

Anthelmintic Activity of Aqueous Leaf Extract of *Clerodendrum Serratum*

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Abstract: *Clerodendrum serratum* Linn. (Family: Verbenaceae) is very widely distributed in tropical and subtropical regions of the world. Ethno-medicinal importance of the plant has been reported in various indigenous systems of medicines like Ayurveda, Siddha and Unani for the treatment of various life-threatening diseases such as syphilis, typhoid, cancer, jaundice and hypertension. Some of the chief constituents found in the plant are D-mannitol, hispidulin, cleroflavone, apigenin, scutellarein, serratagenic acid, acteoside, verbascoside, oleanolic acid, clerodermic acid, γ -sitosterol, β -sitosterol, cholestanol, clerosterol, campesterol and 24-ethyl cholesterol. Traditionally, it has been also used as anti-rheumatic, anti-asthmatic, febrifuge, in cephalgia and ophthalmia. The roots of *C. serratum* are also used as anti-oxidant, anti-bacterial, and anti-fungal. Besides these the antimicrobial value of this herbal plant have also been reported in its stems and leaves. These reports are very encouraging and indicate that herb should be studied more extensively for its therapeutic benefits. *Clerodendrum serratum* is a shrub which is not much branched with stems. The root of the plant is attributed with various activities like anti-inflammatory, digestive and carminative and many more. It is used to treat the conditions like inflammations, anorexia, cough, asthma, hiccough, tubercular glands, skin diseases etc. Various minerals like Na, Mg, Al, Ca etc. saponins, terpenoids, D-mannitol are the phytoconstituents present in the plant. Research works are carried out to study the pharmacognostic, physicochemical, hepatoprotective, anti-oxidant, anti-inflammatory, analgesic, antiasthmatic and various other activities.

Keywords: Anthelmintic , *Clerodendrum Serratum* , Helminthiasis , Anti-inflammatory

1. Introduction

India is known for its traditional medicinal systems—Ayurveda, Siddha, and Unani. Medical systems are found mentioned even in the ancient Vedas and other scriptures. The Ayurvedic concept appeared and developed between 2500 and 500 BC in India. The literal meaning of Ayurveda is “science of life,” because ancient Indian system of health care focused on views of man and his illness. It has been pointed out that the positive health means metabolically wellbalanced human beings. Ayurveda is also called the “science of longevity” because it offers a complete system to live a long healthy life. It offers programs to rejuvenate the body through diet and nutrition. It offers treatment methods to cure many common diseases such as food allergies, which have few modern treatments. However, one should be aware that Ayurvedic nutrition is not a “magic bullet” system but requires the full participation of the patient to succeed. It is an interactive system that is user-friendly and educational. It teaches the patient to become responsible and self-empowered. Ayurveda is not a nutritional system for those seeking an escape or excuse to further abuse their body or mind. It is a system for empowerment, a system of freedom, and long life.

The medicinal plants have various secondary metabolites in them. Because of these principles they are widely used in the entire world by the people to cure various ailments. *Clerodendrum serratum* (Verbenaceae) is a tropical medicinal plants distributed in the forest of western ghates and various forests of India. In Indian system of medicine, that plant is well known as bharangi (Sanskrit) and commonly known as blue glory (English) and Gantu bharangi (Kannada) . As per the traditional claims roots are the potential source of drugs for

ailments such as asthma, body ache, bronchitis, fever, cholera dropsy, eye disease, inflammation, malaria, snake bite, rheumatism, tuberculosis wounds and also used for skin infection, Helminthic infection (roots and leaves).

Helminthiasis

Helminthic infections refer to worms that live as parasites in the human body. Worm infection occurs when infective eggs, or larvae, enter the body, mature, lay eggs and feed off the person. People get infected with worms when living in an unclean environment of poor sanitation and unhygienic habits. The three main types of common intestinal worms that infect humans are large intestinal roundworm (*Ascaris umbricoides*), hookworm (*Ancylostoma duodenale* and *Necator americanus*) and whipworm (*Trichuris trichuira*). Globally, more than 3.5 eople are infected with intestinal worms.

Helminths- (Worms) are multicellular parasites.

Helminthiases

Helminthases also known as worm infection, is any macroparasitic disease of humans in which a part of the body is infected with parasitic worms, known as helminthes. The Leaves extract of *clerodendrum serratum* were evaluated for anthelmintic activity in *pheretima posthuma* (Indian earth worm) of nearly equal size (6+_1cm). *Pheretima posthuma* is used due to its anatomical and physiological resemblance with the intestinal roundworm parasite of human beings. Because of easy availability of earth worms, they have been used widely for evaluation of the anthelmintic drugs.[4]

Plant Profile**Clerodendrum Serratum****Taxonomy:-**

Domain - Eukaryota

Kingdom - Plantae

Sub-kingdom - Viridaplantae

Phylum - Tracheyophyta

Sub-phylum - Euphylllophytina

Division - Angiospermae

Class - Magnoliopsida

Subclass - Lamiidae

Order - Lamiales

Family - Lamiaceae/Verbenaceae

Sub-family - Ajugoideae

Genus – Clerodendrum

Species – Serratum

Vernacular Names:

Bengali - Bamunhatee, Bamanhatee, Bhuijam

English - Blue glory, Beetle killer

Gujarati - Bharangee

Hindi – Bharangi

Kannada-Gantubrangee

Malayalam – Cheruthekkku

Marathi - Bharangee, Bharangi

Oriya – Chinds

Punjabi – Bhdangee

Sanskrit - Brahmanayashtika, Barbura

Tamil – Cheruteku

Telugu – Ganttubrabangee

Habitat:

Clerodendrum serratum is a genus of flowering plants in the Verbenaceae family. Estimates of number of species in Clerodendrum serratum vary widely, about 450 species. The genus is native to tropical and warm temperate regions of the world, with most of the species occurring in tropical Africa and southern Asia, but some in the tropical Americas and northern Australia, and a few extending north into the temperate zone in eastern Asia.

Macroscopic:

Clerodendrum serratum is a slightly woody shrub with bluntly stems and branches. These trees are about 2-8 ft high. It is annual or perennial, usually aromatic (Findmeacure, Ayurvedic, Zipcodezoo).

Root:

Mature root hard, woody and cylindrical; up to 5 cm thick; external surface light brown having elongated lenticels.

Stem:

Usually quadrangular (four angled).

Bark:

Thin and easily separated from a broad wood which shows marked medullary rays and concentric growth rings in a transversely cut surface; short fractures; acrid taste.

Leaf:

Leaves usually three at a node, sometimes opposite oblong or elliptic, serrate, alternate without stipules.

Flower:

Blue, many in long cylindrical thyrsus. They are bisexual, zygomorphic, rarely subactinomorphic, bracteolate or not. Corolla with a slender tube, lobe-5, spreading; stamens epipetalous, 4 or 2, free; anther 1 or 2-celled usually dehiscent longitudinally, disc persistent. Ovary superior, 2-celled and each cell 2-ovuled; and style sub-terminal and gynobasic.

Chemical Constituents:

The major groups of chemical constituents present in the Clerodendrum genus are carbohydrates, phenolics, flavonoids, terpenoids and steroids.

Phenolics:

The phenolic compounds in the genus Clerodendrum are found in both free as well as bound to sugar moiety. Some of the phenolic compounds isolated were serratagenic acid, acteoside, indolizine and verbascoside which possess biologically activities such as anti-oxidant, antimicrobial, anti-proliferative, antihypertensive and anti-cancer activities.

Terpenes:

Terpenoids are generally found to be bound to sugar moieties by a glycoside linkage. Usually they are present as glycosides in their beta-D-glucosidic form. Some of the terpenes isolated from the plant like betulin, oleanolic acid, clerodermic acid, betulinic acid, friedelin and monomelittoside had weak CNS activity, strong molluscicidal and fungi toxic activities.

Steroids:-

Steroids are terpenes based on the cyclopentane perhydroxy phenanthrene ring. Chiefly, gamma-sitosterol, beta-sitosterol, cholestanol, clerosterol, campesterol and 24-ethyl cholesterol were reported to be isolated from the plant.

Traditional Uses

- 1) Roots and leaf extracts of *Clerodendrum serratum* has been used for the Treatment of rheumatism, asthma, anorexia, leucoderma, leprosy, fever and other inflammatory diseases.
- 2) The roots of the plant have been claimed to be used in dyspepsia, seeds in Dropsy and leaves as a febrifuge and in cephalgia and ophthalmia.
- 3) Aqueous extracts of leaves of *Clerodendrum serratum* possess bronchodilator Property Previous studies suggest that apigenin-7-glucoside has demonstrated anti-inflammatory, antimicrobial, hepatoprotective and anti-diarrheal properties. The compound also showed significant protection against Alzheimer's diseases in mice.

2. Materials, Methods and Experimental work:-

Collection of Plant:

The fresh Leaves of *Clerodendrum serratum* were collected during the months November–December from the Wild sources of India. The plant material was taxonomically identified and authenticated by Dr. B.J. Patil Department of Botany, S. G. M. College, Karad. The Leaves of *Clerodendrum serratum* were dried in the shade, milled into coarse powder by a mechanical grinder and stored in air tight closed container.

Study design:

Media was prepared by dissolving extract in normal saline of different concentrations. Helminthes were inserted into this media. Effects were observed.

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Materials

Plant material used	<i>Clerodendrum serratum</i> Leaves
Animal used	Indian earth worm (<i>pheretima posthuma</i>)
Apparatus	Conical flask, Petridis, beaker, measuring cylinder, etc.
Chemicals	Alcohol, Dragondroff's reagent, mayer's reagent, sodium hydroxide, lead acetate, normal saline solution, D/W, etc

Method

Extraction Method – Maceration Extraction Method (Aqueous Extract)

Preparation of plant extract-

Extract prepared by Maceration process:

- Following steps are involved in the maceration process
- Plant material (cut, dried)
- After shade drying aerial part of *clerodendrum serratum* blended into fine powder with a blender. (crushed or moderately coarse powder)
- Aqueous extract prepared by cold maceration process.
- The powder Placed in a closed vessel (conical flask) (135gm drug).
- Whole of the selected solvent added (675 ml water)
- Allow to stand for seven days, shaking occasionally.
- Liquid strained off, solid residue pressed.
- Strained and expressed liquid mixed. Clarified by filtration, evaporation and concentration

Phytochemical Test

1. Test for Carbohydrate

Test	Observations	Inference
i] Molish Test- Take a 2 ml extract in a clean and dry test tube. Add 2 drops Ethanolic Alpha-naphthol and mix. Incline the test tube and add carefully 2 ml of concentrated H ₂ SO ₄ (Sulphuric acid) along the side of the test tube	Purple colour ring form at the junction of two liquid.	Carbohydrate are Present
ii] Benedicts Test- Take a 5ml Benedicts reagent add 8 drop of extract boil it for two minutes and cool it	Green to Yellow precipitate	Carbohydrate are Present
iii] Fehling Test In a test tube add 2 ml of extract and add equal volumes of fehling B solution and place it in boiling water bath for two minute	Brownish Red Precipitate	Carbohydrate are Present
iv] Barfoed's Test - 2ml Barfoed's reagent add 2ml of extract keep the test tube in boiling water bath for 3 minute and cool	A brick red precipitate	Carbohydrate are Present

2. Test for Steroids and Terpenoids

Test	Observations	Inference
i]Salkowski reaction – 2mg of dry extract was shaken with chloroform, to the chloroform layer sulphuric acid was added slowly by the sides of test tube	Formation of Red Colour	Steroid and Terpenoid are Present
ii]Lieberman Burchard's Test – 2mg of dry extract was dissolved in acetic unhydride heated to boiling cool and then 1ml of concentrated sulphuric acid was added along the side of the test tube	Formation of Green Colour	Steroid and Terpenoid are present

3. Test for Flavanoids

Test	Observations	Inference
i]Shinoda Test- The dried extract were dissolved in 5ml Ethanol intest and few drops of concentrated Hcl Were added then add the magnesium turnings	Formation of Pink colour	Flavonoids are Present
ii]Lead acetate test- To small quantity of above residue lead acetate was added	Formation of yellow colour precipitate	Flavonoids are Present
iii]Sodium hydroxide test- Take extract tube and add 5%NaOH	Yellow colour form which is decolourise with addition of Glacial acetic acid	Flavonoids are present

4. Test for saponins-

Test	Observation	Inference
i]Foam Test- Place 1ml extract and 2ml water in test tube shaken well	Development of stable Foam for 10 minute	Saponin are Present

Chemical constituents

The major group of chemical constituent present in the clerodendrum genus is carbohydrates, phenolics, flavonoids, terpenoids, and steroids.

4. Anthelmintic Activity

Experimental Procedure

The leaves extract of clerodendrum serratum were evaluated for anthelmintic activity in pheretima posthuma (Indian earth worm) of nearly equal size (6+ 1cm).

Test sample of extract was prepared at the concentrations 100, 200, 300 and 400mg/ml in normal saline solution.

Standard Albendazole tablet solution was prepared at the concentrations 100, 200mg.

All the test solution and standard drug solution were prepared freshly before starting experiment.

The Indian earth worm (pheretima posthuma) was placed in different 4 petridish containing 20 ml of above test solution of extracts at different concentration.

Observations were made for the time taken for paralysis was noted when no movement of any sort could be observed except when the worms were shaken vigorously.

Time for death of worms were recorded after ascertaining that worms neither moved when shaken vigorously nor when dipped in warm water.

The same above procedure are also used for standard drug.

5. Result and Discussion

Activity of Leaves Extract and Standard Drug

Treatment	Dose mg/ml	Paralysis Time	Death Time
Test drug	100mg	3hrs 4mins 25sec	3hrs 35mins 20sec
Test drug	200mg	2hrs 40mins 22sec	2hrs 58mins 23sec
Test drug	300mg	1hrs 45mins 15sec	2hrs 2mins 9sec
Test drug	400mg	47mins 17sec	1hrs 18mins 1sec
Std.drug	100mg	16mins 4sec	50mins 15sec
Std.drug	200mg	10mins 2sec	25mins 30 sec

The result in above table show that the time taken for paralysis and death of earth worms after treating with test substance. The activity was compared with standard drug (Albendazole). Water extract of Clerodendrum serratum show significant Anthelmintic activity against earth worm. From above our observation, (400mg/ml) higher concentration of extract produced paralysis effect much earlier and the time taken for death was shorter for earth worm. 100mg/ml has taken little more time to cause paralysis and death of earth worm as compared with standard drug. Therefore, further study must be carried out so that the general people can get actual benefit from his important medical plant

6. Conclusion

Conclusion, the use of Leaves of Clerodendrum serratum as an anthelmintic have been confirmed as the roots extracts displayed activity against the earth worms.

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