

Formulation And Evaluation of An Adhesive Herbal Bandage Containing Ethanolic Extract of *Clerodendrum viscosum*

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ABSTRACT

The present study focuses on the formulation and evaluation of an adhesive herbal bandage containing ethanolic extract of *Clerodendrum viscosum* leaves, a plant known for its significant medicinal properties. The extract was prepared using Soxhlet extraction and incorporated into an ointment base with varying concentrations (F1, F2, F3). Physicochemical characteristics were assessed for the formulations, such as pH, homogeneity, spreadability, solubility, and stability. Biological activities including anti-inflammatory, antimicrobial, antioxidant, and drug release studies were performed. All formulations showed good physical features and stability, according to the results. Among them, F3 demonstrated stronger antibacterial efficiency against *Staphylococcus aureus* and *Escherichia coli*, as well as larger percentage inhibition in anti-inflammatory and antioxidant assays. An sticky herbal bandage was created using the refined composition. The study comes to the conclusion that herbal bandages made from *Clerodendrum viscosum* can be a viable, efficient, and all-natural substitute for wound healing applications.

KEYWORDS: *Clerodendrum viscosum*, Herbal bandage, Ethanolic extract, Wound healing, Anti-inflammatory activity, Antimicrobial activity, Antioxidant activity, Adhesive bandage, Herbal formulation, Soxhlet extraction

I. INTRODUCTION

1.1 *Clerodendrum viscosum*

The plant *Clerodendrum viscosum* is an indigenous medicinal plant, and is a member of the "Verbenaceae" family. The plant is known to have antihelminthic and antipyretic properties. *Clerodendrum infortunatum* roots and fresh leaf juice were employed as a laxative and to get rid of tumors and ascarids. *Clerodendrum infortunatum* roots contain two flavonoids that have demonstrated potent antifungal activity: cabruvin and quercetin. Flavonoids have been identified by phytochemical analyses of *Clerodendrum infortunatum* leaves.

Clerodendrum infortunatum leaf extracts in ethanol and petroleum ether exhibit wound-healing properties. The ethanol extracts *Clerodendrum infortunatum* possess potential antibacterial activity. Flavonoids are one of the important biologically active substances present in *Clerodendrum infortunatum* leaves and roots. These polyphenolic compounds shows a remarkable spectrum of biological activities.

Clerodendrum viscosum Vent. (synonym: '*Clerodendrum infortunatum* L.') is a medicinally significant perennial shrub of the "Verbenaceae" family. Taxonomically, it is classified under Kingdom Plantae, Phylum Angiospermae, Class Dicotyledons, and Order Verbenales. The genus *Clerodendrum* comprises numerous species observed in tropical and subtropical areas, including *Clerodendrum viscosum* holds significant ethnomedicinal value. Commonly known as "Hill Glory Bower". It is known as "Titabhat" in English, "Bhat" in Hindi, and "Vattaperuvalam" in Malayalam. The plant is well recognized in traditional systems of medicine for its diverse therapeutic applications and widespread availability across the Indian subcontinent. [1,2,3,4]



Figure no:1 *Clerodendrum viscosum* plant

1.2 HERBAL BANDAGE [5]

Band-Aids, also known as adhesive plaster, are the main material used to prevent wound injuries and

treat those that are acute but not dangerous. They can be administered without consulting a professional or requiring any kind of expertise. However, the effectiveness or start of action of these synthetic adhesive bandages might not be as good as anticipated. Therefore, adhesive bandages based on plant-derived herbal ingredients can be made and applied in order to provide a better alternative and effective method for treating such wounds. These plant-derived bandages may be beneficial with many properties, but synthetic variants may incorporate different medications for healing with just one.

II. MATERIALS AND METHODS

2.1 PREFORMULATION STUDIES FTIR SPECTROSCOPY^[6]

The compatibility tests were conducted at room temperature using FTIR spectroscopy to determine the interaction of drugs with the excipients used in the formulation.

2.2 FORMULATION OF HERBAL OINTMENT^[7,8,9,10,11]

- **Preparation of extracts of *Clerodendrum viscosum* leaf:**

Clerodendrum viscosum leaves were meticulously separated, cleaned, shade dried, mechanically grinded and coarsely powdered. Extraction of dried powder of the *Clerodendrum viscosum* was carried out by using Soxhlet apparatus assembly with a solvent of ethanol.

There were two extraction rounds. Weighing about 25 grams of dry powder and soaking it in the proper solvent and then extracted using 500 milliliters of ethanol in a Soxhlet extractor. After filtration, the solvent was distilled off to get the dry extract. Each extract's yield % was computed.



Figure No:2 Soxhlet apparatus

- **Preparation of ointment:**

Weighing the proper amount of hard paraffin wax and placing it in a porcelain dish submerged in water has prepared the ointment basis. The remaining components, including lanolin, cetyl alcohol, and white soft paraffin, were added once the hard paraffin wax had melted. Levigation has been used to add additional dried *Clerodendrum viscosum* leaf extract to the ointment base. To create a concentrated ointment base with a finely divided powder evenly dispersed throughout, the powder is first rubbed with a little amount of the bottom. Next, using a spatula, dilute the concentrated ointment with the remaining base. Finally, propyl and methyl parabens have been introduced.

INGREDIENTS	F1	F2	F3
<i>Clerodendrum viscosum</i> leaf extract	0.4g	0.5g	1.0g
Lanolin	0.50g	0.50g	0.50g
Cetyl Alcohol	0.30g	0.30g	0.30g
Hard Paraffin	0.30g	0.30g	0.30g
White soft paraffin	8.48g	8.48g	8.48g
Methyl paraben	0.018g	0.018g	0.018g
Propyl paraben	0.02g	0.02g	0.02g

Table no:1 Composition of *Clerodendrum viscosum* ointment

2.3 EVALUATION PARAMETERS [8,12,13]

- **Physical appearance**
Physical parameters like colour, homogeneity were evaluated
- **pH**
The pH of the produced herbal ointment was tested using a digital pH meter. The ointment solution was produced with 100ml of distilled water and placed aside for 2 hours. The pH of the solution was tested in triplicate, and a mean was calculated.
- **Spreadability**
To determine the spreadability, an excess of sample is placed in between two slides which was compressed to uniform thickness by placing a definite weight for definite time. The time required to separate the two slides was measured as spreadability. Reduced time for separating of two slides results better spreadability. Spreadability was calculated by following formula

$$S=M \times L/T$$

Where, S= Spreadability
M= Weight tide to the upper slide
L= Length of glass slide
T= Time taken to separate the slides

$$\text{Percentage inhibition} = \frac{\text{Absorbance control} - \text{Absorbance sample}}{\text{Absorbance control}} \times 100$$

- **Calculation of IC50:**
The graph was used to determine the amount of medication ($\mu\text{g/ml}$) required to denature half of the protein. The IC50 was calculated for concentration of both the sample and standard.

2.5 ANTIMICROBIAL ACTIVITY [16,17]

The "Disc diffusion method" was used to determine antibacterial activity. Determination of zone of inhibition was done by preparing agar plates aseptically. Plates were allowed to cool then *Staphylococcus aureus* and *Escherichia coli* cells were cultured and spread on the agar plates and then 5 wells of 10mm were prepared by sterile cork borer. The formulations were added using the micropipette into the well of inoculated plates and allowed to stand

- **Solubility**
Solubility in boiling water, alcohol, ether, chloroform were evaluated
- **Stability Studies**
The stability test of the *Clerodendrum viscosum* ointment was carried out for four weeks at various temperature conditions like 4°C, 25°C, and 37°C.

2.4 ANTI INFLAMMATORY ACTIVITY [14,15]

- **Inhibition of Protein Denaturation:**
The reaction mixture consisted of 0.05 ml of ointment [F1, F2, F3] with concentrations of 50, 100, 150, and 200 $\mu\text{g/ml}$ and 0.45 ml of bovine serum albumin (5% aqueous solution) (0.5 ml). 1N HCl was used to raise the pH to 6.3. After being incubated at 37°C for 20 minutes, the samples were heated to 57°C for 30 minutes. Ibuprofen (50, 100, 150, 200 $\mu\text{g/ml}$) is the common drug. After the samples cooled, 2.5 milliliters of Saline phosphate buffer (pH 6.3) was added to each tube. Absorbance was measured using spectrophotometry at 660 nm. The control for the test is bovine serum albumin. The proportion of protein denaturation inhibition was calculated using the following formula.

for ten to fifteen minutes then incubated at 37°C for 48 hours. Plates were examined for the presence of a clean area around the ointment-containing wells following the incubation times. A Vernier caliper is used to measure the zone of inhibition.

2.6 ANTIOXIDANT ACTIVITY [18]

- **DPPH Method (2,2-diphenyl-1-picryl hydrazyl radical scavenging method):**

1 ml of 0.3mM alcoholic solution of DPPH was mixed with 2.5 milliliters of the samples with different concentrations (50, 100, 150 and 200 $\mu\text{g/ml}$) of ointment of *Clerodendrum viscosum* / standard ascorbic acid (50,100,150,and 200 $\mu\text{g/ml}$). The samples were kept in the dark at ambient temperature, and after

half an hour, the absorbance was measured at 517nm. By contrasting the test and control

absorbance, the % inhibition of DPPH radical was determined using the following formula:

$$\text{Percentage inhibition} = \frac{\text{Absorbance control} - \text{Absorbance sample}}{\text{Absorbance control}} \times 100$$

- **Calculation of IC₅₀ (50% Inhibitory Concentration):**
 The graph was used to determine the amount of medication (µg/ml) required to scavenge half of the free radical. The IC₅₀ was calculated for concentration of both the sample and standard.

plates. The samples were then placed on the inoculated agar plates and incubated for about 24 hours. Following incubation, a distinct region of interrupted growth underneath and along the sides of the test material indicated antimicrobial effectiveness. The inhibitory zone is quantified and correlated with the rate of release.

2.7 RATE OF RELEASE OF MEDICAMENT [19,20]

To evaluate the medication's rate of release, samples were placed on the surface of nutrient agar contained in a Petri dish. A solution of nutrient powder was prepared and autoclaved. Petri plates were also sterilized. The Petri plates were filled with the sterile nutritional agar solution, which was thereafter allowed to solidify. In the occasion that the drug was bactericidal, the agar plates were previously seeded with a suitable organism such as Escherichia coli. The culture of broth was applied to the AATCC agar

2.8 FORMULATION OF AN ADHESIVE HERBAL BANDAGE [8,9,10,11]

- The woven fabric was cut into suitable dimension 7 × 2.5cm (length × width)
- Wound pad of 2.5 × 1.2 cm size was prepared and fixed on adhesive woven fabric.
- Then prepared herbal ointment was spread over wound pad. The backing plastic material having same size was fixed over the adhesive woven fabric.

III. RESULTS AND DISCUSSION

3.1 PREFORMULATION STUDIES

- **FTIR SPECTROSCOPY**

Functional group (Extract)	Frequency (cm ⁻¹) (Extract)	Functional group (Excipients)	Frequency (cm ⁻¹) (Excipients)	Functional group (Extract + Excipients)	Frequency (cm ⁻¹) (Extract + Excipients)
C-H	2850-2960	C-H	2850-2960	C-H	2850-2960
C-H	1400-1470	C-H	2850-2960	C-H	2850-2960
C-H	700-900	C-H	1400-1470	C-H	3200-2500

Table No:2 Drug-Excipient compatibility study by FTIR

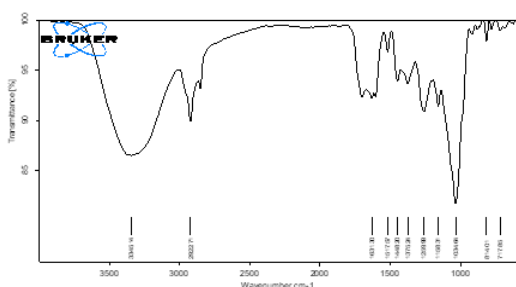


Figure no:3 FTIR spectra of ethanolic extract of *Clerodendrum viscosum* leaf

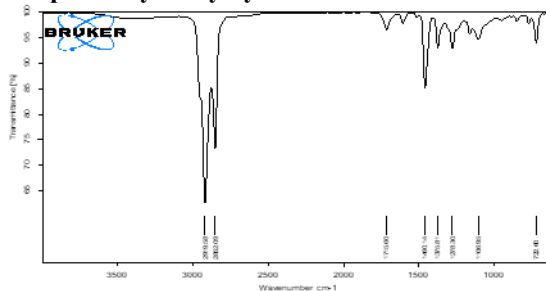


Figure no:4 FTIR spectra of excipients

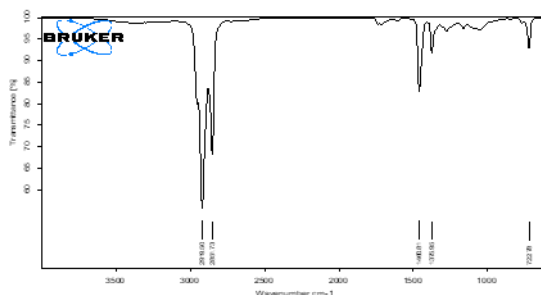


Figure no:5 FTIR spectra of ethanolic extract of *Clerodendrum viscosum* and excipients.

3.2 FORMULATION OF HERBAL OINTMENT

- preparation of extracts of *clerodendrum viscosum* leaf

The coarse powder of *Clerodendrum viscosum* leaf was subjected to ethanolic extraction using Soxhlet apparatus in 2 rounds. After extraction, the percentage yield of the extract was calculated. The yield % and further distinguishing characteristics of the extract are tabulated below.

SI.NO	Weight of powder taken (g)	Color	Consistency	Percentage yield (%w/w)
1.	40	Dark green	Semi-solid	12.87

Table no:3 Results showing the nature and percentage yield of *Clerodendrum viscosum* leaf

- preparation of ointment

Three formulations of herbal ointment (F₁, F₂, and F₃) were prepared using different concentrations of *Clerodendrum viscosum* leaf extract and then evaluated. The amount of the extract varied as 0.4 g in F₁, 0.5 g in F₂, and 1.0 g in F₃, while all other ingredients were kept constant. The formulation base consisted of lanolin (0.5 g), which acts as an emollient and enhances absorption; cetyl alcohol (0.3 g), which serves as a thickening agent; hard paraffin (0.3 g) to provide stiffness; and white soft paraffin (8.48 g) as the main ointment base. Methyl paraben (0.018 g) and propyl paraben (0.02 g) were included as preservatives. Thus, the formulations were designed to study the effect of varying drug concentration while maintaining a uniform base composition.

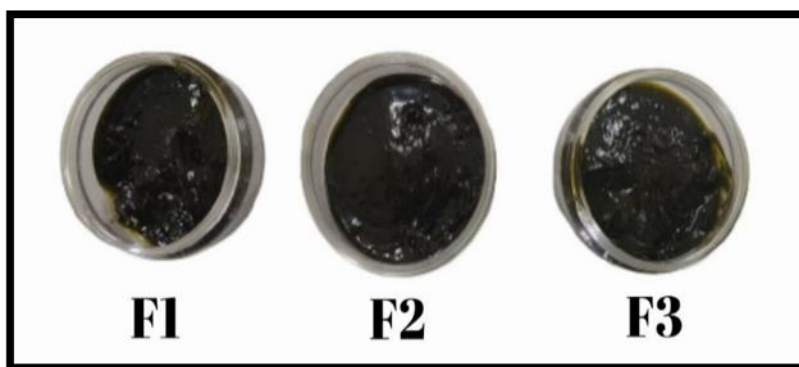


Figure no:6 Formulations (F₁, F₂, F₃)

3.3 EVALUATION PARAMETERS

Batch No.	Color	Odour	Homogeneity	pH	Spreadability (Sec)

F ₁	Dark green	Characteristic	Good	5.2	31
F ₂	Dark green	Characteristic	Good	5.1	29.2
F ₃	Dark green	Characteristic	Good	5.4	26.5

Table no:4 Color, odour, homogeneity, pH, and spreadability of prepared ointment (F₁, F₂, F₃).

• **Solubility studies**

The solubility studies of the ointment using different solvents were conducted.

SOLVENTS	F ₁	F ₂	F ₃
Water	Insoluble	Insoluble	Insoluble
Alcohol	Highly soluble	Highly soluble	Highly soluble
Ether	Highly soluble	Highly soluble	Highly soluble

Table no:5 Solubility studies of prepared ointment (F₁, F₂, F₃).

• **Stability studies**

Batch no.	4°C	25°C	37°C
F ₁	Stable	Stable	Stable
F ₂	Stable	Stable	Stable
F ₂	Stable	Stable	Stable

Table no:6 Stability studies of prepared ointment (F₁, F₂, F₃)

3.4 ANTI INFLAMMATORY ACTIVITY

• **Inhibition of protein denaturation of formulated ointment**

SI. No	Sample	Concentration (µg/ml)	Absorbance at 517nm	% Inhibition
1.	Control	-	0.820	-
2.	Standard (Ibuprofen)	50	0.450	45.12
		100	0.350	57.31
		150	0.236	71.21
		200	0.172	79.02
3.	F ₁	50	0.505	38.40
		100	0.412	49.85
		150	0.319	60.72
		200	0.255	68.90
4.	F ₂	50	0.480	41.46
		100	0.392	52.20
		150	0.310	62.20
		200	0.250	69.51
5.	F ₃	50	0.467	43.10
		100	0.366	55.40
		150	0.276	66.35
		200	0.215	73.80

Table no:7 Results showing inhibition of protein denaturation of formulated ointment.

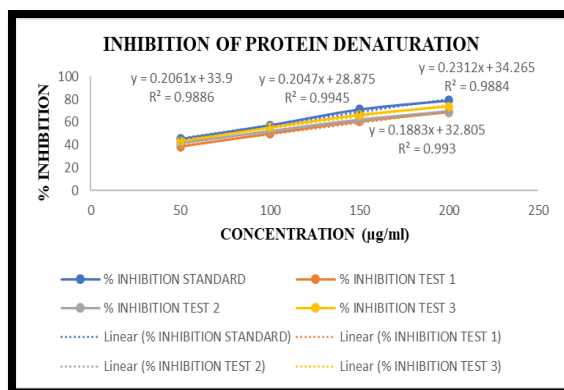


Figure no:7 IC₅₀ values of standard and formulated ointments [Inhibition of protein denaturation method]

3.5 ANTIMICROBIAL STUDY

ORGANISM	ZONE OF INHIBITION DIAMETER(mm)			
	F ₁	F ₂	F ₃	STANDARD
Staphylococcus aureus I	5	6	8	20
Escherichia Coli I	7	9	10	20

Table no:8 Antimicrobial activity of prepared ointment (F₁, F₂ and F₃)

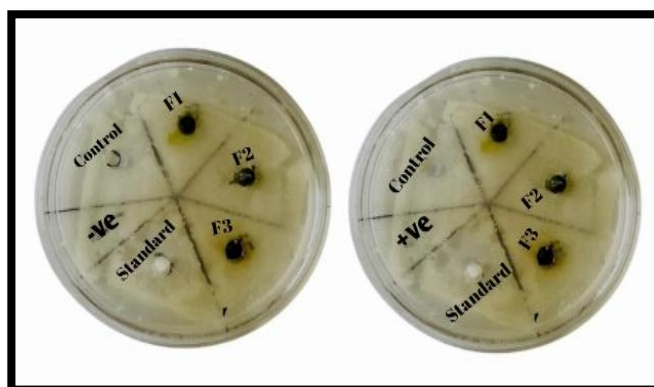


Figure no:8 Antimicrobial activity of formulations (F₁, F₂, F₃)

3.6 ANTIOXIDANT ACTIVITY

- DPPH Radical scavenging activity of formulated ointment.

SI. No	Sample	Concentration (µg/ml)	Absorbance at 517nm	% Inhibition
1.	Control	-	0.507	-
2.	Standard	50	0.304	40.03

	(Ascorbic acid)	100	0.227	55.22
		150	0.186	63.31
		200	0.110	78.30
3.	F ₁	50	0.370	27.02
		100	0.265	47.73
		150	0.205	59.56
		200	0.160	68.44
4.	F ₂	50	0.355	29.98
		100	0.250	50.69
		150	0.195	61.53
		200	0.145	71.40
5.	F ₃	50	0.340	32.94
		100	0.235	53.65
		150	0.185	63.51
		200	0.135	73.37

Table no.9 Results showing in-vitro antioxidant activity of formulated ointment.

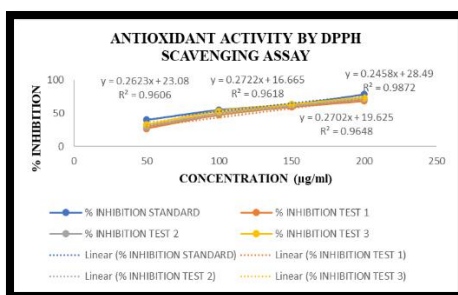


Figure no:9 IC₅₀ values of standard and formulated ointments [DPPH radical scavenging assay]

3.7 RATE OF RELEASE OF MEDICAMENT

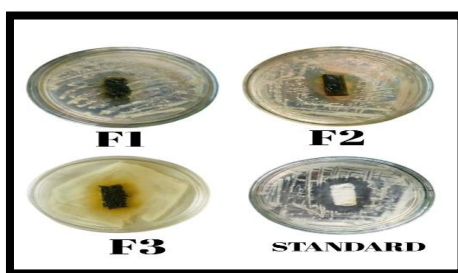


Figure no:10 Rate of release of medicament of formulations (F₁, F₂, F₃)

Organism	Zone of inhibition (mm)			
	F ₁	F ₂	F ₃	Standard
E.coli	13	20	23	25

Table no:10 Result showing rate of release of medicament of formulations (F₁, F₂, F₃)

3.8 FORMULATION OF AN ADHESIVE HERBAL BANDAGE

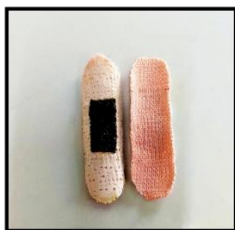


Figure no:11 Herbal adhesive bandage

IV. SUMMARY AND CONCLUSION

The study successfully demonstrated the formulation and evaluation of a herbal ointment and its application in an adhesive bandage using *Clerodendrum viscosum* leaf extract. The prepared formulations showed satisfactory physicochemical properties and stability under different conditions. Among the formulations, F3 exhibited the most significant biological activities, including anti-inflammatory, antimicrobial, and antioxidant effects, along with better drug release characteristics. These findings support the potential of *Clerodendrum viscosum* as an effective natural agent in wound management. The developed herbal adhesive bandage offers a safe, economical, and efficient alternative to conventional synthetic bandages, demonstrating its potential for upcoming pharmaceutical and clinical applications.

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