% and distilled water exhaustively in a soxhelt apparatus. From the yield of the four extracts, three dilutions were prepared (100, 200 and 300 mg/ml). Other (50g) of *S. argel* powdered leaves were then extracted with 200 ml distilled water in soxhelt extractor apparatus. The extracted was sterilized using membrane filtration unit (Sartorius). The resulting sterile filtrate was aseptically transferred in to labelled sterile bottle.

Testing bacterial sensitivity

The cup-plate agar diffusion method (Kavangh, 1972) was adopted to assess the antibacterial activity of the prepared extracts. Two ml of isolated bacteria suspension were mixed with 200 ml of nutrient agar and kept at 40°C. 20 ml aliquots were poured in to sterile petri dishes. The agar was left to settle and then from each of the plates, four cups (5 mm in diameter) were cut using a sterile cork borer (No.4) and agar discs were removed. Cups were filled to

2/3 of their depth with the prepared extract using standard Pasteur pipette, and allowed to diffuse at room temperature for one hour. The plates were then incubated in the up-right position at 37°C for 18 hours. For each extract there were three replications for each tested bacteria.

Results

In the present study, the five extracts of *S. argel* leaves extracted by distilled water, ethanol 80%, Chloroform, ethyl acetate and petroleum ether solvents were screened for antibacterial activity on Gram positive bacterium *Staphylococcus aureus* and Gram negative bacterium *Escherichia coli*. It was observed that *S. argel* exhibited anti-bacterial activities. Apart from the extracts of petroleum ether the other four extracts from the leaves of *S. argel* showed clear inhibitory activities against both bacteria (Table 1).

Leaves extract	Concentration of extract (mg/ml)	Diameter of growth inhibition zone by mm	
		<i>S. aureus</i>	<i>E. coli</i>
Ethyl acetate	100	13	9
	200	14	10
	300	16	12
Chloroform	100	11	12
	200	11.3	13
	300	12	14
Ethanol 80%	100	15	14
	200	18	15
	300	21	17
Distilled water	100	13	12
	200	18	17
	300	32	30

Table 1: Antibacterial activity of *S. argel* leaves extracts as determined by diffusion technique in nutrient agar.

The degree of antibacterial activity was classified as low, medium or high depending on the diameter of the inhibition zones. These zones diameters developed as a result of the amount of the herbal extract, the concentration of the active ingredient in the inoculums, and the resistance of the bacteria to antibacterial constituent in the extract.

Phytochemical screening and TLC separation in this study, seems to confirm the above finding, and prove the presence of phytochemical components: Saponin, flavonoid and cardiac glycoside (Table 2).

Group constituent	Test	Result
Flavonoid	Potassium hydroxide test	+
Saponin	Frothing test	+
	Blood haemolysis	+
Tannin	Ferric chloride test	-
Cardiac glycoside	Legal test	+

Table 2: Phytochemical constituent of *S. argel* extracts.

Discussion

The antibacterial activities exhibited in the four extracts from *S. argel* on both bacteria *Staphylococcus aureus* and *Escherichia coli* could be due to the presence of phytochemical components composed of saponin, flavonoid and cardiac glycoside (Table 1 and 2) Mitcher [1] reported similar results. He proved that certain plant constituent such as alkaloid, tannin; saponin and flavonoid were associated with antibacterial activities. These results also agreed with the findings of Rose (1980) and El Hady [6], who reported different inhibitory effect of the above mentioned components against bacteria and fungi.

The inactivity of the petroleum ether extracts from *S. argel* leaves observed in this study could be due to the fact that the chloroform used in the extraction process might have removed the compounds which act as soluble agents for the active constituents in the petroleum ether extracts. Another possibility, which might explain this loss of activity, could be the synergetic action of more phytochemical components present in different extract fractions.

The low antibacterial activity showed by *S. argel* extracts which has due to the resistance of the bacteria to the antibacterial constituent in the extract agreed with the findings of Sowofora [7], who reported resistance of these bacteria to many pharmaceutical drugs particularly the clinical isolate of *Staphylococcus aureus* which showed a clear resistance to benzyl penicillin. This was found to be due to the production of β -lactam ring.

The phytochemical screening and TLC separation in this results has confirmed and complemented the results of Khalid, *et al.* [5], who proved the presence of antibacterial ingredient such as kaempferol, cardiac glycoside; and those of Maharn, *et al.* (1967) and El Fishawi [4] who also proved the presence of the above mentioned compounds as well as quercetin. However, further detailed investigations are required for complete chemical identification of active constituents of *S. argel* needed to produce new antibacterial agent from the plants [8,9].

Conclusion

Solenostemma argel leaves extracts were subjected to antibacterial assay using cup-plate agar diffusion method against one Gram positive bacterium *Staphylococcus aureus* and one Gram negative bacterium *Escherichia coli*. Four extracts except petroleum ether showed clear inhibitory action against both bacteria. The antibacterial activity is due to presence of phytoconstituents as saponin and flavonoid in different plant extracts which confirmed by Thin Layer Chromatography (TLC). This confirmed the uses of plant in folk medicine.

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Volume 3 Issue 6 June 2019

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