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ORIGINAL RESEARCH



# Formulation and Assessment of a *Ricinus communis* Leaf Extract-Based Shampoo for the Treatment of *Tinea capitis*

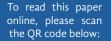
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# ABSTRACT

Tinea capitis, a dermatophytic fungal infection affecting the scalp, represents a significant global public health concern, with particular prominence in regions characterized by limited economic resources. Unlike antibacterial medications, the options for antimicrobial treatments targeting fungal infections are relatively scarce. Furthermore, several of the existing antifungal compounds have been shown to be hazardous, chemically unstable and have poor pharmacologic properties. Therefore, it is imperative to explore alternative therapeutic avenues, particularly those derived from traditional medicinal practices and herbal compounds that have demonstrated antifungal efficacy in laboratory settings. This study aimed to formulate and evaluate a shampoo containing Ricinus communis leaf extract as an adjunctive therapy for Tinea capitis using a laboratory-based experimental design. Ricinus communis leaves were harvested from Kabarak University botanical gardens, dried and ground into coarse powder, followed by Soxhlet extraction with methanol. The extract was concentrated to dryness and used in formulation of a shampoo. The shampoo was evaluated for color intensity, clarity, fluidity, ability to produce foam, homogeneity and odor. The in vitro antifungal activity on fungal isolate of Microsporum canis species that cause Tinea capitis was performed to demonstrate the inhibitory effects of the prepared shampoo and the methanolic extract at 100%, 50% and 25% concentrations. Ketoconazole 2% shampoo was used as a positive control. The quantitative bioassay was performed using disk diffusion method. The results showed that the methanol extract and the formulated shampoo have inhibitory effect against *Microsporum canis*. There was no statistical difference in zone of inhibition caused by the formulated shampoo with that caused by 2% ketoconazole shampoo (P-value = 0.59957; P-value > 0.05). Therefore, the prepared shampoo demonstrated remarkable antifungal activity against the Microsporum canis sample that is comparable to 2% ketoconazole shampoo highlighting its significance as an adjuvant therapeutic alternative for the treatment of *Tinea capitis*.

Keywords: extract, formulation, Ricinus communis, shampoo, Tinea capitis.

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## INTRODUCTION

Fungi are heterotrophic eukaryotic organisms comprising many species, some of which are pathogenic. Fungal infections can be superficial, subcutaneous, or systemic depending on which area of the body is afflicted (Silva-Rocha, et. al., 2017). Tinea infections are caused by dermatophytes (Zhan & Liu, 2021). Superficial *Tinea capitis* affects the scalp and hair, *Tinea barbae* affects the beard, *Tinea cruris* affects the pubic area, *Tinea pedis* affects the feet, *Tinea corporis* affects the skin, with *Tinea unguium* the nail beds, and *Tinea manuun* the hands (Shemer, & Babaev, 2018). The primary etiological pathogens are zoophilic *Microsporum canis*, anthropophilic *Trichophyton violaceum* and anthropophilic *Trichophyton tonsurans* (Zhan & Liu, 2017).

*Tinea capitis* is prevalent in preadolescent children, notably those between 3 to 7 years of age (Leung, et. al., 2020). In many parts of the world, *Microsporum canis* is a common cause of *Tinea capitis* (Chen & Yu, 2023). Although the zoophilic dermatophytes remain to be the most predominant cause of *Tinea capitis*, there was a surge in infections caused by the anthropophilic fungus *Trichophyton tonsurans* and *Trichophyton rubrum*, while there was also a tendency towards a decline in the frequency of zoophilic pathogens (Zhan & Liu, 2017). *Tinea capitis* caused by specific species exhibit variability depending on socio-economic conditions in a particular area, for example, the predominant etiological agent was identified as *Trichophyton tonsurans* in an evaluation study of *Tinea capitis* prevalence in school-going children in Mathare, Kenya. Infections with *Trichophyton spp. (61.3%)*, *Microsporum spp.* (13.3%) and *Epidermophyton spp.* (7.3%) were discovered in 81.3% (122/150) of the children who took part in the study (Moto et al., 2015).

*Tinea capitis* has been successfully treated with systemic antifungal therapy in the recent past. Oral antifungal medications such are griseofulvin, terbinafine, itraconazole, and fluconazole are also prescribed. Antifungal topical medication can be used to decrease spore transmission and as a complement to systemic antifungal medication. Locally applied therapies do not permeate the core of the hair shaft; hence integrating systemic and topical antifungal medications may help to enhance cure rates (Leung et al., 2020). Recent data reveal that novel oral antifungals including terbinafine and new azoles are efficacious, safer, and are applied for shorter length of time to eliminate *Tinea capitis*. Topical adjuvant treatments for *Tinea capitis* include 2% ketoconazole shampoo and 1% selenium sulfide (Kumar, et al., 2014).

Although there is an increasing global burden of *Tinea capitis* infections due to rising prevalence and occurrences, particularly in poor socioeconomic sectors of the population, comparatively few antimicrobial compounds are available to treat fungus infections (Zhan, Lain& Liu, 2021). Furthermore, several of the existing antifungal compounds have been shown to be hazardous, chemically unstable, and have poor pharmacologic properties. The prescription of first-line therapeutic medicines for *Tinea capitis*, particularly griseofulvin, has been shown to pose considerable treatment failure, high cost, and long duration of action (Alkeswani, et. al., 2019). As a result, adjuvant therapy for *Tinea capitis* treatment is required, particularly to combat the disease's rising emergence and documented cases in known endemic areas with low socio-economic living conditions. In an effort to stop the spread of this infection, sporicidal shampoos such as selenium sulfide which can help remove adhering scales and speed the eradication of viable spores from the scalp have been developed (Pomeranz & Sabnis, 2002). Although shampoos are easier to formulate, the products available in the market have some drawbacks. Hair browning occurred in some individuals after application of 2% ketoconazole shampoo, an adjuvant remedy for *Tinea capitis* (Kubicki, et. al., 2019).

The growth of multidrug-resistant fungi, paired with a paucity of antifungals has necessitated the development of novel antifungal agents. Therapeutic exploration of alternatives is vital, particularly in medicinal herbs and extracts with proven antifungal properties (Abad et al., 2006; Lopes and Salgueiro, 2017). According to a study (Al Badi and Khan, 2014; Midddha, 2015), therapy should aim to reduce or eliminate antifungal medication toxicity by providing safe, effective, and cost effective formulations that are relatively non - toxic and suitable with all skin preparations, including natural products. Toxicity profiles of preparations from herbs have been largely acknowledged as minimal. They are also hypoallergenic and hence are safe to use. Their compatibility with various types of skin serves as an added advantage (Bijauliya, et al., 2017). Herbal preparations can therefore be used as alternative to conventional therapy for Tinea infections (Lopes, et. al. 2017).

*Ricinus communis,* commonly known as Castor oil plant is a herb in the Euphorbiaceae (spurge) family (Xu & Deng, 2017). It originates from Africa and has thrived far across most world climates. The seed contains ricin and ricinolein whereas the leaf contains fixed and volatile oils. The hydrocarbon terpenes, sesquiterpenes, and polyterpenes found in the volatile oils are phytochemicals which may be used for both medicinal and cosmetic purposes (Yusuf et al., 2015). Because it is obtained from a natural source, methanolic extract of *Ricinus communis* leaves is skin-compatible and essentially a safe candidate to use in formulation of an antifungal shampoo (Bijauliya, et al., 2017). Due to limited antifungal medications available to combat the increasing prevalence and occurrence of *Tinea* infections (Lopes and Salgueiro, 2017), this study aimed to develop a safe and efficacious herbal based shampoo for *Tinea capitis* treatment. The shampoo was formulated and evaluated as a therapeutic alternative to conventional topical therapy for suppressing *Tinea capitis* spore transmission among exposed person.

# **METHODOLOGY**

#### **Research Design**

This study used a laboratory-based experimental approach to formulate and evaluate a shampoo containing *Ricinus communis* leaf extract. *Ricinus communis* leaves were harvested manually from Kabarak University botanical garden, dried and ground into coarse powder, followed by Soxhlet extraction with methanol. The extract was concentrated to dryness and used in formulation of a shampoo. The shampoo was evaluated for color intensity, clarity, pH, fluidity, ability to produce foam, homogeneity and odor. The in vitro antifungal activity on fungal isolate of *Microsporum canis* species that cause *Tinea capitis* was performed to demonstrate the inhibitory effects of the prepared shampoo and the methanolic extract at 100%, 50% and 25% concentrations. Ketoconazole 2% shampoo was used as a positive control. The quantitative bioassay was performed using disk diffusion method.

#### **Ethical Considerations**

Ethics review was sought from Kabarak University Research Ethics Committee (KUREC-090722). Permission to collect data was obtained from the National Commission for Science, Technology and Innovation (NACOSTI)(Research license no. 182762). All materials used in the formulation and evaluation of the shampoo were of analytical grade standard. *Tinea capitis* inoculum of *Microsporum canis* species was obtained from Kenya Medical Research Institute (KEMRI) in Nairobi, Kenya. The innoculum of *Microsporum canis* was transported in a sealed sterile container under cold chain conditions and maintained in a Biological Safety Cabinet (BSC) throughout the experiment. Appropriate gowning of personal protective equipment was observed to minimize contamination.

#### **Collection of Ricinus communis Leaves**

*Ricinus communis* leaves were harvested and collected from the botanical garden at Kabarak University. Identification of the leaves was performed by Kabarak University botanist. The botanical garden is well maintained by skilled personnel and with proper plant watering, soil treatment and plant health inspection.

#### Preparation of Ricinus communis Methanol Leaf Extract

*Ricinus communis* freshly cut leaves were cleaned, dried and ground into coarse powder using a motor grinding machine. Soxhlet extraction with methanol was performed on the dried ground leaf powder in a thimble at 65°C-70°C for 8 hours. The methanolic extract was concentrated using a rotary vacuum evaporator until all the solvent used was evaporated. The *Ricinus communis* extract was transferred into a universal beaker and the net weight was measured using an analytical balance. The percent yield of the extract was computed as weight of the extract per weight of the crude powder. The extracts were maintained in a refrigerator to avoid degradation.

#### Formulation of Ricinus communis Methanol Leaf Extract Shampoo

*Ricinus communis* leaf extract was mixed with appropriate quantities of established shampoo ingredients presented in Table 1 (Mithal & Saha, 2000; Sharma et al., 2018) to obtain a shampoo formular. Measurements of each quantity of ingredients used was attuned appropriately in order to get the desired acceptance ranges and physicochemical qualities of a shampoo. The pH of the shampoo was adjusted to human skin pH range by adding sufficient quantity of buffer. Physical properties and in-vitro antifungal activity of the formulated shampoo were tested.

| Table 1:   |  |
|--|--|
| Ingredients for formulation of Ricinus communis leaf extract shampoo |  |

| Ingredients        |  |  |  |
|--------------------|--|--|--|
| Primary surfactant | <ul> <li>pH adjusting agent</li> </ul> |  |  |
| Foam booster       | Preservative                           |  |  |
| • Humectant        | Antioxidant                            |  |  |
| Chelating agent    | Perfuming agent                        |  |  |
| Moisturizing agent | Active ingredient                      |  |  |

#### **Physical Appearance**

The *Ricinus communis* shampoo was evaluated for color, transparency and homogeneity by visual inspection.

#### pH Determination

A sample of the shampoo formulation was diluted to a concentration of 10% v/v in a beaker. The pH was measured by a digital microprocessor pH meter at  $25^{\circ}$ C (Sawant et al., 2020).

#### **Rheological Evaluation**

Viscosity of the formulated shampoo was determined using a NDJ SNB Rotary Digital Viscosity Meter, at 25°C and 30 rpm on spindle number 2 and torque 21%.

#### Tinea capitis Culture

Sabouraud Dextrose Agar (SDA) was tested for viability and sterility, then prepared for use. In 100ml of distilled water, 65 grams of SDA medium was suspended. Chloramphenicol was added to the medium. Gentle heat was applied while stirring and then boiled until it completely dissolved. After autoclaving for 15 minutes at 121°C, the mixture was cooled to 45°C and poured into petri dishes. To obtain colonies, *Tinea capitis* inoculums of *Microsporum canis* species were inoculated into the medium with a sterile inoculating loop. The plates were incubated at 25°C in an inverted orientation, and then incubated for 7 days.

#### Maintenance of the Fungal Cultures

*Tinea capitis* inoculum of *Microsporum canis* species was obtained from KEMRI in Nairobi, Kenya. The inoculum was transported in a sealed sterile container under cold chain conditions and maintained in a laminar flow BSC at 25°C throughout the experiment. *Tinea capitis* isolates of *Microsporum canis* species were sub-cultured in SDA and incubated at 25°C. Tests involving handling of *Tinea capitis* microorganisms were performed aseptically under BSC. Appropriate protective gowning of personnel was observed throughout the experiment.

#### Preparation of Disk for Antifungal Activity

Using sterile Whatmann No. 1 filter paper, eighteen 6mm diameter disks were prepared and soaked overnight for 18 hours in test samples of the formulated shampoo, shampoo base, three serial concentrations of *Ricinus communis* leaf extract (100, 50, 25% w/v) and 2% ketoconazole shampoo.

#### Antifungal Assay of the Shampoo

The antifungal profile of formulated shampoo was determined using the disc diffusion method against *Microsporum canis* species of *Tinea capitis*.

#### **Disk Diffusion Method**

Petri plates were filled with 20 ml of SDA and left for 30 minutes to dry in order to solidify. The *Tinea capitis* inoculum containing *Microsporum canis* species was sub-cultured into petri-dishes containing SDA. With sterile forceps on the plate, different disks containing the required test samples were placed right on the petri dish and carefully pushed against the Agar surface to ensure contact. This was performed according to corresponding labels of the test solutions marked on the surface of the petri-dishes. Ketoconazole 2% shampoo and the product formulation base were used as positive and negative controls, respectively. For 30 minutes, the plates were kept in Biological Safety Cabinet to ensure appropriate diffusion of the extract before 72 hours of incubation at 25°C. Zones of inhibition

were measured in millimeters and recorded in equal angles to one other against the corresponding concentration. In millimeters, the mean of triplicate test results was determined.

#### Data Analysis

Tables, line charts and bar graphs with mean of inhibition zones were used to present the results. The results were entered into an excel spreadsheet and evaluated using statistical One Way Analysis of Variance (ANOVA) of data to determine the significant differences among average means.

# RESULTS

#### **Methanol Leaf Extracts Preparation**

The total yield of *Ricinus communis* methanol leaf extract was 9.51 grams as shown in Table 2.

#### Table 2:

Yield of Ricinus communis methanol leaf extract

| Weight of dry powdered leaves before extraction | 16.68 grams |
|---|-------------|
| Weight of extract obtained                      | 9.51 grams  |
| Percentage yield                                | 57.01%      |

# Evaluation Parameters for the Formulated *Ricinus communis leaf extract-based* Shampoo

The results of physical properties and the in-vitro antifungal evaluation of the formulated Ricinus communis shampoo are presented in Tables 3 and 4 and Figure 1.

#### Physical appearance

A light green opaque liquid with a pleasant odor.

#### pН

Slightly acidic, pH 5.63 at 25°C

*Rheological evaluation* Pseudoplastic, viscosity 282 cP at 25°C

#### Foaming ability

It foams easily on agitation. Foam volume after 1 minute of agitation was 140 ml.

#### Table 3:

Physicochemical parameters of Ricinus communis shampoo

| Parameter              | Result   |
|------------------------|--|
| Physical appearance    | Light green<br>Homogeneous<br>Pleasant smell<br>Good foaming |
| рН                     | 5.63   |
| Rheological evaluation | Pseudoplastic<br>Viscosity, 282 cP                           |
| Foam volume            | Average 140 ml   |

#### Zones of inhibition (mm) of test samples on Microsporum canis species

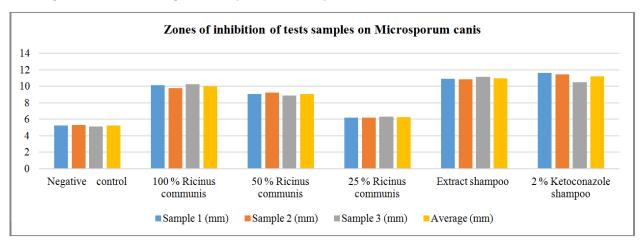
The formulated *Ricinus communis* methanol leaf extract antifungal shampoo showed significant antifungal activity against *Microsporum canis* species of *Tinea capitis*; an average zone of inhibition of 11.01 mm was observed. Zones of inhibition caused by the formulated *Ricinus communis* shampoo were higher than inhibition caused by 100% *Ricinus communis* methanol leaf extracts (10.05 mm). The 100% leaf extracts showed greater inhibition on *Microsporum canis* compared to 50% extracts (9.07 mm). The 25% extracts showed minimal inhibition (6.23 mm). The blank shampoo formulation used as negative control displayed no antifungal inhibitory effect (5.23 mm) while marketed 2% Ketoconazole shampoo showed the greatest inhibitory effect (11.21 mm)

#### Table 4:

#### Zones of inhibition (mm) of test samples on Microsporum canis species

|   | Sample 1 (mm) | Sample 2 (mm) | Sample 3 (mm) | Average (mm) |
|---|---------------|---------------|---------------|--------------|
| Shampoo base (negative control)                 | 5.25          | 5.30          | 5.15          | 5.23         |
| 100% <i>R.communis</i><br>methanol leaf extract | 10.15         | 9.78          | 10.24         | 10.05        |
| 50% <i>R.communis</i> methanol<br>leaf extract  | 9.05          | 9.25          | 8.90          | 9.07         |
| 25% <i>R.communis</i> methanol<br>leaf extract  | 6.17          | 6.22          | 6.30          | 6.23         |
| <i>Ricinus communis</i> leaf<br>extract shampoo | 10.94         | 10.89         | 11.19         | 11.01        |
| Ketoconazole 2% shampoo<br>(positive control)   | 11.65         | 11.45         | 10.53         | 11.21        |

#### Figure 1: Zones of inhibition (mm) of test samples on Microsporum canis



#### One Way ANOVA of mean zones of inhibition

Results presented in Table 5 show that there was a significant difference in inhibition of *Microsporum* canis species by the different test samples of shampoo base, 100% *Ricinus communis* methanol leaf extract, 50% *Ricinus communis* methanol leaf extract, 25% *Ricinus communis* methanol leaf extract, *Ricinus communis* methanol leaf extract, *Ricinus communis* methanol leaf extract, 8 ketoconazole shampoo (F= 235.9127, df=5, P-value=1.51E-11).

#### Sum Variance Groups Count Average 3 15.70 5.23 0.0058 Shampoo base (negative control) 100% extract 3 30.17 10.06 0.06 50% extract 3 27.20 9.07 0.03 25% extract 3 18.69 0.0043 6.23 3 Ricinus communis shampoo 33.02 11.01 0.03 2% Ketoconazole (positive control) 3 33.63 11.21 0.3568 ANOVA Source of SS Df MS F P-value F crit Variation **Between Groups** 94.96 5 18.99 235.91 1.51E-11 3.11

A comparison of zones of inhibition caused by the *Ricinus communis* shampoo and 2% Ketoconazole shampoo showed that both of the formulations exhibit substantial antifungal inhibition. Results analyzed in Table 6 indicate that there was no significant difference in zones of inhibition of the formulated *Ricinus communis* methanol leaf extract shampoo and that of marketed 2% Ketoconazole shampoo (P-value = 0.59957; P-value> 0.05

#### Table 6:

#### Zones of inhibition of the formulated Ricinus communis and 2 % Ketoconazole shampoo

| Groups                        |          | Count |      | Sum   | Average | Variance |
|-------------------------------|----------|-------|------|-------|---------|----------|
| Ketoconazole 2% (<br>control) | positive | 3     |      | 33.63 | 11.21   | 0.36     |
| Ricinus communis s            | shampoo  | 3     |      | 33.02 | 11.01   | 0.03     |
| ANOVA                         |          |       |      |       |         |          |
| Source of<br>Variation        | SS       | Df    | MS   | F     | P-value | F crit   |
| Between Groups                | 0.06     | 1     | 0.06 | 0.32  | 0.60    | 7.71     |

# DISCUSSION

Discovering novel classes of antifungals and chemicals that inhibit resistant mechanisms is essential due to the proliferation of multidrug-resistant fungal strains and the decreasing number of medicines currently on the market. A search for therapeutic alternatives has resulted from this, especially among medicinal plants and chemicals derived from them that are employed for their proven antifungal effects (Abad et al., 2006; Lopes and Salgueiro, 2017). In this study, methanol extracts of dry *Ricinus communis* leaves exhibited inhibitory activity against *Microsporum canis*, a species that causes *Tinea capitis*. Therefore, an antifungal shampoo containing *Ricinus communis* methanol leaf extract was formulated and evaluated for antifungal activity. Soxhlet extraction was preferred since the technique achieves complete extraction with minimum amount of solvent (Gopalasatheeskumar, 2018).

Methanol was chosen as the main solvent in this Soxhlet extraction process based on the phytoconstituents isolation procedure. Methanol is a semi-polar solvent which can extract several phytoconstituents. The solvent needs to be inert and simple to remove in order to obtain a substantial yield (Alam et. al, 2008). Since methanol solvent was used, phytochemicals obtained from the extraction may include polyphenols, polyacetyles, flavonoids, terpenoids, tannins, steroids, alkaloids, glycosides and saponins (Kannadasan et. al, 2011). In this study, the percentage yield of *Ricinus communis* methanol leaf extract was 57.01%. This showed that the application of Soxhlet extraction with methanol solvent for *Ricinus communis* leaves produced a substantial yield of extracts.

#### Table 5: One Way ANOVA of mean zones of inhibition

*Ricinus communis* extract shampoo was designed to achieve the desirable characteristics of an ideal shampoo which include: Optimum viscosity to facilitate easy application; it should be capable of spreading effectively. The shampoo should provide enough lather during usage, be able to remove foreign materials and should not leave any sort of coating on the scalp. It needs to be properly washable. In addition, it should produce lather with both hot and cold water, and make combing simpler after shampooing. After drying, the hair should not have a rough appearance. It should make the hair more lustrous both during and after shampooing and it must have a pleasant smell. The scalp should not itch or become irritated in any manner as a result of its application. Also, no microbial development should be supported by it. The shampoo should be stable with a shelf life of 2 to 3 years. It should be cost-effective (Mithal & Saha, 2000, Sharma et al., 2018).

In this study, the shampoo was designed to deliver the active ingredients effectively and to achieve the intended antifungal activity against *Tinea capitis*. The choice of each ingredient and its quantity was prompted by the purpose of the ingredients. Detergents, conditioners, thickeners, sequestering agents, pH adjusters, preservatives and specialty additions were used to make shampoo (D'Souza &Rathi, 2015; Gottschalck, 2006). These measurements were adjusted in order to produce a shampoo that achieves the basic purpose of cleaning the scalp, skin, and hair and also to condition and beautify hair and as an adjuvant in the treatment of *Tinea capitis*.

During formulation, anionic surfactant was used as the primary surfactant because of the excellent foaming characteristics (Sharma et al., 2018). The most commonly used anionic surfactant is sodium lauryl ether sulphate (Poucher, 2013). Secondary surfactants improve the effectiveness of foam, viscosity and other antistatic and conditioning properties. Foam boasters, sequestrants such as Disodium EDTA and citric acid are used in many shampoos. Vitamin E is used an antioxidant (Sharma et al., 2018; Gottschalck, 2006). The regulatory authorities mandate that each and every batch of shampoos be tested before being marketed. An assessment of activity and safety is part of evaluation. Since synthetic detergents are used to make the majority of shampoos, it also warns about their toxicity, making evaluation a crucial factor (Sharma et al., 2018).

For physical characteristics, the *Ricinus communis* shampoo was observed to be homogenous, consistent in fluidity, good ability to produce foam and with a pleasant odor. Therefore, this showed that the formulated shampoo exhibits the ideal properties of a desirable shampoo. A shampoo should have a pleasant scent and good aesthetic properties (Malviya & Sharma, 2014). The pH of formulated *Ricinus communis* shampoo was 5.63, a slightly acidic pH which is near the normal pH of the skin. This is a preferred pH of a shampoo. The pH levels of shampoo formulations are crucial for increasing and enhancing hair's attributes, reducing eye discomfort, and stabilizing the ecological balance of the scalp. Mild acidity reduces edema and inhibits scaling, which results in shine. Thus, one strategy to reduce damage to the hair is to promote shampoos with a lower pH, which is the current trend as indicated in a similar study on formulation and evaluation of polyherbal shampoo in order to increase hair growth and inhibit fungal growth (Lodha, 2019).

The *Ricinus communis* shampoo demonstrated pseudoplastic behavior since viscosity was observed to decrease with increasing shear stress. Product viscosity is crucial in defining and regulating a variety of characteristics, including stability, product aesthetics, like clarity and flow-ability for packaging, shampoo spread-ability on hair, and product consistency in the package (Gaud et al., 2001).

In vitro antifungal assay against *Microsporum canis* species was performed to determine antifungal activity of the formulated shampoo against *Tinea capitis*. Ketoconazole 2% w/v shampoo was used as positive control, while a blank shampoo base was used as negative control. *Microsporum canis* species was chosen for this experiment based on the fact that it is a worldwide distributed zoophilic dermatophyte that affects many animal species, including humans, and causes clinical disorders that are frequently characterized by multifocal alopecia, scaling, and circular lesions. For the treatment of fungal *Microsporum canis* infection, a wide range of oral and topical antifungal treatments are available. However, the effectiveness of these medications and treatment regimens varies, and up to 40% of patients may not respond to treatment for resistance-related reasons. One of the biggest obstacles to determining microbiological resistance in clinical cases where treatment is not working is the absence of standardized reference methodologies for measuring *Microsporum canis* antifungal susceptibility. As a result, information regarding current treatment for *Microsporum canis* and the procedures used to test the antifungal activity of the most widely used medications, namely, Allylamines, Azoles, Polyenes, and Griseofulvin have been compiled (Aneke et al., 2018).

Both crude extracts of *Ricinus communis* leaves and the formulated shampoo of *Ricinus communis* methanol leaf extracts, showed inhibitory effect against a culture of *Microsporum canis* species. The zone of inhibition caused by the formulated shampoo (11.01 mm) was higher than that of crude extracts alone. Ketoconazole shampoo, which was used as positive control for the experiment showed 11.23 mm zone of inhibition, while the lowest activity was reported for the blank which did not contain *Ricinus communis* extracts.

The results from this present study revealed that there was no significant difference in zones of inhibitions of the formulated shampoo and Ketoconazole shampoo, (P-value=0.59957 and thus P-value> 0.05). However, there was significant difference in comparing measured zones of inhibition of the shampoo base, crude extracts of *Ricinus communis* at different concentration, the prepared shampoo and Ketoconazole shampoo (P-value=1.51E-11 since P-value< 0.05). The different concentration of the crude extracts of *Ricinus communis* produced inhibitory actions, with 25% concentration having the least zone of inhibition. The shampoo formulation showed greater inhibition compared to the neat (100%) extract. This can be attributed to the inhibitory effect of some of the excipients such as surface active agents that were used in formulation.

The inhibitory effects observed in this antifungal assay of both methanol extracts and the developed shampoo containing *Ricinus communis* methanol extracts was due to presence of different contents of active substances found in *Ricinus communis* leaves. According to Jena & Gupta, 2012), steroids, saponins, alkaloids, flavonoids, and glycosides are all found in *Ricinus communis*, according to the preliminary phytochemical analysis. The presence of alkaloids, ricinine (0.55%) and N-demethylricinine; flavones glycosides quercetin-3-O- $\beta$ -D-glucopyranoside, quercetin-3-O- $\beta$ -D-glucopyranoside, quercetin-3-O- $\beta$ -D-glucopyranoside, kaempferol-3-O- $\beta$ -D-glucopyranoside, kaempferol-3-O- $\beta$ -D-glucopyranoside, kaempferol-3-O- $\beta$ -D-glucopyranoside, the antifungal activity observed in the study suggest significant potential of inhibition against fungal *Microsporum canis* species. Similar findings of activity have been reported for methanol leaf extract of dried leaves of *Ricinus communis* showed that the methanol leaf extracts of the chosen plant have significantly more potential to inhibit the growth of pathogenic bacterial and fungal strains than ethanol and aqueous leaf extracts (Naz&Bano, 2012).

# CONCLUSION

This research concludes that a *Ricinus communis* leaf extract-based shampoo was formulated and evaluated for physical properties and antifungal activity. The formulated shampoo demonstrated desirable physical characteristics for its application; it depicts pseudoplastic flow properties, foams easily, thoroughly eliminates filth, easily washable, mildly acidic and has a pleasant odor. The shampoo exhibits antifungal activity against *Microsporum canis species*. Zones of inhibition caused by the formulated shampoo and marketed 2% ketoconazole shampoo showed no significant difference. The *Ricinus communis* leaf extract shampoo is a potential therapeutic alternative to conventional antifungal formulations for adjunct therapy.

# **RECOMMENDATIONS**

Based on the above conclusion, the formulated *Ricinus communis* leaf extract shampoo should be developed further to ensure quality consistency and compliance to Marketing Authorization requirements. Validation of methods and stability studies of the shampoo are necessary. In vitro research on combination of *Ricinus communis* shampoo with synthetic antifungal agents including conventional antifungals to determine synergism and toxicity will provide essential information on product use.

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