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RESEARCH ARTICLE

White Turmeric (*Kaempferia rotunda L.*) extract liquid soap preparation for feminine hygiene and effectiveness against *Candida albicans*

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Keywords
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Abstract

Background: *Kaempferia rotunda* L. (*white turmeric*) contains alkaloids, phenols, saponins, glycosides, steroids, terpenoids, and other ingredients with antimicrobial, anticancer, antiallergic, antioxidant, analgesic, and antifungal properties. Meanwhile, vaginal discharge or fluor albus is an excessive release of non-bloody fluid caused by the *Candida albicans* fungus. **Objective**: This study aimed to formulate white turmeric extracts (*Kaempferia rotunda* L.) into standard liquid soap for feminine hygiene and to determine the level of antifungal activity against *Candida albicans*. **Methods:** The liquid soap was produced by the maceration of white turmeric in 70% ethanol, which was evaporated until a thick extract was formed. This soap preparation included 7% stearic acid, 1% adeps lanae, 2% triethanolamine (TEA), 5% glycerol, white turmeric extract (at concentrations of 20%, 25%, and 30%), vanilla oil, and distilled water added up to 100 mL. Furthermore, the well method was used to test the antifungal activity of the preparations, and analysis was done on SPSS. **Results:** The results showed that the white turmeric ethanol extract was effective against the *Candida albicans* fungus at concentrations of 25% and 30%, with inhibitory diameters of 11 mm and 15 mm (categorised as powerful), respectively. Also, the liquid soap preparations of the extract revealed antifungal activity at a concentration of 30%, with a 9.6 mm inhibition diameter (medium category). The soap met Indonesian National Standard (SNI) requirements, including organoleptic, pH, high foam, and viscosity properties. **Conclusion:** The white turmeric ethanol extract can be formulated into a feminine hygiene soap with antifungal activity at a concentration of 30%.

Introduction

Turmeric is a spice and medicinal plant consumed by Indonesians either as a cooking ingredient, herbal medicine, or to maintain health and beauty. The white turmeric rhizome (*Kaempferia rotunda* L.) in the community is used to treat diarrhoea, heartburn, slimming, and vaginal discharge (Astutiningsih *et al.,* 2014). Generally, the chemical constituents contained in white turmeric include alkaloids, phenols, saponins, glycosides, steroids, terpenoids, and other ingredients with antimicrobial, anticancer, antiallergic, antioxidant, analgesic, and antifungal properties (Sumathi *et al.,* 2013).

However, research on the efficacy of white turmeric as an antifungal agent is limited. Essential oils and simplicia waste extracts from white turmeric rhizome distillation inhibited the growth of *Candida albicans*. The inhibitory power on the growth of *C. Albicans* differs between the oil and the extract (Astutiningsih *et al.,* 2014).

Furthermore, the female reproductive system, such as the vagina, needs to be kept clean because of its hidden location. Women who fail to maintain their genital area tend to experience numerous consequences, including infections caused by fungi, bacteria, parasites, and viruses such as vaginal discharge, unpleasant odours, or others. Vaginal discharge or fluor albus is the excessive

release of non-bloody fluid, with a smell or not, and is accompanied by local itching (Maysaroh & Mariza, 2021).

Generally, antifungal agents prevent yeast infection in the vagina. The antifungal used in this study was derived from herbal plants, namely white turmeric (*Kaempferia rotunda*, L) ethanol extract. Therefore, it was formulated as a liquid soap for easy use to keep the intimate area clean. The feminine hygiene soap is a liquid preparation for cleaning the female area, and it is prepared from basic ingredients that do not cause irritation to the skin (Rahmi *et al.,* 2017). Also, the soap should have a pH similar to the female genital area, which is around 5-8 according to Indonesian National Standard (SNI) standards. This study aimed to formulate white turmeric extracts (*Kaempferia rotunda* L.) into SNI standard female soap preparations and determine the antifungal activity of the ethanol extract and soap preparations against *Candida albicans*.

Methods

Extraction

A sample of 2,000 g of white turmeric simplicia was macerated in an 8 L 70% ethanol solvent. The juice was separated from the pulp 24 hourly using a filter cloth; the pulp was repressed with 70% ethanol and then filtered for three days. The liquid extract obtained was evaporated using a rotary evaporator to produce a thick extract.

Anti-candida activity test

A pure suspension of the fungus *Candida albicans* 0.5 Mc Farland units was prepared in a petri dish containing a sterilised medium on a table with a thickness of 4 mm. The fungal suspension was smeared on the surface of the Sabouraud Dextrose Agar (SDA) medium using a sterile swab until it was evenly distributed, then incubated for 10 minutes. A well was made by pressing against the surface of the media using a sterile blue tip with a diameter of 6 mm. Furthermore, 50 concentrations of white turmeric extract liquid soap were added to each well, using 20%, 25%, and 30% concentrations (Ariami *et al.,* 2017). The negative and positive control used aqua dest and 100% nystatin, respectively. The petri dish was wrapped with coffee paper, then coded and incubated at room temperature 37˚C for 3x24 hours. The growth and clear zone formed were observed, and measurements were taken using a calliper/ruler.

Soap preparations

The materials used were weighed, then dissolved in stearic acid and adeps lanae, and heated at a temperature of 70°C above a water bath. Furthermore, the mixture was placed in a mortar, then glycerol and triethanolamine (TEA) were gradually added while grinding. Then white turmeric extract and vanilla oil were added and stirred until homogeneous. Subsequently, the ingredients were filled with distilled water up to 100 mL.

Liquid soap preparation test

Organoleptic test

Changes in the homogeneity, aroma, and colour of the feminine hygiene soap preparation were observed. Also, observations were made at room temperature weekly, from the first to the 28th day of storage (SNI, 1996).

pH test

The pH measurement was conducted using a pH meter dipped in the soap preparation; the numbers on the meter were read after complete immersion. Also, the pH of the soap preparation must match that of the vagina, which is 3.8-4.5.

Foam power check

The height of the liquid soap solution was measured and stirred at a certain speed using a magnetic stirrer. One gram of liquid soap was placed in a test tube and shaken.

Viscosity check

The instrument used was a Brookfield viscometer, and the liquid soap preparation was placed in a 500 ml beaker glass. Measurements were taken, and the spindle was dipped into the preparation until the boundary line was on the spindle. Then the viscometer was turned on, and examinations were conducted in the first and second week (Lachman *et al.,* 1986).

Testing the antifungal activity

Liquid soap preparations of 20%, 25%, and 30% concentration of white turmeric ethanol extract were planted in wells of the SDA medium containing *Candida albicans*. The positive and negative control used nystatin and a soap base, respectively, and were incubated for 24 hours at 37°C. The antifungal activity was measured according to growth inhibition zones of *Candida albicans*, and data were analysed using SPSS.

Results

Determination of White turmeric plants

The results of the plant identification conducted by the Indonesian Institute of Sciences (LIPI), Bogor, showed that the sample used was a white turmeric plant (*Kaemperia rotunda* L.).

Results of White turmeric simplicia

Dried white turmeric simplicia was produced from 16,000 grams of fresh white turmeric rhizome and subjected to a wet sorting process, washing, slicing, drying, dry sorting, and grinding into dry powder. A total of 2,500 grams dried simplicia yielded 2,000 g of powder.

Extraction results of White turmeric

The white turmeric rhizome extraction was done using the maceration method with 70% ethanol solvent. The 2,000 g of powder yielded 70 grams (3.50%) of thick extract.

Results of the secondary metabolite test for White turmeric

The secondary metabolite test results of the white turmeric extract included saponins, alkaloids, and flavonoids (Kumar *et al.,* 2015). The phytochemical screening showed the presence of a secondary metabolite compound qualitatively, not quantitatively. However, no research has stated the minimum amount of a secondary metabolite compound required to inhibit *Candida albicans*. Therefore, the amount and specifications of the compound required to inhibit the growth of *Candida albicans* are unknown.

Testing the antifungal activity of White turmeric extract

Table I shows the diameter of the inhibition zone of white turmeric extracts (*Kaempferia rotunda* L.) with *Candida albicans*. The result indicated that the white turmeric extract with a concentration of 20% has no antimicrobial activity while 25% and 30% extracts had an inhibition potential of 11 mm and 15 mm, respectively. Furthermore, the negative control Dimethylsulfoxide (DMSO) had no antimicrobial activity, contrary to the positive control, nystatin (inhibition diameter of 15 mm). The inhibition zone produced at the concentration of 30% was classified as powerful. Therefore, the higher the concentration of the formulation, the larger the inhibition zone formed (Ningsih, 2017).

Table I: Results of testing for antifungal activity of extracts

Sample	Concentration	Average diameter (mm)	Category response
Turmeric extract white	20%	-	-
Turmeric extract white	25%	11	Strong
Turmeric extract white	30%	15	Strong
DMSO	Negative control	-	-
Nystatin	Positive control	15.5	Strong

Testing the antifungal activity of White turmeric extract liquid soap

Table II shows the diameter of the inhibition zone of the white turmeric extract (*Kaempferia rotunda*, L.) with *Candida albicans*. The results indicate that the preparations with a concentration of 30% and positive controls have antifungal activity *($p < 0.0001$)*.

Table II: Results of testing for antifungal activity of liquid soap preparations

Sample	Concentration	Average diameter (mm)	Category response
Soap	20%	-	-
Soap	25%	-	-
Soap	30%	9.6	Medium
Soap base	Negative control	-	-
Nystatin	Positive control	16	Strong

Evaluation of White turmeric extract liquid soap for feminine area cleansing

The composition of the formula can be seen in Table III. The following are the evaluation of the liquid soap preparation for feminine hygiene with white turmeric extract (*Kaempferia rotunda*, L.). The organoleptic properties (colour, shape, and odour) of the white turmeric extract liquid soap (*Kaempferia rotunda*, L.) did not change after four weeks of storage. The extract is a thick white liquid with a vanilla fragrance. All three formulations (F1, F2, and F3) had the same pH value of 5. For the negative control (soap base), the pH was 7. After adding the extracts at the concentrations of 20%, 25%, and 30%, the pH became 5. The positive control had a pH of 6.

Table III: Women's liquid soap formula

Ingredient	Formula (%)			Use
	A	B	C	
White Turmeric extract	20%	25%	30%	Active ingredient
Stearic acid	7.5	7.5	7.5	Emulsifier
Adeps lanae	1	1	1	Shaper
Triethanol-amin	2	2	2	Emulsifier
Glycerol	5	5	5	Humectant
Vanilla oil	3 drops	3 drops	3 drops	Fragrance
Aqua dest	Ad 100 mL	Ad 100 mL	Ad 100 mL	Solvent

Observations showed that the positive control (nystatin) produced foam as high as 1 cm, the negative control (soap base) 1.5 cm, F1 (20%) 2 cm, F2 (25%) 2 cm, and F3 (30%) as 2 cm (Figure 1). The preparation of white turmeric extract liquid soap met the requirements of the height of the foam, ranging between 13-220 mm. The viscosity of liquid soap preparations has decreased from day to day, caused by the evaporation of some of the components that make up liquid soap and the effect of storage (Table IV).

Table IV: Viscosity test results on liquid soap turmeric extract

Formula	At week	Viscocity value (cP)
Base form	Week 1	1.299
Extract base		650
Base form	Week 2	840
Extract base		300

Tests were carried out at room conditions of 28°C and sample temperature 25°C

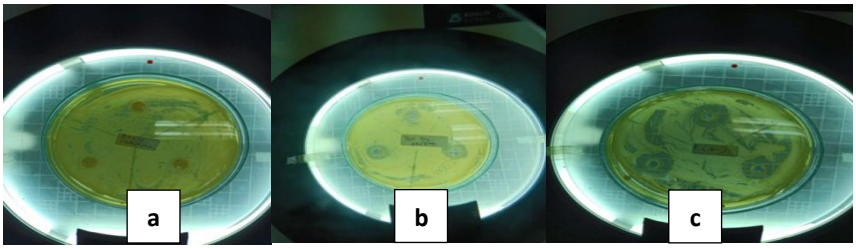

Figure 1: (a) Negative control (soap base); (b) Concentration 30%; (c) Positive control (nystatin)

Discussion

Generally, flavonoids, saponins, tannins, and alkaloids are thought to have antifungal activity. Flavonoids act by binding to microtubule proteins in cells and disrupting the mitotic spindle function, resulting in fungal growth inhibition (Bhaskara, 2012).

The antibacterial effect of alkaloids in the white turmeric extract was due to the ability of the quaternary aromatic groups to intercalate with DNA and disrupt the integrity of the peptidoglycan constituent in bacterial cells. Therefore, the peptidoglycan disruption causes the cell wall layer to be incompletely formed, resulting in cell death (Rahman *et al.,* 2017). The alkaloids cause cell damage as antifungals and bind strongly to ergosterol to form holes, leading to membrane leakage, permanent cell damage, and death in fungi (Jager, 2014).

The flavonoids inhibit fungal growth by interfering with the permeability of fungal cell membranes. Also, the hydroxyl group in flavonoid compounds alters organic components and nutrient transport, leading to toxic effects on fungi (Oliveira *et al.,* 2016).

The antibacterial mechanism of saponins involves the leakage of proteins and enzymes within cells (Madduluri *et al.,* 2013). Saponins play a role in inhibiting or killing *Candida albicans* by lowering the surface tension and increasing the permeability of the fungal cell wall sterol membrane, causing the cell to expel a concentrated intracellular fluid, allowing metabolic substances, nutrients, enzymes, and proteins to escape and result in fungal death (Lolok *et al.,* 2020).

According to the data, white turmeric extract liquid soap at a concentration of 20% and 25% had no antifungal activity because these concentrations of extract in the soap were insufficient. However, a concentration of 30% showed an antifungal potential of 9.6 mm, indicating a moderate antifungal inhibition of the preparation; hence, combining soap-making ingredients affects the active substances in the extract. The antifungal potency of the positive control (nystatin) was 16 mm, indicating a potent inhibitory activity.

The negative control (soap base) showed no antifungal potential, indicating that the ingredients used in soap production lacked an active substance that inhibits moulds. The inhibition zone of the white turmeric extract decreased due to the addition of a dosage base containing no active antifungal substance. Furthermore, the evaluation of the physical aspects of the feminine soap showed that it met the SNI requirements for organoleptic, stability, pH, viscosity, and high foam properties. From the observations, the colour of the soap obtained was milky white in the negative control (soap base) and light brown in the 20%, 25%, and 30% formulations due to the addition of the white turmeric extract. During the four-week test, the brown colour and the liquid state of the soap preparation remained unchanged in the three formulations because it was stored at a stable temperature, i.e., room temperature.

According to the United States Patent, pH values for vaginal douches should range from 5.5 to 8.5. At these values, pH will not interfere with the normal flora of bacteria in the vagina. The preparation is considered stable if the pH does not change significantly. Furthermore, foam height was stable because the material used was within the required limits of 0.5-2.5% (Rowe, 2009). Regarding storage, white turmeric extract was stored at a stable room temperature of 27^{o}C. Further research should improve the formula to achieve optimum antifungal potency against *Candida albicans*, with the highest inhibition zone diameter.

Conclusion

The white turmeric ethanol extract can be formulated into a feminine hygiene soap with antifungal activity at a concentration of 30%.

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Conflicts of interest

There are no conflicts of interest.

References

Anna K Jager, S.H.F. (2014). Correlation between Plant Secondary Metabolites and Their Antifungal Mechanisms–A Review. *Medicinal & Aromatic Plants*. https://doi.org/10.4172/2167-0412.1000154

Ariami, P., Danuyanti, I., & Anggraeni, B.R. (2017). The Effectiveness of Mangosteen Peel Tea (Garcinia mangostana L) as Antimicrobial against the Growth of Methicillin-Resistant Staphylococcus aureus (MRSA) Bacteria. Laboratory Technology

Astutiningsih, C., Octaviani, R., & Suratiningsih, S. (2014). Inhibitory power of essential oil and waste extract from distillation of white turmeric rhizome (Kaempferia rotunda L.) on growth Candida albicans ATCC 10231. *Jurnal Farmasi Sains Dan Komunitas,* **11**(1), 18-22

Bhaskara, G. Y. (2012). Uji Daya Antifungal Ethanol Extract of Bay Leaf (Syzygium polianthum [Wight] Walp.) against Candida albicans Atcc 10231 secara in Vitro. Medical Faculty of Universitas Muhammadiyah Surakarta

Kumar, A., Kumar, S., & Navneet. (2015). Antimicrobial activity and phytochemical analysis of Kaempferia rotunda L. rhizomes. *Der Pharmacia Lettre,* **7**(9), 389-395

Lachman, L., Liebarman, H.A., dan Kanig J.L. (1986). Theory and Practice of Industrial Pharmacy II, translated by Siti Suyatmi, Edition III (III). Jakarta: University of Indonesia

Lolok, N., Awaliyah, N., & Astuti, W. (2020). Formulation and Activity Test of Liquid Soap Preparations for Feminine Cleansing Extracts of Waru (Hibiscus tiliaceus) Leaves Against Fungus Candida albicans. *Jurnal Mandala Pharmacon Indonesia*. https://doi.org/10.35311/jmpi.v6i01.53

Madduluri, S., Babu Rao, K., & Sitaram, B. (2013). In vitro evaluation of antibacterial activity of five indigenous plants extract against five bacterial pathogens of human. *International Journal of Pharmacy and Pharmaceutical Sciences*

Maysaroh, S., & Mariza, A. (2021). Knowledge About White in Young Women. *Jurnal Kebidanan Malahayati,* 7(1), 104-108. https://doi.org/10.33024/jkm.v7i1.3582

Ningsih, D.R. (2017). Leaf extract of mango (Mangifera indica L.) As antifume against candida albicans and identification of compound class. *Jurnal Kimia Riset*. https://doi.org/10.20473/jkr.v2i1.3690

Oliveira, V.M., Carraro, E., Auler, M. E., & Khalil, N.M. (2016). Quercetin and rutin as potential agents antifungal against Cryptococcus spp. *Brazilian Journal of Biology*. https://doi.org/10.1590/1519-6984.07415

Rahman, F.A., Haniastuti, T., & Utami, T.W. (2017). Skrining fitokimia dan aktivitas antibakteri ekstrak etanol daun sirsak (Annona muricata L.) pada Streptococcus mutans ATCC

35668. *Majalah Kedokteran Gigi Indonesia*. https://doi.org/10.22146/majkedgiind.11325

Rahmi, I.W., Nurhikma, E., Badia, E., & Ifaya, M. (2017). Formulasi Sabun Pembersih Kewanitaan (Feminime Hygiene) dari Ekstrak Kulit Buah Durian (Durio zibethinus Murray). *Jurnal Mandala Pharmacon Indonesia*. https://doi.org/10.35311/jmpi.v3i02.8

Raymond C Rowe, P.J.S. and M.E.Q. (2009). Handbook of Pharmaceutical Excipients (6th ed.). London: American Pharmaceutical Association., 6

Sumathi, S., Iswariya, G. T., Sivaprabha, B., Dharani, B., Radha, P., & Padma, P.R. (2013). Comparative Study of Radical Scavenging Activity and Phytochemical Analysis of Fresh and Dry Rhizomes of Curcuma zedoaria. *International Journal of Pharmaceutical Science and Research*