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## Primary phytochemical investigation of *Sphagneticola trilobata* (L.) Puruski

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### Abstract

*Sphagneticola trilobata* (L.) Pruski., commonly referred as *Wdelia trilobata* has been used as traditional folk medicinal plant for the treatment of various ailments. A study is designed to explore the preliminary phytochemical analysis of *Sphagneticola trilobata* (L.) Pruski., which is responsible for its pharmacological properties. Qualitative phytochemical screening of *Sphagneticola trilobata* (L.) Pruski., was studied. Four solvents viz; ethanol, petroleum ether, chloroform and distilled water were used to obtain extracts from powdered leaves, stem and roots. The extracts were subjected to qualitative phytochemical screening using standard procedures. From the results, it was observed that of the thirteen phytochemicals screened, ten were found present in various solvent extracts. They are alkaloids, flavonoids, saponins, terpenoids, steroids, glycosides, tannins, proteins, aminoacids and carbohydrates. In all, more phytochemicals were found present in extract prepared with ethanol. Remarkably, quinones, phlobatannins and oxalates were absent in all extracts from different parts of the plant. Phytochemicals found present in the *Sphagneticola trilobata* (L.) Puruski. indicates its ability as a source of principles that may supply novel medicines.

**Keywords:** *Sphagneticola trilobata*, phytochemicals, plant extract, drugs

### Introduction

Green plants always fascinates us with their remarkable source of biochemical components. Ancient literature tells as their usage as traditional health remedies is the most popular for 80% of world population in Asia, Latin America and Africa with minimal side effects (Bibitha *et al.* 2002 and Maghrani *et al.* 2005) <sup>[1, 10]</sup>. Presence of these compounds makes them commercially significant. Now a days, many pharmaceutical companies have spent a lot of time and money in developing natural products extracted from plants, to produce more cost effective medicines that are affordable to the common man (Uniyal *et al.* 2006) <sup>[14]</sup>.

*Sphagneticola trilobata* (L.) Pruski., is a member of the family *Asteraceae* the sunflower or daisy family. The species is commonly referred to by its former name, *Wdelia trilobata* (L.) Hitchc. *Sphagneticola trilobata* (L.) Pruski., is a soil creeper and forms a thick carpet. The genus *Sphagneticola*, has about 70 species with tropical and subtropical distribution. The species is Native to Mexico, Central America the and throughout the Caribbean, now designated by IUCN as one of World's 100 worst invasive species. The plant species is usually introduce as an ornamental or ground cover in gardens and reproduce by vegetative mode. It rapidly forms a dense ground cover, crowding away and preventing other plant species from regenerating.

It is a perennial herb with a creeping habit. The stems are rounded, green or reddish in color with trichomes on it. They grow up to 2 m long wit adventitious roots at their nodes. Short, semi-upright (ascending), flowering branches are produced of these creeping stems. The leaves are bright green, fleshy simple and shows opposite phyllotaxy. These leaves 2-9 cm long and 2-5 cm wide, acute at the apex and winged and sessile at the base usually have three lobes (hence the name *trilobata*) and irregularly toothed (serrated) margins.

The single attractive bright yellow flower heads are borne on the end of terminal and axillary stalks (peduncles), with 2 to 4 whorls of bracts forming the involucre at the base of the head. Each head has 8-13 ray florets that are 6-15 mm long with 1- to 3 finely toothed tips and pistillate. In the centre of these flower-heads there are numerous tiny yellow tubular disc florets 4-5 mm long, and mixed with chaffy bracts. Both ray and disc florets are yellow. The base of each capitulum is enclosed in a row (involucre) of narrow (lanceolate) green bracts. Flowering occurs throughout the year, but is most common from spring to autumn. The fruit is a 2 to 4-angled achene, with short, narrow pappus scales on the top. The seeds, when present, are 4-5 mm long and topped with a crown of short fringed scales. They are elongated in shape, brown in colour and have a rough surface texture.

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*Sphagneticola trilobata* (L.) Pruski., has been historically used as traditional folk medicinal plant for the treatment of various ailments, (Li *et al.*, 2012)<sup>[9]</sup>. Coe *et al.* (1996)<sup>[2]</sup> have reported that fruits, leaves and stem are used in childbirth and in the treatment of bites and stings, fever and infection. Leaves are used in the treatment of kidney dysfunction, cold, wounds and amenorrhoea and dysmenorrhoea (Tsai *et al.*, 2009; Govindappa *et al.*, 2011; Meena *et al.*, 2011)<sup>[4, 9, 13]</sup>. The present study is designed to explore the preliminary phytochemical analysis of *Sphagneticola trilobata* (L.) Puruski. which is responsible for its pharmacological properties. Hope the results will draw attention towards this unexplored potential of this plant species and may help in developing new phytochemical drug with more clinical value.

## Material and Methods

### Collection of plant material

The leaves, stem and roots of *Sphagneticola trilobata* (L.) Puruski. Were collected from three different locations of Thiruvananthapuram. Collected leaves, stem and roots of *Sphagneticola trilobata* (L.) Puruski. were shade dried and was ground using motor and pestle to obtain a fine powder. The powder was further passed through a 2mm sieve to obtain finer particles. The powdered samples were stored in a clean glassware container until needed for analysis.

### Preparation of plant extracts

5 gram of each sample was taken and extracted in Soxhlet apparatus successively with ethanol, petroleum ether, chloroform, and water. After extraction, the extracts were filtered through Whatman No.1 filter paper and stored for further phytochemical investigations

### Preliminary phytochemical investigations

The major secondary metabolites like, alkaloids, flavonoids, saponins, terpenoids, steroids, glycosides, tannins, quinones, Phlobatannins and oxalate and primary chemical compounds such as proteins, aminoacids and carbohydrates were assessed according to the standard procedure described by Harborne (1998)<sup>[5, 6, 7]</sup>. Following protocols were used for present investigation.

#### Test for Alkaloids (Wagner's reagent)

A fraction of extract was treated with 3-5drops of Wagner's reagent [1.27g of iodine and 2g of potassium iodide in 100ml of water] and observed for the formation of reddish brown precipitate (or colouration) which indicates the presence of alkaloids.

#### Test for Flavonoids (Alkaline reagent test)

2ml of extracts was treated with few drops of 20% sodium hydroxide solution. Formation of intense yellow colour, which becomes colourless on addition of dilute hydrochloric acid, indicates the presence of flavonoids.

#### Test for Saponins (Foam Test)

Test solution was mixed with water and shaken and observed for the formation of froth, which should be stable for 15 minutes. This indicates the presence of Saponins.

#### Test for Terpenoids (Salkowki's test)

1ml of chloroform was added to 2ml of each extract followed by a few drops of concentrated sulphuric acid. A reddish brown precipitate produced immediately indicated the presence of terpenoids.

#### Test for Steroids (Liebermann Burchard test)

Crude extract was mixed with few drops of acetic anhydride, boiled and cooled. Concentrated sulphuric acid was then added from the sides of the test tube and observed for the formation of a brown ring at the junction of two layers. Formation of green coloration of the upper layer indicate the presence of Steroids.

#### Test for Glycosides (Keller Killiani Test)

Test solution was treated with few drops of glacial acetic acid and Ferric chloride solution and mixed. Concentrated sulphuric acid was added, and observed for the formation of two layers. Lower reddish brown layer and upper acetic acid layer which turns bluish green would indicate a positive test for glycosides.

#### Test for Tannins (Braymer's test)

2ml of extract was treated with 10% alcoholic ferric chloride solution. Formation of blue or greenish colour solution shows the presence of Tannins.

#### Test for Quinones

A small amount of extract was treated with concentrated HCL and observed for the formation of yellow precipitate (or colouration).

#### Test for Phlobatannins (Precipitate test)

Deposition of a red precipitate when 2mls of extract was boiled with 1ml of 1% aqueous hydrochloric acid was taken as evidence for the presence of phlobatannins.

#### Test for Oxalate

To 3ml portion of extracts were added a few drops of ethanoic acid glacial. A greenish black colouration indicates presence of oxalates.

#### Test for Proteins (Biuret Test)

Test solution was treated with 10% sodium hydroxide solution and two drops of 1% copper sulphate solution and observed for the formation of violet/pink color.

#### Test for Free Amino Acids (Ninhydrin Test)

Test solution when boiled with 0.2% solution of Ninhydrin, would result in the formation of purple color suggesting the presence of free amino acids.

#### Test for Carbohydrate (Benedict's test)

Test solution was mixed with few drops of Benedict's reagent (alkaline solution containing cupric citrate complex) and boiled in water bath, observed for the formation of reddish brown precipitate to show a positive result for the presence of carbohydrate.

## Results and Discussion

### Phytochemical Investigations

Results obtained for qualitative screening of phytochemicals in different parts of *Sphagneticola trilobata* (L.) Puruski. is presented in Table 1. Of the thirteen phytochemicals screened for, ten were found present in various solvent extracts. They are alkaloids, flavonoids, saponins, terpenoids, steroids, glycosides, tannins, proteins, aminoacids and carbohydrates. In all, more phytochemicals were found present in extract prepared with ethanol. Remarkably, quinones, phlobatannins and oxalates were absent in all extracts from different parts of the plant.

According to Tiwari *et al.*, the factors affecting the choice of solvent are; quantity of phytochemicals to be extracted, rate of extraction, diversity of different compounds extracted, diversity of inhibitory compounds extracted, ease of subsequent handling of the extracts, toxicity of the solvent in the bioassay process, potential health hazard of the extractant. The logic in using different solvents when screening for phytochemicals in plant materials was clearly validated in present study. For instance, the results shows that saponins and tannins were exceptionally present in ethanol and water extracts but absent in other two extracts. Steroids showed their presence in petroleum ether and chloroform extracts. This corroborates the reports of Misra *et al.* Proteins, free amino acids and carbohydrates showed their presence in all extracts irrespective to the solvents and plant parts.

Phytochemical screening of the extracts of *Sphagneticola trilobata* (L.) Puruski revealed the presence of alkaloids, steroids, flavonoids, and aminoacids, (table 1). These compounds have significant application against human pathogens, including those that cause enteric infections (El-Mahmood *et al.*) [3]. The result indicates that *Sphagneticola trilobata* (L.) Puruski can be used as a source of pharmaceutically important phytochemicals. Alkaloids generally present in all extracts which play some metabolic role and control development in living system. They are also

involved in protective function in animals and are used as medicine especially the steroidal alkaloids. Tannins are known to inhibit pathogenic fungi, is present in ethanol and water extract. The flavonoids in plant have been reported to exert multiple biological effects including antioxidant, free radical scavenging abilities, anti-inflammatory, anti-carcinogenic etc.

Presence of biochemical compounds in different parts of *Sphagneticola trilobata* lead the way for the development of different biochemical agents that can be used as future drugs. Thus the plant species can be considered as a valuable plant in both traditional and modern drug development areas for its versatile medicinal uses. The medicines developed from the species can be used as an alternative for modern drugs which is comparatively inexpensive also. Despite a long tradition of use of *Sphagneticola trilobata* for treatment of various problems, it still remains unexplored pharmacologically to prove its traditional claims (Kumudini *et al.* 2013; Neelam *et al.* 2014) [8, 12]. There are no clinical data available that would provide evidence of efficacy of *Sphagneticola trilobata* in humans. Extracts and constituents of *Sphagneticola trilobata* may have considerable clinical potential in humans and need to be studied further in *in vivo* models and ultimately in clinical studies.

**Table 1:** Result of phytochemical screening of *Sphagneticola trilobata* (L.) Puruski. + indicates presence and – indicates absence.

Phytochemicals	Extract											
	Ethanol			Petroleum ether			Chloroform			Distilled Water		
	Leaf	Stem	Root	Leaf	Stem	Root	Leaf	Stem	Root	Leaf	Stem	Root
Alkaloids	+	+	–	+	+	+	–	–	–	+	+	+
Flavonoids	+	+	+	+	+	+	+	+	+	+	+	+
Saponins	–	+	+	–	–	–	–	–	–	–	+	+
Terpenoids	+	+	–	+	+	+	+	+	+	+	+	–
Steroids	–	–	–	+	+	–	+	+	–	–	–	–
Glycosides	+	–	+	–	–	–	–	–	–	–	–	–
Tannins	+	–	+	–	–	–	–	–	–	+	+	–
Quinones	–	–	–	–	–	–	–	–	–	–	–	–
Phlobatannins	–	–	–	–	–	–	–	–	–	–	–	–
Oxalate	–	–	–	–	–	–	–	–	–	–	–	–
Proteins	+	+	+	+	+	+	+	+	+	+	+	+
Free amino acids	+	+	+	+	+	+	+	+	+	+	+	+
Carbohydrates	+	+	+	+	+	+	+	+	+	+	+	+

## Conclusion

It is very necessary to introduce new and biologically safe and active drugs for an eco-friendly life style. Phytochemicals found present in the *Sphagneticola trilobata* (L.) Puruski. indicates their potential as a source of principles that may supply novel medicines. Further studies are therefore suggested to ascertain their antimicrobial, anti-plasmodic and antihelminthic activities. Furthermore, isolation purification and characterization of the phytochemicals found present will make interesting studies.

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