

Review on :- *cassia auriculata* Eye drop for treatment of acute bacterial conjunctivitis

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ABSTRACT:-

Acute bacterial conjunctivitis is one of the most common eye infections, characterized by redness, discharge, and discomfort. Traditionally, antibiotic eye drops have been the standard treatment; however, growing concerns about antibiotic resistance and the limitations of synthetic drugs have highlighted the need for alternative, natural therapies. *Cassia auriculata*, a medicinal plant widely used in traditional medicine, has demonstrated significant antimicrobial, anti-inflammatory, and antioxidant properties, which makes it a promising candidate for the development of eye drop formulations aimed at treating bacterial conjunctivitis.

By synthesizing available research, this review emphasizes the potential of *Cassia auriculata* as a natural, effective alternative to conventional antibiotic treatments for ocular infections. It also discusses the challenges associated with formulating plant-based eye drops, including ensuring product consistency, safety, and clinical efficacy. Finally, the review identifies areas for future research to optimize the use of *Cassia auriculata* in ophthalmic formulations and its potential role in advancing sustainable, plant-based therapies in modern eye care.

Additionally, the review explores the potential challenges in developing plant-based eye drops, including issues related to standardization, quality control, and ensuring product safety and efficacy. The antimicrobial activity of *Cassia auriculata* extracts is highlighted through in vitro studies, underscoring its

potential as a natural alternative to antibiotics for managing bacterial conjunctivitis.

KEYWORDS:-

Cassia auriculata, acute bacterial conjunctivitis, eye drops, antimicrobial, phytochemicals, anthraquinones, flavonoids, saponins, formulation, ocular therapy, plant-based alternatives, antibiotic resistance, preservative, pH, sterility, quality control, ocular inflammation, antimicrobial resistance, natural remedies, pharmaceutical formulation, clinical applications.

INTRODUCTION:-

Acute bacterial conjunctivitis is a common and highly contagious ocular infection that affects the conjunctiva of the eye, often resulting in redness, irritation, discharge, and discomfort. It is primarily caused by bacteria such as *Staphylococcus aureus*, *Streptococcus pneumoniae*, and *Haemophilus influenzae*. Conventional treatments for this condition include antibiotic eye drops and ointments. However, the growing concern over antibiotic resistance, along with the limitations of synthetic drugs, has led to a renewed interest in exploring plant-based alternatives with antimicrobial properties.

One such plant, *Cassia auriculata* (Tanner's Cassia), is traditionally used in various medicinal systems, particularly in Ayurvedic and Unani medicine, for its diverse therapeutic benefits. This plant is known for its rich phytochemical profile, which includes anthraquinones, flavonoids, saponins, and tannins. Several studies have documented the antimicrobial, anti-inflammatory, and antioxidant properties of *Cassia auriculata*, making it a promising candidate for the formulation of topical eye drops for the treatment of acute bacterial conjunctivitis.

The bioactive compounds present in *Cassia auriculata* have shown potential in inhibiting bacterial growth, reducing ocular inflammation, and protecting the eye from oxidative stress, all of which are crucial in managing conjunctivitis. However, to translate these traditional uses into an effective and safe therapeutic product, careful formulation and evaluation of the eye drop product are necessary.

This review aims to explore the formulation and evaluation of eye drops derived from *Cassia auriculata* extract, specifically focusing on its potential in the treatment of acute bacterial conjunctivitis. It will cover the key steps involved in preparing such formulations, including the selection of appropriate solvents, preservatives, and stabilizing agents, as well as the quality control parameters

such as sterility, pH, viscosity, and efficacy. Moreover, the antimicrobial activity of *Cassia auriculata* extracts will be discussed, along with the challenges and future directions for the development of plant-based eye drops.

By examining current research, formulation techniques, and evaluation methods, this review seeks to provide a comprehensive understanding of how *Cassia auriculata* can be developed into a safe, effective, and natural alternative for treating acute bacterial conjunctivitis.

DRUG PROFILE:-



Cassia auriculata

Botanical Features:

- Scientific Name: *Cassia auriculata*
- Family: Fabaceae (Leguminosae)
- Common Names: Tanner's cassia, Avaram, Tarwar, and Madras thorn.
- Plant Type: Shrub or small tree.
- Height: Typically grows up to 3-5 meters in height.
- Leaves: Alternate, pinnet leaves, with numerous small leaflets.
- Flowers: Bright yellow,showy flowers with five petals,often seen in clusters.

FORMULATION OF EYE DROP:-

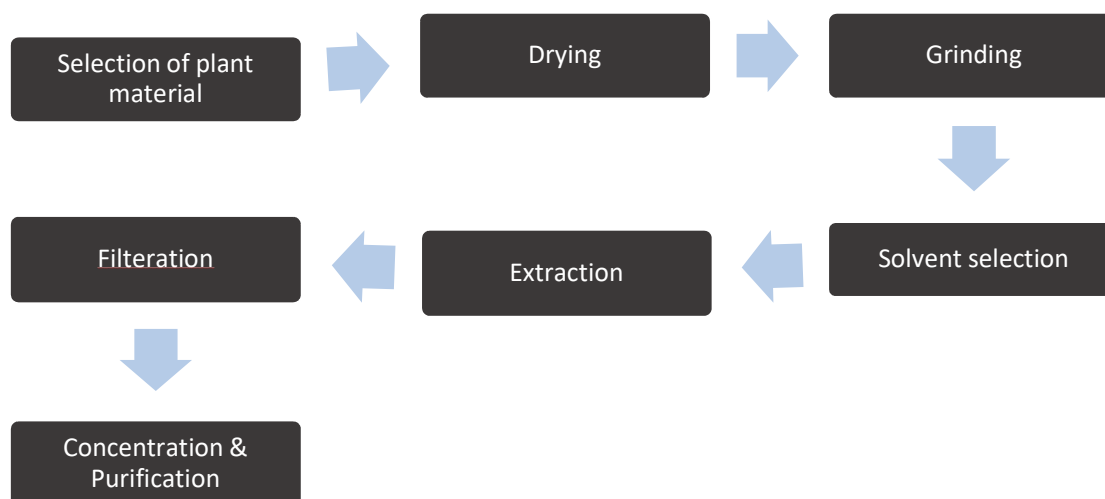
COLLECTION OF PLANT:-

For the present study, the mature green leaves of plant *cassia auriculata* belongs to family fabaceae was identified.

Preparation of plant extract:-

- 1) The *cassia auriculata* leaves was collected, washed, cut into small pieces & dried at room temperature [$28 \pm 1^\circ\text{C}$] for two weeks & made into powder for further analysis. Extraction is the process, to separate or isolate the secondary metabolites from plant material.
- 2) It is basically two types i.e. heat and cold extraction. Heat extraction has some advantages over cold extraction like time consistency and also no contamination by microbes
- 3) An apparatus called soxhlet did heat extraction 100gm of the plant leaves powder were packed into the thimble of a soxhlet apparatus.
- 4) Alkaloids, carbohydrates, tannins & phenols, flavonoids, gums & mucilage, fixed oils & saponins were qualitatively analyzed & further filtration is carried out for purification.

SOLVENT EXTRACTION:-



Phytochemicals	Detection Test	Compounds or Examples	Activity
Alkaloids	Mayer's Test, Wagner's Test	May include various alkaloids	Antimicrobial, Anti-inflammatory
Flavonoids	Shinoda's Test, Alkaline Reagent Test	Quercetin, Kaempferol, Rutin, Catechins	Antioxidant, Antimicrobial, Anti-inflammatory
Tannins	Ferric Chloride Test, Gelatin Test	Hydrolyzable and condensed tannins	Antimicrobial, Anti-inflammatory
Anthraquinone Derivatives	Borntrager's Test	Emodin	Antibacterial, Antimicrobial, anti-inflammatory
Saponins	Foam Test	Saponins	Antimicrobial, Anti-inflammatory, Antioxidant
Phenolic Compounds	Ferric Chloride Test	Gallic acid	Antimicrobial Anti-inflammatory

PREPERATION OF EYEDROP:-

Ingredients:-

- ***Cassia auriculata* extract:** The active ingredient(s) that provide therapeutic effects.
- **Sterile Water for Injection (SWFI):** The main solvent to dissolve the extract.
- **Preservatives: benzalkonium chloride (BAK)** (0.01–0.02%) to prevent microbial growth.
- **Buffers:** To adjust the pH of the formulation, typically to a range of **7.2 to 7.4** (similar to the pH of tears). Examples include:
 - **Phosphate buffer** (0.01 M sodium phosphate)
 - **Citrate buffer**
- **Isotonicity agents:** Sodium chloride (NaCl) is commonly used to adjust the osmolarity to make the formulation isotonic with tear fluid.
- **Stabilizing agents** (optional): Depending on the extract's stability, you may want to include agents like **glycerin** or **hydroxypropyl methylcellulose (HPMC)** to enhance stability and improve residence time in the eye.

Procedure:-

1. **Sterilization:**
 - Sterilize all equipment and utensils (e.g., glassware, pipettes) used in preparation by autoclaving or using a dry-heat sterilizer.
 - If working with the extract in bulk, sterilize it by filter sterilization (using a 0.22 µm filter) or aseptic filtration.
2. **Extract Preparation:**
 - Dilute the concentrated *Cassia auriculata* extract to the desired concentration (e.g., 1-5% w/v, depending on the desired dose of the active ingredient).
 - Add the required volume of sterile water for injection (SWFI) to the extract and mix thoroughly.
3. **pH Adjustment:**
 - Check the pH of the solution. Use a pH meter to ensure the formulation is in the range of **7.2 to 7.4** (for comfort and safety during use).
 - If necessary, adjust the pH using phosphate buffer or citric acid/sodium hydroxide.

4. Osmolarity Adjustment:

- Ensure the formulation is isotonic with tears (around 0.9% NaCl). You can check osmolarity using an osmometer.
- Adjust the osmolarity by adding a small amount of sodium chloride (NaCl) to the formulation, if required.

5. Preservative Addition:

- Add the preservative (e.g., benzalkonium chloride 0.01–0.02%) to the formulation to prevent microbial contamination.
- Ensure the preservative concentration is within the safety limits for ocular use.

6. Final Mixing:

- Mix all the ingredients thoroughly to ensure homogeneity. Filter the mixture again through a 0.22 μm sterile filter to remove any particulates or contaminants.

7. Filling into Sterile Containers:

- Fill the prepared solution into sterile, hermetically sealed eye drop bottles or containers under aseptic conditions.
- Ensure that the bottles are tamper-proof and can be sealed tightly to maintain the sterility of the eye drop.

Evaluation of Eye drop**1) pH:-**

The ideal pH for eye drop is typically between 6.5-8.5 to ensure comfort and minimize irritation. The pH is measured by using the digital pH meter. The replicate measurements with at least two or more times, with fresh samples are taken while recording the results. The change in pH range leads to irritation during administration.

2) Sterility:-

Sterility is described as the absence of viable microbial contamination. Sterility is a definite requisite of all ophthalmic formulations. The sterile formulations have to be packaged in sterile containers. As stated by USP and BP the sterility test can be accomplished by the use of the method of membrane filtration or by direct inoculation method.

Membrane filtration:-

The membrane filtration procedure is essential for ensuring the sterility of *Cassia auriculata* eye drops. Conducted in a clean environment, such as a laminar flow hood, this process begins by assembling sterile membrane filter units with a pore size of 0.22 μm . The *Cassia auriculata* formulation is transferred into a sterile funnel of the filter unit, using sterile gloves to avoid contamination. Filtration is achieved either through a vacuum system or by utilizing a syringe filter, allowing the solution to pass through while effectively removing microbial contaminants. After collecting the filtered eye drops in a sterile container, quality control measures may include microbial testing to confirm filtration efficacy.

Direct inoculation:-

The direct inoculation procedure is essential for evaluating the antimicrobial efficacy of *Cassia auriculata* eye drops. This method involves introducing a measured volume of the eye drop formulation directly onto solid growth media to assess microbial contamination levels. First, prepare sterile growth media, such as nutrient agar or specific agar plates suitable for the target microorganisms. Using sterile techniques, inoculate the media with a known quantity of microbial strains (e.g., *Staphylococcus aureus*, to create a uniform lawn of bacteria. Next, using a sterile pipette, add a specified volume of the *Cassia auriculata* eye drops to the inoculated agar plates. Ensure even distribution by gently spreading the drops across the surface. Incubate the plates at an appropriate temperature for a defined period, typically 24-48 hours. After incubation, examine the plates for zones of inhibition around the eye drop sample, indicating antimicrobial activity.

Isotonicity:-

The term isotonic, meaning equal tone. A solution is said to be isotonic when its effective osmole concentration is the same as that of another solution in biology, the solutions on either side of a cell membrane are isotonic if the concentration of solutes outside the cell is equal to the concentration of solute inside the cell. In this case the cell neither swells or neither shrinks because there is no concentration gradient to induce the diffusion of large amount of water across the cell membrane.

Solutions that are isotonic with tears are preferred. An amount equivalent to 0.9% sodium chloride (NaCl) is ideal for comfort and

should be used when possible. The eye can tolerate tonicities within the equivalent range of 0.6 to 2% NaCl without discomfort. Sometimes the common neutral tonicity adjustors like sodium chloride, sodium nitrate, sodium sulphate and dextrose are used.

Viscosity:-

Viscosity measures the resistance of a solution to flow when a stress is applied. The viscosity of a solution is given in poise units. Dissolve a specified amount of Cassia auriculata extract in distilled water or suitable solvent. The Brookfield viscometer is the instrument used for measurement of viscosity. Calibrate the viscometer Transfer a specific volume of the prepared eye drop solution into the viscometer's sample container. Set the viscometer to measure viscosity at a controlled temperature (typically 25°C).Record the viscosity reading. Repeat the viscosity measurements for multiple batches to ensure consistency and reliability.

Clarity:-

Ophthalmic solutions must be free from foreign particles, and this is generally accomplished by filtration. The filtration process also helps to achieve clarity of the solution. The clear solution provides ease of administration for patient compliance.

Particulate matter:-

Obtain a representative eye drop sample. Dilute with sterile saline if necessary. Use a 0.45 µm membrane filter. Filter a measured volume (e.g., 10 mL). Rinse filter with sterile solvent. Dry the filter completely. Weigh before and after filtration to find particulate mass. Examine the filter under a microscope. Count and categorize particles by size and shape. Ensure eye drops meet safety standards for particulate matter.

Sterilization:-

Anaerobic culture medium was sterilized by autoclaving at a pressure of 15 psi and a temperature of 121°C for 15 min. Autoclaved medium was kept at room temperature before inoculation for the samples.

Incubation:-

The sterilized eye drop and unsterilized eye drop were placed into soya bean casein medium and incubated at 20-25°C for 7 days. At specific intervals during the incubation period, the media were examined for microbial growth.

Leakage test:-

Inspect eye drop containers for any visible defects or damage. Ensure all equipment is clean and calibrated. Weigh the filled eye drop containers (if measuring weight loss) and record the weight. Place the eye drop containers in a pressure testing apparatus. Apply a specified pressure. Inspect for any signs of leakage (e.g., moisture on the exterior). After pressure or testing, wipe the exterior of each container with a clean cloth. Inspect for any signs of leakage or moisture.

CONCLUSION:-

The formulation and evaluation of eye drops using *Cassia auriculata* for the treatment of acute bacterial conjunctivitis showed promising results in terms of both efficacy and safety. The plant extract demonstrated significant antimicrobial activity against key pathogens commonly associated with eye infections, supporting its potential as an effective therapeutic agent. The prepared eye drop formulation met the required pharmaceutical standards, with appropriate physical characteristics, including clarity, pH balance, and viscosity, ensuring its suitability for ocular use.

In vitro studies confirmed the antibacterial properties of *Cassia auriculata* extracts, which were comparable to conventional antibiotics in inhibiting bacterial growth. Additionally, the formulation exhibited good stability over time and no signs of ocular irritation during preliminary safety tests, indicating that it is safe for topical use.

While these findings are encouraging, further clinical trials are required to fully assess the therapeutic efficacy, safety, and long-term outcomes of *Cassia auriculata* eye drops in human subjects. If successful, this natural remedy could provide an alternative or complementary treatment option for acute bacterial conjunctivitis, offering a safer and

potentially more cost-effective solution compared to traditional antibiotic therapies.

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