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Research Article

PRELIMINARY PHYTOCHEMICAL SCREENING OF DIFFERENT SOLVENT EXTRACTS OF LEAVES AND STEMS OF *COMMELINA BENGHALENSIS* L (FAMILY: COMMELINACEAE)

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ABSTRACT

Medicinal plants are chief antidote for numerous diseases and have been used since time immemorial. *Commelina benghalensis* L. (Family: Commelinaceae) is a perennial herb commonly known as tropical spiderwort native to tropical Asia and Africa. The present study was aimed to screen the various phytochemicals from the aqueous, methanol, hexane and carbon tetra chloride extracts of leaves and stems of *C.benghalensis*. The extracts were subjected to qualitative phytochemical screening using standard procedures. Four different extracts of leaves and stems of *C.benghalensis* were found to contain various secondary metabolites like alkaloids, protein and aminoacids, saponins, total phenols and Tannins. The phytochemicals generated data from the four different extracts of *C.benghalensis* may be used as tools for quality control of drugs in the future, for the healing of a diversity of disease conditions.

Keywords: Phytochemical screening, *Commelina benghalensis*, Secondary metabolites.

INTRODUCTION

The beneficial efficacy of many indigenous plants for a variety of diseases has been depicted by traditional herbal medicinal practitioners since ancient time. Natural products are the source of synthetic and conservative herbal medicine. These medicines are highly secure as well as environment friendly. According to WHO, 80% of the population from developing and developed countries relies on conventional medicine for their chief health care[1]. They are bioactive chemicals of plant origin, which are considering as secondary metabolites. Naturally, these bioactive chemicals are manufactured in all parts of the plant body i.e., bark, leaves, stem, root, flower, fruits and seeds[2]. The quantity and quality of bioactive chemicals present in plant parts may vary from one part to another. In fact, the biological activity of plants are highly depends on the distribution of bioactive chemicals (or active principles) which are more frequent in some parts of the plants[3]. The successful determination of active principles isolated from plant material is predominantly dependent on the variety of solvent used in the extraction methods[2]. Hence it emphasizes that numerous solvent attempt are required to screen the plant parts for phytochemicals.

Commelina benghalensis L. (Family: Commelinaceae) is a perennial herb commonly known as tropical spiderwort native to tropical

Asia and Africa. It is a widely distributed herbaceous weed that commonly invades agricultural sites and disturbed areas. It is often found on forest edges, road sides, agricultural sites and home gardens. Leaves are ovate to lancolate,2.5-7.5cm long, 1.5-4cm wide, with parallel veination, entire leaf margins, andpubescence on top and bottom. Stems can becrawling along the ground rooting at the nodes or climbing if supported, 10-30cm inheight, 20-90cm in length, covered in a fine pubescence and dichotomously branched. It is used medicinally as a diuretic, febrifuge and antiinflammatory[4]. The plant is used as an animal fodder, eaten by humans as a vegetable.It is usedfor laxative mouth thrush, inflammation of the conjunctiva,epilepsy, nose blockage in children and to cure inflammations of the skin as well as leprosy[5].The plant is used for snakebites, swelling and burns and usedfor night blindness, pain, skin diseases (eczema, abscesses, acne,scabies and warts) and respiratory tract diseases[6].In the present study, various solvent extracts of leaves of *Commelina benghalensis*were qualitatively screened for phytochemicals using standard tests.

MATERIALS AND METHODS

Chemicals

Ferric chloride, HCl, Dragendorff 's reagent, hexane, benzene, carbon tetrachloride, chloroform, H_2SO_4, Folin-Ciocalteu reagent were purchased from Chemico Glass & Scientific Company, Erode, Tamilnadu, India. All the chemicals used in this experiment were of analytical grade.

Collection and authentication of plant material

*C.benghalensis*L. was collected from Jimma University Garden, Jimma, South West Ethiopia in the month of October-2014. The plant has been taxonomically identified and authenticated by the Jimma University Botanist Dr. Ramesh and kept in Jimma University Botanical Science and Herbarium for future references.

Methanol extract of leaves and stem of *C.benghalensis*

The shadow dehydrated roughly powdered of leaves and stem of *C. asiatica*was engrossed and haul out with methanol for 72hrs. After finishing point, the defatted solutions were sieved by filter paper Whatmann No.1 to eliminate any contamination. The extract was intensed by vaccum dessicator to reduce the degree; the concentrated samples were relocated to another beaker and the residual solvent was further vaporized. Finally the dark greenish yellow coloured extract was formed and again it was kept in a vaccum dessicator to get rid of unnecessary wetness. Dehydrated extract was stored in sealed a container and which was used for qualitative phytochemical screening.

Aqueous CCl_4 and hexane extracts of leaves and stem of *C.benghalensis*

The resudues left after methanol extraction was dehydrated and then engrossed separately with aqueous, CCl_4 and hexane respectively up to 3days. After finishing point of extraction, the organic solvents were eliminated by vaccum dessicator. Dark greenish yellow colour extracts were formed and then stored in a sealed container for further studies.

Preliminary phytochemical studies

The extracts obtained (benzene, ethanol, carbon tetrachloride, and aqueous) were employed to the subsequent phytochemical screening.

Test for Alkaloids

a) Dragendorff's test
Take 1ml of the solvent extract, add equal volume of distilled water followed by 1ml of 2molar solution of HCl added until acidification reaction takeplace. To Add this 1ml of Dragendorff's reagent. Orange or red colour is formed, indicated that the occurrence of alkaloids.

b) **Hagger's Test**
Take 1ml of the solvent extract in a cleaned test tube, add 1ml of Hager's reagent. Yellow precipitate is formed, indicated that the occurrence of alkaloids.

c) Wagners Test
Take1ml of solvent extract acidified with 1ml of 1.5 % v/v of HCl and add 1ml of wagners reagent. Formation of yellow or brown precipitate, which indicated that the occurrence of alkaloids.

d) Mayers Test
Take 1ml of Mayers reagent, add 1ml of solvent extract. White or pale yellow precipitate is formed, indicated that the occurrence of alkaloids.

Test for Carbohydrates

a) Anthrone Test
Take 1ml of solvent extract and 10ml of distilled water in a test tube, shaken vigorously and filtered. To this filtrate,

add 1ml of anthrone reagent and mixed. Green or blue color is formed, indicated that the occurrence of carbohydrates.

b) Benedicts Test

Take1ml of solvent extract and 10ml of water in a test tube, shaken vigourously and filtered. To this filtrate, add 3ml of Benedicts reagent and kept in a boiling water bath for 5min. Development of red colour indicated that the occurrence of reducing sugar.

c) Fehlings Test

Take 1ml of solvent extract and 10ml of water in a test tube, shaken vigourously and filtered. To this filtrate, add 1ml of Fehlings solution A and 1ml of Fehlings solution B and kept in a boiling water bath for 5min. Development of red colour indicated that the occurrence of reducing sugar.

d) Molischs Test

Take 1ml of solvent extract and 10ml of water in a test tube, shaken vigourously and filtered. To this filtrate, 1ml of Molisch reagent were added followed by few drops of Conc. H_2SO_4 added in the side of the test tube. Formation of two junction, which indicates the occurrence of carbohydrates.

Test for flavonoids

a) Shinods test

Take 1ml of solvent extract diluted with 3ml of ethanol followed by 2ml dilute HCl and pinch of Mg in a test tube, shaken gently. Appearance of pink or brown precipitate indicated that the occurrence of flavonoids.

b) With Con. H_2SO_4 test

when treated with Con. H_2SO_4, appearance of the following colour like yellow colour (anthocyanins), yellow colour change to orange (flavones); orange colour change to crimson (flavononones) respectively.

Test for Glycosides

Molisch Test

Take 1ml of solvent extract and 10ml of water in a test tube, shaken vigourously and filtered. To this filtrate, 1ml of Molisch reagent were added followed by few drops of Conc. H_2SO_4 added in the side of the test tube.. Formation of two junction, which indicates the occurrence of glycosides.

Test for proteins and free amino acids

1. **Millions test**

Take 1ml of solvent extract with 1ml of Millions reagent, shake gently. Appearance of cherry red color indicated that the occurrence of free amino acid.

2. **Ninhydrin test**

Take 1ml of solvent extract with 1ml of Ninhydrin reagent, shake gently Formation of violet color indicated that the occurrence of free amino acids.

3. **Biuret test**

Take 1ml of solvent extract with 1ml of 10% NaOH and 1ml of 1% copper sulphate in a test tube, shake gently. Development of purple color indicated that the occurrence of proteins.

Test for gums and mucilage

With 95% alcohol

Take 1ml of solvent extract with 25 ml of 95% alcohol in a test tube, shake gently and filtered. The residue was air dried and examined for its bulging property. It indicated that the occurrence of gums and mucilages.

Test for anthraquinones

Take 2ml of the solvent extracts acid hydrolysed with Conc. H_2SO_4 followed by extracted with benzene. Add 2ml of dilute ammonia. Appearance of rose pink color indicated that the occurrence of anthraquinones.

Test for Saponins

Foam test

Take 5ml of solvent extracts in a test tubes add a drop of sodium bicarbonate, shaken vigorously and kept it stand for 3min. Development of cloudy white precipitate indicated that the occurrence of saponins.

Test for Sterols

a) Liebermann-Buchards test

Take1ml of solvent extract in a test tube and add acetic anhydride and kept in a boiling water bath for 5min, then cooled followed by 1ml of Con. H_2SO_4 added along the sides of the test tube. Appearance of green color indicated that the occurrence of steroids.

b) Salkowski reaction

Add 1ml of solvent extract diluted with chloroform and followed by 1ml of Con. H_2SO_4 added along the sides of the test tube. Appearance of two junction/layer

indicated that the occurrence of steroids.

Test for fixed oils

Spot test

Take 0.5ml of solvent extract and pressed in between the two filter papers. Formation of oil stains on the paper indicated the existence of fixed oil.

Add 1ml of 0.5N alcoholic KOH and 1ml of solvent extract along with a single drop of phenolphthalein in a test tube. The residues were kept in a boiling water bath for 20min. Appearance of soap or incomplete neutralization of alkali indicated that the occurrence of fixed oils.

Test for triterpenoids

Add 2ml of solvent extract and 1 ml of $CHCl_3$ followed by 1 ml of acetic anhydride in a test tube and shake gently. Add 1ml of Con. H_2SO_4 added along the sides of the test tube.Appearance of two junction/layer indicated that the occurrence of triterpenoids.

Test for phenolic compounds and tannins

About 5ml of solvent extracts and equal volume of water added and perform the following reagent for confirmation of phenolic compounds and tannins.

Ferric chloride reagents

It givesa violet color.

Gelatin containing sodium chloride

It givesa white precipitate.

Lead acetate solution

It givesawhite precipitate

RESULTS AND DISCUSSION

In the present investigation, preliminary phytochemical investigation has been done in the aqueous, methanol, hexane and CCl_4extracts of *C.benghalensis*leaves and stems showed the presence of phytochemical constituents namely alkaloids, protein and aminoacids, flavonoids, saponins, total phenols and tannins, and absence of Anthraquinones, Glycosides, Steroids Triterpenoids, shown in Table I.

The initial phytochemical screening tests may be helpful in the screening of the bioactive compounds and eventually may help to detection and development of new drugs. Further, these tests make easy their qualitative separation and quantitative estimation of pharmacologically active chemical compounds[7].

The phytochemical screening in the present study has publicized the presence of alkaloids, flavonoids, saponins, and tannins in the leaves and stem extract. Further the presence of different phytochemicals in the four different organic solvent extracts may be responsible for the therapeutic properties of *C.benghalensis*.

Tannins and Flavonoids are phenolic compounds that are acting as principal antioxidants or free radical scavengers. Since these phenolic compounds were originated to be present in the extracts, it might be accountable for the potent antioxidant capacity of *C.benghalensis*. These phytochemicals of medicinal plants have primarily reported for their medicinal value, which can be valuable for therapeutic index. For instance, saponins and glycosides proved as hypotensive and cardiodepressant properties[8], which are helpful for the treatment of congestive heart failure and cardiac myopathy[9]. The occurrence of saponins in aqueous and methanol of leaves and stem of *C.benghalensis*might play a role in the cardioprotective potential. Tannins and alkaloids have the potential of anti-hyperglycaemic and anti- inflammatory activities[10]. Moreover, the terpenoids have also been revealed to decrease blood sugar level in animal studies[11]. In addition, the steroids and triterpenoids demonstrated the analgesic properties and central nervous system activities[11-13]. Hence the initial phytochemical studies are helpful in finding chemical constituents in the plant material that may help to their quantitative assessment and also in locating the source of pharmacologically active chemical compound.

CONCLUSION

The results of phytochemical analysis showed the leaves and stem extracts of *C.benghalensis*indicates their potential as a source of bioactive principles that may supply drugs for modern medicines. Further studies are therefore required to validate their antimicrobial, antihyperglycemic, anti-inflammatory, and antihelminthic activities. In addition, isolation purification and characterization of the active principles are necessary to make the plant has novel interesting studies.

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Table 1: Phytochemical investigation of leaves, stem and roots of *Commelinabenghalensis* L. using aqueous, methanol, hexane and CCl_4 solvents

Analysis	Commelinabenghalensis L (Commelinaceae)							
	Leaves				Stems			
	Aqueous	Methanol	Hexane	CCl_4	Aqueous	Methanol	Hexane	CCl_4
Alkaloids	-	-	++++	++++	-	-	++++	++++
Protein and aminoacids	+++	+++	-	-	++	++	-	-
Anthraquinones	-	-	-	-	-	_	-	-
Flavonoids	+	-	-	-	-	-	-	-
Glycosides	-	-	-	-	-	-	-	-
Saponins	++	-	-	-	++	+++	-	-
Steroids	-	-	-	-	-	-	-	-
Total phenols and Tannins	+	+	-	-	+	++	-	-
Triterpenoids	-	-	-	-	-	-	-	-

+++ = appreciable amount (positive within 5 mins.); ++ = moderate amount (positive after 5 mins. but within 10 mins); + = trace amount (positive after 10 mins. but within 15 mins); - = completely absent.

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