



PHYTOCHEMICAL ANALYSIS, ANTIBACTERIAL AND ANTIFUNGAL ACTIVITY OF LEAF EXTRACT OF SENNA OCCIDENTALIS

Virology

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ABSTRACT

Senna occidentalis is one of the most widely used herbal plants, in India it is used to treat bone fracture and bone dislocation, and it is also used to treat whooping cough, convulsion, and diabetes. The phytochemicals in the leaf extract were determined using standard protocols, and antimicrobial efficacy was determined by the well diffusion technique. The results of this work conclude that the leaf extract consists of pharmacologically active compounds that might be useful against microbes as traditional medicine.

KEYWORDS

Senna occidentalis, Phytochemical analysis, Antibacterial activity, Antifungal activity, Antioxidant activity.

INTRODUCTION:

The primary source of traditional medicine for the production of drugs is plants [1]. The leaf extract of *Senna occidentalis* is used as a common herbal treatment in India, Nigeria and other parts of the world. According to the World Health Organization, 70% of the world's population are dependent on medicinal plant for public health [2-4]. World health facilities have developed so much but still infections with bacteria, fungi, viruses and parasites are major threats to public health. The impact of these infections is much more in developing countries due to the unavailability of medicines and the evolution of infectious agents to resistant varieties [5]. The drugs found in plants are from the phytochemicals present in a part of that plant [6]. They have certain effects on the physiology of the infectious agents as well as on the human body which will ultimately protect us from these infectious agents [7].

Senna occidentalis L. is a yellow-flowered shrub, that belongs to the family *Fabaceae* and genera *Cassia* and *Senna*. They are native to tropical and subtropical regions [8]. The synonyms for *Senna occidentalis* are *Cassia occidentalis* L., *C. foetida* Persoon, and *Ditremexa occidentalis* (L.) [8]. Vernacular names of the plant are Negro coffee senna, stinking weed in English, Café negre, case-café, case puante in French, menting (Javanese), kasingsat (Sundanese), kopi andelan (Sumatra) in Indonesian, balatong – aso (Tagalog), andadasi (Ilokano), duda (Bisaya) in Philippines, sãndaøk khmaoçh in Cambodian, kh'ët, lang kh'ët in Laos, khilek-thet (northern), khilek-pi (central), chumhet-thet (peninsular) in Thailand, vông giang nam, muông cõt khí in Vietnamese, Kasondi (Hindi), Karinthakara (Malayalam), Adavi Thangedu (Telugu), Nattam Thakira Paayaverai (Tamil) in India [9]. The origin of *Senna occidentalis* is unknown but thought to have originated in tropical South America. Now it is a common weed throughout the tropics and subtropics. It is also widespread in Southeast Asia [9,10]. Roasted seeds of *Senna occidentalis* are used as a replacement for coffee. In Indonesia, young leaves and fruits are consumed as vegetables. The young fruits are also consumed as a side dish with rice [11,12]. The medicinal applications of *S. occidentalis* were numerous in Africa. It is used as an ointment to treat all kinds of skin diseases. It is also used as a medicine against diabetes also used as a diuretic, Hepatonic, liver detoxifier, whooping cough, convulsion, and treatment of bone fracture and bone dislocation as a herbal treatment in India. Puttur is well known for the herbal treatment of bone fracture and bone dislocation using *S. occidentalis* in South India [11,12]. In the past, *Senna occidentalis* was traded between Africa and Europe as a medicinal product. At present the only trade is local there is no statistical data [13].

Description:

Shrub, erect, annual plant that grows up to 2 to 3 meters tall. Stem obtuse angled, branched. Leaves are arranged in spirals, usually in 3 –

7 pairs 15 – 17cm long. Flowers are in 2 – 3 pairs, petals are in yellow with 4 stamens and the ovary is tomentose glabrous style with a small lateral stigma. Seeds are dull brown in colour and seed pods are dark brown in color [14].

Growth and Development:

It grows well under warm humid conditions; it dies with the onset of cold or dry periods. Under favorable conditions it flowers throughout the year [15, 16].

MATERIALS AND METHODS:

Collection of plant:

The plant *Senna occidentalis* was collected from Puttur, Chittoor District, A.P., India. The collected plant leaves are chopped into pieces and air dried for 10 days then ground into fine powder, used for subsequent studies.

Preparation of plant leaf extract:

The dried leaf powder was weighed up to 10 gms and it was dissolved in 100ml of distilled water.

Phytochemical analysis of plant leaf extract:

The phytochemical analysis was performed to determine the bioactive compounds present in the leaf extract of *Senna occidentalis* [17 - 20].

Test for carbohydrates:

Molisch's test:

Molisch's reagent (1ml) and a few drops of concentrated H_2SO_4 were added to 2ml of plant leaf extract, formation of violet colour at the junction of two layers indicates the presence of carbohydrates.

Test for reducing sugars:

Benedict's test:

Benedict's reagent (1ml) was added to 2ml of leaf extract and heated in the water bath for 5 minutes, the formation of brick brick-red colour indicates the presence of reducing sugars.

Test for anthraquinone glycosides:

Borntrager's test:

A few drops of diluted H_2SO_4 were added to leaf extract and heated for 5 minutes, filtered and cooled with an equal volume of dichloromethane, The formation of rose pink to red colour indicates the presence of anthraquinone glycosides.

Test for saponins:

Froth test:

To the leaf extract, 2ml of distilled water was added and shaken for 15 minutes. The formation of foam up to 1cm indicates the presence of saponins.

Test for proteins:**Biuret's test:**

NaOH (10%) (1ml) was added to 2ml of leaf extract and heated, later few drops of CuSO_4 (0.7%) were added, formation of a purplish violet colour indicates the presence of proteins.

Test for steroids:**Liebermann-Burchard test:**

A few drops of acetic anhydride were added to plant leaf extract, boiled and cooled later few drops of concentrated H_2SO_4 were added, The Formation of brown ring at the junctions of two-layer and the upper layer turning green indicates the presence of steroids.

Test for tannins and phenolic compounds:**Iodine test:**

A few drops of diluted iodine solution were added to the leaf extract of *Senna occidentalis*, The formation of transient red colour indicates the presence of tannins and phenolic compounds.

Test for alkaloids:**Wagner's test:**

Diluted HCl (5ml) was added to the plant leaf extract and filtered, later few drops of Wagner's reagent were added, formation of reddish-brown precipitate indicates the presence of alkaloids.

Test for flavonoids:**Shinoda's test:**

Ethanol (95%), 0.5gms of magnesium and a few drops of concentrated HCl were added to the plant leaf extract, the formation of pink colour indicates the presence of flavonoids.

Antibacterial activity:

The antibacterial activity of plant leaf extract was tested by using the well diffusion method against *E. coli*, *Klebsiella pneumonia*, (Gram-negative), *S. aureus*, and *Bacillus subtilis* (Gram-positive). The bacterial cultures are obtained from the Department of Virology, S.V. University, Tirupati. The bacterial cultures were spread evenly on the nutrient agar plate by using an L-rod then different concentration of plant leaf extract was loaded into the wells of the agar medium. Later the plates were incubated at 37°C for 24 hours. Streptomycin was used as a control. After incubation, the zone of inhibition was measured [21].

Antifungal activity:

The antifungal activity of plant leaf extract was tested against *Aspergillus niger* by using the well diffusion method. The fungal cultures are obtained from the Department of Virology, S.V. University, Tirupati. The spore suspension of *A. niger* was spread evenly on a potato dextrose medium by using L-rod. Later, wells were made in the PDA medium using a sterile borer then different concentrations of plant leaf extract were added. The plates were incubated at room temperature for 5 – 7 days. After incubation, the zone of inhibition was measured [22].

RESULTS AND DISCUSSIONS:**Phytochemical analysis of plant leaf extract:**

The phytochemical analysis of the plant leaf extract revealed the presence of carbohydrates, saponins, proteins, steroids, alkaloids, phenolic compounds, tannins and the absence of anthraquinone glycosides, flavonoids and reducing sugars. The phytochemical analysis is performed in duplicates. The results are shown in table-1. Similar reports are studied in A. Medicinal plants and Traditional medicine in Africa, Phytochemical methods. 3rd ed. London: Chapman and Hall, Phytochemical screening of medicinal plants belonging to *Euphorbiaceae* [17 - 20].

Table-1: Phytochemical analysis of leaf *Senna occidentalis* extract

Phytochemicals	Test	Observation	Result
Carbohydrates	Molisch's test	Violet colour	+ve
Reducing sugars	Benedict's test	colour change negative	-ve
Anthraquinone glycosides	Borntrager's test	colour change negative	-ve
Saponins	Froth test	Formation of foam	+ve
Proteins	Biuret's test	Purplish violet colour	+ve

Steroids	Liebermann-Burchard test	Reddish brown-green rings	+ve
Tannins & phenolic compounds	Iodine test	Transient red colour	+ve
Alkaloids	Wagner's test	Reddish brown precipitate	+ve
Flavonoids	Shinoda's test	colour change negative	-ve

Antibacterial activity:

The antibacterial activity of plant leaf extract against the bacterial strains was studied and results are shown in Table 2. The plant leaf extract of *Senna occidentalis* exhibited excellent antibacterial activity against both Gram-positive and Gram-negative bacteria. At the lowest concentration of plant leaf extract the zone of inhibition for gram-positive bacteria was 0.4 to 0.9cm and for gram-negative bacteria was 0.4 to 1.0cm, at the highest concentration the zone of inhibition for gram-positive bacteria was 1.0 to 1.6cm and for gram-negative bacteria was 0.9 to 1.7cm. With increasing the concentration of plant leaf extract, the zone of inhibition also increased. The antibacterial activity of leaf extract was better than the control. Similar reports were seen in Physicochemical Characterization and Antibacterial Activity of *Senna occidentalis* Linn. [21].

Table – 2: Antibacterial activity of *Senna occidentalis* leaf extract

Conc of plant leaf extract (in μl)	Zone of inhibition			
	<i>B. subtilis</i>	<i>S. aureus</i>	<i>E. coli</i>	<i>Klebsiella pneumonia</i>
25	0.4	0.9	1.0	0.4
50	0.7	1.2	1.4	0.4
75	1.0	1.6	1.7	0.9
streptomycin	0.9	0.9	0.9	0.9

Antifungal activity:

The antifungal activity of plant leaf extract against *A. niger* is shown in Table-3. At the lowest concentration, the zone of inhibition was 0.1cm, at the highest concentration the zone of inhibition was 0.6cm. The antifungal activity increased as the concentration of plant leaf extract increased. The results are concluded by referring Antifungal activity of *Senna occidentalis* root extract against *Macrophomina phaseolina* [22].

Table – 3: Antifungal activity of *Senna occidentalis* leaf extract

Conc. of plant leaf extract (in μl)	Zone Of Inhibition (in cm)
50	0.1
75	0.2
100	0.6

CONCLUSION:

Based on this study results it is confirmed that the plant leaf extract of *Senna occidentalis* consists of phytochemicals such as carbohydrates, proteins, saponins, alkaloids, tannins, and phenolic compounds. The leaf extract has the ability to inhibit microbes such as bacteria and fungi and it might be used as antibiotics and antifungal agents clinically up on further research and studies.

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