

Effectiveness of Pisang Raja Peel Extract (*Musa Paradisiaca L*) on Bacterial Growth of *Porphyromonas Gingivalis* as the Cause of Periodontitis

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Abstract

Background. Periodontal disease is a chronic inflammatory infection that causes damage to the tissue supporting the teeth. The most common periodontal diseases are gingivitis and periodontitis. Periodontitis is a complex, localized chronic inflammatory disease characterized by the destruction of connective tissue, periodontal ligaments, and bone supporting teeth. This periodontal disease is caused by plaque bacteria on the surface of the teeth, where the plaque is a thin layer of biofilm containing a collection of pathogenic microorganisms such as *Porphyromonas gingivalis*, *Actinobacillus actinomycetemcomitans*, *Prevotella intermedia*, *Tannerella forsythia* and *Fusobacterium nucleatum* which are soft deposits. *Porphyromonas gingivalis* is a gram-negative, black pigmented, rod-shaped anaerobic bacteria. *Porphyromonas gingivalis* is one of the dominant bacteria in chronic periodontitis which is found in subgingival plaques. Indonesia is a country with a tropical climate which makes it rich in biodiversity, which can be used as antibacterial agents. Plantain (*Musa Paradisiaca L*) is a fruit that is easy to get, but the skin is rarely used. The Pisang Raja peel (*Musa Paradisiaca L*) contains chemical compounds saponins, tannins, flavonoids, and alkaloids which have antimicrobial properties. **Objective:** To determine the effectiveness of Pisang Raja peel (*Musa Paradisiaca L*) extract against the inhibition of *Porphyromonas gingivalis* bacteria as a cause of periodontitis. **Methods:** Using laboratory experimental methods, the test was carried out in the laboratory with the form of research in the form of Posttest Only Control Design and sampling by random sampling through 4 treatments and 6 repetitions. Statistical test was One Way Anova. **Results:** Based on the oneway Anova test, the concentration group of 16%, 32%, and 64% and the control group obtained a p-value of ($p < 0.01$) which means that there is a significant difference. Conclusion: Based on the results of this study, it is shown that the effectiveness of Pisang Raja peel extracts concentrations with concentration 16%, 32%, and 64% in inhibiting *Porphyromonas gingivalis* bacteria and control group obtained p-value ($p < 0,01$) means there was significant correlation. **Conclusion:** Based on this research that there is effective correlation Pisang Raja peel extract with concentration 16%, 32%, dan 64% can inhibit the growth *Porphyromonas gingivalis*.

Keywords: Periodontitis, *Porphyromonas gingivalis*, Pisang Raja Peel (*Musa Paradisiaca L*)

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Introduction

Most people still do not make oral health problems their top priority. However, as we know, teeth and

mouth are “gateways” for the entry of bacteria so that they can interfere with the health of other organs. According to Riskesdas 2007 and 2013, the percentage of the population having dental and mouth problems increased from 23.2% to 25.9%. Of the population with dental and oral health problems, the proportion of the population receiving medical care increased from 29.7% in 2007 to 31.1% in 2013.^[1]

Periodontal disease is a chronic inflammatory infection that causes damage to the supporting tissues of the teeth, characterized by loss of connective tissue adhesions and resorption of alveolar bone. The most common periodontal diseases are gingivitis and periodontitis. Periodontitis is a complex, localized chronic inflammatory disease characterized by the destruction of connective tissue, periodontal ligaments, and bone supporting teeth. This periodontal disease is caused by plaque bacteria on the surface of the teeth, where plaque is a thin layer of biofilm which contains a collection of pathogenic microorganisms such as *Porphyromonas gingivalis*, *Actinobacillus actinomycetemcomitans*, *Prevotella intermedia*, *Tannerella forsythia* and *Fusobacterium nucleatum* which are soft deposits.^{[2], [3],[4], [5]}

Porphyromonas gingivalis is a gram-negative, rod-shaped, black pigmented bacteria found in subgingival plaques. *Porphyromonas gingivalis* produces several virulences, namely fimbria, capsules, polysaccharides, lipopolysaccharides and hemolysis which are pathogenic in the oral cavity. *Porphyromonas gingivalis* is a bacterium that is often associated with the pathogenesis of periodontitis. In the subgingival plaques of chronic periodontitis patients, 85% of these bacteria were found. As an opportunistic bacterium that triggers periodontitis, *Porphyromonas gingivalis* expresses various virulence factors, such as fimbriae, capsules, lipopolysaccharide (LPS), membrane proteins and membrane vesicles. Gingipain and lipopolysaccharide (LPS) were used to invade the periodontium tissue.^{[6],[7]}

Indonesia is a country with a tropical climate which makes it has a lot of biodiversity, which can be used

as antibacterial. One of them is the plantain banana with the scientific name *Musa Paradisaca L* which has anti-inflammatory, antioxidant and antibacterial properties.⁸ Several studies that have been conducted using plantain peels state that plantain peels contain saponins, alkaloids, tannins, quinones and flavonoids. which has activity as an antimicrobial.⁹ Based on this description, the authors are interested in conducting research on the Effectiveness of Banana Peel Extract (*Musa paradisiaca L*) on Inhibition of *Porphyromonas gingivalis* bacteria as a cause of periodontitis.

Methods

The research design used is True Experiment in the laboratory, namely testing is carried out in the form of research in the form of Posttest Only Control Design. This research was conducted at the Phytochemical Laboratory of the Faculty of Marine and Fisheries Sciences and the Laboratory of Microbiology, Faculty of Medicine, Hasanuddin University. This research was conducted from February-April 2021 to completion. The samples in this study were plantain peel extracts with various concentrations, namely 16%, 32%, and 64% which were tested on cultured *Porphyromonas gingivalis*.

A sample of plantain peel was weighed as much as 300 grams and extracted using the maceration method, soaked using 96% ethanol solvent for 24 hours with occasional stirring every 1 hour. The maceration results were filtered using a vacuum filter to extract the filtrate to obtain the ethanol maserate of plantain peels. The ethanol maserate was then concentrated using a rotary evaporator to obtain a thick extract of banana peels. Then the dilution was carried out using DMSO (Dimethyl sulfoxide) which aims to produce a concentration of 16%, 32% and 64%.

Mueller Hinton Agar (MHA) is placed on a petri dish, then the bottom of the petri dish is divided according to the number of paper disks. Before testing the inhibitory power, then making a suspension of *Porphyromonas gingivalis* bacteria by first using a loop needle that was previously heated above a methylated

lamp to pick up bacteria, then the bacteria on the loop needle was inserted into a test tube containing 0.85% NaCl. Then the solution is homogenized. Then enter the bacteria on the loop needle into the MC solution. Farland 0.5 to equalize the turbidity and capacity standard for the number of bacteria that will be taken.

Inhibition test on bacteria using agar disc diffusion method. In this test, replication was carried out 6 times. A total of 6 pieces of petri dishes containing MHA medium were inoculated with Porphyromonas gingivalis bacteria. Soaked the paper disk at each concentration and then put it on each petri dish. Then incubate all the petri dishes containing bacteria and banana peel extract for 1x24 hours with a temperature of 37°C. In each test group with 6 repetitions. The width of the clear zone formed is calculated using

a digital digital term. The clear zone formed is a zone of inhibition of the growth of the bacterium Porphyromonas gingivalis.

Result

The study was conducted by measuring the inhibition zone of plantain peel extract with a concentration of 16%, 32%, 64% and Chlorhexidine 0.2% against the growth of Porphyromonas gingivalis bacteria. After conducting research to measure the inhibition of growth of the Porphyromonas gingivalis bacteria using a 16%, 32%, 64% concentration of Banana Banana Skin (Musa Paradisiaca L) extract solution, and a positive control of 0.2% chlorhexidine with 6 replications, the results were obtained. research can be seen in table 1.

Table 1. Diameter of Inhibitory Power Zone of Banana Banana Skin (Musa Paradisiaca L) Concentrations of 16%, 32%, 64% and Chlorhexidine 0.2% on the Growth of Porphyromonas gingivalis Bacteria

Replication	Banana Banana Skin Extract (Musa Paradisiaca L)						Control (mm)	
	Concentration 16%	Mean ± SD	Concentration 32%	Mean ± SD	Concentration 64%	Mean ± SD	K+	Mean ± SD
1.	12	13,76± 0,47	13,6	15,51± 1,40	15,1	17,11 ± 1,61	14,2	16,00± 1,82
2.	13,3		14,2		15,4		13,5	
3.	13,5		16,2		17,1		16,6	
4.	14,1		17,1		19,2		18,4	
5.	14,2		15,3		18,3		16,9	
6.	15,5		16,7		17,6		16,4	

Table 1 shows that the inhibition zone has been formed in a solution of banana skin extract (Musa paradisiaca L) at a concentration of 16%, 32%, 64% and a positive control solution of 0.2% chlorhexidine. The measurement results in the table above show that

the diameter of the bacterial inhibitory zone in the banana peel extract solution (Musa paradisiaca L), the concentration of 16 %% at replication 1 is 12 mm, replication 2 is 13.3 mm, replication 3 is 13.5 mm, replication 4 is 14.1 mm, replication 5 is 14.2 mm, replication 6 is 15.5 mm. Inhibition zone of bacteria

in plantain peel extract solution (*Musa paradisiaca* l) concentration 32% at replication 1 is 13.6 mm, replication 2 is 14.2 mm, replication 3 is 16.2 mm, replication 4 is 17.1 mm , replication 5 is 15.3 mm, replication 6 is 16.7 mm. Then in the zone of bacterial inhibition in plantain (*Musa paradisiaca* l) peel extract solution, the concentration of 64% at replication 1 is 15.1 mm, replication 2 is 15.4 mm, replication 3 is 17.1 mm, replication 4 is 19, 2 mm, replication 5 is 18.3 mm, replication 6 is 17.6 mm.

In the control solution, a positive control solution

of 0.2% chlorexidine was used which showed an inhibitory power of 14.2 mm in replication 1. in replication 2 it was 13.5 mm, in replication 3 it was 16.6 mm, then in replication 4 it was 18.4 mm, then in replication 5 it was 16.9 mm and in replication 6 it was 16.4 mm. The biggest zone of bacterial inhibition is in a solution of banana skin extract (*Musa paradisiaca* L) with a concentration of 64% in replication 4, which is 19.2 mm, while the lowest zone of inhibition is in a solution of banana skin extract (*Musa Paradisiaca* L) with a concentration of 16 % for replication 1, which is 12 mm.

Tabel 2. Uji Normalitas

Type of solution		Mean ± SD	p-value Shapirol wilk
Banana Peels (<i>Musa Paradisiaca</i> L) extract solution	Concentration 16%	13,76± 0,47	0.913
	Concentration 32%	15,51± 1,40	0.620
	Concentration 64%	17,11 ± 1,61	0.660
Positive Control	<i>chlorexidine</i> 0,2%	16,00± 1,82	0.578

Note: Normality Test; Shapirol Wilk test: p> 0.05, normal data distribution

* Anova One-way test: p <0.01: significant

Based on the results of the Shapirol Wilk normality test, the overall solution of both the banana skin extract solution (*Musa Paradisiaca* L) with a

concentration of 16%, 32%, 64% and a positive control of 0.2% chlorexidine showed a p-value> 0.05 so it can be concluded that all normally distributed data.

Tabel 3. Uji One Way Anova

Group	Comparation	Mean Difference	Std. Error	p-value/ sig.	p-value
Banana Peels (<i>Musa Paradisiaca</i> L) extract solution 16%	Banana Peels (<i>Musa Paradisiaca</i> L) extract solution 32%	-1.75000	0,876	0.060	0.009
	Banana Peels (<i>Musa Paradisiaca</i> L) extract solution 64%	-3.35000	0,876	0.001*	
	Chlorexidine 0,2%	-2.23333	0,876	0.019*	
Banana Peels (<i>Musa Paradisiaca</i> L) extract solution 32%	Banana Peels (<i>Musa Paradisiaca</i> L) extract solution 64%	-1.60000	0,876	0.083	
	Chlorexidine 0,2%	-0.48333	0,876	0.588	
Banana Peels (<i>Musa Paradisiaca</i> L) extract solution 64%	Chlorexidine 0,2%	1.11667	0,876	0.217	
*. The mean difference is significant at the 0.05 level., *Post Hoc test: Low Significant Difference (LSD) test; p<0.05: significant					

Based on the results of the One Way ANOVA statistical test carried out, it showed a p-value of 0.009 ($p < 0.05$) or a p-value less than 0.05, meaning that there was a significant difference between the zones of inhibition of bacterial growth using a solution of bark extract. Banana (*Musa Paradisiaca* L) concentrations of 16%, 32%, 64% and Chlorhexidine 0.2% positive control against the growth of *Porphyromonas gingivalis* bacteria.

Discussion

This study has proven that banana peel extract has an effective inhibitory effect in inhibiting the growth of *Porphyromonas gingivalis* bacteria based on the results of statistical effect tests that have been carried out. This plantain peel extract can inhibit bacteria because of its phytochemical content in the form of effective antibacterial compounds. This is consistent with the research that tested the phytochemical compounds contained in plantain peels and identified the presence of flavonoids and saponins. The mechanism of flavonoids as antibacterials is to inhibit the synthesis of nucleic acids, quercetin, mostly due to inhibition of DNA gyrase. Sophora flavone G and (-) epigallocatechin gallate have been suggested to inhibit cytoplasmic membrane function, whereas licochalcones A and C can inhibit energy metabolism.^[10]

The study was conducted in Nigeria by examining the ratio of plantain between the peel extract and its fruit extract against ten types of gram positive and negative bacteria. They obtained the results that most of the bacteria had more effect on the plantain peel extract than the fruit extract. This is due to the higher percentage of hydrocarbons, monoterpenes and oxygenated monoterpenes as potential antibacterial components in plantain peels.^[11]

Ahmed (2016) states that plantain peels contain chemical compounds contained in plantain peel extract which can inhibit gram-negative and gram-positive bacteria. The chemical compounds found in banana peels are flavonoids and tannins which

have biological and pharmacological properties. Plantain peels contain flavonoids and tannins which are important phytochemicals with various properties including, antibacterial, anticancer, anti-inflammatory, and antioxidant activity.^[12]

Davis and Stout (1971) determined that the inhibition zone criteria showed that the antibacterial strength of 20 mm or more means very strong, 10-20 mm means strong, 5-10 mm means moderate and 5 mm or less means weak. In this study, the inhibition zone results were obtained with a concentration of 16%, 32% and 64% in mm (millimeters), namely, 13.76 mm, 15.51 mm, and 17.11 mm. Results obtained from concentrations of 16%, 32%, and 64% all indicate that above 10mm means that the strength of the antibacterial power is strong. The results of this study are consistent with research conducted at Sam Ratulangi University, Manado, Faculty of Dentistry regarding the inhibitory ability of brown algae against *porphyromonas gingivalis*, which found that the higher the concentration of the extract, the greater the resulting inhibition.^{[13],[14]}

However, the anti-bacterial activity of plantain peel extract concentrations of 16% and 32% were still lower than chlorhexidine which was used as a positive control. Chlorhexidine is a mouthwash that can reduce plaque formation, inhibit plaque growth and prevent periodontal disease. This is due to the nature of Chlorhexidine itself, which is a bactericid and bacteriostatic against various kinds of bacteria. The mechanism of action of chlorhexidine is effective in inhibiting growth and killing gram-positive and gram-negative bacteria, depending on the concentration used. Chlorhexidine molecules have a positive charge (cations) and most of the bacterial molecular charges are negative (anions). This causes the strong adhesion of chlorhexidine to the bacterial cell membrane. Chlorhexidine will cause changes in the permeability of the bacterial cell membrane, causing the release of cell cytoplasm and cell components with low molecular weight from inside the cell to penetrate the cell membrane, causing bacterial death.^[15]

Conclusion

Plantain peel extract (*Musa paradisiaca* L) concentrations of 16%, 32% and 64% were effective in inhibiting the inhibition of *Porphyromonas gingivalis* bacteria and there were significant differences between the three concentrations in inhibiting *Porphyromonas gingivalis* bacteria ($p < 0.05$). However, the concentration of 64% and chlorhexidine 0.2% was more effective in inhibiting *Porphyromonas gingivalis* bacteria.

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Ethical Considerations: Ethical clearance was obtained from Universitas Muslim Indonesia, Makassar; with number" 085/A.1/KEPK-UMI/III/2021. Just before the interview, written (or thumb impression) consent was obtained from each participant in Universitas Muslim Indonesia, Makassar guidelines.

Conflicts of Interest: The authors alone are responsible for the views expressed in this article and they do not necessarily represent the views, decisions, or policies of the institutions with which they are affiliated.

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