Ad-Dawaa' Journal of Pharmaceutical Sciences
ISSN: 2654-7392, E-ISSN: 2654-6973
Vol. 6, No. 1, June 2023, p. 83 – 92
DOI: https://doi.org/10.24252/djps.v6i1.38774

Formulation and Antibacterial Activity of Porang (*Amorphophallus muelleri* Blume) Extract Gel Against Propionibacterium acne

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Article history:
Submited: 21-6-2023
Revised: 3-7-2023
Accepted: 13-7-2023

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Cite this article: Tahar, N., Wahyuni, D., Adawiyah, R., Khaerani, Wahyuddin, M. (2023). Formulation and antibacterial activity of porang (*Amorphophallus muelleri* Blume) extract gel against *Propionibacterium acne*. Ad-Dawaa' J. Pharm. Sci. 6(1): 83-92.

ABSTRACT

Introduction: The increase in bacterial resistance to antibiotics provides an opportunity to obtain antibacterial compounds from natural ingredients. Porang is a plant belonging to the Araceae family that has potential against Propionibacterium acne. **Aims**: This study aims to determine the effectiveness of the preparation of porang tuber extract gel against *Propionibacterium acne*. **Methods**: The porang were extraction by maceration using ethanol. The extract porang were made gel with various concentration and evaluation their stability and antibacterial activity using disc diffusion method. **Result**: The hedonic test showed no significant differenece each of formula and stable during storage in two weeks. On the other hand, only formula F2 and F3 can inhibit of *Propionibacterium acne*. **Conclusion**: The porang extract can made of gel and good stability. The formula F2 and F3 have antibacterial activity.

KEYWORDS: Porang tubers, gel, *Propionibacterium acne*, formulation, antibacterial activity.

INTRODUCTION

The *Amorphophallus muelleri* Blume (porang) plant is a member of the taro family (Araceae). This edible Araceae is a native Indonesian plant that grows wild in tropical forests. The general public in Java knows this plant by the name of porang. This plant thrives in the lowlands, mountains, and forests with tall trees. Porang grows among the tall plants around it, such as teak and sono trees, as a shelter (Wahida, Afiati, & Jumari., 2021).

In a previous study, porang tuber has effectiveness as an antibacterial such as *Staphylococcus aureus and Escherichia coli* bacteria. Porang tubers are not only used as food ingredients but can be used as medicinal ingredients because they contain certain compounds such as alkaloids as antibacterials (Erlina & Muhtadi, 2021). The porang is very promising for export at high price values which can be made into various kinds of processed products, ranging from food, cosmetics, and other industrial raw materials. In addition, it can be processed as an ingredient/additive in various cake products, tablet fillers and binders, microbial growth

media, and thickeners in syrups and fruit juice products (Yasin, et al., 2021).

The problem of acne on the facial skin is also caused by personal hygiene and environmental cleanliness. However, washing your face can reduce and prevent acne which can be done by everyone. Facial soap in gel formulation can used to remove dirt from the surface of the skin, and in general, several facial cleansing products often contain active ingredients or a combination of active ingredients to kill acne-causing bacteria (Marliana, Sartini, & Karim, 2018). Regular use of soap is one of the important components of effective acne management, not only increases antimicrobial activity, but also reduces the risk of infection, removes excess sebum, and prevents hair follicle obstruction (Hastuti, Mustifah, Ulya, Risman, & Mwardi, 2019). This study aims to formulate of porang tuber extract gel and determine antibacterial activity against *Propionibacterium acne*.

MATERIAL AND METHODS

Material

The materials used in this study were porang tuber, carbomer 940 (NIPest® 40), triethanolamine (TEA), sodium luryl sulphate (SLS), methyl paraben, propyl paraben purcashing by Merck, Super Tetra® (Kimia Farma), aquadest (OneMed). *Propionibacterium acne* suspension, NaCl® (Widatra Bhakti), muller hinton agar, nutrient agar, nutrient broth by Merck.

Extraction

Extraction was conducted using the maceration method. Crude drug powder of 450 grams were extracted with ethanol (96%) (ratio 1:10) and the filtrate and residue were then separated. The extracts were evaporated using a rotary vacuum evaporator with a temperature of 50 °C to produce porang tuber extracts (Dirgantara, Fidrianny, & Insanu, 2022).

Phytochemical Screening

Saponin

As much as 0.5 g of porang tuber extract to 5 ml of water in a test tube. Then shake the test tube vigorously and observe a stable froth. The foam was mixed with 3 drops of olive oil and shaken vigorously at 60° after which it was observed that a stable emulsion was formed (Rajendra, Gopal, Mahaboob, Yashoda, & Manjula, 2011).

Tannin

Heat 0.5 g of porang tuber extract and add a little distilled water to a test tube. Then, a solution of 1% gelatin in 10% sodium chloride was added to the porang tuber extract and observed for a white precipitate indicating the presence of tannins (Rajendra, Gopal, Mahaboob, Yashoda, & Manjula, 2011).

Flavonoids

Three methods can be used to test for flavonoids. First, add a few drops of 1% neutral ferric chloride or $FeCl_3$ to a portion of the aqueous filtrate of the extract. Flovanoid compounds are indicated by the formation of a

Table 1. Formula of porang gel

Material	F0 (%)	F1 (%)	F2 (%)	F3 (%)	F4 (%)
Porang tuber extract	-	3	5	7	-
Super tetra	-	-	-	-	250 mg
Carbopol	0.5	0.5	0.5	0.5	0.5
TEA	0.56	0.56	0.56	0.56	0.56
SLS	1	1	1	1	1
Methyl paraben	0.1	0.1	0.1	0.1	0.1
Propil paraben	0.5	0.5	0.5	0.5	0.5
Propylene glicol	5	5	5	5	5
Aquadest (ml)	add 100	add 100	add 100	add 100	add 100

black-green color when standing. Second, a few drops of lead acetate solution or 10% acetic acid are added to a portion of the extract. The presence of flavonoids is indicated by a yellow precipitate. Third, some of the extracts was dissolved in methanol, magnesium powder was added to it, and one ml of concentrated HCl was added from the side of the test tube. Magenta or orange color indicates the presence of flavonoids (Rajendra, Gopal, Mahaboob, Yashoda, & Manjula, 2011).

Alkaloid

As much as 0.5 g of porang tuber extract was diluted separately in 10 ml of 2N HCL, boiled, and filtered. Then 5 ml of the filtrate was added with 2 ml of ammonia, then 5 ml of chloroform was added and shaken slowly to extract the alkaloid base. The chloroform layer was extracted with 10 ml of acetic acid (chloroform solution). A few drops of Dragendroff's solution are added to the chloroform solution, alkoloids can be indicated by the presence of a reddish-brown precipitate. A few drops of Mayer's reagent are added to the chloroform solution, alkoloids can be indicated by the presence of a creamy white or yellowish-white precipitate. A few drops of Wagner's solution are added to the chloroform solution. Alkaloids are characterized by the presence of a brown precipitate (Rajendra, Gopal, Mahaboob, Yashoda, & Manjula, 2011).

Formulation of Porang Gel

Super Tetra and the porang tuber extract in various concentrtion (Table 1) were dissolved in hot (100°C) water to create combination 1. The carbapol and TEA were expanded in 20 ml of hot water (100°C) until they completely inflated to create the gel basis (mixture 2). In 10 ml of hot water, the SLS, methylparaben, and propylparaben were dissolved (combination 3). Stir the mixtures 1, 2, and 3 until they are homogenous. As much as 100 mL of hot water was added, and the mixture was then homogenized while being agitated. The solution is then placed in a sterile container or tube after cooling, accelerated temperature storage and room temperature storage of preparations (Susianti, Juliantoni, & Han, 2021).

Characterization of Gel

Organoleptic

This test is carried out visually and can see directly the shape, color, smell of the gel that is made.

pH

The pH value of the porang tuber extract gel soap which was carried out using a pH meter. The pH measurement was carried out using a pH meter by taking 0.5 g of the gel preparation which had been diluted into 50 mL of distilled water and measured with a pH meter, let stand for a few moments and the results were visible on the pH meter information. Preparations that meet the skin criteria are in the interval 4.5 – 6.5 (Rahmawati & Setiawan, 2019).

Viscosity

This evaluation was carried out using a Brookfield viscometer. The spindle speed and no 3, namely at a certain size until it reaches 100% torque at room temperature or accelerated temperature. The viscosity test was conducted by placing 100 ml of gel in a tubular container and installing a certain spindle. The spindle must be immersed in the test preparation. The viscometer is turned on, and confirmed that the rotor can rotate at a certain rpm speed. Observe the pointer of the viscometer 64, which points to the number on the viscosity scale and then record it and multiply it by a factor of 100. The requirement for a good viscosity value for gel preparations is 50-1000 dPa.s with an optimal viscosity of 200 dPa.s, or 500-5000 cps (Hasanah, Indah, Anggraeni, Ismaya, & Puji, 2020).

Hedonic

This evaluation was carried out on 20 probands. The probands were asked to judge based on the color, aroma, texture and irritation of the gel soap preparation samples. Furthermore, the Probandus is expected to fill out the questionnaire paper that has been provided. To determine skin irritation, it is done by applying a gel soap preparation to the hands for 10 minutes, then observing (Susianti, Juliantoni, & Han, 2021).

Stability Test

Physical stability test was carried out for one month by storing the preparation at room temperature and accelerated temperature for 2 weeks. The preparations were stored at 4±2oC for 24 hours, then transferred to an oven at 40 ± 2ºC for 24 hours (one cycle).

Organoleptic evaluation, homogeneity, pH and viscosity were carried out according to the procedure above (Husni, Fitriani, & Baitariza, 2021).

Antibacterial Activity of Gel

Before testing the antibacterial activity the tools used that have been sterilized, nutrient media to be prepared, and bacteria have been cultured and made in the form of suspension with turbidity according to McFarland standards. The positive control used in this study was erythromycin which was dissolved with aqua pro injection with a concentration of 25 mg/l (0.005%). Antibacterial activity tests were carried out by means of 0.1 ml of bacterial suspension put into sterilized petri

dishes. Add agar nutrient media as much as 20 ml, stir until homogeneous, and let it solidify. After the media is solidified, a well hole of samples and controls is needed to be tested, then enter the 50 mg gel preparation, then incubate for 24 hours at a temperature of 35-37ºC. After 24 hours the diameter of the inhibition zone is measured (Pertiwi, Hafiz, & Salma, 2019).

RESULTS AND DISCUSSION

Extraction

The yield of porang tuber viscous ethanol extract was 4.4%. Maceration is a simple method of extracting simplicia powder by immersing the simplicia powder in the solvent liquid with the mechanism of the liquid solvent penetrating the cell wall, the active substance will be dissolved due to the difference in concentration between the solution of the active substance inside the cell and outside the cell so that the solution with high concentration will be pushed out of the cell. The solvent used is 96% ethanol, because it can penetrate more easily into cells, is universal in nature can attract all types of active substances, both polar, semipolar, and non-polar, and also has a low level of toxicity (Sarlina, Razak, & Tandah, 2017).

Phytochemical Screening

Phytochemical screening tests were carried out to determine the content of compounds in porang tuber extract. The result showed in Table 2. Phytochemical screening was carried out qualitatively on secondary metabolites that were considered to have antibacterial effectiveness, such as alkaloids, flavonoids, saponins, and tannins.

Table 2. Results of phytochemical screening of porang tuber ethanol extract

Screening metabolites	Result
Saponin	+
Tanin	-
Flavonoid	+
Alkaloid	+

Based on the saponin test, a positive result was obtained, namely that there were saponin compounds that had effectiveness as an antibacterial characterized by the presence of stable foam and emulsion. In the tannin test, a negative result was obtained, namely, there were no tannin compounds in the ethanol extract of porang tubers. This is in line with previous research where porang tuber extract does not contain tannin (Erlina & Muhtadi, 2021).

In the alkaloid test, positive results were obtained containing alkaloid compounds that have antibacterial effectiveness marked by a change in color after being reacted with a certain corrector. Previous research also reports that porang tuber extract positive contain alkaloid (Erlina & Muhtadi, 2021).

In the flavonoid test, positive results were obtained for containing flavonoid compounds that have antibacterial effectiveness marked by a change in color after being reacted with a certain corrector. The mechanism for the antibacterial activity of flavonoids. The red

Table 3. Characterization of gel

Parameter	F0	F1	F2	F3	F4
Color	Clear white	Milk brown	Brown	Dark brown	Yellow
Smell	Typical	Typical	Typical	Typical	Typical
Texture	Soft	Soft	Soft	Soft	Soft
Clarity	Transparent	Transparent	Transparent	Transparent	Transparent
pH	9.0	8.0	7.3	6.5	7.1
Viscosity (cP)	NA	351	108.6	346.6	NA

color change in the flavonoid test was caused by the addition of HCl and Mg. The purpose of using Mg and HCl metals is to reduce the benzopyran nucleus contained in the flavonoid structure to form red or orange flavilium salts (Sani, Subaidah, & Andayani, 2021).

Characteristic of Gel

An organoleptic test was carried out to describe gel preparations by visual and odor observation. From the observations, the color, shape, and smell were obtained as shown in the Table 3. The brown color of this gel comes from porang root extract. The brown color of the extract is due to the extraction of natural polar coloring compounds, especially from phenolic or polyphenolic polymers such as tannins, melanins, lignins, and quinones in plants (Sani, Subaidah, & Andayani, 2021). This gel has a distinctive smell of porang tuber extract which comes from the thick extract of porang tubers used. This dosage form is a gel, where the form is by the gel dosage form, which is a semisolid preparation.

pH

pH testing was carried out using the HI-2020 pH meter for the 3 formulas. Based on the results of the gel soap screening test, the pH value was obtained in the range of 6.5 - 8.0. This means that the pH of the gel soap-based formula meets the requirements according to SNI 06-4085-1996 which states that the pH of soap-based is 8-11 and the pH of detergent-based is in the range of pH 4-8 (Pertiwi, Desnita, & Luliana, 2020). It can be concluded that the pH test results have met the test requirements.

Hedonic Assay

The hedonic test is a test in organoleptic sensory analysis that is used to determine the magnitude of the difference in quality between several similar products by providing an assessment or score of certain properties of a product and to determine the level of preference of a product. This level of liking is called the hedonic scale, for example very like, like, rather like, somewhat dislike, do not like, really don't like, and so on. The one-way ANOVA statistical test showed that the texture, color, and aroma were $p>0.05$ which indicates that there is no significant difference between each formula.

Viscosity

Viscosity testing aims to determine the consistency of the preparation. The higher the

viscosity value, the more difficult it is for the drug to be applied to the skin, the lower the viscosity value, the easier it is for the drug to be used (Susianti, Juliantoni, & Han, 2021). A good viscosity value for gel preparations is 50-1000 dPa.s with an optimal viscosity of 200 dPa.s, or 500-5000 cps (Hasanah, Indah, Anggraeni, Ismaya, & Puji, 2020). In formula I, the viscosity values are obtained in the range of 182 – 441 cps. In formula II, viscosity values are obtained in the range of 108-110 cps, and III, viscosity values are obtained in the range of 324-390 cps. Based on the results of the viscosity test, it does not meet the viscosity requirements of the gel.

According to Wahyudin et al (2018), carbopol influenced the spreadability response and the viscosity of the preparation, because the addition of Carbopol concentration increased the viscosity response and decreased the spreadability response (Wahyuddin, Kurniati, & Aridewi, 2018). The higher the carbopol concentration, the higher the viscosity value, so the viscosity level of preparation is also higher because the amount of polymer that will form the gel base increases. The higher the concentration of carbopol, the greater the number of polymers. Vice versa, the lower the Carbopol content, the lower the viscosity value.

According to BPOM Regulation Number 13 of 2018 it states that the quality of a medicine (for example this gel soap as an acne medicine) depends on the starting material, packaging material, production process, and so Formulation and antibacterial activity of porang on. Thus, the influence of the quality and brand of the ingredients used also affects the results of the preparations made.

Stability Test

Based on the results of the organoleptic test (Table 4), the color of the formulas F1, F2, and F3 did not change. As for the color change, only the positive control formula containing tetracycline.

The results of pH measurements at room temperature and accelerated temperature showed that there was an increase and decrease in the pH of the gel preparation during storage which indicated that the preparation was less stable during storage. Even though the pH value has increased and decreased, it is not significant so it still meets the requirements. This means that the pH of the gel soap from each formula meets the requirements according to SNI 06-4085-1996 which states that the pH of soap is 8-11 and the pH of topical preparations is in the range of pH 4-8 (Pertiwi, Desnita, & Luliana, 2020).

Viscosity affects the spreadability of the preparation. The higher the viscosity, the lower the spreading power, and vice versa. Viscosity is affected by temperature, where increasing temperature will reduce the force between atoms by increasing the distance between atoms so that the viscosity of the preparation decreases. The formula after 2 weeks of storage becomes thicker. During the storage process, the water content in the gel decreases, causing the gel to become thicker.

Table 4. The results of the organoleptic stability test of storage at room temperature

Time	Parameter	F1	F2	F3
1 week (RT)	Color	Milk brown	Brown	Dark brown
	Smell	Typical	Typical	Typical
	Texture	Soft	Soft	Soft
	Clarity	Transparent	Transparent	Transparent
1 week (AT)	Color	Milk brown	Brown	Dark brown
	Smell	Typical	Typical	Typical
	Texture	Soft and tick	Soft, slightly runny	Soft and liquid
	Clarity	Transparent	Transparent	Transparent
2 week (RT)	Color	Milk brown	Brown	Dark brown
	Smell	Typical	Typical	Typical
	Texture	Slightly sticky	Soft and sticky	Soft and sticky
	Clarity	Transparent	Transparent	Transparent
2 week (AT)	Color	Milk brown	Brown	Dark brown
	Smell	Typical	Typical	Typical
	Texture	Soft and sticky	Soft, slightly viscous	Soft, slightly viscous
	Clarity	Transparent	Transparent	Transparent
1 week (RT)	pH	8.0	7.3	6.5
1 week (AT)	pH	6.6	5.8	6.0
2 week (RT)	pH	7.8	7.0	6.5
2 week (AT)	pH	7.2	6.4	6.7
1 week (RT)	Viscosity (cP)	351	108.6	346.6
1 week (AT)	Viscosity (cP)	2821	429.6	157.3
2 week (RT)	Viscosity (cP)	1201	560.6	432
2 week (AT)	Viscosity (cP)	1048	788.6	407.6

The high concentration of water content in the 3% formula is more than in other formulas, allowing the viscosity of the gel to change to be higher (Irianto, Purwanto, & Mardan, 2020).

Antibacterial Activity of gel

The results of the antibacterial effectiveness test on porang tuber extract facial soap gel preparation against Propionibacterium acne bacteria showed that porang tuber extract gel preparation had effectiveness against Propionibacterium acne bacteria which was marked by the formation of a clear circle indicating an inhibition zone around the disc paper. The gel preparations tested at a concentration of 3% had an inhibition zone of 0 mm, at a concentration of 5% it produced an average inhibition zone of 10.5 mm and at a concentration of 7%, it produced an average inhibition zone of 16 mm.

This shows that the antibacterial effectiveness of porang tuber extract is in the strong category. The categorization is based on the diameter classification of the inhibition zone which states that it is classified as weak if < 5 mm, if 5-10 mm it is included in the medium category, the strong category if > 10-20 mm, and if > 20-30 mm it is classified as very strong (Rahayu, Lahay, & Jamilah, 2021). The antibacterial effectiveness test showed that the negative control used, namely the formula without the active substance did not provide a zone of inhibition. These results indicate that the formula without the active substance does not affect antibacterial effect-

iveness. The negative control is used to ensure that the diameter of the inhibition zone is formed purely by the active compound isolated from porang tubers, not the influence of other substances.

According to previous research, porang tuber extract with a concentration of 3% produced an average inhibition zone of 7.6 ± 2.0 mm, then at a concentration of 5% it produced an average inhibition zone of 11.3 ± 1.1 mm, and at a concentration of 7% produced an average inhibition zone of 15.6 ± 3.0 mm (Erlina & Muhtadi, 2021). Based on the results, the higher the concentration of food, the greater the value of the inhibition zone. The value of the resulting inhibition zone is directly proportional to the concentration of the extract. From the results of the phytochemical screening, porang tubers contain saponins, flavonoids, and alkaloids which have an antibacterial mechanism.

CONCLUSION

The porang tuber extract gel preparation has effectiveness against *Propionibacterium acne* bacteria. The concentration of porang tuber extract facial soap gel that is most effective in inhibiting *Propionibacterium acne* bacteria is a formula with a concentration of 7%. Further research is needed from all the formulas that have been made so that this preparation can be developed on a larger scale as a facial wash gel for treating acne.

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