

## RESEARCH ARTICLE

## Preliminary phytochemical and antimicrobial screening of solvent extracts of roots of *Andrographis paniculata* and stem bark of *Bombax ceiba*

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**ABSTRACT**

Medicinal plants are used to cure common ailments by the people of the tribal area. *Bombax ceiba* and *Andrographis paniculata* are selected for the antimicrobial and phytochemical analysis on the basis of medicinal folklore reports and literature data. The selection of plants for evaluation was based on traditional use for treatment of common ailments. *Bombax ceiba* is known Katesawari, semal, savari. It is used in diarrhoea, dysentery & menorrhagia. *Andrographis paniculata* belongs to the family of Acanthaceae and is popular worldwide with the name of "King of Bitters" in English. It is an annual herbaceous plant which is widely cultivated in Southern Asia, India, China and some parts of Europe. Qualitative phytochemical screening of stem bark *Bombax ceiba* and roots *Andrographis paniculata* was studied. The methanol solvent was used to obtain extracts from powdered plant parts. The extracts were subjected to qualitative phytochemical screening using standard procedures. Results show that six of seven phytochemicals screened for, were present in stem bark of *Bombax ceiba*. They are; carbohydrate, flavonoids, phenols, saponins, sterols and tannins. Five of seven phytochemicals screened for, were present in the roots *Andrographis paniculata*. They are; carbohydrates, phenols, saponins, alkaloids and tannins. The diversity of phytochemicals present suggests that *Bombax ceiba* stem bark and *Andrographis paniculata* roots could serve as a source of useful drugs.

**Keywords:** *Andrographis paniculata*, *Bombax ceiba*, Phytochemicals, Antibacterial, Roots, Stem bark.

**INTRODUCTION**

Phytochemicals are bioactive chemicals of plant origin. They are regarded as secondary metabolites and are naturally synthesized in all parts of the plant i.e. any part of the plant may contain active components (Tiwari *et al.*, 2011). The quantity and quality of phytochemicals present in plant parts may differ from one part to another. In fact, there is lack of information on the distribution of the biological activity in different plant parts essentially related to the difference in distribution of active compounds (or active principles) which are more frequent in some plant parts than in others (Lahlou, 2004). Phytochemicals have been recognized as the basis for traditional herbal medicine. The presence of a phytochemical of interest may lead to its further isolation, purification and characterization. Then it can be used as the basis for a new pharmaceutical product.

*Bombax ceiba* (Bombacaceae) is large handsome deciduous tree, trunk with prickles. Lokal name of *Bombax ceiba* is Katesawari, semal, savari. The root has stimulant & tonic properties. The bark & the root are emetic. The roots of saplings up to about three years old are know as 'semarkanda' in the Central Provinces & are used as a nerve tonic & as an astringent . The gum is used as a tonic & astringent .It is also used in diarrhoea, dysentery & menorrhagia (Caius, 1939).

The genus *Andrographis* consists of 28 species, only a few species are medicinal. *Andrographis paniculata* is an annual herb extremely bitter in taste. The plant is known in India by various vernacular names. It is also known as Kalmegh or Kalamegha. Local name of this plant is Bhui-neem. It has a strong bitter taste similar to the Neem tree. *Andrographis paniculata* has an important place in the Indian Pharmacopoeia and is one of the most widely used plants in ayurvedic formulations (Hooker, 1885). The whole plant has variety of therapeutic values. It has immune-suppressive properties and is useful in treatment of wounds, ulcers, leprosy, sore throat, and hypertension, (Puri *et al.*, 1996). Panchang (stem, leaves, flowers, root and seeds) of the plant is being used in various formulation of Indian system of medicine for the treatment of fever, malaria and sore throat (Chopra *et al.*, 1956). It has been used in the treatment of some skin infections in India by folkloric medicine practitioners (Jain, 1991).

In the present study, extracts of stem bark of *Bombax ceiba* and root of *Andrographis paniculata* were qualitatively screened for phytochemicals and antimicrobial activities using standard tests.

## MATERIALS AND METHODS

**Collection of Plant Materials:** Bark of *Bombax ceiba* and roots of *Andrographis paniculata* were collected from different parts of the Chandrapur district. The material for the present investigation was collected from villages inhabiting tribals in the Chandrapur & Gadchiroli districts of Maharashtra. The information was gathered by questioning local healers and knowledgeable villagers.

**Preparation of sample:** Collected samples are immediately chopped into small fragments. Stem, root, & bark are chopped into 3-5 cm pieces & splint longitudinally into several sections. Herbaceous

samples & large leaves & fruits are also cut into small pieces.

Roots were washed thoroughly to remove soil before chopping. Chopping of fresh samples after collection hasten drying & its advantages are reduction in volume & increase in drying speed. Chopping permitted rapid drying of samples, otherwise they did not dry for weeks (Mehrotra, 1996)

The dried material pulverized by a mechanical grinder. The material is coarsely ground by a mechanical grinder. The coarsely powder was stored in plastic container. The powdered material was kept in air tight plastic container & stored at low temperature till required for further study.

**Solvent extraction:** The shade dried powder of the bark of *Bombax ceiba* and root of *Andrographis paniculata* (50 gm each) was packed well in Soxhlet apparatus and was subjected with water and methanol by continuous hot extraction for about 24 hrs. The extracts were filtered through Whatman filter paper and concentrated on a water bath. The final concentrated extracts were stored in refrigerator in bottles. The above prepared methanol extracts were used for phytochemical and anti-bacterial investigation. The dried extracts were tested for their phyto constituent's by standard methods.

## Phytochemical Screening

### Test for Alkaloids

**Mayers test:** To a little of the test filtrate taken in watch glass a few drops of the Mayer's reagent reagent were added. Formation of cream colored precipitate showed the presence of alkaloids.

**Test for carbohydrates: (Molisch's test)** Few drops of Molisch's reagent + 2ml of the extracts. Then add 2ml of conc. H<sub>2</sub>SO<sub>4</sub> down the side of the test tube. The mixture was then allowed to stand for few minutes. Formation of a red or violet colour at the Junction of the two layers indicate positive test.

**Tests for flavonoids:** A small quantity of test residue was dissolved in 5 ml ethanol (95%) and treated with few drops of concentrated hydrochloric acid and 0.5 gm of magnesium. The pink or magenta colour is developed within three minutes, if flavonoids are present.

**Test for Phenols (Ferric chloride test):** A fraction of the extracts was treated with aqueous 5% ferric

chloride and observed for formation of deep blue or black colour.

**Test for Amino acids and Proteins (1% ninhydrin solution in acetone):** 2ml of filtrate was treated with 2-5 drops of ninhydrin solution placed in a boiling water bath for 1-2 minutes and observed for the formation of purple colour.

**Test for Saponins (Foam test):** To 2mls of extract was added 6ml of water in a test tube. The mixture was shaken vigorously and observed for the formation of persistent foam that confirms the presence of saponins.

**Test for Sterols (Liebermann-Burchard test):** 1ml of extract was treated with drops of chloroform, acetic anhydride and conc.H<sub>2</sub>SO<sub>4</sub> and observed for the formation of dark pink or red colour.

**Test for Tannins (Braymer's test):** 2ml of extract was treated with 10% alcoholic ferric chloride solution and observed for formation of blue or greenish colour solution. The results obtained were recorded as --, +, ++, +++ signs, indicating their approximate concentrations. The results are given in Table 1.

**Anti-bacterial screening:** Agar well-diffusion method (Okeke, 2001) was used for the anti-bacterial study. The overnight culture grown was used for inoculation. For working stock 1 mL of each bacterial strain was initially inoculated in 100 mL of sterile nutrient broth and incubated for 37° ± 1°C for 24 hr respectively. Then 0.2 mL of the each test organisms from the working stock was seeded into 100 mL sterile nutrient agar medium and cooled to 48°C to 50°C in a sterile Petri dish respectively. When the nutrient agar

medium solidifies, four holes of uniform diameter (6mm) were made using sterilized cork borer. Then, 0.2 mL of each methanol extracts and standard solution were placed in each hole separately. The plates were maintained at room temperature for 2 hr to allow the diffusion of the solution into the medium.

All the bacterial plates were incubated at 37° ± 1°C for 18 hr and the zone of inhibition was measured. Triplicates were maintained for each sample of the extracts respectively. For each bacterial strains control were maintained where pure solvents were used. All the diameters of inhibition zone were measured in mm. The results are shown in Table 2.

## RESULTS AND DISCUSSION

**Phytochemical Screening:** Results obtained for qualitative screening of phytochemicals in stem bark of *Bombax ceiba* and roots of *Andrographis paniculata* are presented in table 1.

Of the seven phytochemicals screened for, five were found present in solvent extracts of *Andrographis paniculata*. They are alkaloid, phenols, carbohydrates, saponins and tannins. Phytochemical screening of stem bark of *Bombax ceiba* found to contain carbohydrate, flavonoids, phenols, saponins, sterols and tannins. In all, more phytochemicals were found present in the stem bark than in the roots. Remarkably, flavonoids were not present in roots but present in stem bark. This suggests that the stem bark offers a wider array of phytochemicals than the root. The diversity of phytochemicals present suggests that *Bombax ceiba* stem bark and *Andrographis paniculata* roots could serve as a source of useful drugs.

**Table 1: Phytochemical Analysis of Plant Extracts**

Plant Species	Part	Alkaloid	Saponins	Flavonoids	Tannins	Phenols	Sterols	Carbohydrates
<i>Andrographis paniculata</i>	Root	+++	++	-	+	++	-	+
<i>Bombax ceiba</i>	Bark	-	++	+	+++	+	+	++

Abbr: - = Absent, + = Presence, ++ = Moderate, +++ = Good

**Table No 2: Results showing the antibacterial activity of plant extracts**

Plant	Plant part	Extract	M.O.								
			S.a.	E.c.	B.s.	V.c.	S.d.	K.p.	S.t.	P.a.	P.v.
<i>Andrographis paniculata</i>	Root	ME	12	10	12	11	17	11	14	15	13
<i>Bombax ceiba</i>	Bark	ME	14	--	11	--	--	--	--	9	--

ME=Methanol; M.O.= Microorganism; S.a.=Staphylococcus aureus; E.c.=Escherichia coli; B.s.=Bacillus subtilis; V.c.=Vibrio cholerae; S.d.=Shigella dysenteriae; K.p.=Klebsiella pneumoniae; S.t.= Salmonella typhi; P.a.=Pseudomonas aeruginosa; P.v.=Proteus vulgaris.

**Anti-bacterial screening:** Results of the antibacterial screening of methanol extracts of roots of *Andrographis paniculata* revealed significant antibacterial activity against all tested bacterial strains. The results are shown in table 2.

The methanol extract of roots of *Andrographis paniculata* showed more significant activity against all tested bacterial organisms than that of the stem bark extract of *Bombax ceiba*. The maximum antibacterial activity of methanol extracts of *Andrographis paniculata* was exhibited against *Shigella dysenteriae* and of *Bombax ceiba* was exhibited against *Staphylococcus aureus*. The inhibitory effects of *Andrographis paniculata* and of *Bombax ceiba* on the test micro-organisms may be due to the presence of the above phytochemical components. The various phytochemical compounds detected are known to have beneficial importance in medicinal science. The phenols along with antimicrobial activity also show astringent properties (Adelheid Brantner' 1994). Flavonoids and other polyphenols have been shown to exhibit significant antioxidant activity. Alkaloids have been used to treat diseases like malaria and glycosides serve as defense mechanisms against many micro-organisms. Alkaloids generally present in both parts play some metabolic role and control development in living system (Lalitha *et al.*,2012). They are also involved in protective function in animals and are used as medicine especially the steroidal alkaloids (Sharma *et al.*,2011). Saponin protects the plant against microbes and fungi.

## CONCLUSION

Phytochemicals found present in root and stem bark extracts of *Andrographis paniculata* and *Bombax ceiba* indicates their potential as a source of principles that may supply novel medicines. Hence the crude methanol extract of *Andrographis paniculata* Root and bark extract of *Bombax ceiba* can be used for further purification and preparation of new anti-microbial for the more resistant type of micro-organism. The above findings recommend the further investigation of *Andrographis paniculata* and *Bombax ceiba* to evaluate their chemical potential.

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