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# ANTIOXIDANT AND ANTIDERMATOPHYTIC EVALUATION OF THE TOPICAL HERBAL GEL FORMULATION

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## ABSTRACT

In the present study, the flavonoids viz. Quercetin and Kaempferol were selected and formulated in different ratios and were screened for Antioxidant andAnti-dermatophyte activities.The gel was prepared in different ratios like 1:2, 1:1 and 2:1 of QuercetinKaempferol& among them the 1:1 ratio of Quercetin: Kaempferol has a good response. The prepared various combinations of gels were evaluated for various physicochemical properties. From the results it is clearly evident that all the gel formulations showed good gelling property and homogeneity. The pH of all the formulation was in the range of 6.60 to 7.33, which lies in the normal pH range of the skin.Formulation was showed zone of clearance and percentage of inhibition at higher concentration (1000 µg) against human pathogenic dermatophyte. Maximum zone of inhibition exhibited formulation of different organisms:*Trichophytonmentagrophtye* (19 mm) and *Trichophytonrubrum*(18 mm).

**Key Words:** Quercetin, Kaempferol, Anti-dermatophyte activities

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## INTRODUCTION

Medicinal plants have been identified and used throughout human history. Plants have the ability to synthesize a wide variety of chemical compounds that are used to perform important biological functions, and to defend against attack from predators such as insects, fungi and herbivorous mammals. At least 12,000 such compounds have been isolated so far; a number estimated to be less than 10% of the total. Chemical compounds in plants mediate their effects on the human body through processes identical to those already well understood for the chemical compounds in conventional drugs; thus herbal medicines do not differ greatly from conventional drugs in terms of how they work.This enables herbal medicines to be as effective as conventional medicines, but also gives them the same potential to cause harmful side effects. The use of herbs to treat disease is almost universal among non-industrialized societies, and is often more affordable than purchasing expensive modern pharmaceuticals. The World Health Organization (WHO) estimates that 80 percent of the population of some Asian and African countries presently use herbal medicine for some aspect of primary health care. Studies in the United States and Europe have shown that their use is less common in clinical settings, but has become increasingly more in recent years as scientific evidence about the effectiveness of herbal medicine has become more widely available (1-3).Pharmacologists,

microbiologists, botanists, and natural-products chemists are combing the Earth for phytochemicals and leads that could be developed for treatment of various diseases. In fact, according to the World Health Organisation, approximately 25% of modern drugs used in the United States have been derived from plants. Among the 120 active compounds currently isolated from the higher plants and widely used in modern medicine today, 80 percent show a positive correlation between their modern therapeutic use and the traditional use of the plants from which they are derived. More than two thirds of the world's plant species - at least 35,000 of which are estimated to have medicinal value - come from the developing countries.[verification needed] At least 7,000 medical compounds in the modern pharmacopoeia are derived from plants.In many medicinal and aromatic plants (MAPs) significant variations of plants characteristics have been ascertained with varying soil traits, and the selective recovery and subsequent release in food of certain elements have been demonstrated. Great attention must be paid to choose soil and cropping strategies, to obtain satisfactory yields of high quality and best-priced products, respecting their safety and nutritional value (4). Aim is to develop a topical application using poly hydroxy natural flavonoids (Quercetin and Kaempferol) for antioxidant and Antidermatophytic activity.

## MATERIALS AND METHODS

### Antidermatophytic activities of Formulated gel

### Preparation of dermatophyteinocula (5, 6)

Dermatophyte inoculum was prepared from a 7 day old culture on Saboraud's Dextrose Agar medium. With a sterile loop, the tops of plate spores were transferred to a tube containing 5 mL of Saboraud's dextrose Broth. The tube was then incubated at 28ºC for 5 day. The turbidity of the culture suspension was adjusted with broth or sterile saline solution (0.85 - 0.9%). The density of this culture was adjusted with 0.5 McFarland standards such that final inoculum would contain 5 x $10^5$ CFU/mL.

### Well diffusion method

The *in vitro*dermatophyte activities of test extracts were determined by the well diffusion method as described by Perez *etal*. (1990). The well diffusion test was performed using Saboraud's Dextrose Agar (SDA) medium for dermatophyte. The medium was prepared and autoclaved at 15 lbs pressure (121ºC)for 5 min. The medium was cooled to 50-55º C and poured into sterile petriplates to a uniform depth of 4 mm which is equivalent to approximately 25-30 mL in a 90 mm plate. Once the medium was solidified, standardiseddermatophyte suspension was swabbed on the medium within 15 min of adjusting the density of the inoculum. The plates were undisturbed for 3 to 5 min to absorb the excess moisture. Sterilized 9 mm cork borer was used to make agar wells, 50 µL, 100 µL and 200 µL sample extracts of concentrations 250 µg, 500 µg, 1000 µg from 5 µg/µL stock solution was dispensed into each well and 100% DMSO as a control. Fluconazole (30 µg) for fungi suspended in sterile glass distilled water were used as positive control.Zone of Inhibition (ZI) were measured by 1 mm accuracy caliber and percentage of inhibition was calculated by the formula,

$$\text{Percentage of inhibition (\%)} = [\text{Zone of inhibition/}\}(\text{mm})]\times 100$$

### Antidermatophytic activity by Poison Plate method

Antidermatophytic activity of formulation was determined by food-poisoned technique (Schmitz, 1930). Formulation concentration (1000 µg/mL) was mixed with sterilized Sabouraud Dextrose Agar medium in separate flasks, and transferred into Petri plates. The media was allowed to solidify. The seven day old fungal culture disk (*Trichophytonrubrum and Trichophytonmentagrophytes*) of 9 mm diameter was taken and inoculated to the center of Petri plates containing formulated gel in aseptic condition. SDA medium, without formulated gel served as control. All plates were incubated at 28 ± 2°C and radial growth of colony was measured after seven days of incubation. Each test was performed in triplicate.

## RESULTS AND DISCUSSION

### Antidermatophytic activity of Formulated Gel

### Well diffusion method

Formulated gel showed various ranges of antidermatophytic activity at concentrations of 250 µg/mL, 500 µg/mL and 1000 µg/mL.Antidermatophytic activity of formulation revealed that the zone of inhibition (percentage of inhibition) against dermatophyte ranges between 10.00 mm (11.11%) and 19 mm (21.11%) (Table-1). Formulation was showed zone of clearance and percentage of inhibition at higher concentration (1000 µg) against human pathogenic dermatophyte. Maximum zone of inhibition exhibited formulation of different organisms:*Trichophytonmentagrophtye* (19 mm) and *Trichophytonrubrum*(18 mm).

**Table-1MIC of formulated gel and standard antibiotics Griseofulvin against human pathogenic dermatophytes**

| Concentration (µg/mL) | MIC (µg/mL) | |
|---|---|---|
| | Formulated Gel | Griseofulvin |
| *Trichophytonmentagrophtye* | 500 | 31.51 |
| *Trichophytonrubrum* | 500 | 31.51 |

### Food poison method

At highest concentration (1000 µg/mL) of formulated gel were used to test arrest of mycelia growth on solidified agar medium. The results (Fig-1, 2) were compared with control without drugs, after 7 day incubation mycelia growth was occurred in control, whereas treatment was arrested.

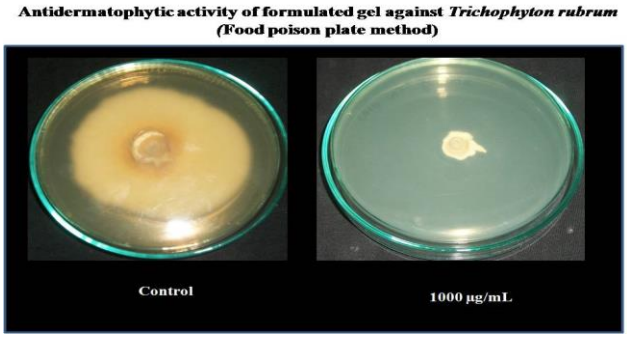


**Fig-1Antidermatophytic activity of formulated gel against *Trichophytonmentagrophtye*- Food poison plate method**

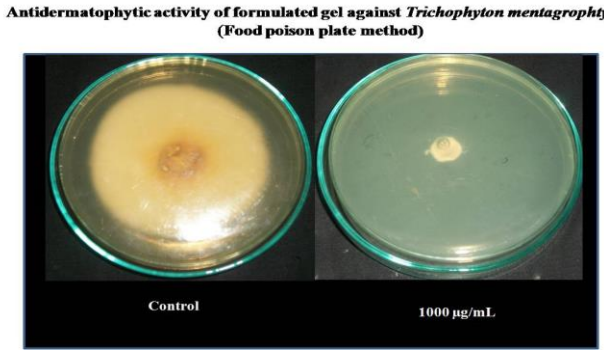


**Fig-2Antidermatophytic activity of formulated gel against *Trichophytonmentagrophtye*- Food poison plate method**

## CONCLUSION

Natural remedies are more acceptable in the belief that they are safer with fewer side effects than the synthetic ones. Herbal formulations have growing demand in the world market. Herbal medicines to be as effective as conventional medicines, but also gives them the same potential to cause harmful side effects. The use of herbs to treat disease is almost universal among non-industrialized societies, and is often more affordable than purchasing expensive modern pharmaceuticals. The World Health Organization (WHO) estimates that 80 percent of the population of some Asian and African countries presently uses herbal medicine for some aspect of primary health care.In the present study, the flavonoids viz. Quercetin and Kaempferol were selected and formulated in different ratios and were screened for Antioxidant andAnti-dermatophyte activities.Formulated gel showed various ranges of antidermatophytic activity at concentrations of 250 µg/mL, 500 µg/mL and 1000 µg/mL.Formulation was showed zone of clearance and percentage of inhibition at higher concentration (1000 µg) against human pathogenic dermatophyte. Maximum zone of inhibition exhibited formulation of different organisms:*Trichophytonmentagrophtye* (19 mm) and *Trichophytonrubrum*(18 mm).

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