ASIAN JOURNAL OF PHARMACEUTICAL AND CLINICAL RESEARCH

INNOVARE ACADEMIC SCIENCES
Knowledge to Innovation

Vol 7, Issue 3, 2014

ISSN - 0974-2441

Original Article

PHYTOCHEMICAL SCREENING AND ANTIBACTERIAL ACTIVITY OF *SCOPARIA DULCIS* EXTRACTS

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Received: 21 April 2014, Revised and Accepted: 10 May 2014

ABSTRACT

Objective: The present study was carried out to investigate the phytochemical screening and antibacterial activity of different extracts of *Scoparia dulcis*.

Methods: Preliminary phytochemical screening in the different extracts of *Scoparia dulcis* for the detection of alkaloids, carbohydrates, flavonoids, phenolic compounds, resin, saponins, steroids, tannins, terpenoids, protein, cardiac glycosides, reducing sugars, proteins and volatile oils and antibacterial activity against *Bacillus* sp., *Corynebacterium* sp. (Gram positive) and *Escherichia coli, Klebsiella pneumoniae, Acinetobacter* sp. (Gram negative) by disc diffusion method.

Results: The phytochemical investigation showed the presence of alkaloids, carbohydrates, flavonoids, phenolic compounds, resin, saponins, steroids, tannins, terpenoids, protein and volatile oils. The antibacterial activity of the different concentrations (25 µl, 50 µl, 75 µl, 100 µl, and 125 µl) of different extracts of *Scoparia dulcis* against both the Gram positive and Gram negative bacteria showed activity against both the bacteria.

Conclusion: *Scoparia dulcis* can act as potent anti-microbial agent as well as it has many phytochemicals.

Keywords: *Scoparia dulcis*, phytochemical, antibacterial activity

INTRODUCTION

Scoparia dulcis, a sweet broom weed, a perennial herb, is widely distributed in tropical and subtropical regions [1]. Many studies reported that the phytochemicals has powerful anticancerous, antitumorous, antileukemia and antiviral properties. The potent phytochemical has displayed selected cytotoxic activity against malignant brain tumours, bone cancer and melanomas [2,3]. Phytochemical screening of *Scoparia dulcis* have found that it contains diterpenoids, flavonoids, tannins, triterpenes, hexacosonal, β-sitosterol, ketone-dulcitone and amellin, an antidiabetic compound [4,5,6]. *Scoparia dulcis* is rich in terpenoids (24 compounds), flavonoids (20 compounds) and steroids (4 compounds) and some miscellaneous compounds (14 compounds) have also been reported from this plant [7,8,9].

In many parts of the world, fresh or dried *Scoparia dulcis* have been traditionally used to treat stomach troubles [10], hypertension [11], diabetes [12], bronchitis [13] and as analgesic and antipyretic agents [14]. *Scoparia dulcis* powder was used in the treatment of bronchitis, fever and kidney disorders in the traditional medicines [15,16]. The objective of the present study was to explore the various phytochemicals and the antibacterial activity of *Scoparia dulcis* collected from Kanyakumari District, Tamil Nadu, India.

MATERIALS AND METHODS

Collection of plant: The investigated plant *Scoparia dulcis* was collected from Kanyakumari district during the month of December, 2012. The whole plants were washed with tap water, the parts were separated and dried at room temperature and ground into a coarse powder. The powder was stored in an airtight container and kept in a cool, dark and dry place until all the analysis were commenced.

Preparation of extract: The powdered plant material was successively extracted with the following solvents of increasing polarity – petroleum ether, toluene, chloroform, methanol, ethanol, and water.

All the solvents and aqueous extracts were subjected to phytochemical analysis described by 17,18,19,20,21,22,23.

Preliminary phytochemical screening

Test for phenols by ferric chloride test

To 1 ml of the different solvent extracts of *Scoparia dulcis*, 3 ml of distilled water and few drops of neutral 5% $FeCl_3$ solution were added. A dark green colour confirmed the presence of phenolic compounds [20].

Test for terpenoids by Salkowski test

To 1 ml of each solvent extract of the *Scoparia dulcis*, 2 ml of chloroform was added, followed by 3 ml of conc. H_2SO_4 to form a layer. A reddish brown colouration of the border indicated the presence of terpenoids [21].

Test for tannins

To 1 ml of solvent extracts of sample, few drops of 1% $FeCl_3$ solution was added. Presence of tannins was confirmed by the formation of blue, black, green or blue green precipitate [22].

Test for saponins

To 2 ml of distilled water, 1 ml of different solvent extracts were mixed and shaken vigorously. The presence of saponins was indicated by a stable persistent froth [18].

Test for flavonoids

For the detection of flavonoids, 1 ml of each solvent extract and 1 ml of NaOH was taken and few drops of H_2SO_4 were added. The occurrence of yellowish brown colour indicated the presence of flavanoids [23].

Test for alkaloids Mayer's test:

To 1 ml of the different solvent extracts of the sample, 1 ml of Mayer's reagent was added on the sides of the test tube followed by the addition of few drops of ethanol. A white or creamy precipitate indicted the test as positive for alkaloids [21].

Test for cardiac glycosides by Keller Killiani's test:

About 1 ml of glacial acetic acid containing 1 drop of $FeCl_3$ solution was added to 1 ml of each solvent extracts of the sample and mixed well, followed by the addition of 1 ml of conc. H_2SO_4. A brown ring at the interphase indicated the presence cardiac glycosides [17,18].

Test for volatile oils:

To 1 ml of the each extracts, added 1 ml of 90% ethanol, followed by addition of few drops of $FeCl_3$. Presence of volatile oils was confirmed by the green colour formation [18].

Test for steroids:

To the 1 ml of each solvent extracts, 2 ml of distilled water was added and shaken vigorously and observed for a stable persistent froth. The frothing was mixed with 3 drops of olive oil and shaken vigorously. The presence of steroids was confirmed by the formation of an emulsion [17,18].

Test for resins

To the 0.5 gm of sample, 5 ml of boiling ethanol was added and filtered through Whatman No. 1 filter paper. The filtrate was diluted with 4 ml of 1% aqueous HCl. Presence of resins was confirmed by the formation of a heavy resinous precipitate [18].

Test for carbohydrates by Molish's test:

To 1 ml of each extract, added few drops of Molish's reagent and 1 ml of conc. H_2SO_4 by the sides of the test tubes. This was then allowed to stand for 2 minutes and then diluted with 3 ml of distilled water. Presence of carbohydrate was confirmed by the formation of red or dull violet colour at the interphase of the two layers [19].

Test for reducing sugars by Fehling's test:

Mix 1 ml of Fehling A and 1 ml of Fehling B solutions boiled for 1 minute in 6 test tubes and added equal volume of extracts. The test tubes were then boiled in a boiling water bath for 5-10 minutes. The presence of brick red precipitate confirmed the presence of reducing sugar [19].

Test for proteins by Biurette Test:

To1 ml of each solvent extracts and added equal volume of biurette reagent and heated for 2 minutes in boiling water bath. Appearence of bluish green colour confirmed the presence of proteins.

Antibacterial assay

The six different solvent extracts of *Scoparia dulcis* were tested for antibacterial activity using well diffusion assay. The bacterial strains tested for the study were *Bacillus* sp., *Corynebacterium* sp. (Gram positive) and *Escherichia coli, Klebsiella pneumoniae, Acinetobacter* sp. (Gram negative). Five wells of 6.5 mm in diameter were prepared on plate of each dish and different concentration (25, 50, 75, 100 and 125 µl) of each solvent extract of the sample was added into the five wells. The assay dishes were then left for 1 hour and subsequently incubated for 24 hours at 37^0C. The diameter of the inhibition zone was then measured. Ampicillin was used as standard (100 µg/well) for comparison.

RESULTS

The preliminary phytochemical examination of various extracts of *Scoparia dulcis* indicated the presence of tannins, flavonoids, terpenoids, phenolic compounds, steroids, glycosides, saponins, alkaloids, carbohydrates, proteins and volatile oil. Some secondary metabolites have been reported in the literature to have pharmacological activities.

The result obtained in the study was presented in the table 1. Flavonoids were screened in toluene, chloroform and ethanolic extracts. Tannins were found in all the extract except in petroleum ether and toluene. Terpenoids were screened in the extracts of chloroform, methanol and ethanol. Phenolic compounds were screened in all the extracts except petroleum ether. Steroids were screened in the entire extract sample. Glycosides were found in all the extracts except in petroleum ether and toluene. Saponins were screened in petroleum ether and toluene extracts only. Alkaloids were screened in all the extracts except methanol and ethanol extract. Carbohydrates were screened in all the extracts. Protein was screened in all the extract except in chloroform extract. Volatile oils were screened in ethanol and methanol extract only.

Table 1: Preliminary phytochemical screening of *Scoparia dulcis*

No.	Test	Petroleum ether extract	Toluene extract	Chloroform extract	Methanol extract	Distilled water extract	Ethanol extract
1.	Tannins	-	-	+	+	+	+
2.	Flavonoids	-	+	+	-	-	+
3.	Terpenoids	-	-	+	+	-	+
4.	Phenolic compounds	-	+	+	+	+	+
5.	Steroids	+	+	+	+	+	+
6.	Glycosides	-	-	+	+	+	+
7.	Saponins	+	+	-	-	-	-
8.	Alkaloids	+	+	+	-	+	-
9.	Molish's test	+	+	+	+	+	+
10.	Biurete	+	+	-	+	+	+
11.	Resin	-	-	-	-	-	-
12.	Volatile oil	-	-	-	+	-	+

"+" – Presence"-" – Absence

The antibacterial activity of different extracts of *Scoparia dulcis* against pathogens were described in the figure 1. In the different extracts of *Scoparia dulcis* at 25 µl concentration, ethanolic extract showed maximum activity against *E.coli*. In 50 µl concentration of different extracts of *Scoparia dulcis*, the toluene extract showed maximum activity against *Klebsiella pneumoniae*. 75 µl of aqueous extract of *Scoparia dulcis* showed maximum activity against *Klebsellia pneumoniae*. *Klebsellia pneumoniae* showed maximum activity at 100 µl of ethanolic extract of *Scoparia dulcis.* 125 µl of methanolic extract of showed maximum activity against *Corneybacterium* sp.

DISCUSSION

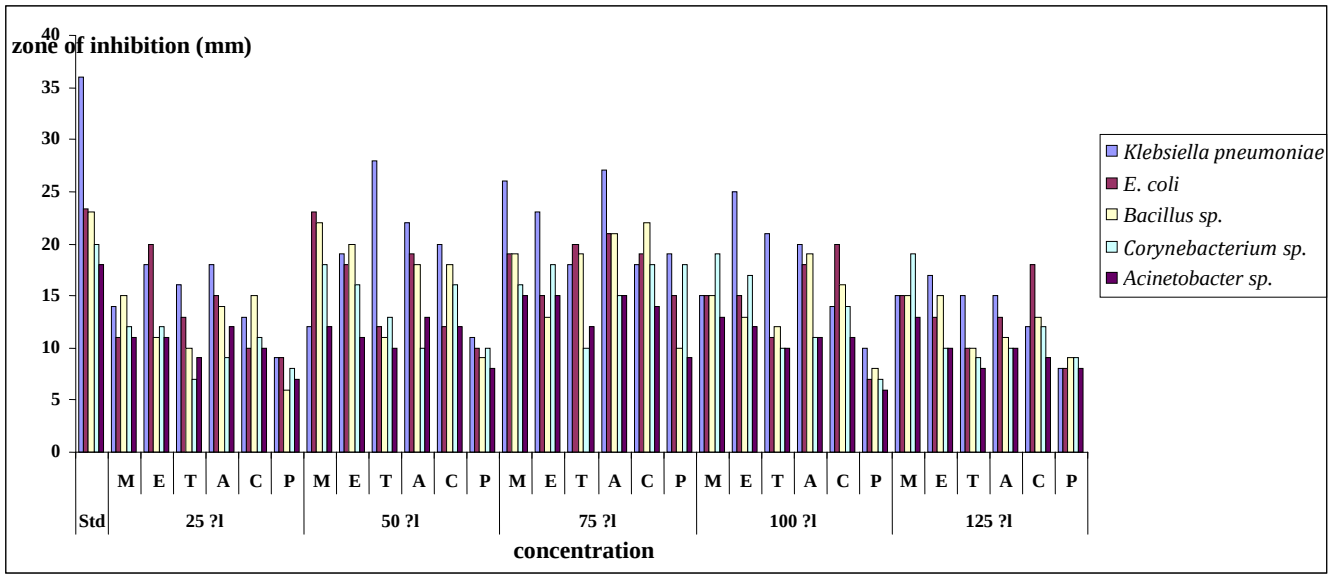
Scoparia dulcis was rich in flavonoids, terpenoids, steroids, saponins and phenols [23, 24]. Some alkaloids had been reported to have anticancer and antiviral activity. Flavanoids had anti-inflammatory activity [25]. The huge reservoir of phytochemicals in *Scoparia dulcis* makes it a successful source of antidiabetic drug. The presence of tannins was responsible for the *Scoparia dulcis* to cure diseases such as diabetes, diarrhea, sore throat, skin ulcer and dysentery. The presence of flavonoids was responsible to cure cancer, inflammation and allergies [26].

Secondary metabolism produced by plants constitutes a source of bioactive substances and now a days the scientist interest has increased due to the search for new drugs from plant origin. Extracts of *Scoparia* dulcis had been shown to posses analgesic and anti-inflammatory properties due to the presence of flavonoid and glutinol [9]. *Scoparia dulcis* had been shown to contain the flavonoid 7-o-methyl scutellarein [5]. Gopalakrishnan *et al* [27] carried out the

phytochemical screening in hexane and methanolic extracts of *Zinimmonii* rhizomes.

In the present study, all the extracts of *Scoparia dulcis* showed 10-26 mm zone of inhibition. Most of the extracts showed maximum activity against *Klebsiella pneumoniae*, a Gram negative bacteria. From the above result it was found that antibacterial action of the different extracts of *S. dulcis* were more on Gram negative bacteria. But these findings did not correlate with the previous findings where most of the active plant extracts showed activity against Gram positive strains only. But, the different extracts of *Scoparia dulcis* showed activity against both the Gram positive and Gram negative bacteria, which indicated the presence of broad spectrum of antibiotic compounds [28,29,30,31,32]. Bharathi *et al* [33] studied the effect of medicinal plants on HIV related opportunistic bacterial and fungal pathogens. Samson Guenne *et al* [34] investigated the antibacterial activities of three *Asteraceae* (*C.americanum*, *E.alba* and *V.colorata*) and found that anti-bacterial compounds can be isolated from these plants.

Fig. 1: Antibacterial activity of different extracts of *Scoparia dulcis* against pathogens



CONCLUSION

Scoparia dulcis is rich in secondary metabolite and has numerous uses in traditional medicine to treat several ailments, ethnomedicinally reputable as anti-diabetics. It has potential for development into a phytomedicine. *S. dulcis* is a good reservoir of diverse type of phytochemicals and makes it a potent antidiabetic agent. Many works are carried out in many parts of the world on *S. dulcis* but this plant has not yet developed as a drug by pharmaceutical industries. A detailed study on the plant about the phytochemical analysis and antibacterial activity helped to promote the traditional knowledge to scientific one.

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