

Formulation of Ointment By Using *Lawsonia Inermis* and their Pytochemical Analysis

**Ms.Janani.J¹, Ms. Kishore Jahan.A², Ms. Lavanya.P³,
Ms.AyishaSithika.S⁴**

¹*Assistant Professor, Department of Chemistry, TBAKC, Kilakarai, Ramanathapuram, Tamilnadu, India.*

^{2 3 4}*PG students, Department of Chemistry, TBAKC, Kilakarai, Ramanathapuram, Tamilnadu, India.*

Corresponding Author: Ms. JANANI

E-mail:janujega1995@gmail.com

Abstract

Our skin shields our body's interior organs from infections and pathogens. Injury that breaks the skin or other body tissues is called a wound. The process of a wound healing is organic and dynamic. Antibacterial cream (or ointment) is one of the treatments that aids in the faster healing of wounds. Medical treatments have been demonstrated to be effective in treating skin conditions like burns and wounds. A particular class of antimicrobial agent that works against bacteria is an antibiotic. Antioxidants, on the other hand, are substances that aid in shielding the skin's surface from oxidative harm brought on by free radicals and external aggressors like UV light and pollution. Due to their antibacterial, antiseptic, and anti-inflammatory characteristics, "LAWSONIA INERMIS" is a traditional medicinal plant that is frequently used for wound healing. By looking at the phytochemical study' findings, we may infer that LAWSONIA INERMIS is a highly effective medicinal and therapeutic source. In this study, we created an antibacterial cream and examined LAWSONIA INERMIS's phytochemical composition.

Keywords: *LAWSONIA INERMIS, antibacterial, anti-oxidant, wound healing ointment.*

1. Introduction

Phytochemicals are a collection of biologically active compounds that are naturally found in various parts of plants, including flowers, buds, roots, leaves, fruits, and barks. The process of phytochemical screening serves the purpose of not only identifying the constituents present in plant extracts, but also determining the most abundant and influential compounds among them. This screening process is invaluable in the search for bioactive agents that hold the potential to be utilized in the development of beneficial pharmaceuticals.

Wounds refer to injuries that result in the breaking of the skin or damage to other body tissues. These injuries can occur either externally or internally. Internal wounds typically arise from impaired circulation, neuropathy, or medical conditions.

On the other hand, external wounds result from external forces or traumatic incidents, leading to the formation of either open or closed wounds.

When a wound occurs, harmful bacteria and germs have the opportunity to invade the body. If these wounds are not properly attended to, the bacteria can rapidly reproduce, leading to various discomforts such as pain, redness, swelling, itching, and oozing.

While wounds naturally heal over time, there are various methods to expedite the healing process. One effective approach involves the application of antibacterial cream or ointment, which not only prevents infections but also accelerates the recovery of wounds.

Medicinal plants have been demonstrated to have a significant impact on the treatment of skin conditions such as cuts and burns. However, the careful selection of plants based on their specific healing properties is crucial in order to determine their true value. Traditional medicinal plants are frequently utilized to produce preparations that effectively aid in the process of wound healing, addressing a diverse range of skin ailments including *LAWSONIA INERMIS*.

LAWSONIA INERMIS, commonly known as the Henna plant, belongs to a family consisting of approximately 620 species. This plant can grow up to a height of 2-6 meters, featuring a smooth stem with leaves growing opposite each other. It holds great significance worldwide for a multitude of purposes, including the dyeing of fabrics, leather, and hair, as well as for the intricate art form known as *MEHANDI*, which involves adorning hands with Henna designs. The Henna plant contains an active ingredient known as *LAWSONE* (2-Hydroxy-1,4-naphthoquinone), which imparts the characteristic orange-red coloration. Additionally, this plant exhibits various phytochemical and pharmacological properties, further enhancing its value.

Medicinal plants are known to possess various phytochemical properties, including alkaloids, *FLAVONOIDS*, tannins, and *TERPENOIDS*, which exhibit antimicrobial and antioxidant characteristics. In an experiment conducted on *LAWSONIA INERMIS* (henna), a phytochemical analysis revealed the presence of several residues or components, such as tannins, *FLAVONOIDS*, alkaloids, *SAPONINS*, glycosides, reducing sugar, amino acids, and proteins.

2. MATERIALS AND METHODS

MATERIALS REQUIRED

- Laboratory flask
- Conical funnel
- Glass stirring rod
- Experiment tubes
- Filtration paper
- Support stand
- Washing bottle
- Bunsen burner flame

CHEMICALS REQUIRED

The ingredients for the task include

- ✓ powdered henna leaves,
- ✓ yellow soft paraffin,
- ✓ beeswax,
- ✓ regular paraffin,
- ✓ ethanol,
- ✓ Wagner's reagent,
- ✓ concentric hydrochloric acid ,
- ✓ Magnesium ribbon,
- ✓ Sodium hydroxide,
- ✓ Fehling A and B solution,
- ✓ Benedict's reagent,
- ✓ lead acetate solution,
- ✓ Chloroform,
- ✓ Concentric sulfuric acid

SAMPLE COLLECTION

Henna leaves are harvested from a garden located near the house and are left to dry for approximately two weeks in a dimly lit area. During this time, the leaves undergo a transformation, turning into a lovely yellow or brown hue. Once fully dried, they are finely ground into a powder-like consistency. This finely ground powder can then be utilized for subsequent procedures or applications.

Figure1. Dried henna leaves



Figure2. Henna powder



EXTRACTION

The extraction of henna leaves is accomplished through the process of ethanol extraction.

ETHANOL EXTRACTION

To begin the process, carefully measure 10 grams of henna powder and place it in a pristine 250ml beaker. Approximately 100ml of ethanol is then poured into the beaker. Next, securely cover the top of the beaker with aluminum foil and set it aside for 1 to 2 hours. Once some time has passed, the mixture is filtered through filter paper to separate any impurities. The resulting extract, which consists of ethanol, is utilized for the subsequent phytochemical analysis.



Figure3. Ethanol extracts

PHOTOCHEMICAL ANALYSIS

Comprehensive examination of phytochemicals was undertaken to determine the occurrence or non-occurrence of secondary metabolites, specifically ALKALOIDS, FLAVONOIDS, GLYCOSIDES, SAPONINS, TANNINS, QUINONES, CARBOHYDRATES, STEREOIDS, and LIPIDS. This analysis was conducted using the subsequent techniques.

DETECTION OF ALKALOIDS

To detect the existence of alkaloids, pour 2 ml of an ethanol-extracted sample into a test tube, followed by the addition of a few drops of Wagner's reagent. This will result in the formation of a cream-colored precipitate.

DETECTON OF TERPENOIDS

In order to detect the existence of terpenoids, combine 2 ml of the extract with 2 ml of chloroform and cautiously introduce concentrated sulfuric acid to form a distinct layer. This process will result in the formation of a reddish-brown color.

DETECTION OF SAPONNINS

In order to detect the existence of SAPONINS, a solution consisting of approximately 2 ml of extract is mixed with an equal amount of Benedict's reagent. This combination results in the formation of a precipitate that appears blue-black in color.

DETECTION OF GLYCOSIDES

In order to detect the existence of glycosides, the extract is subjected to hydrolysis using a solution of hydrochloric acid. Subsequently, the hydrolyzed extract is neutralized with sodium hydroxide and FEHLING A and B solutions are introduced. This process results in the formation of a red precipitate, which confirms the presence of glycosides.

DETECTION OF STERIODS

In order to detect the existence of steroids, a solution is prepared by mixing approximately 1ml of extract obtained from plant leaves with approximately 2ml of acetic acid. Following this, approximately 1ml of sulfuric acid is added to the mixture. The formation of a blue or green color in the solution indicates the presence of steroids.

REDUCING SUGAR

The process involves diluting the extract with water and heating it together with the FEHLING A and B solution. Consequently, a precipitate with an orange-red color is formed.

DETECTION OF TANNINS

In a test tube, take a sample extract measuring 2 ml and combine it with lead acetate solution. As a result, a white precipitate will be obtained.

DETECTION OF FLAVNONIDS

To detect the existence of FLAVONOIDS, follow these steps: Take a test tube and pour 2 ml of sample extract into it. Next, add a few drops of concentrated hydrochloric acid to the test tube. Then, place 3g of magnesium ribbon into the same test tube. If the solution turns purple in color, it indicates the presence of FLAVONOIDS.



Figure3. Phytochemical analysis

FORMULATON OF OINTMENT

The henna extract ointment was meticulously crafted using the following ingredients:

- A majority of 52% Yellow Soft Paraffin
- A small portion of 3% Beeswax
- A quarter composition of 25% Liquid Paraffin
- A significant dose of 15% Natural Extract

A precise quantity of yellow soft paraffin, yellow beeswax, and liquid paraffin were combined in a 250 ML beaker. The contents of the beaker were gently heated using a water bath until reaching a temperature of 70°C, after which the heat source was removed. Subsequently, the temperature of the mixture in the beaker was gradually reduced to 50°C. The measured quantities of natural extract were then introduced into the beaker and vigorously mixed for 5 minutes using a hand mixer. Following this, the resulting mixture was carefully poured into a clean petri dish and left to dry for a period of 2 days.

Table1. Compositions of ointment

Sl.no	Ingredients	Amount
1	Yellow soft paraffin	52%
2	Bees wax	3%
3	Liquid paraffin	25%
4	Natural extract	15%



Figure4. Water bath system of paraffin and wax

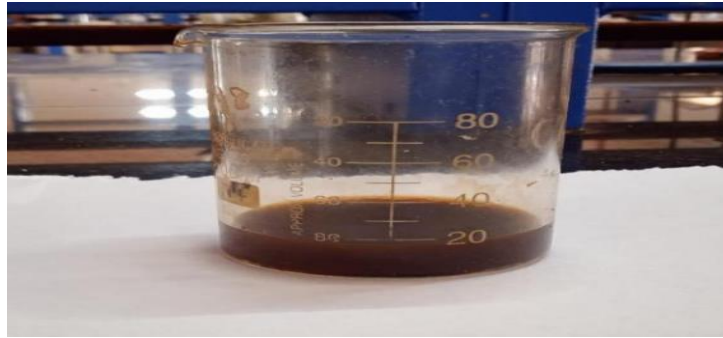


Figure5. Natural henna extract



Figure6. Mixture of paraffin and henna



Figure7. Henna Ointment

Evaluation of P H:

- In a dry beaker, 2.5 g of the formulation and 50 ml of water were added. Ointments were cooked in a beaker on a water bath at 60 to 70°C. Using a pH meter, determine the ointment's PH.

Specific gravity

After washing and drying a beaker, the weight was precisely calculated using a weighing balance. The beaker was filled with 50 ml of water, and the weight was again calculated. After being cleaned and dried, the beaker was filled with 50 grams of prepared ointment. Accordingly, the weight of the beaker containing the creams was calculated. The following formula was used to obtain the formulation's specific gravity:

Specific Gravity = weight of ointment / weight of water.

RESULTS

After analyzing the phytochemical data, it can be deduced that *LAWSONIA INERMIS* is a valuable source of medicinal and therapeutic properties. The compounds isolated from the henna plant have the potential to be developed into pharmaceutical drugs. The henna plant plays a significant role in various pharmacological activities. In today's era, there is a growing demand for non-toxic medications, making traditional medicine derived from *LAWSONIA* an appealing option due to its lack of toxicity and side effects. *LAWSONIA* exhibits inhibitory effects on a wide range of microorganisms and can be beneficial for wound healing, antiviral, antibacterial, antifungal, and anti-inflammatory purposes. Considering these promising results, further experimentation should be conducted to explore the potential of *LAWSONIA* for producing additional medicinal and industrial products.

Table2. Phytochemical analysis

S.no	Phytochemical properties	Present	Absent
1.	Alkaloids	+	
2.	Flavonoids	+	
3.	Tannins	+	
4.	Saponin	+	
5.	Terpenoids	+	
6.	Glycosides	+	
7.	Reducing sugar	+	
8.	Quinones	+	
9.	Steroids		-

The synthesis of chemical elements from *LOWSONIA* leaf extract is shown in Table 1. *ALKALOIDS*, *FLAVONOIDS*, *TANNIS*, *GLYCOSIDES*, *REDUCING SUGAR*, *QUINONES*, *SAPONINS*, *TERPENOIDS* are all present in it. Additionally, *LOWSONIA*'s steroid extraction with ethanol results in their absence.

- The ointment's pH was found to be 7.27 using a pH meter, and its specific gravity

Beaker's empty weight = 99.43 grams.

Water in the beaker weighs = 153.75 grams.

Beaker with ointment weighs = 149 grams.

With the formulas above, swapping out the measured values,

$$\text{SPECIFIC GRAVITY} = \frac{149.38}{153.75} = 0.97$$

SUMMARY AND CONCLUSION

The study conducted on the extract of *LAWSONIA INERMIS* revealed its phytochemical properties, while the ointment derived from it exhibited strong antioxidant and antibacterial properties. The initial phytochemical assessment of this species indicated that the extract of *LAWSONIA INERMIS* contains glycosides, tannins, alkaloids, flavonoids, and reducing sugar. The antibacterial activity of the extract was particularly effective against bacteria such as *E. coli*, enterococcus, streptococcus, and klebsiella, specifically in the treatment of wound infections. The antioxidant activities were evaluated at various concentrations using the DPPH method, demonstrating antioxidant properties comparable to the standard value. These findings suggest that the ethanol extract of henna ointment is a promising candidate for further research on its antioxidant activity.

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