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Preliminary Phytochemicals Screening of *Calligonum polygonoides* Linn.

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ABSTRACT

The present paper was designed to carry out preliminary phytochemicals screening of different parts (roots, stems, buds, flowers and seeds) of *Calligonum polygonoides* Linn. (Polygonaceae) using standard methods. The aqueous extracts and powder of *C. polygonoides* revealed that flavonoids were present in buds, seeds, flowers and stems whereas their absence was noted in roots. Alkaloids were present in roots, buds and seeds whereas their absence was noted in stems and flowers. Proteins were present in flowers and seed whereas these were absent in roots, stems and buds. The tannins, steroids, phenols, carbohydrates and terpenoids were present in roots, stems, buds, flowers and seeds.

Key words: *Calligonum polygonoides*, Polygonaceae, Phytochemical screening.

INTRODUCTION

Everything of this world directly or indirectly depends on plants. According to world health organization (WHO) survey, more than 65-80% of the world's population depends on traditional medicines for their health care and treatments [1-3]. Plants contain lot of phytochemicals and have medicinal properties. Phytochemicals are non-nutritive plant chemicals that have disease preventive properties [4]. Most important phytochemicals extracted from medicinal plants includes alkaloids, tannins, flavonoids, phenolic compounds and lot of other chemicals [5]. Mostly the phytochemicals shows biological activities [6]. Tannins have antimicrobial properties; flavonoids have antibacterial, antifungal, anti-inflammatory, bactericidal and antimicrobial activities; terpenes and steroids are known to have bactericidal and antimicrobial properties against several pathogens [7]. Phytochemicals are the sources of oils, tannins, gums and starting materials of the synthesis of complex substances and important for the discovery of therapeutics agents and drugs [1,8-10].

Calligonum genus (family Polygonaceae) has over 80 species in the world. *Polygonoides* is represented by only one species from this genus in Pakistan. *C. polygonoides* Linn, is commonly called Phog, grows on dry sandy soil of desert. It is a leafless plant has branches and smooth stems, small succulent fruits and bearing abortive flowers. The flowers of *C. polygonoides* contain high amount of protein, possess digestive and tonic properties, and are useful against asthma, cough, and cold. Latex is used for treating eczema and juice of shoot is used for eyes as an antidote to scorpion sting [11-14]. The compounds isolated from *C. polygonoides* are Calligonolides, tetracosan-4-olide, steroidal ester, ß-sitosterol, ß-sitosterol glucoside and ursolic acid [11]. Information concerning the presence of phytochemical constituents in the plant is limited [15]. However, literature survey revealed that, no work has been done on phytochemicals analysis of *C. polygonoides*. The present study was designed to carry out the phytochemicals screening of *C. polygonoides*.

MATERIALS AND METHODS

Collection and identification of plant materials

Fresh roots, stems, buds, flowers and seeds (200 g of each part) of *C. polygonoides* were collected from Village Mahendri-Jo-Par (longitude: N 25° 34' 2'' and latitude: E 70° 11' 20''), District Umerkot, Sindh, Pakistan in January 2011. A voucher specimen (15173) of the plant was deposited in the herbarium of Institute of Plant Sciences, University of Sindh, Jamshoro, Pakistan. The plant was identified by a Taxonomist of the same institution.

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Processing of plant material

Each part of the fresh plant material was washed thoroughly 3 times with sterile water, dried in shadow, and then finely ground to a suitable size and stored in plastic container before analysis.

Preparation of aqueous extracts

5 g powdered of different parts of *Calligonum polygonoides* (roots, stem, buds, flowers and seeds) were crushed in 30 ml of sterile water, heated at 50-60°C for 10 minutes and then filtered (using Whattman No.1 filter paper). The filtrate was centrifuged at 3000 rpm for 20 minutes and analyzed for phytochemical tests [6].

Preliminary phytochemicals analysis

Phytochemicals analysis was carried out according to the methods specified by Siddiqui [6] and Edeoga [16]. The phytochemicals analyzed were alkaloids, tannins, saponins, flavonoids, steroids, phenols, cardiac glycoside, carbohydrates, amino acids, proteins, terpenoids and phlobatannins.

Screening Procedure

Test for Tannins:
About 200 mg of powdered plant was taken in 10 ml water (distilled), stirred and then filtered. 2 ml of $FeCl_3$ was added to the filtrate. Blue-black precipitate observed for tannins and phenols.

Test for Alkaloids:
About 200 mg of powdered plant was stirred with 10 ml CH_3OH and then filtered. 1 ml HCl (1%aqueous) was added to 2 ml filtrate and warmed in a steam bath while stirring. 6 drops of Dragendeff's reagent added to the 1 ml of the filtrate. Reddish-brown precipitate indicates the presence of alkaloid.

Test for Saponins:
About 1 ml of filtrate was stirred with 10 ml distilled water. Frothing persistence indicates the presence of saponins.

Test for Terpenoids:
Added 2 ml of acetic anhydride and few drops of H_2SO_4 (concentrated) in 2ml filtrate. Blue, green ring indicates the terpenoids are present.

Test for Steroids:
About 200 mg of powdered plant material was stirred with 10 ml $CHCl_3$ and filtered then added 2 ml acetic anhydride and few drops of H_2SO_4 (concentrated) in 2 ml filtrate. Green ring indicates the presence of steroids.

Test for Flavonoids:
About 200 mg of powdered plant material was stirred in 10 ml ethanol and filtered. Few pieces of magnesium ribbon were added in 2 ml filtrate and then few drops of concentrated HCl were carefully added. Red color indicates the presence of flavonoids.

Test for Phlobatannins:
About 1 ml of filtrate was boiled with 1% aqueous HCl. Deposition of a red color indicates the phlobatannins are present.

Test for Cardiac glycosides: (Keller-Kiliani test)
Added 1 ml glacial acetic acid, 3 drops $FeCl_3$ and 3 drops H_2SO_4 (concentrated). Green/blue precipitate indicates the presence of cardiac glycosides.

Test for Carbohydrates:
5-8 drops of Fehling's (A and B) solution was added to 1 ml of filtrate. The resultant mixture was boiled for 2 minutes; brick red precipitate indicates the presence of carbohydrates.

Test for Amino acids (Ninhydrin test):
5-7 drops of ninhydrin reagent was added in 2 ml of filtrate and heated content in a boiling water bath for about 5 minutes. Purple indicates the presence of amino acid.

Test for Proteins (Biuret test):
5-7 drops of 5% NaOH and 5-7 drops of 1% $Cu(SO_4)_2$ was added in 2 ml of filtrate. Violet color indicates the presence of proteins.

RESULTS AND DISCUSSION

The curative and pharmacological properties of medicinal plants are due to the presence of secondary metabolites such as flavonoids, sterols, alkaloids, phenols, glycosides, saponins *etc*.

The results of phytochemical screening of powder as well as the aqueous extracts of different parts of *Calligonum polygonoides* (roots, stems, buds, flowers and seeds) were screened, because natural products vary in both concentration and type of phytochemicals in different parts of the plant. Table 1 indicates the presence of remarkable concentrations of flavonoids, tannins, phenols and carbohydrates while cardiac glycoside, amino acids, and phlobatannins were absent.

Table 1: Phytochemical constituents of different parts of *Calligonum polygonoides*

Constituents	Roots	Stems	Buds	Flowers	Seeds
Alkaloids	±	–	±	–	±
Tannins	++	++	++	++	++
Saponins	+	+	+	+	–
Flavonoids	–	±	++	++	++
Steroids	+	+	+	+	+
Phenols	++	++	++	++	++
Cardiac glycoside	–	–	–	–	–
Carbohydrates	++	++	++	++	++
Amino acids	–	–	–	–	–
Proteins	–	–	–	+	±
Terpenoids	+	+	+	+	+
Phlobatannins	–	–	–	–	–

(++) =Higher concentration, (+) =High concentration, (±) =Moderate concentration, (–) =Absence

C. polygonoides contains high concentration of tannins, flavonoids and phenolic compounds than other phytochemicals due to this *C. polygonoides* can be used as a strong antioxidant.

Many researchers have found different types of antioxidants in various kinds of higher plants. Antioxidants enhance the self-life of food, therefore, the addition of antioxidant in food materials and consumption of antioxidant protect the body as well as foods against oxidative deterioration [17]. The medicinal properties of plants have been explored all over the world, due to their strong antioxidant activities, no economic viability and side effects [18].

The phytochemicals play a role in preventing renal calculi, hyperchloestrolcamia and colorectal carcinoma. The literature survey revealed that saponins can reduce cholesterol and control human cardiovascular disease. Tannins give protection against microbiological degradation of dietary proteins in the semen [19].

Phytochemical screening plays a vital role in the pharmacological and chemical study of the medicinal plants. It may suggest feasible pharmacological effects of its extracts or fractions in comparison of identified phytochemicals groups, highlighting a close relationship with its main therapeutic uses [20].

CONCLUSION

Phytochemicals screening results revealed that certain parts of the plant gave a positive test for a particular class of secondary metabolites whereas other parts gave negative test for the same class of compounds. Higher amount (tannins, carbohydrates, phenolic compounds in roots, stems, buds, flowers, seeds; flavonoids in buds, flowers, seeds), high amount (saponins, steroids, terpenoids in roots, stems, buds, flowers, seeds; proteins in flower), a moderate amount (alkaloids in roots, buds, seeds; flavonoids in stems; proteins in seeds) and negative results (cardiac glycoside, amino acids, phlobatannins in roots, stems, buds, flowers, seeds; alkaloids in stems, flowers; saponins in seed; flavonoids in roots; proteins in roots, stems, buds) of phytochemical constituents was found in *C. polygonoides*.

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