



**FORMULATION AND EVALUATION OF ANTIBACTERIAL HERBAL
OINTMENT FROM ETHANOLIC EXTRACT OF *CURCUMA LONGA*
RHIZOME AND *CATHARANTHUS ROSEUS* FLOWER**

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ABSTRACT

Medicinal plants used for the antimicrobial treatment started in ancient times. Even in all over the world modern medicine is available, the interest on plant medicines and their utilization have been increasing rapidly in recent years. Several medicinal plant extracts and its phytochemicals have shown activity against all types of microorganisms including both Gram-negative and Gram-positive bacteria. Herbal medicines have recently attracted the great interest towards their versatile application, the present work is to formulate and evaluate the ointment of *C.longa* rhizome and *C.roseus* flower extract. The extracts were prepared by using maceration method. The ointment base was prepared and formulation of ointment was done by incorporating the extract in the base by levigation method. After completion of formulation it was evaluated for its physicochemical parameters and for its stability. Thus it could become a media to use the medicinal properties of *C.longa* rhizome and *C.roseus* flower effectively and easily as a simple dosage form. The antimicrobial activity of optimized formulation was showed significant activity against the tested bacterial pathogens. The present study confirmed the antimicrobial activity of the formulations. The results are the justification for the use of the herbal medicine.

Keywords; Curcuma longa, Cathranthus roseus, Maceration, Levigation, Spredability

1. INTRODUCTION

Infectious diseases are an increasingly greater extent in recent years and this is a world-leading cause for premature death [1]. Bacterial infections have a large impact on public health [2]. All of the human organs are susceptible to bacterial infection. Some kind infections cover a small area on the top of skin. Antibacterial substances help to inhibit or kill bacterial pathogens [3, 4]. Antibiotics and other chemical antimicrobial agents play a big role against infections, but they lead to various side effects. Still, any of the ingredients in these products can cause an allergic reaction [5].

The golden fact is medicinal plants are used as antimicrobial agents from ancient times and they have effective remedies and no side effects. Use of herbal treatment is independent of any age group [6, 7]. Natural products may possess a new source of an antimicrobial agent with a possible novel mechanism of action. The secondary metabolites of plants such as alkaloids, tannins, terpenoids, and flavonoids mainly responsible for these activities. Several medicinal plant extracts and its phytochemicals have shown activity against infection including both Gram-negative and Gram-positive bacteria [1].

Ointments are semisolid preparation used for the external application. Plant drugs can also be formulated in the form of ointment. The effective ratio of active

ingredients incorporating with ointment base by trituration and after completion of the formulation, the ointment quality is assessing in terms of diffusion, irritancy, stability, and spreadability [8]. Herbal drugs are also available in the form of ointment which is semisolid preparation used topically for several purposes such as protectants, antiseptics, emollients, antipruritics, keratolytics and astringents. Numerous studies have been proved the therapeutical uses of *C. rhizome* and *C.roseus* flower [9, 10].

Curcuma longa, perennial herbaceous plant of the Zingiberaceae family. *Curcuma* rhizomes are pale yellow to brown-orange. In ancient times it was used as a perfume as well as a spice. The rhizome has bitter warm taste. In parts of Asia, turmeric water is applied as a cosmetic to lend a golden glow to the complexion. Reputed to have anti-inflammatory properties. Turmeric is consumed as a pill form for a variety of ailments, and intestinal problems. It also used as an herbal medicine for rheumatoid arthritis, chronic anterior uveitis, conjunctivitis, skin cancer, small pox, chicken pox, wound healing, urinary tract infections, and liver ailments [11]. It is used as antiseptic, expectorant, condiment etc. It is rich in antioxidants, research conducted has demonstrate uses of turmeric in the treatment of arthritis, liver diseases,

Alzheimer and depression management [12]. *Catharanthus roseus* is known as Madagascar periwinkle is a popular ornamental plant in gardens and homes across the world. All parts of the plant including leaf, root, shoot and stem contains more amount of alkaloids, which are used for therapeutic purposes against several diseases. The most important alkaloids vinblastine and vincristine exhibits anti-cancer and anti-diabetic property [13, 14].

Antibacterial activity is the most important characteristics of medical textiles, to provide adequate protection against microorganism, biological fluids and aerosols, as well as disease transmission. Antibacterial products help to prevent or kill bacteria on surfaces. Antibacterial products may have labels that say they disinfect, kill bacteria or sanitize [1, 15]. The aim of the study is to formulate the Herbal Ointment Using ethanolic extract of *C.longa* rhizome and *C.roseus* flower and Assessment of its Anti-Bacterial Activity.

2. MATERIALS AND METHODS

2.1 Collection of Plant Material

Dried *Curcuma longa* rhizomes were purchased from the local market and *Catharanthus roseus* flowers were collected from Viralimalai village area of Tiruchirappalli district.

2.2 Preparation of *C.longa* Rhizome and *C.roseus* Flower Extract

Curcuma rhizomes were washed under running water and cutted in to small pieces. They were shadow dried for another 10-15 days. The dried specimens were made a fine powder using grinder mixture and stored in air tight container. The 50g powder material of sample was mixed with 100ml of ethanol at room temperature and kept in hot perculator for about 30 minutes. After 30 minutes, the mixture was filtered and the solution alone was collected in a separate conical flask. The extract obtained from the above method was stored in glass vial for further processes [16]. The fresh flowers of *C.roseus* were collected and allowed them shade dried for another 5-10 days. The dried specimens were made a fine powder using grinder mixture and stored in air tight container. The 50g powder material of sample was mixed with 100ml of ethanol at room temperature and kept in hot perculator for about 30 minutes. After 30 minutes, the mixture was filtered and the solution alone was collected in a separate conical flask. The extract obtained from the above method was stored in glass vial for further processes [16].

2.3. Formulation of Simple Ointment

Initially ointment base was prepared by weighing accurately grated hard paraffin which was placed in evaporating dish on water bath. After melting of hard paraffin remaining ingredients were added and stirred gently to aid melting and mixing

homogeneously followed by cooling of ointment base [6].

2.4 Formulation of Herbal Ointment

Herbal ointment was prepared by mixing accurately weighed turmeric rhizome and *Caranthus roseus* flowers extract to the

ointment base by levigation method to prepare a smooth paste with 2 or 3 times its weight of base, gradually incorporating more base until to form homogeneous ointment, finally transferred in a suitable container [6].

Table 1: Formulation of Simple Ointment Base

S. No.	Ingredients	Quantity
1.	Wool fat	0.5g
2.	Cetostearyl alcohol	0.5g
3.	Hard paraffin	0.5g
4.	Yellow soft paraffin	8.5g

Table 2: Formulation of Herbal Ointment

S. No.	Ingredients	Quantity
1.	Prepared <i>Curcuma longa</i> rhizome extract	0.06g
2.	Prepared <i>Caranthus roseus</i> flowers extract	0.06g
3.	Simple Ointment base q.s.	10 g

2.5 Physico Chemical Evaluation of Herbal Ointment

Colour, Odour, Consistency, pH, Spreadability, Homogeneity, Loss of drying, Solubility, Washability, Non irritancy Test, Stability study [17].

2.6 Assesment of Antibacterial Activity

Assessment of anti – bacterial activity followed by Agar well – diffusion method [18].

3. RESULTS AND DISCUSSION

The present study was carried out to prepare herbal ointment and evaluate its physico chemical parameters and its antibacterial activity. For this the *Curcuma longa* rhizome and *C. roseus* flowers extract were prepared. A good yield of extract was obtained and there was no any harm to the chemical constituents and their activity. The levigation method was used to prepare

ointment so that uniform mixing of the herbal extract with the ointment base was occurred which was stable during the storage.

3.1 Physicochemical evaluation

The physicochemical properties were studied which shown in Table 3.1 was satisfactory results for spreadability, extrudability, washability, solubility, loss on drying and others. Also the formulation was placed for a stability study at different temperature conditions like 20⁰C, 25⁰C and 37⁰C within four weeks. There were no changes observed in spreading ability, diffusion study as well as irritant effect.

The colour of the formulated ointment illustrates that the gradual changes of *C.longa* rhizome and *C.roseus* flowers extract. Ointment from light yellow to dark yellow was visually observed. The colour is

produced due to the natural colour pigment from the *Curcuma longa* rhizome during the infusion process. It can be concluded that the darker yellow colour of the ointment is due to higher amount of *C.longa* rhizome and *C.roseus* flowers extract applied in the formulation [19]. The viscosity of the ointment can be defined as the ability of mixed materials to stick together. Frequently, the viscosity characteristic is correlated with the smoothness of the ointment. Thus, the active ingredient contributes to the viscosity value of the ointment. The more concentrate the active ingredient *C.longa* rhizome and *C.roseus* flowers extract in the ointment, the more viscous the ointment is. This study proved that the value of ointment has good viscosity [19]. The pH value of the ointment was influenced by the pH of the herb used. The pH value of the ointment relates the amount of active ingredient of *C.longa* rhizome and *C.roseus* flowers extract. As stated in **Table 3.1** it is because pH values indicates the amount of active ingredient utilized in the

ointment formulation. The pH remains the same as the original pH of the ointments [19]. The stability studies for formulated herbal ointment which proved that all physical characteristics were well maintained during the period of long term stability studies at temperature of 20°C, 25°C and 37°C. Long term stability test showed a stable homogenous appearance for 30 days storage period and no separation phase occurred. Thus indicated that the ointment condition is stable [20]. Evaluations of the prepared herbal ointment showed good results in spreadability, loss of drying, solubility, washability, non-irritancy, tube extrudability [15].

3.2 Antibacterial Activity of *Curcuma longa* Rhizome and *Catharanthus roseus* flower Extract

The result of study shows that the *Curcuma longa* rhizome, *Catharanthus roseus* extract have an antibacterial activity against *Escherichia coli*, *Staphylococcus* at the concentration of about 50µg (**Plate 1, 2**).



Figure 1a & b: Ethanollic extract of *Catharanthus roseus* flowers and *Curcuma longa* rhizome
Figure 2a & b: Ointment base and Formulation of herbal ointment

Table 3.1: Physicochemical evaluation of formulated herbal ointment

S. No.	Physicochemical Parameters	Observation
1	Colour	Yellow
2	Odour	Characteristic
3	Viscosity	Good
4	Consistency	Smooth
5	pH	5.4
6	Spreadability(seconds)	7
7	Loss on drying	30%
8	Solubility	Soluble in boiling water, miscible with alcohol, ether, chloroform
9	Washability	Good
10	Non irritancy	Non irritant
11	Stability at 20°C, 25°C and 37°C	Stable
12	Homogeneity	Good
13.	Tube extrudability	Good extrudable in nature

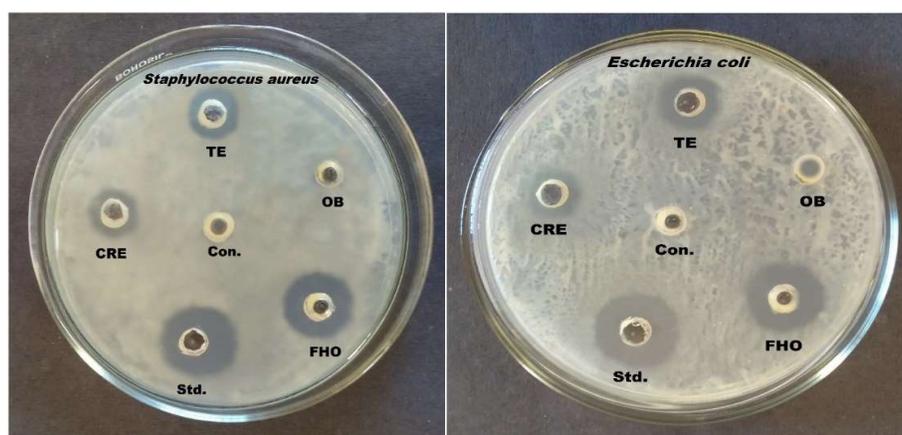


Plate 1&2: Antibacterial activity of Herbal ointment

CRE: *Catharanthus roseus* extract; TE: Turmeric extract; OB: Ointment base; FHO: Formulation of Herbal ointment

Table 3.2: Antibacterial activity of Herbal ointment

Microbial Strains	<i>C. roseus</i> extract	Turmeric extract	Ointment base	Formulation of Herbal ointment	Std. (20µl)	Control (50µl)
<i>Staphylococcus aureus</i> (mm)	2.05	4.30	0.00	8.50	10.15	0.00
<i>Escherichia coli</i> (mm)	2.30	4.50	0.00	8.75	10.25	0.00

Results were recorded as the presence or absence of inhibition zone. The inhibitory zone around the well, indicated absence of tested organism. The diameters of the zones were measured using diameter measurement. The zone of inhibition activity shows the moderate sensitivity scale. Triplicates were maintained and the average values were recorded for antimicrobial activity [15].

4. SUMMARY

The present study was done to prepare and evaluate the herbal ointment. For this, the herbal extracts were prepared to obtain a good yield of extract and there was no harm to the chemical constituents and their activity. The levigation method was used to prepare ointment so that uniform mixing of the herbal extract with the ointment base was occurred which was stable during the storage. The physicochemical properties were studied which shows satisfactory

results for spreadability, extrudability, washability, solubility, loss on drying and others. Also the formulation was placed for a stability study at different temperature conditions like 20⁰C, 25⁰C and 37⁰C within four weeks. There were no changes observed in spreading ability, diffusion study as well as irritant effect. From the above results, it shows moderate zone of inhibition against *Staphylococcus aureus* and *Escherichia coli*.

5. CONCLUSION

From the ancient time medicinal plants have various medicinal properties like antibacterial, antifungal, anti-inflammatory etc. Thus this ointment could become a media to use these medicinal properties effectively and easily as a simple dosage form. Plant which we have chosen for evaluation was turmeric rhizome and *Catharanthus roseus* flower extract. Physico chemical parameters like colour, odour, pH, Homogeneity, spreadability, washability, loss of drying, stability shows the good results and non-irritable in nature. Ointment was performed to antibacterial activity. The zone of inhibition of about 20µl was found to be 10.15 for *Staphylococcus aureus* strain and 10.25 for *Escherichia coli* strain. The reaction was found to be moderately sensitive against the bacterial infections.

CONFLICTS OF INTEREST

The authors declare that there are no conflicts of interest regarding the publication of this paper.

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