The Antibacterial Activities of Durian Rinds Extract (*Durio Zibethinus*) Against *Propionibacterium acne*

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Abstract. Acne vulgaris is a kind of skin diseases which caused by *Propionibacterium acne* bacterias. Nowadays, the treatments of acne are still using the antibiotics as a first protocol, but the overuses of this in a long duration can be resitants. Therefore, it is very important to find some candidates of natural products to become antibacterial agents in the acne treatments. One of the natural products is Durian (*Durio zibethinus* (Linn)), a part of durian which is called rinds of durian known as mesocarpium, it contains a lot of organic chemical compounds, there are alkaloids, saponins, flavonoids, tannins, and phenolics which have antibacterial activities especially in flavonoids. This study was aimed to determine the antibacterial activity of durian rinds extracts to the *Propionibacterium acnes* bacterias. This study was used as a randomized completed trial design. The test of antibacterial activities uses the dilution method in 96 well plate and diffusion method, then the amount of the colony was measured by colony counter. The treatments were using a series concentrations 2.5%, 5%, 7.5%, and 10%, the negative control using 10% of DMSO and the positive control using clindamycine. The Results showed that the rinds of durian have given the moderates antibacterial activities at the 7.5% concentrations with the MIC value of 5% and the colony number was 67 colonies. Based on this results, it might conclude that the rinds of durian have the potential as an antibacterial agent which was bacteriostatic.

1. Introduction
Acne is a chronic inflammatory disease of the polycbaceal glands characterized by blackheads, papules, pustules and nodules [1]. Nowadays, the acne treatments use some antibacterial compounds, namely benzoyl peroxide, retinoids also sulfur, hence the prolonged use can cause dermatitis and long-term use of antibiotics caused resistance. *Propionibacterium acnes* has been reported having resistance to tetracyclines and erythromycin [2], so, it is needed to find other candidates of acne treatments to solve the problems.

One of the alternative natural compounds used as natural products which have potentiality as an antibacterial were rinds of Durian (*Durio zibethinus* (Murr.)). In 2010, the material harvest of Durian yielded about 7,037 ton [3]. The rinds of Durian which have been reported to have antibacterial
activities against *Staphylococcus epidermidis* in 6.25% with inhibition zone was 9.66 mm [4], and *Bacillus cereus* in 35% with inhibition zone was 30 mm [5].

![Figure 1. Durian (D. zibethinus (Linn.)) rinds](image)

The colonies numbers have been found in MIC assay against *Escherichia coli*, and *Staphylococcus aureus* with the concentration of 5% are 29 colonies and 36 colonies. Then, the MBC (minimum bactericidal colony) against *E. coli* and *S. aureus* with the concentrations of 30% are one colony)[6]. However, the researches about the durian rinds have not been explored yet. So, to make it has economic values, we need to examine the durian plants and use the wastes to give more contribution to peoples.

2. Material and Methods

2.1. Materials

Durian (*D. zibethinus* (Linn)) rinds, bacterial tests *Propionibacterium acnes*, ethanol 96%, Nutrient Agar, Nutrient Broth.

2.2. Methods

2.2.1. The making of Simplicia and Extractions

The making of simplicias and extraction are done in several stages as follows: the durian (*D. zibethinus* (Linn.)) rinds are washed and then dried using an oven at 60°C. Simplicia of Durian (*D. zibethinus* (Linn.)) rinds are made of powder by grinding. The durian (*D. zibethinus* (Linn.)) rinds which have dried as much as 1 kg are mashed to become powder. Extraction with 96% ethanol done by maceration. The maserate concentrated with a rotary evaporator at a temperature of 50°C with a speed of 50 rpm and then dried in an oven at 40°C until a fixed weight was obtained [7].

2.2.2. The Preparations of Bacterial Tests

a. The Rejuvinations of bacterial tests

The bacterial test was Propionibacterium acnes. The Pure culture stock was taken as much as one ose then inoculated by scraping on NA medium, then incubated at 37°C for 24 hours. After that, the bacteria can be used as a microbial test [8].

b. The Making of bacterial suspensions

The rejuvenation results of Propionibacterium acnes were suspended with a physiological NaCl solution of 0.9% sterile and put into cuvette, then measured transmittance (T 25%) using a spectrophotometer with a wavelength of 580 nm in 25% bacteria and as a blank used physiological NaCl 0.9 [8].

c. The making of Solution tests

Extracts made in various series of concentrations are 2.5%, 5%, 7.5% and 10% as much as 0.1 gram extract are weighed then each is dissolved into 1 ml of DMSO. The positive control used was clindamycin 1% antibiotic which was made by weighing 1 gram of clindamycine powder and dissolving it in 100 ml of distilled water.
d. Antibacterial Assays
The antibacterial activity assay
Extracts made in various series of concentrations are 2.5%, 5%, 7.5%, and 10% as much as 0.1 gram extract are weighed then each dissolved into 1 ml of DMSO. The positive control used was clindamycin 1% antibiotic which was made by weighing 1 gram of clindamycin powder and dissolving it in 100 ml of distilled water.

Antibacterial activity assay was carried out by using the disc diffusion method. Here, a clear zone produced around the disc paper. Inoculated the bacterias into agar media, then drops the test solution with various concentrations and also the control solutions as much as 20 μl into sterile disc paper with a diameter of 6 mm. Then, the disc paper placed on the agar media that has been marked. The cup is tightly closed and incubated anaerobically at 37˚C. After 24 hours incubation was observed and measured the diameter of the inhibition area of the clear zone formed using a ruler so that the diameter of the inhibition area of the extract was known [5].

2.3. Data Analysis
The data of inhibition zone diameters were analyzed using SPSS, and the interpretation referred to [9]. Meanwhile, the Data of colony number will be analyzed statistically with variance analysis using SPSS that related to Standard Plate Count [10]

3. Results and Discussion
3.1. Phytochemical screening
Based on the results of the phytochemical screening, it was shown that the extract of durian rinds contained a class of chemical compounds such as flavonoids, phenolics, alkaloids, steroids and tannins.

3.2. Extraction
The total of wet samples are 11.7 kgs of durian rinds were obtained from the Selat Village of Muaro Jambi Regency, Jambi. Then, dried using an oven at 50°C to obtain 2.3 kgs of dried samples from the durian rinds. Furthermore, maceration and re-maceration of three times were carried out for three days using 96% ethanol and 94 grams of concentrated extract of durian rinds were obtained which was separated by macerated solvent using a rotary evaporator. The yield value obtained is 4.087%. The last, the ethanol extract of the durian rinds tested for its antibacterial activity [6].

3.3. Antibacterial Activities Assay
Based on the results of preliminary tests of antibacterial activity, the MIC values of durian rinds were obtained as shown in figure 2 and table 1.

![Figure 2. The preliminary tests of antibacterial activity](image)

The dilution method is suitable in identifying the MIC values, which can estimating each concentrations of antibacterial agent that poured to the agar media. The MIC value is the lowest concentration that can
inhibit the bacterial growth and can be seen visually. Hence, the MIC values were stated in mg/ml atau mg/L.

Table 1. Antibacterial Activities of Durian Rinds Extracts against \( P. \text{acnes} \)

| Consentations (%) | Inhibition Zone (mm) | Average Inhibition Zone (mm)±SE M |
|-------------------|---------------------|----------------------------------|
| \( R1 \)          | \( R2 \)            | \( R3 \)            | \( R4 \)            |
| P1                | 4.25                | 6.25               | 1                   | 3                   | 3.63±1.1 |
| P2                | 5.25                | 8.25               | 3.75                | 5.5                 | 5.68±0.9 |
| P3                | 5.75                | 6.75               | 10.5                | 5.25                | 7.06±1.1 |
| P4                | 5.25                | 8                  | 8                   | 7.75                | 7.25±0.6 |
| K+                | 29.7                | 29.2               | 5                   | 29.2                | 29.67±0.19 |
| K-                | 1                   | 1                  | 1                   | 1                   | 1±0.00 |

Acknowledgement: The differences Superscripts in the same column show the significant value (P<0.05). P1(Extracts of 2.5%), P2(Extracts of 5%), P3(Extracts of 7.5%), P4(Extracts of 10%), K-(DMSO 10%), K+ (clyndamicine 1.2%), SEM (Standard Error Mean)

The antibacterial activity has shown at a concentration of 5% with moderate levels of antibacterial activity interpretation related to Davis and Stout [9]. It can be concluded from the overall test concentration which provides antibacterial activity in the moderate category is the concentration of 5%, 7.5% and 10% with the diameters of inhibition zone between 5–10 mm but from all treatments, the strength of the antibacterial activity is still not comparable to the positive control which give a strong antibacterial activity.

3.4. The calculation of colony number

The data were analyzed statistically using the One-Way Anova, and it showed that the concentration of extract used had a significant effect (P<0.05) on the number of bacterial colonies). Next, for further test continued with Tukey HSD's, it showed a significant difference in colony number (P <0.05) of all treatments for positive controls and negative controls. It was seen that the number of bacterial colonies throughout procedure was lower compared to negative controls. It was indicated the negative controls did not give any inhibition and antibacterial activity of durian rinds extract is not comparable with positive ones. From the overall results of testing the number of colonies obtained is still in the range of the standard plate count, which is 30-300 colonies [10].

Table 2. Colony number of \( D. \text{zibethinus (Linn.)} \) against \( P. \text{acnes} \)

| Concentration of Test Solution | Colony Number | Average Colony Numbers ± SEM |
|-------------------------------|---------------|-------------------------------|
| \( R1 \)                      | 52            |                               |
| \( R2 \)                      | 49            |                               |
| \( R3 \)                      | 51            |                               |
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| Concentration of Test Solution | Colony Number | Average Colony Numbers ± SEM |
|-------------------------------|---------------|------------------------------|
|                               | R1  | R2  | R3  |                             |
| P1 (Extracts of 10%)          | 50.67 ± 0.88 |
| P2 (Extracts of 7.5%)         | 57  | 53  | 56  | 55.33 ± 1.20                |
| P3 (Extracts of 5%)           | 69  | 66  | 68  | 67.67 ± 0.88                |
| P4 (Extracts of 2.5%)         | 77  | 80  | 81  | 79.33 ± 1.22                |
| K+ (Clindamycin 1.2%)         | 14  | 12  | 15  | 13.67 ± 0.88                |
| K- (DMSO 10%)                 | 93  | 96  | 95  | 94.67 ± 1.22                |

Acknowledgement: The differences Superscripts in the same column show the significant value (P < 0.05). P1 (Extracts of 10%), P2 (Extracts of 7.5%), P3(Extracts of 5%), P4(Extracts of 2.5%), K+ (Clindamycin 1.2%), K- (DMSO 10%), SEM (Standard Error Mean).

There are a lot of active compounds that can be obtained from durian plants such as the flavonoids, alkaloids, steroids, which have many potential activities like antimicrobial, antiinflammation, antioxidants, etc. Therefore, the durian rinds have particular problems which are the lipids. So, it is needed to continue by fractinations, isolation, and purifications to get the active compounds by bioassay guidance.

4. Conclusion

Based on this results, it is conclude that the durian rinds have the potential as an antibacterial agent which was bacteriostatic. So for the next studies, it can be continuing by bioassay guidance to get the isolates.

References

[1] Harper, J. C, “Acne Vulgaris”, Birmingham: Departement of dermatology, University of Alabama, 2007.
[2] Hassanzadeh, P., M. Bahmani., and Mehrabani, D, “Bacterial Resistance to Antibiotics in Acne Vulgaris : an in Vitro study”. Indian J Dermatol, 2008, Vol 53 : 122-4.
[3] http://www.jambiprov.go.id/images/jambi_angka/5930BAB5.PERTANIAN.pdf Jambi in the number 2010 accessed on September 7th, 2017.
[4] Azhari, F, “Aktivitas Antibakteri Ekstrak Etanol Kulit Buah Durian (Durio zibethinus Murr.) terhadap Staphylococcus epidermidis dan Shigella sonnei serta Bioautografinya”, Skripsi, Surakarta : Universitas Muhammadiyah Surakarta, 2015.
[5] Jamal, K. P, “Uji Aktivitas Antibakteri Ekstrak Etanol Daging Kulit Buah Durian (Durio zibethinus Murr.) terhadap Bakteri Salmonella Thypi ATCC 14028 dan Bacillus cereus ATCC 11778 Penyebab Diare” Skripsi, Jambi : Universitas Jambi, 2017.
[6] Fitrianingsih, Diah Tri Utami, Indri Maharini, “Antimicrobial potency of Durian Rind Extracts (*Durio zibethinus*) on some microbes test in vitro” was presented at the ICCSCP on 5-6 July 2018 in Padang-West Sumatra, Universitas Jambi, Jambi, *unpublished*, 2018.

[7] Fitrianingsih, Diah Tri Utami, Indri Maharini, “Skrining Fitokimia dan Uji Aktivitas Sitotoksik Ekstrak Daun Dadap Serep (*Erythrina subumbrans*) Terhadap Sel Hela Secara In Vitro” Prosiding Seminar Nasional APTFI II Banjarmasin, 17-18 November 2017,ISBN 978-602-7321-2-8, pp. 149-155.

[8] Andi Nur Aisyah, Nurul Arfiyanti Yusuf, Hasliah, “Pengaruh variasi konsentrasi emulgator *phytocream* Terhadap kestabilan fisik formula krim ekstrak etanol daun Kelor (*moringa oleifera l.*) Dalam menghambat *Propionibacterium acnes*”, Prosiding Seminar Nasional APTFI II Banjarmasin, 17-18 November 2017,ISBN 978-602-7321-2-8, pp. 29-60.

[9] Davis, W.W. dan T.R.Stout. 1971. Disc Plate Method of Microbiological Antibiotic Assay. Journal of Microbiology. 22 (4) : 666-670.

[10] Dart, R. K. 1996. Microbiology For the Analytical Chemist, The Royal society of Chemistry, UK.