

**FORMULATION AND EVALUATION OF EUCALYPTUS OIL LOADED  
IN HERBAL GEL FOR ITS ANTIBACTERIAL ACTIVITY**

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**ABSTRACT**

Originally, the majority of antibacterial compounds was derived from plants. The term "herbal medicine" describes the use of any seed or leaf oil for therapeutic purposes. In addition to conventional dosage forms, herbal medications are also made in gel form. A gel is a semisolid substance that has a gel-like consistency and is applied topically to various body parts. Formulating and evaluating an antibacterial herbal gel using local medicinal plants was the aim of the project. For the purpose to formulate gel, the oil from the chosen plants was extracted in various ratios at random. The topical formulation was created and evaluated for a uniformity drug content, physical parameters, and Spreadability (SP). As shown by the results, the MIC values for coriander and eucalyptus herbal gel against *Bacillus subtilis* and *Staphylococcus aureus* were 50% v/v and 50% v/v, respectively.

The second formulation (ECG50) displayed the highest drug content (65%), as well as the formulation's maximum stability and zone of effect. A human pathogen that can lead to infections of the skin or mucous membranes is the herpes simplex virus. Antibacterial, antimicrobial, antiviral, anti-inflammatory, and local analgesic properties are possessed by geranium, licorice, rosemary, sumac, and mellifluous plants. These herbs also have the effect of controlling the synthesis of viral proteins. The design, formulation, and assessment of the gel comprising these eucalyptus oils are the objectives of this study.

**KEYWORDS:** Antibacterial, Herbal Gel, Eucalyptus oil, Formulation, Evaluation.

## INTRODUCTIONS

The gels as semisolid systems consisting of either suspensions made up of small inorganic particles, or large organic molecules interpenetrated by a liquid. Where the gel mass consists of a network of small discrete particles, the gel is classified as a two-phase system. Single phase gels consist of organic macromolecules uniformly distributed throughout a liquid in such a manner in that no apparent boundaries between the dispersed macromolecules and the liquid. Single phase gels and jellies can be described as three-dimensional networks formed by adding macromolecules such as proteins, polysaccharides, and synthetic macro molecules to appropriate liquids. Many polymer gels exhibit reversibility between the gel state and sol, which is the fluid phase containing the dispersed or dissolved macromolecules. However, formation of some polymer gels is irreversible because their chains are covalently bonded.

The three-dimensional networks formed in two phase gels is and jellies is formed by several inorganic colloidal clays. Formation of these inorganic gels is reversible. This review focuses mainly on water-based gels and jellies. Gel structure, the basis for understanding the physical properties associated with gels, is examined first, followed by the rheology of gels. The physical properties of gels and jellies can be classified based on two groups.

- A) Transitional properties and rheological properties, yield point and rupture.
- B) Spectrophotometric and thermal techniques are used to identify gel microstructures (physical junction zones) and their related transitional properties.

For example, nuclear magnetic resonance (NMR) spectroscopy measures the structural and dynamic characteristics of the polymer just prior to aggregation and gel formation and circular dichroism (CD) spectroscopy measures the conformational changes of the polymer during network formation.

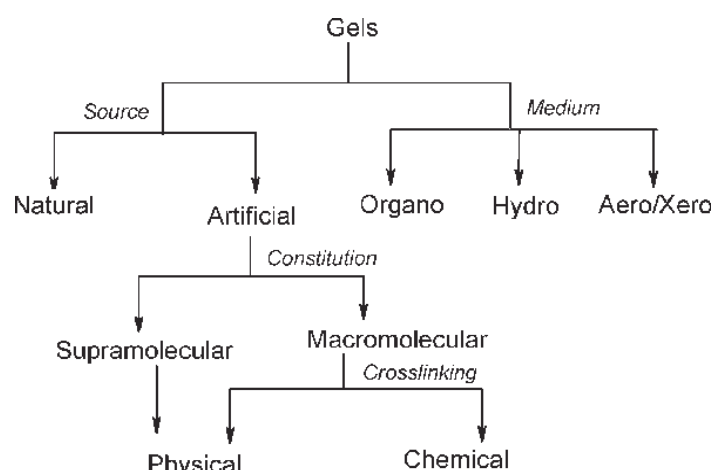
Mechanical techniques are used to determine rheological properties of gels. These techniques employ either small deformation measurements that yield viscoelastic parameters or large deformation measurements that generate complete stress strain profile, which include failure parameters.

The majority of gels are formed by the aggregation of colloidal sol particles, the solid or semisolid system so formed being interpenetrated by a liquid.

Gelation of lyophilic sols Gels formed by lyophobic sols can be divided into two group depending on the Nature of the bond between the chain of the network of gel.

Type1 are irreversible system with a three- dimensional network formed by covalent bond between the micro molecules.

Type 2 gels are held together by much weaker intermolecular bonds such as Hydrogen bond. These gels are heat reversible, a transition from the sol to gel occurring on either heating or cooling. Poly solution, for example, gel on cooling below a certain temperature referred to as the gel point. Because of their gelling properties poly are used as jellies for the application of drugs to the skin. On the application gel dried rapidly leaving a plastic film with the drug in intimate contact with the skin.



**Fig. No. 01: Classification of Gel.**

### Topical Drug Delivery System

The goal of any drug delivery system is to provide a therapeutic amount of drug to the proper site in the body to promptly achieve and then maintain the desired drug concentrations. The route of administration has a significant impact on the therapeutic outcome of a drug. Skin is one of the most readily accessible organs on human body for topical administration and is main route of topical drug delivery system. Topical delivery can be defined as the application of a drug containing formulation to the skin to directly treat cutaneous disorders (e.g.acne) or the cutaneous manifestations of a general disease (e.g. psoriasis) with the intent of containing the pharmacological or other effect of the drug to the surface of the skin or within the skin. Semi-solid formulation in all their diversity dominates the system for topical delivery, but

foams, spray, medicated powders, solutions, as well as medicated adhesive systems are also in use.

#### Advantages of Topical Drug Delivery System

1. Avoidance of first pass metabolism.
2. Convenient and easy to apply.
3. Reduction of doses as compared to oral dosage forms.
4. Improve patient compliance.
5. Provide suitability of self-medication.
6. Ability to easily terminate the medications, when needed
7. Ability to deliver drug more selectively to a specific site

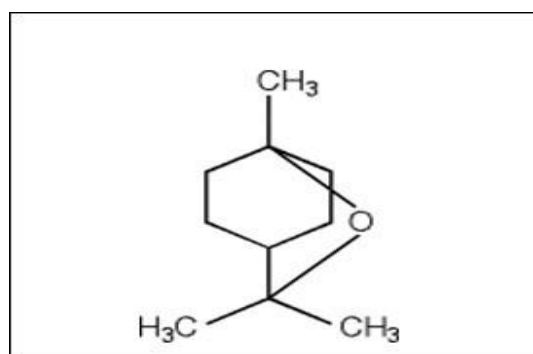
#### Disadvantages of Topical Drug Delivery System

1. Skin irritation or contact dermatitis may occur due to the drug and/or excipients.
2. Drug molecule with poor skin permeability cannot be given through topical drug delivery system.
3. Allergic reactions may develop at the application site.
4. Drug molecule with larger particle size is difficult to absorb through the skin.

#### A. Drug Profile

##### ➤ Eucalyptus

Eucalyptus is a fast-growing evergreen tree native to Australia. As an ingredient in many products, it is used to reduce symptoms of cough, flu, and congestion. It is also found in creams, gels and ointments aimed at relieving muscle and joint pain.<sup>[7]</sup> There are more than 400 different species of eucalyptus. Eucalyptus globules, also known as Blue Gum, are a major source of eucalyptus oil used worldwide.



**Fig. 1: Eucalyptol.**

Eucalyptol is an organic compound with a formula: (structure). It is a major component of Eucalyptus globules oil. Nature has been a source of healing agents for thousands of years. More recently, research has been done on medicinal plants around the world. The plants were used to treat disease years ago before the use of the latest clinical drugs. Such medicinal plants are also recognized as having medicinal properties or as a precursor to useful drugs. More than 50% of these synthetic drugs come from natural products. These natural products play an important role in drug development. With the increasing use of chemicals, many antibiotic antibiotics have developed resistance to them; which is why there is a great need to develop a new anti-agent with improved performance and comprehensive applications. Certain extracts from plants have shown antimicrobial properties and have been studied to be used as agricultural chemicals with excellent antibacterial activity. Essential oils from the Myrtaceae family show a variety of biological functions such as bacteriostatic, fungi static and anti-inflammatory effects. Eucalyptus is one of the most important lists of the Myrtaceae family, a large evergreen tree and some 700 species of shrubs. Although many plants of this genus have their origins in Australia and Tasmania, these have always been grown in many other countries, including Tunisia and used for its various properties. About 500 species of eucalyptus produce essential oils. Eucalyptus is highly valued for its wood and is a good source of protein, tannins, and dye although eucalyptus oil is a very important product. The oil can easily be wasted on its leaves. Eucalyptus oil is exported to many countries such as China, India, South Africa, Portugal, Brazil and Tasmania in terms of trade. Another name for this plant is "red forest", which was traditionally used to treat wounds, abscesses, and other ailments. The leaves can only be found on trees planted to produce oil. Eucalyptus is known as a good source of many natural substances that exhibit activities that are resistant to a few microorganisms. Various flexible phytochemicals such as isoprenoids found in eucalyptus leaves indicate a number of medicinal / antibacterial properties. Eucalyptus extracts are approved as food additives and are currently used in the manufacture of various cosmetics. Saponins, tannins, steroids and flavonoids are found in eucalyptus leaf extract. Alkaloids and flavonoids have antimicrobial activity. Traditionally, eucalyptus leaves have been used to heal wounds and fungal infections.

## AIM

The aim of the present work was to formulate the after shaving gel by using Eucalyptus oil.

## OBJECTIVES OF RESEARCH

1. To formulate the herbal gel of eucalyptus oil for topical drug delivery system.
2. To evaluate the herbal gel for topical drug delivery system.
3. To study antibacterial activity for eucalyptus oil loaded herbal gel by disc diffusion assay.
4. The objective of this research work was to formulate the Herbal gel which does not cause any side effects or adverse reactions.
5. To study use of genus eucalyptus & to formulate safe and stable anti-bacterial gel.
6. To develop topical drug delivery system and to prevent first pass metabolism.
7. To overcome the limitation of conventional oral and parenteral route of drug administration.
8. Externally, the antiseptic, slightly anesthetic, anti-bacterial, and warming properties of Eucalyptus make it a valuable resource treatment of burns, sores, ulcers, scrapes, boils, and wounds.

## METHODOLOGY

### INSTRUMENTS AND GLASSWARES

**Table No. 1: Ingredients and quantity.**

Sr. No	Instruments	Glasswares
1	Lab stirrer (mechanical stirrer)	Beaker
2	Sonicator	stirrer
3	UV-visible spectroscopy	Volumetric flask
4	Digital pH meter	Measuring cylinder
5	Brookfield Viscometer	Funnel

## MATERIAL AND METHODS

### A. Material

**Table No. 02: Name of Instruments and Glassware.**

Ingredients	Role
Ethanol	Preservative
Water	Solubilizer
Carbopol 940	Gelling agent
Eucalyptus	Antiseptic
Peppermint	Cooling sensation
Glycerin	Humectant
Rose oil	Fragrance

#### ➤ Ethanol

Ethanol is a common ingredient in many cosmetic and cosmetic products. It acts as an astringent to help cleanse the skin, as a barrier to lotions and to help ensure that the

ingredients do not break down, and it helps the hairspray to adhere to the hair. Because ethanol is effective in killing germs such as bacteria, fungi and bacteria, it is a common ingredient in many hand sanitizers.<sup>[11]</sup> The U.S. The Centers for Disease Control and Prevention (CDC) recommends the use of hand cleaners in cases where soap and water are not readily available. Practicing hand hygiene is also an important part of helping to stop the spread of COVID-19. Using hand sanitizers or hand rubs (ABHR) can help disable SARS-CoV-2, a type of coronavirus that causes COVID-

19. Ethanol is a natural product of plant fermentation and can be produced by the addition of ethylene.<sup>[12]</sup>

#### ➤ **Water**

Water is the most common ingredient in cosmetic products, often appearing first in the list of ingredients. Depending on the category of product, average beauty products can contain anywhere from 60% to 85% water.

Water has been called the 'solvent universal' in cones. In combination with emulsifiers, water can be mixed with 'thick' ingredients, such as butter and oil, to facilitate the formation of emulsions used to form creams, lubricants, and gels. Water also plays a key role in the production and production of many natural resources, sometimes combined with other solvents such as ethanol.

#### ➤ **Carbopol 940**

Carbopol is a water-soluble polymer, which is used as an emulsifying, stabilizing, stabilizing, thickening in many industries and is used as a grinding agent after shaving gels and other products. Carbopol is found in several different categories, widely used in cosmetics and cosmetics, including gels, creams and cosmetics, detergents, and air conditioners. The gelling effect is activated in two phases, the first dispersing and hydration of Carbopol, and the second "neutralizing" the solution by adding chemicals that increase the pH. Neutral agents include triethanolamine (TEA).<sup>[13]</sup>

#### ➤ **Eucalyptus**

As Eucalyptus has properties that help fight dandruff, improve blood circulation, and eliminate any inflammation or fungal attacks on the skin, it retains its integrity to promote significant hair growth. Its cool and cool effects reduce even stress, which is often cited as

one of the most common causes of hair loss. It provides the nutrients and nutrients needed for healthy hair growth.

As mentioned in the brief English history shown at the beginning of this article, this ingredient has the potential to kill germs and start wound healing for many years. When using oils, it is important to clean them thoroughly before removing any germs. Minor injuries, scratches, or abrasions can benefit from these oils and keep them away from any disease attacks as they contain anti-bacterial and anti-bacterial components.

### ➤ **Peppermint**

"Peppermint oil naturally cleanses the skin and has antibacterial and antibacterial properties. It has a cooling effect that relieves irritation and inflammation due to acne," peppermint oil can be a real skin preservative if used properly. It has a skin- lightening properties and a cool feeling.<sup>[14]</sup>

The use of peppermint oil can also serve as part of the fragrance of cosmetic and skin care products. However, research has found peppermint oil to provide a cooling feeling that can help create a cooling effect on sensitive and uncomfortable skin. Peppermint oil mainly contains menthol and menthone, followed by nutrients such as pulegone, menthofuran, and limone. It can be found in rinse-off formulations with a concentration of 3% or less.<sup>[15]</sup>

### ➤ **Glycerin**

After water and odor, glycerin is one of the most commonly reported ingredients in mammals. It is also a great ingredient in moisturizers, lotions and gels. The safe use of glycerin is growing in popularity but there are things consumers should be aware of when choosing to do this. Studies show that glycerin can have a positive effect on your skin in many ways. Glycerin, also known as glycerol, is a natural compound found in vegetable oils or animal fats. It is a clear, colorless, odorless, and sweet-smelling liquid. Glycerin is a humectant, a type of moisturizing agent that draws water to the outer part of your skin from the deeper levels of your skin and air.<sup>[16]</sup> According to a reliable research source (2016), glycerin is "the most effective humectant" available to increase water flow to the upper layer of your skin. In skin care products, glycerin is often used with an occlusive, another type of moisturizing agent, to soothe the skin.



### ➤ Rose oil

First and foremost, Pure Rose Oil is an antiseptic. This is one of the reasons why it works so well on acne-prone skin. It can nourish and disinfect the skin. It is so effective that it is also used to treat scars and other skin imperfections.<sup>[18]</sup> Natural, pure Rose Oil contains more than 50 beneficial compounds on the skin. These naturally occurring molecules cannot make it into a lab. Therefore, synthetic Rose Oil (also known as Artisan oil) has absolutely no benefits on the skin (smells just like roses). In addition, pure Rose Oil (steam distilled from the leaves of the Bulgarian Rosa Damascena) is plentiful. Richer in these nutrients are cheaper natural varieties such as Rose Hip Oil (produced from Rose buds from any rose tree) or Rose Absolute Oil (a solvent extracted from Rose oil) and other cheaper alternatives.<sup>[19]</sup>

## B. METHODS

### C. Preformulation Research

Preliminary testing is the investigation of the physical and chemical properties of a substance alone and when combined with auxiliary substances. It is the first step in the logical development of dosage forms. These studies should focus on those physicochemical structures of the new compound that may have an impact on drug performance and the development of an effective dosage form. A complete understanding of these structures may ultimately give a reason for the architectural design.

### D. How to prepare herbal extract

- The cold maceration process is used for the preparation of herbal extract.
- Collected plants of eucalyptus leaves are washed in water and dried under direct sunlight for three consecutive days after which they are mixed into a separate powder.
- 20 grams of strong powdered leaves and flower petals are immersed in 100 ml of ethanol 99.9% v / v and stored for maceration separation for approximately 3-4 days with occasional shaking.
- After maceration the extract is filtered using Whatman and 1 and the filtrate was collected and stored in the refrigerator.
- The extract is used to make the gel.

### E. Qualitative Methods

The phytochemical tests were performed by the methods given by Harborne, 1973.

- Hanch test: Two ml extract was taken in a test tube .one ml of concentrated H<sub>2</sub>SO<sub>4</sub> was added from the side walls of the test tube and the formation of a brown ring suggested the presence of carbohydrate.
- Tannin test: To 0.5ml of extract solution one ml of water and 1-2drops of ferric chloride solution was added. blue Colour was observed for Gallic tannins and green black for cate cholic tannins.
- Phenols test: in two ml of extract, a pinch of ferric chloride was added .appearance of green Colour indicates the presence of phenol.
- Protein test: Two ml of extract was taken, and one to two drops of nitric acid was added development of yellow Colour indicates the presence of proteins.
- Quinone test: two ml extract was taken, few drops of concentrated H<sub>2</sub>SO<sub>4</sub> were added and Appearance of red Colour indicates the presence of quinones.
- Fat test: the extract was tapped on the filter paper. appearance of oil on the filter paper .appearance of oil on the filter paper showed the presence of fat in the extract of eucalyptus.
- Triethanolamine was added drop wise to the formulations to balance the skin pH (6.8-7) and to get the gel at the desired consistency.



**Fig. No. 05: Test no 1.2.3.4.5.6.**



**Fig. No. 06: Test no 7How to prepare the gel after.**

## **I. RESULT AND DISCUSSION**

The formulation was developed with eucalyptus oil using Carbopol 934 as gelling agent. The formulation was Colorless and have characteristic odor of eucalyptus. The formulation was glossy and translucent, and had good consistency. The pH of the formulation ranged from 5.7 to 6.0, which was relevant with human skin ph. Hence it may be suitable for topical application without discomfort. Spreadability test was performed for evaluation of gel. The

viscosity of formulation increased with increase in the concentration of Carbopol content. The Spreadability of the formulation was found to be good. The increase in viscosity was observed with decrease in Spreadability and vice versa. The skin irritation test was performed to evaluate the skin irritation of the formulated gel on the skin of human volunteers. The results of these tests were shown that the formulated herbal Anti-Bacterial gel was safe to use.

Physical appearance

1. **Colour:** The Colour of the formulation was checked out against white background. No any different colour particle are shown.
2. **Odour:** The odour of the gel was checked by mixing the gel in water and taking the smell camphor us scent that is sharp and highly pungent.
3. **Consistency:** The consistency was checked by applying gel on skin. Smoothly and easily sprayed on skin.
4. **Grittiness:** The formulation was evaluated microscopically under 40 x magnifications thire is no any presence of any particulate matter or aggregates.
5. **Homogeneity:** Homogeneity was tested by visual inspection by naked eye after allowing them to set in a container they were evaluated for their appearance and presence of aggregates
6. **Clarity:** The clarity of various formulations was determined by visual inspection under white background there is no any particular matter.
7. **Skin irritancy test:** This test was performed on 10 healthy human volunteers of either sex after obtaining consent for the same. About 0.5 gm. of gel was applied to an area of about 6cm<sup>2</sup> on skin of hand covered with a gauze patch. The patch was held in contact with the skin for period of 1hr, the gauze was removed and residual test substance was scrapped, without altering the existing response or integrity of the epidermis. The skin was observed at

1 hr., 3hrs, 6hrs, and 12hrs.24hrs. 48hrs. and 72hrs.For any visible response on the skin. No any irritation can takes place.

### 1. Measurement of pH

The pH of developed gel formulations was determined using digital pH meter. 1 gm of gel was dissolved in 100 ml distilled water and kept aside for two hours. The measurement of pH of each formulation was done in triplicate and average values are calculated.

**Table No. 04: Measurement of PH.**

Sr. No	Observation time	pH observed
1	After 12 hrs.	5.6
2	After 24 hrs.	6.0

**8. Spreadability (SP)**

Spreadability (SP) of formulations was determined by an apparatus suggested by Multimer 45, which was fabricated itself in laboratory and used for slide fixed on wooded block and upper slide with one end tide to glass slide and other end tied with other end tied to weight pan. An excess of gel (2-5 gm.) was placed in between two glass slides and then 1000 gm. weight was placed on slides for 5 min to compress the sample to a uniform thickness, Weight (80 gm.) was added to pan. They show good spreadability (SP).

$$\text{Spreadability} = m \times l / t$$

Where,

m= weight tied to upper slide, l= length of the glass slide (6cm)

t= time in seconds

**Table No. 05: Spreadability.**

Test	Observation
Spreadability	Easily spreadable

**9. Viscosity**

The viscosity of prepared gel will be measured with Brookfield viscometer at a setting of 100 RPM at 25°C. viscosity is good.

**Table No. 06: Viscosity Observation.**

Sr no.	Formulation code	Viscosity
1.	F1	1675 dyne/cm
2.	F2	1695 dyne/cm
3.	F3	1633 dyne/cm
4.	F4	1722 dyne/cm
5.	F5	1755dyne/cm

**10. UV VISIBLE SPECTROSCOPY**

Sample: Eucalyptus oil gel formulation

**Stock Solution**

1) Make a 1000micro gram/mL solution. Take 1gm of drug and dissolve in 100mL of water.

- 2) Stir continuously up to small particle dissolve, if any particle seen in solution filter out the solution.
- 3) After filtration take a solution in volumetric flask placed in sonicator bath for 10 min.

#### Reference Sample

The formulation is soluble in distilled water is the reference sample.

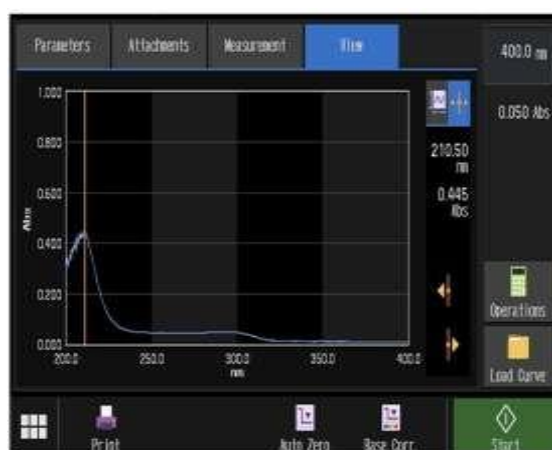
10 microgram/mL solution

Take 1mL solution into stock solution and volume make up to 10 mL

While identify UV rang of sample is Wavelength- 210.50 nm

Absorbance – 0.445.

Graph 10 microgram/mL



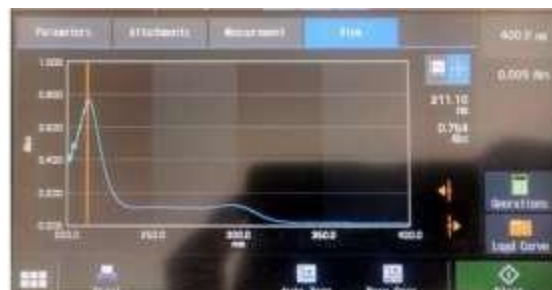
**Graph No. 02: 10 microgram/mL 20 microgram/mL solution.**

Take 2mL solution into stock solution and volume make up to 10mL

While identify UV rang of sample is Wavelength- 211.10 nm

Absorbance – 0.76

#### Graph No.03: 20 microgram/mL:



**Graph No. 03: 20 microgram/mL.**

## CONCLUSION

In the present study, antibacterial gels made from eucalyptus oil were formulated and evaluated. Eucalyptus oil, a sample substance from the eucalyptus plant, was utilized in the compositions' manufacture. A green leafy plant in the Myrtaceae family, eucalyptus is also used medicinally. The main reason behind this investigation was to formulate a stable and safe Anti-Bacterial gel. To assess the gel's performance, several evaluation tests were conducted. Such as physical appearance, colour, consistency, odour, greasiness, grittiness, homogeneity, skin irritancy test, pH determination, spreadability (SP), and viscosity. From the result of performed test we conclude that gel formulation from eucalyptus oil is safe to use.

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