

# Efficacy of Banana Leaf Oil Extract Against Candida Albicans Growth in Diaper Rash

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**Abstract.** The baby's skin is very sensitive, so it is prone to infection or suffering from allergies or fungi and bacteria. Skin disorders that often occur in babies are characterized by the appearance of prickly heat, irritation, diaper rash, or eczema. The prevalence of diaper rash in infants aged 0-12 months is quite high at 25% of the 6,840,507,000 babies in the world. Candidiasis dermatitis is a diaper caused by a fungal infection of candida albicans. The use of antifungal drugs causes candida albicans to become more resistant and causes side effects. The use of natural materials began to be developed as safer natural antifungals. Banana leaves contain alkaloids, flavonoids, and polyphenols that are antifungal. The study aimed to analyze the effectiveness of banana leaf oil extract in inhibiting the growth of candida albicans fungi.

Method: This was experimental research. The sample of this study was a pure culture of candida albicans totaling 30 samples, divided into 5 groups of treating concentrations of plantain leaf oil extract. The variables of this study were the concentration of plantain leaf oil extract 0%, 25%, 50%, 75%, and 100% as free variables, and the diameter of the bat growth zone of candida albicans fungus as a bound variable. Data analysis was carried out with the ANOVA test and the Tukey LSD test to analyze the effectiveness of banana leaf oil extract against the growth of candida albicans fungi.

Results and Discussion: Phytochemical test results show that banana leaf oil extract contains alkaloid compounds, flavonoids, and polyphenols. The results of the inhibition zone data analysis with the ANOVA test and the Tukey test showed a significant influence (p < 0.05) on each concentration of the extract on the growth of candida albicans. The results of measuring the diameter of the inhibition zone of the largest plantain leaf oil extract were at a concentration of 25%. Thus, the extract concentration of plantain leaf oil is effective for inhibiting the growth of candida albicans fungi at a concentration of 25%.

Conclusion: Based on the results of this study, it can be concluded that banana leaf oil extract can effectively inhibit the growth of candida albicans at a concentration of 25%.

**Keywords:** Diaper rash · Candidiasis Dermatitis · Candida albicans · Banana leaf extract

#### 1 Introduction

The health of the baby is the most important aspect that parents must pay attention to; if this problem is ignored, it will become a serious problem. Babies have different needs related to the hygiene of daily life, such as nutrition or care. Diapers are important accessories needed by babies. Usually, diapers, when used, are in direct contact with the skin 24 h a day, seven days a week. Frequent use of diapers in babies can cause problems for the health of their skin [1].

Diaper dermatitis (DD), or diaper rash, is a common dermatological problem in newborns, infants, and young children characterized by red and thickened-looking skin that often arises on diaper-covered areas of the skin. Various factors cause (etiology) the occurrence of diaper rash (diaper rash, diaper dermatitis, napkin dermatitis), including irritation or friction between the diaper and the skin, moisture factor, lack of maintaining hygiene, use of diapers that are rarely changed or for too long, not immediately replaced after peeing or defecation (feces), microorganism infection (especially fungal and bacterial infections), diaper material allergies, disturbances in the sweat glands in diaper-covered areas, due to wet areas, injury or sensitivity to certain chemicals, eating new foods, and the use of antibiotics [2–4].

At a very young age (baby), the development of the skin is not yet mature enough, and its functions have not been fully developed, as is its immune system. Therefore, this baby can be said to have skin sensitive to anything that is outside his body, such as skin barrier disorders or percutaneous absorption from irritants [3, 4].

Diaper rash, also called diaper dermatitis, is an inflammatory eruption in the area covered by diapers, namely the thigh, buttocks, and anal areas [5]. The skin in the diaper area is prone to prolonged contact with urine and feces, as well as diaper occlusion, which causes overhydration and changes in the pH of the skin. This environment increases damage to the structure of the stratum corneum, which directly contributes to the disruption of barrier function. Prolonged exposure to urine and feces results in a more alkaline pH of diaper-covered skin resulting in changes in microbial colonization, activation of protease enzymes and fecal lipase, and disruption of the stratum corneum. Friction from wet diapers can cause skin protection damage and increased skin permeability to potential irritation or inflammatory triggers. These factors which contribute to diaper dermatitis [3, 4].

Clinical symptoms of diaper rash or diaper dermatitis on the baby's skin are characterized by reddish skin, which is one of the symptoms of the reaction in the baby's body. Diaper dermatitis causes perianal redness with scales and folds of the thigh folds, while fungal dermatosis is characterized by bright red lesions with multiple satellite lesions and erosion. This can cause itching, soreness, and heat in the baby, making the baby cry or uncomfortable [6, 7].

Dermatitis Diapers (DD) can be caused by candida albicans fungal infection called candida dermatitis, which develops rapidly in tropical areas, such as Indonesia, due to dusty air conditions, warm temperatures, and humid temperatures so that microbes can thrive in this region. [8]. Infections caused by fungi increase significantly for those with weak immunity, including newborns, who are very susceptible to fungal infections, such as candida dermatitis [9].

The infection caused by candida is called candidiasis. Candidiasis can be found worldwide and affect various ages, both men and women. The prevalence of candidiasis in Indonesia, which is around 20–25%, can attack hair, skin, nails, mucous membranes, and other organs, such as the mouth and esophagus [10].

Meanwhile, the incidence of diaper rash dermatitis varies by country, depending on the baby's hygiene and the parents' knowledge (caregivers) about how to use diapers, and may also be related to weather factors. According to research, 16% of babies have diaper rash in the UK, and most are between the ages of 12 to 24 months [11]. DD levels among hospitalized infants vary from 4 to 67% [11–14]. Esser [15] reported that babies have DD at about 42 days and a birth weight of 2500 g. In addition, babies born at a gestational age of enough months are roughly five times more likely to develop DD when they receive food than when not fed [15].

Based on data from WHO in 2012, the prevalence of skin irritation (diaper rash) in babies aged 0–12 months is quite high 25% of the 6,840,507,000 babies born worldwide mostly suffer from diaper rash due to diaper use. Frilasari, 2016 proves that the incidence of diaper rash in Indonesia reaches 7–35%, which affects male and female babies. Putra, 2012, data according to Kimberly A. Horii and John Mersch in 2010 states that 10–20% of dermatitis diapers are found in pediatric practices in America. Prevalence in infants ranges from 7–35% [16]. Rimba, 2015 mentioned that most numbers were found at 6–12 months old. In Indonesia, according to statistical data from the Pediatric Dermatology Division of the Children's Skin Polyclinic of Cipto Mangunkusumo Hospital Jakarta in 2005–2009, one in three babies or toddlers has experienced diaper rash. The incidence of diaper rash reaches 7–35%, which affects both male and female babies [17, 18].

Candida albicans is the most common fungus that causes disease in neonates. Other species of candida that also cause neonatal infections include C.parapsilosis and C.glabrata. Candida mukokutan infections in neonates often occur in the form of thrush (oropharyngeal candidiasis) and diaper rash [19]. The candida albicans species is the most abundant species worldwide, representing a global average of 66% of all candida sp [10, 20]. This species is generally a normal microorganism that exists in the human body, namely in the digestive tract, reproductive tract, oral cavity, and human skin. Candida yeast is usually in the intestinal tract and can be found on the mucous membranes and skin without causing infection [21]. In individuals who have a healthy immune system, Candida albicans are often harmless. However, predisposition factors, such as changes in the host's microbiota, changes in the host's immune response, or environmental consequences, can allow this fungus to grow and cause infection [22].

Candida fungus has predilection in moist and warm areas of the body folds, supported by hot weather and a tropical climate, excessive sweat production, and lack of maintained personal hygiene so that it can cause the skin to become susceptible to candidiasis. Candidiasis is the result of the overgrowth of candida fungal organisms. The signs and symptoms of candidiasis vary depending on the area of the infected body [23]. This pathogenic fungi mainly infects hair, skin, and nails (dermatophytes) and causes the most common mycotic skin disease (dermatophytosis) worldwide [21]. Candidiasis causes the infected area to become red and moist, and to vesiculation [10]. Most candida infections cause minimal complications, such as redness, itching, and discomfort, although complications can become severe or even fatal if left untreated in certain populations [23].

The clinical presentation can be broadly divided into cutaneous candidiasis, mucosal candidiasis, and systemic candidiasis. In immunocompetent people, candidiasis is usually a local infection of the skin, toenails or toenails (onychomycosis), or mucous membranes. Candidiasis of the skin and mucous membranes occurs in all age groups [21, 23].

Candida mukokutan infections in infants are the most common infections, such as oral candidiasis and diaper rash, to serious infections. Diaper rash in babies hits moist skin, such as the buttocks and genitalia, thigh folds, most often on the skin area affected by the diaper. Babies with diaper rash usually have candida colonies in the digestive tract with positive culture results from feces. When defecating, infected feces transmit yeast to the skin around the anus. The occlusive and humid diaper environment supports the growth of candida [19].

One of the management of people with diseases caused by infection is by treatment using antibiotics and antifungals. Antifungal therapy is usually carried out topically and systemically. Topical anti-fungal therapeutics have several advantages over systemic therapies, namely lower side effects and drug interactions, localized therapies, and lower costs. The group of topical antifungals used in candidiasis include imidazoles and polyene. Imidazole is antifungal with a broad spectrum. Nystatin and amphotericin B are the two main types of topical drugs of antifungal polyene. The systemic antifungal group used in mucocutaneous candidiasis includes fluconazole and amphotericin B. (Kusumaputra and Zulkarnain, 2014). Research in several countries shows that the use of antibiotics is increasing, but there are many uses that are not yet rational. The constant use of antifungal drugs for candidiasis, such as fluconazole, itraconazole, ketoconazole, nystatin, griseofulvin, or amphotericin B can cause the Candida albicans fungi to become resistant to such antifungals [24, 25]. Therefore, it is necessary to study other alternatives to treat, one of which is to use traditional medicines.

Using efficacious plants for medicine is one of the efforts to overcome the problem of fungal infections. In addition, medicinal plants also have minimal side effects. Today, medicinal plants are widely studied scientifically as a substitute for chemical drugs. One of the plants with the potential as a medicinal plant is the banana plant. Parts of banana plants consisting of roots, stems, midribs, leaves, flowers, and fruits have varying potential. Previous studies reported that banana stem extracts have activity in inhibiting the growth of candida albicans. Banana stems also contain several types of phytochemical substances, such as saponins and tannins, that function as antibiotics, and antifungals, accelerate the growth of new cells, stimulate the formation of fibroblasts, and inhibit the growth of bacteria [26, 27].

To date, relatively little research has been done on the benefits and content of banana leaves (Musa sp.). Banana leaf extract contains active phytochemical compounds and secondary metabolites of the tannin group, phenols, flavonoids, alkaloids, steroids, glycosides, and saponins. Some of the benefits of the content of secondary metabolite compounds that are usually present in plants have the potential as antioxidants, anticancer, anti-inflammatory, and antimicrobial. The content can also treat various types of diseases [8, 28].

Based on the description above, this study was conducted to analyze the effect of banana leaf extract (moses sp.) against the growth of candida albicans fungus that causes diaper rash in babies, which can be applied as an active ingredient in anti-diaper rash drugs.

## 2 Method

This was experimental research. The study sample was purely candida albicas with a sample size of 36, divided into six treatment groups with a concentration of banana leaf oil extract, which was 0% and 25%. The study variable consists of the free variable and bound variable. The free variable is in the form of the concentration of banana leaf oil extract. Medium-bound variable bound is a fungal growth rash (Candida albicans). The growth examination stage of candida albicans was carried out laboratory with a series of examination processes as follows:

## 2.1 Banana Leaf Oil Extract Preparation

Clean and fresh plantain leaves washed and dried in the sun to dry. Leaves that had been dried were immediately mashed for further extraction. Banana leaf oil extract was infused with a socleation process with a 96% alcohol solvent and given a little orange peel to give it a scent. The simplicia powder was placed in an extraction bag coated with filter paper in such a way and then put into alcohol-stripping water in a round base flask. The coating liquid was heated to give the active substance of simplicia. During the heating process, the vapor of the igniting liquid and the active substance would be formed and condensed in the condenser until it turned into a liquid substance again containing the active substance of simplicia. These drops of fallen liquid substance were accommodated in a container as an extract of plantain leaf oil. The extraction yield of pure plantain leaf oil (100%) was then diluted with 96% alcohol to a concentration of 25%, 50%, and 75%.

## 2.2 Evaluation of the Physical Properties of Banana Leaf Oil Extract

The physical properties of banana leaf extract oil from socleation were carried out using organoleptic tests and pH measurements of banana leaf oil extract.

1. Organoleptic/Macroscopic Assay

This test was carried out by observing color, smell, and shape (texture). The color test was carried out visually with the direct eye, the odor test was by smelling the aroma of this banana leaf oil, and the shape test was by observing the shape, whether it is liquid or viscous.

2. Measurement of pH extract

PH determination was carried out using universal pH paper. The finished oil was dripped on the pH paper, and then its pH was observed. The pH requirement for the skin was 4.5 to 6.5.

#### 2.3 Phytochemical Test of Banana Leaf Oil Extract Content

Phytochemical tests were used to determine the content of active antifungal compounds that can inhibit the growth of fungi. Phytochemical tests were carried out with the following procedures [29]:

1. Alkaloid test

The oil extract was weighed 0.5 g, put into a test tube, dissolved with 1 ml of HCl 2N and 9 ml of water, and then divided into three tubes. The result was positive that it contained alkaloids; if added, Mayer reagents would form a white (yellowish-white) precipitate, and Wagner reagents would produce a brown precipitate.

2. Test saponins

The oil extract was weighed 0.5 g, and put in a test tube. 10 ml of hot water was added and shaken for 10 min until foam or more was formed and then dripped with HCl 2N. If the foam did not disappear with the addition of HCl 2N, then the extraction was positive for containing saponins.

3. Tannin test

The oil extract was put in a test tube, 10 ml of hot water was added and shaken, and then 20 ml of 10% NaCl was added and filtered. The resulting filtrate was added FeCl3, and if there was a dark blue or black color change, it was positive to contain tannins.

4. Flavonoid Test

Oil extract weighed as much as 0.5 g added with ethanol. Then 5–6 drops of concentrated HCl were added, forming a red color indicating the presence of flavonoids and the formation of an orange color indicating the presence of flavone compounds (Tiwari et al. 2011).

5. Polyphenol test

Banana leaf oil extract of 1 ml was reacted with a 10% solution of iron (III) chloride, in case of dark blue, blue-black, or greenish-black color indicating polyphenol compounds.

#### 2.4 Candida Albicans Sensitivity Test

This test aims to determine the sensitivity or sensitivity of the fungus to the test material. The welling method is more commonly used in antifungal power testing because it is more effective in inhibiting the growth of fungi. The diameter of the clear zone is a clue to the sensitivity of the test fungus, where the wider the clear zone, the better the antifungal power possessed [30].

The susceptibility test used McFarland standard 0.5 with 9.95 ml H2SO4 added with BaCl2 1% 0.05 ml. Then, the Candida albicans suspension was mixed with the PZ solution. After that, it was compared with McFarland's turbidity standards. Furthermore, fungi suspense was planted on natural resources media with full streaking. After that, disc paper that was given various concentrations of banana leaf oil extract 25%, 50%, 75%, and 100% and sterile aquades (0%) stuck to the fungus in the SDA medium. And as an appeal of the positive inhibition zone, disc paper was dipped in a 2% solution of the antibiotic ketoconazole. Disc paper was then incubated at room temperature for 24–48 h to wait or see the formation of clear zones around the disc paper to be observed and steamed clear zones.

## 2.5 Data Analysis

To study the effect of differences in banana leaf oil extract concentrations on the growth of candida albicans, inhibition zone data were analyzed using the ANOVA test with  $\alpha$  0.05. Meanwhile, to find out the efficacy of the concentration of banana leaf oil extract, it was analyzed with the Tukey LSD test.

## 3 Results

The results of this study can be presented in the form of test results of the following test parameters:

A. Evaluation of the physical properties of plantain leaf oil extract The results of the observation of the organoleptic test in a macroscopic manner on the results of banana leaf oil extraction showed an oil extract in the form of a thick liquid, clear, and scented natural banana leaves fragrance. The pH measurement result of the oil extract is about 6.5.

- B. Phytochemical Test Content of Banana Leaf Oil Extract The results of the phytochemical test of banana leaf oil extract content are presented in Table 1.
- C. Test of growth sensitivity of candida albicans

The results of the candida albicans growth sensitivity test on natural resources were measured as the diameter of the colorless (clear) inhibition zone observed on disc paper which had been treated with various concentrations of plantain leaf oil extract. The results of the shrinkage of the inhibition zone are presented in Table 2.

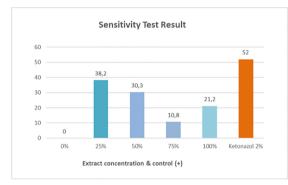
To know the differences in inhibition zones at each concentration of banana leaf oil extract, the average value of the diameter of the inhibition zone is presented in the form of a bar graph in Fig. 1.

Content	Reagents	Result
Alkaloids	Mayer reagent	+
	Wagner reagent	+
Saponins	HCl 2N	_
Tannins	NaCl 10%, FeCl3	_
Flavonoids	Ethanol, concentrated HCl	+
Polyphenols	FeCl3	+

Table 1. Phytochemical test of banana leaf oil extract content

No.	The concentration of Plantain Leaf Oil Extract	Block Zone Diameter
1	0%	0 mm
2	25%	$38.2 \text{ mm} \pm 0.8$
3	50%	$30.3 \text{ mm} \pm 1.0$
4	75%	$10.8 \text{ mm} \pm 1.0$
5	100%	$21.2 \text{ mm} \pm 1.6$
Control (+)	Ketoconazole 2%	$52.0 \text{ mm} \pm 1.0$

Table 2. Results of measurement of the growth inhibition zone of the candida albicans



**Fig. 1.** The Diameter of the growth inhibition zone of candida albicans in each banana leaf extract (source: research results)

## 4 Discussion

The phytochemical test results prove that the content of banana leaf oil extract is alkaloids, flavonoids, and polyphenols. From various literature, this substance has the ability to inhibit the growth of candida albicans fungi, so it has the potential to be an antifungal for candida albicans to overcome diaper rash.

In this study, the preparation of variations in the concentration of banana leaf oil extract was carried out by the dilution method using 96% alcohol. This is done because the oil extract can dissolve well in organic solvents, such as 96% alcohol. The sensitivity test of banana leaf oil extract to the growth of candida albicans was carried out by the welling method. This method is commonly used in antifungal power tests because it is more effective for measuring the inhibitory power of large fungal growth, and the active substance can diffuse directly without obstruction. In addition, by using this method, it can be known the area of the inhibitor zone. The sensitivity test of candida albicans in banana leaf extract was measured as an inhibitory zone which was done by measuring the diameter of the clear zone of banana leaf extract on disc paper. The diameter of the inhibitory zone indicates the test fungus's sensitivity to an active antifungal ingredient. The wider the inhibition zone, the substance has better antifungal power [30].

The results of the sensitivity test of banana leaf oil extract with a concentration of 0%, 25%, 50%, 75%, and 100% obtained the average value of the diameter of the inhibitory zone successively was 0.0 mm, 38.2 mm, 30.3 mm, 10.8 mm, 2 1.2 Mm. Based on this average value, the concentration of banana leaf oil extract with a concentration of 25% has the greatest inhibitory power.

The results of the ANOVA test showed that the difference in extract concentration had a significant effect ( $\alpha < 0.05\%$ ). The Tukey HSD test showed that the inhibitory power of each banana leaf extract concentration was significantly different. Therefore, based on the results of statistical tests in the study, the concentration of 25% banana leaf oil extract had the largest inhibition zone that was significantly different compared to other concentrations. At higher concentrations of the extract, 50%, 75% and 100% of the inhibitory power of candida albicans growth are getting lower, which is indicated by the smaller inhibition zone diameter. This happens because the diffusion ability of the active substance in the extract is inversely proportional to the viscosity of the extract. The higher the concentration of the extract, the greater its viscosity, causing the active substance to be difficult to diffuse (Son et al., 2018). Therefore, the ability of banana leaf oil extract to inhibit the growth of candida albicans fungus is most effective as an antifungal at a concentration of 25%. The anti-fungal ability of banana peel oil extract is 25%, close to the positive control capacity, namely ketoconazole 2%, with an inhibitory zone diameter of 52.0 mm. Thus, banana leaf extract is proven to be a natural anti-fungal candida albicans, is safe to overcome diaper rash and does not cause chemical reactions on the baby's skin, which is sensitive to chemicals.

The antifungal ability of plantain leaf oil extract is due to alkaloid compounds, flavonoids, and polyphenols. According to Sun et al. 2021, antifungal compounds inhibit the growth of candida albicans fungi by several mechanisms, including 1). Inhibition of Candida virulence factors related to the formation of fungal hyphae to form pathogenic fungal colonies; 2) Inhibition of the formation of candida albicans biofilms. Fungal biofilm is a dense 3-dimensional polymer structure that can form a persistent layer to protect fungal cells from external factors so that the fungus will be resistant to drugs antifungal. Inhibition of biofilm formation will destroy the cell defenses of the fungus; 3). Changes in the cell membrane candida, membrane cell fungi consist mainly of sterols, glycerophospholipids, and sphingolipids. All three are the basic ingredients for various functional proteins and maintain the structural integrity of the cell membranes of fungi. Antifungal compounds will interfere with the biosynthesis stages of the three substances so that the availability of the basic ingredients of functional proteins and the defense of membrane structures are reduced, and fungal cells will die; 4). Candida induction apoptosis, apoptosis is a programmed process of cell death in eukaryotes, which involves a series of enzymes under the control of certain genes. Induction of apoptosis in candida is a mechanism for increasing the program of cell death through the inhibition of reactive oxygen species (ROS), which plays an important role in the formation of hyphae [20].

Alkaloids are active substances from plants that serve as powerful drugs and activators for immune cells that can destroy bacteria, viruses, fungi, and cancer cells. Alkaloids have antifungal activity with the mechanism of changing the cell membrane of fungi by inhibiting esterase, DNA, RNA polymerase, and cell respiration and play a role in DNA intercalation, which causes membrane s damage. The alkaloid will bind strongly to ergosterol, forming a hole that causes cell membrane leakage. This permanently damages the cells, and the fungal cells will die [20, 31].

Besides alkaloids, the content of flavonoid compounds in banana leaf oil extract has antifungal compounds. The mechanism of flavonoid antifungal was carried out in several ways, namely inhibiting the formation of hyphae, inhibiting the formation of biofilms, and reducing the generation of ROS to change the morphology of fungal cell membranes. Flavonoids break down the extension of fungal hyphae, inhibiting the formation of protective biofilms of fungal cells against the attack of antifungal substances. The generation of ROS will increase the apoptosis of fungal cells, which will accelerate the death of fungal cells [32, 33].

The mechanism of action of the antifungal effect of phenol compounds was done by damaging the biosynthesis of ergosterol and disrupting the integrity of the fungal membrane. Ephemeral phenol denatures protein bonds on the cell membrane so that the cell membrane is lysed, and possibly phenol can penetrate the cell nucleus. The entry of phenols into the cell nucleus is what causes the fungus not to develop [34, 35].

#### 5 Conclusion

In this study, banana leaf oil extract was shown to contain alkaloid compounds, flavonoids, and polyphenols. In the sensitivity test, banana leaf extract has the power to inhibit the growth of rash fungi (Candida albicans) starting at a concentration of 25% to a concentration of 100%. At a dilution concentration of 25%, using 96% alcohol was effective in inhibiting the growth of the fungus but still less effective when compared to positive controls.

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