



A REVIEW ON ANTI-DIARRHOEAL ACTIVITY OF *CAPPARIS ZEYLANICA*

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ABSTRACT

Diarrhoea is typically a sign of intestinal disorders, which can be brought on by a wide range of bacterial, viral, and parasitic (protozoa and helminths) organisms (including *E. coli*, *Vibrio cholerae*, *Shigella* species, and others). According to ethnopharmacological research, *L. camara* has been shown to have antibacterial, antifungal, and anthelmintic properties against those microorganisms that cause diarrhoea. Additionally, it has properties that are antimycobacterial, antioxidant, antinociceptive & anti-inflammatory, antimalarial, antiulcerogenic, and anti-leishmaniasis. The leaves of *L. camara* and its components (Lanthadne A) have been shown in in vivo investigations to exhibit antimotility and anti-diarrhoeal action in mice model of diarrhoea. Additionally, an in vitro investigation on an excised rat ileum revealed this plant has antispasmodic activity by opposing the effects of acetylcholine and compounds that can reduce gastrointestinal muscular spasm have been hypothesised to have antidiarrheal properties.

Since ancient times, people all over the world have employed plants and plant extracts in homoeopathic medicines, herbal treatments, and traditional therapies. The raw medications made from various medicinal plant components have a variety of therapeutic characteristics. The study of phytochemicals based on ethnopharmacological data is widely regarded as an efficient method for discovering novel bioactive principles. Therefore, there is a perceived need to find innovative drugs that are less expensive, less expensive economically, and have fewer severe side effects.

Keywords- *E.Coli*, *Vibrio*, Diarrhoea, Antimotility.

Introduction- One of the most typical health issues is diarrhoea. It might range from a minor, transient ailment to one that could be fatal. Each year, there are reportedly 2 billion instances of diarrhoeal illness worldwide. Additionally, around 1.9 million children under the age of five die from diarrhoea each year, predominantly in underdeveloped nations. As a result, it is now the second most common cause of mortality for people in this age range.

Stools that are unusually loose or watery are signs of diarrhoea. Bacteria, viruses, or parasites are the most common causes of diarrhoea. Chronic diarrhoea can also be caused by digestive system diseases.

It is not diarrhoea if a person often passes faeces that are of a normal consistency. Similar to this, breastfed infants frequently pass loose, sticky faeces. The causes and remedies of diarrhoea are examined in this article. Symptoms, diagnosis, prevention, and when to consult a doctor are also covered.

An infection in the digestive tract is the cause of diarrhoea in many cases. The bacteria that caused this illness include the following:

- bacteria
- viruses parasitic organisms

The most commonly identified causes of acute diarrhoea in the United States are the bacteria *Salmonella*, *Campylobacter*, *Shigella*, and *Escherichia coli*.

Even if all of the digestive organs seem to be operating normally, certain forms of chronic diarrhoea are referred to as "functional" instances because of this. Irritable bowel syndrome (IBS) is the most prevalent cause of functional diarrhoea in the industrialised world.

Constipation, diarrhoea, or both may be among the changed bowel habits that IBS patients experience, in addition to cramping and stomach pain.

The condition known as IBD is another reason why people have persistent diarrhoea. IBD refers to Crohn's disease or ulcerative colitis. Blood in the stool is another symptom of either illness.

Some other major causes of chronic diarrhoea include:

- **Microscopic colitis:** This is a persistent type of diarrhoea that usually affects older adults. It develops due to inflammation and occurs often during the night.
- **Malabsorptive and maldigested diarrhoea:** The first is due to impaired nutrient absorption, and the second is due to impaired digestive function. Celiac disease is one example.
- **Chronic infections:** A history of travel or antibiotic use can be clues in chronic diarrhoea. Various bacteria and parasites can also be the cause.
- **Drug-induced diarrhoea:** Laxatives and other drugs, including antibiotics, can trigger diarrhoea.
- **Endocrine-related causes:** Sometimes, hormonal factors cause diarrhoea. This is the case in Addison's disease and carcinoid tumours.
- **Cancer-related causes:** Neoplastic diarrhoea is associated with a number of guts cancers[39].

Plant Profile-; Name: - *Capparis zeylanica* L.

Capparis zeylanica is an evergreen climbing shrub producing stems 2 - 5 metres long, occasionally to 10 metres. The plant is harvested from the wild for local use as a medicine and occasionally as a food.

Description:-

Stragglers, branchlets adpressed tomentose. Leaves 7-9 x 5-6 cm, ovate, apex mucronate, base truncate, pubescent; petiole to 1 cm, densely pubescent, stipular spines small, in pairs, recurved. Flowers in supra-axillary rows, 3-4 cm across, white, buds densely pubescent; pedicels 2-4 cm, pubescent; stamens numerous, long exerted, white, turns to brown, gyn androphore as long as or longer than filaments; ovary 2.5 mm, ellipsoid.

Natural History

Cyclicality:- Flowering and fruiting: March-May

Flowering occurs in March and fruits ripen during October-November

Flower In axillary clusters; stamens cream when anthesis, red to purple in the evening. Flowering from February-April.

Fruit An ovoid berry, pendulous, smooth, pustulate; blood red when ripe; seeds many. Fruiting April onwards.

Field tips Branchlets stellate tomentose, with recurved spines.

Leaf Arrangement:- Alternate-spiral

Leaf Type:-Simple

Leaf Shape:- Ovate, elliptic or lanceolate

Leaf Apex:- Obtuse-retuse or mucronate

Leaf Base:- Cuneate-obtuse

Leaf Margin: - Entire

A shrub leaves simple with stipulary thorn found generally in upper Assam. Fruit is hairy and flowers are generally white. The leaves are employed as a counter-irritant, and are made into a poultice for treating boils, swellings and haemorrhoids They are also used to reduce perspiration and to improve the appetite. A decoction of the root-bark is used as a remedy for vomiting and to improve the appetite

Habitat and Distribution

Common in plains from coast to 1000m. India, Burma, Sri Lanka, Indo Chinese peninsula, Andamans, Malaysia.

Global Distribution: - Indo-Malesia and China

Indian distribution:- India: Assam, Maharashtra, Meghalaya, Odisha, Tamil Nadu, Uttar Pradesh, Eastern Himalayas. Very common in the deciduous forests from foothills to 400m. India, Sri Lanka to Myanmar, Thailand, Indo-China and Malesia .

EXPERIMENTALWORK: Preparation of plants herbarium sheet and authentication of *Capparis zeylanica* roots:

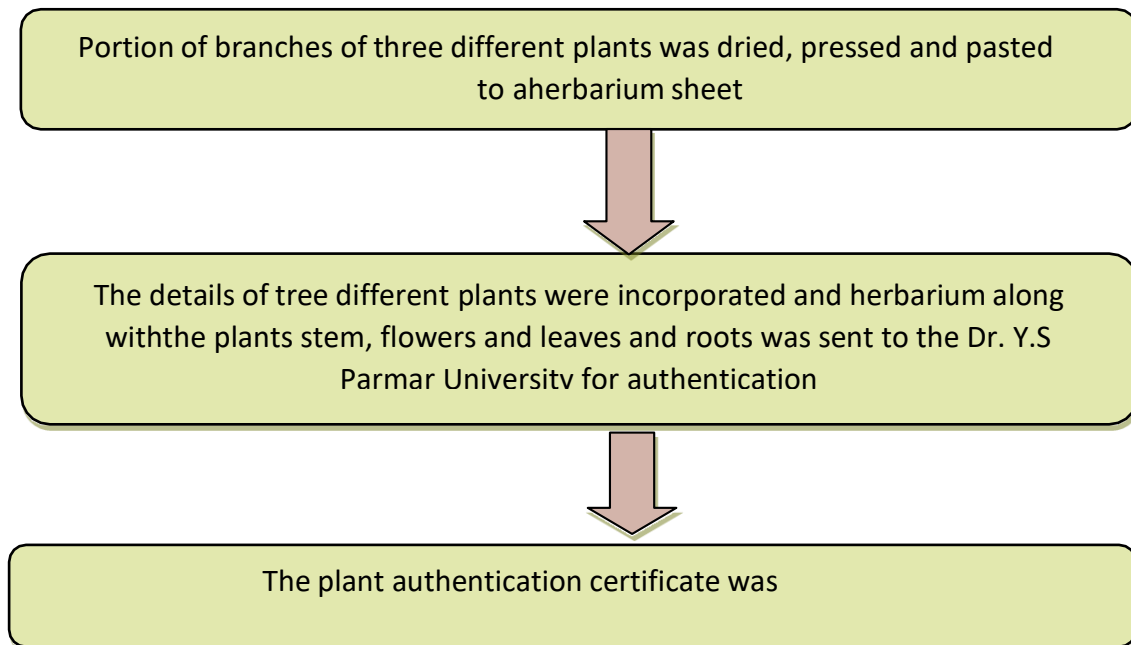


Figure No. 5: Schematic representation for the process of preparation of plant herbarium and authentication of *Capparis zeylanica*

5.1 Preparation of leaves and roots powder: A fresh leaves of *Capparis zeylanica* were collected, washed thoroughly under running water to remove dirt and finally washed with distilled water. Leaves of *Capparis zeylanica* were shade dried at room temperature and were powdered. The dry powder (25g) was exhaustively extracted with suitable solvent using three different soxhlet apparatus (14 extraction cycles) at different temperatures.

The extract obtained was filtered using Whatman filter paper. Then, the extracted solvent was partially distilled out at 80 ± 2 °C to get the semisolid extract.

5.2 Optimization of solvents and temperature for the preparation of different plant extracts:

Extraction solvent was optimized by extracting the *Capparis zeylanica* highest polarity solvents Methanol using soxhlet apparatus. methanolic extract was subjected to phytochemical screening. Solvent amount and extraction temperature were optimized to get the optimum condition for maximum yield. Solvent concentration was optimized by varying amounts of solvent used (50, 75, 100, 150 ml) and the temperature at which at which the extraction was to be carried out was optimized by varying temperature conditions 25, 30, 40, 50 °C of heating mantle of soxhlet apparatus. Time/cycles was optimized at 10, 12, 14 and 16 extractions.

5.3 Phytochemical screening: Preliminary qualitative phytochemical screening of the methanolic extract was done by using standard phytochemical screening methods. Change in color was noted for the result.

Test for Carbohydrates: Small methanol extract volumes were dissolved and filtered in distilled water. The filtrate subsequently underwent a molish test, the test of Benedict and the test of Fehling for the presence of carbohydrate.

- i. **Molish Test:** 2 ml extract solution was carefully placed along- side the tubes in a few drops of 15 percent ethanol α -naphthol (Molish reagent) and 2 ml concentrated sulphuric acid. At the crossroads of two layers the forming of a reddish violet ring shows carbs.
- ii. **Benedict's test:** Benedict reagent equivalent volume and test solution were combined in a test tube and heated for 5 minutes in a boiling water bath. Green/yellow/red precipitates formation reflects the sugar reduction in the test solution.
- iii. **Fehling's Test:** 1ml of Fehling's A and 1 ml of Fehling's B solution was mixed and boiled for 1 minute. The same test solution volume was added and 5-10 minutes hot bath water. Precipitates of first yellow and subsequently brick red suggest carbohydrate content.

Test for amino acids:

i. Ninhydrin Test: The extract has been processed and boiled with a 4-8 pH Ninhydrin reagent. Forming purple colour means that amino acids are present.

Test for Tannins and Phenolic compounds: A small amount of methanolic extract was dissolved in water. The ferric chloride and lead acetate tests were then performed on this solution.

- i. **Ferric Chloride Test:** 1 ml of 5 percent ferric chloride solution was added to 5 ml of extract solution. The presence of tannins is indicated by a greenish black hue.
- ii. **Lead acetate Test:** 5 mL of the extract solution was mixed with 1 mL of 10% aqueous lead acetate solution. The presence of tannins is shown by the development of a yellow colour precipitate.

Test for Saponin Glycosides:

i. Foam Test: 1 mL of methanolic extract was diluted to 20 mL with distilled water and shaken for 15 minutes in a graduated cylinder. Saponin can be detected by a 1 cm layer of foam.

Test for Alkaloids: A small portion of solvent-free methanolic extract was filtered after being agitated with a few drops of dilute hydrochloric acid.

- i. **Mayer's Test:** A few drops of Mayer's reagent were applied to 2 ml of filtrate. The presence of alkaloids is indicated by the formation of a cream-coloured precipitate.
- ii. **Wagner's Test:** A few drops of Wagner's reagent were applied to 2 ml filtrate, and a reddish-brown coloured precipitate indicated the presence of alkaloids.

iii. Hager's Test: A few drops of Hager's reagent were added to 2ml of filtrate. The presence of alkaloids is indicated by the presence of yellowish precipitate.

Test for Flavonoids:

i. Lead acetate Test: 1 ml of 10% aqueous lead acetate solution was allowed to react with 1.5ml of extract solution. The presence of tannins is shown by the formation of yellow coloured precipitate.

Test for Cardiac Glycosides:

i. Keller-Killiani Test: Glacial acetic acid, one drop 5 percent Ferric Chloride, and con. Sulphuric acid was added to 2 ml of extract. The presence of cardiac glycosides is indicated by the formation of a reddish-brown hue at the intersection of two liquid layers and the advent of a bluish green colour in the upper layer.

ii. Legal's Test: 1 mL pyridine and 1 mL sodium nitroprusside were added to the extract. The presence of cardiac glycosides is indicated by the presence of a pink to red tint.

Test for Terpenoids:

i. Salkowski Test: 5 mL of each extract was combined with 2 mL chloroform, and 3 mL of concentrated H₂SO₄ was carefully added to form a layer. The presence of terpenoids is indicated by a reddish brown colouring of the interface.

Physico-chemical characterization of extract:

i. Density: A pycnometer/specific gravity bottle was used to determine density.

ii. pH: pH metre was calibrated using pH 4, 7, and 9.2 buffers, and the pH was then calculated by dipping the electrode directly into the extract until a steady pH was reached.

iii. Determination of Total Ash: The total Ash of powder of *Capparis zeylanica* was determined by increasing 1 gm of accurately weighted crude powder in a tared silica crucible. It was incinerated in a muffle furnace at a temperature not exceeding 450°C until free from carbon, then cooled and weighed.

Z – X

$$\text{Total ash} = \frac{Y - X}{Y - X} \times 100$$

X = Weight of empty crucible

Y = Weight of crucible + Sample

Z = After complete ashing, weight of Residue + Weight of crucible

5.4 TLC: Step involve in performing TLC of three different plant extracts:

Capparis zeylanica

Pre-coated TLC plate: Silica gel- G

5.4.1 : Sample application: The capillary was immersed in the solution and the sample was applied to the capillary at a location about 2cm from one end of the plate to the thin layer plate. The plate was dried out by air.

5.4.2 : Chamber saturation: For aegeline detection, a TLC chamber was saturated with mobile phase toluene: ethyl acetate: formic acid (6: 4: 0.1 v/v) at room temperature (20 °C - 25 °C). The mobile phase was

poured into the chamber, which was then covered with a lid and left to saturate for 30 minutes.

5.4.3 :Chromatogram Development: The plate was kept in the chamber after the chamber had been saturated and the sample had been spotted on the plate. Otherwise, the spotted material would dissolve in the pool of solvent instead of undergoing chromatography, so the solvent level at the bottom of the chamber was kept below the level of spot put to the plate. The solvent was allowed to flow roughly a third of the way around the silica plate.

Visualization: Plates were removed and were examined visually and in iodine chamber and after the retention factor was calculated by following formula.

Distance travelled by solute from origin line

$$RF = \frac{\text{Distance travelled by solute from origin line}}{\text{Distance travelled by solvent from origin line}}$$

5.5 FTIR (Fourier Transform Infra Red Spectroscopy): Fourier transform infrared spectroscopy originates from the fact that this technique is appropriate to convert organic data into actual spectrum. At different wave numbers every functional group acquires one or more distinctive peak and one specific frequency range. Vibration, chemical bond movement, bending and stretching can be detected by this technique. The FTIR spectrum of sample was obtained using FTIR Spectro photo meter (Make:) 10 mg of extract was directly placed on the sample compartment (made up of selenium coated diamond) and the spectrum was obtained in the range of 4000-650 cm^{-1} at a resolution of 8 cm^{-1} for the characterization of chemical functional groups. The FTIR data gives the estimation of compound present in the extract. FTIR helps in determination of compatibility between excipients and active ingredients as presence of all the peaks at respective places indicates active ingredient's compatibility with excipients. However, minor shifting or change in intensity of peak is acceptable due to mixing with other components.

5.6 : Animals Studies 5.6.1: Selection of animals

Wistar albino rats weighing around 150-350gms were selected for the experiment. The animals were checked to confirm that they were free from any disease. The rats were collected from the animal house of Himalayan Institute of Pharmacy.

5.6.2: Maintenance of animals

The rats that were selected were brought into the laboratory 2 days before the commencement of experiment for acclimatization. They were provided with standard laboratory rodent chow diet obtained from (Kalia agro industries Ltd, Kala-Amb) and free access to water. A 12-hour day and dark cycle and room temperatures at 27°C were maintained.

The plant was identified by botanist, Noni Horticulture and forestry, Nauni Himachal Pradesh, bearing reference number 1027 and deposited in herbarium for future reference.

CONCLUSION

The present study "Evaluation of anti-diarrhoeal activity by using methanolic leaf extract of *Capparis zeylanica*." in experimental animal models was found effective in the anti-diarrhoea activity due to the presence of tannins. This study strongly implies that the methanolic leaf extract of *Capparis zeylanica* can be used to treat diarrhoea and its effects.

Further studies are required to evaluate the anti-diarrhoea activity of *Capparis zeylanica* methanolic leaf extract in other models. The isolation of the active compound, its evaluation in experimental model and study of mechanisms of action will eventually yield and develop new drugs.

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