

Phytochemical Activity and Quantitative Analysis of Senna Alata and Senna Hirusta Medicinal Plants

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Abstract:

Preliminary phytochemical screening is the key step in finding the chemical constituents that leads to the isolation of medicinally important lead compounds. Senna Alata (L.) Roxb and Senna Hirusta is a popular Indian medicinal tree. The purpose of the present study was to evaluate the phytochemical constituents of the extracts left by Senna Alata and Senna Hirusta. The extraction was carried out in three different solvents to identify and understand the bioactive chemical constituents of the leaf extracts by subjecting the powder to Soxhlet extraction in various solvent systems according to the polarity wise. The yield of each extract was calculated and the methanolic extract found to be more in both. The phytochemical screening was tested for three different extracts (methanol, diethyl ether, and hexane) from two different plant sources in which the two methanolic extracts showed more activity compared to the other two. In Senna Alata methanolic extract shows the presence of alkaloids, flavonoids, steroids, terpenoids, phenols, tannins and Carbohydrates while in Senna Hirusta the phytoconstituents present are alkaloids, flavonoids, steroids, terpenoids, phenols and Carbohydrates. In the quantitative phytochemical analysis, the methanolic extracts of Senna Alata and Senna Hirusta have alkaloid content (26.40 and 4.20), flavonoid content (6.76 and 0.84) and Phenol content (37.48 and 28.76). It shows that Senna Alata's methanolic extract has the potential to scavenge free radicals and can provide leads from multiple medicinal crops in the ongoing quest for natural antioxidants to be used in the treatment of free radical reaction-related diseases that contribute several pharmaceutical applications to humans.

Keywords: Phytochemical analysis, Senna Alata, Senna Hirusta, Alkaloid, Flavonoid, Phenols.

1. Introduction

Man, since time immemorial, herbal or plant products have been used as a medicine to develop immunity or resistance to cold, joint pains, fevers, etc. Scientific data has been well established in a good number of investigated medicinal plants [1]. Nevertheless, only very few plant-origin medicines could enter clinical use and only a dozen medicinal plants could not be approved by the National Formulators. For this purpose, special efforts are required for the production of therapeutic effective herbal drugs [1]. Extraction is separation of the constituents required from plant materials [2]. In recent years, plant-derived compounds have gained more interest in the prevention and cure of human diseases as they are considered more bio-friendly. Over 6,000 plants are commonly believed to be used in traditional folk and herbal medicine in India and Africa, representing about 75 percent of third world countries' medicinal needs [3]. Large amounts of evidence have accumulated to show the promising potential of medicinal plants used in specific conventional, complementary and alternative human disease treatment methods [4]. Numerous plant-derived therapeutic agents have been introduced to modern medicine by medicinal plants [5]. Most plants contain a variety of phyto pharmaceutical products which have found very important applications in the field of agriculture, human and veterinary medicine. Natural products play a leading role in developing novel drug leads for disease treatment and prevention [6, 7].

The natural bioactive compounds found in plants are phyto constituents. Such phyto constituents function with nutrients and fibers to form an integrated part of the system of protection against different diseases and stress conditions [8]. Phytochemicals are generally divided into two classes, i.e. primary and secondary constituents; according to their plant metabolism functions. Primary constituents include common carbohydrates, amino acids, proteins, and chlorophyll while secondary constituents are alkaloids, terpenoids, steroids, and flavonoids, etc. Phytochemical research of plant extracts shows the presence of active principles in plant parts such as flower, bark, leaves, root, fruit,

etc. Phytochemicals are non-nutritious plant chemicals with protective or disease-preventive properties. These chemicals are produced by plants to protect themselves, but research shows that many phytochemicals can protect people from disease [9-11]. Knowledge of plant chemical constituents is important due to the value of such knowledge for the synthesis of complex chemical substances. Plants are rich in a wide range of secondary metabolites, such as alkaloids, flavonoids, saponins, and tannins, etc., found to have anti-microbial properties in vitro [4].

The family Fabaceae belongs to *Senna Alata* and *Senna Hirusta*. *Senna Alata* is a common shrub used to treat parasitic skin disease infections and ringworm infection. Morphological studies describe plants where they grow between 2-5 m in height and possess branches with 10-20 pairs of leaflets distributed horizontally. Flowers are auxiliary racemes with short pedicels and green sepals, bright yellow petals, small calved stamens and dense fruit with quadrangular seeds [12]. To be clear of side effects on diabetic mellitus and its complication is to find the alternative form of medication. *Senna Hirusta* plant [13, 14] is commonly called hairy senna and stinking Senna. It is a perennial terrestrial, erect shrub in tall, stem rounded, solid glabrous up to 150 cm, flowering period from September to December, and fruiting period from November through January [13]. Plant tomentose is soft. Branches grown, leaves with a gland at the base of the petiole 15-20 cm long; stipulations leaner; leaves 4-6 pairs, ovate elliptic, acuminate, rounded or cuneate at the base, 10 x 4 cm. Terminal corimas in penicles; lanceolate bract, acuminate. The antheriferous stamens 6-7, 2 larger. Many were seeding 14 x 0.5 cm [15]. The main purpose of this study was to study the preliminary phytochemical qualitative and quantitative analysis of various extracts of *Senna Alata* and *Senna Hirusta*.

2. Materials and Methods

2.1 Collection of Plant Materials

The fresh samples of *Senna Alata* and *Senna Hirusta* from the yercaurd Hills, Tamil Nadu, were collected randomly. Under running tap water, the sample materials were washed, air dried and then homogenized to fine powder and stored in refrigerated airtight bottles.

2.2 Preparation of Extracts

Extract of the crude sample was prepared by extraction method Soxhlet. About 20gm of powdered sample material was uniformly packed into a thimble and extracted separately with 250ml of various solvents including methanol, diethyl ether, and hexane. The extraction process will proceed for 24 hours, or until the extractor solvent in the siphon tube is colourless. The extract was taken in a beaker afterwards and kept on a hot plate and heated at 30-40°C till all the solvent was evaporated. Dried extract was kept at 4°C in the fridge for future use.

2.3 Phytochemical Screening

Preliminary phytochemical analysis was performed for *Senna Alata* and *Senna Hirusta* methanol, diethyl ether, and hexane extract, as defined by Brain and Turner, 1975 and Evans, 1996[16,17] standard methods.

2.4 Detection of alkaloids

Extracts were dissolved individually in dilute hydrochloric acid and filtered.

- a) **Mayer's test:** Filtrates have been treated with reagent Mayer. Formation of a precipitated yellow cream indicates the presence of alkaloids.
- b) **Wagner's test:** Wagner's reagent was treated with filtrates. Brown/ reddish brown precipitate formation indicates the presence of alkaloids.

2.5 Detection of Flavonoids

- a) **Lead acetate test:** Extracts were treated with a few drops of a solution of lead acetate. Formation of precipitated yellow colour indicates the presence of flavonoids.
- b) **H₂SO₄ test:** Few drops of H₂SO₄ were treated with extracts. Orange colour formation indicates flavonoid presence.

2.6 Detection of Steroids

Liebermann- Burchard test: The extract of 0.5 g was combined with 2ml of acetic anhydride, each with 2ml of H_2SO_4 . In some samples the colour changed from violet to blue or green shows the presence of steroids.

2.7 Detection of Terpenoids

Salkowski's test: The whole plant sample of 0.2 g extract was carefully applied to form a sheet, mixed with 2ml of chloroform and concentrated H_2SO_4 (3ml). A reddish-brown interior coloration suggested the presence of terpenoids.

2.8 Detection of Anthroquinones

Borntrager's test: Approximately 0.2 g of the extract was boiled for a few minutes in a water bath with 10 per cent HCl. It was filtered, and cooling allowed. The filtrate was supplemented with equal volume of $CHCl_3$. A few drops of 10 per cent NH_3 have been added and heated to the mixture. The presence of anthraquinones indicates the formation of pink colour.

2.9 Detection of Phenols

- a) **Ferric chloride test:** With few drops of 5 per cent ferric chloride solution, extracts were treated. Bluish black colour formation indicates presence of phenol.
- b) **Lead acetate test:** Extract was treated with a few drops of a solution of lead acetate. Precipitate yellow colour formation represents the presence of phenol.

2.10 Detection of Saponins

Froth test: Distilled water of quantity 5 ml was taken and around 0.2 g of the extract was shaken. Frothing formation (persistent appearance of creamy stable small bubbles) shows the presence of saponins.

2.11 Detection of Tannins

Ferric chloride test: A small amount of extract was mixed with water and heated on a bath of water. The mixture was filtered and added to the filtrate 0.1 per cent ferric chloride. The presence of tannins indicates a dark green colour.

2.12 Detection of Carbohydrates

Fehling's test: Each of Fehling solutions A and B in a water bath of 0.2gm filtrate is boiled with 0.2ml. A red precipitate indicates sugar content.

Fehling's solution A: Copper sulphate (34.66g) is dissolved in distilled water and made up to 500ml using distilled water.

Fehling's solution B: Potassium sodium tartarate (173g) and sodium hydroxide (50g) is dissolved in water and made up to 500ml.

2.13 Detection of Oils and Resins

Spot test: Test solution was applied on filter paper. It develops a transparent appearance on the filter paper. It indicates the presence of oils and resins.

3. Quantitative Phytochemical Analysis

3.1 Estimation of Alkaloids

Determination of alkaloids using method Harborne (1973). 5ml of the Senna Alata and Senna Hirusta methanolic extract was weighed in a 250 ml beaker and 200 ml of 10 percent ethanol acetic acid was added and covered and allowed to stand for 4 hours. This was purified and one fifth of the original volume concentrated the extract on a water bath. Once the precipitation was complete, concentrated ammonium hydroxide was applied drop wise to the extract. The entire solution was allowed to settle, and the precipitated was collected, washed and filtered with diluted ammonium hydroxide. The residue is the alkaloid which has been weighed and dried.

3.2 Estimation of Flavonoids

Senna Alata and Senna Hirusta of each 5 ml of methanolic extracts were repeatedly collected at room temperature with 100 ml of 80 per cent aqueous methanol. The mixture was then filtered into a pre-weighed 250ml beaker with a filter paper. The filtrate was transferred and weighed into a water bath and allowed to evaporate to dryness. The flavonoid percentage was computed by difference Vellingiri V, Aruna N, Hans KB. 2011. Antioxidant Potential and Health Relevant Functionality of Traditionally Processed Cassia hirsuta L. Seeds: An Indian Underutilized Food Legume [18].

3.3 Determination of Total phenols

For extraction of the phenolic component, the fat-free sample was boiled with 50 ml of ether for 15 min. 5 ml of Senna Alata methanolic extract and Senna Hirusta was pipetted into a 50 ml flask, then 10 ml of distilled water was added. 2 ml of ammonium hydroxide solution and 5 ml of amyl alcohol concentrate were also added. For color growth, the plant samples were made up to label and left to respond for 30 min. That was at 505 nm [19].

4. Result and Discussion

In any phytochemical screening procedure, the preliminary step is extraction. This involves the extraction by standard procedures of medicinally active portions of plant tissues using selective solvents. The extractive value of Senna Alata and Senna Hirusta leaf powder in different solvent system showed the maximum yield percentage was shown by the methanolic extract of both plants. The products thus derived from plants are relatively complex metabolite mixtures, in liquid or semi-solid state or in dry powder form, and are intended for pharmacological uses. Solvents penetrate into the solid plant material during extraction and solubilize compounds with equal polarity [20].

4.1 Qualitative Phytochemical analysis

The result of the preliminary phytochemical screening of the leaves of Senna Alata and Senna Hirsuta are summarized in Table 1. The leaf extracts of Senna Alata methanolic extracts reveals the presence of alkaloid, flavonoid, steroid, terpenoid, phenols, tannin and carbohydrates, where as in diethyl ether terpenoids, carbohydrates, oils and resins, however only carbohydrates, oils and resins is only present in hexane extract. In Senna Hirsuta methanolic extract (alkaloid, flavonoid, steroid, terpenoid, phenols and carbohydrates), diethyl ether (steroids, terpenoids, carbohydrates, oils and resins) and in hexane extract (flavonoids, carbohydrates, oils and resins) were present. Alkaloids are a category of naturally occurring chemical compounds that contain mainly essential nitrogen atoms. Their analgesic properties have been identified [21]. The alkaloids in plants may be used as anesthetic agents in medicine [22]. Once administered to animals, alkaloids have significant physiological effects and hence their wide use in drug development medicine [23, 24].

Table 1: Qualitative Phytochemical Analysis for Senna Alata and Senna Hirusta

Phytochemicals	Senna Alata			Senna Hirusta		
	Methanol	Diethyl ether	Hexane	Methanol	Diethyl ether	Hexane
Alkaloids Mayer's test Wagner's test	+	-	-	+	-	-
Flavonoids Lead acetate test H ₂ SO ₄ test	++	-	-	+-	-	+
Steroids Liebermann-Burchard test	+	-	-	+-	+	-
Terpenoids Salkowski test	+	+	-	+-	+	-
Arthroquinone Borntrager's test	-	-	-	-	-	-
Phenols Ferric chloride test Lead acetate test	+	-	-	+	-	-
Saponin	-	-	-	-	-	-
Tannin	+	-	-	-	-	-
Carbohydrates	++	+	+	+	+	+
Oils & Resins	-	+	+	-	+	+

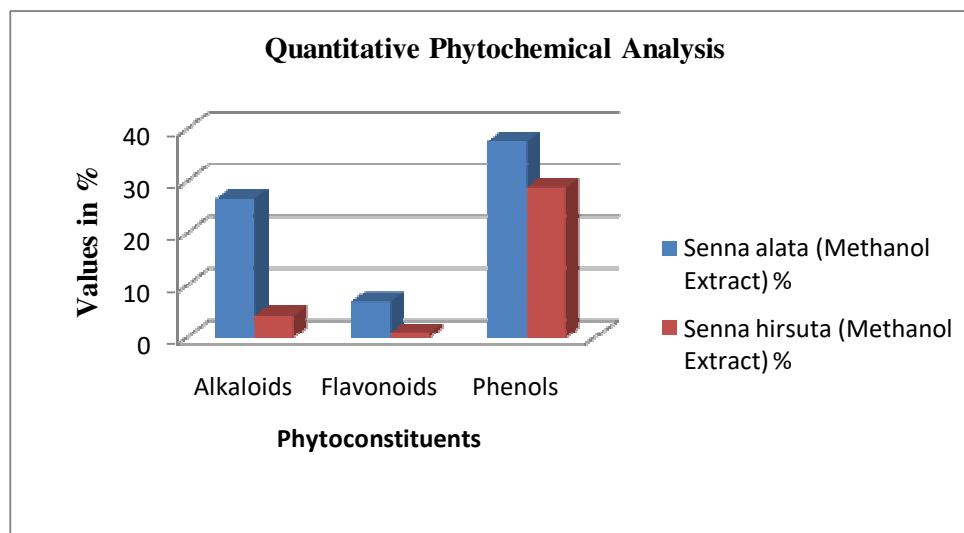
Natural antioxidants come primarily from plants in the form of phenolic compounds such as flavonoids, phenolic acids, etc., [25]. Flavonoids and tannins are a significant group of compounds which act as primary antioxidants or free radical scavengers [26]. Flavonoids are water soluble phytochemicals which, by quenching, up-regulating or protecting antioxidant defences and chelating radical intermediate compounds, reduce free radicals [27]. Phenolic compounds are of great importance because they have a high antioxidant potential which protects the human body from oxidative stress, which can lead to diseases such as cancer, cardiovascular problems and ageing [28]. Tannins contribute to the astringency properties, i.e. faster wound healing and inflamed mucous membrane [29]. Steroids were also identified, and their medicinal value may depend on their relationship to compounds such as sex hormones [30].

4.2 Quantitative Phytochemical Analysis:

The objective assessment of Senna Alata and Senna Hirsuta coated, as described in Table 2. Only methanolic extract has been checked on both plant samples. The findings of quantitative alkaloid content analysis obtained from both the plant extract were 26.40% and 4.20% respectively. In root bark the alkaloid content was lower than in wood. These values are similar to those reported for Senna Alata [31] for (8.50±0.01mg/100 g). Alkaloids are compounds that are more or less toxic that mostly function on the central nervous system [32]. The flavonoid values measured from the two samples were respectively 6.76 percent and 0.84 percent.

Table 2: Quantitative Phytochemical Analysis of Senna Alata and Senna Hirusta

Phytoconstituents	Senna Alata (Methanol Extract) %	Senna Hirusta (Methanol Extract) %
Alkaloids	26.40	4.20
Flavonoids	6.76	0.84
Phenols	37.48	28.76

**Figure 1: Quantitative Phytochemical Analysis of Senna Alata and Senna Hirsuta**

In Senna Alata leaf the flavonoid content was higher than in the Senna Hirsuta leaf extract. The phenol content measured from this study was 37.48 percent for both Senna Alata and Senna Hirsuta leaf extract, and 28.76 percent. The root bark concentration was found small as opposed to the value of the skin. Such values were high when compared with Senna Alata at 2.00 ± 0.21 mg/100 g [29] research on leaf. Alkaloid, flavonoid, and phenols have the ability to cure the disease. Compared with Senna Hirsuta used for medicinal purposes, the content of Senna Alata methanolic extracts was found to be high.

Consequently, the results obtained in this study suggest that the identified phytochemical compounds may be the bioactive constituents responsible for the effectiveness of the studied plants Senna Alata and Senna Hirsuta. It has also been confirmed that the presence of some of these compounds has antimicrobial, antioxidant and anticancer properties. It could therefore be concluded from the analysis that the leaf extracts could be a source of useful in the chemotherapy of some microbial infection for the industrial manufacture of drugs. The presence of these phytochemicals could be attributed to the bioactive principles of most medicinal plants, including the plants under study, which are responsible for ethno pharmacological activities.

5. Conclusion

The presence of these phytochemicals has been confirmed by several studies contributing medicinal properties to the plants. Extracts from this plant may therefore be viewed as a good source for useful drugs. Preliminary qualitative testing is useful in the detection of bioactive principles, and may result in the discovery and development of drugs. Phytochemical studies have revealed that the methanolic extract *Senna Alata* and *Senna Hirsuta* is rich in many active phytoconstituents that give physiological reaction. Nevertheless, to explore this plant's secret therapeutic efficacy, a detailed analysis of plant material is needed in-order. It is also important to follow several phytochemical methods to isolate, purify and classify the active constituents present in this plant, which could later become promising for the production of drugs.

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